

**A002*****Methylobacterium carri* sp. nov., Isolated from Automotive Air Conditioning System**

Jigwan Son and Jong-Ok Ka\*

*Department of Agricultural Biotechnology and Research Institute of Agriculture and Life Sciences, Seoul National University*

A bacterial strain, designated DB0501<sup>T</sup>, with Gram-stain-negative, aerobic, motile, and rod-shaped cell, was isolated from an automotive air conditioning system collected in the Republic of Korea. 16S rRNA gene sequence analysis indicated that the strain DB0501<sup>T</sup> grouped in the genus *Methylobacterium* and closely related to *Methylobacterium platani* PMB02<sup>T</sup> (98.8%), *Methylobacterium currus* PR1016A<sup>T</sup> (97.7%), *Methylobacterium variabile* DSM 16961<sup>T</sup> (97.7%), *Methylobacterium aquaticum* DSM 16371<sup>T</sup> (97.6%), *Methylobacterium tarhaniae* N4211<sup>T</sup> (97.4%) and *Methylobacterium frigidaeris* IER25-16<sup>T</sup> (97.2%). Genomic relatedness between strain DB0501<sup>T</sup> and its closest relatives was evaluated using average nucleotide identity, digital DNA-DNA hybridization and average amino acid identity with values of 86.4–90.8%, 39.3 ± 2.6–48.2 ± 5.0% and 87.8–89.5% respectively. The strain grew 15–30°C, pH 5.5–8.0 and in 0–1.0% w/v NaCl. Summed feature 3 (C<sub>16:1</sub> ω7c and/or C<sub>16:1</sub> ω6c) and summed feature 8 (C<sub>18:1</sub> ω7c and/or C<sub>18:1</sub> ω6c) were the predominant cellular fatty acids in strain DB0501<sup>T</sup>. Q-10 was the major ubiquinone. The major polar lipids were phosphatidylethanolamine, phosphatidylglycerol, and phosphatidylcholine. The DNA G+C content of strain DB0501<sup>T</sup> was 70.8 mol%. Based on phenotypic, genotypic and chemotaxonomic data, strain DB0501<sup>T</sup> represents a novel species of the genus *Methylobacterium*, for which the name *Methylobacterium carri* sp. nov. (type strain DB0501<sup>T</sup> = KACC 21434<sup>T</sup> = JCM 34003<sup>T</sup>) is proposed.

**A003*****Raineyella fluvialis* sp. nov., an Actinobacterium Isolated from Freshwater Sediment**Yeon Bee Kim<sup>1,2</sup>, Joon Yong Kim<sup>1</sup>, Hye Seon Song<sup>1,2</sup>, Juseok Kim<sup>1</sup>, Seung Woo Ahn<sup>1</sup>, Tae Woong Whon<sup>1</sup>, Se Hee Lee<sup>1</sup>, Jin-Kyu Rhee<sup>2</sup>, and Seong Woon Roh<sup>1\*</sup><sup>1</sup>World Institute of Kimchi, <sup>2</sup>Ewha Womans University

A novel facultative anaerobic actinobacterium, designated strain CBA3103<sup>T</sup>, was isolated from sediment of the Geum River in South Korea. Phylogenetic analysis indicated that strain CBA3103<sup>T</sup> is most closely related to *Raineyella antarctica* LZ-22<sup>T</sup> (98.47% 16S rRNA gene sequence similarity). The genome of strain CBA3103<sup>T</sup> was 3,649,865 base pairs with a 69.6 mol% G+C content. The average nucleotide identity value between strain CBA3103<sup>T</sup> and *Raineyella antarctica* LZ-22<sup>T</sup> was 79.22%. Cells of strain CBA3103<sup>T</sup> were Gram-positive, rod-shaped, 0.6–0.9 μm wide, and 1.4–2.4 μm long. Growth occurred at 15–40°C (optimum, 35°C), at pH 6.0–7.0 (optimum, pH 7.0), and 0–2% (w/v) (optimum, 0–1% [w/v]) NaCl concentrations. The major cellular fatty acids in strain CBA3103<sup>T</sup> were anteiso-C<sub>15:0</sub>, anteiso-C<sub>15:1</sub> A, and iso-C<sub>14:0</sub>. The major respiratory quinone was menaquinone-9(H<sub>4</sub>). The polar lipids of strain CBA3103<sup>T</sup> were diphosphatidylglycerol, phosphatidylglycerol, phosphatidylethanolamine, five unidentified glycolipids, and three unidentified phospholipids. Based on the genotypic, phenotypic, and chemotaxonomic analyses, strain CBA3103<sup>T</sup> represents a novel species of the genus *Raineyella*, for which the name *Raineyella fluvialis* sp. nov. (type strain CBA3103<sup>T</sup> = KACC 21446<sup>T</sup> = DSM 110288<sup>T</sup>) is proposed.

[Supported by World Institute of Kimchi funded by the Ministry of Science and ICT (KE2001-2), NRF (2018R1D1A1A09082921 and 2018R1D1A1B07045349), and the Ewha Womans University Research Grant of 2019.]

**A004*****Collinsella acetigenes* sp. nov., an Anaerobic Actinobacterium Isolated from Human Feces**

Kook-Il Han<sup>1</sup>, Mi Kyung Eom<sup>1</sup>, Ji-Sun Kim<sup>1</sup>, Min Kuk Suh<sup>1</sup>, Han Sol Kim<sup>1</sup>, Seung-Hwan Park<sup>1</sup>, Ju Huck Lee<sup>1</sup>, Se Won Kang<sup>1</sup>, Jam-Eon Park<sup>1</sup>, Byeong Seob Oh<sup>1</sup>, Seoung Woo Ryu<sup>1</sup>, Seung Yeob Yu<sup>1</sup>, Seung-Hyeon Choi<sup>1</sup>, Dong Ho Lee<sup>2</sup>, Hyuk Yoon<sup>2</sup>, Byung-Yong Kim<sup>3</sup>, Je Hee Lee<sup>3</sup>, and Jung-Sook Lee<sup>1,4\*</sup>

<sup>1</sup>Korean Collection for Type Cultures, Korea Research Institute of Bioscience and Biotechnology, <sup>2</sup>Seoul National University Bundang Hospital, <sup>3</sup>ChunLab, Inc., <sup>4</sup>University of Science and Technology (UST)

A novel actinobacterial strain, Gram-positive, anaerobic, non-motile and rod-shaped, designated KGMB02528<sup>T</sup>, was isolated from healthy human feces. Cells of strain KGMB02528<sup>T</sup> grew optimally at pH 7.0 and 37°C and in the presence of 0% (w/v) NaCl. Phylogenetic analysis based on 16S rRNA gene sequences revealed that strain KGMB02528<sup>T</sup> belonged to the family *Coriobacteriaceae*, and was most closely related to *Collinsella aerofaciens* DSM 17552<sup>T</sup> (95.8%). The phylogenetic and phenotypic data indicate strain KGMB02528<sup>T</sup> is clearly distinguished from the *Collinsella* lineage. The DNA G+C content was 58.0 mol%. The major cellular fatty acids (>10 %) were C<sub>16:0</sub> DMA, C<sub>16:0</sub> ALDE, C<sub>14:0</sub> DMA and C<sub>12:0</sub>. The predominant end products of carbohydrate fermentation was acetic acid. Strain KGMB02528<sup>T</sup> contained *meso*-diaminopimelic acid as the diamino acid in the peptidoglycan. Based on the phenotypic, chemotaxonomic, and phylogenetic properties, strain KGMB02528<sup>T</sup> represents a novel species of the genus *Collinsella*, for which the name *Collinsella acetigenes* sp. nov. is proposed. The type strain is *Collinsella acetigenes* KGMB02528<sup>T</sup> (= KCTC 15847<sup>T</sup>).

[This work was supported by the Bio & Medical Technology Development program (Project No. NRF-2016M3A9F3947962) of the National Research Foundation of Korea (NRF) funded by the Ministry of Science and ICT (MSIT) of the Republic of Korea and a grant from the Korea Research Institute of Bioscience & Biotechnology (KRIBB) Research initiative program]

**A006****Taxonomic Characterization and Antimicrobial Potential of *Micromonospora* Strains Isolated from River Side Soil**

Dong Hyeon Lee, Min Ji Kim, and Seung Bum Kim\*

*Department of Microbiology and Molecular Biology, College of Bioscience and Biotechnology, Chungnam National University*

*Micromonospora* is a promising actinobacterial group that has the potential to produce secondary metabolites. In this study, strains R2-23, R2-29 and R3-6 were separated from soil samples around the riverside of Geum River. The taxonomic status of these isolates was determined using a polyphasic approach and all three strains presented chemotaxonomic and morphological characteristics consistent with the classification in *Micromonospora*. For strains R2-23, R2-29 and R3-6, the highest 16S rRNA gene sequence similarities were observed with *Micromonospora phytophila* SG15<sup>T</sup> (99.29%), *Micromonospora rifamycinica* AM105<sup>T</sup> (99.37%) and *Micromonospora wenchangensis* CCTCC AA 2012002<sup>T</sup> (99.42%), respectively. However, each of the strains were readily distinguished genetically and phenotypically from their phylogenetic neighbors, indicating that they represent three new *Micromonospora* species. R2-23 and R2-29 were found to have the PKS1 and NRPS type biosynthetic gene clusters while R3-6 had only PKS1 type, based on PCR detection results. All three strains showed antibacterial activity against Gram positive bacteria including *Micrococcus luteus*, *Staphylococcus aureus*, *Corynebacterium diphtheria*, *Bacillus subtilis*, and *Enterococcus faecalis*, and also against yeast, namely *Candida krusei* and *Candida albicans*. Strain R2-29 was also active against *Saccharomyces cerevisiae*, and R3-6 against broad group of filamentous fungi.

**A007****Polyphasic Taxonomic Analysis for *Streptomyces bomunensis* sp. nov. Isolated from Mountain Soil Around a Dead Tree**

Jae Ha Lee, Ji Won Jeong, Dong Hyeon Lee, Chung Mi Kim, and Seung Bum Kim\*

*Department of Microbiology and Molecular Biology, College of Bioscience and Biotechnology, Chungnam National University*

This study aims to classify a new species candidate designated MMS17-BM035<sup>T</sup> belonging to the genus *Streptomyces*, which was isolated from soil under a rotten tree using a polyphasic taxonomic approach. Phylogenetic analysis based on 16S rRNA gene sequence comparisons revealed that the strain showed the highest sequence similarity to *Streptomyces fuscigenes* JBL-20<sup>T</sup> (99.45%), *Streptomyces tremellae* Js-1<sup>T</sup> (98.27%), *Streptomyces camponoticapitis* 2H-TWYE14<sup>T</sup> (97.71%), *Streptomyces gelaticus* NRRL B-2928<sup>T</sup> (97.58%), and *Streptomyces pulveraceus* LMG 20322<sup>T</sup> (97.51%). Growth occurred at 15–30°C (optimum, 30°C), at pH 5.0–7.0 (optimum, pH 6.0–7.0), and in the presence of 0–6% (w/v) NaCl (optimum, 0%). Good growth occurred on ISP media 2 and 3, and also on Bennett's agar. The predominant menaquinones were MK-9(H<sub>4</sub>) and MK-9(H<sub>6</sub>). The major polar lipids were diphosphatidylglycerol, phosphatidylethanolamine, phosphatidylinositolmannoside, three unidentified aminophospholipids, two unidentified glycolipids and two unidentified lipids. The major cellular fatty acids were anteiso-C<sub>15:0</sub>, a summed feature consisting of C<sub>18:1</sub>ω7c and/or C<sub>18:1</sub>ω6c, iso-C<sub>15:0</sub>, C<sub>20:0</sub> and C<sub>16:0</sub>. It is evident from this study that strain MMS17-BM035<sup>T</sup> merits recognition as a new species of *Streptomyces*, for which the name *Streptomyces bomunensis* sp. nov. (type strain=MMS17-BM035<sup>T</sup>) is proposed.

[Supported by a research grant from the National Institute of Biological Resources (NIBR).]

**A008*****Amanita brunneofolia*, a New Species in Section *Roanokenses* from South Korea**Jong Won Jo<sup>1</sup>, Young-Nam Kwag<sup>1</sup>, Hyung So Kim<sup>1</sup>, Hyun Lee<sup>1</sup>, Sang-Kuk Han<sup>2</sup>, Jae-Gu Han<sup>3</sup>, Seung Hwan Oh<sup>1</sup>, and Chang Sun Kim<sup>1\*</sup><sup>1</sup>*Division of Forest Biodiversity, Korea National Arboretum*, <sup>2</sup>*Research Planning and Coordination, Korea National Arboretum*, <sup>3</sup>*Mushroom Research Division, National Institute of Horticultural and Herbal Science, Rural Development Administration*

A new species of *Amanita* sect. *Roanokenses*, *A. brunneofolia*, from South Korea, is described based on morphological and molecular evidences. The species is characterized by medium- to large-sized basidiomata, a greenish white pileus covered with brownish, floccose pyramidal volval remnants, an appendiculate margin, reddish brown lamellae, a long radicating stipe, and ellipsoid to elongate amyloid basidiospores. Based on both nrLSU and combined dataset (nrLSU, *rpb2* and *tef1-α*), *A. brunneofolia* formed a monophyletic clade and clearly separated from other *Amanita* species. In addition, we describe other *Amanita* species in *A.* sect. *Roanokenses*, namely, *A. caojizong*. This is the first report of this species for South Korea.

[This work was supported by research grants of the Korea National Arboretum (project no. KNA 1-1-25, 19-2) and the National Institute of Horticultural and Herbal Science (project no. PJ01476601).]

**A009*****Bacillus bambusae* sp. nov., Isolated from Bamboo Grove Soil**

Ji won Jeong, Yeong Seok Kim, and Seung Bum Kim\*

*Department of Microbiology and Molecular Biology, College of Bioscience and Biotechnology, Chungnam National University*

Strain BG109 was isolated from Bamboo grove soil in Korea (35°16'25.5 N 127°28'34.5 E), and subjected to polyphasic taxonomic characterization. Strain BG109 was an aerobic, Gram-positive and endospore-forming bacterium. BG109 showed growth at 10°C–45°C (optimally 37°C), at pH 4–10 (optimally 8), and in the presence of 0–7% NaCl concentration (optimally 0–1%). The predominant menaquinone of BG109 was MK-7, and the cell wall peptidoglycan contained major amount of *meso*-diaminopimelic acid. The main fatty acids were anteiso-C<sub>15:0</sub> (24.55%), iso-C<sub>15:0</sub> (23.89%), iso-C<sub>14:0</sub> (11.99%), iso-C<sub>16:0</sub> (11.84%), and C<sub>16:0</sub> (10.50%). BG109 shared 98.17%, 97.13%, 97.00% 16S rRNA similarity with *Bacillus litoralis*, *Bacillus endolithicus*, *Bacillus niabensis*. Thus, it is evident that strain BG109 merits recognition as a new species of *Bacillus*, for which the name *Bacillus bambusae* sp. nov. is proposed.

[Supported by a research grant from the National Institute of Biological Resources (NIBR).]

**A010*****Paenibacillus artemisicola* sp. nov. and *Paenibacillus lignolyticus* sp. nov., Isolated from Plant Roots**

You Ju Ham, Ji Won Jeong, Dong Hyeon Lee, and Seung Bum Kim\*

*Department of Microbiology and Molecular Biology, College of Bioscience and Biotechnology, Chungnam National University*

Strain MWE-103<sup>T</sup> was isolated from the root of mugwort and strain DLE-14<sup>T</sup> from the root of daylily. Based on the 16S rRNA gene sequencing analysis, MWE-103<sup>T</sup> was mostly related with *Paenibacillus sacheonensis* SY01<sup>T</sup> (97.81%), and DLE-14<sup>T</sup> with *Paenibacillus rhizoryzae* IZS3-5<sup>T</sup> (98.33%). The optimal growth media of both strains were R2A and NA. The growth temperature range of both strains was 10°C to 37°C (optimum, 25°C to 30°C), and the NaCl concentration was 0-2% (optimum, 0%) for MWE-103<sup>T</sup> and 0-1% (optimum, 0%) for DLE-14<sup>T</sup>. The major fatty acids of MWE-103<sup>T</sup> were anteiso-C<sub>15:0</sub>, iso-C<sub>16:0</sub>, iso-C<sub>15:0</sub>, and C<sub>16:0</sub>, and those of DLE-14<sup>T</sup> were anteiso-C<sub>15:0</sub>, iso-C<sub>15:0</sub>, C<sub>16:0</sub>, iso-C<sub>16:0</sub>, and iso-C<sub>17:0</sub>. The major quinone of both strains was menaquinone 7. Amylase and cellulase activities were positive for both strains, and DLE-14<sup>T</sup> also degraded lignin. The strains could be differentiated from related species in enzyme activities and fatty acid composition. Based on these results, each of the two strains MWE-103<sup>T</sup> and DLE-14<sup>T</sup> merits recognition as a new species, for which the names *Paenibacillus artemisicola* sp. nov. (type strain=MWE-103<sup>T</sup>) and *Paenibacillus lignolyticus* sp. nov. (type strain=DLE-14<sup>T</sup>) are proposed. Further studies will include tests for plant growth promoting properties.

[Supported by a research grant from the National Institute of Biological Resources (NIBR).]

**A011****Three Species New to Korea Belonging to the Poisonous Genus *Gyromitra***

Hyun Lee, Jong Won Jo, Young-Nam Kwag, Sang-Kuk Han, and Chang Sun Kim\*

*Forest Biodiversity Division, Korea National Arboretum*

Species belonging to the genus *Gyromitra* are called as false morels and form cerebriiform ascocarps. It is well known that many *Gyromitra* species are poisonous. In Korea, only two species (*G. esculenta* and *G. infula*) are recorded. However, molecular phylogenetic analyses about this genus has never been conducted in Korea so far. In this study, we used specimens deposited in Korea National Arboretum. A phylogenetic tree inferred from nuclear rDNA internal transcribed spacers (ITS) was obtained, and four species were belonging to three clades. Three species (*G. gigas*, *G. perlata*, and *G. tianshanensis*) were new to Korea and *G. esculenta* also detected. Herein, we provide identification keys of Korean *Gyromitra* species and detailed description of the species new to Korea. [This study was supported by the Korea National Arboretum (KNA 1-1-25, 19-2).]

**A012*****Pseudomonas guryensis* sp. nov. and *Pseudomonas ullengensis* sp. nov., Isolated from Soil**

Chung Mi Kim, Ji Won Jeong, Dong Hyeon Lee, and Seung Bum Kim\*

*Department of Microbiology and Molecular Biology, College of Bioscience and Biotechnology, Chungnam National University*

Two Gram-staining-negative, aerobic, rod-shaped bacteria designated strains SR9<sup>T</sup> and UL070<sup>T</sup>, were isolated from soil and subjected to taxonomic characterization. Strain SR9<sup>T</sup> grew at 10–37°C (optimum 30°C), at pH 4.0–10.0 (optimum pH 8.0) and in the presence of 0-1% NaCl (optimum 0%), and UL070<sup>T</sup> at 4–33°C (optimum 30°C), at pH 4.0–10.0 (optimum pH 7.0) and in the presence of 0-2% NaCl (optimum 0%), respectively. Strain UL070<sup>T</sup> was motile with flagella. Analysis of 16S rRNA gene sequences indicated that the two strains fell into a phylogenetic cluster belonging to the genus *Pseudomonas*. Strain SR9<sup>T</sup> and UL070<sup>T</sup> were mostly related to *Pseudomonas peli* with 98.3% and 97.7% sequence similarities, as the similarity between the two isolates was 98.9%. The major fatty acids of the two strains were C<sub>18:1</sub>ω7c, a summed feature consisting of C<sub>16:1</sub>ω7c/C<sub>16:1</sub>ω6c, C<sub>16:0</sub> and C<sub>12:0</sub>. The major respiratory quinone was Q9, and the major polar lipids were phosphatidylethanolamine (PE) and diphosphatidylglycerol (DPG) for both strains. On the basis of both phenotypic and phylogenetic evidences, the isolated strains should be classified as a novel species, for which the name *Pseudomonas guryensis* sp. nov. (type strain= SR9<sup>T</sup>) and *Pseudomonas ullengensis* sp. nov. (type strain= UL070<sup>T</sup>) are proposed.

[Supported by a research grant from the National Institute of Biological Resources (NIBR).]

**A013****Description and Antimicrobial Potential of *Micromonospora humidus* sp. nov., Isolated from Riverside Soil**

Jun Sic Ra, Min Ji Kim, and Seung Bum Kim\*

*Department of Microbiology and Molecular Biology, College of Bioscience and Biotechnology, Chungnam National University*

*Micromonospora* is a genus considered a rare actinobacterial group and is the second largest producers of antimicrobial and other bioactive substances after *Streptomyces*, and their metabolites includes aminoglycoside, macrolide, and anthracycline antibiotics. Therefore, they were chosen as the main targets for experiments to screen new antibiotic substances. In this experiment, based on the previous leading researcher's experiment, *Micromonospora* strain R1-14<sup>T</sup> was isolated from a riverside soil sample, and its antimicrobial potential as well as taxonomic status was examined. The phylogenetic analysis of 16S rRNA gene sequence of strain R1-14<sup>T</sup> showed high similarity with '*Micromonospora wenchangensis* (99.51%), *Micromonospora rifamycinica* (99.37%)'. The gyeB gene sequence analysis showed high similarity with '*M. wenchangensis* (98.08%), *M. rifamycinica* (95.85%)'. Strain R1-14<sup>T</sup> formed an independent phylogenetic line, and thus was considered as a new species candidate. Strain R1-14<sup>T</sup> grew at pH 5–11 (optimum 8.0), 15–40°C (optimum 37°C), and in the presence of 0–4% NaCl (optimum 1%). The major cellular fatty acids were C<sub>18:1</sub>ω9c, iso-C<sub>15:0</sub>, C<sub>15:0</sub>-3OH, anteiso-C<sub>15:0</sub>, and iso-C<sub>16:0</sub>. The strain showed antagonistic activity against bacteria and yeast. Based on this study, a new species of *Micromonospora* is proposed, for which the name *Micromonospora humidus* sp. nov. is proposed (type strain=R-14<sup>T</sup>).

**A015*****Ferrimonas lipolytica* sp. nov., a Facultatively Anaerobic Bacterium Isolated from Seawater**

Seung Seob Bae, Yoon-Hee Jung, Yong Min Kwon, Dawoon Chung, Dae-Sung Lee, and Kyunghwa Baek\*

*National Marine Biodiversity Institute of Korea*

A Gram-stain-negative, motile, facultative anaerobic rod-shaped, designated strain S7<sup>T</sup> was isolated from a surface seawater sample collected at Uljin in the East Sea, South Korea. Phylogenetic analysis based on the 16S rRNA gene sequences revealed that strain S7<sup>T</sup> was affiliated with members of genus *Ferrimonas*, *Ferrimonas balearica* PAT<sup>T</sup> (95.7%), *Ferrimonas senticii* P2S11<sup>T</sup> (95.6%) and *Ferrimonas pelagia* CBA4601<sup>T</sup> (95.1%), respectively. The strain S7<sup>T</sup> has a single circular chromosome of 4.13 Mbp with a DNA G+C content of 49.4 mol%. The average nucleotide identity (ANI) and digital DNA-DNA hybridization (dDDH) between strain S7<sup>T</sup> and type strains of *F. balearica* PAT<sup>T</sup>, *F. senticii* P2S11<sup>T</sup> yielded ANI values of 70.6 and 71.9%, and dDDH values of 13.9 and 15.1%, respectively. Growth was observed at 8–31°C (optimum 27°C), at pH 7-9 (optimum pH 7), and with 1-6% NaCl (optimum 2). The respiratory quinones were Q-7 and MK-7 and the major fatty acids (>10%) were C<sub>16:0</sub>, C<sub>16:1</sub>ω9c, C<sub>17:1</sub>ω8c, and summed feature 3 (C<sub>16:1</sub>ω7c and/or C<sub>16:1</sub>ω6c). The major polar lipids were identified as phosphatidylethanolamine, phosphatidylglycerol, two unidentified phospholipids, and three unidentified lipids. On the base of this polyphasic approach, it was determined that the strain represents a novel species of the genus *Ferrimonas*, for which the name *Ferrimonas lipolytica* sp. nov. is proposed. Type strain is S7<sup>T</sup> (=KCTC 72490<sup>T</sup> =JCM 33793<sup>T</sup>).

[Supported by a grant from MABIK in-house program (2020M00500).]

**A016*****Nakamurella aerolatum* sp. nov., Isolated from Air Conditioner**Dae Young Kim<sup>1</sup>, Ki Eun Lee<sup>2</sup>, In Tae Cha<sup>2</sup>, and Dong Uk Kim<sup>1\*</sup><sup>1</sup>Department of Biological Sciences, Sangji University, <sup>2</sup>Microorganism Resources Division, National Institute of Biological Resources Biological Resources Research Department

A Gram-stain-positive, aerobic, white coloured coccus-shaped bacterium, designated DB0629<sup>T</sup>, was isolated from air conditioner, Republic of Korea. Phylogenetic analysis based 16S rRNA gene sequences indicated that strain DB0629<sup>T</sup> was affiliated with the genus *Nakamurella* in the family *Nakamurellales* and shared 93.66–97.57% sequence similarities with *Nakamurella* species. Whole genome sequencing of strain DB0629<sup>T</sup> revealed genome size of 4.3 Mbp and the G+C content of 69.4 mol%. The strain contained C<sub>15:0</sub> anteiso, C<sub>17:0</sub> anteiso, C<sub>16:0</sub> iso as major fatty acids and the predominant respiratory quinone was MK-8(H4). The major polar lipids were diphosphatidylglycerol (DPG), phosphoethanolamine (PE), phosphatidylethanolamine (PI), phospho-glycolipid (PGL). On the basis of phylogenetic and phenotypic characteristics, strain DB0629<sup>T</sup> is considered to represent a novel species of the genus *Nakamurella*, for which the name *Nakamurella aerolatum* (type strain DB0629<sup>T</sup>= KCTC 72726<sup>T</sup>) sp. nov. is proposed.

[This work was supported by a grant from the National Institute of Biological Resources (NIBR), funded by the Ministry of Environment (MOE) of the Republic of Korea (NIBR202002108)]

**A017*****Flexivirga aerolatum* sp. nov., Isolated from Air Conditioner**Dae Hyun Kim<sup>1</sup>, Ki Eun Lee<sup>2</sup>, In Tae Cha<sup>2</sup>, and Dong-Uk Kim<sup>1\*</sup><sup>1</sup>Department of Biological Science, Sangji University, <sup>2</sup>Microorganism Resources Division, Biological Resources Research Department, National Institute of Biological Resources

A Gram-stain-positive, aerobic, pale yellow-coloured, non-motile and coccus-shaped bacterium, designated ID2601S<sup>T</sup>, was isolated from air conditioner, Phylogenetic analysis based 16S rRNA gene sequences indicated that strain ID2601S<sup>T</sup> was affiliated with the genus *Flexivirga* in the family *Dermacoccaceae* and shared 97.92% sequence similarities with *Flexivirga endophytica* (type strain YIM 7505<sup>T</sup>) species. Whole genome sequencing of strain ID2601S<sup>T</sup> revealed genome size of 4.00 Mbp and the G+C content of 69.79 mol%. The strain contained summed feature 3 (C<sub>16:1</sub> ω6c/C<sub>16:1</sub> ω7c), summed feature 9 (C<sub>17:1</sub> iso ω6c/C<sub>16:0</sub> 10-methyl) as the major fatty acids and MK-8(H4) and MK-8(H6) as the major respiratory quinone. The polar lipids detected in the strain were diphosphatidylglycerol, phospholipid, phosphoglycolipid, unidentified aminolipid, unidentified amino phospholipid, two unidentified glycolipids and four unidentified lipids. On the basis of phylogenetic and phenotypic characteristics, strain ID2601S<sup>T</sup> is considered to represent a novel species of the genus *Flexivirga*, for which the name *Flexivirga aerolatum* (type strain ID2601S<sup>T</sup>= KCTC 49353<sup>T</sup>) sp. nov., is proposed.

[This work was supported by a grant from the National Institute of Biological Resources (NIBR), funded by the Ministry of Environment (MOE) of the Republic of Korea (NIBR202002108)]

**A018*****Oricola thermophila* sp. nov., a Marine Bacterium Isolated from Tidal Flat Sediment and Emended Description of the Genus *Oricola* Hameed et al. 2015**Sung-Hyun Yang<sup>1</sup>, Mi Jeong Park<sup>1,2</sup>, and Kae Kyoung Kwon<sup>1,2\*</sup><sup>1</sup>Marine Biotechnology Research Center, Korea Institute of Ocean Science & Technology, <sup>2</sup>University of Science and Technology

A Gram-negative, aerobic, rod-shaped (1.8-4.4  $\mu\text{m}$   $\times$  0.5-0.7  $\mu\text{m}$ ) and motile marine bacterium, designated as MEBiC13590<sup>T</sup> was isolated from tidal flat sediment of the Incheon City, West seaside of Korea. The 16S rRNA gene sequence analysis revealed that strain MEBiC13590<sup>T</sup> showed high similarity with the *Oricola cellulosilytica* CC-AMH-0<sup>T</sup> (98.0%) and followed by *Oceaniradius stylonematis* StC1<sup>T</sup> (97.2%), however, clustered with *Orc. cellulosilytica*. Growth was observed at 22-50°C (optimum 45°C), at pH 5-9 (optimum pH 7) and with 1-6% (optimum 3%) NaCl. The predominant cellular fatty acids were C<sub>16:0</sub> (7.6%), C<sub>18:0</sub> (12.2%), 10-methyl C<sub>18:1</sub> $\omega$ 7c (5.7%), C<sub>19:0</sub> cyclow6c and summed feature 8 (comprised of C<sub>18:1</sub> $\omega$ 7c and/or C<sub>18:1</sub> $\omega$ 6c; 38%). The DNA G+C contents is 63.5 mol%. The major respiratory quinone is Q-10. Several phenotypic characteristics such as urease, gelatinase and enzyme activities of lipase (C14),  $\alpha$ -chymotrypsin, acid phosphatase,  $\beta$ -galactosidase and  $\beta$ -glucosidase differentiate strain MEBiC13590<sup>T</sup> from *Oricola cellulosilytica* CC-AMH-0<sup>T</sup>. Based on this polyphasic taxonomic data, strain MEBiC13590<sup>T</sup> should be classified as a novel species in the genus *Oricola* and it is proposed as *Oricola thermophila* sp. nov. The type strain is MEBiC13590<sup>T</sup> (=KCCM 43313<sup>T</sup> =JCM 33661<sup>T</sup>).

**A019*****Flavobacterium humi* sp. Nov., a Flexirubin-type Pigment Producing Species Isolated from Soil**

Inhyup Kim, Geeta Chhetri, Jiyoun Kim, Minchung Kang, and Taegun Seo\*

Department of Life science, Dongguk University-Seoul

A yellow pigmented, Gram-stain-negative, rod-shaped, strictly aerobic, motile by means of gliding, oxidase and catalase positive bacterium, designated strain DS2-A<sup>T</sup>, was isolated from soil. Phylogenetic analysis of 16S rRNA gene sequence revealed that strain DS2-A<sup>T</sup> belonged to the genus *Flavobacterium* and was most closely related to *F. aquatile* LMG 4008<sup>T</sup> (96.4%), *F. terrae* DSM 18829<sup>T</sup> (95.6%), *F. vireti* THG-SM1<sup>T</sup> (95.5%), *F. inkyongense* IMCC27201<sup>T</sup> (95.4%), *F. brevivitae* TTM-43<sup>T</sup> (95.2%), and *F. cucumis* DSM 18830<sup>T</sup> (95.2%). Strain DS2-A<sup>T</sup> produces flexirubin-type pigments. The major polar lipid was found to be phosphatidylethanolamine. The average nucleotide identity values between strain DS2-A<sup>T</sup> and selected taxa, *F. aquatile* LMG 4008<sup>T</sup>, *F. terrae* DSM 18829<sup>T</sup>, and *F. cucumis* DSM 18830<sup>T</sup>, were 72, 72.7, and 71.6%, respectively. The draft genome of strain DS2-A<sup>T</sup> has a number of 14 contigs, scaffold N50 of 476,310 bp and a total size of 3,563,867 bp. Additionally, strain DS2-A<sup>T</sup> contains 3,127 of gene, 41 of tRNA, 6 of rRNA, and 3 of ncRNA. The DNA G + C content of stain DS2-A<sup>T</sup> was 40.7 mol%. Based on phylogenetic and phenotypic analyses, strain DS2-A<sup>T</sup> is considered as a novel species of the genus *Flavobacterium*, for which the name *Flavobacterium humi* sp. nov. (= KACC 19715<sup>T</sup> = JCM 32786<sup>T</sup>) has been proposed.

[This work was supported by a grant from the National Institute of Biological Resources (NIBR), funded by the Ministry of Environment (MOE) of the Republic of Korea (NIBR202002203).]

**A020*****Flavobacterium baculatum* sp. nov., a Flexirubin-type Pigment Producing Bacteria Isolated from the Soil of Jeju-do**

Geeta Chhetri, Jiyou Kim, Inhyup Kim, Minchung Kang, and Taegun Seo\*

*Department of Life Science, Dongguk University Seoul*

An aerobic, Gram-stain-negative, dull yellow-pigmented, oxidase and catalase-negative, non-motile, non-spore forming, long rod-shaped, designated strain SNL9<sup>T</sup>, was isolated from soil of crossroads of Jeju Island in South Korea. Flexirubin-type pigments were present. Phylogenetic tree analysis based on the 16S rRNA gene sequence revealed that strain SNL9<sup>T</sup> formed a lineage within the family *Flavobacteriaceae* of the phylum *Bacteroidetes*, it was most closely related to *Flavobacterium ummariense* DS-12<sup>T</sup> and *Flavobacterium viscosum* (96.2% and 95.3% similarity, respectively). The major isoprenoid quinone was MK-6. The major fatty acids were iso-C<sub>15:0</sub> (38.5%), summed feature 3 (15.3%) and summed feature 9 (20.4%). The polar lipid profile of strain SNL9<sup>T</sup> showed the presence of Phosphatidylethanolamine (PE) as major lipid with several other minor lipids. The DNA G + C content was 39.4 mol%. The values of average nucleotide identity and DNA-DNA hybridization were calculated as 76.9% and 20.4% between the novel strain SNL9<sup>T</sup> and *Flavobacterium ummariense* DS-12<sup>T</sup>. Thus the data accumulated in this study support the suggestion that strain SNL9<sup>T</sup> is considered to represent a novel species of the genus *Flavobacterium*, for which the name *Flavobacterium baculatum* sp. nov. is proposed.

[This work was supported by a grant from the National Institute of Biological Resources (NIBR), funded by the Ministry of Environment (MOE) of the Republic of Korea (NIBR202002203)].

**A021*****Lewinella aurantiaca* sp. Nov., Carotenoid Pigments Producing Bacterium Isolated from Surface Seawater**

Inhyup Kim, Geeta Chhetri, Jiyou Kim, Minchung Kang, and Taegun Seo\*

*Department of Life science, Dongguk University-Seoul*

A Gram-stain-negative, aerobic, flexible rod or filament-shaped, carotenoid-pigmented, non-motile bacterium, which was designated as SSH13<sup>T</sup> was isolated from surface seawater sample collected from Sehwa Beach, South Korea. The novel isolate required NaCl for growth and grew optimally at approximately 2% NaCl. Chemotaxonomic and morphological characteristics were consistent with members of the genus *Lewinella*. Furthermore, phylogenetic analysis conducted based on 16S rRNA gene sequence studies revealed that strain SSH13<sup>T</sup> was most closely related to the type strains of the genus *Lewinella*. Strain SSH13<sup>T</sup> showed 16S rRNA gene sequence similarities with *L. persica* DSM 23188<sup>T</sup> (95.3%), *L. agarilytica* KCTC 12774<sup>T</sup> (95.0%) and *L. cohaerens* KACC 14388<sup>T</sup> (89.7%). Menaquinone-7 was the predominant respiratory quinone. The average nucleotide identity values between strain SSH13<sup>T</sup> and selected taxa, *Lewinella persica* T-3<sup>T</sup>, *Lewinella agarilytica* SST-19<sup>T</sup> and *Lewinella cohaerens* KACC 14388<sup>T</sup> were 72.6%, 72.9% and 66.1%, respectively. The DNA G+C content in stain SSH13<sup>T</sup> was 52.8 mol%. Based on phylogenetic and phenotypic analyses, strain SSH13<sup>T</sup> is considered to be a novel species of the genus *Lewinella*, for which the name *Lewinella aurantiaca* sp. nov., has been proposed.

[This work was supported by a grant from the National Institute of Biological Resources (NIBR), funded by the Ministry of Environment (MOE) of the Republic of Korea (NIBR202002203).]

**A022*****Pontibacter terrigena* sp. nov., Isolated from Soil**

Geeta Chhetri, Jiyou Kim, Inhyup Kim, Minchung Kang, and Taegun Seo\*

*Department of Life Science, Dongguk University-Seoul*

A taxonomic study using a polyphasic approach was performed on a Gram-stain negative, red-pink, aerobic, non-motile, asporogenous, rod-shaped bacterium, designated strain JH31<sup>T</sup>, isolated from soil collected from Jeju-do, South Korea. The 16S rRNA gene sequence analysis showed that strain JH31<sup>T</sup> is phylogenetically related to *Pontibacter virosus* W14<sup>T</sup>, *Pontibacter amylolyticus* 9-2<sup>T</sup>, *Pontibacter ramchanderi* LP43<sup>T</sup> and *Pontibacter lucknowensis* DM9<sup>T</sup> (98.3, 97.2, 97.2 and 96.4% sequence similarity, respectively). The major fatty acids of strain JH31<sup>T</sup> were identified as iso-C<sub>15:0</sub>, iso-C<sub>15:0</sub> 3-OH and summed feature 4. The presence of the major menaquinone MK-7 supported the affiliation of the strain to the genus *Pontibacterium*. The polar lipid profile was found to consist of phosphatidylethanolamine. The genome of strain JH31<sup>T</sup> has a G+C content of 48.3 mol%. The *in silico* DNA-DNA hybridization and average nucleotide identity values between strain JH31<sup>T</sup> and the closely related strain *Pontibacter virosus* W14<sup>T</sup> were 38.5% and 72.3% respectively. Based on the genotypic, phenotypic and chemotaxonomic analyses, strain JH31<sup>T</sup> represents a new species belonging to the genus *Pontibacter*, for which the name *Pontibacter terrigena* sp. nov. is proposed. The type strain is JH31<sup>T</sup> (=KACC 21705<sup>T</sup> =NBRC 32880<sup>T</sup>).

[This work was supported by a grant from the National Institute of Biological Resources (NIBR), funded by the Ministry of Environment (MOE) of the Republic of Korea (NIBR202002203)].

**A023*****Reinekea thalattae* sp. nov., a New Species of the Genus *Reinekea* Isolated from Surface Seawater in Sehwa Beach**

Inhyup Kim, Geeta Chhetri, Jiyou Kim, Minchung Kang, and Taegun Seo\*

*Department of Life science, Dongguk University-Seoul*

A Gram-stain-negative, non-pigmented, curved rod-shaped, single polarly flagellated, facultatively anaerobic bacterium, designated as SSH23<sup>T</sup>, was isolated from surface seawater sample collected at the Sehwa Beach in South Korea. The novel isolate required NaCl for growth and grew optimally between 2–3% NaCl. Strain SSH23<sup>T</sup> showed high 16S rRNA gene sequence similarities with *R. marinisedimentorum* DSM 15388<sup>T</sup> (96.4%), *R. marina* KACC 17315<sup>T</sup> (96.2%), *R. blandensis* KACC 17315<sup>T</sup> (95.9%) and *R. aestuarii* KCTC 22813<sup>T</sup> (95.6%). The major polar lipids of strain SSH23<sup>T</sup> were phosphatidylethanolamine, diphosphatidylglycerol and phosphatidylglycerol. The predominant respiratory quinone was found to be ubiquinone-8. The average nucleotide identity values of strain SSH23<sup>T</sup> with *R. marinisedimentorum* DSM 15388<sup>T</sup> and *R. blandensis* MED297<sup>T</sup> were determined to be 72.2% and 69.8%, respectively. The G+C content of the genomic DNA was 45.5 mol%. Based on genotypic, phenotypic, chemotaxonomic, and phylogenetic analyses, strain SSH23<sup>T</sup> was considered to represent a novel member of the genus *Reinekea*, for which the name *Reinekea thalattae* sp. nov. is proposed. The type strain of *Reinekea thalattae* is SSH23<sup>T</sup>.

[This work was supported by a grant from the National Institute of Biological Resources (NIBR), funded by the Ministry of Environment (MOE) of the Republic of Korea (NIBR202002203).]

**A024*****Simulacricoccus soli* sp. nov., a Casein Hydrolyzing Bacterium Isolated from Soil**Geeta Chhetri<sup>1</sup>, Inhyup Kim<sup>1</sup>, Jiyouon Kim<sup>1</sup>, Minchung Kang<sup>1</sup>, Myung Kyum Kim<sup>2</sup>, and Taegun Seo<sup>1\*</sup><sup>1</sup>Department of Life Science, Dongguk University-Seoul, <sup>2</sup>Department of Bio & Environmental Technology, College of Natural Science, Seoul Women's University

A bacterial strain, designated 17bor-14<sup>T</sup>, was isolated from gamma ray-irradiated soil. Cells of this strain were Gram-stain negative, strictly aerobic, rod shape and catalase-positive and oxidase-negative. The major fatty acids of strain 17bor-14<sup>T</sup> were C<sub>16:1</sub> ω5c, iso-C<sub>15:0</sub> and iso-C<sub>14:0</sub> 3-OH. The polar lipid profile contained phosphatidylethanolamine, phosphatidylglycerol, four unidentified aminolipid and two unidentified lipid. The G+C content of the genomic DNA of 17bor-14<sup>T</sup> was 73.0 mol%. The 16S rRNA gene sequence analysis showed that strain 17bor-14<sup>T</sup> was phylogenetically most closely related to *Simulacricoccus ruber* MCy10636<sup>T</sup> (99.4% sequence similarity). DNA-DNA relatedness between strain 17bor-14<sup>T</sup> and its closest relative was below 70%. The results of genotypic and phenotypic data showed that strain 17bor-14<sup>T</sup> could be distinguished from its phylogenetically related species, and that this strain represented novel species within the genus *Simulacricoccus*, for which the name *Simulacricoccus soli* sp. nov. (type strain 17bor-14<sup>T</sup> =KCTC 52883<sup>T</sup> =NBRC 112881<sup>T</sup>) is proposed.

[This work was supported by a grant from Seoul Women's University (2018) and the National Institute of Biological Resources (NIBR), funded by the Ministry of Environment (MOE) of the Republic of Korea (NIBR202002203)].

**A026*****Pistacia*-associated flexivirus 1 Identified in the Mastic Tree Transcriptome**

Dongbin Park, Chul Jun Goh, and Yoonsoo Hahn\*

Department of Life Science, Chung-Ang University

A genome sequence of a novel virus *Pistacia*-associated flexivirus 1 (PAFV1) was identified from a mastic tree (*Pistacia lentiscus*) transcriptome dataset. PAFV1 encodes three proteins: a replicase (REP) with RNA-dependent RNA polymerase activity, a movement protein (MP), and a hypothetical protein (HP). The PAFV1 REP sequence showed high similarity to those of three known members of the mycovirus family Gammaflexiviridae: Entoleuca gammaflexivirus 1 (EnFV1), Entoleuca gammaflexivirus 2 (EnFV2), and Botrytis virus F (BVF). Sequence comparison and phylogenetic analysis showed that PAFV1, EnFV1, and the endogenous virus of the fungus *Monosporascus cannonballus* formed a distinct clade (apart from EnFV2 and BVF), and may be the founding members of a novel genus in the family Gammaflexiviridae. Interestingly, the PAFV1/EnFV1 MP sequences showed similarity to the tobamo-like mycovirus MP sequences, implying a genomic recombination between members of the family Gammaflexiviridae and tobamo-like mycoviruses. PAFV1 may be a mycovirus infected a fungus associated with the mastic tree sample, which is evidenced by the presence of fungal ribosomal RNA sequences in the mastic tree transcriptome. The PAFV1 genome sequence is a useful resource for studying the mycovirus genome evolution.

[This research was supported by grants from the National Research Foundation of Korea funded by the Government of Korea (grant Nos. NRF-2017R1A1B4005866 and NRF-2018R1A5A1025077)]

**A027****Trichosanthes Associated Rhabdovirus 1 Identified in the *Trichosanthes kirilowii* Transcriptome**

Chul Jun Goh, Dongbin Park, and Yoonsoo Hahn\*

*Department of Life Science, Chung-Ang University*

A novel RNA virus, Trichosanthes associated rhabdovirus 1 (TrARV1), was identified in a transcriptome dataset isolated from a root sample of *Trichosanthes kirilowii*. *T. kirilowii* is a flowering plant belonging to the family Cucurbitaceae, of which fruits, seeds, and root tubers have been used clinically in traditional Chinese medicine. The TrARV1 genome sequence has six open reading frames encoding five canonical structural proteins of the family Rhabdoviridae (N, nucleocapsid; P, phosphoprotein; M, matrix protein; G, glycoprotein; and L, polymerase), and an accessory protein. Sequence comparisons and phylogenetic analyses of L and N proteins indicated that TrARV1 is a novel member of the genus *Cytorhabdovirus* of the family Rhabdoviridae. TrARV1 is most closely related to Wuhan insect virus 5 and persimmon virus A. The putative *cis*-regulatory elements involved in transcription termination and polyadenylation, commonly found in the gene junction regions of rhabdoviruses, were identified in the TrARV1 genome with the consensus sequence 3'-ACUAAAUUUUUGAUCUUU-5'. The genome sequence of TrARV1 may be useful for studying the evolution and molecular biology of cytorhabdoviruses.

[This research was supported by grants from the National Research Foundation of Korea funded by the Government of Korea (grant Nos. NRF-2017R1A1B4005866 and NRF-2018R1A5A1025077)]

**A028*****Arachidicoccus soli* sp. nov., a Bacterium Isolated from Soil**

Tae-Wan Kim, Shin Ae Lee, Mee-Kyung Sang, Jaekyeong Song, Soon-Wo Kwon, and Hang-Yeon Weon\*

*Agricultural Microbiology Division, National Institute of Agricultural Sciences, Rural Development Administration*

A Gram-stain-negative, aerobic, non-motile and rod shaped bacterium, designated KIS59-12<sup>T</sup>, was isolated from the soil sample collected at Hodo island in Boryeong city, Republic of Korea. The strain grew at the temperature range of 10-33 °C, pH 6.0-7.5, and 0-4 % NaCl (w/v). The phylogenetic analysis based on 16S rRNA gene sequences showed that strain KIS59-12<sup>T</sup> was in the same clade with *Arachidicoccus rhizosphaerae* Vu-144<sup>T</sup> and *Arachidicoccus ginsenosidivorans* Gsoil809<sup>T</sup> with 97.5% and 97.2% sequence similarity, respectively. Comparative genome analysis between strain KIS59-12<sup>T</sup> and *A. rhizosphaerae* Vu-144<sup>T</sup> showed that average nucleotide identity (ANI) value was 69.4% and digital DNA-DNA hybridization value was 19.1 %. The major respiratory quinone was menaquinone-7 (MK-7). The major polar lipids were phosphatidylethanolamine and an unknown polar lipid. The predominant cellular fatty acids were iso-C<sub>15:0</sub> (42.4%), iso-C<sub>15:1</sub> G (16.0%), and iso-C<sub>17:0</sub> 3-OH (18.3%), which supported the affiliation of strain KIS59-12<sup>T</sup> with the genus *Arachidicoccus*. The genomic DNA G+C content was 36.4 %. On the basis of phylogenetic, physiological, and chemotaxonomic characteristics, strain KIS59-12<sup>T</sup> represents a novel species of the genus *Arachidicoccus*, for which the name *Arachidicoccus soli* sp. nov. is proposed. The type strain of *Arachidicoccus soli* is KIS59-12<sup>T</sup> (= KACC 17340<sup>T</sup> = NBRC 113161<sup>T</sup>).

**A029*****Paenibacillus lycopersici* sp. nov. and *Paenibacillus rhizovicinus* sp. nov., Isolated from the Rhizosphere of Tomato (*Solanum lycopersicum*)**

Shin Ae Lee Lee, Jun Heo, Mee-Kyung Sang, Jaekyeong Song, Soon-Wo Kwon, and Hang-Yeon Weon\*

*Agricultural Microbiology Division, National Institute of Agricultural Sciences, Rural Development Administration*

Two Gram-stain-positive, rod-shaped, endospore-forming bacteria, designated 12200R-189<sup>T</sup> and 14171R-81<sup>T</sup> were isolated from the rhizosphere of tomato plants. The 16S rRNA gene sequence similarity between strains 12200R-189<sup>T</sup> and 14171R-81<sup>T</sup> were 97.2%. Both strains showed the highest 16S rRNA gene sequence similarities to *Paenibacillus sacheonensis* SY01<sup>T</sup> (96.3% and 98.0%, respectively). Comparative genome analysis revealed that average nucleotide identity (ANI) and digital DNA-DNA hybridization (dDDH) values among 12200R-189<sup>T</sup>, 14171R-81<sup>T</sup>, and other closely related species were below the cut-off levels 95% and 70%, respectively. Strain 12200R-189<sup>T</sup> grew at a temperature range of 15-40°C, pH 6.0-9.0, and 0-3% NaCl (w/v), whereas strain 14171R-81<sup>T</sup> grew at a temperature range of 10-37°C, pH 6.0-8.0, and 0-1% NaCl (w/v). Menaquinone-7 (MK-7) was the only isoprenoid quinone detected in both strains. The predominant cellular fatty acids (>10%) were iso-C<sub>15:0</sub>, anteiso-C<sub>15:0</sub>, and iso-C<sub>16:0</sub>. Based on phylogenetic, genomic, phenotypic, and chemotaxonomic analyses, strains 12200R-189<sup>T</sup> and 14171R-81<sup>T</sup> represent two novel species of the genus *Paenibacillus*, for which the names *Paenibacillus lycopersici* sp. nov. and *Paenibacillus rhizovicinus* sp. nov. are proposed. The type strains are 12200R-189<sup>T</sup> (= KACC 19916<sup>T</sup>) and 14171R-81<sup>T</sup> (= KACC 19915<sup>T</sup>).

**A030*****Chitinophaga agri* sp. nov., a Bacterium Isolated from Soil of Reclaimed Land**

Hang-Yeon Weon, Shin Ae Lee, Jun Heo, Tae-Wan Kim, Mee-Kyung Sang, Soon-Wo Kwon, and Jaekyeong Song\*

*Agricultural Microbiology Division, National Institute of Agricultural Sciences, Rural Development Administration*

A Gram-negative, aerobic, and long rod-shaped bacterium, designated as H33E-04<sup>T</sup>, was isolated from the soil of reclaimed land, Republic of Korea. The strain grew at a temperature range of 15-40°C, pH 5.0-10.0, and 0-2% NaCl (w/v). The phylogenetic analysis based on 16S rRNA gene sequences showed that strain H33E-04<sup>T</sup> was in the same clade with *Chitinophaga pinensis* DSM 2588<sup>T</sup>, *Chitinophaga filiformis* IFO 15056<sup>T</sup>, and *Chitinophaga ginsengisoli* Gsoil 052<sup>T</sup> with 98.4%, 97.9%, and 97.8% sequence similarities, respectively. The de novo genome assembly revealed that the DNA G+C content of the strain was 46.2 mol%. Comparative genome analysis between strain H33E-04<sup>T</sup> and *C. pinensis* DSM 2588<sup>T</sup> showed that the average nucleotide identity (ANI) and digital DNA-DNA hybridization (dDDH) values were 79.9% and 23.4%, respectively. The major respiratory quinone was menaquinone-7 (MK-7) and the predominant cellular fatty acids were iso-C<sub>15:0</sub> (31.7%), C<sub>16:1</sub> ω5c (31.2%), and iso-C<sub>17:0</sub> 3-OH (11.8%), supporting the affiliation of strain H33E-04<sup>T</sup> with the genus *Chitinophaga*. Based on phylogenetic, physiological, and chemotaxonomic characteristics, strain H33E-04<sup>T</sup> represents a novel species of the genus *Chitinophaga*, for which the name *Chitinophaga agri* sp. nov. is proposed. The type strain of *Chitinophaga agri* is H33E-04<sup>T</sup> (= KACC 21303<sup>T</sup>).

**A031*****Lysobacter arenosi* sp. nov., Isolated from Gangwondo Yeongwol Soil**

Kyeong Ryeol Kim and Che Ok Jeon\*

*Department of Life Science, Chung-Ang University*

A Gram-stain-negative and mesophilic bacterium, designated strain R7<sup>T</sup>, was isolated from gangwondo yeongwol soil in South Korea. Cells of strain R7<sup>T</sup> was non-motile rods showing catalase and oxidase-positive activities. Growth of strain R7<sup>T</sup> was observed at 10–37°C (optimum, 30°C) and pH 6.0–10.0 (optimum, pH 7–8), and in the presence of 0–1.5% (w/v) NaCl (optimum, 0%). Strain R7<sup>T</sup> contained iso-C<sub>14:0</sub>, iso-C<sub>15:0</sub>, iso-C<sub>16:0</sub>, and summed feature 9 (comprising C<sub>16:0</sub> 10-methyl and/or C<sub>17:1</sub> iso ω9c) as major cellular fatty acids (>5%) and ubiquinone-8 as the sole isoprenoid quinone. Phosphatidylethanolamine, phosphatidylglycerol and an unidentified phospholipid were detected as major polar lipids. The G+C content of strain R7<sup>T</sup> calculated from the whole genome sequence was 67.1 mol%. Strain R7<sup>T</sup> was most closely related to *Lysobacter panacisoli* C8-1<sup>T</sup> with a 98.7% 16S rRNA sequence similarity and shared less than 98.5% 16S rRNA sequence similarities with other type strains. Phylogenetic analysis based on 16S rRNA gene sequences showed that strain R7<sup>T</sup> formed a phyletic lineage with *Lysobacter panacisoli* C8-1<sup>T</sup> within the genus *Lysobacter*. On the basis of phenotypic, chemotaxonomic and molecular analysis, strain R7<sup>T</sup> clearly represents a novel species of the genus *Lysobacter*, for which the name *Lysobacter arenosi* sp. nov. is proposed. The type strain is R7<sup>T</sup> (= KACC 21663<sup>T</sup>)

**A032*****Nocardiopsis pacificus* sp. nov., Isolated from the East Pacific Seawater**

Mirae Kim, Yeonjung Lim, and Jang-Cheon Cho\*

*Department of Biological Sciences, Inha University*

A Gram-stain-positive, streptococci-shaped, non-motile, pale yellow-pigmented, facultatively anaerobic bacterium, designated *Nocardiopsis* sp. CNT189<sup>T</sup>, was isolated from the East pacific seawater close to the San Diego coastal area. Cellular growth occurred at 30–37°C, pH 7.0–8.0, and with 2–6% (w/v) NaCl. Phylogenetic analysis based on the 16S rRNA gene sequences revealed that strain CNT189<sup>T</sup> belonged to the genus *Nocardiopsis* and shared 98.8–99.6% sequence similarities with other *Nocardiopsis* species. Whole genome sequencing of strain CNT189<sup>T</sup> revealed genome size of 7.16 Mbp and DNA G+C content of 74.1 mol%. The CNT189<sup>T</sup> genome shared the average nucleotide identity (ANI) of 92.8% with *Nocardiopsis compsta* KS9<sup>T</sup> and 95.06% with *Nocardiopsis potens* DSM 45234<sup>T</sup>. The major respiratory quinone was menaquinone-9 (MK-9) and predominant cellular fatty acids were C<sub>18:1</sub> ω9c (25.0%), C<sub>16:0</sub> (13.5%), and anteiso-C<sub>15:0</sub> (12.7%). Major polar lipids were diphosphatidylglycerol, phosphatidylglycerol, phosphatidylmonomethylethanolamine. Based on phylogenetic distinction and differential phenotypic characteristics, strain CNT189<sup>T</sup> is considered to be assigned to the genus *Nocardiopsis* as the type strain of a new species, for which the name *Nocardiopsis pacificus* sp. nov. is proposed.

[This study was supported by a grant from the Collaborative Genome Program of the KIMST funded by the Ministry of Oceans and Fisheries (No. 20180430).]

**A033*****Halomonas locisalis* sp. nov., a Moderately Halophilic Bacterium Isolated from a Salt Lake**Ruan Wenting<sup>1</sup>, Shehzad Abid Khan<sup>1</sup>, Mohammad Saeid Hejazi<sup>2</sup>, and Che Ok Jeon<sup>1\*</sup><sup>1</sup>Department of Life Science, Chung-Ang University, <sup>2</sup>Molecular Medicine Research Center, Biomedicine Institute, Tabriz University of Medical Sciences

In the course of screening halophilic bacteria in Urmia Lake in Iran, which is being threatened by dryness, a novel Gram-negative, moderately halophilic, heterotrophic and short rod-shaped bacterium, designated as TBZ9<sup>T</sup>, was isolated and characterized. Colonies were found to be creamy yellow, with catalase- and oxidase-positive activities. The growth of strain TBZ9<sup>T</sup> was observed to be at 15–37°C (optimum, 30°C), at pH 6.0–10.0 (optimum, pH 7.0) and in the presence of 1.0–17% (w/v) NaCl (optimum, 3%). Ubiquinone-9 was detected as the only respiratory isoprenoid quinone. Phosphatidylglycerol, phosphatidylethanolamine, unidentified phospholipids, and unidentified polar lipids were detected as the major polar lipids. Strain TBZ9<sup>T</sup> was found to be most closely related to *Halomonas arcis* AJ282<sup>T</sup>, *Halomonas songnenensis* NEAU-ST10-39<sup>T</sup> and *Halomonas lutescens* Q1U<sup>T</sup> with the 16S rRNA gene sequence similarities of 98.43%, 98.01%, and 97.77%, respectively, but strain TBZ9<sup>T</sup> formed a distinct phylogenetic lineage from the closely related strains within the genus *Halomonas*. Based on phenotypic, chemotaxonomic and molecular properties, strain TBZ9<sup>T</sup> represents a novel species of the *Halomonas* genus, for which the name *Halomonas locisalis* sp. nov. is proposed. The type strain is TBZ9<sup>T</sup> (= KACC 21783).

**A034****Description of a Novel Propionogenic Gut Bacterium AP1 and Analysis of Propionate-producing Pathway**Ji-Sun Kim<sup>1</sup>, Min Kuk Suh<sup>1</sup>, Kook-Il Han<sup>1</sup>, Mi Kyung Eom<sup>1</sup>, Keun Chul Lee<sup>1</sup>, Han-Sol Kim<sup>1</sup>, Ju Huck Lee<sup>1</sup>, Seung-Hwan Park<sup>1</sup>, Se Won Kang<sup>1</sup>, Jam-Eon Park<sup>1</sup>, Byeong Seob Oh<sup>1</sup>, Seung Yeob Yu<sup>1</sup>, Seung-Hyeon Choi<sup>1</sup>, Dong-Ho Lee<sup>2</sup>, Hyuk Yoon<sup>2</sup>, Byungyong Kim<sup>3</sup>, Je Hee Lee<sup>3</sup>, and Jung-Sook Lee<sup>1,4\*</sup><sup>1</sup>Korean Collection for Type Cultures, Korea Research Institute of Bioscience and Biotechnology, <sup>2</sup>Seoul National University Bundang Hospital, <sup>3</sup>ChunLab, Inc., <sup>4</sup>University of Science and Technology

A novel Gram-stain-positive, non-motile, non-spore-forming, rod-shaped and strictly anaerobic bacterium, designated AP1<sup>T</sup>, was isolated from a healthy Korean feces. Comparative analysis of 16S rRNA gene sequences showed that strain AP1<sup>T</sup> was most closely related to *Merdimonas faecis* BR31<sup>T</sup> (94.3 %) in the family *Lachnospiraceae*. The average nucleotide identity (ANI) value between strain AP1<sup>T</sup> and related species, *M. faecis* BR31<sup>T</sup>, was also 75.6 %, indicating that strain AP1<sup>T</sup> was a novel genus within the family *Lachnospiraceae*. Strain AP1<sup>T</sup> utilized a wide variety of carbohydrates including glucose, galactose, glycerol, D-cellobiose, D-melezitose and D-sorbitol. By analyzing metabolites produced by strain AP1<sup>T</sup>, it was determined that strain AP1<sup>T</sup> formed a high concentration of propionate. Genomic analysis was also supported that this novel strain produced a large amount of propionate through the amino acid catabolic pathway, not through succinate-and/or propanediol-pathways, which are adopted by most gut bacteria. The level of propionate production was increased by adding L-threonine and L-methionine in media. Taken together, strain AP1<sup>T</sup> represents a novel species in a novel genus within the family *Lachnospiraceae* and produces propionate using amino acids not using carbohydrates. It is also expected that propionogenic bacterium AP1<sup>T</sup> can be used as new generation probiotics to target various metabolic syndrome as propionate-producing consortium does. [This research was supported by a Bio & Medical Technology Development program of the National Research Foundation of Korea (NRF) funded by the Ministry of Science and ICT (MSIT) of the Republic of Korea (NRF-2016M3A9F3946674), and a grant from the Korea Research Institute of Bioscience & Biotechnology (KRIBB) Research initiative program].

**A035****A Novel Bacterium of the Family Comamonadaceae, Isolated from Forest Soil**

Sang Hee Han, Ja Yeon Yu, Mi Sun Kim, Bo Ra Shin, Ji Won Lee, and Chi Nam Seong\*

*Department of Biology, College of Life Science and Natural Resources, Suncheon National University*

A Gram-stain-negative, aerobic, motile and white pigmented bacterium, designated strain JDB68<sup>T</sup> was isolated from soil collected from Mt. Jeokdaebong in Goheung, Republic of Korea. Phylogenetic analysis based on 16S rRNA gene sequences revealed that strain JDB68<sup>T</sup> formed a distinct lineage within the genus *Paucibacter* and was most closely related to *Paucibacter aquatile* CR182<sup>T</sup> (98.27% 16S rRNA gene sequence similarity), *Paucibacter oligotrophus* CHU3<sup>T</sup> (98.13%) and *Pelomonas saccharophila* DSM 654<sup>T</sup> (98.06%). Strain JDB68<sup>T</sup> grow at 10-30°C (optimally at 25°C), 6-10 pH (optimally at 7 pH), and 0-0.5% of NaCl (optimally at 0% NaCl). Cells grew on R2A, NA but not on MA, TSA, LB agar. Tyrosine (0.5%) and tween20 were hydrolysed. Catalase-negative and oxidase-positive. The DNA G+C content of the strain was 65.5 mol%. The major fatty acids of JDB68<sup>T</sup> were summed feature 3 (C<sub>16:1</sub> ω7c and/or C<sub>16:1</sub> ω6c) and C<sub>16:0</sub>. The major polar lipid was phosphatidylethanolamine. Ubiquinone 8 (Q-8) was the predominant respiratory quinone. Strain JDB68<sup>T</sup> was proposed as a new species of the genus *Paucibacter*, for which the name *Paucibacter soli* sp. nov. is proposed.

[This work was supported by a grant from the National Institute of Biological Resources (NIBR), funded by the Ministry of Environment (MOE) of the Republic of Korea (NIBR202002203).]

**A036****Characterization Extended-spectrum Cephalosporin (ESC) Resistance in *E. coli* and *Salmonella* on a Broiler Chicken**

Bai Wei, Bo-Ram Kwon, Ke Shang, Jun-Feng Zhang, Se-Yeoun Cha, Hyung-Kwan Jang, and Min Kang\*

*Department of Veterinary Infectious Diseases and Avian Diseases, College of Veterinary Medicine and Center for Poultry Diseases Control, Jeonbuk National University*

To investigate the extended-spectrum cephalosporin (ESC) resistance determinants in *Salmonella* and *E. coli* obtained from the same chicken. From a national study, 16 (2.9%) isolates of *Salmonella* from 8 farms and 42 (7.6%) isolates of *E. coli* from 15 farms showed reduced susceptibility to ceftazidime, respectively. Fifteen (93.8%) isolates of *Salmonella* were CTX-M-15 positive, whereas one CMY-2 positive. Meanwhile, 29 (69.0%) isolates of *E. coli* carried CTX-M-55, 3 CTX-M-15, one CTX-M-27 and 9 (21.4%) CMY-2. Whole genome sequence for the transferable plasmid showed genomic diversity of the plasmid from *Salmonella* and *E. coli* source from same chicken. The plasmid from *Salmonella* showed high homology with plasmid from canine *E. coli* (UK), while the plasmid from *E. coli* showed high homology with turkey's *Salmonella* (USA). Furthermore, transfer of the CMY-2 gene was by a common transposon of ISEcp1 downstream with CMY-2/blc/SugE. In conclusion, this important information should contribute to a greater understanding of the complex evolution and growing problem of ESC resistance in food-producing animals.

[This work was supported by Korea Institute of Planning and Evaluation for Technology in Food, Agriculture and Forestry (IPET) through Animal Disease Management Technology Development Program, funded by Ministry of Agriculture, Food and Rural Affairs (MAFRA)(119059-2, 716002-7)].

**A037**

**Two Novel Bacteria Isolated from Korean Indigenous Vertebrate**

So-Yeon Lee, Woorim Kang, and Jin-Woo Bae\*

*Department of Biology and Department of Life and Nanopharmaceutical Sciences, Kyung Hee University*

Two putative novel species, strain S11R28<sup>T</sup> and H21T32<sup>T</sup>, were isolated from fecal and gut samples of oriental stork (*Ciconia boyciana*) and Korean shiner (*Coreoleuciscus splendidus*).

Strain S11R28<sup>T</sup> was Gram-stain-negative bacterium, which possessed ten polar lipids, ubiquinone Q-8, and C16:0 as main cellular fatty acids. Strain H21T32<sup>T</sup> was Gram-stain-positive cocci occurring in pairs. Three sugar components and five amino acids of the cell wall, eight polar lipids, main cellular fatty acids were detected. Strain S11R28<sup>T</sup> and H21T32<sup>T</sup> formed a monophyletic clade with *Undibacterium parvum* DSM 23061<sup>T</sup> and *Jeotgalibaca arthritidis* CECT 9157<sup>T</sup>, respectively. ANI values of two strains with the closest relative species were 78.66% and 77.25%, respectively. Based on these various properties of two strains, strain S11R28<sup>T</sup> and H21T32<sup>T</sup> are considered to represent a novel species, for which the name *Undibacterium piscinae* sp. nov. and *Jeotgalibaca ciconiae* sp. nov. is proposed.

[This work was supported by Ministry of Food and Drug Safety (20172MFDS195), Republic of Korea.]

**A038**

**Detection and Molecular Characterization of Avian Coronavirus in Wild Bird Population**

Jong Yeol Park, Jun-Feng Zhang, Yea Jin Lee, Min Kang, Se-Yeoun Cha, and Hyung-Kwan Jang\*

*Department of Veterinary Infectious Diseases and Avian Diseases, College of Veterinary Medicine and Center for Poultry Diseases Control, Jeonbuk National University*

Coronaviruses (CoVs) are continuously circulating in mammals and birds that pose a threat to humans and livestock, which are classified into four genera (Alpha, Beta, Gamma, and Deltacoronavirus). Wild bird species serve as a natural reservoir of many emerging zoonotic pathogens. The main purpose of the present study is to monitor the prevalence and molecular characterization of avian coronavirus in wild birds in Korea. A total of 2,070 fecal samples were collected from wild birds in 2019, and ten samples obtained from same area were pooled into one test sample. Samples (n=207) were tested for the presence of coronaviruses by using nested reverse transcription-PCR (RT-PCR). First, all samples were submitted to RT-PCR and nested PCR. Next, only the positive nested-PCR samples were propagated in specific-pathogen-free (SPF) embryonated chicken eggs for virus isolation. Samples were then sequenced and analyzed using a molecular phylogeny approach. 117 samples out of 207 (56.5%) were positive. Our data showed that positive rate was significantly higher than previous studies. Phylogenetic analysis of the RNA-dependent RNA polymerase (RdRp) gene resulted in Gammacoronavirus.

[This work was supported by Korea Institute of Planning and Evaluation for Technology in Food, Agriculture and Forestry (IPET) through Animal Disease Management Technology Development Program, funded by Ministry of Agriculture, Food and Rural Affairs (MAFRA)(716002-7).]

**A039****Diversity and Bioprospecting of Cold Adapted Arctic Fungi**

Yu Ri Choe<sup>1</sup>, Gyeong Deok An<sup>2</sup>, Gi Mo Park<sup>2</sup>, Hyewon Kim<sup>2</sup>, So yun Lee<sup>2</sup>, Yeon jung Lee<sup>2</sup>, Hyuncheol Oh<sup>3</sup>, Joung Han Im<sup>4</sup>, and Jae Hak Sohn<sup>2,5\*</sup>

<sup>1</sup>Seafood Research Center, Silla University, <sup>2</sup>Major in Food Biotechnology, Division of Bioindustry, College of Medical and Life Sciences, Silla University, <sup>3</sup>Institute of Pharmaceutical Research and Development, College of Pharmacy, Wonkwang University, <sup>4</sup>Korea Polar Research Institute, KORDI, <sup>5</sup>Research Center for Extremophiles and Marine Microbiology, Silla University

We surveyed the diversity of Arctic fungi isolated from sample collected in the region of Norway and their capability to produce bioactive compounds. Sample diluted by 10 fold dilution method after homogenization, were inoculated on nutrient agar (NA), 0.1% NA, R2A agar, PDA (Potato Dextrose Agar) and YMA (Yeast Malt Agar) plate and then incubated at 10°C for 20 days. 88 fungal colonies isolated and preserved in 15% (v/v) glycerol solution. Fungal isolates were tested for their ability to grow at temperatures (10 and 25°C). As a result, the 9 fungi strain were psychrophilic form, and 37 fungi strains were psychrotolerant form. Fungal extract were cultured using PDA at 15°C for 10-15 days and extracted with ethyl acetate. From the PTP1B inhibitory assay using fungal extracts, 49 extracts displayed strong inhibitory activity. From the results of identification using internal transcribed spacer (ITS) sequences, the fungal strains investigated in this study consisted of 55 taxa in 15 genera [*Alternaria* (1), *Aspergillus* (2), *Camarosporula* (1), *Chaetomium* (1), *Chrysosporium* (9), *Coniochaeta* (2), *Geltingia* (1), *Lachnellula* (1), *Leptosphaeria* (1), *Mucor* (6), *Penicillium* (21), *Pseudogymnoascus* (6), *Rhizoctonia* (1), *Tolypocladium* (1), *Torulasporea* (1)]. These results suggest that marine fungi isolated from the sediment of region of Norway might be a valuable resource for the screening of bioactive compound.

**A040****The Effect of Cold Temperature on the Growth and Diversity of Arctic Bacteria**

Yu Ri Choe<sup>1</sup>, Eun Gyoung Kim<sup>2</sup>, A-Yeong Park<sup>2</sup>, Ji Hyeon Lee<sup>2</sup>, Rye Gyeong Park<sup>2</sup>, Ji sun Kim<sup>2</sup>, Jin Sol Park<sup>2</sup>, Hyuncheol Oh<sup>3</sup>, Joung Han Im<sup>4</sup>, and Jae Hak Sohn<sup>2,5\*</sup>

<sup>1</sup>Seafood Research Center, Silla University, <sup>2</sup>Major in Food Biotechnology, Division of Bioindustry, College of Medical and Life Sciences, Silla University, <sup>3</sup>Institute of Pharmaceutical Research and Development, College of Pharmacy, Wonkwang University, <sup>4</sup>Korea Polar Research Institute, KORDI, <sup>5</sup>Research Center for Extremophiles and Marine Microbiology, Silla University

We survey the cold adapted characteristic and the diversity of a culturable bacteria isolated from the region of Norway. Sample diluted by 10 fold dilution method after homogenization, were inoculated on nutrient agar (NA), 0.1% NA, R2A agar, PDA (Potato Dextrose Agar) and YMA (Yeast Malt Agar) plate and then incubated at 10°C for 20 days. The 130 bacterial strain were isolated by morphological characteristics of grown colonies. For the growth of isolated bacteria in different temperature, 130 bacterial strain were inoculated in NA and then cultured at 10°C and 25°C for 20 days. As a result, the 1 bacterial strain were psychrophiles, and 113 bacterial strains were psychrotolerant bacteria, and 13 bacterial strains were mesophile. From the phylogentic analysis based on 16S rRNA gene sequence, bacteria consisted of 27 taxa in 9 genera [*Bacillus* (1), *Burkholderia* (1), *Caballeronia* (1), *Corynebacterium* (1), *Frondehabitans* (2), *Pseudomonas* (15), *Rahnella* (2), *Rathayibacter* (1), *Staphylococcus* (3)].

**A041*****Flaviflexus ciconiae* sp. nov., Isolated from the Feces of the Oriental Stork**

Jae-Yun Lee, Woorim Kang, Pil Soo Kim, So-Yeon Lee, Hojung Sung, June-Young Lee, and Jin-Woo Bae\*

*Department of Life and Nanopharmaceutical Sciences and Department of Biology, Kyung Hee University*

A novel Gram-stain-positive, non-spore-forming, non-motile, coccobacillus-shaped, strictly aerobic bacterium, designated strain H23T48<sup>T</sup>, was isolated from the feces of oriental storks collected from the Seoul Grand Park Zoo, Republic of Korea. Optimal growth of the strain was observed at 37°C, pH 8, and with 3% (w/v) NaCl. 16S rRNA gene sequence comparison revealed that strain H23T48<sup>T</sup> was closely related to *Flaviflexus salsibiostraticola* EBR4-1-2<sup>T</sup> (97.0% sequence similarity) and *Flaviflexus huanghaiensis* H5<sup>T</sup> (96.7 % sequence similarity). Strain H23T48<sup>T</sup> contains MK-9(H<sub>4</sub>) as the major menaquinone and C<sub>16:0</sub> (42.4%), C<sub>18:1</sub> ω<sub>9c</sub> (31.3%), and C<sub>14:0</sub> (17.7%) as the major cellular fatty acids. The polar lipids included phosphatidylglycerol, two unidentified lipids, six unidentified phospholipids, and two unidentified glycerophospholipids and the amino acid composition of the cell wall peptidoglycan was glycine, L-alanine, L-lysine, D-glutamic acid, and L-aspartic acid. The genomic G+C content of the strain is 59.5 mol%, and the average nucleotide identity (ANI) value between strain H23T48<sup>T</sup> and *F. salsibiostraticola* KCT C33148<sup>T</sup> (=EBR4-1-2<sup>T</sup>) is 75.5%. [This study was supported by a grant from the Mid-Career Researcher Program (NRF-2020R1A2C3012797) through the National Research Foundation of Korea.]

**A042*****Pseudorhodobacter turbinis* sp. nov., Isolated from the Gut of the Korean Turban Shell, *Turbo cornutus***

Yun-Seok Jeong, June-Young Lee, Hojun Sung, Jeong-Eun Han, Euon Jung Tak, So-Yeon Lee, Jae-Yun Lee, Pil Soo Kim, Dong-Wook Hyun, Mi-Ja Jung, and Jin-Woo Bae\*

*Department of Biology and Department of Life and Nanopharmaceutical Sciences, Kyung Hee University*

A novel Gram-stain-negative, coccus-shaped, aerobic and motile bacterial strain designated S12M18<sup>T</sup> was isolated from the gut of the Korean turban shell, *Turbo cornutus*. The phylogenetic analysis based on 16S rRNA gene sequence showed that strain S12M18<sup>T</sup> belong to the genus *Pseudorhodobacter* and showed highest 16S rRNA gene sequence similarity with *Pseudorhodobacter aquimaris* HDW-19<sup>T</sup> (98.63 %). The OrthoANI value between strain S12M18<sup>T</sup> and *P. aquimaris* HDW-19<sup>T</sup> was 86.96%. The major cellular fatty acids of strain S12M18<sup>T</sup> was summed feature 8 (C<sub>18:1</sub> ω<sub>7c</sub> or C<sub>18:1</sub> ω<sub>6c</sub>). The major components of polar lipids were phosphatidylcholine, phosphatidylglycerol and phosphatidylethanolamine. The predominant isoprenoid quinone was Q-10. The DNA G + C content was 57.8 mol%. The polyphasic analyses indicated that strain S12M18<sup>T</sup> represents a novel species of the genus *Pseudorhodobacter*, for which the name *Pseudorhodobacter turbinis* sp. nov. is proposed. The type strain is S12M18<sup>T</sup> (=KCTC 62742<sup>T</sup> =JCM 33168<sup>T</sup>).

[This work was supported by a grant from the Mid-career Researcher Program (NRF-2016R1E1A1A02921587), Bio & Medical Technology Development Program (NRF-2017M3A9F3046549), SRC (NRF-2018R1A5A1025077) through the National Research Foundation (NRF) funded by the Ministry of Science, ICT and Future Planning (MSIT); the National Institute of Biological Resources (NIBR201801106), funded by the Ministry of Environment of Korea.]

**A043*****Tumebacillus avium* sp. nov., Isolated from the Gut of a Cinereous Vulture, *Aegypius monachus***

Hojun Sung<sup>1</sup>, Hyun Sik Kim<sup>1</sup>, June-Young Lee<sup>1</sup>, Woorim Kang<sup>1</sup>, Pil Soo Kim<sup>1</sup>, Dong-Wook Hyun<sup>1</sup>, Euon Jung Tak<sup>1</sup>, Mi-Ja Jung<sup>1</sup>, Jeong Rae Rho<sup>2</sup>, Sun Duk Park<sup>2</sup>, Hyung Eun Shim<sup>2</sup>, and Jin-Woo Bae<sup>1\*</sup>

<sup>1</sup>Department of Life and Nanopharmaceutical Sciences and Department of Biology, Kyung Hee University, <sup>2</sup>Seoul Grand Park

A Gram-stain-positive, facultatively aerobic, spore-forming and rod-shaped bacterial strain, AR23208<sup>T</sup>, was isolated from the gut of a cinereous vulture (*Aegypius monachus*), collected at the Seoul Grand Park Zoo, Republic of Korea. Strain AR23208<sup>T</sup> grew optimally at 25–30°C, at pH 7, in the absence of NaCl. The phylogenetic analysis revealed that the 16S rRNA gene of strain AR23208<sup>T</sup> shared 98.2% and 97.1% sequence similarity with the corresponding sequences of *Tumebacillus algifaecis* THMBR28<sup>T</sup> and *T. lipolyticus* NIO-S10<sup>T</sup>, respectively. The predominant fatty acids (>10%) of strain AR23208<sup>T</sup> were iso-C<sub>15:0</sub> (46.5%), summed feature 4 (anteiso-C<sub>17:1</sub> B and/or iso-C<sub>17:1</sub> I, 11.7%) and anteiso-C<sub>15:0</sub> (11.1%). The primary isoprenoid quinone was menaquinone-7. The polar lipids were phosphatidylethanolamine, phosphatidylmonomethylethanolamine, phosphatidylglycerol, six unidentified phospholipids, an unidentified amino phospholipid and ten unidentified lipids. The OrthoANI value based on the complete genome sequence of strain AR23208<sup>T</sup> and the closest related strain, *T. algifaecis*, was 80.4%. The genomic DNA G+C content of strain AR23208<sup>T</sup> was 56.0 mol%. Based on the current study, strain AR23208<sup>T</sup> is proposed to be a novel species candidate of the genus *Tumebacillus*, with the strain name *Tumebacillus avium* sp. nov. and strain AR23208<sup>T</sup> (=KCTC 33929<sup>T</sup> =JCM 32188<sup>T</sup>) as the type strain.

[This study is supported by Ministry of Food and Drug Safety (20172MFDS195), Republic of Korea.]

**A044*****Chitinibacter bivalvium* sp. nov., a Bacterium Isolated from Gut of Freshwater Mussel *Anodonta arcuiformis***

Jee-Won Choi, Jae-Yun Lee, Dong-Wook Hyun, Pil Soo Kim, Yun-Seok Jeong, June-Young Lee, Jeong Eun Han, So-Yeon Lee, Hojun Sung, Euon Jung Tak, Hyun Sik Kim, Mi-Ja Jung, and Jin-Woo Bae\*

Department of Biology and Department of Life & Nanopharmaceutical Sciences, Kyung Hee University

A novel Gram-negative, aerobic, rod-shaped bacterium, designated strain 2T18<sup>T</sup>, was isolated from the gut of freshwater mussel, *Anodonta arcuiformis*, collected from Seosan-si, Chungcheongnam-do, Republic of Korea. Phylogenetic analyses based on 16S rRNA gene sequences showed that the strain belonged to the genus *Chitinibacter*. Strain 2T18<sup>T</sup> formed a monophyletic clade with *Chitinibacter suncheonensis* SK16<sup>T</sup>, *Chitinibacter fontanus* STM-7<sup>T</sup>, *Chitinibacter tainanensis* BCRC 17254<sup>T</sup>, *Chitinibacter alvei* TNR-14<sup>T</sup>, with sequence similarities of 98.74%, 98.52%, 98.35% and 95.8%, respectively. Strain 2T18<sup>T</sup> grew optimally at 30°C, pH 8, with 0.5% (w/v) NaCl. The predominant isoprenoid quinone is ubiquinone-8 (Q-8) and major fatty acids are summed feature 3 (C<sub>16:1</sub> ω6c and/or C<sub>16:1</sub> ω7c) and C<sub>16:0</sub>. The G+C content of the genomic DNA is 50.58 mol%. The average nucleotide identity (ANI) value between strains 2T18<sup>T</sup> and *C. fontanus* STM-7<sup>T</sup> (=KCTC 42982<sup>T</sup>) is 77.84%. Based on phenotypic, genotypic, phylogenetic, and chemotaxonomic characteristics, strain 2T18<sup>T</sup> represents a novel species of the genus *Chitinibacter* for which the name *Chitinibacter bivalvium* sp. nov. is proposed. The type strain is 2T18<sup>T</sup> (=KCTC 72821<sup>T</sup>).

[This work was supported by Korea Institute of Planning and Evaluation for Technology in Food, Agriculture and Forestry (IPET) through Agricultural Microbiome R&D Program, funded by Ministry of Agriculture, Food and Rural Affairs (MAFRA) (918011-04-1-SB010).]

**A045*****Paenibacillus gonggeomjia* sp. nov., Isolated from Sediment of the Reservoir Gonggeomji**Ji-Hye Han<sup>1</sup>, Seoni Hwang<sup>1</sup>, Jaesoo Lim<sup>2</sup>, Jing-Young Lee<sup>2</sup>, and Sang Deuk Lee<sup>1\*</sup><sup>1</sup>Nakdonggang National Institute of Biological Resources, <sup>2</sup>Korea Institute of Geoscience and Mineral Resources

Gram-stain positive, motile, ivory colored and designated strain 19GGS1-52<sup>T</sup> was isolated from sediment (depth 90 cm) of the reservoir Gonggeomji, Sangju-si, Republic of Korea. Reservoir Gonggeomji is a historical irrigation facility and designated as a wetland protected area by Ministry of Environment. Phylogenetic analysis based on 16S rRNA gene sequences revealed that the strain 19GGS1-52<sup>T</sup> belongs to the genus *Paenibacillus* of the family *Paenibacillaceae*. Strain 19GGS1-52<sup>T</sup> was closely related to *Paenibacillus donghaensis* KCTC 13049<sup>T</sup> (97.30%), *Paenibacillus wynnii* DSM 18334<sup>T</sup> (97.03%) and showed less than 97.0% sequence similarity to other members of the genus *Paenibacillus*. The average nucleotide identity values of strain 19GGS1-52<sup>T</sup> with *P. donghaensis* KCTC 13049<sup>T</sup> and *P. wynnii* DSM 18334<sup>T</sup> were 75.52% and 75.56%, respectively. *In silico* DNA-DNA hybridization values between *P. donghaensis* KCTC 13049<sup>T</sup> and *P. wynnii* DSM 18334<sup>T</sup> were 20.9% and 21.6%, respectively. The novel strain grew over a temperature range of 10-37°C, at pH values of 6.0-8.0 and in the presence of 0-1.5% (w/v) NaCl. The major cellular fatty acids were anteiso-C<sub>15:0</sub> and C<sub>16:0</sub>. The genomic DNA G+C content was 45.6 mol%. On the basis of the phylogenetic inference, phenotypic and chemotaxonomic data, strain 19GGS1-52<sup>T</sup> should be classified as a novel species of the genus *Paenibacillus*, for which the name *Paenibacillus gonggeomjia* sp. nov. is proposed.

**A046****Phenotypic and Genetic Properties of *Brachybacterium vulturis* sp. nov. and *Brachybacterium avium* sp. nov., Isolated from Feces of an Andean Condor**Euon Jung Tak and Jin-Woo Bae<sup>\*</sup>

Kyung Hee University

Two strains VM2412<sup>T</sup> and VR2415<sup>T</sup> were isolated from feces of an Andean condor (*Vultur gryphus*) in Seoul Grand Park, Gyeonggi-do, South Korea. They shared 99.3% 16S rRNA gene sequence similarities with each other, but they were identified as two distinct species according to 89.0-89.2% ANI<sub>b</sub>, 90.3% ANI<sub>m</sub>, 89.7% OrthoANI and 38.0% dDDH values using whole genome sequences. Among species with validly published names, *Brachybacterium ginsengisoli* DCY80<sup>T</sup> shared the highest 16S rRNA gene sequence similarities with strains VM2412<sup>T</sup> (98.7%) and VR2415<sup>T</sup> (98.4%) and showed the closest genomic relatedness with strains VM2412<sup>T</sup> (83.3-83.5% ANI<sub>b</sub>, 87.0% ANI<sub>m</sub>, 84.3% OrthoANI and 27.8% dDDH) and VR2415<sup>T</sup> (82.8-83.2% ANI<sub>b</sub>, 86.7% ANI<sub>m</sub>, 83.9% OrthoANI and 27.2% dDDH). The genomic G+C contents of strains VM2412<sup>T</sup> and VR2415<sup>T</sup> were 70.8 and 70.4 mol%, respectively. Genome-based phylogenetic tree constructed by up-to-date bacterial core gene set (UBCG) showed that members of the genus *Brachybacterium* formed a clade with the isolated strains, supporting taxonomic classification of the strains into the genus *Brachybacterium*. Based on phenotypic and genotypic analyses in this study, strains VM2412<sup>T</sup> and VR2415<sup>T</sup> are considered to represent two novel species of the genus *Brachybacterium*.

[This work was supported by a grant from the Mid-Career Researcher Program (NRF-2020R1A2C3012797) through the National Research Foundation of Korea (NRF).]

**A048****Isolation of Beneficial Bacteria from Infant Feces**Seong-Hee Kim<sup>1</sup>, Yun Kyung Lee<sup>2</sup>, Dong-Woo Lee<sup>3</sup>, Myung Hee Nam<sup>4</sup>, Soo-Jong Hong<sup>5</sup>, and Bong-Soo Kim<sup>1\*</sup>

<sup>1</sup>Department of Life Science, Multidisciplinary Genome Institute, Hallym University, <sup>2</sup>Soonchunhyang Institute of Medi-bio Science, Soonchunhyang University, <sup>3</sup>Department of Biotechnology, Yonsei University, <sup>4</sup>Seoul Center, Korea Basic Science Institute, <sup>5</sup>Department of Pediatrics, Childhood Asthma Atopy Center, Environmental Health Center, University of Ulsan College of Medicine

The trillions of microorganisms live in the human intestinal tract, however most of them are still uncultured yet. Isolation of bacteria is important for understanding physiological and functional characteristics and for applying in various fields. In this study, we isolated and obtained bacteria strains from healthy infants comparing to infants with atopic dermatitis. We analyzed the difference of gut microbiome between healthy infants and infants with atopic dermatitis for isolating different bacteria between two groups. We collected fecal samples from 18 healthy infants. Isolation was performed under anaerobic condition using seven different media [TSA, BHI (+Yeast extract + Hemin), TSA (+5% Sheep blood), TSA (+5% Sheep blood + L-glutamine), Columbia blood agar, KCTC MEDIA No.978, Reinforced Clostridial Medium]. We obtained 369 strains and classified them based on 16S rRNA gene sequencing and phylogenetic analyses. Isolated 369 strains were classified into 39 genera and 67 species. We conducted immune assay using mouse immune cell for selecting beneficial strains. The effect of each strain on immune cell was analyzed using splenic cells. Isolated beneficial bacteria will be applied to develop pharmabiotic candidates for atopic dermatitis and immunologic diseases.

[This research was supported by the Bio & Medical Technology Development Program of the National Research Foundation (NRF) funded by the Korean government (MSIT) (No. NRF-2017M3A9F3043837)]

**A050****A Novel Species Isolated from the Gut of Stork**

Jeong Eun Han, Woorim Kang, and Jin-Woo Bae\*

Department of Life and Nanopharmaceutical Sciences and Department of Biology, Kyung Hee University

A novel facultative aerobic, non-motile and Gram-negative bacterium, designated strain H23M31<sup>T</sup>, was isolated from the gut of *Oriental stork*. Phylogenetic analysis based on the 16S rRNA gene sequence similarity indicated that strain H23M31<sup>T</sup> is most closely related to *Aequorivita capsosiphonis* DSM 23843<sup>T</sup>, sharing 96.36% similarity with that strain. Strain H23M31<sup>T</sup> belongs to the genus *Aequorivita*, which shares a clade with *A. capsosiphonis* DSM 23843<sup>T</sup> in a phylogenetic tree based on the 16S rRNA sequences. Based on genome sequencing, the DNA G+C content of strain H23M31<sup>T</sup> is 38.25 mol%, with an average nucleotide identity value, calculated by a comparative genomic analysis of strains H23M31<sup>T</sup> and *A. capsosiphonis* DSM 23843<sup>T</sup>, of 73.93%. Based on the phylogenetic, genotypic, and phenotypic information, strain H23M31<sup>T</sup> is proposed to be a novel species of the genus *Aequorivita*. The type strain is H23M31<sup>T</sup> (= KCTC 62809<sup>T</sup> = JCM 33229<sup>T</sup>).

**A051*****Planobacterium gilvum* sp. Nov., Flexirubin Type Pigments Producing Bacterium Isolated from Rice Plant Root**

Inhyup Kim, Geeta Chhetri, Jiyou Kim, Minchung Kang, and Taegun Seo\*

*Department of Life Science, Dongguk University-Seoul*

A Gram-stain-negative, short rod-shaped, aerobic, non-motile, non-spore-forming, yellow-pigmented, catalase positive, oxidase negative bacterium, designated as strain legu1<sup>T</sup>, was isolated from rice plant root. Phylogenetic analysis conducted based on 16S rRNA gene sequence studies revealed that strain legu1<sup>T</sup> was most closely related to the type strains of the genus *Planobacterium*. Strain legu1<sup>T</sup> was closely related to *Planobacterium salipaludis* KCTC 52835<sup>T</sup> (97.1%). Strain legu1<sup>T</sup> produces flexirubin-type pigments. Growth occurred at pH 6.0–10.0 (optimum, pH 7.0–8.0), with 0–2% NaCl (optimum, 0%; w/v), and at 2–37°C (optimum, 30°C). Menaquinone-7 was the predominant respiratory quinone. The major polar lipid was found to be phosphatidylethanolamine. The genome of strain legu1<sup>T</sup> has a number of 6 contigs, N50 of 727,146 and a total size of 2,765,419 nt. The DNA G + C content was 40.7 mol%. Based on phylogenetic and phenotypic analyses, strain legu1<sup>T</sup> is considered to be a novel species of the genus *Planobacterium*, for which the name *Planobacterium gilvum* sp. nov., (type strain legu1<sup>T</sup> = KACC 21166<sup>T</sup> = NBRC 113747<sup>T</sup>) has been proposed.

[This work was supported by a grant from the National Institute of Biological Resources (NIBR), funded by the Ministry of Environment (MOE) of the Republic of Korea (NIBR202002203).]

**A052****An Alternative Model to Evaluate the Virulence of *Salmonella enterica* serovar Gallinarum**

Jun-Feng Zhang, Ke Shang, Bai Wei, Min Kang, Se-Yeoun Cha, and Hyung-Kwan Jang\*

*Department of Veterinary Infectious Diseases and Avian Diseases, College of Veterinary Medicine and Center for Poultry Diseases Control, Jeonbuk National University*

Chicken pathogenicity test is the traditional method for assessing the virulence of *Salmonella enterica* serovar Gallinarum. However, this method is limited by several factors, including ethical considerations, costs, and the need for specialized facilities.

We established a chicken embryo lethality assay (ELA) model to determine the virulence of *S. Gallinarum*. Three virulent and three avirulent representative strains, which were confirmed by the chicken pathogenicity test, were used to perform the ELA. The most significant difference between the virulent and avirulent strains could be observed when 13-day-old embryos were inoculated via the AC route and incubated for 5 days. Based on a 50% embryo lethal dose (ELD<sub>50</sub>), isolates considered to be virulent had a Log<sub>10</sub>ELD<sub>50</sub> of ≤ 4.0, moderately virulent strains had a Log<sub>10</sub>ELD<sub>50</sub> of 4.0–6.1, and avirulent isolates had a Log<sub>10</sub>ELD<sub>50</sub> of ≥ 6.1. The ELA results of 42 field strains showed that thirty-two strains (76.2%) were virulent, nine were moderately virulent (21.4%), and one strain was avirulent (2.4%). In conclusion, these results suggest that the ELA can be used as an alternative method to assess the virulence of *S. Gallinarum*.

[This work was supported by Korea Institute of Planning and Evaluation for Technology in Food, Agriculture and Forestry (IPET) through Animal Disease Management Technology Development Program, funded by Ministry of Agriculture, Food and Rural Affairs (MAFRA) (120005-2, 716002-7).]

**A053****Occurrence of Antimicrobial-resistant *Salmonella* in Hatchery and Dissemination in an Integrated Broiler Chicken Operation in Korea**

Ke Shang, Bai Wei, Jun-Feng Zhang, Se-Yeoun Cha, Min Kang, and Hyung-Kwan Jang\*

*Department of Veterinary Infectious Diseases and Avian Diseases, College of Veterinary Medicine and Center for Poultry Diseases Control, Jeonbuk National University*

Horizontal and vertical transmission are both important routes of *Salmonella* dissemination in chicken. The present study is to identify the antimicrobial-resistant (AMR) *Salmonella* occurrence in breeder farm and hatchery during 2015–2016, and to reveal the transmission route of *Salmonella* along broiler production. Positive identification rates in breeder farm and hatchery were 3.0% (6/200) and 16.4% (36/220), respectively. Two serotypes of *S. Montevideo* (3/6) and *S. Virchow* (3/6) were detected in breeder farm. The most common serotype recovered from hatchery was *S. Albany* (17/36), followed by *S. Montevideo* (11/36). All 42 isolates were resistant to at least one antimicrobial, with 100% (6/6) and 52.8% (19/36) being multidrug resistant in breeder farm and hatchery, respectively. Further analysis through pulsed-field gel electrophoresis (PFGE) indicated that *S. Montevideo* isolated from hatchery showed 100% genomic identity with isolates from breeder farm, broiler farm, slaughterhouse, and retail market.

In conclusion, *Salmonella* from hatchery with a high prevalence. The AMR *Salmonella* emergence in breeder and hatchery could spread to the following chicken production stages.

[This work was supported by Korea Institute of Planning and Evaluation for Technology in Food, Agriculture and Forestry (IPET) through Animal Disease Management Technology Development Program, funded by Ministry of Agriculture, Food and Rural Affairs (MAFRA) (317009-3, 716002-7).]

**A054****Comparative Genomic Analysis of Four Probiotic *Lactobacillus* Species, *L. acidophilus*, *L. helveticus*, *L. rhamnosus*, and *L. casei***Yusook Chung<sup>1,2</sup>, Min-Jung Kwak<sup>1</sup>, Jisu Kang<sup>3</sup>, Won-Hyong Chung<sup>3</sup>, Young-Do Nam<sup>3\*</sup>, Hongman Kim<sup>2</sup>, Sanghyun Lim<sup>2\*</sup>, Myung Jun Chung<sup>2</sup>, and Jihyun F. Kim<sup>1,4\*</sup><sup>1</sup>Department of Systems Biology and Division of Life Sciences, Yonsei University, <sup>2</sup>R&D Center, Cell Biotech Co., Ltd.,<sup>3</sup>Research Group of Gut Microbiome, Korea Food Research Institute, <sup>4</sup>Strategic Initiative for Microbiomes in Agriculture and Food, Yonsei University

Several species in the genus *Lactobacillus* are widely used as starters in fermented foods and probiotics because of its high metabolic function to produce various bioactive compounds. Here, we determined complete genome sequences of *Lactobacillus helveticus* LH5 and *Lactobacillus rhamnosus* LR5, each of which had been isolated from a healthy Korean adult and compared with those of other strains in the species, along with those of *Lactobacillus casei* and *Lactobacillus acidophilus*. Through genome comparison we analyzed several factors involved in adaptation to the host intestinal conditions that include S-layer proteins, antioxidants, and bacteriocins. We also found that strains possessing a high ratio of strain-specific genes tend to have more genes for adaptation in different environments

**A055****Two New Recorded Species of Macrofungi from Mt. Munsu, Korea**Hae Jin Cho<sup>1,2</sup>, Yu Jin Oh<sup>1</sup>, Dae Wook Kim<sup>1</sup>, Jun Gi Byeon<sup>1</sup>, Yeong-Su Kim<sup>1</sup>, and Young Woon Lim<sup>2\*</sup><sup>1</sup>Baekdudaegan National Arboretum, <sup>2</sup>Seoul National University

Macrofungi play important roles in forest ecology as wood decayers, symbionts, and pathogens of living trees. For the effective forest management, it is imperative to have a comprehensive overview of macrofungi diversity in specific areas. We investigated macrofungi diversity of Mt. Munsu from May to November in 2019. More than 175 specimens were collected. The specimens were classified into 2 phyla, 4 classes, 14 orders, 41 families, 77 genera, and 116 species, and were deposited in the herbarium of Baekdudaegan National Arboretum. These specimens were identified based on morphological characteristics and sequence analysis of internal transcribed spacer (ITS) or the nuclear large subunit rRNA (LSU) region. We discovered 2 new species to Korea: *Gloiodon nigrescens* and *Leucoagaricus sublittoralis* and provided the detailed morphological descriptions.

**A056*****Novosphingobium fluvii* sp. nov., Isolated from the Nakdonggang River**

Hye Kyeong Kang, Ji Young Jung, Sang-Soo Han, Bok Yeon Jo, Eu Jin Chung, and Hyun Mi Jin\*

Nakdonggang National Institute of Biological Resources (NNIBR)

An aerobic, Gram-stain-negative, rod-shaped, DBP-degrading bacterium, designated strain EMRT-2, was isolated from the Nakdong River. Strain EMRT-2<sup>T</sup> was able to grow at 15–40°C, at pH 5.5–10.0, with 1.0% (w/v) NaCl. 16S rRNA gene sequence analysis of strain EMRT-2<sup>T</sup> showed highest sequence similarity to *Novosphingobium aromaticivorans* DSM 12444<sup>T</sup> (97.93%) and *Novosphingobium stygium* IFO 16085<sup>T</sup> (97.43%), and lower levels of similarity (<97.0%) to other species of the genus *Novosphingobium*. The major fatty acid profile consisted of C14:0 2-OH (16.53%), C16:0 (8.89%), C18:1 ω7c 11-methyl (6.35%), C16:1ω7c/C16:1ω6c (summed feature 3, 8.60%) and C18: 1ω7c/C18 :1ω6c (summed feature 8, 53.71%). The polar lipid profile constitutes Phosphatidylethanolamine (PE), phosphatidylglycerol (PG), diphosphatidylglycerol (DPG), unidentified phospholipid (PL1-3), unidentified lipid (L). The predominant quinone system was ubiquinone Q-9 and Q-10. The DNA G+C content was 66.07 mol%. Using polyphasic taxonomic analysis, we have shown that strain EMRT-2<sup>T</sup> is a novel species in the genus *Novosphingobium*, for which we propose the name *Novosphingobium fluvii* sp. nov. The type strain is EMRT-2<sup>T</sup> (= KACC 21769<sup>T</sup>).

**A057****The Study on the Unrecorded Genus and Species, *Cyanobium gracile* (Synechococcales, Cyanophyceae)**

Dae Ryul Kwon, Seung Won Nam, Sang-Soo Han, Hye Kyeong Kang, Ji Young Jung, Eu Jin Chung, and Bok Yeon Jo\*

*Nakdonggang National Institute of Biological Resources*

*Cyanobium* is a genus of the picocyanoprokaryote belonging to 14 species worldwide. In this study, we study the unrecorded genus and species, *Cyanobium gracile*. A new isolate of *C. gracile* from Adong Reservoir was established in culture under phototrophic conditions. The morphology and ultrastructure of this strain were studied with light and transmission electron microscopy. *C. gracile* lives as solitary cells without gelatinous envelopes and is symmetrical oval, ellipsoid to shortly rod-shaped. Thylakoids laid along the cell walls, and three thylakoid membranes were parallel to each other. The nucleoplasm was present in the center of the cell. A molecular phylogeny based on 16S rRNA sequences showed that the three strains of *C. gracile* formed a strongly supported monophyletic lineage (ML = 100, pp = 1.00) and, which included type strain (PCC6307) and newly recorded strain (Adong101619). Based on morphological, ultrastructural, and molecular data, we reported the newly recorded *C. gracile* in Korea.

**A058****Genome Insight and Description of Antibiotic Producing *Paraburkholderia antibiotica* sp. nov., Isolated from Arctic Soil**

Ram Hari Dahal and Jaisoo Kim\*

*Ecology Laboratory, Department of Life Science, College of Natural Sciences, Kyonggi University*

A white colour, non-motile, Gram-stain-negative, rod bacterium, designated strain RP-4-7<sup>T</sup> was obtained from soil sampled at the Arctic region. Strain RP-4-7<sup>T</sup> showed antimicrobial activity against gram-negative pathogens (*Pseudomonas aeruginosa* and *Escherichia coli*). A phylogenetic analysis based on its 16S rRNA gene sequence revealed that strain RP-4-7<sup>T</sup> formed a lineage within the genus *Paraburkholderia*. The closest members were *Paraburkholderia metrosideri* DNBP6-1<sup>T</sup> (98.8% sequence similarity), *Paraburkholderia fungorum* NBRC 102489<sup>T</sup> (98.4%) and *Paraburkholderia madseniana* RP11<sup>T</sup> (98.4%). The genome was 4,674,594 bp long with 8,304 protein-coding genes. The anti-SMASH analysis of genome showed 12 putative biosynthetic gene clusters responsible for various secondary metabolites. The sole respiratory quinone was ubiquinone-8 (Q-8). The major cellular fatty acids were summed feature 8 (C<sub>18:1</sub> ω7c and/or C<sub>18:1</sub> ω6c), summed feature 3 (C<sub>16:1</sub> ω7c and/or C<sub>16:1</sub> ω6c), C<sub>16:0</sub> and cyclo-C<sub>17:0</sub>. The principal polar lipids were phosphatidylethanolamine, diphosphatidylglycerol, phosphatidylglycerol and unidentified aminolipid. The DNA G + C content of the type strain was 61.3 mol%. Based on genomic, chemotaxonomic, phenotypic and phylogenetic analyses, strain RP-3-3<sup>T</sup> represents novel species in the genus *Paraburkholderia*, for which the name *Paraburkholderia antibiotica* sp. nov. is proposed. The type strain is RP-4-7<sup>T</sup> (= KACC 21621<sup>T</sup>).

[Supported by grants from NRF-2019R1F1A1058501]

**A059*****Paraburkholderia flava* sp. nov., Isolated from Cool Temperate Forest Soil**

Ngoc Hoang Trinh and Jaisoo Kim Kim\*

*Department of Life Science, College of Natural Sciences, Kyonggi University*

A Gram-stain-negative, aerobic and short rod-shaped bacterial strain LD6<sup>T</sup>, was isolated from cool temperate forest soil in Suwon, South Korea. The strain could grow well on R2A, TSA, NA and Mueller-Hinton agar. LD6<sup>T</sup> tolerated 2% NaCl (w/v), pH 8.0 and also grew in temperature range from 10-37°C (optimal, 28°C). The strain is most closely related to *Paraburkholderia azotifigens* NF2-5-3<sup>T</sup> (98.2%), *P. megapolitana* A3<sup>T</sup> (97.9%), *P. ginsengiterrae* DCY85<sup>T</sup> (97.9%), and *P. caribensis* MWAP64<sup>T</sup> (97.7%). The fatty acids profile mainly consisted of C<sub>17:0</sub> cyclo, C<sub>16:0</sub>, C<sub>16:0</sub>-3OH, C<sub>19:0</sub> cyclo w8c, and C<sub>12:0</sub>. The DNA G+C content of isolated strain was 63.4 mol%. The average nucleotide identity and digital DNA-DNA hybridization values between strain LD6<sup>T</sup> and its reference type strains ranged from 80.3 to 82.4%, and from 23.7 to 33.7%, respectively. LD6<sup>T</sup> represents a novel species of the genus *Paraburkholderia*, for which the name *Paraburkholderia flava* sp. nov. is proposed.

[This work was supported by a NRF 2019R1F1A1058501]

**A060*****Lysobacter solisilvae* sp. nov., Isolated from Soil**

Chae Yung Woo and Jaisoo Kim\*

*Department of Life Science, Kyonggi University*

A Gram-stain negative, aerobic, yellow-coloured, rod-shaped bacterium, designated strain II4, was isolated from soil in Seongnam city, Gyeonggi-do, Republic of Korea using 6 well plate and cultured routinely on R2A agar at 28°C for 3 days. The novel strain grew on R2A and TSA. Based on 16S rRNA gene sequence comparisons, strain II4 had close similarity with *Lysobacter terrae* THG-A13<sup>T</sup> (97.88%), *Lysobacter niabensis* GH34-4<sup>T</sup> (97.82%), *Lysobacter oryzae* YC6269<sup>T</sup> (97.74%), *Lysobacter yangpyeongensis* GH19-3<sup>T</sup> (97.53%), *Lysobacter panacisoli* CJ29<sup>T</sup> (97.24%), *Lysobacter rhizosphaerae* THG-DN8.3<sup>T</sup> (97.22%), and *Lysobacter tabacisoli* C8-1<sup>T</sup> (97.14%). Chemotaxonomic data revealed that strain II4 possesses Q8 as a predominant quinone, iso-C15:0, iso-C16:0, iso-C17:1 w9c as the major fatty acids and phosphatidylethanolamine, phosphatidylglycerol and diphosphatidylglycerol as the major polar lipids. Caseinase, tween 40, tween 60 and dnase are hydrolysed. Catalase and oxidase activities are positive. The type strain is II4 (=KEMB 9005-732<sup>T</sup> =KACC 21196<sup>T</sup> =NBRC 113956<sup>T</sup>).

[Supported by grants from NRF (2019R1F1A1058501)]

**A061*****Anaerocolumna sedimenticola* sp. nov., Isolated from Fresh Water Sediment**

Juseok Kim, Joon Yong Kim, Hye Seon Song, Yeon Bee Kim, Tae Woong Whon, Seong Woo Ahn, Se Hee Lee, and Song Woon Roh\*

*Microbiology and Functionality Research Group, World Institute of Kimchi*

Strain CBA3638<sup>T</sup> was isolated from the sediment of the Geum River, the Republic of Korea. The cells of strain CBA3638<sup>T</sup> were Gram-negative, strict anaerobic, rod-shaped (0.5–1.0 μm in width, and 4.0–4.5 μm in length). Optimal growth of strain CBA3638<sup>T</sup> occurred at 37 °C, pH 7.0 and with 1.0% NaCl concentration. Based on the 16S rRNA gene sequences, phylogenetic analysis showed that strain CBA3638<sup>T</sup> belongs to the genus *Anaerocolumna* in the family *Lachnospiraceae* and is closely related to *Anaerocolumna cellulosilytica* (94.6–95.0%). The genome of strain CBA3638<sup>T</sup> was 5,500,435 bp with 36.7% G+C content. Strain CBA3638<sup>T</sup> had seven 16S rRNA genes and specialty genes related to two transporter genes (*mefE* and *cmpA*), and did not have antibiotics resistance genes. The quinone was not observed and the predominant fatty acids were C<sub>14:0</sub> and C<sub>16:0</sub> and the major polar lipid was phosphatidylcholine. Based on phylogenetic inference and phenotypic data, we conclude that strain CBA3638<sup>T</sup> is proposed as a novel species in the genus *Anaerocolumna*, with the name *Anaerocolumna sedimenticola* CBA3638<sup>T</sup> sp. nov. (type strain CBA3638<sup>T</sup> = KACC 21652<sup>T</sup> = DSM 110663<sup>T</sup>).

**A062*****Sphingobium arcticum* sp. nov., Isolated from Arctic Soil**

Hyoyong Park, Ram Hari Dahal, and Jaisoo Kim\*

*Kyonggi University*

A Gram-negative, non-motile, aerobic bacterium, designated strain AR-3-1<sup>T</sup> was isolated from Arctic soil in Cambridge Bay, Canada. The strain AR-3-1<sup>T</sup> grew optimally at 15–28°C. Phylogenetic analysis based on 16S rRNA gene sequence showed that strain AR-3-1<sup>T</sup> formed a lineage within the family *Sphingomonadaceae* and clustered as members of the genus *Sphingobium*. The closest members were *Sphingobium cupriresistens* CU4<sup>T</sup> (98.08% sequence similarity), *Sphingobium vermicomposti* VC-230<sup>T</sup> (97.59%) and *Sphingobium lactosutens* DS20<sup>T</sup> (97.52%). The genome was 5,162,327 bp long with 4,822 protein-coding genes. The major cellular fatty acids were summed feature 8 (C<sub>18:1</sub> ω7c and/or C<sub>18:1</sub> ω6c), summed feature 3 (C<sub>16:1</sub> ω7c and/or C<sub>16:1</sub> ω6c), C<sub>14:0</sub> 2-OH, and C<sub>16:0</sub>. The major quinone was Q-10. The principal polar lipids were phosphatidylethanolamine (PE), sphingoglycolipid (SGL), phosphatidylglycerol (PG), and phosphatidylmethylethanolamine (PDE). The average nucleotide identity (ANlu) and *in silico* DNA-DNA hybridization (dDDH) relatedness values between strain AR-3-1<sup>T</sup> and phylogenetically closest members were below the threshold value for species delineation. The DNA G + C content of strain AR-3-1<sup>T</sup> was 63.15mol%. Based on genomic, chemotaxonomic, phenotypic and phylogenetic analyses, strain AR-3-1<sup>T</sup> represents novel species in the genus *Sphingobium*, for which the name *Sphingobium arcticum* sp. nov. is proposed. The type strain is AR-3-1<sup>T</sup> (=KACC21613<sup>T</sup>).

[Supported by NRF (2020M3F8A1080198).]

**A063****Production of Antimicrobial Agents from *Bacillus* spp. Isolated from Soil**

Thi Tuyet Nhan Le and Jaisoo Kim\*

*Ecology Laboratory, Department of Life Science, College of Natural Sciences, Kyonggi University*

Antimicrobial agents have been applied in many different fields such as food preservation, medicine, and environment care. The study aims to determine the biological potential of *Bacillus* strains isolated from soil against pathogenic microbes. The potential antibacterial strains were isolated in soil collected in Vietnam. The isolates were screened against *Candida albicans*, *Escherichia coli*, *Pseudomonas aeruginosa*, and *Staphylococcus aureus*. The results indicated 10 isolates exhibited strong inhibition against above pathogenic microbes. Molecular identification showed these isolates belonged to *Bacillus* genus. The strongest inhibition isolate was picked up for antimicrobial production. The antimicrobial agents were extracted using ethyl acetate, further purified, and identified the stability of antimicrobial to temperature, pH, and enzyme.

[Supported by grants from NRF (2020M3F8A1080198)]

**A064****Genome Mining and Description of *Massilia arctica* sp. nov., Isolated from Arctic Soil**

Min Gyeong Park, Ram Hari Dahal, and Jaisoo Kim\*

*College of Kyonggi University*

A Gram-stain-negative, straw colour and flexirubin test-positive bacterium, designated RP-1-19<sup>T</sup> was isolated from soil sampled at the Arctic region in Spitsbergen, Svalbard, Norway. Strain RP-1-19<sup>T</sup> hydrolysed casein, starch and CM-cellulose. Cells were aerobic, psychrophilic and grew well at 4–32°C. A phylogenetic analysis based on its 16S rRNA gene sequence revealed that strain RP-1-19<sup>T</sup> formed a lineage within the family *Oxalobacteraceae* and clustered as members of the genus *Massilia*. The closest members were *Massilia violaceinigra* B2<sup>T</sup> (98.62% similarity), *Massilia eurypsychrophila* JCM 30074<sup>T</sup> (98.34%). The genome was 4,522,469 bp long and contained 4,084 protein-coding genes. The major cellular fatty acids were summed feature 3 (C<sub>16:1</sub> ω7c and/or C<sub>16:1</sub> ω6c), C<sub>16:0</sub>, and C<sub>10:0</sub> 3-OH. The predominant isoprenoid quinone was Q-8. The principal polar lipids were phosphatidylethanolamine, phosphatidylglycerol, and diphosphatidylglycerol. The DNA G + C content was 63.3 mol%. In addition, the average nucleotide identity (ANIu) between strains RP-1-19<sup>T</sup>, B2<sup>T</sup> and JCM 30074<sup>T</sup> were 79.6 and 79.9%, respectively and DNA-DNA hybridization relatedness value were 22.9 and 23.0%, respectively. Based on genomic, chemotaxonomic, phenotypic and phylogenetic analyses, strain RP-1-19<sup>T</sup> represents novel species in the genus *Massilia* for which the name *Massilia arctica* sp. nov. is proposed. The type is RP-1-19<sup>T</sup> (= KACC 21619<sup>T</sup> = NBRC 114359<sup>T</sup>).

[Supported by grants from NRF (2020M3F8A1080198)]

**A067*****Rhizobium fluminis* sp. nov., Isolated from River in South Korea**

Se Bin Kim, Jae Young Park, and Hyo Jung Lee\*

*Department of Biology, Kunsan National University*

A Gram-staining-negative, cream colour, and strictly aerobic bacteria, designated strain KS-G21T, was isolated from stream. Strain was motile rods by a single polar flagellum showing catalase and oxidase positive activities and optimally grew at 30°C, pH 7–8, and 0% (w/v) NaCl. Strain contained ubiquinone-10 and phosphatidylethanolamine, phosphatidylglycerol, phosphatidylcholine, diphosphatidylglycerol as the major respiratory quinone and polar lipid, respectively. As the major cellular fatty acid (>5%), strain KS-G21T contained C16:0 3-OH, C16:0, cyclo-C19:0 ω8c, summed feature 8 (comprising C18:1 ω6c and/or C18:1 ω7c). The DNA G+C contents of strains KS-G21T calculated from genome was 62.8 mol%. Strain KS-G21T was most closely related to *Rhizobium alvei* LMG 26895T 96.08% 16S rRNA gene sequence similarities. In conclusion, strain KS-G21T represent novel species of the genus *Rhizobium*, for which the name *Rhizobium fluminis* sp. nov. is proposed. The type strain is KS-G21T (=KACC 01216 T = JCM 33384T).

**A068****Development of Genome-based Diagnostic Technique Using Universal Primers of Rift Valley Fever Viruses**Juseong Kim<sup>1</sup>, MinJi Kim<sup>1</sup>, and Sungmi Choi<sup>2\*</sup>*<sup>1</sup>Department of Public Health Sciences, Graduate School, Korea University, <sup>2</sup>BK21PLUS Program in Embodiment: Health-Society Interaction, Department of Public Health Sciences, Graduate School, Korea University*

The Rift Valley fever viruses (RVFV) was first identified in 1931 during an investigation into an epidemic among sheep on a farm in the Rift Valley in Kenya. It caused by an arbovirus belonging to the *Phlebovirus*. In this study, we are developing fast genome amplification and sequencing protocol for RVFV. A universal primer set designed to construct amplicons with suitable numbers and lengths for sequencing RVFV genomes will be designed and screened. The experimental procedure for reverse transcription, amplification, and construction of sequencing library will be optimized too. To verify the specificity and universality of developed primer sets and optimal protocols, the synthesized genome segments of mutated viruses will be produced and used. The expected genome amplicon will be sequenced using MinION and it will be take less than an hour. As a result of this study, we hope to develop a rapid and accurate diagnostic method to be applied to clinical samples to determine variations of RVFV.

[This work was supported by the National Research Foundation of Korea (NRF) grant funded by the Korea government (MSIT) (No.2019R1F1A1059925) and the Korea Health Technology R&D Project through the Korea Health Industry Development Institute (KHIDI), funded by the Ministry of Health & Welfare, Republic of Korea (No.HI20C0558).]

**A069****Comparison of PFGE-RFLP and 16S rRNA Gene Sequence Data for Typing of Bacillaceae Isolates from Korean Traditional Fermented Fish Sauce, Jeotgal**

Jeeyoung Lee and Eungbin Kim\*

*Department of Systems Biology, Yonsei University*

Jeotgal is salty and fermented traditional Korean fish sauce. Two-year-old jogi (small yellow croaker) jeotgal was selected based on information obtained from interviews with the local craftsmen and literature review. One hundred-microliter aliquots of the sample were plated out on Marine Broth agar plates adjusted to 5% NaCl and incubated at 25°C. A total of eighty colonies were randomly picked for PCR amplification of 16S rRNA genes followed by sequencing. All of the strains sequenced were found to belong to the single family Bacillaceae. At genus level, a total of seven genera were identified: *Bacillus*, *Virgibacillus*, *Pontibacillus*, *Halobacillus*, *Oceanobacillus*, *Piscibacillus*, and *Paraliobacillus*. Pulsed-field gel electrophoresis (PFGE) analysis demonstrated that a *Bacillus* sp. harbors a plasmid of about 200 kb. PFGE combined with restriction cleavage with different enzymes was performed to detect genetic variation in the related species. The present work suggests that PFGE-RFLP (restriction fragment length polymorphism) is a relatively easy, cheap, and effective molecular fingerprinting technique for differentiating bacteria.

[This research was financially supported by the Strategic Initiative for Microbiomes in Agriculture and Food (918011-4) funded by the Ministry of Agriculture, Food and Rural Affairs, Republic of Korea.]

**A070****Genomic and Taxonomic Study of a Novel Strain RR4-56, a Candidate for Novel Genus in the Family Rhodobacteraceae from a Seawater Recirculating Aquaculture System**Jeeun Park<sup>1,2</sup>, Young-Sam Kim<sup>1,3</sup>, and Kyoung-Ho Kim<sup>2,4\*</sup>

<sup>1</sup>Department of Microbiology, Pukyong National University, <sup>2</sup>School of Marine and Fisheries Life Science, Pukyong National University, <sup>3</sup>School of Marine and Fisheries Life Science, Pukyong National University, <sup>4</sup>Department of Microbiology, Pukyong National University

A novel bacterium, designated strain RR4-56<sup>T</sup>, was isolated from a biofilter of the seawater recirculating aquaculture system. The 16S rRNA gene sequence analysis showed that the isolate was closely related to *Halovulum dunhuangense* YYQ-30<sup>T</sup> (92.6%), *Albimonas donghaensis* DS2<sup>T</sup> (91.3%), and *Pontivivens insulae* GYSW-23<sup>T</sup> (91.3%) that all belong to the family *Rhodobacteraceae*.

The strain is irregular-rod-shaped, gram-negative, aerobic, oxidase-positive, and catalase-negative. The growth occurred at 15–35°C, pH 5.0–9.5, and 0–7% NaCl (w/v). The optimum temperature, pH, and salinity for growth were 25–30°C, pH 8.5, and 2–3% NaCl (w/v), respectively. It contained ubiquinone-10 (Q-10) and the major fatty acids were 11-methyl C<sub>18:1</sub>ω7c (31.8%), Cyclo C<sub>19:0</sub>ω8c (16.1%), and summed feature 8 (C<sub>18:1</sub>ω7c and/or C<sub>18:1</sub>ω6c, 30.4%). The polar lipids present in strain were phosphatidylglycerol, unidentified phospholipid, and unidentified aminolipid. The strain has one 4,373,045 bp chromosome with G+C contents of 65.8 mol% including 4,169 genes, 4,118 CDSs, 3 rRNAs, and 45 tRNAs. The genome annotation predicted some gene clusters related to the degradation of several types of organic matter. The result of this study demonstrated that the strain RR4-56<sup>T</sup> represents a novel genus in the family *Rhodobacteraceae*.

[This research was supported by Basic Science Research Program through the National Research Foundation of Korea (NRF) funded by the Ministry of Education (2016R1D1A3B04935909).]

**A071*****Pedobacter yeochunensis* sp. nov., Isolated from Sediment of Yecheon**Hong Sik Im<sup>1</sup>, Ha Jung Moon<sup>2</sup>, Hye Kyoung Yang<sup>2</sup>, and Sang-seob Lee<sup>1,2\*</sup><sup>1</sup>Department of Life Science, Graduate School, Kyonggi University, <sup>2</sup>Division of Bio-Convergence, Kyonggi University

A Gram-staining-negative, aerobic, motile by gliding, rod-shaped, designated strain SW-16<sup>T</sup> was isolated from the sediment of Yecheon in Suwon, Republic of Korea. Phylogenetic analysis based on the 16S rRNA gene sequence revealed that strain SW-16<sup>T</sup> formed a lineage within the genus *Pedobacter* of the family Sphingobacteriaceae. Phylogenetic analysis also showed that strain SW-16<sup>T</sup> was most closely related to *Pedobacter soli* 15-51<sup>T</sup> (98.0% 16S rRNA gene sequence similarity), along with *Pedobacter kyungheensis* THG-T17<sup>T</sup> (97.0%). The growth was observed at 10-37 °C (optimum at 30°C), pH 6-8 (optimum at pH 7), 0-2.0% NaCl (optimum at 0%). It was distinct from various species of the genus *Pedobacter*, including type strains tested in this study. The G+C content of the genomic DNA was 42.21 mol%. The major fatty acids of the bacterial strain were iso-C<sub>15:0</sub>, iso-C<sub>17:0</sub> 3-OH, and summed feature 3 (iso-C<sub>15:0</sub> 2-OH and/or C<sub>16:1</sub>ω7c). The predominant respiratory quinone was menaquinone-7 (MK-7), and the major polar lipid were phosphatidylethanolamine (PE), the unidentified lipid, L1 and L2, unidentified amino lipids, AL1 and AL2. Based on phenotypic, genotypic and phylogenetic analysis, strain SW-16<sup>T</sup> represents a novel species of the genus *Pedobacter*, for which the name *Pedobacter yeochunensis* sp. nov. is proposed. The type strain is SW-16<sup>T</sup> (= KEMB 1602-396<sup>T</sup>= KCTC 82079<sup>T</sup>).

[Supported by the grant from KGU]

**A072****Taxonomic Study of Strain RR4-35 Representing a Novel Genus in the Family *Rhodobacteraceae*, Isolated from a Seawater Recirculating Aquaculture System**Hyun-Kyoung Jung<sup>1,2</sup>, Young-Sam Kim<sup>1,2</sup>, and Kyoung-Ho Kim<sup>1,2\*</sup><sup>1</sup>Department of Microbiology, Pukyong National University, <sup>2</sup>School of Marine and Fisheries Life Science, Pukyong National University

Taxonomic study of a novel bacterial strain RR4-35<sup>T</sup> which isolated from a seawater recirculating aquaculture system (RAS) in Busan, South Korea was done. Strain RR4-35<sup>T</sup> was aerobic, gram-negative, non-motile, oxidase-positive, catalase-negative, rods cells which grew optimally at 25–30°C, pH 7.0 on marine agar, required 2% (w/v) NaCl for growth. Strain RR4-35<sup>T</sup> contained Q-10 as the predominant ubiquinone, contained summed feature 8 (C<sub>18:1</sub> ω6c, 66.22%) and C<sub>19:0</sub> ω8c cyclo (14.77%) as major fatty acids. The major polar lipids of strain RR4-35<sup>T</sup> are phosphatidylethanolamine, phosphatidylglycerol, unknown aminolipids and undefined lipids. The DNA G+C content of the type strain is 47.64 mol%. Strain RR4-35<sup>T</sup> was most closely related to *Pelagicola litorisediminis* CECT 8287<sup>T</sup> with 96.47% sequence similarity within the family *Rhodobacteraceae*. PacBio RS II sequencing yielded one complete chromosome (3,833,345 bp with 59.3% G + C content) and six plasmids which contained a total of 4,487 genes, 4,436 CDSs, 47 tRNAs, and 3 rRNA. KEGG analysis detected gene clusters related to pathways such as nitrate and nitrite ammonification, nitrosative stress, denitrifying reduction, and denitrification. Based on the polyphasic analysis, it was concluded that strain RR4-35<sup>T</sup> represents a novel genus and belonging to the family *Rhodobacteraceae* (=KCTC 72134<sup>T</sup>=MCCC 1K03753<sup>T</sup>).

[Supported by Basic Science Research Program through the National Research Foundation of Korea (2016R1D1A3B04935909).]

**A074*****Pedobacter aquadulcis* sp. nov., Isolated from Fresh Water**

Tien Chau Le Tran, Do-Hoon Lee, and Chang-Jun Cha\*

*Department of Systems Biotechnology, Chung-Ang University*

A novel bacterial strain, designated CJ43<sup>T</sup>, was isolated from fresh water located in Jeongseon-gun, Gangwon-do, South Korea. The isolate was gram-staining negative, aerobic, and rod-shaped. Strain CJ43<sup>T</sup> grew optimally at 30°C and pH 7.0 on R2A agar in the absence of NaCl. Phylogenetic analyses based on 16S rRNA gene sequences showed that strain CJ43<sup>T</sup> belongs to the family *Sphingobacteriaceae* and was the most closely related to *Pedobacter glucosidilyticus* 1-2<sup>T</sup> (98.1% sequence similarity). Based on the whole-genome sequence of strain CJ43<sup>T</sup>, the DNA G+C content was 34.9%. The average nucleotide identity and *in silico* digital DNA-DNA hybridization values between strain CJ43<sup>T</sup> and *P. glucosidilyticus* 1-2<sup>T</sup> were 88.7% and 49.4%, respectively. Strain CJ43<sup>T</sup> contained phosphatidylethanolamine as a major polar lipid. The sole detected respiratory quinone was menaquinone-7 (MK-7). The major fatty acids (> 10%) of strain CJ43<sup>T</sup> were *iso*-C<sub>15:0</sub>, *iso*-C<sub>17:0</sub> 3OH and summed feature 3 (C<sub>16:1</sub>ω6c and/or C<sub>16:1</sub>ω7c). Based on the polyphasic taxonomy data, strain CJ43<sup>T</sup> represents a new species of the genus *Pedobacter*, for which the name *Pedobacter aquadulcis* sp. nov. is proposed with the type strain CJ43<sup>T</sup> (= KACC 21350<sup>T</sup> = JCM 33709<sup>T</sup>).

[This work was supported by a project of the National Institute of Biological Resources (NIBR) to survey Korean indigenous species and funded by the Korea Ministry of Environment (MOE) as 'the Environmental Health Action Program (2016001350004)'.]

**A075*****Diaphorobacter caeni* sp. nov., Isolated from Sludge in Wastewater Plant**Hyekyoung Yang<sup>1</sup>, Hong Sik Im<sup>2</sup>, Youngmi Lee<sup>3</sup>, Yochan Joung<sup>3</sup>, and Sang-Seob Lee<sup>1,2,3\*</sup><sup>1</sup>*Life Science Major, Division of Bio-Convergence, Kyonggi University,* <sup>2</sup>*Department of Life Science, Graduate School, Kyonggi University,* <sup>3</sup>*Korea Environmental Microorganisms Bank (KEMB)*

A Gram-stain-negative, non-motile, aerobic short-rod, designated NR2-3-3-1<sup>T</sup>, was isolated from sludge in Gwacheon wastewater plant, Republic of Korea. The isolate was catalase-positive and oxidase-negative. Strain NR2-3-3-1<sup>T</sup> grew at 15-30°C (optimum 30°C) and pH 5-8 (optimum pH 7) and in the presence of 0-1.0% (w/v) NaCl. Phylogenetic analyses based on the 16S rRNA gene sequence showed that strain NR2-3-3-1<sup>T</sup> belonged to the genus *Diaphorobacter* and sharing highest sequence similarities with *Diaphorobacter aerolatus* 8604S-37<sup>T</sup> (97.4% gene sequence similarity), *Diaphorobacter nitroreducens* NA10B<sup>T</sup> (96.7%) and *Diaphorobacter oryzae* RF3<sup>T</sup> (96.3%). The only respiratory quinone was ubiquinone-8 (Q-8). The polar lipid profile comprised phosphatidylethanolamine, phosphatidylglycerol, diphosphatidylglycerol and two unknown aminolipid. The major fatty acids were C<sub>16:0</sub>, summed feature 3 (comprising C<sub>16:1</sub>ω7c and/or C<sub>16:1</sub>ω6c), summed feature 8 (C<sub>18:1</sub>ω7c or C<sub>18:1</sub>ω6c), cyclo-C<sub>17:0</sub>. The DNA G+C content was 63.2 mol%. Strain NR2-3-3-1<sup>T</sup> represents a novel species of the genus *Diaphorobacter*, for which the name *Diaphorobacter caeni* sp. nov. is proposed. The type strain is NR2-3-3-1<sup>T</sup>.

**A076*****Glaciecola mytili* sp. nov., Isolated from Mussel**Ha Jung Moon<sup>1</sup>, Hong Sik Im<sup>2</sup>, hyeyoung Sung<sup>3</sup>, Yochan Joung<sup>3</sup>, and Sang-Seob Lee<sup>1,2,3\*</sup><sup>1</sup>Life Science Major, Division of Bio-Convergence, Kyonggi University, <sup>2</sup>Department of Life Science, Graduate School, Kyonggi University, <sup>3</sup>Korea Environmental Microorganisms Bank (KEMB)

A novel bacterium, designated MH2013<sup>T</sup>, was isolated from mussel in Wando, Republic of Korea. Cells were Gram-staining-negative, aerobic, straight rod, non-motile, oxidase- and catalase- positive. Growth occurred at 4–30°C (optimum 30°C), at pH 7–8 (optimum 8) and in the presence of 0.5–8% (w/v) NaCl (optimum 2%). Comparative analysis of 16S rRNA gene sequences revealed that strain MH2013<sup>T</sup> is a member of the genus *Glaciecola*, sharing highest sequence similarities with *Glaciecola amylolytica* THG-3.7<sup>T</sup> (96.7%, 16S gene sequence similarity), and *G. nitratireducens* FR1064<sup>T</sup> (94.7%). The major fatty acids were Summed Feature 3 (C<sub>16:1</sub>ω7c and/or C<sub>16:1</sub>ω6c), C<sub>16:00</sub>, Summed Feature 8 (C<sub>18:1</sub>ω7c or C<sub>18:1</sub>ω6c). The predominant respiratory quinone was Q-8. The major polar lipids were phosphatidylethanolamine, phosphatidylglycerol, diphosphatidylglycerol and two unidentified lipid. The DNA G+C content was 44.5 mol%. On the basis of the evidence presented in this study, strain MH2013<sup>T</sup> represents a novel species of the genus *Glaciecola*, for which the name *Glaciecola mytili* sp. nov. is proposed. The type strain is MH2013<sup>T</sup>.

**B001**

**Differential Impacts of the Different Types of Soybean Pathogens on Phyllosphere and Rhizosphere Bacterial Communities**

In-Jeong Kang<sup>1</sup>, Amy Welty<sup>2</sup>, Sunggi Heu<sup>1</sup>, Hyung Kwon Shim<sup>1</sup>, and Gwyn A. Beattie<sup>2\*</sup>

<sup>1</sup>Crop Cultivation & Environment Research Division, National Institute of Crop Science, <sup>2</sup>Dept of Plant Pathology & Microbiology, Iowa State University, USA

Bacterial communities inhabiting soybean are highly diverse and expected to play pivotal roles in responding to and defending against pathogens. Here, we examined how distinct plant pathogens, including stem infections with *Phytophthora sojae* and foliar infections with *X. axonopodis* pv. *glycines* and *P. syringae* pv. *tabaci*. We also examined the extent to which these interactions were influenced by the reaction of soybean cultivars to the three pathogens, and by soybean growth in soils after a 1-year transition from paddy soil versus after at least 9 years of upland cultivation. Soybean infection with *P. sojae* strongly impacted the rhizosphere communities on both R and S hosts and in both soils, and strongly impacted the phyllosphere communities but in a soil- and cultivar-dependent manner; *P. sojae* infection had little effect on the endosphere communities. Interestingly, despite the similarity in the disease etiology and symptoms of the two foliar pathogens, these pathogens differentially impacted the endosphere communities as well as the phyllosphere communities, with minor impacts of the cultivar resistance detected only for the phyllosphere communities. These findings demonstrated distinct impacts of pathogens, even pathogens inducing similar symptoms, on soybean microbiomes, and a differential ability of distinct leaf spot pathogens to influence microbiomes within roots.

[Supported by grants from Rural Development Administration (project no. PJ01255301)]

**B002**

**Amylolytic Activity by a Marine-derived Yeast *Sporidiobolus pararoseus* PH-Gra1**

Yong Min Kwon, Hyun Seok Choi, Hyeong Seok Jang, and Dawoon Chung\*

National Marine Biodiversity Institute of Korea

Amylase accounts for approximately 30% of world enzyme production, and is utilized in the textile, food, detergent, and fermentation industries. Our laboratory have isolated a coral-red yeast strain from sea algae, designated PH-Gra1. Based on the morphological and phylogenetic analyses using sequences of internal transcribed spacer (ITS) and a D1/D2 domain of large subunit of ribosomal DNA, PH-Gra1 was identified as *Sporidiobolus pararoseus*, which is a frequently isolated yeast from marine environments. The optimum growth of this strain was observed at 22°C, pH 6.5 without addition of NaCl to the media. The extracellular crude enzyme showed the maximum amylolytic activity at 55°C, pH 6.5, and 0%-3.0% (w/v) NaCl under the examined conditions. Moreover, its activity was stable at temperatures from 15°C to 45°C for 2 h. This study provides fundamental knowledge of an amylase-producing yeast *S. pararoseus* and will facilitate industrial applications of marine yeasts.

**B004****Biodegradation of DDT by a Soil Bacterium, *Rhodococcus* sp. D7-11**

Jae-Cheol Lee, Geun-Hyoung Choi, InCheol Park, Jaehong You, Chang-Muk Lee, Si-Hyun An, and Jae-Hyung Ahn\*

*National Institute of Agricultural Sciences, Rural Development Administration*

DDT [1, 1, 1-trichloro-2, 2-bis (4-chlorophenyl) ethane] is one of the Persistent Organic Pollutants (POPs) on the Stockholm Convention list, which can cause adverse effects to human health as well as eco-systems. Although many DDT-degrading bacterial and fungal strains have been reported in the world, no such microorganisms have been reported in Korea. In this study, we isolated a DDT-degrading bacterium from a DDT-contaminated site in Korea. Through repetitive enrichment cultures using DDT as sole source of carbon and energy, a DDT-degrading bacterial strain, D7-11 was pure-cultured. The 16S rRNA gene of strain D7-11 shows a high similarity (99.35%) to that of *Rhodococcus zopfii* (NBRC 100606). Strain D7-11 degraded 46.8% of 1 mg/L of 4,4-DDT in minimum medium within 2 weeks and only 10% of degraded DDT was converted to 4,4-DDE [1,1-Dichloro-2,2-bis(4-chlorophenyl) ethylene], thus the part of DDT is supposed to be converted to less toxic substances than DDT and DDE. This result shows that strain D7-11 has a high capability to bioremediate DDT-contaminated soils. Further studies should be focused on the investigation of degradation pathway using mass spectra and toxicity reduction to soil biota.

[This work was supported by the National Institute of Agricultural Sciences, Rural Development Administration, Republic of Korea (project no. PJ04120002)]

**B006****Seasonal Dynamics of the Bacterial Communities Associated with Cyanobacterial Blooms in the Han River**Minkyung Kim<sup>1</sup>, Jaebok Lee<sup>1</sup>, Dongwoo Yang<sup>2</sup>, Hye Yoon Park<sup>3</sup>, and Woojun Park<sup>1\*</sup>

<sup>1</sup>Laboratory of Molecular Environmental Microbiology, Department of Environmental Science and Ecological Engineering, Korea University, <sup>2</sup>Department of Ecology and Conservation, National Marine Biodiversity Institute of Korea, <sup>3</sup>National Institute of Biological Resources

DNA-based analyses of bacterial communities were performed to identify the bacteria co-occurring with cyanobacterial blooms in samples collected at a single site over 2 years. *Microcystis aeruginosa* was the most predominant species within the phylum Cyanobacteria, and microcystins were detected during all cyanobacterial blooms. Culture-independent analyses of filtered bacterial communities showed that the *Flavobacterium* species in phylum Bacteroidetes was dominant in the cyanobacterial phycosphere, followed by the *Limnohabitans* species in Betaproteobacteria. To identify key bacterial species that develop long-term symbiosis with *M. aeruginosa*, another culture-independent analysis was performed after the environmental sample had been serially subcultured for 1 year. Interestingly, *Brevundimonas* was the most dominant species, followed by *Porphyrobacter* and *Rhodobacter* within the Alphaproteobacteria. Screening of 100 colonies from cyanobacterial bloom samples revealed that the majority of culturable bacteria belonged to Gammaproteobacteria and Betaproteobacteria, including *Pseudomonas*, *Curvibacter*, and *Paucibacter* species. Several isolates of *Brevundimonas*, *Curvibacter*, and *Pseudomonas* species could promote the growth of axenic *M. aeruginosa* PCC7806. The sensitivity of PCC7806 cells to different environmental conditions was monitored in bacteria-free pristine freshwater, indicating that nitrogen addition promotes the growth of *M. aeruginosa*.

[Supported by a grant from NIBR]

**B008****Isolation of Actinobacteria Growing on Plastics – Polyethylene and Polypropylene**Ha Pham<sup>1</sup>, Jae-hyung Ahn<sup>2</sup>, Hor-Gil Hur<sup>3</sup>, and Yong-Hak Kim<sup>1\*</sup>

<sup>1</sup>Department of Microbiology, Daegu Catholic University School of Medicine, <sup>2</sup>Agricultural Microbiology Division, National Institute of Agricultural Sciences, Rural Development Administration, <sup>3</sup>School of Earth Science and Environmental Engineering, Gwangju Institute of Science and Technology

The consumption of plastics rises significantly in a few decades, leading to the global contamination of plastic wastes. The waste treatment and reduction of plastic products are one of the most urgent issues all around the world. Among various kinds of plastic materials, polyethylene (PE) and polypropylene (PP) make up more than 55% in South Korea (Data from FEPIC, 2014). However, the biodegradation of PE and PP is very slow due to the crucial characteristics of high-molecular-weight, nonhydrolyzable hydrocarbon chains lacking functional groups accessible to microorganisms. We investigated plastic degrading microorganisms in activated sludge samples of industrial wastewater treatment facilities and an artificial mixture of various hydrocarbon-degrading bacteria containing different types of bacterial alkane hydroxylases and CYP450 monooxygenases. From enrichments of plastic degrading bacteria, we isolated three actinobacteria from activated sludge and an artificial mixture of hydrocarbon-degrading bacteria, which were able to grow on PE and PP as the sole carbon sources. Further experiments of fluorescence and scanning electron microscopy, denaturing gradient gel electrophoresis, 16S rRNA gene sequencing, and mass spectrometry showed that the isolated actinobacteria can play roles in the bacterial growth, biofilm formation, and partial degradation of PE and PP.

[Supported by the grant [PJ01497402] of the cooperative research program of the Rural Development Administration, Republic of Korea].

**B009****Microbial Niches in Raw Ingredients Determine Microbial Community Assembly during Kimchi Fermentation**Hye Seon Song<sup>1,2</sup>, Tae Woong Whon<sup>1</sup>, Se Hee Lee<sup>1</sup>, Jin-Kyu Rhee<sup>2</sup>, and Seong Woon Roh<sup>1\*</sup>

<sup>1</sup>Microbiology and Functionality Research Group, World Institute of Kimchi, <sup>2</sup>Department of Food Science and Engineering, Ewha Womans University

Fermented foods constitute hubs of microbial consortia differentially affecting nutritional and organoleptic properties, quality, and safety. Here we show the origin source of fermentative microbes and fermentation dynamics of kimchi. We partitioned microbiota by raw ingredient (kimchi cabbage, garlic, ginger, and red pepper) to render kimchi fermented by each source-originated microbe pool and applied multi-omics (metataxonomics and metabolomics), bacterial viability, and physiochemical analyses to longitudinally collected samples. Only kimchi cabbage- and garlic-derived microbial inoculums yielded successful kimchi fermentations. The dominant fermentative microbial taxa and subsequent metabolic outputs differed by raw ingredient type: the genus *Leuconostoc*, *Weissella*, and *Lactobacillus* for all non-sterilized ingredients, garlic, and kimchi cabbage, respectively. Gnotobiotic kimchi inoculated by mono-, di-, and tri- isolated fermentative microbe combinations further revealed *W. koreensis*-mediated reversible microbial metabolic outputs. The results suggest that the raw ingredient microbial habitat niches selectively affect microbial community assembly patterns and processes during kimchi fermentation.

**B010****'Who' and 'How' are They Getting Energy in the Ultra-oligotrophic Environment?!**

Mi-Jeong Park<sup>1,2</sup>, Sung-Han Kim<sup>1,3</sup>, Jin-Sook Mok<sup>3</sup>, Hye Youn Cho<sup>3</sup>, Jung-Ho Hyun<sup>3</sup>, Jung-Hyun Lee<sup>1,2</sup>, and Kae Kyoung Kwon<sup>1,2\*</sup>

<sup>1</sup>Korea Institute of Ocean Science and Technology, <sup>2</sup>University of Science and Technology, <sup>3</sup>Hanyang University

Sub-seafloor microorganisms play an important role in the Earth's redox evolution and are involved in ocean alkalinity. Despite these importances, little research has been done due to poor accessibility. Among them, the South Pacific Gyre (SPG) has a cell number as low as  $10^2$  per  $1\text{ cm}^3$  because the poor nutrition of the surface layer limits the amount of organic matter delivered to the seafloor. This makes studying about the biosphere a big challenge. For this reason, it has been difficult to have an integrated view of which and how microorganisms influenced environmental change in spite of the well-established studies of environmental factors. So, we tried to get DNA using samples obtained during IODP EXP329 and 16S rRNA amplicon analysis at 3 sites (U1365, U1370, and U1371) was conducted. And we could predict metabolisms which achieved in each samples through PICRUSt. For U1365 and U1370, where oxygen penetration occurs at all depths, aerobic respiration was predicted to occur at all depths, but for U1371, where oxygen is consumed entirely at the surface, aerobic respiration rarely occurs and it was predicted that anaerobic respiration involved with nitrate, manganese, and sulfate would occur actively instead. Therefore, we focus on microorganism's energy metabolisms in the SPG sub-seafloor and furthermore, help to understand how they can adapt in the harsh condition by suggesting energy cycles referring to changes of environmental factors.

[Supported by KIOST(PE99824)]

**B011****Diving deep: Exploring the Diversity of Microorganisms in Deep-sea Sediments of Hydrothermal Vent Fields in the Central Indian Ridge**

Teddy Namirimu<sup>1,2</sup>, Yun Jae Kim<sup>1</sup>, and Kae Kyoung Kwon<sup>1,2\*</sup>

<sup>1</sup>Marine Biotechnology Research Center, Korea Institute of Ocean Science & Technology, <sup>2</sup>Major of Applied Ocean Science, University of Science and Technology, Daejeon

Little is known about the community structure and functional capability of microbial communities in hydrothermal fields in the Central Indian Ridge (CIR). In this study, we explored the microbial diversity in sediment samples collected during the 2019 expedition from three hydrothermal vent fields; Invent B, Invent E and Onnuri Vent Field (OVF), across the CIR. Microbial diversity was determined by 16S ribosomal rRNA gene sequencing using Illumina MiSeq platform. *Proteobacteria* followed by *Chloroflexi*, as well as *Euryarchaeota*, *Woesearchaeota* and *Thaumarchaeota* were dominant in the all samples. Comparative analysis showed that (i) Invent E differed significantly in microbial community composition and relative abundance; (ii) samples from Invent E and OVF clustered together based on principle component analysis. Predictive functional profiling suggested that the chemoautotrophic microbes in the vents might possess the reverse tricarboxylic acid cycle and the Calvin–Bassham–Benson cycle for carbon fixation in response to carbon dioxide highly enriched in the environment, which is possibly fueled by geochemical energy with sulfur and hydrogen. This study may serve as a basis for deeply understanding the genetic diversity and functional capability of the microbial communities of Indian Ocean hydrothermal systems.

[Supported by KIOST (PE99822) and MOF (20170411)].

**B013**

**Investigation of the Groundwater Bacterial Community in Agricultural Area Based on Dilution-to-Extinction Culturing and 16S rDNA Amplicon Sequencing**

Jaeho Chang, Jaeho Song, Innam Kang, and Jang-Cheon Cho\*

*Department of Biological Sciences, Inha University*

While most of the research on groundwater microorganisms have focused on the aspects of bioremediation, pathogenic microorganisms, and microbial dark matters, cultivation of groundwater microorganisms has been less studied. Cultivation of groundwater prokaryotes, especially the uncultured, is important to understand their physiology, metabolism, and ecological roles. High-throughput-cultivation (HTC) based on dilution-to-extinction method has been proven to be highly efficient in isolating representative bacterial groups. To isolate uncultured groups of groundwater microorganisms, we investigated the microbial community of the groundwater in Yang-gu, Korea, using the seasonal HTC and 16S rRNA gene amplicon sequencing. A total of 1,042 bacterial strains were successfully cultivated using the HTC method. Phylogenetic analysis of 16S rRNA genes of the bacterial strains showed that they belong to diverse taxa such as the phyla *Proteobacteria* (*Alpha*-, *Beta*-, and *Gammaproteobacteria*), *Bacteroidetes*, and *Actinobacteria*. Notably, isolates belonging to several previously uncultured lineages, such as the OPB56 and SJA-28 clade, were obtained in this study. Through the 16S rDNA amplicon sequencing analysis, the abundance and distribution of cultured bacterial groups were also determined. This study will be a valuable resource for further studies on the microbial community dwelling in groundwater environments.

[This study was supported by a grant of the Mid-Career Research Program through the NRF (NRF-2019R1A2B5B02070538)]

**B014**

**Metabolic Features of Ganjang (a Korean Traditional Soy Sauce) Fermentation Revealed by Genome-centered Metatranscriptomics**

Byung Hee Chun and Che Ok Jeon\*

*Department of Life Science, Chung-Ang University*

Ganjang samples were prepared and major organic compounds and bacterial and fungal abundances and communities were analyzed periodically during fermentation, both before and after the removal of meju. Through metagenome and microbial isolate sequencing, 17 high-quality genomes or metagenome-assembled genomes representing all major ganjang microbiota were recovered. The absolute abundances and normalized transcriptional expression of the ganjang microbiota revealed that microbial metabolism might primarily occur while meju bricks were in the ganjang solution and decrease after the meju bricks are removed from the solution. Metabolic pathways for carbohydrates, proteins, and lipids were reconstructed and their metabolic genes were transcriptionally analyzed, revealing that facultative lactic acid fermentation by *Tetragenococcus* was the major fermentation process and that aerobic respiration by facultative aerobic bacteria was also important during ganjang fermentation. Although the fungal abundances and transcriptional expression were generally much lower than those of bacteria, our analysis suggests that yeasts such as *Debaryomyces* and *Wickerhamomyces* might be majorly responsible for producing biogenic amines and flavors.

[This work was supported by the National Research Foundation (2017M3C1B5019250 and 2018R1A5A1025077) of the Ministry of Science and ICT, Republic of Korea.]

**B015****Killing Effect of Deinoxanthin on Cyanobacterial Bloom-forming *Microcystis aeruginosa***

Wonjae Kim and Woojun Park\*

*Laboratory of Molecular Environmental Microbiology, Department of Environmental Science and Ecological Engineering, Korea University*

Cyanobacterial blooms caused mainly by *Microcystis aeruginosa* could be controlled using application of antagonistic bacteria. Killing effect of *Deinococcus*-derived carotenoids have not been tested on *M. aeruginosa* cells. *Deinococcus metallilatus* MA1002 cultured under the light increased the production of several carotenoid-like compounds. When *D. metallilatus* MA1002 cells were cocultured with axenic *M. aeruginosa* PCC7806, growth defect of *M. aeruginosa* was observed. Ethyl acetate-concentrated compounds from ethyl alcohol extracts of *Deinococcus* cells were tested to find an active compound in *Deinococcus* cell-death products. Carotenoid-like compounds were identified using thin-layer chromatography, high-performance liquid chromatography, and liquid chromatography-mass spectrometry. Deinoxanthin-like compounds could inhibit growth of axenic *M. aeruginosa* cells probably due to its interference with *Microcystis*-membrane synthesis during cell elongation. Scanning electron microscopic images showed complete collapse of *M. aeruginosa* cells under deinoxanthin-derivative added condition. Combinatory treatment of our deinoxanthin-derivative with H<sub>2</sub>O<sub>2</sub> is more effective against *M. aeruginosa* cells. Deinoxanthin-derivative can be used as a novel candidate for preventing cyanobacterial blooms.

[Supported by a Korea University grant (K2006821).]

**B016****Isolation and Characterization of a Cypermethrin-degrading Soil Bacterium, *Bacillus* sp. 3T-2**

Si-Hyun An, In Cheol Park\*, Jae Hyung Ahn, and Jae-Cheol Lee

*National Institute of Agricultural Sciences, Rural Development Administration*

Cypermethrin, one of the pyrethroid insecticides, is widely used to protect crops and animals from infestation by insects throughout the world due to its high effectiveness. However, it has been sometimes reported as a residue in soils and agricultural products thus can cause adverse effects to aquatic biota, bees and humans. In this study, five cypermethrin-degrading bacterial strains were isolated through enrichment culture using the mixture of the 49 agricultural soils and characterized for use as detoxifying agents of the insecticide. These five cypermethrin-degrading strains were identified as *Paenibacillus*, *Bacillus*, *Rhodococcus*, and *Terrabacter* based on their 16S rRNA gene sequences. One of the strains, *Bacillus* sp. 3T-2, degraded 44% of the 10 mg/L of cypermethrin in mineral medium within 2 weeks. Future works will be to identify the degradation products of cypermethrin and determine whether strain 3T-2 can degrade other pyrethroid insecticides. Strain 3T-2 might be useful for the bioremediation of cypermethrin-contaminated soil.

**B017****Temperature and Bacterial Biofilm Formation**

Suran Kim, Xi-Hui Li, Youngsun Shin, Hyeon-Ji Hwang, and Joon-Hee Lee\*  
Department of Pharmacy, College of Pharmacy, Pusan National University

*Pseudomonas aeruginosa*, an opportunistic human pathogen experiences a big change in temperature during infection into human body. In this study, we investigated the temperature effect on the biofilm formation of *P. aeruginosa* and revealed that the biofilm formation increased rapidly at temperatures lower than 25°C. *P. aeruginosa* formed the most robust biofilm of conspicuous mushroom-like structure at 20°C. However, when the temperature increased to 25°C, the biofilm formation rapidly decreased. Above 25°C, as the temperature rose, the biofilm formation increased again little by little despite its less structured form, indicating that 25°C was the low point of biofilm formation. When the intracellular c-di-GMP levels were measured with temperature, it decreased rapidly as the temperature rose from 20 to 25°C, and there was no significant change above 25°C. The expression of *pel*, *alg*, and *psl* genes encoding Pel, alginate, and Psl, respectively, were also dramatically affected by temperature, but the pattern was different. Transcription of *pel* was regulated by temperature in a pattern such as a change in intracellular c-di-GMP levels. The expression of *alg* gene rapidly decreased from 20 to 25°C, reaching a low point at 25°C, and then slightly increased again which was the most similar to the actual biofilm formation pattern. However, the expression of *psl* gradually increased with raising temperature between 20 and 37°C. Interestingly, the temperature range causing a dramatic increase in biofilm formation is different depending on the strain of *P. aeruginosa*. In PA14, a dramatic decrease in biofilm formation and c-di-GMP was observed when the temperature rose from 30 to 37°C. In conclusion, in *P. aeruginosa*, temperature is a very important factor that determines the amount and structure of biofilm formation.

**B018****Maturation of the Gut Microbiome and Its Dysbiosis in Infants with Atopic Dermatitis**

Min-Jung Lee<sup>1</sup>, Yun Kyung Lee<sup>2</sup>, Dong-Woo Lee<sup>3</sup>, Myung Hee Nam<sup>4</sup>, Soo-Jong Hong<sup>5</sup>, and Bong-Soo Kim<sup>1\*</sup>

<sup>1</sup>Department of Life Science, Multidisciplinary Genome Institute, Hallym University, <sup>2</sup>Soonchunhyang Institute of Medi-bio Science, Soonchunhyang University, <sup>3</sup>Department of Biotechnology, Yonsei University, <sup>4</sup>Seoul Center, Korea Basic Science Institute, <sup>5</sup>Department of Pediatrics, Childhood Asthma Atopy Center, Environmental Health Center, University of Ulsan College of Medicine

The establishment of the gut microbiota during infancy affects immune system development and health status in adulthood. Perturbations of the gut microbiota during this period can be contribute to development of immune-mediated diseases. The alteration of infant gut microbiome with atopic dermatitis (AD) has been reported: however, the difference of gut microbiome maturation and its role in infants with AD have not yet analyzed. We analyzed the composition and functional gene profiles of infant (6 to 36-month age) gut microbiome according to AD severity. Gut microbiome was analyzed from 348 infants by 16S rRNA gene and metagenomic sequencing (112 samples from healthy infants, 110 samples from mild AD, and 124 samples from severe AD patients). We compared the composition and functional genes of gut microbiome at different infant chronological age according to AD severity. For Indicator species analysis, the 27 species and 44 functional genes were assigned to chronological age of infants and their developments were different among infants according to AD severity. Infant with AD severity groups were impaired development of their gut microbiota, leaving them with communities that appear immature gut ecosystem than those of chronologically age-matched healthy individuals. These results can help to understand the maturation of gut microbiome in infants and its potential roles in the pathogenesis of AD according to severity.

[Supported by grants from NRF (No. NRF-2017M3A9F3043837)]

**B019****Analysis of Carbon Metabolic Profiles of *Leuconostoc mesenteroides* through Metabolomics and Metaproteomics Using LC-Q-Tof-MS**

Ju hye Baek, Kyung Hyun Kim, and Che Ok Jeon\*

Department of Life Science, Chung-Ang University

*Leuconostoc (Leu.) mesenteroides* is a representative kimchi lactic acid bacterium having an ability to produce mannitol. To investigate the carbon metabolic profiles of *Leu. mesenteroides* depending on carbon sources, *Leu. mesenteroides* DRC 1506 was cultured in chemically defined medium supplemented with glucose, fructose, galactose, mannose, arabinose, xylose, ribose, sucrose, lactose, maltose, trehalose, and cellobiose as single or mixed carbons for 60hrs and their fermentation products, intracellular metabolites, and proteomes were analyzed using LC-Q-Tof-MS, along with the measurement of pH, during cultivation. Strain DRC 1506 grew well on glucose, fructose, xylose, lactose, maltose, sucrose, and trehalose as carbon source, but showed a poor growth on other carbon sources. The growth of strain DRC 1506 reached an optical density (600nm) of approximately 0.6 for 0.3% (w/v) glucose, fructose, xylose, lactose, maltose, and trehalose, but it reached around 0.8 for sucrose, due to the production of dextran. The pH values of the cultures containing fructose and sucrose decreased to 3.8-9 and 4.0-4.1, respectively, and those of the cultures containing other carbon sources were 4.3-4.5, which suggests that *Leu. mesenteroides* may have a better metabolic ability for fructose and sucrose compared with other carbon sources. In addition, metabolome and proteome were analyzed and we will present metabolic profiles of *Leu. mesenteroides* in the poster presentation place.

**B020****Adjuvant Effect of *Silene armeria* Extract with Polymyxin B against *Acinetobacter baumannii* ATCC17978**

Min Gyeong Kang, Jae Bok Lee, Bo Ra Shin, Jae Eun Park, and Woo Jun Park\*

Laboratory of Molecular Environmental Microbiology, Department of Environmental Science and Ecological Engineering, Korea University

Non-toxic supplements are desirable to reduce amounts and side effects of polymyxins for the treatment of *Acinetobacter baumannii*. Among the 120 kinds of plant extracts, only *Silene armeria* extract (SAE) showed the synergistic effect with polymyxin B (PMB) in our fractional inhibitory concentration and time-kill analyses. Pretreatment of cells with SAE made *A. baumannii* more susceptible to PMB in exposure time- and concentration-dependent manner, indicating that SAE induced alteration of cells. To confirm the synergistic effects of PMB and SAE *in vivo*, the killing assay of *Galleria mellonella* was performed and showed the highest survival rate of *G. mellonella* infected with *A. baumannii* cells under synergistic condition. Fluorescence and scanning electron microscopic observation confirmed alteration on surface of SAE-treated *A. baumannii* cells. SAE triggered an increase in cell width and total negative charge of surface area of cells. The *pmrA* gene linked to PMB stress was overexpressed in *A. baumannii* cells under synergistic condition. Addition of osmolytes canceled synergistic effect of SAE with PMB. Morphological alteration of cells might increase the effectiveness of PMB by enhancing PMB binding to surface of cells. This study provides a promising approach for utilization of plant extracts to reduce the toxicity of PMB in clinical trials.

[Supported by grants from the National Research Foundation of Korea (No. NRF-2019R1A2C1088452).]

**B021****Functional Single Cell Approach for Probing the Drought-tolerant Bacteria by Raman Microspectroscopy and Stable Isotope Probing**

Jee Hyun No, Nishu Susmita Das, and Tae Kwon Lee\*

*Department of Environmental Engineering, Yonsei University*

A few of rhizobacteria have a capability of plant growth promotion under drought which can cause the agricultural problems. However, the conventional methods are a great hurdle to quantify abundance of unclassified drought-tolerant bacteria in environments. We assessed the abundance of drought-tolerant bacteria in the soil microbiome using Raman microspectroscopy with heavy water. The C-D bands ( $2040\text{-}2300\text{ cm}^{-1}$ ) in Raman spectra can be used as a semi-quantitative indicator of metabolic activity of drought-tolerant bacterial cells. The six model bacterial strains and three types of soil samples were incubated for 60 h in nutrient broth medium containing 40% deuterium supplemented with 20% Polyethylene glycol (PEG). In the model bacteria, the drought-sensitive bacteria with PEG did not show any detectable C-D bands, whereas the drought-tolerant bacteria with PEG were appeared the C-D bands. The metabolic activity of bacterial cells of forest soil in drought condition was significantly higher than those from agricultural field soil. This novel approach has enormous potential for assessing and comparing the abundance of drought-tolerant bacteria in the complex soil environment.

[This research was supported by the Strategic Initiative for Microbiomes in Agriculture and Food, Ministry of Agriculture, Food and Rural Affairs, South Korea (Project No. 918014-4) and National Institute of Agricultural Sciences (NAS) funded under Rural Development Administration (No. PJ013176).]

**B022****Analysis of Functional Roles of Airway Microbiome in Childhood Asthma Using *In Vitro* System and Multi-omics**

Min-Hee Kim, Min-Jung Lee, and Bong-Soo Kim\*

*Department of Life Science, Multidisciplinary Genome Institute, Hallym University*

The human airway microbiome has an important role in asthma. Various studies reported that diverse microbes coexist in the human airway and they are always interacted with each other and host. However, most studies have reported only different microbes in airway microbiome between non-asthma subjects and asthma patients. In this study, we analyze the functional role of airway microbiome based on metatranscriptomics and metabolomics using polymicrobial in vitro system. We tested various culture conditions for maintaining and growth of diverse airway microbiota, and we modified culture conditions for growing diverse airway microbes. We analyzed the role of airway microbiome in arachidonic acid metabolism and lysine degradation, which were previously reported as potentially different metabolisms by airway microbiome in children with asthma. Metabolites and metatranscriptome were analyzed during 36 h in anaerobic conditions at 37°C. Genes of airway microbiome were differently expressed among non-asthma, asthma, and remission subjects. In particular, *cyp4A* were upregulated in the airway microbiome of asthma and remission subjects after 36 h. Therefore, 20-Hydroxyeicosatetraenoic acid could generate more in asthma and remission subjects, and this molecule is involved in hypersensitivity of airway. These results help to understand the role of airway microbiome in children with asthma.

[Supported by grants from NRF (2017R1D1AB03029282).]

**B023****Dynamics of Microbial Communities and Metabolites of Doenjang, a Traditional Korean Fermented Soybean Paste, during Fermentation**

Dong Min Han, Byung Hee Chun, and Che Ok Jeon\*

*Department of Life science, Chung-Ang University*

Eleven doenjang (traditional Korean fermented soybean paste) batches were prepared using raw materials that were obtained from eleven different local manufacturers and their microbial communities and metabolites were investigated during fermentation. The initial pH values of doenjang samples were in the range of 4.7–6.1 and the pH values gradually increased to 5.6–7.4 until the end of fermentation. Bacterial and fungal community analysis using an Illumina MiSeq showed that the phyla *Firmicutes*, *Proteobacteria*, and *Ascomycota* were abundant in all doenjang batches. Bacterial community analysis at the genus level showed that *Tetragenococcus* and *Bacillus* were abundantly identified from the doenjang batches. Fungal community analysis at the genus level showed that *Aspergillus* and *Debaryomyces* were abundantly identified from the doenjang batches. The statistical analysis showed that the relative abundance of *Tetragenococcus*, *Staphylococcus*, *Idiomarina*, *Weissella*, *Millerozyma*, *Meyerozyma*, and *Wickerharmomyces* had a correlation with the amount of meju bricks and NaCl concentration. Metabolite analysis using  $H^1$ -NMR showed that the concentration of organic acids was low, while the concentration of free sugars was high in the doenjang batches containing low NaCl concentration. GC/MS analysis showed that 3-methylbutanoic acid and tetramethyl pyrazine were detected as major volatile compounds in all doenjang batches, suggesting that they are common flavor compounds of doenjang.

**B024****Molecular Evidence for the Cause of 2-Methylisoborneol (2-MIB) Odor in North-Han River, South Korea**Ju-Yong Jeong<sup>1</sup>, Sang-Hoon Lee<sup>2</sup>, Mi-Ra Yun<sup>1</sup>, Seung-Eun Oh<sup>1</sup>, Seon-Min Hwang<sup>1</sup>, Ji-Soo Park<sup>1</sup>, Chan-Won Hwang<sup>1</sup>, Tae-Hwa Kim<sup>1</sup>, Mi-Hye Yun<sup>1</sup>, and Hee-Deung Park<sup>2\*</sup>*<sup>1</sup>Department of Water Environment Research, Gyeonggi-do Institute of Health and Environment, <sup>2</sup>School of Civil, Environmental and Architectural Engineering, Korea University*

The outbreak of 2-Methylisoborneol contamination in the drinking water source causes inconvenient odor issues in the water distribution system. To investigate the major player of 2-MIB odor production in the freshwater system during the 2-MIB odor issue from the North-Han River in the fall of 2018, microscopic isolation method, and molecular phylogenetic approach were performed. In microscopic observation, a member of *Pseudanabaena* species, a kind of benthic cyanobacterium, was isolated. Phylogenetic analysis based on the sequences of the 16S rRNA gene and 16S-23S rRNA ITS region revealed that the isolated cyanobacterial strain showed 98% identity with that of *Pseudanabaena yagii*, which recorded in South Korea, for the first time. The *mibC* gene was also found in their genome and showed 100% similarity with that detected in the *Pseudanabaena* isolate from Japan. The 2-MIB production was increased slowly during mat formation on the vessel wall, however, it rapidly increases after the temperature was dropped. These results suggested that the 2-MIB odor issue in North-Han River might be caused by the release of 2-MIB from the mat formed *Pseudanabaena yagii* under the low temperature of the freshwater environment.

**B025****Analysis of Microbial Communities and Metabolite Changes of Meju, a Traditional Korean Fermented Soybean Brick, during Fermentation**

Hyung Min Kim, Byung Hee Chun, and Che Ok Jeon\*

*Department of Life Science, Chung-Ang University*

Doenjang meju samples were periodically obtained from six different local meju manufacturers during the entire fermentation period and their bacterial and fungal community dynamics, along with pH, water content, and metabolites, were analyzed to investigate the fermentation features of doenjang meju. The initial water contents of meju were approximately 60-70% and the water contents constantly decreased to 10-43% until the end of fermentation. The initial pH values were in the range of 6.6-7.0. Doenjang meju samples had two distinguished pH profiles during fermentation: in some meju, the pH values decreased rapidly during the early fermentation periods and in other meju, the pH values were relatively constantly maintained during fermentation, suggesting that they might have different microbial communities. The bacterial community analysis revealed that the genera *Bacillus*, *Enterococcus*, and *Leuconostoc* were majorly abundant during fermentation, however, their abundances varied depending on the meju batches. The fungal community analysis revealed that the genera *Aspergillus* and *Cladosporium* were abundant during fermentation. The metabolite analysis of meju samples revealed that acetate, lactate, and butyrate were identified as major organic acids. The amounts of amino acids in meju dominated by *Bacillus* were higher than that in meju dominated by lactic acid bacteria. These results will provide insights into a better understanding of Korean traditional doenjang meju fermentation.

**B026****Cell Lysis Effects of Mannitol on a Toxic Cyanobacterium, *Microcystis aeruginosa***

Ye Lin Seo, Jaejoon Jung, and Che Ok Jeon\*

*Department of Life Science, Chung-Ang University*

Harmful cyanobacterial bloom (HCB) is a global issue threatening water quality and human health. *Microcystis aeruginosa*, one of dominant species causing HCB, produces a toxic metabolite, microcystin. Many researchers have been studied to develop a strategy to control the growth of *M. aeruginosa*. While studying the interaction of *M. aeruginosa* and symbiotic bacteria via their metabolites, we found that mannitol and sorbitol caused a rapid cell lysis within 24 h at a concentration of  $> 25 \mu\text{M}$ , however, sugar alcohols such as arabitol, ribitol, xylitol, and inositol were shown no effect on *M. aeruginosa*. Microscopic observation showed that cytosol leaked out of the ruptured cell by mannitol whereas the untreated cells were bleached first and cellular structure was lost. In addition, the transcriptional analysis showed that many genes encoding photosystem I and other photosynthesis-related genes were downregulated, which means that photosynthesis might be inhibited by mannitol. Furthermore, sigma factor F which is involved in stress response were also downregulated with mannitol treatment. In contrast, most of the genes encoding ribosomal proteins and chaperone protein were upregulated to restore damaged proteins. Therefore, our results suggest that mannitol can be an effective substance to suppress the growth of *M. aeruginosa* and control cyanobacterial blooms.

**B027****Distribution Patterns of Anaerobic Ammonia-oxidizing (Anammox) Bacteria in Full-scale Wastewater Treatment Plants Located in South Korea**Sang-Hoon Lee<sup>1</sup>, Hoang Phuc Trinh<sup>1</sup>, Garam Jeong<sup>2</sup>, You-Jung Jung<sup>2</sup>, Hyeokjun Yoon<sup>2</sup>, and Hee-Deung Park<sup>1\*</sup><sup>1</sup>*School of Civil, Environmental and Architectural Engineering, Korea University*, <sup>2</sup>*Biological and Genetic Resources Assessment Division, National Institute of Biological Resources*

The application of the anaerobic ammonia-oxidizing bacteria (Anammox) process to the mainstream of municipal wastewater brings a promising scenario for the self-sufficient and energy-saving treatment plants. The limitation of the Anammox process application in the mainstream process of wastewater treatment plants (WWTP) is known as their relatively slow growth. For the successful application, it is essential to obtain a high number of the Anammox biomass by understanding their ecological roles in WWTPs and enriching locally distributed species. To understand distribution patterns of the Anammox bacteria in the WWTPs, microbial communities in the influent wastewaters, and activated sludge bioreactors were analyzed. The wastewater samples were collected from 15 full-scale WWTPs operated with media process, located in South Korea. The 16S rRNA genes based microbial community analyses revealed that the microbial communities in the influent wastewater showed significantly different distribution patterns compare to that in the activated sludge and distinctly located in the PCoA plot. The candidate *Brocadia* species was the only Anammox species observed in the media samples. The qPCR quantification revealed that the abundance of the Anammox species in the media showed up to  $10^7$ , while that in the influent wastewater and activated sludge showed less than  $10^5$ , suggesting that the Anammox species in the WWTP might be favored the surface-attached forms to overcome their slow growth rate.

**B028****A Culture Broth of *Methylosinus* sp. M13-3 Stimulates the Growth of Methane Oxidizing Bacteria Which Cannot Grow without Vitamin B<sub>12</sub>**Jaewon Ryu<sup>1</sup>, Rajendra Singh<sup>2</sup>, and Si Wouk Kim<sup>1,2\*</sup><sup>1</sup>*Department of Energy Convergence, Chosun University*, <sup>2</sup>*Department of Environmental Engineering, Chosun University*

In our previous study, several methane-oxidizing bacteria (MOB) were isolated from the rice paddy fields soils collected from Jella-namdo province. MOB was cultured in a 160 ml glass vial containing 20 ml nitrate mineral salts (NMS) medium with methane/oxygen mixture (50/50, v/v) in the headspace. In this study, we tried to investigate the mutual interaction on growth between MOB and MOB, or MOB and heterotrophs co-existed in soils. The growth of each isolated MOB was monitored in a NMS medium with or without vitamin B<sub>12</sub> as a growth factor. Among MOB isolates, a culture broth of *Methylosinus* sp. M13-3 stimulates the growth of other MOB which did not grow on NMS medium without vitamin B<sub>12</sub>. The culture broth was filtered, concentrated and fractionated by HPLC. It was found that some fractions collected from HPLC showed the same growth promoting effect on the same MOB strain. In a next study, the growth stimulating component will be characterized.

[This work was supported by a research grant of Ministry of Science and ICT (NRF-2018R1A2B2001006)]

**B029****Biodegradation and Mineralization of Styrofoam (Polystyrene) by the Microbial Communities in Gut of Mealworm**Sang-Hoon Lee<sup>1</sup>, Jeong-Hoon Park<sup>2</sup>, Hyun-Jin Kang<sup>1</sup>, and Hee-Deung Park<sup>1\*</sup><sup>1</sup>School of Civil, Environmental and Architectural Engineering, Korea University, <sup>2</sup>Sustainable Technology and wellness R&D Group, Korea Institute of Industrial Technology (KITECH)

In recently, it was reported that styrofoam was able to efficiently degraded by mealworm (the larvae of *Tenebrio molitor Linnaeus*), and approximately 50% of styrofoam was able to be converted to the CO<sub>2</sub> gas in the gut of mealworms. However, despite their life cycles (four life stages: egg, larva, pupa, and adult), the mealworms were limitedly used in the biodegradation and mineralization of styrofoam continuously. In our previous the study, the results of the comparison of microbial communities in the mealworm gut microbiota with and without feeding of styrofoam revealed that the *Proteobacteria*, especially genus *Hafinia* belonging to the class Gammaproteobacteria (49.6%), were most dominantly distributed in the mid-gut of mealworms, followed by phyla *Tenericutes* (14.7% to 24.4%) and *Firmicutes* (11.5% to 26.5%) in the mid-gut of mealworm feed with styrofoam. To specifically enrich the styrene degradable microbial consortia, enrichment culture of mealworm gut microbiota from styrofoam fed condition was performed by supplementation of styrene in the minimal media. After a month of enrichment culture, the members belonging to the several groups of microbial communities including the members belonging to the previously enriched enterobacterial members were predominated in the styrene supplemented media, suggesting that the specific microbial communities from mealworm, gut might be involved in the biological degradation of styrene in the mealworm gut microbiota.

**B031****Enhanced Micropollutants Removal by Immobilized *Bjerkandera* sp. TBB-03 with Lignocellulosic Substrate**

Bo Ram Kang and Tae Kwon Lee\*

Department of Environmental Engineering, Yonsei University

Micropollutants (MPs) are one of hazardous materials, which have adverse impacts on the public health and ecosystem even at low concentrations (ng/L to ug/L). Nevertheless, MPs are hardly removed in conventional WWTPs due to its original design to remove macronutrients. Fungal based biological treatment is regarded as a promising approach to degrade persistent MPs. There is still, however, little information on the application of fungal system on the WWTPs. In this study, we used solid state fermentation (SSF) to immobilize *Bjerkandera* sp. TBB-03 in lignocellulosic substrates. Acetaminophen, bisphenol A (BPA) and doxycycline hyclate were removed upto 90% within four hours whereas the concentrations of carbamazepine (CMZ) and sulfamethoxazole were not reduced. These results demonstrated that removal by fungal enzyme depends on the functional group of each MPs, as the fungal laccase was specialized to catalyze the oxidation of aromatic compounds containing phenol or amine group. Compared to free laccase, SSF enhanced removal rates, especially of BPA and CMZ. Complete removal of BPA through SSF was twice as fast as free laccase, and CMZ was at least captured as much as 50%. Taken together, our results suggested the enhanced removal capacity of immobilized TBB-03, which can be adaptable for a further application of fungal bioreactor in WWTPs.

[This research was funded by NRF (2017R1D1A3B03029787).]

**B032****Methionine-derivatives Metabolized by Symbiotic Bacteria Delays the Death of *Microcystis aeruginosa***Jaejoon Jung<sup>1</sup>, Yunho Lee<sup>2</sup>, Ye Lin Seo<sup>1</sup>, Ju Hye Baek<sup>1</sup>, and Che Ok Jeon<sup>1\*</sup><sup>1</sup>Department of Life Science, Chung-Ang University, <sup>2</sup>Department of Food Science and Biotechnology, CHA University

Cyanobacterial bloom is an increase of cyanobacterial abundance with harmful effects on the ecosystem and human health. Recent studies revealed that cyanobacteria and co-existing heterotrophic bacteria have complex interactions such as antagonisms or mutualism. However, the mechanisms of symbiosis are not elucidated. We revealed that the growth of a toxic cyanobacterium, *Microcystis aeruginosa* was not sustained while xenic *M. aeruginosa* culture maintained its growth for an extended period, implicating the role of co-existing bacteria in the growth of *M. aeruginosa*. *Pseudomonas* sp. MAE1-K isolated from xenic *M. aeruginosa* delayed the abrupt death of axenic *M. aeruginosa* NIES-298. Co-cultivation of NIES-298 with transposon mutant library of MAE1-K determined that knock-out of *metZ* gene coding for *O*-succinylhomoserine sulfhydrylase, participating methionine biosynthesis, accelerated the death of NIES-298. Interestingly, exogenous methionine restored the death-delay effect only in the presence of MAE1-K wild type or *metZ* mutant. This data suggested that methionine-derivatives produced by symbiotic heterotrophic bacteria supported the growth of *M. aeruginosa*. Transcriptomic analysis revealed that NIES-298 up-regulated the expression of the methionine salvage pathway, chlorophyll biosynthesis, and vitamin B12 biosynthesis in the presence of MAE1-K. Our data suggested that *M. aeruginosa* benefited essential nutrients and modulated metabolism by symbiotic heterotrophic bacteria.

**B033****Isolation and Characterization of Etofenprox-degrading Bacteria Isolated from Soils**

Ji Yeon Han and Dong-Uk Kim\*

Department of Biological Science, Sangji University

Twelve etofenprox-degrading bacteria were isolated from 73 soil samples obtained from various agricultural fields in South Korea, and their genetic and phenotypic characteristics were investigated. The isolates were able to utilize etofenprox as a sole source of carbon and energy. Analysis of 16S rRNA gene sequence indicated that the isolates were related to members of the genera, *Arthrobacter*, *Rhizobium*, *Bacillus*, *Cupriavidus*, *Bradyrhizobium*, *Mycolicibacterium* and *Sphingomonas*. Seven different chromosomal DNA patterns were obtained by polymerase-chain-reaction (PCR) amplification of repetitive extragenic palindromic (REP) sequences from the 12 isolates. This is the first report that microorganisms involved in the degradation of etofenprox have been isolated.

[This work was supported by Rural Development Administration (Project No. PJ014897032020)]

**B034**

**Isolation and Characterization of Azoxystrobin-degrading Bacteria Isolated from Agricultural Soils**

SoYi Chea and Dong-Uk Kim\*

*Department of Biological science, College of Natural Science, Sangji University*

Forty three azoxystrobin-degrading bacteria were isolated from 5 soil samples obtained from various agricultural fields in South Korea and their genetic and phenotypic characteristics were investigated. The isolates were able to utilize azoxystrobin as a sole source of a carbon and energy during the complete degradation of azoxystrobin. Analysis of 16S rRNA gene sequence indicated that the isolates were related to members of the genera, *Microvirga*, *Mycolicibacterium*, *Pseudorivibacter*, *Lysobacter*. Five different chromosomal DNA patterns were obtained by polymerase-chainreaction (PCR) amplification of repetitive extragenic palindromic (REP) sequence from the 43 isolates. This is the first report that microorganisms involved in the degradation of azoxystrobin have been isolated. [This work was supported by Rural Development Administration (Project No. PJ014897032020)]

**B035**

**Genome Sequencing of Skin-originated *Staphylococcus* and *Cutibacterium* by Nanopore Using MinION**

Jubin Kim, Han Na Oh, and Woo Jun Sul\*

*Department of Systems Biotechnology, Chung-Ang University*

Most of *Staphylococcus* is harmless and reside normally on the human skin and also *Cutibacterium* is largely commensal and part of the skin microbiome present on most healthy adult human skin. Thus, the interaction between *Staphylococcus* and *Cutibacterium* is important because skin conditions can change depending on their interaction. Technique we used for genome sequencing is Nanopore sequencing. Nanopore sequencing is a unique, scalable technology that enables direct, real-time analysis of long DNA or RNA fragments. Through Nanopore DNA sequencing using MinION device, we can get immediate data streaming for rapid, actionable results and generate short to ultra-long reads for ultimate experimental flexibility. All samples for Nanopore sequencing are isolated from human skin with cotton swabs and nose strips. Nanopore sequencing was performed with ten *Staphylococcus* and nine *Cutibacterium* samples by one flow cell. We did assembly with flye assembler and did genome annotation with Prokka. Through genome sequencing by Nanopore, we construct genome map and found factors such as antimicrobial peptide, ARG or virulence factors of each samples. This will provide the clue of interaction between *Staphylococcus* and *Cutibacterium*.

**B036****Aquatic Environment Drives Convergence in Gut Microbiome across Invertebrates and Vertebrates**

Pil Soo Kim<sup>1</sup>, Na-Ri Shin<sup>1,2</sup>, Jae-Bong Lee<sup>3</sup>, Woorim Kang<sup>1</sup>, Min-Soo Kim<sup>1,4</sup>, Tae Woong Whon<sup>1,5</sup>, Dong-Wook Hyun<sup>1</sup>, Ji-Hyun Yun<sup>1</sup>, Mi-Ja Jung<sup>1</sup>, Joon Yong Kim<sup>1,5</sup>, and Jin-Woo Bae<sup>1\*</sup>

<sup>1</sup>Department of Biology and Department of Life and Nanopharmaceutical Sciences, Kyung Hee University, <sup>2</sup>Korean Collection for Type Cultures, Biological Resource Center, Korea Research Institute of Bioscience and Biotechnology, <sup>3</sup>Distant-water Fisheries Resources Division, National Institute of Fisheries Science, <sup>4</sup>Department of Microbiology and Molecular Biology, Chungnam National University, <sup>5</sup>Microbiology and Functionality Research Group, World Institute of Kimchi

All animals on Earth have biological symbiotic associations with bacteria. However, most of the gut microbiome studies are focused on terrestrial vertebrates, especially human and mice. More than 70% of surface of Earth is covered by the aquatic environments and it is play great role as a home for more than 220,000 living species. To better understand the potential for microbiomes to influence the health, physiology, behavior and ecological basis of symbiotic relationship between animal host and indigenous microbes with numerous environmental challenges, it is necessary to perform an investigation of gut microbiota on various aquatic animals. Here, we comprehensively characterized gut bacterial communities of more than 90 vertebrate and invertebrate species, which are living in aquatic environment. The gut microbial community of fish was strongly shaped by host habitat compared to host taxonomy and trophic level and the gut microbial diversity and composition of wild cephalopods were differentiated and formed distinctly different clusters with the host phylogeny. Principal coordinate analysis revealed that the gut bacterial community of fish significantly differs from those of other vertebrates (reptiles, birds, and mammals). Collectively, these data provide a reference for future studies on the gut microbiome of aquatic animals, and insights into the relationship between fish and their gut bacterial community.

[Supported by grants from NRF and NIBR]

**B037****Metacommunity Structure in River-connected Lakes: the Importance of Water Quality in Algal Community Varied?**

Seok Hyun Ahn, Min Sung Kim, In Jae Jung, and Tae Kwon Lee\*

Department of Environmental Engineering, Yonsei University

Currently, there is a big issue of algal bloom. In this study, we have approached algal communities in metacommunity structure. The metacommunity shows how the distribution of algal communities depends on the environment. This study analyzed the algal community in the six points of the Han River by metacommunity method. The streams at all six sites showed both Clementsian and quasi-Clementsian structures, which means that the algal community responds to changes in the environment. It was classified as upstream, midstream, and downstream according to the topographical flow of the river, and it was confirmed that the water quality and algal community changed accordingly. *Asterococcus* only existed upstream, and *Aulacoseira*, *Cyclotella*, *Stephanodiscus* existed more downstream than other regions, *Rhodomonas* were found to exist a lot in the midstream. Through the canonical correspondence analysis (CCA), it was confirmed that chemical oxygen demand, biological oxygen demand, and conductivity are important environmental variables for the algal community. Temperature was also confirmed that it is an important variable in all of the upstream, midstream, downstream. Through this study, it was confirmed that the algal community in the Han River is closely related to changes in the environment. Depending on the characteristics of the region, each algal community and water quality are different. Therefore, the countermeasures for each region should be different for algal bloom.

**B038****Microbiome Taxonomic Profiles of Bacteria in the Soil around the Fire Blight-diseased Apple and Pear Trees Buried**

Ye Eun Kim, Hyeong Jin Noh, In Hee Jung, and Seong Hwan Kim\*

*Department of Microbiology and Institute of Biodiversity, Dankook University*

The Gram-negative bacterium *Erwinia amylovora* causes fire blight disease and has been destroying apple and pear orchards every year in Korea. To control this disastrous disease, all apples and pears in the orchard where an infected host was found had been buried 2 meters deep in orchard soil. The buried plants are expected to be decayed by soil microorganisms. However, there is no information about the soil microorganisms. Therefore, in order to obtain basic data on the microorganisms in buried soil, we examined the bacterial diversity present in the buried soil. Last year, apple trees buried at the Jecheon site in 2015 and apple trees buried at the Anseong site in 2016 were excavated using an excavator. Soil samples were taken at two points at each location about 50 cm above and below the buried host plant. The collected samples were subjected to microbiome taxonomic profile (MTP) analysis of 16S rDNA from bacteria. 215,863 readings were made in Jecheon. Among them, *Clostridium*, *Bacillus*, *Arthrobacter* dominated. In the Anseong soil samples, 338,953 readings were obtained, with *Lysinibacillus*, *Arthrobacter* and *Psychrobacillus* accounting for the largest proportion of the total samples. It was confirmed that the two buried places differed in the diversity of bacteria, but *Arthrobacter* appeared commonly in two regions. This is the first report on the MTP profiling of bacteria in soils around fire blight diseased plants buried.

**B039****Analysis of Microbial Communities on Waste Plastic Films Collected from Four Landfill Sites**

Jae-Hyung Ahn, Jae-Cheol Lee, Myoung-Hwa Jung, Yu-Na Jeon, Jaehong You, Chang-Muk Lee, Si-Hyun An, and Incheol Park\*

*Agricultural Microbiology Division, National Institute of Agricultural Sciences, RDA*

Microbial degradation of plastics is a potential solution to the plastic pollution. To identify and isolate the microorganisms with potentials to degrade plastics, we analyzed the microbial communities on waste plastic films and nearby soils collected from four landfill sites in Jeju, Jeonguep, and Sunchang in Korea, in which the wastes were buried before more than 10 years. Principal component analysis showed that the bacterial and fungal communities on the waste plastic films were highly different from those in the nearby soils, but they also showed distinct patterns depending on their geographical sources. Among the bacteria on the waste plastic films, the relative abundances of *Arthrobacter*, *Myxococcales*, and *Bradyrhizobiaceae* increased compared to those in the nearby soils in more than three landfill sites. Among the fungi on the waste plastic films, the relative abundances of *Mortierella* and *Thanatephorus* increased compared to the nearby soils in more than two landfill sites. These results suggest that the microorganisms enriched on the plastic films may have the ability to degrade the plastic films. At present we are trying to isolate these microorganisms and examine their degradative potentials of polyethylene films.

[This work was supported by the National Institute of Agricultural Sciences, Rural Development Administration, Republic of Korea (project no. PJ01497401).]

**B041****Enhanced Removal of Cesium by Green Microalga, *Desmodesmus armatus* SCK, under Photoheterotrophic Condition with Magnetic Separation**

Hye Kyeong Kang, Seung Won Nam, Ji Young Jung, Sang-Soo Han, and Byung-Gon Ryu\*  
*Nakdonggang National Institute of Biological Resources (NNIBR)*

This present study investigated the feasibility of using an isolated novel microalga, *Desmodesmus armatus* SCK, for treating cesium ( $\text{Cs}^+$ ) and radioactive  $^{137}\text{Cs}$  from aqueous solution with effects of different temperature, potassium ( $\text{K}^+$ )-starvation of seed cells, and organic substrate including volatile fatty acids (VFAs) on bioaccumulation of the  $\text{Cs}^+$ . The obtained results revealed that *D. armatus* SCK accumulated a relatively higher  $\text{Cs}^+$  than other green microalgae, yielding uptake of  $2.08 \text{ nmol } \text{Cs}^+ 10^6 \text{ cells}^{-1}$  at  $25^\circ\text{C}$  under illuminated condition. At lower temperature, 3.7-fold higher  $\text{Cs}^+$ -accumulation than that observed in  $25^\circ\text{C}$  was also obtained, probably due to excessive accumulation phenomena of exterior  $\text{K}^+$  by inhibitory effect of chilling stress. The biologically engineered *D. armatus* SCK grown in  $\text{K}^+$ -depletion condition allowed to increase 26% of its maximum uptake capability observed in  $\text{K}^+$ -sufficient condition. Furthermore, the capability of  $\text{Cs}^+$ -uptake by *D. armatus* SCK was significantly enhanced when the VFAs are added as organic substrates in algal culture. The result of radionuclide experiment in this study also indicated that this strain can eliminate a wide level of the radioactive  $^{137}\text{Cs}$  ranged from very lower (10 Bq/ml) to higher level (1000 Bq/ml) of its radioactivity.

[This work was supported by a grant from the Nakdonggang National Institute of Biological Resources grant funded by the Ministry of Environment (NNIBR202002104), Republic of Korea]

**B043*****In Vitro* Investigation into the Impacts of *Lactobacillus paracasei* NSMJ15 with Probiotic Potentials on the Populations of Cattle Intestinal Microbiota**

Ji Young Jung, Sang-Soo Han, Z-Hun Kim, Hye Kyeong Kang, and Eu Jin Chung\*  
*Microbial Research Department, Nakdonggang National Institute of Biological Resources (NNIBR)*

Lactic acid bacteria (LAB) was being studied to probiotics development in human and animal health for restoring the balance of intestinal microbiome in parallel with controlling pathogen. However, information about population dynamics of bacterial community including pathogens under intestinal microbiome by probiotic LAB-treatment is still limited. In this study, the main goals are to screen LAB with probiotic potential and access its impact on the bacterial population of cattle intestinal microbiome. *Lactobacillus paracasei* NSMJ15 was screened as potential probiotics from fermented product through acid and bile salt tolerance, cell surface hydrophobicity, and anti-pathogenicity determinant assays. And effects of cell-free supernatant (CFS) of strain NSMJ15 on the intestinal microbiota population, after 8 and 16 h of CFS-treatment, in 10% (v/v) fecal microbiome suspensions, were investigated by *in vitro* cultures. The growth curve analysis, from control to CFS-treated samples, showed that strain NSMJ15 could influence the intestinal microbiota population. 16S amplicon sequencing revealed that members of *Escherichia* and *Morganella*, which could possess pathogenicity, was decreased compared to control. This study showed the impact of *L. paracasei* NSMJ15 with probiotic potential on cattle intestinal microbiota population through *in vitro* assay, which might be experimental results in future probiotics development in animal use.

[Supported by grant from NNIBR funded by MOE.]

**B044****Isolation of *Sphingobium* sp. A3 Harboring the Degrading Ability of Bisphenol A**

Daewon Jeong, Hye Kyeong Kang, Ji Young Jung, Bok Yeon Jo, Eu Jin Chung, Byung-Gon Ryu, Hyun Mi Jin, and Sang-Soo Han\*

*Nakdong National Institute of Biological Resources*

Bisphenol A (BPA) is used in a wide variety of products such as plastic bottles and food packages. However, this chemical is being extensively studied, the endocrine effects observed first in animal studies, and now in humans. BPA mimics the structure and function of the hormone estrogen, it can interfere with normal bodily processes including cell growth, repair, fetal developments, and reproduction. This study was isolated and characterized the microorganism, which has the degrading ability about BPA. To isolate the microorganism, the soil samples derived from dumpsite were collected, and then enrichment culture was conducted with BPA. We finally obtained *Sphingobium* sp. A3, which was able to grow at 15-35°C, at pH 5.0–9.5. *Sphingobium* sp. A3 was closed with *Sphingobium yanoikuyae* (ATCC 51230) by analysis of the phylogenetic tree based on the 16S rRNA gene sequence. Strain A3 exhibited a high BPA-degrading ability. In conclusion, we successfully isolated the BPA-degrading bacteria strain-*Sphingobium* sp. A3-from a dumpsite. Members of the family *Sphingomonadaceae* have been previously found to possess unique systems for metabolizing various organic contaminants such as BPA. Our study suggested that strain A3 can also metabolize the BFA, which are interesting properties. Further study to gain a more detailed understanding of the properties of the BPA-degrading enzyme, mechanism, and metabolic pathway from A3 is now underway.

**B046****Biodegrading Effects of Soil Residual Pesticides by Co-inoculation of Bacterial Strains**

In-Cheol Park<sup>1</sup>, Jae-Hyung Ahn<sup>1</sup>, Si-Hyun An<sup>1</sup>, Jae-hong Yoo<sup>2</sup>, Byung-Hak Han<sup>2</sup>, and In-Cheol Park<sup>1\*</sup>

<sup>1</sup>Bioremediation Team, National Institute of Agricultural Sciences, RDA, <sup>2</sup>Agricultural Microbiology Division, National Institute of Agricultural Sciences, RDA

The pesticide degrading bacteria *Sphingomonas* sp. C8-2 and *Sphingobium* sp. Cam5-1 were isolated from agricultural soil in Korea by enrichment culture. The *Sphingomonas* sp. C8-2 is able to degrade triazole fungicide difenoconazole. The strain *Sphingobium* sp. Cam5-1 degrade cadusafos as well as 6 kinds of organophosphorus insecticides with P-S bonds. To determine the pesticides degrading activities of these bacterial strains, we conducted soil residual pesticide remove test in plastic house and field condition. The difenoconazole and 6 kinds of organophosphorus insecticide were mixed with soil at concentration of 10 mg/kg, then *Sphingomonas* sp. C8-2 and *Sphingobium* sp. Cam5-1 were co-inoculated into the soil at concentration of 10<sup>6</sup> cfu/g soil, respectively. The soil residual pesticide concentrations were analyzed at a week by high-performance liquid chromatography (HPLC). By treated with bacterial strains, difenoconazole was 65.3% (after 7 weeks) in plastic house, and 98.1% (after 2 weeks) decrease in field. The organophosphorus insecticide cadusafos was 99.7% (after 5 weeks) decreased in plastic house, and almost all (after 5 weeks) removed in field condition. Also these strains were able to reduce 82% (after 3 weeks) of ethoprosfos in plastic house and 69% (after 2 weeks) in field condition. However, reducing effects of methidathion, phenthoate, profenofos, and phorate were not shown as bacteria treatment.

**B047****Gut Microbiome Change Occurred by Diet Control in Atopic Dermatitis**Min-gyung Baek<sup>1</sup> and Hana Yi<sup>1,2\*</sup>*<sup>1</sup>Department of Public Health Sciences, Graduate School, Korea University, <sup>2</sup>School of Biosystems and Biomedical Sciences, Korea University*

Atopic dermatitis is a chronic inflammatory skin disease primarily affects young children. Recent microbiome studies repetitively reported the correlation between the disease severity and microbiome structures. Because atopic dermatitis has no definitive cure, patients often seek alternative therapies. Diet control has been used as one of the efficient therapeutic methods for atopic dermatitis, but the principle has not been revealed yet. To gain a basic knowledge on the effective mechanism of diet control in atopic dermatitis, the human gut microbiome was studied before and after designated diet control in patient and control groups. The 16S rRNA amplicon sequencing was performed using Miseq and the sequencing data were analyzed with QIIME pipeline. The microbiome profile of patient group was very similar to that of control group. The microbiome similarity was observed both samples obtained before diet control and samples obtained after diet control. The results indicate that atopic dermatitis and designed diet control did not affect the overall microbiome composition of human intestine. However, the abundance of several bacterial species was significantly different depending on diet control. Our findings revealed the characteristics of the intestinal microbiome in atopic dermatitis patients, which is very similar to the control microbiome regardless of designed diet control.

**B048****16S rRNA-based Metagenomics Reveals Diversity and Phylogenetic Relationship of Bovine Digital Dermatitis Microbiome**

Hector Espiritu, Lovelia Mamuad, Sujeong Jin, Sangsuk Lee, and Yongil Cho\*

*Department of Animal Science and Technology, Suncheon National University*

Digital dermatitis (BDD) is a polybacterial foot disease causing lameness in bovine due to painful lesions, posing serious impacts on productivity and welfare of affected animals. *Treponema* is the known agent due to high abundance but the presence of diverse microbiota suggests a synergistic role in disease development. This study investigated the diversity and phylogeny of BDD microbiome in Korea by 16S rRNA metagenomics. One normal and three BDD samples were subjected to this study. Sequencing was carried out by Illumina Miseq. Sequences were binned to operational taxonomic units (OTU) by UCLUST. The microbiome and phylogeny were analyzed. A total of 129 and 185 OTUs were observed in normal and BDD, respectively, wherein 47 OTUs overlapped. Aside from the 138 species, 15 species in the overlap presented increased abundance in BDD. Chao1 diversity index showed a more diverse microbiome in BDD. Spirochaetes and Proteobacteria were the most abundant phyla in BDD and normal skin, respectively, suggesting close relationship in each sample. The p-distance of nucleotide sequences also suggested a high divergence between OTUs. The phylogeny showed the evolutionary relationship of OTUs. Firmicutes was the most diverse phylum. We concluded that microbiome shift leading to richer diversity, and the increase in the dominance of opportunistic species are factors in disease development.

[Supported by the National Research Foundation of Korea (NRF-2018R1C1B6004589)]

**B051****Identification of *Trueperella pyogenes* Causing *Lymphadenitis* in Korean Native Black Goat**

Md Aftabuzzaman, Seon-Ho Kim, Hector M. Espiritu, Sang-Suk Lee, and Yong-il Cho\*

*Department of Animal Science and Technology, Suncheon National University*

Caseous lymphadenitis (CLA) is a chronic, contagious disease of worldwide distribution caused by *Corynebacterium pseudotuberculosis*. Abscess formation within superficial lymph nodes and internal organs were common characteristic of the disease in goat and sheep. Adult animals are more susceptible causing economic losses due to the carcass condemnation as well as animal welfare. The objective of the study is to assess the current prevalence of CLA and its etiology in Korean native black goat. The lesions were obtained from 315 animals from 31 farms within different region. Out of 315 animals, 93 carcasses presented CLA infection. All cultured samples were identified by 16S rRNA gene sequencing. As expected, majority of the isolates were *C. pseudotuberculosis* (85.5%). Interestingly, *Trueperella pyogenes* (*T. pyogenes*), in a few cases, was isolated as causative agent in CLA. This study provides the evidence in the possible involvement of *T. pyogenes* as a minor cause of CLA in Korean native goat. Nationwide survey of CLA is needed for further characterization and risk factors assessment of *T. pyogenes* infection to reach an accurate disease management in flocks.

[Supported by grants from "Cooperative Research Program for Agriculture Science and Technology Development (Project No. PJ01496003)" Rural Development Administration, Republic of Korea.]

**B052****Microbiome of Organs of Ascidian *Halocynthia roretzi*, a Sea Pineapple of Korea**Seong-Jin Kim<sup>1,2</sup> and Kyoung-Ho Kim<sup>1,2\*</sup>*<sup>1</sup>Department of Microbiology, Pukyong National University, <sup>2</sup>School of Marine and Fisheries Life Science, Pukyong National University*

An ascidian *Halocynthia roretzi* is one of economic marine invertebrate which aquacultured and consumed in Korea. Most of studies on the ascidian are usually focused on methods of aquaculture. In this study, we investigated the microbiome of three organs, gill-slit, digestive gland, and intestine, in *H. roretzi* individuals. DNA was extracted and amplified by using barcoded PCR of 16S rRNA genes. Microbial communities of three organs were identified after amplicon sequence analyses with QIIME2 pipeline. Total 43 samples contained *Parcubacteria*, *Tenericutes*, *Proteobacteria* and *Firmicutes* as a core-bacteria. Different bacterial taxa were identified in different organ as predominant groups as follows: *Gammaproteobacteria* (17.85%) in the gill-slit, *Entomoplasmatales* (26.15%) and *Hepatoplasma* (14.73%) in the digestive gland, and *Bacillus* (15.29%) in the intestine. Intestine generally showed higher values of species richness and Shannon index than the other organs in alpha indices. Gill-slit, on the other hand, comprised the simplest microbial composition. As a result of beta diversity analysis microbial community of each organ formed distinguished cluster. These results will provide to basic data with studies of microbiome in *H. roretzi*.

[This research was supported by Basic Science Research Program through the National Research Foundation of Korea (NRF) funded by the Ministry of Education (2016R1D1A3B04935909).]

**B054****Changes in the Bacterial Communities during Decomposition of Livestock Carcasses**

Michelle Miguel, Seon-Ho Kim, Sang-Suk Lee, and Yong-Il Cho\*

*Department of Animal Science and Technology, Suncheon National University*

Microorganisms play a vital role during the decomposition process. Thus, the changes in the bacterial community during decomposition of livestock carcasses was assessed in a soil with an intact microbial community and soil which was sterilized. A 2 × 2 laboratory burial set-up was made using swine and poultry carcasses in two types of soil (i. soil with an intact microbial community; ii. sterilized soil), incubated under two conditions (i. with oxygen access; ii. without oxygen access), and was observed for a period of 60 days. In this study, a total of 16 and 18 phyla were identified in decomposing swine and poultry carcasses using Illumina Miseq sequencing of 16S rRNA gene amplicons. Phylum Firmicutes was predominant in all burial set-up at all sampling period. In decomposing swine carcass set-up, *Bacillus* was the dominant genus at day 60 in all burial set-ups. Meanwhile, in the decomposing poultry carcass set-up, *Pseudogracilibacillus* and *Lentibacillus* were the dominant genera at day 60 in burial set-ups under aerobic and anaerobic condition, respectively. In conclusion, the study showed that shifts in the bacterial communities were observed during carcass decomposition.

[This work was supported by Korea Institute of Planning and Evaluation for Technology in Food, Agriculture and Forestry (IPET) through Animal Disease Management Technology Development Program, funded by Ministry of Agriculture, Food and Rural Affairs (MAFRA) (118099-03).]

**B055****Chickens Gut Microbial Community Analysis to Create Microbial Indicators of Antibiotics Substitutes**

Bo Yun Choi, Kyu Chan Lee, Dong Yong Kil, and Woo Jun Sul\*

*Dept. of Systems Biotechnology, Chung-Ang University*

In the poultry industry, antibiotics were used commonly because using them was better for chickens growth than not using them. However, Substitutes were essential as the use of antibiotics in feed was prohibited. Therefore, we intend to find out the changes in the bacterial communities of chickens fed with antibiotics and create indicators for them to lay the microbial foundation for antibiotics substitutes. Microbial community analysis of 16S rRNA genes was conducted with a total of 56 chickens over 10, 21, and 35 days after birth and according to feed with supplements (N; feed with no supplements, A; feed with antibiotics, P; feed with probiotics). Firmicutes and Bacteroidetes were the dominant bacterial phyla regardless of the groups. *Lactobacillus* was the dominant genus in N, *Alistipe CHKCI003* was the most in A, and *Bacteroides* was the most in P. As a result of the LEfSe analysis in 35 days, *Oscillibacter* occupies the most in A. *Alitipes*, dominate in A, are commonly found in chicken intestines. So, it seems that we should focus the 35 days after the birth of chickens in A to find microbial indicators of antibiotics substitutes. The physiological role of *Oscillibacter*, which occupies the most in this group, should be studied further.

**B056**

**Survivability and Effect of Beneficial Bacterium (*Bacillus* sp. KJ40) on Pepper Rhizosphere Soil Microorganisms**

Ju Hee An, Songhwa Kim, Mee Kyung Sang, Hang-Yeon Weon, and Jaekyeong Song\*

*Agricultural Microbiology Division, National Institute of Agricultural Sciences, RDA*

Recently, as the usefulness of microorganisms in agriculture has emerged, it has been applied to crops to promote growth and control disease and reduce damage caused by environmental disturbances. We selected and isolated the *Bacillus* sp. KJ40, which alleviates the drought stress of crops. The present study was conducted to observe the microbial community change of KJ40-treated pepper rhizosphere. First, by quantitative analysis using real time PCR with specific primer, survivability was analyzed for 2 months after inoculation of KJ40. Second, the pepper was inoculated with KJ40 and treated with drought stress to investigate effect on rhizosphere microorganism. In the results, KJ40 survived even after 60 days in plastic house. Quantitative PCR analysis showed that quantification of Archaea increased significantly in the soil treated drought stress regardless of the inoculation of microorganisms. To confirm the community changes in the Archaea, the MiSeq Illumina sequencing platform based on 16S rRNA gene will be performed. Keywords: Beneficial Bacterium, KJ40, Drought, Rhizosphere, qPCR

[Supported by grants from RDA]

**B057**

**Bacterial Diversity of Digestive Tracts of White Leg Shrimp, *Litopenaeus vannamei*, and Rearing Water during Biofloc Aquaculture**

Young-Sam Kim<sup>1,2</sup> and Kyoung-Ho Kim<sup>1,2\*</sup>

*<sup>1</sup>Department of Microbiology, Pukyong National University, <sup>2</sup>School of Marine and Fisheries Life Science, Pukyong National University*

Bacterial diversity in digestive tracts of whiteleg shrimp and rearing water during biofloc aquaculture was investigated using amplicon sequencing of 16S rRNA gene. The effects of temperature and carbon source on microbial community was monitored in the zero-water exchange shrimp biofloc technology (BFT) system. Unifrac PCoA analysis showed that samples were clustered according to sampling sites and culture periods. In the rearing water, the major groups of *Proteobacteria* showed positive correlation with the concentration of nitrogen compounds but negative with alkalinity, pH, and carbon source, while opposite patterns were observed in other major core groups. *Planctomycetes* and *Actinobacteria* groups were positively correlated with culture period, while *Proteobacteria* and *Verrucomicrobia* groups were negatively correlated in the shrimp digestive tract. Most *Actinobacteria* were positively correlated with temperature, while *Bacteroidetes* were negatively correlated with it in all habitats. The present study will provide useful information for development of effective biofloc aquaculture of white leg shrimp, *Litopenaeus vannamei* through understanding of effect of temperature and wheat flour treatment on bacterial communities.

[Supported by Basic Science Research Program through the National Research Foundation of Korea (2016R1D1A3B04935909).]

**B058****Screening and Isolation of Diazinon Degrading Bacteria**Songhwa Kim<sup>1,2</sup>, Ju Hee An<sup>1</sup>, Mee Kyung Sang<sup>1</sup>, Hang-Yeon Weon<sup>1</sup>, Ho-Jong Ju<sup>3</sup>, and Jaekyeong Song<sup>1\*</sup>

<sup>1</sup>Agricultural Microbiology Division, National Institute of Agricultural Sciences (NAS), Rural Development Administration (RDA), <sup>2</sup>Division of Agricultural Biology, College of Agriculture and Life Sciences, Chonbuk National University, <sup>3</sup>Division of Agricultural Biology, College of Agriculture and Life Sciences, Chonbuk National University

The excessive use of pesticides has generated a number of environmental problems such as contamination of air, water and terrestrial ecosystems. Diazinon has been widely used throughout the world with applications in agriculture and horticulture for controlling insects in crops, ornamentals, lawns, fruit and vegetables. Pesticide residues may cause serious problems in agricultural field. In this study, to isolate a highly effective diazinon-degrading microorganism, we carried out enrichment from plastic house soil and analysis of degrading-activity of 1,800 strains which isolated previously. Degrading activity was checked using thin-layer chromatography (TLC) and high-performance liquid chromatography (HPLC).

These results showed that 5 isolates degraded 70–80% of the diazinon in 5 days. And a total of 110 isolates grown well onto minimal medium (MM) plates containing diazinon were selected. For field application, we plan to find the better strains that degrade diazinon and construct diazinon-degrading microorganism consortium.

Keywords: Diazinon, degrading-bacteria

[Supported by grants from RDA]

**B059****High and Low Level Mupirocin Resistance in *Staphylococcus***

Haeseong Lee and Jong-Chan Chae\*

Division of Biotechnology, Jeonbuk National University

*Staphylococcus aureus*, *Staphylococcus epidermidis*, and *Staphylococcus pseudintermedius* are Gram-positive bacteria which are normal flora on the skin of humans and mammals. Among these strains, *S. aureus* and *S. epidermidis* are found mainly in human skin, and *S. pseudintermedius* is found mainly in animal skin. Mupirocin acts on isoleucyl tRNA synthetase of bacteria, inhibiting protein synthesis. It is used very effectively in the treatment of infection of methicillin-resistant *S. aureus*. However, mupirocin resistant *Staphylococcus* have been emerged and two levels of resistance for mupirocin have been known: low-level resistance and high-level resistance. While low-level resistance is conferred by the *ileS* gene located in the chromosome of bacteria, the *mupA* gene located in plasmid is responsible for high-level resistance. Therefore, high-level mupirocin resistance can be transferred by conjugation between *Staphylococcus* and mupirocin resistance in *Staphylococcus* has currently increased worldwide. In this study, 5 strains of methicillin resistant *S. epidermidis* and 3 strains of methicillin resistant *S. pseudintermedius* were isolated for mupirocin resistance from companion animals and residential environment. As results of the whole genome sequencing, *mupA* and *ileS* genes were detected in chromosome and plasmid, and low- and high-level resistances were consistent with the genotypes and gene locations. Also, the analyzed genome contexts were compared in this study.

**B060****Prevalence of Methicillin-Resistant *Staphylococcus* and Their Genotypic Identifications**

Haeseong Lee, Hyeonbin Kim, and Jong-Chan Chae\*

*Division of Biotechnology, Jeonbuk National University*

*Staphylococcus* is a normal flora in the skin and nasal of humans and mammals. *Staphylococcus* is one of the most important pathogens related with antibiotic resistance. Thus, in this study, we investigated the prevalence of *Staphylococcus aureus* (MRSA), *Staphylococcus epidermidis* (MRSE) and *Staphylococcus pseudintermedius* (MRSP) showing resistance to methicillin from companion animals and host humans. While *S. aureus* is one of normal flora found mainly in human skins, *S. epidermidis* and *S. pseudintermedius* are known as normal flora found mainly in animal skins. However, the same clones of *S. aureus*, *S. epidermidis*, and *S. pseudintermedius* were shared in the skins of humans and animals. As a result of genotypic investigation of *S. aureus*, they were ST72 clones harboring SCCmec type IV. But *S. epidermidis* and *S. pseudintermedius* were various ST type clones containing various SCCmec types. The shared strains were selected and their genomes were analyzed. The compared genomes showed 100 percent similarities containing the same antibiotic resistant genes. This result suggests that the same clones are transmitting each other between different hosts.

**B061****Isolation and Phylogenetic Analysis of Geosmin and 2-MIB-Producing Actinobacteria in North-Han River Watershed**

Seung-Hyeok Soung and Song-Ih Han\*

*Department of Microbiology & Resouces, College of Science & Technology, Mokwon University*

Occurrence of the geosmin and 2-methylisoborneol (MIB) in water environments indicates that odour-producing organisms are commonly occurring. To address these issues, actinomycetes, the major producers of geosmin and 2-MIB, have been investigated extensively. To effectively respond to the odor material produced by actinobacteria, it is important to specify the odor-producing potential, in which actinobacteria are distributed. This investigation evaluates the enumeration and isolation of actinobacteria from the water and sediment soils in North-Han River Watershed and then phylogenetic characterization of the isolated actinobacteria. We obtained 343 actinobacteria from January to June 2020. Among 343 actinobacteria, we selected 58 representative isolates. The isolates categorized as 3 genera (*Streptomyces*, *Nocardia*, *Kitasatospora*) by 16S rRNA gene sequence analysis. The potential and expression of odor synthesis genes were analyzed by using the PCR methods. The genes producing geosmin and 2-MIB were detected large number of isolates. This study confirmed that actinobacteria significantly affect the production of geosmin and 2-MIB that causes odors in North-Han River Watershed.

**B062****Identification of Cyanobacterial Community Structure and Variation of *mibC* Gene Expression in the Lake Uiam**Keonhee Kim<sup>1</sup>, Sejin Lee<sup>2</sup>, Kyunghwa Seo<sup>2</sup>, and Soon-Jin Hwang<sup>1\*</sup><sup>1</sup>Human and Eco-Care Center and Department of Environmental Health Science, Konkuk University, <sup>2</sup>Department of Water Resources Management, Hangang River Regional Head Office, K-Water

Harmful cyanobacteria producing 2-MIB, an odorous material, occur in many eutrophic freshwater systems around the world and cause both socio-economic and environmental problems in water use. The Lake Uiam located in the upstream part of the North Han River has been reported to have high concentrations of 2-MIB since 2018, but the responsible organism(s) which produces 2-MIB is remained unclarified. This study analyzed the whole cyanobacterial community and potentially 2-MIB-synthesizing cyanobacteria community in the Lake Uiam. Sediment and waters sample were taken by a grab sediment sampler and Vandonn sampler, respectively. eDNA was extracted from the both sediment and water samples by a commercial extraction kit. Also, pyrosequencing and *mibC* gene cloning were performed. Total cyanobacterial community composed of 14 genera and 28 species in the sediment accounted for only a minute proportion (0.01%) of the whole bacterial community. *Tychonema bourrellyi* was found to be most dominant, followed by *Cyanobium gracile* and *Calothrix* sp. In addition to *Tychonema*, *Leptolyngbya* found to be a rare genus in the study site is known to produce 2-MIB but their *mibC* genes were found only in the sediment sample. While in the water column, the *mibC* gene of planktonic *Pseudanabaena* genus accounted for the majority in which the *mibC* genes of *Pseudanabaena redekei* and *P. yagii* showed a large proportion. The proportion of *mibC* gene of *Planktothricoides* genus was very low. Among the seven cyanobacteria strains isolated from the lake area (both water column and the sediment) where the Gong-Ji Stream merged, six strains possessed the *mibC* gene. However, only two strains, *Pseudanabaena redekei* and *Planktothricoides raciborskii* were identified to actually produce 2-MIB substances. Therefore, we suspect that major producer of 2-MIB in the Lake Uiam is *Pseudanabaena* in the water column and *Tychonema* and *Leptolyngbya* in the sediment. [This study was supported by the 2020 Han River Basin Environmental Survey Project]

**B063****Catabolism of 2-Hydroxyphenylacetate in *Comamonas testosteroni* Strain P19**

Hyeonbin Kim, Haeseong Lee, Kui-Jae Lee, and Jong-Chan Chae\*

Division of Biotechnology, Jeonbuk National University

*Comamonas testosteroni* strain P19 has a 5 Mb chromosome with one cryptic plasmid. The genome contained gene clusters encoding proteins for metabolism of several aromatic hydrocarbons, including anthranilate, benzoate, biphenyl, *m*-hydroxybenzoate, *p*-hydroxybenzoate, *p*-methoxybenzoate, phenol, phthalate, protocatechuate, and terephthalate. A mutant of P19 strain lost the ability to degrade 2-hydroxyphenylacetate (2HPA). Whereas the mutant strain did not grow in the presence of 2HPA as a sole carbon source, it was capable of metabolizing analogous compounds, 3-hydroxyphenylacetate (3HPA) and 4-hydroxyphenylacetate (4HPA). The strain had a mutation on a gene for cytochrome P450 (CYP). The deduced polypeptide consisted of domains for cytochrome P450, ferredoxin reductase, and ferredoxin indicating multifunctional property of hydroxylation and electron transfer. The transcription of CYP gene was induced by 2HPA, but not by 3HPA or 4HPA. In this study, the novel CYP was found to play a key role in hydroxylation of 2HPA in strain P19.

**B064**

**Antibiotic Resistance in Streams and the Effect of Discharged Wastewater**

Haeseong Lee<sup>1</sup>, Hyeonbin Kim<sup>1</sup>, Woo Jun Sul<sup>2</sup>, Chang-Jun Cha<sup>2</sup>, Kui-Jae Lee<sup>1</sup>, and Jong-Chan Chae<sup>1\*</sup>

<sup>1</sup>*Division of Biotechnology, Jeonbuk National University,* <sup>2</sup>*Department of Systems Biotechnology, Chung-Ang University*

In this study, we monitored genes for antibiotic resistance and planktonic bacteria in natural streams and discharged waters of wastewater treatment plants. The diversity of bacteria and antibiotic resistance were analyzed by culture dependent and independent methods. The dominant families were *Aeromonadaceae*, *Enterobacteriaceae*, and *Flavobacteriaceae* and bacterial community was affected by discharged waters. The abundance of antibiotic resistant genes (ARG) and mobile genetic elements (MGE) was higher in discharged waters than in streams. Especially, higher abundance of tetracycline resistant genes was detected in water discharged from aquaculture farm and bacterial isolates showed more strong resistance against tetracycline. Getting together, these results indicate that discharged waters are affecting on the bacterial community and resistome in natural environment.

**C001****The Effect of Red Clover (*Trifolium pratense*) Extract on Gut Microbiota and the Bone Quality in Ovariectomized (OVX) Rats**

Yixian Quah and Seung-Chun Park\*

*Laboratory of Applied Pharmacokinetics & Pharmacodynamics, College of Veterinary Medicine, Kyungpook National University*

Prolonged consumption of herbal extract has shown to regulate the composition of gut microbiota. Red clover (*Trifolium pratense*) extract was known to be taken as dietary supplement to ease menopausal symptoms. Thus, the aim of this study was to elucidate the effect of red clover (RC) extract on the gut microbiota as well as the bone metabolic and microstructure in the OVX model. Gut microbiota analysis revealed that the RC treatment improved significantly the intestinal microbiota composition in the OVX rats by recovering the abundance of Verrucomicrobia and reducing Firmicutes compared to the NC group. *Akkermansia muciniphila* was the highest relative abundance strain in the taxonomic composition and its abundance was highest in RC group. MicroCT analysis showed that RC treated groups showed improvement in the BMD and BV/TV values compared to the NC group. Our real-time PCR analysis results showed that RC treatment was able to suppress the osteoclastic gene RANKL expression but did not show significant effects on osteoblastic genes (OPG, OCN, ALP and type 1 collagen  $\alpha$ ). Besides, serum biomarker analysis revealed that RC reduced the level of triglyceride and LDL/VLDL and increased HDL level. Taken together, RC treatment have shown changes in the gut microbiota taxonomic composition and the relationships between gut microbiota and bone quality have been studied in this study.

**C002****Enrofloxacin-sulfamethoxazol/trimethoprim Combination Shows Synergistic Effects against Pathogenic Bacteria Isolated from Pigs**

Eon-Bee Lee, Biruk Tesfaye Birhanu, and Seung-Chun Park\*

*Laboratory of Veterinary Pharmacokinetics and Pharmacodynamics, College of Veterinary Medicine, Kyungpook National University*

Due to the indiscriminate use of a single antibacterial, bacteria that are resistant to certain antibiotics increase significantly. Therefore, research to inhibit the appearance of resistant bacteria remains an important challenge. Previous studies have shown that combinations of antibacterial with different efficacy can reduce drug resistance without side effects, so combination therapy can be applied to veterinary medicine. Enrofloxacin (ENFX) is effective for the bactericidal activity of various bacteria in pigs. Sulfamethoxazole /trimethoprim (5:1) mixture (ST) is widely used as an antibacterial to inhibit microorganisms. In this study, the combinatorial effect of the ENFX-ST combination on five bacterial species was designed whether the combination exhibited a synergistic effect, or not. To know this, MIC, MBC, FIC and the mortality rate were performed for these bacteria. Meanwhile, the optimal dose was selected to be the product by conducting a single oral toxicity test in rats and an efficacy test in pigs. Finally, ENFX 25 + ST 75 ratio was selected as the final formulation considering antibacterial pharmacodynamics and production cost. The combination showed significant efficacy in pigs with digestive and respiratory symptoms. When this combination was administered as a therapeutic dose, the withdrawal period was calculated as 10 days. From the above results, it can be suggested that the optimal ENFX-ST combination is promising for veterinary antibacterial treatment.

**C004****Anti-inflammatory and Anti-fibrotic Effects of *Nocardiosis* sp. 13G027 Extract in Transforming Growth Factor  $\beta$ 1-Induced Nasal Polyp-derived Fibroblasts**Grace Choi<sup>1</sup>, Hyukjae Choi<sup>2</sup>, Dae-Sung Lee<sup>1</sup>, and Il-Whan Choi<sup>3\*</sup><sup>1</sup>Department of Genetic Resources, National Marine Biodiversity Institute of Korea, <sup>2</sup>College of Pharmacy, Yeungnam University, <sup>3</sup>Department of Microbiology and Immunology, Inje University College of Medicine

The genus *Nocardiosis* produce a variety of bioactive compounds such as antimicrobial agents, anticancer substances, toxins and etc. In this study, we investigated the anti-inflammatory and anti-fibrotic effects of *Nocardiosis* sp. 13G027 extract in lipopolysaccharide (LPS) induced RAW 264.7 macrophages and transforming growth factor  $\beta$ 1- induced nasal polyp-derived fibroblasts. Treatment with the 13G027 extract significantly repressed the production of inflammatory mediators such as nitric oxide (NO) and reactive oxygen species. We tested the expression levels of  $\alpha$ -smooth muscle actin ( $\alpha$ -SMA), collagen type-1 (Col-1), fibronectin and phosphorylated-small mothers against decapentaplegic (Smad)3 in NP tissues were measured by western blot analysis and immunohistochemistry for anti-fibrotic effect of 13G027 extract. TGF- $\beta$ 1 induced the expression  $\alpha$ -SMA, Col-1 and fibronectin, and stimulated fibroblast-mediated contraction of collagen gel. However, pre-treatment with 13G027 extract inhibited the expression of these proteins. The inhibitory effects were mediated through the suppression of Smad3 signaling pathways in TGF- $\beta$ 1-induced NPFDs. These results suggested that 13G027 extract may be important in inhibiting myofibroblast differentiation and extracellular matrix protein accumulation in NP formation through the Smad3 signaling pathway.

[This work was supported by a research grant (2020M00500) from the National Marine Biodiversity Institute of Korea, Republic of Korea.]

**C005****Characterization of Various Fungi Isolated from Korean Traditional Meju for Taste Enhancement of Fermented Soybean Foods**So-Won Lee<sup>1</sup>, Oh-Cheol Kim<sup>1</sup>, Jiye Hyun<sup>1</sup>, Sunyeong Kim<sup>1</sup>, Hyewon Mok<sup>1</sup>, Jaejung Lee<sup>1</sup>, Seung-Bum Hong<sup>2</sup>, and Yong-Ho Choi<sup>1\*</sup><sup>1</sup>Sempio Fermentation Research Center, Sempio Foods Company, <sup>2</sup>Agricultural Microbiology Division, National Academy of Agricultural Science, Rural Development Administration

Low-molecular-weight peptides and amino acids are key ingredients that impart the deep, rich taste of Jang. In order to increase the taste ingredient, fungi with high enzyme activity such as protease, LAP and GGT should be used as meju starter. Generally, *A. oryzae* is industrially used as a single starter because it has high protease activity. But there is a limit to making simple taste and flavor.

So to find a novel meju starter except of *A. oryzae* that will enrich the flavor of the jang, we researched fermentation of soybean using various fungi obtained from traditional meju.

We conclude that *Scopulariopsis brevicaulis*, *Penicillium solitum* and *Aspergillus luchuensis* was excellent in ability to produce taste ingredients. Meju made with *S. brevicaulis* showed high enzyme activity in Protease, CAP, DAP, and glutaminase. And the soy sauce had high amino acid content and total nitrogen content. Meju made with *P. solitum* showed the highest GGT activity and the high protease activity. This soy sauce had a high content of amino acids and total nitrogen. *A. luchuensis* had lower enzymatic activity than the two types of fungi, so the content of amino acid and total nitrogen in soysauce was low. However, the proportion of glutamic acid was very high.

[This work was carried out with the support of "Cooperative Research Program for Agriculture Science and Technology Development (Project No. PJ01453902)" Rural Development Administration, Republic of Korea.]

**C006****Selection of *Lactococcus lactis* HY7803 for Glutamate Production Based on Comparative Genomic Analysis**

Jungmin Lee<sup>1</sup>, Sojeong Heo<sup>1</sup>, Jihoon Choi<sup>2</sup>, Minsoo Kim<sup>2</sup>, Eunji Pyo<sup>2</sup>, Myounghee Lee<sup>2</sup>, Sangick Shin<sup>2</sup>, Jaehwan Lee<sup>2</sup>, Jaehun Sim<sup>2</sup>, and Do-Won Jeong<sup>1\*</sup>

<sup>1</sup>Department of Food and Nutrition, Dongduk Women's University, <sup>2</sup>R&BD Center, Korea Yakult Co., Ltd.

Eight lactic acid bacteria species, *Lactococcus lactis*, *Lactobacillus plantarum*, *Lactobacillus casei*, *Lactobacillus brevis*, *Leuconostoc mesenteroides*, *Lactobacillus fermentum*, *Lactobacillus buchneri*, and *Lactobacillus curvatus* were performed the comparative genomic analysis to determine glutamate, the purest taste of umami, production pathway. *In silico* prediction of the amino acid biosynthesis pathways revealed that eight LAB species did not possessed genes for the glutamate production, except conversion glutamate from glutamine in *L. lactis* and *Leu. mesenteroides*. Moreover, only *L. lactis* IL1403 contained related genes for glutamate production from glucose via citrate. Based on comparative genomic analysis, *L. lactis* was suggested the proper species for glutamate production. *L. lactis* HY7803 strain was selected among 17 strains for glutamate production. Strain HY7803 produced glutamate at 0.619  $\mu\text{mol/ml}$  concentration level and citrate, precursor for glutamate production, increased into the glutamate production at 0.734  $\mu\text{mol/ml}$  level. Our results indicate that HY7803 strain has been selected based on comparative genomic analysis for glutamate production.

**C008****Functional and Localization Analysis of Alcohol Acetyltransferases Responsible for Aroma Ester Production in *Saccharomyces fibuligera***

Hyeon Jin Kim, Ki Seung Kim, Hye Yun Moon, Su Jin Yoo, Dong Wook Lee, and Hyun Ah Kang\*

Department of Life Science, College of Natural Science, Chung-Ang University

Aroma ester components are responsible for the fruity character of fermented alcoholic beverages. Acetate esters are produced by fermenting yeast cells via alcohol acetyltransferase (ATF). By bioinformatics analysis of the whole genomes of the amylolytic yeast *Saccharomyces fibuligera* KJJ81, isolates from Nuruk we identified 12 multiple ATF orthologues (*SfATF*) in the diploid genome of *S. fibuligera* KJJ81. The identified *SfAtf* proteins display low sequence identities to *S. cerevisiae* Atf1p from 13.3 to 27.0%. All of them, except *SfAtf(A)4p* and *SfAtf(B)4p*, contain the activation domain (HXXXD) conserved in other Atf proteins. Western blot analysis showed that *SfATF(A)2*, *SfATF(B)2* and *SfATF(B)6*, which showed relatively high mRNA levels in *S. fibuligera*, were highly expressed in the heterologous host *Saccharomyces cerevisiae*. Analysis of culture supernatants by headspace gas chromatography GC-MS confirmed that the *S. cerevisiae* strains expressing *SfATF(A)2*, *SfATF(B)2* and *SfATF(B)6* produced isoamyl acetate and phenylethyl acetate at high levels. Cellular localization analysis using GFP fusion revealed the cytoplasmic expression of *SfAtf* proteins, in contrast to the localization of *S. cerevisiae* Atf proteins in lipid particles. This is the first report on systematic characterization of *S. fibuligera* ATF genes and proteins responsible for high-level production of volatile acetate esters.

[Supported by NRF (Grant No. NRF-2017M3C1B5019295) and AFRA (Grant No. 918010042HD030)]

## C009

### **Extracellular Enzyme and Antimicrobial Activity of *Bacillus* sp. Isolated from Soil of Saryeoni Oreum**

Doseung Lee, Jong-Du Lee, Yoonji Lee, Kyoung Sik Yang, Jong Chul Lee, Weon-Jong Yoon, and Yong-Hwan Jung\*

*Biodiversity Research Institute, Jeju Technopark*

The purpose of this study was to discover industrial microorganisms with extracellular enzyme activity and antimicrobial activity from specific area in Jeju Island. The 16S rRNA analysis revealed that SRN-011 strain isolated from soil of Saryeoni Oreum exhibited 98.4% homology with *Bacillus pseudomycooides* NBRC 101232. As a result of the analysis of extracellular enzyme activity using the liquid culture media as a crude enzyme, protease activity was 522 unit/ml and cellulase activity was 140 unit/ml, but amylase activity and lipase activity showed low activity. Also, antibacterial activity was evaluated by disk diffusion method against pathogenic bacteria and the growth inhibition was observed *Staphylococcus aureus* and *Candida albicans*, respectively. Therefore, these results indicated that SRN-011 strain has shown the possibility of the application for the bioindustry.

[This work was supported by a research grant from Jeju Special Self-Governing Province.]

## C010

### **A Functional Genomic Approach to *Pseudomonas fluorescens* DR397 Reveals the Gene Networks Evolving Plant Drought Resistance**

Susmita Das Nishu, Jee Hyun No, and Tae Kwon Lee\*

*Department of Environmental Engineering, Yonsei University*

Drought poses a major threat to global food security by damaging crops. A few rhizospheric bacteria can promote plant drought tolerance by synergetic interaction yet the mechanisms in genome level are poorly understood. We investigated the genetic pathway of *Pseudomonas fluorescens* DR397 evolving plant drought resistance. The strain DR397 was isolated from soybean plant grown in dry region of Gangwon-do, Korea. Drought alleviation capacity in soybean was evaluated by pot test using direct inoculation of DR397 and proceeded for genomic and transcriptome analysis. *P. fluorescens* DR397 significantly promoted stem length, root length, fresh and dry weight (1.3, 1.7, 2.0 and 1.2 times than control) under drought. The genome analysis revealed gene clusters specific to Trehalose synthase (*TreS*), glutamate synthase (*ProBJ*), Proline importer (*OpuE*), ACC deaminase (*RimM*) reduce drought stress and tryptophan synthesis, P-solubilization (*PstABCS*), siderophore genes to promote plant growth. RNASeq supported that strain DR397 robust compatible solute synthesis to employ root exudates and organic materials as energy sources. The metabolic regulatory pathway of *P. fluorescens* DR397 to promote crop drought resistance give insight into plant-microbe interaction in extreme environments.

[Supported by grants from National Institute of Agricultural Sciences funded under RDA (PJ013176) and Strategic Initiative for Microbiomes in Agriculture and Food, MOA, Food and Rural Affairs, South Korea (No.918014-4)]

**C011****Ergosterol Pathway Engineering to Optimize Recombinant *Saccharomyces cerevisiae* Producing Cholesterol**Hye Yun Moon<sup>1</sup>, Hae Eun Park<sup>1</sup>, Dong Wook Lee<sup>1</sup>, Hui Jeong Jang<sup>2</sup>, and Hyun Ah Kang<sup>1\*</sup><sup>1</sup>Department of Life Science, College of Natural Science, Chung-Ang University, <sup>2</sup>Daewoong Pharmaceutical

Yeast produce ergosterol as a major sterol product, while animals produce cholesterol. In this study, several synthetic biology approaches were performed in *Saccharomyces cerevisiae* to develop recombinant yeast strains producing cholesterol. As the first step to block the ergosterol production, the yeast endogenous genes *ERG5* and *ERG6* were deleted with simultaneous insertion of two heterologous genes encoding 7-dehydrocholesterol reductase (*DHCR7*) and 3 $\beta$ -hydroxysterol D24-reductase (*DHCR24*). The codon-optimization of *Drosophila* *DHCR7* and *DHCR24* genes in two ways, codon usage (CU) and codon context (CC), revealed that the CC-optimized *DHCR24* increased protein stability, leading to the remarkably enhanced production of cholesterol compared to the CU-optimized *DHCR24*. Noticeably, HA tagging exerted a negative effect on the activity of *DHCR24*, thus drastically decreasing the conversion efficiency to cholesterol while accumulating several intermediates including zymosterol, dehydrodesmosterol, and desmosterol. Subsequently, multiple integration of CU-optimized *DHCR7* and *DHCR24* increased the cholesterol yield up to five folds. Additional introduction of genes involved in biosynthesis of ergosterol production, such as *tHMG1*, *ERG2*, *ERG3*, *ERG27*, and *UPC2-1*, into the cholesterol producing yeast further increased overall production of cholesterol and its precursor sterols, which are high-valued products with various industrial applications.

[Supported by Grant No. NRF2018R1A5A1025077]

**C012****Identification and Characterization of Bacteria Majorly Responsible for Metabolizing Aromatic Amino Acids in Human Gut**

Hye Su Jung, Kyung Hyun Kim, and Che Ok Jeon\*

Department of Life Science, Chung-Ang University

It has been reported that indoleacetic acid, cinnamic acid, and dopamine that are formed from the metabolisms of aromatic amino acids play important roles as ligands of aryl hydrocarbon receptors or neurobiological molecules. Metabolic bacteria and genes of aromatic amino acids have been partially studied based on cultured bacteria and major bacteria and genes in the human gut have been yet explored. Here, we used sequence similarity network and gene neighborhood analyses against public protein database and human metagenome database and found a new gene cluster responsible for aromatic amino acid metabolism in the human gut. The relative abundance of the representative genes of each cluster in human gut metagenome showed the novel group that may play a major role in aromatic amino acid metabolism. The novel group formed a distinct gene cluster from the previously known species group such as *Clostridium sporogenes*. Abundance analysis using public gut metagenome cohort data of human metagenome database showed that the abundance had a negative correlation with the occurrence of Crohn's disease, ulcerative colitis, liver cirrhosis, and non-alcoholic fatty liver disease. These findings suggest that the newly discovered groups of aromatic amino acid metabolizing bacteria may have a possibility as a new pharmabiotics to alleviate human diseases.

**C013****Longitudinal Evaluation of Fecal Microbiota Transplantation for Ameliorating Calf Diarrhea and Improving Growth Performance**

HyunSik Kim<sup>1</sup>, Tae Woong Whon<sup>2</sup>, Hojun Sung<sup>1</sup>, Yun-Seok Jeong<sup>1</sup>, Na-Ri Shin<sup>3</sup>, Dong-Wook Hyun<sup>1</sup>, Pil Soo Kim<sup>1</sup>, and Jin-Woo Bae<sup>1\*</sup>

<sup>1</sup>KyungHee University, <sup>2</sup>World Institute of Kimchi, <sup>3</sup>Korea Research Institute of Bioscience and Biotechnology

Calf diarrhea is associated with enteric infections, and also provokes the overuse of antibiotics. Therefore, proper treatment of diarrhea represents a therapeutic challenge in livestock production and public health concerns. Here, we evaluated the ability of an alternative to antibiotic treatment, fecal microbiota transplantation (FMT), to ameliorate diarrhea and restore gut microbial composition in growing calves. We transferred fresh feces ( $n = 20$ ) from six healthy donor calves to diarrheic calves. Age-matched diarrheic calves treated with saline ( $n = 14$ ) or antibiotics ( $n = 23$ ) were included as negative and treatment controls. We found that the FMT ameliorated the calf diarrhea based on Bristol stool score (BSS) with lower liquidity. To investigate the temporal development of the gut microbiota associated with diarrhea and FMT, we conducted multi-omics analyses of longitudinally collected 450 fecal samples and found that FMT-induced alterations in the gut microbiota (an increase of the family *Porphyromonadaceae*) and metabolomic profile (a decrease in fecal amino acid concentration) strongly correlated with the remission of diarrhea. During the continuous follow-up study over 24 months, we found that FMT further improved the growth performance of the cattle. This first FMT trial for ruminants suggest that the alterations in the gut microbiota may be useful for the treatment of diarrheic calves, and that FMT may have potential role of improvement of growth performance.

**C014****Longitudinality-based Improvement of Microbial Correlation for Rare Bacteria**

Ji-Won Huh and Jihyun Kim\*

*Department of Systems Biology, College of Life Science and Biotechnology, Yonsei University*

Most of microbial correlation analysis relies on the co-occurrence, requiring many samples to find significant correlations. However, diverse repertoire of microbiota and distinct host factors interfere to discern true correlation. Based on the fact that gut microbiome is longitudinally stable, we compared microbial correlations measured from longitudinal or cross-sectional data by using human metagenomic dataset from the Human Microbiome Project (HMP). Notably, correlations measured by intra-personal sampling were less spurious but consistent than those by random sampling.

We assumed that even if not detected, microbes may exist in samples that were temporally proximal to the microbes-detected samples. To exploit the longitudinality, pseudo-abundance was added to the microbe-deficient samples depending on proximity, which rescued the missed correlation that was likely to be related such as pre-documented partners in oral commensals or bacteriophages and preys. Furthermore, IBD or non-IBD biomarkers were grouped better in a correlation network with pseudo-abundance compared to original network.

Here, we showed that longitudinal approach greatly assists identification of true microbial correlation.

**C015****Determination of *Escherichia coli* O-serotype Using Real-time PCR**

Eun-Ji Shin, Jeong-Eun Kwak, Ho-Jeong Choi, Hoon-Jae Jeong, and Jo-Hoon Lee\*

*Department of Food Science and Biotechnology, Graduate School of Biotechnology, Kyung Hee University*

O-Antigens are part of the lipopolysaccharide which constitutes the outer layer of Gram-negative bacteria. O-Antigens are responsible for classifying *Escherichia coli* strains and determining the O-serotype. *E. coli* O-serotype is important for tracking the source of foodborne illness for epidemiological studies. Conventional serotyping was performed based on the agglutination reaction between the O-antigen and antisera. This method is a delicate, difficult, time-consuming, and expensive procedure. Therefore, the purpose of this study was to develop a fast, accurate, and economic serotype identification method using Real-time PCR (RT-PCR) with high specificity and a low detection limit. Target genes from 11 serotypes (O6, O25, O26, O55, O78, O103, O104, O111, O125, O145, and O157) causing the high frequency of food poisoning were selected and determined O-serotype to design primers using pan-genome analysis. The *wzy* gene was selected as a target gene. Primer sets were designed and were verified using singleplex, cross-check, multiplex PCR to clarify if they amplify properly the specific target genes. For the improvement of rapid serotyping, singleplex RT-PCR and Multiplex RT-PCR were developed using primer sets with new probes for serotyping in 30 min. Therefore, this PCR and RT-PCR methods for rapid *E. coli* serotyping provided the rapid, accurate, and robust serotyping which may substitute the inaccurate and expensive antiserum serotyping.

**C016*****Streptococcus kimchii* sp. nov., Isolated from Pogi-Kimchi, a Traditional Korean Fermented Food**

Ye-Jin Jung, Do-Hee You, and Ju-Hoon Lee\*

*Department of Food Science and Biotechnology, Graduate School of Biotechnology, Kyung Hee University*

The strain ZB199<sup>T</sup> (ZB) was isolated from Korean traditional fermented food. 16S rRNA gene sequencing using BLASTN showed that strain ZB199<sup>T</sup> had similarities to *S. parasanguinis* ATCC 15912<sup>T</sup> (97.27%), *S. rubneri* LMG 27207<sup>T</sup> (96.99%), and *S. australis* ATCC 700641<sup>T</sup> (96.78%). Phylogenetic tree analysis based on 16S rRNA sequence and multilocus sequence type analysis of concatenated partial *atpA*, *sodA*, *pheS*, *tuf*, *gki* gene sequences revealed that ZB was closed to those of strains which showed high identity score with ZB. However, PCR fingerprints techniques showed that strain ZB could be separated from its nearest phylogenetic relatives in the molecular level. The biochemical identification of ZB from the closest type strains was performed by API and VITEK analyses. The phenotypic characteristics of the strain was distinguished from other *Streptococcus* species with respect to carbohydrate fermentation and enzyme activity. In order to comparative genome analysis with *S. parasanguinis* ATCC 15912<sup>T</sup>, which showed the closest relationship, the complete genome sequencing was conducted by PacBio. The complete genome of ZB was 2.1 Mb. The comparative genome analysis revealed that the ZB had more genes associated with phenotypic analyses results than *S. parasanguinis* ATCC 15912<sup>T</sup>. It suggested that ZB was discriminated from *S. parasanguinis* ATCC 15912<sup>T</sup>, which showed 3.5% difference in ANI results. Based on these results, strain ZB was classified as a novel species of the genus *Streptococcus*.

**C017****Metagenomic and Statistical Study of Clinical Effects on Inflammatory Bowel Disease Using Synbiotics**You-Tae Kim<sup>1</sup>, Joon-Gi Kwon<sup>1</sup>, Jo-Seph Lee<sup>1</sup>, Nam-Su Oh<sup>2</sup>, Yon Ho Choe<sup>3</sup>, and Ju-Hoon Lee<sup>1\*</sup>

<sup>1</sup>Department of Food Science and Biotechnology, Institute of Life Sciences and Resources, Kyung Hee University, <sup>2</sup>Department of Food and Biotechnology, Korea University, <sup>3</sup>Department of Pediatrics, Samsung Medical Center, Sungkyunkwan University School of Medicine

Inflammatory bowel disease (IBD) is a common disease caused by intestinal inflammation with high fat and low fiber diets. Notably, scientific evidence supports that IBD may result from inappropriate immune response from the compositional balance in the gut microbiota. For control of IBD, synbiotics treatment has been suggested as an efficient approach to modulate the gut microbiota. The synbiotics were prepared using *Lactobacillus gasseri* 505 fermented with *Cudrania tricuspidata* leaf extract in milk. For IBD patients, 82 patients were recruited and randomly divided into control group and synbiotics group. The feeding was for six weeks and additional six weeks were for washout period. The samples for gut microbiome analysis were collected at the start of feeding (Visit 1), right after the termination of feeding (Visit 2), and after six weeks of washout period (Visit 3). Metagenomic analysis showed that the diversity of gut was significantly different only between Visit 1 and Visit 3 of synbiotics group. In the change of bacterial composition, beneficial bacteria including *Bifidobacterium* increased and potential pathogens were reduced. Interestingly, these beneficial bacterial and potential pathogens showed negative and positive correlation, respectively, with ESR (erythrocyte sedimentation rate), CRP (C-reactive protein), and Calprotectin which are indication of marker for inflammation.

**C018****Targeted Delivery of the Mitochondrial Target Domain of Noxa to Tumor Tissue via Synthetic Secretion System in *E. coli***Daejin Lim<sup>1,2</sup>, Woong Chae Jung<sup>3</sup>, Ji-Hyeon Lee<sup>3</sup>, and Miryoung Song<sup>3\*</sup>

<sup>1</sup>Department of Microbiology, Chonnam National University Medical School, <sup>2</sup>Department of Molecular Medicine, Chonnam National University Graduate School, <sup>3</sup>Department of Bioscience and Biotechnology, Hankuk University of Foreign Studies

Targeted delivery of drugs is a key aspect of the successful treatment of untreatable diseases such as tumors. In the pursuit of accurate delivery with high specificity and low size limit for peptide drugs, a synthetic type 3 secretion system (T3SS) has been rebuilt from a native genetic system encoded in *Salmonella* pathogenicity island-1 (SPI-1) with no virulence effectors. Here, we tested the potential of synthetic T3SS as drug delivery machinery for the peptide-based drugs. First, the genetic system for synthetic T3SS was introduced into non-native host *E. coli*, which was chosen for safety due to the lack of *Salmonella*-driven virulence factors and for the modularity test. And, the mitochondrial targeting domain (MTD) of Noxa was tested as a cargo protein with anti-tumor activity. To this end, the gene encoding MTD was engineered for secretion through synthetic T3SS, resulting in the tagged MTD at the N-terminus. When *E. coli* carrying synthetic T3SS and MTD on plasmids was administered into tumor-bearing mice, MTD with a secretion tag at the N-terminus was clearly detected in the tumor tissue after induction. Also, the tumor growth and mortality of tumor-bearing animals were mitigated by the delivered MTD with its cytotoxic activity. Thus, this work potentiates biotherapeutic bacteria for incurable diseases including tumors by implanting a dedicated delivery system.

**C019****Purification and Characterization of Type II Restriction Endonuclease (*Fisl*) from Thermophilic Bacterium *Fervidobacterium islandicum* AW-1**

Dariimaa Ganbat<sup>1</sup>, Bo Gyoung Choi<sup>1</sup>, A Young Jin<sup>1</sup>, Dong-Hoon Won<sup>1</sup>, Dong-Woo Lee<sup>2</sup>, Seong-Bo Kim<sup>3</sup>, and Sang-Jae Lee<sup>1\*</sup>

<sup>1</sup>Major in Food Biotechnology and Research Center for Extremophiles & Marine Microbiology, Silla University,

<sup>2</sup>Department of Biotechnology, Yonsei University, <sup>3</sup>Bio-Living Engineering Major, Global Leaders College, Yonsei University

Identification of Restriction Modification (R-M) system is important for development of genetic tools. Genome sequence analysis of thermophilic *Fervidobacterium islandicum* AW-1 revealed a presence of a *DpnII*-like restriction endonuclease. Gene (*fisl*) encoding a type-II restriction endonuclease (*Fisl*) of R-M system was cloned and the recombinant enzyme was expressed in *Escherichia coli* with His-tag at the N-terminus. The open reading frame of the *fisl* gene consists of 930 bp that encodes a protein of 310 amino acid residues with a calculated molecular mass of 36 kDa. Artificial DNA substrate containing two 5'-GATC-3' recognition sites was amplified from plasmid vector pUC19 and used as substrate. *Fisl* exhibited a broad pH optimum range between 5.0 to 10.0 and was active at temperature range between 65-70°C. Manganese and cobalt could replace magnesium as a cofactor for activity. MgCl<sub>2</sub> optimum concentration required for enzyme activity was 5-10 mM. Recombinant *Fisl* was stable at 70°C for more than two hours and at 75°C for 30 min. *Fisl* has been identified as a heat stable isoschizomer of type II restriction endonucleases, *DpnII* and *Sau3AI*. It is a novel thermostable type II endonuclease enzyme from *Fervidobacterium islandicum* AW-1.

**C020****A Research on the Diversity and Enzyme Productivity of Halophilic Microorganisms Isolated from the Soil around the Ranch for the Exploration of Fermentation Strains**

Yujeong Yeom<sup>1</sup>, Ga Eul Jeong<sup>1</sup>, Min-Seok Jeong<sup>1</sup>, Hae Rang Lee<sup>1</sup>, Seung-Yeon Yoo<sup>1</sup>, Yong-Jik Lee<sup>2</sup>, Seok-Cheol Cho<sup>3</sup>, Han-Seung Lee<sup>1</sup>, and Sang-Jae Lee<sup>1\*</sup>

<sup>1</sup>Major in Food Biotechnology and Research Center for Extremophiles & Marine Microbiology, Silla University,

<sup>2</sup>Department of Bio-cosmetics, Seowon University, <sup>3</sup>Food science & Engineering Major, Seowon University

The diversity and characterization of microorganisms isolated from the soil around several ranches in Korea were confirmed in this study. To isolate halophilic microorganisms, the marine agar medium was basically used and cultivated at 37°C for several days. After single colony isolation, a total of 116 pure colonies were isolated and phylogenetic analysis was carried out based on the result of 16S rDNA sequencing, indicating that isolated strains were divided into 4 phyla, 23 families, 30 genera and 51 species. To confirm whether isolated strain can be a candidate for the fermentation of diverse food ingredients, amylase, lipase, and protease enzyme assays were performed individually, showing that 92 strains possessed at least one enzyme activity. Especially 4 strains, identified to *Jonesia quinghaiensis* (isolate name: JSF 19-2), *Halomonas alkaliantarctica* (isolate name: JSF 21), *Bacillus velezensis* (isolate name: NWFY-36), and *Staphylococcus capitis* subsp. *urealyticus* (isolate name: MSY-5), showed all enzyme activity tested. Moreover, 17 strains showed the ability for auxin production. Therefore, this study has shown the potential to secure domestic biological resources and to use them to apply them to the food and feed industry.

**C021**

**A Study of the Diversity and Profile for Enzyme Production of Aerobically Cultured Halophilic Microorganisms from the Various Solar Salterns**

Ga Eul Jeong<sup>1</sup>, YuJeong Yeom<sup>1</sup>, Ji O Kim<sup>1</sup>, Joo Young Yang<sup>1</sup>, Yong-Jik Lee<sup>2</sup>, Gae-Won Nam<sup>2</sup>, Dong-Woo Lee<sup>3</sup>, Han-Seung Lee<sup>1</sup>, and Sang-Jae Lee<sup>1\*</sup>

<sup>1</sup>Major in Food Biotechnology and Research Center for Extremophiles & Marine Microbiology, Silla University,

<sup>2</sup>Department of Bio-cosmetics, Seowon University, <sup>3</sup>Department of Biotechnology, Yonsei University

This research confirmed the diversity and characterization of halophilic microorganisms isolated from the various solar salterns, collected on the inside and outside of the country. To isolate strains, the marine agar medium was basically used and cultivated at 37°C for several days aerobically. After single colony isolation, a totally of 230 pure colonies were isolated and phylogenetic analysis was carried out based on the result of 16S rDNA sequencing, indicating that isolated strains were divided into 4 phyla, 12 families, 27 genera and 64 species. Firmicutes phylum, the main phyletic group, comprised 89.6% with 3 families, 17 genera and 52 species of Bacillaceae, Staphylococcaceae and Carnobacteriaceae. To confirm whether isolated strain can produce industrially useful enzyme or not, amylase, lipase, and protease enzyme assays were performed individually, showing that 177 strains possessed at least one enzyme activity. Especially 17 strains showed all enzyme activity tested. This result indicated that isolated strains have shown the possibility of the industrial application. Therefore, this study has contributed to securing domestic genetic resources and the expansion of scientific knowledge of the halophilic microorganism community in solar salterns.

**C022**

**Anti-tumor Effects of an Immunotoxin by a Non-inducible Promoter in *Salmonella enterica* serovar Gallinarum**

Mai Thu Phuong<sup>1,2</sup>, Daejin Lim<sup>1,2</sup>, Tran Quang Thanh<sup>1,2</sup>, and Jae-Ho Jeong<sup>1,2\*</sup>

<sup>1</sup>Department of Microbiology, Chonnam National University Medical School, <sup>2</sup>Department of Molecular Medicine (BK21plus), Chonnam National University Graduate School

The efficient anticancer therapeutics relies on a mode of action of protein drugs, which are specifically released from bacteria to tumors and can eliminate cancer cells by its toxicity. However, the remedial proteins require strict regulation of gene expression in tumorigenic and healthy tissues due to risky systemic effects. *rrnB P1* promoter, comprising a *Fis* protein-binding upstream activation region (UAR), has been found significantly activated in *Escherichia coli* MG1655 chromosome in the exponential growth phase *in vitro* and in the tumor *in vivo*. It is proved that the TGF $\alpha$ -PE38 immunotoxin under the control of the non-inducible *rrnB P1* promoter carried by the  $\Delta$ ppGpp *Salmonella enterica* serovar Gallinarum (SG $\Delta$ ppGpp) had an effective anti-tumor influence on some mouse tumor models.

**C023****Bacterial Cancer Therapy Using a Modified Fowl-adapted *Salmonella enterica* serovar Gallinarum**Daejin Lim<sup>1,2</sup>, Hyung-Ju Lim<sup>1,3</sup>, Eun A So<sup>1,3</sup>, Ha Young Kim<sup>1,3</sup>, Miryoung Song<sup>4</sup>, Hyon E. Choy<sup>1,2</sup>, and Jae-Ho Jeong<sup>1,2,3\*</sup>

<sup>1</sup>Department of Microbiology, Chonnam National University Medical School, <sup>2</sup>Department of Molecular Medicine(BK21plus), Chonnam National University Graduate School, <sup>3</sup>Medical Research Center (MRC) - Combinatorial Tumor Immunotherapy Research, <sup>4</sup>Department of Bioscience and Biotechnology, Hankuk University of Foreign Studies

During last two decades, bacterial cancer therapy using live attenuated bacteria has emerged as a promising alternative medicine. Here, we report novel anticancer bacteria based on an avian host specific serotype *Salmonella enterica* serovar Gallinarum in which ppGpp synthesis has been disabled. The bioluminescent DppGpp *S. gallinarum* were injected intravenously into mouse bearing grafted tumors. Strong bioluminescent signals from this bacteria were detected in all of these grafted tumor exclusively two days after the injection which decreased gradually as time passed, as determined by *in vivo* imaging system. Anticancer effect of DppGpp *S. gallinarum* injected through intravenous route was examined using the BALB/c mice with grafted CT26 colon cancer cells. A superb anticancer effect was observed which could be ascribed to the reduced virulence that allowed administration of greater dose than with other attenuated strains of *S. typhimurium*. Superior advantage of *S. gallinarum* in cancer therapy, therefore, would include i) the avian host specific serotype *S. gallinarum* should be safer than *S. typhimurium* to use in human as well as in rodents for treatment and analysis, ii) *S. gallinarum* poses no health threat to the persons handling it in laboratories and clinics, iii) using the balanced-lethal host-vector system of *glmS* gene of *S. gallinarum*, those plasmids encoding any anti-tumor protein gene can be stably maintained in the tumor-associated *S. gallinarum* for clinical use.

**C024****Removal of Malodorous Gases from Pig Manure by Bacterial-surfactant Foam**

Thi Tuyet Nhan Le and Jaisoo Kim\*

Ecology Laboratory, Department of Life Science, College of Natural Sciences, Kyonggi University

Livestock and poultry production farms emit large amounts of greenhouse gases, including most nitrogen-containing compounds, sulfur-containing compounds, and volatile fatty acid compounds. In which, ammonia and hydrogen sulfide are the major odors found in the emissions of these farms. In this study, the ammonia-oxidizing bacteria strains and hydrogen sulfur-oxidizing strains isolated from animal manure which were applied as a bacterial-surfactant foam type to prevent the release of malodorous gases and reduction of their concentration emitted from pig manure. Results have shown that initial ammonia and the hydrogen sulfide concentration in the pig manure of the experiment was about 500 ppm and 60 ppm, respectively, they were reduced to 0 ppm after 15 h of foam spray with ammonia and 12 h with hydrogen sulfide.

[Supported by grants from NRF (2020M3F8A1080198)]

**C025****Isolation Strategy to Enhance Antibiotic Discovery from Uncultured Soil Bacteria**

Ngoc Hoang Trinh and Jaisoo Kim\*

*Department of Life Science, College of Natural Sciences, Kyonggi University*

The Multi-drug resistant (MDR) bacteria is one of the most medical emergency problems currently facing mankind and unfortunately, researchers have struggled to identify new antibiotic drugs. The pursuit of new antibiotic compounds in bacterial resources from traditional cultivation method often results in a high rediscovery rate of recognized compounds and a low chance for detection of truly novel structure. Consequently, in the 1990s the pharmaceutical industry companies abandoned the traditional ways in pursuit of antibiotics.

Most of the currently available antibiotics are originated from soil bacteria which are abundance, but more than 99% of these bacteria cannot be cultured by traditional cultural techniques. To overcome this difficulty, our researches modified traditional cultivation methods and developed new cultivation methods such as transwell plate, diffusion bioreactor, and new soil extraction. The recovery rate of isolation efficiency in each method increases 20%, 30% and 49% respectively

[This work was supported by a research grant NRF 2019R1F1A1058501]

**C026****Occurrence and Inhibition of White Colony-forming Yeast in Kimchi Fermentation**Seong Eun Kang<sup>1,2</sup>, Mi-Ju Kim<sup>1</sup>, Sung-Gi Min<sup>1</sup>, Sung Wook Hong<sup>1</sup>, Hyelyeon Hwang<sup>1</sup>, Seong Woon Roh<sup>1</sup>, Deok-Young Jhon<sup>2</sup>, and Tae-Woon Kim<sup>1\*</sup>*<sup>1</sup>Research and Development Division, World Institute of Kimchi, <sup>2</sup>Division of Food and Nutrition, Chonnam National University*

The presence of white colony-forming yeast (WCFY) on kimchi surfaces indicates a reduction in kimchi quality. This study aimed to investigate the effect of different fermentation temperatures (4, 10, and 20°C) and packaging conditions (open or closed) on WCFY diversity. Community analysis revealed that *Kazachstania servazzii* and *K. barnettii* were most prevalent in kimchi fermented under closed packaging condition at 4, 10, and 20°C. In open packaging condition, *Candida sake*, *K. servazzii*, *K. barnettii*, and *Tausonia pullulans* were the predominant yeast species at 4°C, and *C. sake*, *K. servazzii*, *K. barnettii*, and *Debaryomyces hancenii* were predominantly detected at 10°C. The diversity of the WCFY community was higher under the open rather than the closed packaging condition. These results indicate that fermentation temperature and air exposure can alter WCFY diversity on kimchi surface. In addition, antimicrobial activities of grapefruit seed extract (GSE) against WCFY was investigated to inhibit WCFY growth on kimchi surface. The GSE showed high antimicrobial activity against 4 WCFY strains except for *C. sake* on an agar well diffusion assay. However, when GSE was applied to kimchi inoculated with WCFY strains respectively, the inhibition of growth of WCFY was not observed. It seems that further study is needed to select new natural preservatives with antimicrobial activity against WCFY.

[Supported by grants from IPET (119118-01) and WIKIM (KE2001-2)]

**C027****Value Evaluation of Actinobacterial Resource Based on Microbial Characteristics**

Chun-Zhi Jin<sup>1</sup>, Jong Min Lee<sup>1</sup>, Min-Kyoung Kang<sup>1,2</sup>, So Hee Park<sup>1,3</sup>, Dong-Jin Park<sup>1</sup>, and Chang-Jin Kim<sup>1,3\*</sup>

<sup>1</sup>Industrial Bio-material Research Center, Korea Research Institute of Bioscience and Biotechnology, <sup>2</sup>College of Pharmacy, Chungnam National University, <sup>3</sup>Department of Bio-Molecular Science, KRIBB School of Bioscience, Korea University of Science and Technology (UST)

Actinomycetes, as a producer of diverse secondary metabolites which have the primary importance in medicine, cosmetic, agriculture and food production until now. Though the utilization and application of these actinomycetes were still insufficient due to less information. It is necessary to evaluate microbial resources so that the researchers can do further experiments quickly and efficiently as possible without screening work.

The purpose of this study is to support and help researchers through infrastructure construction and quantitative analysis of microbes. We focus on 16S rRNA sequencing for taxonomic analysis, enzyme (protease, amylase, lipase, cellulase) assay, physiological characters of each stain (growth temperature, media pH, salt tolerance) and LC/MS profiles of culture broth. We have more than 15,000 actinomycetes' information and culture broth could be supplied. It is helpful for high-throughput screening and speed up experiments.

Visit us: <http://mrsc.net/>.

[This research was supported by a grant (NRF-2013M3A9A5076601) from a study on the strategies of improving the value of microbial resources funded by Ministry of Science and ICT of the Korea Government and a grant from KRIBB Research Initiative Program]

**C028****Biosorption of Copper(II) in Aqueous Solution Using *Bacillus* sp. SRCM112835**

Jinwon Kim, Hee-Jong Yang, Gwangsu Ha, Su-Ji Jeong, Sua Im, Su-Jin Shin, Myeong-Seon Ryu, Ji-Won Seo, and Do-Youn Jeong\*

*Microbial Institute for Fermentation Industry (MIFI)*

In this study, biosorption of the Copper(II) by *Bacillus* sp. SRCM 112835 (GenBank accession No. MT626036) in aqueous solution was investigated. *Bacillus* sp. SRCM 112835 was isolated from *Doenjang* (Korean Fermented Soy Paste). Isolated strain effectively removed 30% of the Copper(II) from a 52 mg/L within 1 h. The properties of the *Bacillus* sp. SRCM 112835 were investigated by Fourier transform infrared spectroscopy (FT-IR), point of zero charge (pH<sub>pzc</sub>), and phylogenetic analysis. Furthermore, the influence of initial pH (2.08–9.98) and biomass dosage (0.005–0.07 g) in Copper(II) biosorption were investigated. Isotherm and kinetic experiment results suggest that the Langmuir isotherm and pseudo-second-order kinetic models well fitted the experimental data, respectively. The biosorption experiments showed that *Bacillus* sp. SRCM 112835 could be used as effective biosorbent for the Copper(II) biosorption in aqueous solution.

[This research was supported by Traditional Culture Convergence Research Program through the National Research Foundation of Korea (NRF) funded by the Ministry of Science and ICT (NRF-2016M3C1B5907049).]

**C029****Comparison of Microbial Community Using Traditional Fermented Doenjang and Gochujang Products Produced in Korea**

Su A Im, Gwangsu Ha, Su-Jin Shin, Jinwon Kim, Hee-Jong Yang, and Do-Youn Jeong\*

*Microbial Institute for Fermentation Industry (MIFI)*

In the present study, the diversity of microbial population of Doenjang and Gochujang (Korean fermented products) in Korea area was investigated. Microbial communities were analyzed using Illumina 16S metagenomic sequencing protocol. The 16S rRNA gene (V3-V4 region) phlotypes were classified at the genus, family order, class, and phylum levels. Diversity was measured by inverse Simpson, Shannon index, richness and evenness were measured by the number of observed the operational taxonomic units (OTUs) using CD-HIT. The sequences were analyzed to four phylum (*Firmicutes*, *Proteobacteria*, *Cyanobacteria*, *Actinobacteria*). In this result, the dominant phylum of doenjang (85.05%) and gochujang (74.46%) were *Firmicutes*. The *Bacillus* was the most dominant in both doenjang (39.33%) and gochujang (61.38%) in genus level. *Bacillus velezensis* was the most dominant bacteria of doenjang (30.81%). But, most dominant of gochujang, *Bacillus subtilis* was 49.04%, which was significantly different from doenjang. Therefore, these results showed significant differences microbiota between types of traditional foods.

**C030****Isolation of *Bacillus subtilis* Having Antifungal Activity against Brown Spot and Sheath Rot of Rice**

Myeong Seon Ryu, Ji-won Seo, Su-ji Jeong, Hee-Jong Yang, and Do-Youn Jeong\*

*Microbial Institute for Fermentation Industry (MIFI)*

Brown spot and sheath rot of rice are caused by the pathogens such as *Curvularia lunata*, *Cochliobolus miyabeanus* and *Sarocladium oryzae*, and cause losses such as reduced rice yield and quality, which is an enormous problem with serious long-term effects. Therefore, five kinds of useful *Bacillus*-like isolates which are excellent in extracellular enzyme activity and produce siderophore were selected from paddy soil of Sunchang in Korea for biological control of phytopathogenic fungi. And then, five selected isolates had excellent antifungal activity against three of the phytopathogenic fungi that frequently occur in rice, and JSRB 177 strain had the most excellent antifungal activity. Based on the experimental results, JSRB 177 is finally selected as a strain for biological control and identified to *Bacillus subtilis* through 16S rRNA sequence analysis. In addition, physiological characteristics of JSRB 177 confirmed by analysis of carbohydrate fermentation patterns and enzyme production ability. In the future, further studies related to industrialization such as port test and establishment of mass production process are needed, but based on the above results, JSRB 177 is expected to be used as a biological control agent for the rice pathogenic fungi.

[This research was supported by Traditional Culture Convergence Research Program through the National Research Foundation of Korea (NRF) funded by the Ministry of Science and ICT (NRF-2016M3C1B5907049)]

**C031****Structural Insights into Thioredoxin-thioredoxin Reductase System of Thermophilic Feather Degrading *Fervidobacterium islandicum* AW-1 Reveals Its Plausible Role in Keratinolysis**Immanuel Dhanasingh<sup>1</sup>, Dong-Woo Lee<sup>2</sup>, and Sung Haeng Lee<sup>1\*</sup><sup>1</sup>Department of Cellular and Molecular Medicine, Chosun University School of Medicine, <sup>2</sup>Department of Biotechnology, Yonsei University

Reduction of disulfide bridges in complex feather keratin is supposed to be the prerequisite for microbial keratin degradation. In accordance with that, to unveil the insights of the electron transfer between feather keratin and thioredoxin system in thermophilic keratin-degrading *F. islandicum* AW-1, the structural studies of *FiTrxR* and *FiTrx* were implemented. The genes corresponding to *FiTrxR* and *FiTrx* were cloned, expressed, purified and crystallized. The crystal structures of *FiTrxR* and *FiTrx* were determined at 2.45 Å and 1.25 Å, respectively. The *FiTrxR* structure existed as a homodimer (~70 kDa) with each monomer composed of NADPH and FAD binding domains bound to FAD co-factor. Interestingly, the chains in the homodimer existed in two different conformations: Flavin oxidized (FO) state and Flavin Reduced (FR) state. The presence of calcium metal ion at the dimer interface is unique feature in *FiTrxR*. Furthermore, *FiTrx* structure contained typical thioredoxin fold comprising the CXXC motif in the oxidized state and located at the surface for easy binding with *FiTrxR*. To conclude, we propose *FiTrx* to be a mediator in electron transfer cycle by initially reducing the disulfide bonds from the feather fragments, followed by accepting proton from *FiTrxR* during the thioredoxin cycle. Thus, our study facilitates in deeper understanding of the redox chemistry involved during the reduction of disulfide bridges in keratin.

**C032****Structural Insights of Cytochrome *c<sub>L</sub>* from the Aquatic Methylophilic Bacterium *Methylophilus aminisulphidivorans* MP<sup>T</sup> Reveals Distinctive Features for Efficient Electron Transfer during Methanol Oxidation**Suparna Ghosh<sup>1,2</sup>, Immanuel Dhanasingh<sup>1,2</sup>, Jaewon Ryu<sup>4</sup>, Si Wouk Kim<sup>3,4</sup>, and Sung Haeng Lee<sup>1,2\*</sup><sup>1</sup>Department of Cellular and Molecular Medicine, Chosun University School of Medicine, <sup>2</sup>Department of Biomedical Sciences, Graduate School of Chosun University, <sup>3</sup>Department of Environmental Engineering, Chosun University, <sup>4</sup>Department of Energy Convergence, Graduate School of Chosun University

Cytochrome *c<sub>L</sub>* (Cyt<sub>c<sub>L</sub>) is an abundant periplasmic protein, takes part in methanol oxidation process in methylophilic, in which electron transfer (ET) take place from the pyrroloquinoline (PQQ) cofactor of the methanol dehydrogenase (MDH) to Cyt<sub>c<sub>L</sub>. Due to lack of structural information, the direct ET between the MDH and Cyt<sub>c<sub>L</sub> is still elusive. In this study we determine the first crystal structure of Cyt<sub>c<sub>L</sub> from the marine methylophilic bacterium *Methylophilus aminisulphidivorans* MP<sup>T</sup> at 2.13 Å resolution. The distinctive features include the presence of three metal ion binding sites for Na<sup>+</sup>, Ca<sup>+</sup>, and Fe<sup>2+</sup> were found, wherein the ions mostly formed coordination bonds with the amino acid residues on the loop (G93-Y111) that interacts with heme. These ions seemed to enhance the stability of heme insertion by increasing the loop's steadiness. The basic N-terminal end, together with helix α4 and loop (G126 to Y136), contributed positive charge to the region. In contrast, the acidic C terminal end provided a negatively charged surface, yielding several electrostatic contact points with partner proteins for electron transfer. These exceptional features of Ma-Cyt<sub>c<sub>L</sub>, along with the structural information of MDH, led us to hypothesize the need for an adapter protein bridging MDH to Cyt<sub>c<sub>L</sub> within appropriate proximity for ET. With this knowledge in mind, the methanol oxidation complex reconstitution in vitro could be utilized to produce metabolic intermediates at the industry level.</sub></sub></sub></sub></sub></sub>

**C033**

**Genomics Analysis and Functional Characterization of *Debaryomyces hansenii* Isolated from Korean Soybean Fermented Food**

Da Min Jeong, Su Jin Yoo, Min-Seung Jeon, Byung Hee Chun, Che Ok Jeon, Seong-il Eyun, Young-Jin Seo and Hyun Ah Kang\*

*Department of Life Science, Chung-Ang University, Seoul*

Fermented soybean products have been getting the spotlight in the international market due to their nutritive values and many health benefits. During soybean fermentation, yeasts play salient roles in the production of diverse flavor compounds that are important in determining the quality of the soybean products. In this study, genome analysis and physiological characterization were performed for *Debaryomyces hansenii* KD2 and C0-11-Y2, which were isolated from the traditional Korean fermented soybean product, 'Jang'. Both *D. hansenii* 'Jang' strains showed much higher halotolerance than the conventional yeast *Saccharomyces cerevisiae*. Especially, the growth of *D. hansenii* KD2 and C0-11-Y2 was improved in the presence of salts, indicating that they are halophilic. The ploidy analysis by FACS and whole genome sequencing revealed that the genome of strain KD2 is haploid with the size of approx. 13 Mb, whereas for the strain C0-11-Y2, it appeared to be diploid with the size of 26 Mb. Whereas the *D. hansenii* strains produced flavors of butter, caramel, and cheese in common, but they also exhibited different flavor profiles in the SPME-GC/MS analysis. Besides the viability in the presence of bile salt and at low pH, *D. hansenii* KD2 and C11 showed the immune-modulating activity to induce higher levels of IL-10, an anti-inflammatory cytokine, than the established probiotic yeast *Saccharomyces boulardii*. Altogether, our results strongly indicated that the *D. hansenii* 'Jang' strains with an enhanced halotolerance, a fine flavor, and a probiotic activity, have high potential to be used either as a fermentation starter or as a probiotic candidate.

**D001****Skin Microbiome on the Hypertrophic Scars of Burn Patients: a Pilot Study**

Cheong Hoon Seo, So Young Joo, and Yoon Soo Cho\*

*Department of Rehabilitation Medicine, Burn Center, Hangang Sacred Heart Hospital, Hallym University College of Medicine*

Hypertrophic scar formation following burn injury might be associated with the changes in the diversity and distribution of skin flora. We investigated the distribution of skin microbiome in burn patients. Ten burn patients who did not have taken antibiotics or applied to the skin within one month were targeted. Swab for 16S rRNS based next-generation sequencing of the skin microbiome and biomechanical properties evaluation were performed on burn scars and normal skin around the burn scars. Hypertrophic burn scars showed higher mean values of erythema ( $P < 0.001$ ) and Uv/Ue (viscoelasticity,  $P = 0.005$ ), and lower mean values of trans-epidermal water loss ( $P = 0.001$ ) and Uf (distensibility,  $P < 0.001$ ). *Cellulomonas chitinilytica*, *Sanguibacter keddieii*, and *Bacillus hisashii* species were more on burn scars. In particular, the five microbial species of *Cellulomonas chitinilytica*, *Sanguibacter keddieii*, *Micrococcus aloeverase*, *Haemophilus parahaemolyticus*, and *Hydrogenophilus thermoluteolus* showed significant differences in the distribution of burn scars and normal skin ( $P = 0.001$ ,  $P = 0.041$ ,  $P = 0.013$ ,  $P = 0.015$ ,  $P = 0.05$ ). There was a significant difference in the community distribution ratio of skin microbiome on burn scars and normal skin around the burn scars.

[This research was supported by Basic Science Research Program through the National Research Foundation of Korea (NRF) funded by the Ministry of Education (2020R111A3074150).]

**D002****Profiles of Antimicrobial Resistance and Enterotoxigenicity of Non-aureus Staphylococci Isolated from Retail Chicken Meat**

Sun Do Kim, Ji Heon Park, Soo In Lee, and Soo-Jin Yang\*

*School of Bioresources and Bioscience, Chung-Ang University*

*Staphylococcus aureus* and non-aureus staphylococci (NAS) are commensal colonizers of the skin and mucous membranes of humans and farm animals. Staphylococcal food poisoning (SFP) results from ingestion of foods contaminated with staphylococcal enterotoxins (SEs) produced by staphylococci. In addition to their enterotoxigenic potential, antimicrobial resistant (AMR) staphylococci have frequently been detected in retail meat samples. In this study, we investigated i) the profiles of NAS species in 129 retail chicken meat samples collected in Korea, ii) prevalence of 19 different SE genes in the NAS, and iii) AMR phenotypes and genetic factors associated with the AMR phenotypes. Seven different species of 62 NAS (62/162, 38.3%) were isolated from raw chicken meat samples: *S. hyicus* ( $n = 29$ , 46.8%), *S. saprophyticus* ( $n = 13$ , 20.9%), *S. sciuri* ( $n = 7$ , 11.3%), *S. simulans* ( $n = 6$ , 9.7%), *S. warneri* ( $n = 3$ , 4.8%), *S. lentus* ( $n = 2$ , 3.2%), and *S. epidermidis* ( $n = 1$ , 1.6%). Forty-five NAS (72.6%) carried at least one SE gene, 28 (62.2%) of which were positive for multiple SE genes. The most prevalent SE genes in the 62 NAS were *seh* (35.5%), *sep* (25.8%), and *sej* (19.4%). Although none of the 62 NAS displayed ceftiofur, relatively high levels of resistance to fusidic acid (54.8%) and tetracycline (38.7%) were observed.

Of note, 16 *S. hyicus*, 1 *S. lentus*, and 1 *S. simulans* displayed quinolone-resistant phenotype, which were usually associated with point mutations in *gyrA* (S84L) and *grrA* (S80L) genes. These results suggest that NAS, especially *S. hyicus* strains, in raw chicken meat may serve as a source for transmission of antimicrobial resistance and SE genes.

[Supported by Research of Korea Centers for Disease Control and Prevention (Project No. 2017NER54060 and 2020ER540500 to S.J.Y)]

**D003****One Health Approach for Investigation of Livestock-associated Methicillin-Resistant *Staphylococcus aureus* (LA-MRSA) in Broiler Chickens and Their Environments throughout the Chicken Meat Production System**

Gi Yong Lee, Haeng Ho Lee, Hong Sik Eom, and Soo-Jin Yang\*  
*School of Bioresources and Bioscience, Chung-Ang University*

Occurrence of LA-MRSA has recently been reported in human patients via direct or indirect contact with infected animals. MRSA has frequently been isolated from mucosal surfaces of humans and animals, foods of animal origin, and farm environment, causing a significant threat to public health. Thus, a holistic One Health approach involving farm workers, animals, and the environment is necessary to investigate prevalence and transmission of LA-MRSA among different sectors. The current investigation was designed to assess the nationwide prevalence of LA-MRSA and LA-MSSA in the chicken meat production system involving broiler chickens, chicken meat, workers, and environment of the facilities in broiler farms, slaughterhouses, and retail markets. A total of 200 *S. aureus* (43 MRSA and 157 MSSA) strains were isolated and examined for their genotypic characteristics and antimicrobial resistance (AMR) phenotypes. Multilocus sequence type (MLST) analysis revealed that the most prevalent clone was clonal complex (CC) 5 (n=76, 38%), followed by CC398 (n=56, 28%), CC8 (n=34, 17%), CC1 (n=24, 12%) and CC30 (n=2, 1%). While only two sequence types (STs), ST692 and ST188 (CC385 and CC1), were confirmed in MRSA strains, various STs were observed in MSSA strains. ST692 and ST188 MRSA strains displayed higher levels of AMR and multidrug resistance phenotypes than MSSA strains. Of note, all of the ST692 and ST188 MRSA strains exhibited quinolone resistance through the mutations in *gyrA* (S84L) and *griA* (S80F). In addition to the AMR, 60% of *S. aureus* strains (n = 120), particularly all of ST188 *S. aureus* (n = 10), showed resistance to zinc chloride. These results suggest that AMR, especially quinolone resistance, and zinc resistance may have played a significant role in the prevalence and persistence of ST692 and ST188 LA-MRSA in broiler farms in Korea.

[This work was carried out by the support of "Research of Korea Centers for Disease Control and Prevention (Project No. 2017NER54060 and 2020ER540500 to S.J.Y)"]

**D004****Anti-bacteria Screening against *S. aureus* and *P. aeruginosa* Using Natural Products of NNIBR Library**

Jinyeong Heo<sup>1</sup>, Hyung Jun Kim<sup>2</sup>, Honggun Lee<sup>1</sup>, Kideok Kim<sup>1</sup>, David Shum<sup>1</sup>, Young Taek Oh<sup>3</sup>, and Soojin Jang<sup>2\*</sup>  
<sup>1</sup>Screening Discovery Platform, Institut Pasteur Korea, <sup>2</sup>Antibacterial Resistance Research Laboratory, Institut Pasteur Korea, <sup>3</sup>Nakdonggang National Institute of Biological Resources

*S. aureus* (*Staphylococcus aureus*) is a Gram-positive, coccal-shaped bacterium and causative agent of wide range of infectious diseases such as skin infections, bacteremia, endocarditis, pneumonia and food poisoning. *S. aureus* is well known for its ability to acquire resistance to various antibiotic classes. Especially, the emergence and spread of methicillin-resistant *S. aureus* (MRSA) strains in hospitals and subsequently in community resulted in significant mortality and morbidity.

*P. aeruginosa* (*Pseudomonas aeruginosa*) is a Gram-negative, rod-shaped, asporogenous, and monoflagellated bacterium. It is a ubiquitous microorganism which has the ability to survive under a variety of environment conditions. It not only causes disease in plants and animals, but also in humans, causing serious infection in hospital-acquired infections, especially immunocompromised patients.

The major problem leading to high mortality is the appearance of drug-resistant strains. Therefore, a vast number of approaches to develop novel anti-infectives is currently needed.

Herein, we attempted to screen using provided natural products from NNIBR (Nakdonggang National Institute of Biological Resources) for potential of alternative drugs.

[This work was supported by grant from the National Research foundation of Korea (NRF; MSIT NRF-2017M3A9G7072864 and MIST NRF-2017M3A9G6068257) and the Nakdonggang National Institute of Biological Resources (NNIBR; NNIBR202002101)]

**D005****Effects of *flaC* Mutation on Stringent Response-mediated Bacterial Growth, Toxin Production, and Motility in *Vibrio cholerae***Hyeon Ju Nam<sup>1</sup>, Pyo Yun Cho<sup>1</sup>, Hye-won Yang<sup>1</sup>, Sang Sun Yoon<sup>2</sup>, and Young Taek Oh<sup>1\*</sup><sup>1</sup>*Nakdonggang National Institute of Biological Resources*, <sup>2</sup>*Department of Microbiology and Immunology, Yonsei University College of Medicine*

The stringent response (SR), which is activated by accumulation of (p)ppGpp under conditions of growth-inhibiting stresses, plays an important role on growth and virulence in *Vibrio cholerae*. Herein, we carried out a genome-wide screen using transposon random mutagenesis to identify genes controlled by SR in a (p)ppGpp-overproducing mutant strain. One of the identified SR target genes was *flaC* encoding flagellin. Genetic studies using *flaC* and SR mutants demonstrated that FlaC was involved in bacterial growth, toxin production, and normal flagellum function under conditions of high (p)ppGpp levels, suggesting FlaC plays an important role in SR-induced pathogenicity in *V. cholerae*. [This work was supported by a grant from the Nakdonggang National Institute of Biological Resources (NNIBR) funded by the Ministry of Environment (MOE) of Republic of Korea (NNIBR202002101).]

**D006****Role of ppGpp-regulated Efflux Genes in *Acinetobacter baumannii***

Kyeongmin Kim, Hye-Won Jung, Md. Maidul Islam, Je Chul Lee, and Minsang Shin\*

*Department of Microbiology, School of Medicine, Kyungpook National University*

Treatment of infections caused by *Acinetobacter baumannii* nosocomial strains has become increasingly problematic owing to their resistance to antibiotics. ppGpp is a secondary messenger involved in growth control and various stress responses in bacteria. The mechanism for inhibition of antibiotic resistance via ppGpp is still unidentified in various pathogenic bacteria including *A. baumannii*. Here, we investigated the effects of ppGpp on efflux pump (EP)-related genes in *A. baumannii*. ppGpp-deficient and -complementary strains were constructed by conjugation and we confirmed (p)ppGpp measurements by TLC analysis. We observed that the ppGpp-deficient strain showed abnormal stretching patterns by TEM analysis. The MICs of antimicrobial agents for the WT, ppGpp-deficient and complementary strains were determined by the E-test and broth dilution assay methods. The expression levels of EP-related genes were determined by q-PCR. Dramatic reductions of MICs in the ppGpp-deficient strain compared with the WT were observed for gentamicin, tetracycline, erythromycin and trimethoprim. Expression of the EP-related genes was also decreased in the ppGpp-deficient strain. This study demonstrates that ppGpp regulates EP-related gene expression in *A. baumannii*, affecting antibiotic susceptibility. To date, treatment for MDR *A. baumannii* has had no new antimicrobial agents, so the A1S\_0579 gene could be a novel therapeutic target for rational drug design by affecting ppGpp production.

**D007****Transcriptional Regulation of the Outer Membrane Protein A (OmpA) in *Acinetobacter baumannii***Kyu-wan Oh<sup>1</sup>, Kyeongmin Kim<sup>1</sup>, Md. Maidul Islam<sup>1</sup>, Hye-Won Jung<sup>1</sup>, Daejin Lim<sup>2</sup>, Je Chul Lee<sup>1</sup>, and Minsang Shin<sup>1\*</sup><sup>1</sup>Department of Microbiology, School of Medicine, Kyungpook National University, <sup>2</sup>Department of Microbiology, School of Medicine, Chonnam National University

*Acinetobacter baumannii* is known for its virulence toward severe patients in hospitals. Along with its multi-drug resistance, treating *A. baumannii* infection has become a serious problem in many clinical environments. The Outer Membrane Protein A (OmpA) of the *Acinetobacter* genus is involved in bacterial virulence. The regulating factors of *A. baumannii* OmpA (AbOmpA) in the post-transcription stage have been identified in past studies. However, the regulation factors behind the transcriptional stage of AbOmpA expression are yet unclear.

We investigated A1S\_0316, a gene that encodes the putative transcription factor for AbOmpA gene expression by DNA-affinity chromatography. We purified A1S\_0316 and examined using size-exclusion chromatography, indicating it forms as in dimer. The binding affinity of A1S\_0316 on AbOmpA promotor region (AbOmpAp) was examined through EMSA. We compared the binding affinity between A1S\_0316 and H-NS protein, a well-known global transcriptional repressor, on AbOmpAp. The A1S\_0316 showed higher binding affinity to AbOmpAp than H-NS protein. We examined regulatory effect of these proteins on AbOmpAp using real-time qPCR and various in vitro tools. Our results indicate that the A1S\_0316 acts as an anti-repressor to AbOmpA gene, inhibiting the binding of H-NS protein. This study is the first demonstration of the transcriptional regulation on OmpA gene. Further details will be discussed at the meeting.

**D008****ppGpp Signaling Plays Critical Role in Biofilm Formation and Virulence of *Acinetobacter baumannii***Md. Maidul Islam<sup>1</sup>, Kyeongmin Kim<sup>1</sup>, Hye-won Jung<sup>1</sup>, Kyu-wan Oh<sup>1</sup>, Daejin Lim<sup>2</sup>, Kwang Soo Kim<sup>2</sup>, Sung-Gwon Lee<sup>3</sup>, Chungoo Park<sup>3</sup>, Je Chul Lee<sup>1</sup>, and Minsang Shin<sup>1\*</sup><sup>1</sup>Department of Microbiology, School of Medicine, Kyungpook National University, <sup>2</sup>Department of Microbiology, Chonnam National University Medical School, <sup>3</sup>School of Biological Sciences and Technology, Chonnam National University

*Acinetobacter baumannii* is a major cause of nosocomial infections which can survive in different hospital environments and its multidrug-resistant capacity is major concern now-a-days. Previous our result showed that ppGpp regulates EP-related gene expression in *A. baumannii*, affecting antibiotic susceptibility [J Antimicrob Chemother. 2020 May 1;75(5):1130-1134]. Here we investigate whether ppGpp are involved in the pathogenesis of *A. baumannii* through biofilm formation, surface motility, adhesion, invasion assays and in-vivo mouse study. Transcriptome analysis of early stationary phase cultures represents that a total of 891 genes were differentially expressed (fold change  $\geq 2$ ), (among them 114 up-regulated and 777 down-regulated genes) in ppGpp deficient strain compared with wild-type. A ppGpp-deficient *A. baumannii* strain exhibited reduction in biofilm formation by more than 3-fold and reduced surface motility. Adhesion to and invasion into human epithelial cells also significantly reduced in ppGpp deletion mutant. In-vivo mouse model reveals that reduced bacterial number in blood of ppGpp deficient strain treated mice than WT. Our findings showed that ppGpp signaling plays critical role in biofilm formation and virulence of *A. baumannii*. This study is the first demonstration of the association between ppGpp and pathogenicity of *A. baumannii*. So, ppGpp could be a potential target for developing anti-virulence strategies against *A. baumannii*.

[Supported by NRF]

**D009****The Novel Functions of a Small Alarmone Synthetase, a Potential (p)ppApp Producer, in *Vibrio cholerae***Hwa Young Kim<sup>1,2</sup>, Ji-Eun Kim<sup>1</sup>, and Sang Sun Yoon<sup>1,2,3\*</sup><sup>1</sup>Department of Microbiology and Immunology, <sup>2</sup>Brain Korea 21 PLUS Project for Medical Sciences, <sup>3</sup>Institute for Immunology and Immunological Diseases, Yonsei University College of Medicine

The stringent response is mediated by a stress alarmone, (p)ppGpp. (p)ppGpp is produced from ATP and either GTP or GDP and controlled by RelA and SpoT. Recent research revealed that an interbacterial toxin, Tas1, in *P. aeruginosa* has a homologous domain with RSH, which produces (p)ppApp, not (p)ppGpp. We found that RelV, an additional SAS of *V. cholerae* is highly homologous to the RSH domain of Tas1. The substrate binding site of RelV is almost identical to that of Tas1, thereby leading us to hypothesize that RelV would synthesize (p)ppApp rather than (p)ppGpp. To demonstrate this hypothesis, we overexpressed *relV* or *relA* gene in *V. cholerae*. We determined that each strain showed different survival fitness. These results suggest that the product from RelV would exert different functions than RelA and it could result in the different overexpression phenotypes. In *V. cholerae*, a small gene (*vc1223*) is conserved and located immediately downstream of *relV* gene. We test whether the VC1223 has the role of an antagonistic effects on RelV. The toxicity of RelV overexpressed in *V. cholerae* was partially inhibited by co-expressed VC1223. We confirmed that the SpoT, a (p)ppGpp-hydrolase, could recover the phenotypes by alleviating the toxicity of RelV in *V. cholerae*. Further study is necessary to reveal the regulatory network of RelA/RelV/SpoT/VC1223 in *V. cholerae* for homeostasis of (p)ppGpp and (p)ppApp.

[This work was supported by National Research Foundation of Korea Grants.]

**D010****Genetic Characteristics and Pathogenicity of Low Pathogenic Avian Influenza H5 Viruses Isolated from Wild Birds in South Korea**

Yu-Na Lee, Sun-Ha Cheon, Yoon-Gi Baek, Yu-Ri Park, Young-Jae Si, Soo-Jeong Kye, Eun-Kyoung Lee, Gyeong-Beom Heo, Myoung-Heon Lee, and Youn-Jeong Lee\*

*Avian Influenza Research & Diagnostic Division, Animal and Plant Quarantine Agency*

Wild birds (WBs) are considered the natural reservoir for avian influenza viruses (AIVs). During the national surveillance programs in South Korea between 2010 and 2017, a total of 770 AIVs isolated from the 89,163 samples in WB habitats. Among these viruses, 59 low pathogenic AIVs (LPAIVs) of the H5 subtype were isolated. Phylogenic analysis revealed that the HA gene of the H5 LPAIVs isolated from WBs since 2012 belonged to Eurasian lineage, classified into three subgroups (HA-II, HA-III, and HA-IV) and were distinguished from those of the previous H5 LPAIVs (HA-I) isolated until 2010. To investigate the replication and transmission capacities of the representative strains of 3 subgroups in chicken, virus shedding was monitored in both challenged and direct-contact groups for 2 wks post-infection. Chickens from HA-II subgroup shed virus by both the oropharyngeal and cloacal route and were 40% seropositive. By contrast, viral shedding from the oropharyngeal route was detected in only one of five chickens from HA-III and HA-IV subgroup respectively. The seroconversion rate was 20% in each group. No transmission to direct-contacts was detected in all groups. Our results suggest that the H5 LPAIVs circulating in WBs were not sufficiently adapted for gallinaceous poultry. Nevertheless, given that WBs harbor novel strains that could affect poultry, our results highlight the need for enhanced AIV surveillance in both WBs and poultry in Eurasia.

[Supported by grants from the APQA.]

**D011****Antimicrobial Susceptibility and Serogroup of Avian Pathogenic *Escherichia coli* from Chickens and Ducks in South Korea between 2018 and 2019**

Jiyeon Jeong, Min-Su Kang, Jin-Hyun Kim, Ok-Mi Jeong, Hye-Jin Lee, Yong-Kuk Kwon, and Ji-Youn Lee\*  
*Animal and Plant Quarantine Agency*

Avian pathogenic *E. coli* (APEC) is the major cause of colibacillosis in poultry, leading to significant economic losses. We investigated distribution of serogroups, antimicrobial resistance patterns and resistance gene retention among the recent APEC isolates from chickens and ducks between 2018 and 2019 in Korea. O2 and O78 were the predominant serogroups in chicken and duck isolates, respectively. APEC isolates showed high resistance to nalidixic acid, tetracycline, and ampicillin, whereas most isolates were susceptible to colistin and azithromycin. In particular, chicken isolates showed higher resistance to ceftiofur, ceftriaxone, and gentamicin in comparison with duck isolates ( $p < 0.05$ ). Most ampicillin resistant isolates harbored  $\beta$ -lactamase encoding genes *bla*<sub>TEM</sub>, *bla*<sub>CTX-M</sub>, and *bla*<sub>SHV</sub>. The plasmid-mediated quinolone genes *qnrB*, *qnrS* and *aac6'-1b-cr* were identified in nalidixic acid resistant isolates. One isolate resistant to colistin (MIC 16  $\mu$ g/ml) carried *mcr-1*, colistin resistance gene, which is the first reported in APEC in Korea. This report indicates that recent APEC isolates were highly resistant to antibiotics used in humans and animals and harbor various antimicrobial resistant genes; these genes can be transferred between food-producing animal and human pathogens, which is a public health concern and should be monitored to track the spread of antimicrobial resistant APEC in poultry farms.

[Supported by grants from Animal and Plant Quarantine Agency]

**D013****Different Growth Kinetics among Subgroups of Highly Pathogenic Avian Influenza H5N8 Virus**

Yoon-Gi Baek, Yu-Na Lee, Yu-Ri Park, Young-Jae Si, Soo-Jeong Kye, Myoung-Heon Lee, and Youn-Jeong Lee\*  
*Avian Influenza Research & Diagnostic Division, Animal and Plant Quarantine Agency*

In early 2014, a novel reassortant clade 2.3.4.4 H5N8 viruses which have circulated in china since 2013 were introduced into South Korea through migratory wild bird. The H5N8 viruses had been spread worldwide and then reintroduced into South Korea in late 2014, while some of the primitive H5N8 viruses (C0) had remained in South Korea. These viruses has evolved further into four subgroups (C1-C4). In this study, to investigate the different growth kinetics in the cell culture among the representative viruses of C0, C1, C2 and C4 group, DF-1 cell was infected with each virus. Supernatants were collected at 8 h, 24 h and 48 h post infection (hpi), and inoculated into DF-1 to measure TCID<sub>50</sub>. The result of growth kinetics showed a statistical significant difference at 24 hpi. A/broiler duck/Korea/Buan2/2014 (Buan2) of C0 group ( $3.9 \pm 0.4 \log_{10} \text{TCID}_{50}/0.1\text{ml}$ ) and A/broiler duck/Korea/H1731/2014 (H1731) of C1 group ( $4.4 \pm 0.1 \log_{10} \text{TCID}_{50}/0.1\text{ml}$ ) were statistically different from A/mallard duck/Korea/H2102/2015 (H2102) of C2 group ( $6.0 \pm 0.5 \log_{10} \text{TCID}_{50}/0.1\text{ml}$ ) and A/domestic mallard duck/Korea/H1924/2014 (H1924) of C4 group ( $6.7 \pm 0.3 \log_{10} \text{TCID}_{50}/0.1\text{ml}$ ) in growth kinetics ( $P < 0.001$ ). These results suggest that each subgroup has a different replication activity according to genetic subgroup. Considered that each subgroup showed a different pathogenicity in chicken, further studies are needed to investigate a correlation between growth kinetics and pathogenicity of the H5N8 viruses in chicken.

**D014****Genetic and Biochemical Equivalent Characterization for Staphylococcus and Micrococcus NCCP Resources as Reference Strains**Yewon An<sup>1,2,3</sup>, Youngsil Choi<sup>1,2,3</sup>, and Suyeon Kim<sup>1,2,3\*</sup><sup>1</sup>Korea Centers for Disease Control & Prevention, <sup>2</sup>National Institutes of Health, <sup>3</sup>National Culture Collection for Pathogens (NCCP)

The National Culture Collection for Pathogens (NCCP), has conducted an equivalent analysis to replace ATCC reference strains with the Korean isolates in NCCP. In order to find alternative strains for ATCC strains, we conducted equivalent experiments with these three strains and 45 Korean strains. First, we analyzed their molecular, biological, and biochemical properties using MALDI-TOF, VITEK-2, and antibiotic disc diffusion method. Additionally, 16S rRNA sequencing, MLST analysis, and NGS analyses were performed to investigate their molecular genetic characteristics. Korean candidates selected were *Staphylococcus aureus* NCCP 16830, *Staphylococcus epidermidis* NCCP 16828, and *Micrococcus luteus* NCCP 16831. They were evaluated for their usefulness by conducting an experiment applying the Microbial Assay for Antibiotics from the general test methods of the KP. The mean titers of the three selected NCCP strains were considered useful because they were within the effective range. In addition, whole genome sequencing was used to compare genetic homology between the ATCC and NCCP strains. *S. aureus*, *S. epidermidis*, and *M. luteus* showed 99.00%, 99.60%, and 98.10% homology between the two strains, respectively. Through this study, the possibility of using NCCP strains to replace ATCC strains was confirmed.

[This study is supported by the funds of the Korea Centers for Disease Control & Prevention, Ministry of Health and Welfare (Funding no. 2017-NG45005-00).]

**D015****Newly Synthesized Photosensitizer, CHLO-Cl, Enhances the Antimycobactericidal Activity of Macrophages against Mycobacterium tuberculosis Infection**Seung-Hwan Baek<sup>1</sup>, Hyo-Ji Lee<sup>1,2</sup>, Chang-Hee Lee<sup>3</sup>, and Yu-Jin Jung<sup>1,2\*</sup><sup>1</sup>Department of Biological Sciences Kangwon National University, <sup>2</sup>Institute of Life Sciences, Kangwon National University, <sup>3</sup>Department of Chemistry, Kangwon National University

*Mycobacterium tuberculosis* (Mtb) is the causative agent of tuberculosis (TB) and infects host macrophages. After Mtb infection, macrophages induce various host defense mechanisms to control Mtb survival. Recent studies have shown that photosensitizer DH-I-180-3 can effectively control Mtb survival in the presence of light exposure, which indicates that it could be developed as a potential drug for photodynamic therapy (PDT) of TB patients. In this study, we sought to assess the bactericidal activity of newly synthesized CHLO-Cl derived from DH-I-180-3 during Mtb infection. Survival of Mtb significantly decreased by 100-fold in CHLO-Cl-treated Mtb with light exposure compared to Mtb treated with CHLO-Cl without light exposure. Repeated treatment of CHLO-Cl with light exposure suppressed Mtb growth, which was comparable to survival of Mtb treated with anti-TB drug, rifampicin (RMP). Treatment of CHLO-Cl with light exposure inhibited Mtb-induced host cell death in Raw264.7 cells. Intracellular Mtb growth was reduced in CHLO-Cl-treated Raw264.7 cells in the presence of light exposure, compared with cells treated with CHLO-Cl without light exposure. Overall, these results suggest that CHLO-Cl can enhance the ability of macrophages to kill bacteria as well as antibiotics that directly kill Mtb. Therefore, CHLO-Cl could be developed as a new drug to improve efficacy of TB treatment.

[Supported by grant from through the National Research Foundation of Korea (NRF-2018R1D1A1B07049097)]

**D016**

**Characteristic Analysis for Clinical Isolates of *Pseudomonas aeruginosa* NCCP Strains from Korea**

Joon Ki Kim, Chi-Hwan Choi, Su Yeon Kim, and Young Sill Choi\*

*Pathogen Resource TF, National Culture Collection for Pathogens, Korea National Institute of Health, Center for Infectious Diseases Research*

51 strains of *P. aeruginosa* registered and quality managed by NCCP, with minimum species identification and culture condition information. This study aimed to provide information to researchers on the characterization analysis of *P. aeruginosa*. The analysis of antibiotic susceptibility, genotype, toxin type, and serotype for 51 *P. aeruginosa* NCCP strains was performed. Antibiotic susceptibility analysis was performed using the disk diffusion method according to the CLSI guidelines and revealed 18 groups; 39.2% of *P. aeruginosa* showed non-susceptibility to all six antibiotics used in the experiment. Sequence type analysis was performed using MLST to confirm the allele number of a house keeping gene and the result was confirmed using PubMLST (<https://PubMLST.org>). Twenty-six groups were identified and ST 235 was the most common at 37.3%. Toxin type analysis was performed by PCR using the toxin genes *exo* (S, T, U, Y) and *toxA*. All strains contained more than one toxin gene, and *P. aeruginosa* showed 98%, 51%, 92.2%, 51%, and 88.2% of *toxA*, *exo* (S, T, U, Y) genes, respectively. For serotype analysis, A~N type was analyzed using a *P. aeruginosa* antisera commercial kit. The serotype analysis revealed that 8 of the 14 serotypes were present. The most common serotype was E type (45.1%), followed by untype (23.5%).

[This study was supported by the funds of the Korea Centers for Disease Control & Prevention, Ministry of Health and Welfare (Funding no. 2019-NG046-00).]

**D017**

**N-Glycan Dependent Endoplasmic Reticulum-associated Glycoprotein Quality Control and Degradation in the Pathogenic Yeast *Cryptococcus neoformans***

Catia Mota, Ki Seung Kim, Eun Jung Thak, Su-bin Lee, and Hyun Ah Kang\*

*Department of Life Science, Chung-Ang University*

Eukaryotic cells evolved a N-glycan-dependent endoplasmic reticulum (ER)-mediated glycoprotein quality control (QC) and ER-associated degradation (ERAD) pathway. The human-pathogenic yeast *Cryptococcus neoformans* has a unique N-linked glycosylation pathway lacking glucosyltransferases but carrying multiple mannosidases. To investigate the molecular assembly of *C. neoformans* specific-QC pathway and its impact on pathogenicity, we disrupted the homologue genes to *UGGT*, *MNS1* (*MNS1A* and *MNS1B*), *MNL1* (*HTM1*) and *MNL2*. The N-glycan profile based on HPLC analysis indicated *MNS1A* as a major *MNS1* homolog, while *MNS1B* as a minor homolog. Thus, simultaneous disruption of both genes led to the total impairment of the *MNS1* function. Although *mns1AΔ*, *mns1BΔ* and *mns1Amns1BΔ* displayed mild growth retardation under several stress conditions, *mns1BΔ* showed particularly enhanced resistance to SDS, suggesting the possibility of *MNS1B* having an additional function. Deletion of *MNL1* and *MNL2*, encoding putative ERAD-associated mannosidases, resulted in increased sensitivity to ER, cell wall stressors and to antifungal drugs. *uggtΔ* showed a noticeable growth defect even at normal growth conditions and severe growth retardation under several stress conditions. Collectively, this shows the importance of protein QC and degradation pathways for proper cell growth and ability to endure various stress conditions in *C. neoformans*.

[Supported by grants NRF-2019R1A2C1084942 and NRF2018R1A5A102507]

**D018****Effect of Altered N-Glycan Structures of Cell Surface Mannoproteins on the Interaction of *Cryptococcus neoformans* with Host Cells**

Su-bin Lee, Eun Jung Thak, Jung Ho Kim, Seung Yeon Chung, and Hyun Ah Kang\*

*Department of Life Science, Chung-Ang University*

*Cryptococcus neoformans* is an opportunistic fungal pathogen that can cause life-threatening meningoencephalitis in immunodeficiency patients. The cell wall of *C. neoformans* includes highly N-glycosylated mannoproteins (MPs), such as MP98, 88, and 84, known as immunoreactive antigens. To investigate the N-glycan structures specific to each Cryptococcal MP and their defined roles in host interaction, MPs with truncated N-glycan chains were produced using an *alg3* null mutant with a defective N-glycosylation. HPLC and MALDI-TOF analysis revealed the distinctive N-glycan profiles of each MPs. *In vitro* adhesion assay with the purified His-tagged MPs revealed that MPs display higher adhesion affinity to lung epithelial cells than macrophages, and that N-glycan truncation resulted in almost no detectable change in the adhesion activity of MPs, despite that methyl- $\alpha$ -D mannopyranoside, a mannose receptor competitive inhibitor, inhibited the binding of MPs to BMDC effectively. To examine the effect of absence of the N-glycans in MPs, a mutant MP84 protein lacking N-glycans completely was generated by site-mutagenesis and analyzed for its immune induction activity. The absence of N-glycans recovered the ability to active cytokine production, although the truncated N-glycans decreased the immune induction activity, indicating complicated effects of N-glycan structures of MPs on the interaction with host cells.

[Supported by grant from the National Research foundation of Korea]

**D019****Antibacterial Effects of Recombinant Endolysin to Sterilize Medical Devices : A Pilot Study**Yoon-Jung Choi<sup>1</sup>, Shukho Kim<sup>1</sup>, Hyun-Ha Jang<sup>2</sup>, Jong-Sook Jin<sup>1</sup>, Yujin Lee<sup>1</sup>, Shin-Woo Kim<sup>2</sup>, and Jungmin Kim<sup>1\*</sup>*<sup>1</sup>Department of Microbiology, School of Medicine, Kyungpook National University, <sup>2</sup>Department of Allergy and Infectious Diseases, Kyungpook National University*

Bacterial contamination of medical devices in hospitals is a very serious problem. Endolysins are enzymes of bacteriophages which can degrade the peptidoglycan of the host bacterial cell wall at the stage of burst-out of the progeny virion. The purpose of this study is to investigate whether recombinant endolysins are able to eradicate bacteria contaminating medical devices. The cultured bacteria were subjected to 16S rRNA sequencing for identification. A recombinant endolysin, LysSS, with a C-terminal 6-His tag was expressed using a His-trap column installed in a fast protein liquid chromatography system. Minimum inhibitory concentrations (MICs) of LysSS against bacteria were determined according to CLSI method. The antibacterial activity of LysSS was determined by measuring the optical density (OD) and counting colony forming unit (CFU) after incubation for 18, 24, 30, 48, and 72 h. A total of 34 bacterial species were identified from 10 catheters. Among them, three bacterial species were dominated; *Staphylococcus epidermidis*, *Corynebacterium striatum*, and *Enterococcus faecium*. LysSS could inhibit *S. epidermidis* and *C. striatum*, but not *E. faecium*. Catheters in which *S. epidermidis* and *C. striatum* were dominant showed a dramatic decrease of OD and CFU after 24 h treatment of LysSS at a concentration of 125-250  $\mu$ g/ml. These results indicate that LysSS endolysin can be used to sterilize medical devices to replace chemical and/or physical antibacterial agents.

**D020****Changes of Gut Microbiota in Carbapenem-resistant *Enterobacteriaceae* Patients after Fecal Microbiota Transplantation**Jin-Jae Lee<sup>1</sup>, Seung Soon Lee<sup>2</sup>, and Bong-Soo Kim<sup>1\*</sup><sup>1</sup>Department of Life Science, Hallym University, <sup>2</sup>Dept of Internal Medicine, Hallym University College of Medicine

The emergence of antibiotic resistant bacteria has become a worldwide concern. Infection with Carbapenem-resistant *Enterobacteriaceae* (CRE) has become an important challenge in health care settings and a growing concern worldwide. Fecal microbiota transplantation has been proposed as a new strategy to promote decolonization in order to reduce the risk of ARB. Therefore, we analyzed the effects of FMT for decolonization of CRE and the different shifts of gut microbiota by decolonization status. We analyzed the gut microbiota from rectal swab samples obtained from CRE patients before and after FMT treatment (n = 56). The observed OTUs and Shannon diversity indices were increased more in patients with CRE decolonization before 8 weeks than in patients with CRE decolonization after 8 weeks by FMT. The bacterial compositions were significantly changed by FMT ( $P$ -value = 0.003). Genera in gut microbiota were differently changed between two groups during follow-up period after FMT. In addition, the microbiota in patients with CRE decolonization before 8 weeks was changed similar to that of the donor ( $P$ -value = 0.003), and the microbiota differently in patients with CRE decolonization after 8 weeks ( $P$ -value = 0.029). These results showed that the gut microbiota could indicate the effects of FMT on decolonization of CRE, and it can be applied to develop predictive method for decolonization of CRE by gut microbiota information.

[Supported by grant from NRF (NRF-2019R111A3A01060465).]

**D021****Contribution of Zur-regulated Lipoprotein A to Bacterial Morphogenesis and Production of Outer Membrane Vesicles in *Acinetobacter baumannii***Hyo Jeong Kim<sup>1</sup>, Nayeong Kim<sup>1</sup>, So Hyun Jun<sup>1</sup>, Joo Hee Ryu<sup>1</sup>, Man Hwan Oh<sup>2</sup>, Seung Il Kim<sup>3,4</sup>, Minsang Shin<sup>1</sup>, Yoo Chul Lee<sup>1</sup>, and Je Chul Lee<sup>1\*</sup><sup>1</sup>Department of Microbiology, School of Medicine, Kyungpook National University, <sup>2</sup>Department of Nanobiomedical Science, Dankook University, <sup>3</sup>Drug & Disease Target Team, Korea Basic Science Institute, <sup>4</sup>Department of Bio-Analytical Science, University of Science and Technology (UST)

Zinc uptake-regulator (Zur)-regulated lipoprotein A (ZrIA) plays a role in bacterial fitness and surmounting antimicrobial exposure in *Acinetobacter baumannii*. This study further characterized the *zrIA* gene and its encoding protein and investigated the roles of the *zrIA* gene in bacterial morphology, antimicrobial susceptibility, and production of outer membrane vesicles (OMVs) in *A. baumannii* ATCC 17978. *In silico* and polymerase chain reaction analyses showed that the *zrIA* gene was conserved among *A. baumannii* strains with 97%-100% sequence homology. Recombinant ZrIA protein exhibited a specific enzymatic activity of D-alanine-D-alanine carboxypeptidase. Wild-type *A. baumannii* exhibited more morphological heterogeneity than a  $\Delta zrIA$  mutant strain at stationary phase. The  $\Delta zrIA$  mutant strain was more susceptible to gentamicin than the wild-type strain. Sizes and protein profiles of OMVs were similar between the wild-type and  $\Delta zrIA$  mutant strains, but the  $\Delta zrIA$  mutant strain produced 9.7 times more OMV particles than the wild-type strain. OMVs from the  $\Delta zrIA$  mutant were more cytotoxic in cultured epithelial cells than OMVs from the wild-type strain. The present study demonstrated that *A. baumannii* ZrIA contributes to bacterial morphogenesis and antimicrobial resistance, but its deletion increases OMV production and OMV-mediated host cell cytotoxicity. These findings provide opposing perspectives of ZrIA for anti-virulence strategies against *A. baumannii*.

**D022****RNA Polymerase-binding Transcription Factor DksA Negatively Regulates Fitness of *Acinetobacter baumannii***

Nayeong Kim, Hyo Jeong Kim, Joo Hee Son, Minsang Shin, and Je Chul Lee\*

*Department of Microbiology, School of Medicine, Kyungpook National University*

*Acinetobacter baumannii* is an important opportunistic pathogen that is aerobic, Gram-negative bacillus associated with nosocomial infection. The DksA is a crucial factor of (p)ppGpp-, guanosine pentaphosphate or tetrphosphate, dependent regulation for optimal control of RNA polymerase through direct interaction with RNA polymerase to alter transcription initiation at target promoters. Although (p)ppGpp is an important transcription factor for regulation of antimicrobial susceptibility and virulence in *A. baumannii*, the role of DksA has not been determined yet. This study investigated the role of DksA in antimicrobial susceptibility and pathogenesis of *A. baumannii* using wild-type *A. baumannii* ATCC 17978,  $\Delta dksA$  mutant, and *dksA*-complemented strains. The  $\Delta dksA$  mutant strain exhibited higher minimum inhibitory concentrations of cephalosporins and quinolones than the wild-type strain. Biofilm formation was significantly increased in the  $\Delta dksA$  mutant compared to the wild-type strain. The RNA-seq and qRT-PCR analyses revealed that RND efflux pump genes and biofilm-associated genes such as *csu* operon were significantly up-regulated in the  $\Delta dksA$  mutant. In conclusions, (p)ppGpp positively regulates the antimicrobial resistance and pathogenesis of *A. baumannii*, whereas DksA negatively regulates bacterial fitness.

**D025****Reshaping of the Gut Microbiota in Peroxisome Proliferator-activated Receptor Alpha Deficient Mice**June-Young Lee<sup>1</sup>, Sang Min Jeon<sup>2</sup>, Eun-Kyeong Jo<sup>2</sup>, and Jin-Woo Bae<sup>1\*</sup><sup>1</sup>*Department of Life and Nanopharmaceutical Sciences and Department of Biology, Kyung Hee University,*<sup>2</sup>*Department of Microbiology, Chungnam National University School of Medicine*

The Peroxisome proliferator-activated receptor alpha (PPAR $\alpha$ ) is a nuclear receptor that regulates the oxidation and transport of fatty acids. Recently, PPAR $\alpha$  is known as an essential factor for antimycobacterial responses via activation of transcription factor EB transcription and inhibition of lipid body formation. Also, PPAR $\alpha$  deficiency in mice led to dysbiosis in the gut, resulting in a microbiota-dependent increase in the expression of inflammatory cytokines and enhanced susceptibility to intestinal inflammation. But, the role of gut microbiota in mycobacterial infection with PPAR $\alpha$  deficiency is largely unknown. We investigated how mycobacterial infection and PPAR $\alpha$  deficiency affects gut microbiota profile. We found that microbial diversity was significantly increased in PPAR $\alpha$  KO mice. Gut microbiota profile between WT and PPAR $\alpha$  KO mice were significantly different and clustered separately by infection of mycobacteria. Before mycobacterial infection, compared to WT, *Butyrivimonas*, *Bilophila* and *Parabacteroides* were depleted and *Lactobacillus* and *Akkermansia* were enriched in PPAR $\alpha$  KO mice. After infection, *Lactobacillus* was still enriched in PPAR $\alpha$  KO mice. From these data, we conclude that PPAR $\alpha$  and mycobacterial infection can modify the host gut microbiota and gut microbiota may be associated with response to mycobacterial infection.

[This work supported by Mid-career Researcher Program (NRF-2020R1A2C3012797) through the National Research Foundation of Korea]

**D026**

**Development of Nanozyme-linked Immunosorbent for the Detection Foot and Mouth Disease Virus (FMDV) Antibodies Using Goat Sera**

Ho-Seong Cho

*College of Veterinary Medicine and Veterinary Diagnostic Center, Jeonbuk National University*

Foot-and-Mouth Disease (FMD) is the first class legally notifiable communicable disease determined by the Office of International Epizootics (OIE). It is an international epidemic disease that infects cloven-hoofed animals such as cattle, goat and swine. It is a very contagious disease that requires an intensive surveillance program. The diagnostic potential of this nanozyme-based ELISA, for detecting the antibody titers to the FMDV O type, was compared with that of conventional ELISA using 100 goat sera taken from 10 goat farms. The nanozyme-based ELISA system was found to be highly specific and sensitive. Moreover, there was a strong positive correlation between the nanozyme-based and conventional ELISA systems ( $n=100$ ,  $r=0.806$ ,  $P < 0.01$ ). Therefore, this method is expected to be a valuable and reproducible tool in the serological monitoring of FMDV vaccination.

[This work was supported by Korea Institute of Planning and Evaluation for Technology in Food, Agriculture, Forestry and Fisheries (IPET) through Animal Disease Management Technology Development Program, funded by Ministry of Agriculture, Food and Rural Affairs (MAFRA) (118091031).]

**D027**

**TLR5-induced Lipocalin 2 Protects Mice against Cyclophosphamide-induced Neutropenic Sepsis**

Daejin Lim<sup>1,2</sup>, Jae-Ho Jeong<sup>1,2</sup>, Hyung-Ju Lim<sup>1</sup>, Miryoung Song<sup>3</sup>, and Hyon E. Choy<sup>1,2\*</sup>

*<sup>1</sup>Department of Microbiology, Chonnam National University Medical School, <sup>2</sup>Department of Molecular Medicine (BK21plus), Chonnam National University Graduate School, <sup>3</sup>Department of Bioscience and Biotechnology, Hankuk University of Foreign Studies*

Neutropenic sepsis is a fatal consequence of chemotherapy, and septic complications are a principal cause of mortality. Chemotherapy-induced neutropenia leads to the development of microscopic ulcers in the gastrointestinal epithelium that serve as entry for intraluminal bacteria, which translocate across the intestinal mucosal barrier for systemic distribution. Here, we developed a mouse model mimicking the pathophysiologic sequence of events occurring in patients with neutropenic sepsis due to administration of cyclophosphamide (500 mg/kg). The model was used to demonstrate that bacterial flagellin, a TLR5 agonist, extended the survival of cyclophosphamide-treated mice by reducing the bacterial load in internal organs. The antimicrobial protein lipocalin 2 is induced by TLR5-NF- $\kappa$ B activation in hepatocytes to confer protection, presumably by sequestering iron from infected bacteria. Lipocalin 2 administration (100  $\mu$ g/mouse) at 12 h after cyclophosphamide treatment effectively extended survival in mice. These findings indicate that the inclusion of lipocalin 2 in the treatment regimen of neutropenic sepsis may improve the therapeutic outcome of cancer chemotherapy.

D028

**Inactivated *Corynebacterium glutamicum* Administered via Oral Route Enhanced Memory T Cell Responses in Mouse Vaccinated with Inactivated *Escherichia coli* K99**Narae Kim<sup>1</sup>, Dobin Ju<sup>1</sup>, Yang-Su Kim<sup>2</sup>, Han Wool Kim<sup>2</sup>, and Cheol-Heui Yun<sup>1\*</sup><sup>1</sup>Department of Agricultural Biotechnology and Research Institute of Agriculture and Life Sciences, Seoul National University, <sup>2</sup>CJ BIO Research & Technology, CJ Cheiljedang Corporation

*Corynebacterium glutamicum*, a non-pathogenic Gram-positive bacterium, is a useful microorganism in the industrial process, such as producing L-glutamate that used as animal feed supplement. While in-depth studies are being conducted for the final product-based approaches, the effects of *C. glutamicum* on the host immune system are yet to be addressed. In the present study, we investigate the safety and adjuvanticity of oral administration of heat-inactivated *C. glutamicum*.

In the first set of study, body weight, diarrhea score, serum cytokine, length of spleen and large intestine, and number of Payer's patches in mouse administered orally with inactivated *C. glutamicum* were examined. The results showed that no significant changes were found in all doses (0, 1, 10, or 100 mg/kg BW) tested. Next, the adjuvanticity of *C. glutamicum* for *E. coli* K99 vaccination model was investigated. The administration of *C. glutamicum* increased the proliferation of antigen-specific memory CD8<sup>+</sup> T cell. Furthermore, expression of IL-17A, known to be required for the elimination of extracellular bacteria, in antigen-specific memory CD8<sup>+</sup> T cells in mouse orally administered with inactivated *C. glutamicum* was increased.

Collectively, the present study suggested that inactivated *C. glutamicum* is seemingly safe in the host when orally administered together with. Moreover, it is probable that *C. glutamicum* as a vaccine adjuvant might be useful for inducing host memory CD8<sup>+</sup> T cell responses.

D029

**Genomic Insight on the Serotype 3 *Streptococcus suis* INT-01 Isolated from a Domestic Pig in South Korea**Seon Young Park<sup>1,2</sup>, Se Ra Lim<sup>3</sup>, Hyemin Kwon<sup>1</sup>, Jee Soo Son<sup>4</sup>, Jee Eun Han<sup>5</sup>, and Ji Hyung Kim<sup>1\*</sup><sup>1</sup>Infectious Disease Research Center, Korea Research Institute of Bioscience and Biotechnology, <sup>2</sup>Division of Animal and Dairy Sciences, College of Agriculture and Life Science, Chungnam National University, <sup>3</sup>Infectious Disease Research Center, Korea Research Institute of Bioscience and Biotechnology, <sup>4</sup>iNtRON Biotechnology, <sup>5</sup>Laboratory of Aquatic Biomedicine, College of Veterinary Medicine, Kyungpook National University

*Streptococcus suis* a major pathogen of pigs, causes severe economic losses to the swine industry. Several virulence factors have been associated with swine-pathogenic strains of *S. suis*, and the most prevalent virulence-associated genes were extracellular protein factor (*epf*), muraminidase-released protein (*mpr*), and suilyisin (*sly*). Although recent study have reported that the serotype 3 with the *epf*-/*mpr*+/*sly*- genotype was the most prevalent among the Korea isolates, its genomic characteristics have not been investigated until yet. Here, we report the genome of *S. suis* strain INT-01 isolated from a domestic pig in Korea. The newly obtained 2,092,054 bp genome of *S. suis* strain INT-01 showed the capsular polysaccharides synthesis locus of INT-01 was almost identical to another serotype 3 *S. suis* strains, thus suggesting that the isolate INT-01 could be classified as *S. suis* serotype 3. The strain INT-01 was phenotypically resistant to several antibiotics, however, the corresponding resistance genes could not be detected in its genomic analysis, thus implying that other unknown resistance determinants are present in the isolate and additional investigations will be needed in our future study. The genomic information of strain INT-01 might provide important insights into the development of control strategies against *S. suis* infections in Korea.

[Supported by grants from the National Research Foundation (NRF) (NRF-2020R1I1A2068827) and KRIBB Research Initiative Programs.]

**D030**

**Performance Evaluation of Virus Transport Medium, CTM™ Kit through the Recovery and Viability Study of Various Viruses Including SARS-CoV-2**

Bong Soo Jeong<sup>1</sup>, Kye Seung Baek<sup>1</sup>, Taewan Kim<sup>2</sup>, Wonil Kim<sup>3</sup>, and Ki Tae Kim<sup>1\*</sup>

<sup>1</sup>R&D Center of Noble Biosciences, Inc., <sup>2</sup>School of Chemical and Biological Engineering and Institute of Chemical Process (ICP), Seoul National University, <sup>3</sup>Veterinary Diagnostic Center, Jeonbuk National University

The novel COVID-19 outbreak started in Wuhan, China and it has now become a global pandemic. The virus's transmission rate is affecting Asia, Europe and the United States, and the global economy, society as well as diplomatic relations have come to standstill. Suitable specimen transport with the most appropriate medical device from collection site to the laboratory is essential for accurate laboratory diagnostics. We have evaluated manufactured CTM™ Kit containing clinical virus transport medium and 2 swabs, nasopharyngeal and oropharyngeal swab with PhiX174, Enterovirus, AIV, HSV-1, *Mycoplasma pneumoniae* and SARS-CoV-2 from infected patients. Swab samples were placed in their respective transport medium tubes and held for 0, 24, 48 and 72 h at either 4°C or room temperature, 20–25°C followed by each assay method, quantitative Polymerase Chain Reaction and Enzyme Linked Immunosorbent Assay (ELISA) method. Holding time up to 10 days was set up to determine the extent of endpoint viability. Recovery studies using CTM™ Kit and comparative commercial product to determine the ability of the products to maintain viability of various strains of viruses have been processed. CTM™ Kit retain nucleic acid in the transport medium under different storage conditions and does not inhibit PCR reaction with high efficacy. The test procedures for quality control are based upon the quality control methods described in CLSI M40-A2 and others.

**D031**

**Outbreak of the Fowlpox in South Korea from 1998 to 2019**

Tuyet-Ngan Thai, Il Jang, Yong-Kuk Kwon, and Hye-Ryeong Kim\*

*Avian Disease Division, Animal and Plant Quarantine Agency*

Fowlpox is a common disease of chickens and cause clinical signs usually including development of skin lesions on the unfeathered areas as a cutaneous form, mucosal lesions involving the mouth, esophagus and trachea as a diphtheritic form. Fowlpox positive cases have been diagnosed 0.2~1.8% per year. However, in recent years, outbreaks of fowlpox have occurred in previously vaccinated flocks and increased in layer and breeder flocks. From 2016 to 2019, we diagnosed twenty cases of fowlpox through passive surveillance performed at Avian Disease Division of Animal and Plant Quarantine Agency. We analyzed outbreak situation of farms and the insertion of reticuloendotheliosis virus into fowlpox virus genomes.

**D032****Expression of a MARTX Toxin RtxA is Controlled by a CRP-coordinated Regulatory Network in *Vibrio vulnificus***

Zee-Won Lee, Seung-Ho Hwang, Garam Choi, and Sang Ho Choi\*

*Laboratory of Food Safety and Toxicology, Department of Agricultural Biotechnology, Seoul National University*

A multifunctional-autoprocessing repeats-in-toxin (MARTX) toxin is an essential virulence factor in many pathogens, including a fulminating human pathogen *Vibrio vulnificus*. It has been reported that H-NS and HlyU repress and derepress the expression of the MARTX toxin gene *rtxA* in *V. vulnificus*, respectively. However, little is known about other regulatory proteins and environmental signals involved in the *rtxA* regulation. In this study, we found that a leucine-responsive regulatory protein (Lrp) activates *rtxA* by binding directly and specifically to the *rtxA* promoter,  $P_{rtxA}$ . Phased hypersensitivity resulting from the DNase I cleavage of the  $P_{rtxA}$  regulatory region suggests that Lrp probably induces DNA bending in  $P_{rtxA}$ . Lrp activates *rtxA* in an independent manner with H-NS and HlyU, and leucine inhibits the Lrp binding to  $P_{rtxA}$  and reduces the Lrp-mediated activation. Furthermore, a cyclic AMP receptor protein (CRP) represses  $P_{rtxA}$ , and exogenous glucose alleviates the CRP-mediated repression. Biochemical and mutational analysis demonstrated that CRP binds directly and specifically to the upstream region of  $P_{rtxA}$ , which presumably alters DNA conformation in  $P_{rtxA}$  and thus represses *rtxA*. Moreover, CRP represses the expression of Lrp and HlyU by binding directly to their upstream regions, forming coherent feedforward loops with Lrp and HlyU. In conclusion, the expression of *rtxA* is controlled by a regulatory network comprising CRP, Lrp, H-NS, and HlyU in response to changes in host environmental signals such as leucine and glucose. This collaborative regulation enables the precise expression of *rtxA*, thereby enhancing the fitness and pathogenesis of *V. vulnificus* during the course of infection.

**D033****Establishment of Full-length Zika Virus Infectious cDNA Clone**

Jeong Yoon Lee, Sojung Bae, and Jinjong Myoung\*

*Molecular Virology, Korea Zoonosis Research Institute, Chonbuk National University*

Zika virus (ZIKV), belongs to Flaviviridae, is one of mosquito-borne emerging viruses and is associated with global pandemic. ZIKV raises severe diseases such as microcephaly in newborns and Guillain-Barré syndrome in adults. However, there is no medicines or vaccines for ZIKV infection. Furthermore, ZIKV pathogenesis and the evasion mechanisms against host immune responses have been unclear. The purpose of this study is to create a full-length ZIKV infectious cDNA clone as a great tool to develop a ZIKV vaccine and investigate host-virus interactions. ZIKV Puerto Rico strain (GenBank: KX087101.2) genome was divided into four fragments synthesized using reverse genetics techniques. The four fragments were assembled on a customized low-copy plasmid, pACYC184SB, using Gibson Assembly technique. Moreover, ZIKV deletion recombinants were constructed to examine as potential vaccine candidates. GFP-expressing ZIKV recombinants were also built as valuable tools for researches of ZIKV pathogenesis *in vitro* and *in vivo* models.

**D034****Modulation of the IFN- $\beta$  and NF- $\kappa$ B Signaling Pathways by Chikungunya Virus-encoded Genes**

Sojung Bae, Jeong Yoon Lee, and Jinjong Myoung\*

*Molecular Virology Laboratory, Korea Zoonosis Research Institute, Jeonbuk National University*

Chikungunya virus (CHIKV) is single-stranded positive-sense RNA virus, belonging to a member of the genus alphavirus and *Togaviridae* family. Since the first outbreak occurred at Tanzania in 1952, there have been multiple outbreaks throughout the half of the 20th century in Asia and Africa countries. CHIKV is transmitted from *Aedes aegypti* and *Aedes albopictus* mosquitoes. It generates multiple symptoms, including headache, fever, muscle pain, joint swelling, and a rash. However, the pathogenesis of chikungunya infection and phenomenon of the viral immune evasion has been poorly understood. Moreover, the roles of individual CHIKV gene against host innate immune responses have been poorly studied. To identify modulation of the type I interferon (IFN) by CHIKV-encoded genes, each CHIKV gene was screened for their effects on immune responses with determining IFN- $\beta$  promoter and NF- $\kappa$ B promoter activities. Here we show that CHIKV nsP2, E2 and E1 strongly antagonize MDA5/RIG-I mediated molecules which are distinct downstream of the signaling pathway. In addition, frame-shifting event resulting in production of TF protein is able to suppress MDA5 and TBK1 resulting in down-regulation of IFN- $\beta$  promoter activity. Understanding the modulation of the IFN- $\beta$  and NF- $\kappa$ B pathways by CHIKV infection may help develop virus-specific therapeutics and vaccines.

**D035****Changes in Fecal Microbiota Composition by Viral Infection in Korean Calves**Yeonsu Oh<sup>1</sup>, Dohyeon Yu<sup>2</sup>, Hak-Jong Choi<sup>3</sup>, and Jinho Park<sup>4\*</sup>*<sup>1</sup>College of Veterinary Medicine and Institute of Veterinary Science, Kangwon National University, <sup>2</sup>College of Veterinary Medicine, Gyeongsang National University, <sup>3</sup>Microbiology and Functionality Research Group, World Institute of Kimchi, <sup>4</sup>College of Veterinary Science, Chonbuk National University*

To compare the changes in fecal microbiota composition by the cause of the viral infection, diarrhea caused by bovine rotavirus (BRV) or bovine corona virus (BCoV) infection was analyzed by comparing the feces of 4 Korean cattle calves and 7 of normal calves. A total of 864,232 reads were obtained (average of 57,6158 leads per head), and a rarefaction curve showing the rate of increase in the number of OTUs according to the number of sequences was confirmed. As a result, a sufficient number of OTUs for analysis was secured. As a result of confirming the rarefaction curve showing the increase rate of the number of Operational Taxonomic Units (OTUs) according to the number of acquired sequences, a sufficient number of OTUs required for analysis was secured. Through the results of alpha- and beta-diversity, taxonomic composition analysis, and LEfSe calculation, the structural differences in the microbial community due to the diarrhea caused by the BRC or BCoV were observed. It is considered that it is necessary to discover candidate microorganisms for preventing diarrhea by selecting and securing useful microorganisms to promote intestinal health condition of Korean calves.

[This research was supported by Technology Development Program (Project No. 1116043-1) for Bio-industry, Ministry for Agriculture, Food and Rural Affairs, Republic of Korea.]

**D036****Selection of Useful Probiotic Candidates to Improve Diarrheic Condition through Microbiota Analysis**Yeonsu Oh<sup>1</sup>, Dohyeon Yu<sup>2</sup>, Hak-Jong Choi<sup>3</sup>, and Jinho Park<sup>4\*</sup>

<sup>1</sup>College of Veterinary Medicine and Institute of Veterinary Science, Kangwon National University, <sup>2</sup>College of Veterinary Medicine, Gyeongsang National University, <sup>3</sup>Microbiology and Functionality Research Group, World Institute of Kimchi, <sup>4</sup>College of Veterinary Science, Chonbuk National University

*Lactobacillus* was selected and isolated as a useful microorganism group in the intestine through analysis of the intestinal microbial community of the normal calf and diarrhea calf. In order to isolate various types of *Lactobacillus*, the *Lactobacillus gasseri* group, *L. amylovorus* group, and *L. reuteri* group having a relative frequency of 10% or more in normal calves were selected as the main isolation targets, and the culture conditions were accomplished. *L. amylovorus* and *L. helveticus*, which were previously integrated into the *Lactobacillus helveticus* group, as the database was upgraded, were currently identified.

Through these results, *Lactobacillus* was selected as a useful microorganism candidate group in the intestine of Hanwoo calves, and 7 *Lactobacillus* spp. were successfully isolated and cultured. In order to supply to Korean calves, additional analysis is remained for further study; microbial growth characteristics, stability including antibiotic susceptibility testing, hemolysis, etc., and full-length genetic analysis.

[This research was supported by Technology Development Program (Project No. 1116043-1) for Bio-industry, Ministry for Agriculture, Food and Rural Affairs, Republic of Korea.]

**D037****An Integrative Systems Genetic Analysis of Atherosclerosis and Gut Microbiota**Myungsuk Kim<sup>1,2</sup>, M. Nazmul Huda<sup>2</sup>, Excel Que<sup>2</sup>, Erik R. Gertz<sup>2</sup>, and Brian J. Bennett<sup>1,2\*</sup>

<sup>1</sup>Department of Nutrition, University of California, Davis, CA, 95616, USA, <sup>2</sup>USDA-ARS-Western Human Nutrition Research Center, Davis, CA, 95616, USA

Atherosclerosis is a precipitating event in the development of cardiovascular disease. Recent studies report that gut microbiota contributes to the pathogenesis of cardiovascular disease, including metabolic syndrome. While host genetic variants are known factors that affect atherosclerosis development and gut microbiota composition, the mechanisms underlying genetic variations are not yet clear. Here, we interrogated atherosclerosis regulatory networks in hyperlipidemic Diversity Outbred mice to reveal key insights into control of atherosclerosis using system genetic approaches of cardio-metabolic traits, microbiome and liver transcriptome. We collected offspring (238 female and 234 male mice) from a cross between transgenic male C57BL/6J mice, which were made susceptible to atherosclerosis by microinjection of human apolipoprotein E-Leiden and cholesterol ester transfer protein genes, and ~200 female DO mice, a population derived from 8 inbred strains. We fed the offspring a high fat/cholesterol diet for 12 weeks. Our results include identifying abundance of fecal microbial taxa associated with atherosclerotic traits, defining the functionality of genes associated with the atherosclerotic traits and gut microbiota, and identifying signatures of functional gene variants predicted to modulate those traits. Trans-omic analysis facilitated identification of Ptpk as a previously unknown regulatory gene for atherosclerotic traits and *Lactococcus* abundance. Collectively, this study provides a rich resource for investigating the pathogenesis of atherosclerosis and suggests an opportunity to discover therapeutics and biomarkers in the setting of hyperlipidemia.

**E001**

**Microbiota is Associated with Anti-stress in the Intestine and Brain**

Junho Lee

*Chonnam National University*

During psychological stress, the intestinal microflora changes, causing serious health problems worldwide. Integrity of the intestinal barrier and blood brain barrier can regulate the process of bacterial translocation and provide the nervous system with real-time information about the environment. We use a rodent model to explore the association between damaged intestinal and blood brain barriers and altered fecal microflora under psychological stress to improve understanding of the camphor axis. Here, clues converge to control the basic developmental processes of the intestine and brain, such as barrier function. This study presents a new direction for investigating the etiology of emotional disorders and the formula of clinical treatment.

**E002**

**Microbiota is Permeability Regulator in Brain**

Junho Lee

*Chonnam National University*

Microbial guns and microbial guns and destruction of the intestinal-brain axis have been associated with various metabolic, immunological, physiological, neurodevelopmental and neuropsychiatric disorders. After a brief review of the relevant literature, we have intestinal serotonin that is produced by intestinal intestinal cell cells, and is picked up and stored by circulating platelets in the intestines, brain and other organs. Intestinal serotonin can also act as a lasting regulatory signal, such as hormones for the whole body, including the brain. This regulatory signal function is mediated by platelets and depends primarily on the actual state of health of the intestine. This hypothesis can partially explain why intestinal dysbiosis can be linked to neurodevelopmental and neuropsychiatric disorders, as well as to various human pathological conditions.

**E003****Microbiome Regulate the Depression**

Junho Lee

*Chonnam National University*

The human intestinal microflora were used to assess whether individuals with a depression phenotype can be identified from healthy reference subjects. The gastrointestinal tract is stimulated by exogenous neurons that connect with its intestinal and central nervous systems. Nerve distribution detects and responds to various stimuli in the gastrointestinal tract, which not only regulates motility and secretion, but also affects physiology, behavior and immunity. The basic mechanisms of gastrointestinal neuron formation have begun to emerge. Research on the things that keep plastics over the life of the organism still remains.

**E004****Identification and Characterization of VapBC Toxin-antitoxin Systems in *Bosea* sp. PAMC26642**Hyerin Jeon<sup>1</sup>, Changmin Oh<sup>1</sup>, and Jihwan Hwang<sup>1,2\*</sup><sup>1</sup>*Department of Integrated Biological Science, Pusan National University,* <sup>2</sup>*Department of Microbiology, Pusan National University*

Toxin-antitoxin (TA) systems are genetic elements encoding a toxin and its antidote, antitoxin. Toxin proteins interfere with diverse cellular functions and noncoding RNA or protein antitoxins counteract the activity of the toxin. These systems are widely spread in bacterial and archaeal genomes. TA systems are involved in diverse cellular events, such as stress response, programmed cell death, and bacterial pathogenicity. Depending on the biochemical nature of the antitoxin (protein or RNA) and how they control the activity of the toxin, TA systems are currently divided into six types.

*Bosea* is a Gram-negative bacterium belonging to family *Bradyrhizobiaceae*, which has been isolated from diverse environments such as soils, sediments, and hospital water. *Bosea* sp. PAMC26642 strain used in this study was isolated from the Arctic lichen *Streocaulon* sp. Twelve putative type II TA systems were predicted in the complete genome of *Bosea* sp. PAMC26642, and three VapBC homologs, AXW83\_01400-01405, AXW83\_11460-11465, and AXW83\_12680-12685 characterized in this study. One of them, the putative VapC toxin AXW83\_01405 inhibited cell growth only in low temperature, and the growth was recovered by co-expression of putative antitoxin AXW83\_01400. Furthermore, overexpression of AXW83\_01405 induced morphological changes that the cells were elongated. Purified VapC was able to degrade 23S and 16S rRNA *in vitro*, and when overexpressed, had activity of mRNA degradation *in vivo*.

**E005****Elucidation of a Novel Role of YebC in the Colanic Acid Production of *Escherichia coli* *bipA*-deletion**Eunsil Choi<sup>1,2</sup>, Hyerin Jeon<sup>3</sup>, Chang Min Oh<sup>3</sup>, and Jihwan Hwang<sup>3,4\*</sup><sup>1</sup>*Microbiological Resource Research Institute, Pusan National University*, <sup>2</sup>*Department of Microbiology, Pusan National University*, <sup>3</sup>*Department of Integrated Biological Science, Pusan National University*, <sup>4</sup>*Department of Microbiology, Pusan National University*

The BipA protein is ubiquitously conserved in various bacterial species and belongs to the translational GTPases. The function of *Escherichia coli* BipA is not essential for cell growth under normal growth conditions. However, cultivation of  $\Delta bipA$  cells at 20°C leads to not only cold-sensitive growth defects but also several phenotypic changes such as ribosome assembly, capsule production, lipopolysaccharide (LPS) and motility, suggesting its global regulatory roles. Previously, our genomic library screening also revealed that the overexpression of r-protein L20 partially suppresses the cold-sensitive growth defects by recovering ribosomal abnormality in the  $\Delta bipA$  cells at 20°C.

Here, we explored another genomic library clone containing *yebC* whose function was predicted to be a transcriptional factor not associating with ribosome biogenesis. Interestingly, the overexpression of *yebC* in the  $\Delta bipA$  cells diminished capsule synthesis at 20°C without recovering defects in ribosome and motility, indicating that YebC may be involved in the regulation of capsule production.

In this study, we collectively investigated the impacts of *bipA*-deletion on LPS, capsule, biofilm, and motility, and using this mutant strain revealed the novel role of YebC in capsule production at 20°C. Furthermore, our findings suggest that the ribosomal defects and increased capsule synthesis independently contribute to the cold-sensitivity of the  $\Delta bipA$  cells, implying its multiple regulatory roles.

**E006****Functional Analysis of Ornithine Lipid Derivatives in *Pseudomonas aeruginosa***Xi-Hui Li<sup>1</sup>, Soo-Kyoung Kim<sup>2</sup>, and Joon-Hee Lee<sup>1\*</sup><sup>1</sup>*Department of Pharmacy, College of Pharmacy, Pusan National University*, <sup>2</sup>*Department of Cell Biology & Molecular Genetics, University of Maryland, USA*

Ornithine lipids (OLs) are bacteria-specific lipids that are found in the outer membrane of Gram (-) bacteria and increased as surrogates of phospholipids under phosphate-limited conditions. In *P. aeruginosa*, bacterial surface charge and hydrophobicity were changed with increased OLs levels, which reduced bacterial susceptibility to antibiotics and antimicrobial peptides (AMPs), interfered with the binding of macrophages to bacterial cells, and enhanced bacterial biofilm formation. According to our previous works, when grown under low phosphate conditions, *P. aeruginosa* became more persistent in the treatment of antibiotics and AMPs in an *olsBA* (OL synthesis operon)-dependent manner. The exogenous addition of OLs provided bacteria with persistence and attenuated *P. aeruginosa* virulence. In host cells, OLs reduced the productions of inflammatory factors (iNOS, COX-2, PGE<sub>2</sub>, and NO) and increased intracellular Ca<sup>2+</sup> release. All these results suggest that bacterial OL plays important roles in bacteria-host interaction in a way that enhances bacterial persistence and develops chronic adaptation of infection. Lyso-OLs (LOLs), an intermediate product of OL biosynthesis increased the membrane potential dramatically, indicating that the LOLs, not OL, may affect the membrane potential mainly. Meanwhile, increased LOLs also reduced the pyocyanin levels and enhance the biofilm formation of *P. aeruginosa*, suggesting that LOLs may play important roles in bacteria-host interaction.

**E007****Anthranilate Effects on Virulence and Antibiotic Resistance of *Pseudomonas aeruginosa***

Hyeon-Ji Hwang, Xi-Hui Li, and Joon-Hee Lee\*

*Department of Pharmacy, College of Pharmacy, Pusan National University*

Anthranilate is an important intermediate for the synthesis of tryptophan and *Pseudomonas* quinolone signal (PQS), and metabolized by anthranilate dioxygenase complex (*antABC* gene products) via TCA cycle. We previously demonstrated that anthranilate causes biofilm dispersal in various bacteria. We now demonstrate that anthranilate is a key factor in pathogenicity-related physiology of *P. aeruginosa* (biofilm formation, antibiotic resistance, and virulence). We found that the level of anthranilate appears to peak because it suddenly increased at stationary phase and decreased again as the stationary phase lasted. Biofilm formation, antibiotic susceptibility, and virulence of *P. aeruginosa* were compared before-and-after the anthranilate peak. Our results demonstrated that this anthranilate peak blocks early biofilm formation and the mature biofilm forms only after the anthranilate peak disappears. Antibiotic resistance and virulence was increased after the anthranilate peak. Deletion of the *antABC* gene blocked the degradation of anthranilate, so did not decrease the anthranilate level, which abolishes the anthranilate peak. *antABC* mutant showed less biofilm formation, increased susceptibility to various antibiotics, and attenuated virulence even after the anthranilate peak. In addition, the exogenous addition of anthranilate from early stage of growth enhanced the susceptibility to antibiotics. Interestingly, this increased anthranilate-mediated susceptibility to antibiotics was disappeared in *antR* mutant. Our results show that there are anthranilate-mediated physiological changes in stationary phase that we have not known so far.

**E008****Engineering of Reductase Enzymes for the Characterization of Electron Transfer**

Chungwoon Yoon, Dong-Heon Lee, and Seung Jae Lee\*

*Department of Chemistry and Institute for Molecular Biology and Genetics, Jeonbuk National University*

Methane is considered as one of the most important greenhouse gases owing to its 20-fold higher heat capacity compared with that of CO<sub>2</sub>. Preliminary studies have aimed to elucidate the mechanisms of soluble methane monooxygenase (sMMO) involved in this extremely stable C–H activation (104.9 kcal/mol) through intermediate studies, advanced spectroscopies, and structural researches, mostly in *Methylosinus trichosporium* OB3b and *Methylococcus capsulatus* Bath, although mechanistic studies are still required.

In this study, *M. sporium* 5 was cultured in a tightly regulated NMS media by supplying methane and air to understand its growth and the expression levels of multi-component enzymes. MMOH, MMOB and MMOR were found to be highly expressed in *M. sporium* 5, and it was purified to measure specific enzyme activities (SEA). These results showed that *M. sporium* 5 exhibits optimal activity at pH 7.5. The electron transfer environment of MMOR is crucial for the activity of sMMO, and different acidities may change the electron transfer environment. *In vitro* activity measurements demonstrated that alkanes, halogens, benzene, and toluene are oxidized through sMMO, and 2 mol equivalents of MMOB showed optimal activity.

[This research was supported by the C1 Gas Refinery Program (NRF-2015M3D3A1A01064876) and Basic Science Research Program of NRF Korea by the Ministry of Education (2017R1A6A1A03015876).]

## E009

### **Structural Conformation of sMMO through Interactions with MMOG**

Chae Min Lee, Ka Young Son, Dong-Heon Lee, and Seung Jae Lee\*

*Department of Chemistry and Institute for Molecular Biology and Genetics, Jeonbuk National University*

The catalytic activities of soluble methane monooxygenases (sMMO) are regulated by the interactions of between hydroxylase and other auxiliary enzymes including regulatory components and reductase. The physical map of sMMO genes has confirmed that *mmoG* gene positioned 5'- or 3'-region in sMMO and the expressed sequence of amino acids show high sequence homologies with reported chaperone.

This chaperone, MMOG, is successfully expressed to understand its functions in sMMO system from synthesized gene from *M. sporium* 5. MMOG is purified with high purity and its biophysical aspects were investigated. The UV-vis spectroscopic studies proved that MMOG does not interact with NADH. The binding affinity is measured through the fluorescence measurements through Trp quenching. These results indicate that MMOG will share the binding site, di-iron active site, like regulatory or inhibitory enzymes and reductases. The functional consequence of MMOG is important to elucidate its physiological functions and catalytic activities are investigated whether it accelerates or retards specific enzyme activity. In this presentation, the biophysical aspect of MMOG will be discussed to understand functional and structural roles in sMMO system.

[This research was supported by the C1 Gas Refinery Program (NRF-2015M3D3A1A01064876) and Basic Science Research Program of NRF Korea by the Ministry of Education (2017R1A6A1A03015876).]

## E010

### **Influence of Substrate in the Hydroxylase Activities and Its Functional Consequences to Product-formations**

Yun Ha Hwang, Chungwoon Yoon, Heeseon Yoo, Dong-Heon Lee, and Seung Jae Lee\*

*Department of Chemistry and Institute for Molecular Biology and Genetics, Jeonbuk National University*

The specific enzymatic activities (SEA) of sMMO are widely investigated to understand its enzymatic activity and turnover rate to understand the influence by substrates including hydrocarbons, O<sub>2</sub>, electrons, and proton. The ratio between substrates and enzymes need to be further investigated to understand its possible influences during expression of hydroxylase and other auxiliary enzymes.

The expression of MMOH is described based on the ratio of two substrates including CH<sub>4</sub> and O<sub>2</sub>. The expression level in sMMO is important for the purification of this homodimer ( $\alpha_2\beta_2\gamma_2$ ) and the complex formation needs to discover further mechanism studies. The methane oxidation is performed in  $\alpha$ -subunit and  $\beta$ -subunit generates specific interactions for the dimeric formation of MMOH. The function of  $\gamma$ -subunit is not well quite understood, but this small-subunit only detected in water-soluble region after cell-lysis. The injection volume of CH<sub>4</sub> and O<sub>2</sub> affect the volume of soluble portion of MMOH based on the quantitative analysis. In addition, the differences of these species depend on the concentration of  $\gamma$ -subunit. The catalytic activities of enzymes are also influenced by the ratio of reductase components and affected by the concentration of CH<sub>4</sub> and O<sub>2</sub> in buffer.

[This research was supported by the C1 Gas Refinery Program (NRF-2015M3D3A1A01064876) and Basic Science Research Program of NRF Korea by the Ministry of Education (2017R1A6A1A03015876).]

**E012****Electron Transfer of sMMO and Its Functional Effects in FAD Binding Domain from *M. sporium* 5**

Ka Young Son, Yun Ha Hwang, Dong-Heon Lee, and Seung Jae Lee\*

*Department of Chemistry and Institute for Molecular Biology and Genetics, Jeonbuk National University*

The resting state of MMOH is reduced by the addition of electrons through MMOR and reduced di-iron active site can activate O<sub>2</sub> for C–H activation. The essential intermediates can be generated by the O<sub>2</sub> activation from reduced MMOR, but the accurate electron-transfer pathways need to be further elucidated. MMOR has FAD/NADH-binding domain and [2Fe-2S] cluster to transfer electrons from NADH to di-iron active site. Although the structural information is available from solution NMR of each domain, the overall structure needs to be further discovered.

MMOR is successfully expressed and purified to understand biophysical aspects. The presence of FAD and [2Fe-2S] cluster is confirmed by the measurement of FAD content, ferrozine assay and UV-Vis spectra. The crystallization of MMOR provides structural insight of truncated version of MMOR-FAD binding domain generated by protease activity. The residues including S159, Y160, S161, and S184 generated hydrogen bonds with FAD. The mutated MMOR was successfully purified and confirmed the modification of iron contents and concentration of FAD. These results confirmed that the expression of reductase components affects the cofactor interactions of MMOR and the modification of MMOR-FAD affects the structural components of iron-sulfur clusters.

[This research was supported by the C1 Gas Refinery Program (NRF-2015M3D3A1A01064876) and Basic Science Research Program of NRF Korea by the Ministry of Education (2017R1A6A1A03015876).]

**E013****Identification and Functional Analysis of Small Protein CydY in *Salmonella enterica* Serovar Typhimurium**

Choa Lee and Iel Soo Bang\*

*Department of Microbiology and Immunology, Chosun University School of Dentistry*

Cytochrome *bd* quinol oxidases, a terminal oxidase enzyme complex, is necessary for the survival of *Salmonella enterica* serovar Typhimurium under low O<sub>2</sub> conditions. The cytochrome *bd* complex contains three subunits, CydA, CydB and CydX. Recently, it has been reported that the small protein CydX is required for the assembly and function of the cytochrome *bd* complex in *S. Typhimurium* under stress conditions, including exposure to β-mercaptoethanol, nitric oxide. We investigated whether a putative gene, *cydY*, located behind *cydABX* operon could affect the function of cytochrome *bd* subunits. To test this question, we constructed  $\Delta cydY$  mutant and *cydY*-overexpressing clone, and investigated whether the *S. Typhimurium* CydY is required for cytochrome *bd* complex activity and cytochrome *bd*-related metabolism. When grown on Luria-Bertani medium,  $\Delta cydX$  mutant exhibited mixed colony sizes, with some small-colony variants, but  $\Delta cydY$  mutant showed colony size comparable to those of the WT. And there was no significant difference between  $\Delta cydY$  mutant and the complementation of the  $\Delta cydY$  mutation with a *cydY*-encoding plasmid, and it was similar to WT. The growth of the  $\Delta cydY$  mutant with β-mercaptoethanol did not impair compared with that of the  $\Delta cydX$  mutant. Based on this finding, compared to previously studied about CydX, the small protein CydY has no significant effect to the role of cytochrome *bd* subunits.

[Supported by NRF of Korea (grants NRF-2018R1D1A1B07044085, NRF-2013R1A1A2012937).]

**E014****Important DNA Recognition Helix Site of the NsrR Regulator for Nitric Oxide Sensing in *Salmonella enterica* serovar Typhimurium**

Hee Jeong Park and Iel Soo Bang\*

*Department of Microbiology and Immunology, Chosun University School of Dentistry*

Bacteria can be exposed to antimicrobial radicals, nitric oxide (NO) and NO-mediated reactive nitrogen species that can damage various bacterial macromolecules, causing abnormal metabolism and consequent bacteriostasis. To cope with nitrosative stress, many bacteria have evolutionally conserved *hmp* gene encoding an ancestral globin, flavohemoglobin Hmp that metabolize NO to less toxic molecules. And flavohemoglobin expression is mainly regulated by the [Fe-S] cluster-containing repressor protein NsrR that binds to the *hmp* promoter. This NsrR-*hmp* interaction has been proved essential for bacterial fitness and virulence in mouse infection models under NO-producing conditions. But little is known about the molecular details by which NsrR senses NO and derepresses *hmp* transcription. By using methodology including random and site-directed mutagenesis techniques, gene transcription analyses, and NO metabolism kinetics, we identified key amino acid residues in the N terminus of the DNA recognition helix site of NsrR for activation of *hmp* transcription and NO metabolism of *S. Typhimurium* in response to NO exposure, and revealed divergence of these amino acids in different bacterial species. Results suggest that divergence of amino acid residues in the DNA recognition helix of NsrR may provide new insight on how bacterial species have evolved to occupy their ecological niches in terms of NO pressure.

[Supported by the NRF of Korea (grants NRF-2013R1A1A2012937, NRF-2016R1A2B1015928).]

**E015****The Periplasmic Chaperone Spy Affects the Iron Transport of *Salmonella enterica* Serovar Typhimurium**

Hye Won Jeong and Iel Soo Bang\*

*Department of Microbiology and Immunology, Chosun University School of Dentistry*

Spy is known as a chaperone protein expressed in the periplasm when spheroplasts were formed by the partial removal of the cell wall of the *Salmonella enterica* Serovar Typhimurium and plays essential role for *Salmonella* pathogenesis. It has been reported that iron acquisition is essential for the growth and virulence of *S. Typhimurium*. TonB is located on the inner membrane, plays an important regulatory role for the activity of iron-transporting proteins including CirA, FhuA and IroN in periplasm. CirA, FhuA, IroN is located in the outer membrane and recognizes iron and accepts it to periplasm. Our proteomic analysis of secreted proteins showed that in *spy* mutant iron transporter proteins are less secreted, compared to those from WT. In this study, we examined if Spy affected iron-dependent growth of *S. Typhimurium*. All salmonella strains used in this study are WT, *spy* and *spy* clone. WT, *spy* and *spy* clone strains were grown in minimal media, and 2-2' bipyridyl was treated with 50  $\mu$ M and 75  $\mu$ M, respectively. The growth of *spy* was lower than that of WT and *spy* clone at 75  $\mu$ M, but it was similar to that of WT at 50  $\mu$ M. Transcription of genes related to iron-transport showed no significant differences in WT, *spy*, and *spy* clones, suggesting that Spy would be directly involved in the TonB-dependent regulation of iron transporters.

[Supported by the National Research Foundation of Korea (grants NRF-2018R1D1A1B07044085 and NRF-2016R1A2B1015928).]

**E016****Functional Investigation of Organic Hydroperoxide Resistance-related Protein Ohr in *S. aureus***

Jang Wan Son, Maria Florencia Colavita, Su hyun Ryu, and Jin Won Lee\*

*Hanyang University*

*Staphylococcus aureus* is major human pathogen of increasing importance as a result of the spread of antibiotic resistance. Research on the survival of *S. aureus* is likely to be the solution to many of the diseases or problems caused by the bacteria. ROS, reactive oxygen species, is one of the environmental stress that affects organism survival. ROS include hydrogen peroxide, superoxide anion, hydroxyl radical, and organic hydroperoxides. Since ROS can damage DNA, proteins, and lipids, resistance to ROS is important for bacterial survival. Bacteria have some enzyme-mediated pathway to resist ROS such as catalase, superoxide dismutase and so on. Among them, organic hydroperoxide resistance protein Ohr is known as involving in organic hydroperoxide protection. We found *B. subtilis* homolog in *S. aureus*. If we can accurately understand structure and function of Ohr, that will present the potential for a viable therapeutic drug. Thus, we want to know what Ohr function is, especially whether Ohr has defense mechanism for the organic hydroperoxides. Here we show that Ohr has a function related to defense against the organic hydroperoxide. This study was supported by Science Research Center grant (NRF-2018R1A5A1025077) and 2019R1F1A1061890.

**E018****Elucidation of Distinct Functions of Four Peptidoglycan Endopeptidase, MepM, MepS, MepH, and PbpG, through Their Specific Phenotypes**Si Hyoung Park<sup>1</sup>, Yung Jae Kim<sup>1</sup>, Han Byeol Lee<sup>1</sup>, Yeong-Jae Seok<sup>2</sup>, and Chang-Ro Lee<sup>1\*</sup><sup>1</sup>*Department of Biological Sciences, Myongji University,* <sup>2</sup>*Department of Biological Sciences and Institute of Microbiology, Seoul National University*

Peptidoglycan (PG) is an essential component of the bacterial exoskeleton that plays a pivotal role in the maintenance of cell shape and resistance to cell lysis under high turgor pressures. PG is composed of repetition of N-acetylmuramic acid and N-acetylglucosamine, and a mesh-like structure with cross-linking between pentapeptide stem of N-acetylmuramic acid of glycan strands and pentapeptide stem of other glycan strands. The synthesis and degradation of PG must be tightly regulated during bacterial cell growth and division. PG hydrolases show high redundancy in many bacteria, including *Escherichia coli*. In this study, we showed that specific phenotype of PG endopeptidases, MepM and MepS. Deletion mutant of *mepM* and *mepS* has salt sensitivity and EDTA sensitivity phenotype, respectively. In complementation test, *mepM* mutant was restored by MepM only, whereas *mepS* mutant was restored by MepS and overexpression of MepH, PbpG, and MepM. Using domain swapped MepM variates and MepS variates, we find out that phenotypes of *mepM* and *mepS* mutant were related to their specific cellular localization and specific domain. Finally, using specific phenotypes of *mepM* and *mepS* mutants, we found that PBP1a can be affected by MepM and MepH, and that PBP1b can be affected by MepM and MepH as well as MepS and PbpG. Especially, deletion mutants of PBP1a or PBP1b shows a similar phenotype to *mepM* mutant, suggesting the importance of MepM on PG synthesis. Therefore, our results indicate that each PG endopeptidases have a distinct role in PG synthesis, depending on its distinct domains and cellular localizations.

**F001**

**Molecular Interaction between Methicillin-Resistant *Staphylococcus aureus* (MRSA) and Chicken Breast Reveals Enhancement of Pathogenesis and Toxicity for Food-borne Outbreak**

Han-Young Chung, Tae Young Kim, Hokyung Byun, and Sang Ho Choi\*

*National Research Laboratory of Molecular Microbiology and Toxicology, Department of Agricultural Biotechnology, Seoul National University*

To study pathogenesis and toxicity of *Staphylococcus aureus* in foods, FORC\_062 was isolated from a human blood sample and complete genome sequence has a type II SCCmec gene cluster and a type II toxin-antitoxin system, indicating an MRSA strain. Its mobile gene elements has many pathogenic genes involved in host infection, biofilm formation, and various enterotoxin and hemolysin genes. Clinical MRSA is often found in animal foods and ingestion of MRSA-contaminated foods causes human infection. Therefore, it is very important to understand the role of contaminated foods. To elucidate the interaction between clinical MRSA FORC\_062 and raw chicken breast, transcriptome analysis was conducted, showing that gene expressions of amino acid biosynthesis and metabolism were specifically down-regulated, suggesting that the strain may import and utilize amino acids from the chicken breast, but not able to synthesize them. However, toxin gene expressions were up-regulated, suggesting that human infection of *S. aureus* via contaminated food may be more fatal. In addition, the contaminated foods enhance multiple-antibiotic resistance activities and virulence factors in this clinical MRSA. Consequently, MRSA-contaminated food may play a role as a nutritional reservoir as well as in enhancing factor for pathogenesis and toxicity of clinical MRSA for severe food-borne outbreaks.

**F002**

**Protein Kinase Pathways for Translational Regulation Mediated by mRNA Decapping Activator Dhh1**

Jae Hee Hwang, Dae Hee Jung, and Jin Mi Kim\*

*Department of Microbiology and Molecular Biology, College of Bioscience and Biotechnology, Chungnam National University*

DEAD-box RNA helicase Dhh1 is an mRNA decapping activator that performs translation repression and mRNA decay through interaction with both decapping and deadenylase complexes. Dhh1 has an important influence on the translational regulation of the mating-specific transcription factor, Ste12. Phosphoproteomic analysis have identified three phosphorylation sites (Thr10, Ser14, and Thr16) in the N-terminal domain of Dhh1. Our previous study analyzed the phosphor-deficient (T10A, S14A, and T16A) and phosphor-mimetic (T10E, S14E, and T16E) mutations which have the amino acid residue alanine or glutamate at the threonine or serine residue of the N-terminal Dhh1. Phosphorylation of Thr16 residue decreased the interaction of Puf6 and Dhh1 and affected Ste12 protein expression. Thr10 phosphor-deficient mutation (T10A) showed a defect in mating-specific process. To identify the upstream protein kinases responsible for Dhh1 phosphorylation, we analyzed the kinase overexpression mutant strains (*TPK2*, *FUS3*, *SNF1*, *STE20* overexpression). Ste12 protein expression increased in *FUS3*, *SNF1* and *STE20* overexpression and decreased in *TPK2* overexpression. Phosphorylation of Thr16 by Tpk2 was confirmed and this phosphorylation appeared to affect the interaction of Puf6 and Dhh1. Effects of Fus3 on Ste12 protein expression and interaction of Puf6 and Dhh1 are under investigation.

**F003****Roles of Apoptosis Related Genes in Atg8-mediated Autophagy**

Jang Won Lee

*Department of Bioscience and Biotechnology Graduate School, Major of Microbiology, Chungnam National University*

Autophagy, one of the programmed cell death, breaks down the cell's small organs on its own and commits suicide in situations where cells are not suitable for living, such as lack of nutrients or infection with pathogens. This creates a double membrane called autophagosome that wraps around the organelle trying to decompose. Checking the formation of autophagosome, which appears during the autophagy process, is typical method to observe autophagy. To analyze the autophagosome and follow that process, we conducted the experiment of using GFP-ATG8. ATG8 is known primarily as the hall marker of autophagy. The exact mechanism of interaction between apoptosis and autophagy has not yet been revealed, but they are not completely independent elements. In this study, *CaMCA1*, known as the metacaspase, *KEM1*, and *LPD1*, the genes that appear to affect the apoptotic phenotype, have been investigated. Using GFP-ATG8 strains, western blot assay and florescent microscopic observation will be conducted for measuring autophagic phenotypes.

**F004****mRNA Localization and Translation Mediated by Puf Protein Families**

Eunyeong Hwang, Daehee Jung, and Jinmi Kim\*

*Department of Microbiology and Molecular Biology, College of Bioscience and Biotechnology, Chungnam National University*

The Puf family of RNA-binding proteins play critical roles in post-transcriptional gene regulation by modulating the target mRNAs. PUFs contain Pum-HD domain which directly binds to specific recognition sequences in the 3' or 5' untranslated region (UTR) of mRNA. The roles of PUFs include differentiation, development, germline function, cell cycle, and mitochondrial biogenesis throughout yeast, *Drosophila*, *C. elegans*, and Humans. In yeast *Saccharomyces cerevisiae* Puf6 protein directly interacts with *ASH1* mRNA and regulates its localization and translation. These binding and translation inhibition require the interaction of the Puf6 protein with the UUGU elements in the 3'UTR of the *ASH1* mRNA. In our previous study, Puf6 protein has been shown to mediate the translational repression of Ste12 transcription factor. Our sequence analysis of the *STE12* mRNA identified UUGU sequences located at 9, 15, and 124 nucleotides upstream of the *STE12* start codon. We investigated whether the putative Puf-binding sites in the 5'UTR of the *STE12* mRNA are important for the translation and localization. Three UUGU sequences are mutated to UACU. The Ste12-HA protein level was significantly higher in a wild-type strain carrying the mutant 5'UACU-*STE12*-HA than the wild-type 5'UUGU-*STE12*-HA. These results suggest that three UUGU elements in the 5'UTR of the *STE12* mRNA are critical for the regulation of *STE12* translation. Ste12 protein localization is under investigation.

**F005**

**Cohesin-associate Factors Modulate Meiotic Recombination and Chromosome Morphogenesis during Yeast Meiosis**

Jeong Hwan Joo and Keun Pil Kim\*

*Department of Life Sciences, Chung-Ang University*

Cohesion is composed of 3 main subunit, Smc1, Smc3 and klesin subunit, Scc1 and cohesion associated protein. Cohesins are ring-shaped multifunctional protein complex which regulates stabilization of chromosome structure and allows proper segregation of chromosomes during cell division. During meiosis, cohesins not only mediate sister chromatid cohesion but also are prominent components of chromosome structural axes and important mediators of the local DNA events of recombination. In this study, we explore the roles of cohesion-associated protein, Pds5, during meiosis, and its functional relationships with the meiosis-specific  $\alpha$ -kleisin, called Rec8, and Rad61/WAPL which negatively regulates cohesin functions. First, we analyzed Zip3 focus which interacts initiation of synapsis to meiotic recombination. in the presence or absence pCLB2-PDS5, rec8 $\Delta$  or rad61 $\Delta$ . Rec8 and Pds5 have affected in formation of COs but Rad61/WAPL has not affected in formation of COs. We further observe formation of synaptonemal complexes. pCLB2-PDS5, rad61, and pCLB2-PDS5 rad61 double mutant exhibits very short synaptonemal complexes, suggesting that Pds5 and Rad61/WAPL are involved in axis length determination.

**F006**

**A *Salmonella* Virulence Protein Activated Pho-regulon to Stimulate Phosphates Transport**

Gyunghwa Jeong and Eun-Jin Lee\*

*School of Life Sciences and Biotechnology Division of Life Sciences, Korea University*

*Salmonella enterica* serovar Typhimurium is a foodborne intracellular bacterial pathogen that causes typhoid fever, paratyphoid fever, and food poisoning. Within the phagosome, *Salmonella* senses low  $Mg^{2+}$ , low pH, and antimicrobial peptides as signals to express virulence genes. Phosphate is essential in bacteria for several important biological processes such as the inheritance of genetic materials, energy metabolism, membrane integrity, and intracellular signaling. *Salmonella* has the unique mechanism responding to Pi starvation, the Pho regulon. It is controlled by the PhoB/PhoR two-component regulatory system. The PhoB/PhoR two-component system regulates phosphate transport through the interaction with the Pst high-affinity phosphate transporter complex and the PhoU regulator. PhoR is a histidine kinase that senses environmental phosphate levels and mediates signaling through an interaction with Pst system and phoU.

We used bacterial two hybrid and immunoprecipitation to detect protein interactions within the phosphate regulatory system. Gentamicin protection assay to measure replication efficiency within macrophages. Here we found that a *Salmonella* virulence protein stimulates expression of *pho* regulon and activates phosphate transport by a direct interaction. We determined key residues for the interaction and the substitution studies suggest that the interaction is required for *Salmonella* pathogenesis.

**F007****The Homologous Recombination and Non-homologous End Joining Systems Cooperatively Modulate DNA Damage Response in the Radiation-resistant Fungus, *C. neoformans***

Kwang-Woo Jung

*Korea Atomic Energy Research Institute*

Genome integrity is continuously encountered with DNA lesions caused by intracellular processes and exogenous genotoxic stresses. DNA double-strand breaks (DSB) is the most deleterious damage of DNA lesions because it induces loss of genetic information if not properly repair. In eukaryotes, homologous recombination (HR) and non-homologous end joining (NHEJ) are involved in the DSB repair. Our previous studies reported that Rad53 regulates expression levels of DNA repair genes through the Bdr1 transcription factor in the DNA damage stress. In this study, we found that expression levels of HR and NHEJ were highly induced in a Rad53-Bdr1 pathway-dependent manner in the genotoxic stress. Deletion of *RAD51*, which is one of the components in the HR, resulted in growth defects in the diverse types of DNA damage stress, whereas perturbations of *KU70* and *KU80* did not affect growth in the genotoxic stresses. However, the *rad51Δ ku70Δ* and *rad51Δ ku80Δ* mutants showed significant sensitivity in response to UV exposure and  $\gamma$ -radiation than each single mutant. Furthermore, both deletion of *RAD51* and *KU70/80* renders cells susceptible to oxidative stress induced by *tert*-butyl hydroperoxide and menadione. Collectively, HR and NHEJ cooperatively regulate genotoxic and oxidative stresses in the *C. neoformans*.

[This work was supported by the Nuclear R&D program of Ministry of Science and Information and Communications Technologies (ICT) (Republic of Korea).]

**F008****Exonuclease I and Cohesin Rec8 Promote Establishment of Homolog Interaction in Meiotic Recombination of Budding Yeast**

Soogil Hong and Keun Pil Kim\*

*Department of Life Sciences, Chung-Ang University*

During meiosis, recombination is comprised by two steps, DNA resection and inter-homolog strand exchange. Exonuclease 1 (Exo1) is an evolutionarily conserved eukaryotic DNA nuclease that plays a role in homologous recombination (HR)-mediated double strand break repair. After Spo11-mediated DSB formation, Exo1 executes DNA end resection procedure from 5' end to 3' end. Exposed-single strand DNA tails of DSB ends is used to search for homolog partner template and exchange their DNA strands.  $\alpha$ -kleisin cohesin Rec8 is an essential for meiosis-specific cohesion that is required to generate proper chromosome segregation. Additionally, Rec8 cohesin involves in maintenance of homolog bias, specifically at the double-Holliday junction formation stage. Here, we examined roles of Exo1 and Rec8 through physical analysis of recombination. Exo1 expression exhibited hyperresection of DSBs and is defective in homologous bias, increasing intersister joint molecules. Exo1 expression in *rec8* deletion strain increased joint molecules and CO. We will discuss more detail about that both Exo1 and Rec8 are involved in homologous recombination, especially in early of homologous bias step.

**F009****Pharmacodynamics Study of Ceftiofur and Florfenicol against Fish Isolated *Streptococcus parauberis* as Accessed by Its Genomic and Proteomic Analysis**

Naila Bobby, Muhammad Aleem Abbas, and Seung-Chun Park\*

*Laboratory of Veterinary Pharmacokinetics and Pharmacodynamics, College of Veterinary Medicine, Kyungpook National University*

In the present study, a comparative and subtractive genomic analysis aimed to identify potential therapeutic target proteins using different metabolic pathways of *Streptococcus parauberis*. By a predefined system a total of 102 annotated metabolic pathways were extracted which involves 112 essential non-homologous proteins out of which only 6 proteins have druggability and the genes for these are *mtnN*, *penA*, *pbp2*, *murB*, *murA*, *coaA* and *fni*. Furthermore, based on these findings, the new antimicrobial against *S. parauberis* infections in *Paralichthys olivaceus* was suggested. For inhibitory effects against identified targets based on FDA drug bank database pharmacodynamics of ceftiofur was studied and for comparative study florfenicol has been used. According to invitro susceptibility studies, all tested strains of fish isolated *Streptococcus parauberis* are susceptible to ceftiofur and florfenicol with MIC of 0.0039–1 and 0.5–8 µg/ml and IC<sub>50</sub> of 0.001–0.5 and 0.7–2.7 µg/ml, respectively. Minimum biofilm eradication concentration (MBEC) range for ceftiofur and florfenicol, against the biofilm forming strains, is 2–256 and 4–64 µg/ml, respectively. Thus, ceftiofur has been found as potentially effective antimicrobial against both planktonic and biofilm forming strains of fish pathogenic *S. parauberis* and can be used in aquaculture industry. Novel drugs for the identified targets can be tested and developed for their efficacy against identified target of this pathogen.

**F010****Genomic and Metabolic Features of *Enterococcus faecium* Revealed by Pan-genomic and Metatranscriptomic Analysis**

Shehzad Abid Khan, Byung Hee Chun, and Che Ok Jeon\*

*Department of Life Science, Chung-Ang University*

*Enterococcus faecium* includes both harmful and beneficial strains, and are ubiquitously present in the natural environments. Here in this study, we sequenced the genomes of 15 *E. faecium* strains and compared the genomes with 148 high quality representative genomes of *E. faecium* strains. We analyzed their phylogenetic relationships based on core-genome, which revealed that *E. faecium* strains were separated into two main clades. All food isolates belonged to clade A, while clade B comprised strains mainly isolated from clinical samples. Genome analysis based on antibiotic resistance and virulence factors showed a clear separation between clade A and clade B. *E. faecium* genomes belonging to clade A did not carry any antibiotic resistance genes except for some strains isolated from human samples. Genomic and metabolic features indicated that genes associated with glycolysis/gluconeogenesis, pentose phosphate pathway, pentose and gluconate interconversions and citrate cycle (TCA cycle) were most abundant and equally distributed among all members of clade A. However, in clade B, along with carbohydrates metabolism, the functional genes associated with amino acid metabolisms were most abundant, which suggested that the members of clade B may metabolize amino acids more efficiently than the strains of clade A. Our study, including genomic and metabolic features, will provide insights into the genetic and metabolic diversity of two clades of *E. faecium* strains.

**F011****Crosstalk of Hog1, Mpk1 and Cpk1 MAPK Pathways Governs the Cell Wall Integrity in *Cryptococcus neoformans***

Yu-Byeong Jang, Jin-Tae Choi, and Yong-Sun Bahn\*

*Department of Biotechnology, College of Life Science and Biotechnology, Yonsei University*

*Cryptococcus neoformans* causes meningoencephalitis regardless of immune system disruption and is responsible for approximately 600,000 deaths annually. Mitogen-activated protein kinases (MAPK), the pivotal kinases of eukaryotes, play major roles in metabolism and stress response. There are three major MAPK pathways including Hog1, Mpk1 and Cpk1 pathways in *C. neoformans*. Even though the study of individual MAPK pathways have progressed extensively, the research concerning the crosstalk among three MAPK pathways have yet to be elucidated. We aim to understand the crosstalk among three MAPKs to explain the complex signals regulating the virulence. We constructed the double and triple MAPK deletion mutants (*mpk1Δ hog1Δ*, *cpk1Δ hog1Δ*, *mpk1Δ cpk1Δ*, *mpk1Δ cpk1Δ hog1Δ*) and verified characterizing how MAPK crosstalk regulate the downstream factors. We discovered all three MAPKs have roles in thermosensitivity and cell wall and membrane stress response. Specifically, Mpk1 is known to play key roles in cell wall integrity (CWI), and we discovered that Hog1 and Cpk1 cooperatively contribute to CWI. To identify the regulatory mechanism in CWI with the crosstalk of MAPK, we observed the changes of phosphorylation of MAPKs in all double MAPK mutants under cell wall disturbing conditions. In addition, we discovered that MAPKs distribute to cell membrane susceptibility independently. Therefore, we aim to provide insight into the regulation of cell wall integrity by complex MAPK crosstalk.

**F012****Unveiling the Role of the Casein Kinase 2 in the Pathogenicity of the Human Fungal Meningitis Pathogen *Cryptococcus neoformans***

Yeseul Choi and Yong-Sun Bahn\*

*Department of Biotechnology, College of Life Science and Biotechnology, Yonsei University*

The opportunistic human fungal pathogen *Cryptococcus neoformans* causes fatal meningoencephalitis both in immunocompromised patients and immunocompetent individuals. However, the therapeutic options for treatment of cryptococcosis are currently highly limited. As a potential antifungal drug target, kinases have been considered to be good candidates and play important regulatory roles in cellular mechanisms and virulence of fungal pathogens. In previous studies, we found Cka1, a serine/threonine eukaryotic kinase, is involved in regulation of cell cycle, cellular morphology and pathogenicity of *C. neoformans*. In this study, we aim to figure out the regulatory mechanism of Cka1. We found one catalytic subunit Cka1 and two regulatory subunits which are Ckb1 and Ckb2 as a putative complex subunit. We identified the physical interactions between each subunit of casein kinase 2 using co-IP. We also constructed single and double knockout mutants of regulatory subunits to compare the phenotypes with *cka1Δ*. Interestingly, when *CKA1* was overexpressed in *ckb2Δ ckb1Δ*, it restored the growth defect of *ckb2Δ ckb1Δ*. We also constructed *cka1Δ ckb1Δ ckb2Δ* triple deletion mutant and it showed severe growth defect like *cka1Δ* mutant. As a result, Cka1 plays major roles and Ckb1/Ckb2 have minor roles in casein kinase 2 complex. This study will provide a comprehensive Cka1 cellular mechanism to develop an antifungal drug.

**F013****Systematic Analysis of Host-derived Cues for the Regulation of Pathogenicity-related Transcription Factors in *Cryptococcus neoformans***

Seong-Ryong Yu, Minjae Lee, Kyung-Tae Lee, and Yong-Sun Bahn\*

*Department of Biotechnology, College of Life Science and Biotechnology, Yonsei University*

*Cryptococcus neoformans* is a causative agent of fungal meningoencephalitis, which results in more than 180,000 deaths annually. Although a number of signaling pathways involved in the pathogenicity of *C. neoformans* have been characterized in past years, it remains elusive how complex signaling pathways are coordinated and regulated during the whole infection process. Previously we performed NanoString-based *in vivo* transcription profiling of 183 kinases, 178 transcription factors, and 139 phosphatases during the whole infection process of *C. neoformans*. Here we focused on 12 transcription factors, including *PDR802*, *BZP4*, *HOB5*, *ZNF2*, *FZC39*, *FZC30*, *SRE1*, *HLH1*, *STB4*, *MLN1*, *MET32*, and *GAT201* of which *in vivo* expression were highly induced during host infection but did not exhibit evident *in vitro* phenotypes. To elucidate their *in vivo* functions, the expression level of the 12 genes were measured in host mimic condition (HMC): RPMI cell culture media supplemented with 10% fetal bovine serum at 37°C under 5% CO<sub>2</sub>. We found that all of them were highly induced by HMC. Next we dissected the HMC signals that trigger the induction of the 12 transcription factors. We found that *PDR802* was highly induced by body temperature and carbon starvation. To further investigate the gene interaction, we constructed the double knockout mutants of body temperature, carbon starvation. In conclusion, we provided insight into complex signaling pathways modulating the pathogenicity of *C. neoformans*.

**F014****Identification of the Role of cAMP Pathway in the Emerging Fungal Pathogen *Candida auris***

Ji-Seok Kim, Tae-Hyun Kim, and Yong-Sun Bahn\*

*Department of Biotechnology, College of Life Science and Biotechnology, Yonsei University*

*Candida auris* is a globally emerging multidrug-resistant fungal pathogen causing invasive human infection and associated with high fatality diseases in immunocompromised patients. Accordingly, the importance of research on *C. auris* is increasing. In this study, we characterized cyclic adenosine monophosphate (cAMP) pathway of *C. auris* which has been considered to be one of the most important signal transduction pathway and related to the growth and virulence of pathogenic fungal species. Among the various genes associated with the cAMP pathway, we constructed knockout strains for the adenylyl cyclase *CYR1*, PKA gene catalytic subunit *TPK1*, *TPK2*, and regulatory subunit *BCY1* which are expected to play an important role in the signaling process and related to pathogenicity of *C. auris*. We also constructed double knockout strains of *TPK1* and *TPK2* to figure out how the two genes interact. Then we conducted a phenotypic analysis of the mutants, and we found that four genes were involved in multiple stress responses. Also we confirmed the difference in biofilm formation between WT and mutants. These findings revealed that PKA genes are involved in the formation of *C. auris* biofilm. In addition, the four mutant strains showed different expression level of drug resistance related genes. Consequently, these results will indicate that targeting cAMP pathway genes in *Candida auris* could serve as an effective alternative to antifungal therapy against emerging fungal pathogen *C. auris*.

**F015****The Unraveling of Complex Signaling Networks Involved in the Developmental Process of *Cryptococcus neoformans***

Jin-Young Kim, Yeonseon Lee, and Yong-Sun Bahn\*

*Department of Biotechnology, College of Life Science and Biotechnology, Yonsei University*

The fungal pathogen *Cryptococcus neoformans* causes cryptococcosis by the inhalation of infectious spores generated by unisexual or bisexual reproduction. To understand complex signaling networks modulating the developmental process, a complete understanding of genome-scale transcription factors (TFs) and kinases is needed. Previously we reported that 37 TFs and 42 kinase mutants constructed in *C. neoformans* MAT $\alpha$  H99 strain background exhibited altered mating efficiency. To further elucidate the mating regulatory mechanism, we constructed knockout mutants of the mating-regulating TFs and kinases in YL99 strain—MAT $\alpha$  isogenic strain of H99 strain—to monitor unilateral and bilateral mating, and to perform an analysis of their function in the developmental process. We constructed 22 gene-deletion strains representing eleven TFs and are currently constructing gene-deletion strains for the remaining mating-regulating TFs and kinases. For confirmed mutant strains, we are examining mating phenotypes during bilateral mating. Furthermore, we are examining transcript profiles of mating-regulating TFs and kinases at different developmental stages of sexual reproduction. Ultimately, this study will focus on mapping and discovering the functions of the mating-regulating TFs and kinases, and elucidating complex signaling networks in the developmental process of *C. neoformans*.

**F016****Deciphering the Role of Pseudouridylation in Human Fungal Pathogen *Cryptococcus neoformans***Seung-Heon Lee<sup>1</sup>, Jin-Young Kim<sup>1</sup>, Seong-Ryong Yu<sup>1</sup>, Anna F. Averette<sup>2</sup>, Joseph Heitman<sup>2</sup>, and Yong-Sun Bahn<sup>1\*</sup><sup>1</sup>*Department of Biotechnology, College of Life Science and Biotechnology, Yonsei University,* <sup>2</sup>*Department of Molecular Genetics and Microbiology, Medicine, and Pharmacology and Cancer Biology, Duke University Medical Center, Durham, North Carolina, USA*

*Cryptococcus neoformans* is a basidiomycetous fungal pathogen that causes systemic cryptococcosis and meningoencephalitis in immunocompromised individuals. Due to its clinical importance, revealing the factors that can affect its life cycle is critical for development of antifungal and anticryptococcal drugs. Among the various factors, pseudouridylation of RNA is the most abundant type of post-transcriptional modification. Pseudouridylases isomerize uridine into pseudouridine, which subsequently affects the stability of RNA structure. In *Saccharomyces cerevisiae*, eight proteins exist as stand-alone pseudouridylases, and each protein has specific catalytic sites and roles. To unravel the biological functions of the enzymes in *C. neoformans*, we identified six putative pseudouridylases in *C. neoformans* by using BLAST search in the FungiDB database with protein sequences of the known pseudouridylase genes. To characterize the function of pseudouridylases, we constructed more than two independent strains for 5 putative pseudouridylase genes and examined their phenotypic traits under various conditions. *CBF1*, which is essential gene in *S. cerevisiae*, is also suspected to be essential in *C. neoformans*. Among the proteins, *DEG1* and *PUS7* seemed to have major roles in stress responses and virulence of *C. neoformans*. By using pseudouridylation RNA-sequencing, we will identify pseudouridylated mRNA transcripts and characterize their role in pathogenicity of *C. neoformans*.

**F017**

**Complete Genome Sequence of Halo-tolerant, Alkaline Protease Producing Strain of *Micrococcus luteus* SB1254**

Bo Gyoung Choi<sup>1</sup>, Yong-Jik Lee<sup>2</sup>, Dariimaa Ganbat<sup>1</sup>, Gi Seob Hong<sup>1</sup>, Min Hee Jung<sup>1</sup>, Joon Young Heo<sup>1</sup>, Jae Hak Shon<sup>1</sup>, Han-Seung Lee<sup>1</sup>, and Sang-Jae Lee<sup>1\*</sup>

<sup>1</sup>Major in Food Biotechnology and Research Center for Extremophiles & Marine Microbiology, Silla University,

<sup>2</sup>Department of Bio-cosmetics, Seowon University

A *Micrococcus luteus* SB1254 strain was isolated from mussel at Geoje Island in Korea for finding alkaline protease producing extremophiles. This strain can grow a diverse concentration of NaCl (0 to 10%) and pH (7 to 10) together with the production of protease. The complete genome comprises 2,553,002 bp with a G + C content of 72.9%, 2,126 coding genes and 54 RNA genes. Strain SB1254 was identified as *M. luteus* using TrueBac™ service offered from ChunLab and the IMG-ER platform analysis revealed it harbored 32 protease related genes, meaning that it can be used as a source of the protease production for the eco-friendly industrial application.

**F018**

**Systematic Analysis on Transcription Unit Architecture of *Streptomyces avermitilis***

Yongjae Lee<sup>1</sup>, Yujin Jeong<sup>1</sup>, Namil Lee<sup>1</sup>, Soonkyu Hwang<sup>1</sup>, Woori Kim<sup>1</sup>, Suhyung Cho<sup>1</sup>, Bernhard Palsson<sup>2,3,4</sup>, and Byung-Kwan Cho<sup>1,5\*</sup>

<sup>1</sup>Department of Biological Sciences and KAIST Institute for the BioCentury, Korea Advanced Institute of Science and Technology, <sup>2</sup>Department of Bioengineering, University of California San Diego, La Jolla, CA 92093, USA,

<sup>3</sup>Department of Pediatrics, University of California San Diego, La Jolla, CA 92093, USA, <sup>4</sup>Novo Nordisk Foundation Center for Biosustainability, 2800 Kongens Lyngby, Denmark, <sup>5</sup>Intelligent Synthetic Biology Center

*Streptomyces avermitilis* holds industrial importance as the producer of avermectin, a widely used anthelmintic agent. Despite its industrial importance, *S. avermitilis*' genome organization and regulation of gene expression remain poorly understood. To understand the genome-scale gene expression landscape, four types of next-generation sequencing techniques, including dRNA-Seq, Term-Seq, RNA-Seq, and Ribo-Seq, were exploited, determining a total of 1601 transcription units (TUs) in *S. avermitilis*. Integrative analysis of the TU information with gene expression datasets revealed diverse regulatory elements for transcriptional and translational control of individual TUs. The conserved promoter motifs were identified from 2361 transcription start sites as 5'-TANNNT and 5'-TGAC for the -10 and -35 elements, respectively. Analysis of the 3'-end of RNA transcript revealed that stem structure formation is a major determinant for transcription termination. The TU architecture suggests the presence of novel small RNAs and *cis*-regulatory elements in the genome. Our findings will elevate *S. avermitilis*' potential as a production host for a diverse set of secondary metabolites.

[This work was supported by the Novo Nordisk Foundation (NNF10CC1016517). This work was also supported by the Intelligent Synthetic Biology Center of Global Frontier Project (2011-0031957) and the Bio & Medical Technology Development Program (2018M3A9F3079664) through the NRF funded by the MSIT.]

**F019****Zinc Regulates Gliotoxin Biosynthesis in *Aspergillus fumigatus***

Hyewon Seo, Suzie Kang, Yong-Sung Park, and Cheol-Won Yun\*

*School of Life Sciences and Biotechnology, Korea University*

*Aspergillus fumigatus* requires micronutrient zinc because zinc performs a wide range of biological functions, including fungal pathogenicity. Among many factors that affect fungal toxicity, gliotoxin is a representative secondary metabolite that attacks the host's immune cells. We found that the gliotoxin production was inversely proportional to the zinc concentration. To unveil the detailed mechanism, we performed microarrays and northern blots. The results of these experiments showed that the expression between the zinc-dependent transcriptional activator ZafA and the gliotoxin Zn2-Cys6 transcription factor GliZ showed the same pattern for zinc concentration. Interestingly, in the promoter region of *GliZ*, there were two conserved ZafA-binding motifs, 5'-CAAGGT-3'. When these motifs were lost, the gliotoxin production and their pathogenicity in murine models were attenuated. Taken together, the zinc greatly contributes to the fungal toxicity by regulating gliotoxin production of *A. fumigatus*. [This work was supported by the Cooperative Research Program for Agriculture Science and Technology Development (Project No: PJ01368101), Rural Development Administration, Republic of Korea.]

**F020****The First Report of Genetic and Structural Diversities in the *SPRN* Gene in the Horse, an Animal Resistant to Prion Disease**Sae-Young Won<sup>1,2</sup> and Byung-Hoon Jeong<sup>1,2\*</sup><sup>1</sup>*Department of Bioactive Material Sciences, Jeonbuk National University,* <sup>2</sup>*Korea Zoonosis Research Institute, Jeonbuk National University*

Prion diseases are fatal neurodegenerative diseases and are characterized by the accumulation of abnormal prion protein (PrP<sup>Sc</sup>) in the brain. During the outbreak of the bovine spongiform encephalopathy (BSE) epidemic in the United Kingdom, prion diseases in several species were reported; however, horse prion disease has not been reported thus far. In previous studies, shadow of prion protein (Sho) has contributed to an acceleration of conversion from normal prion protein (PrP<sup>C</sup>) to PrP<sup>Sc</sup>, and shadow of prion protein gene (*SPRN*) polymorphisms have been significantly associated with the susceptibility of prion diseases. We investigated the genotype, allele and haplotype frequencies of the *SPRN* gene using direct sequencing. In addition, we analyzed linkage disequilibrium (LD) and haplotypes among polymorphisms. We also investigated LD between *PRNP* and *SPRN* SNPs. We compared the amino acid sequences of Sho protein between the horse and several prion disease-susceptible species using ClustalW2. To perform Sho protein modeling, we utilized SWISS-MODEL and Swiss-PdbViewer programs. We found a total of 4 polymorphisms in the equine *SPRN* gene; however, we did not observe an in/del polymorphism, which is correlated with the susceptibility of prion disease in prion disease-susceptible animals. The *SPRN* SNPs showed weak LD value with *PRNP* SNP.

**F021****Absence of Single Nucleotide Polymorphisms (SNPs) in the Open Reading Frame (ORF) of the Prion Protein Gene (*PRNP*) in a Large Sampling of Various Chicken Breeds**Sae-Young Won<sup>1,2</sup> and Byung-Hoon Jeong<sup>1,2\*</sup><sup>1</sup>Department of Bioactive Material Sciences, Jeonbuk National University, <sup>2</sup>Korea Zoonosis Research Institute, Jeonbuk National University

Prion diseases are zoonotic diseases with a broad infection spectrum among mammalian hosts and are caused by the misfolded prion protein (PrP<sup>Sc</sup>) derived from the normal prion protein (PrP<sup>C</sup>), which encodes the prion protein gene (*PRNP*). Currently, although several prion disease-resistant animals have been reported, a high dose of prion agent inoculation triggers prion disease infection in these disease-resistant animals. However, in chickens, natural prion disease-infected cases have not been reported, and experimental challenges with prion agents have failed to cause infection. Unlike other prion disease-resistant animals, chickens have shown perfect resistance to prion disease thus far. Thus, investigation of the chicken *PRNP* gene could improve for understanding the mechanism of perfect prion-disease resistance. Here, we investigated the genetic characteristics of the open reading frame (ORF) of the chicken *PRNP* gene in a large sampling of various chicken breeds. We found only tandem repeat deletion polymorphisms of the chicken *PRNP* ORF in the 4 chicken breeds including 106 Dekalb White, 100 Ross, 98 Ogotgye and 100 Korean native chickens. In addition, the distribution of chicken insertion/deletion polymorphisms was significantly different among the 4 chicken breeds. Finally, we found significant differences in the number of *PRNP* SNPs between prion disease-susceptible species and prion disease-resistant species.

**F022****Bovine Spongiform Encephalopathy (BSE)-associated Polymorphisms of Shadow of Prion Protein Gene (*SPRN*) in Korean Native Cattle (Hanwoo) and Holstein Cattle**Yong-Chan Kim<sup>1,2</sup> and Byung-Hoon Jeong<sup>1,2\*</sup><sup>1</sup>Department of Bioactive Material Sciences, Jeonbuk National University, <sup>2</sup>Korea Zoonosis Research Institute, Jeonbuk National University

Bovine spongiform encephalopathy (BSE) is a fatal infectious neurodegenerative disease caused by the accumulation of pathogenic prion protein (PrP<sup>Sc</sup>) in the central nervous system (CNS), particularly in the brain. In a recent study, the shadow of prion protein (Sho), encoded by the shadow of prion protein (*SPRN*) gene, accelerates the progression of prion diseases, and a 12-bp insertion/deletion polymorphism in the coding region of the *SPRN* gene is associated with susceptibility to atypical BSE-affected Polish cattle. To date, the genetic study of the *SPRN* gene in Korean cattle has not been performed. In this study, we investigated the genotype and allele frequencies of *SPRN* polymorphisms in 193 Hanwoo and 168 Holstein cattle and analyzed the linkage disequilibrium (LD) and haplotypes of *SPRN* polymorphisms. In addition, we compared the distribution of the 12-bp insertion/deletion polymorphism between atypical BSE-diagnosed Polish cattle and Korean cattle to evaluate the susceptibility of atypical BSE. Furthermore, we estimated a deleterious effect of polymorphisms on the Sho protein using PROVEAN. We found a total of 7 polymorphisms, including 1 novel single nucleotide polymorphism (SNP), c.231G>A. We also found significantly different distributions of genotype, allele and haplotype frequencies of 7 polymorphisms between Hanwoo and Korean Holstein cattle.

**F023****Strong Association of Regulatory Single Nucleotide Polymorphisms (SNPs) of the *IFITM3* Gene with Influenza H1N1 2009 Pandemic Virus Infection**Yong-Chan Kim<sup>1,2</sup> and Byung-Hoon Jeong<sup>1,2\*</sup><sup>1</sup>Department of Bioactive Material Sciences, Jeonbuk National University, <sup>2</sup>Korea Zoonosis Research Institute, Jeonbuk National University

*IFITM3* gene is an important host immunological effector against viral invasion for various viruses, including influenza A viruses. Previous studies have reported that single nucleotide polymorphisms (SNPs) of the *IFITM3* gene, such as the rs12252 SNP in the splicing receptor and the rs34481144 SNP in the promoter region, play a pivotal role in the pathogenesis of pandemic influenza A 2009 in several ethnic groups. In the present study, the purpose was to conduct fine mapping of the *IFITM3* gene and compare the genetic differences of the *IFITM3* gene between those of healthy Koreans and H1N1 2009 pandemic influenza-infected patients. Significant differences between healthy controls and H1N1 pandemic influenza 2009 affected patients were observed in the genotype frequencies of 5 regulatory SNPs, c.-204G>T ( $P = 0.039$ ), c.-188T>C ( $P < 0.0001$ ), c.-181T>C ( $P = 0.0003$ ), c.-178A>C ( $P = 0.0002$ ) and c.-175T>C ( $P = 0.0002$ ), and 1 intronic SNP, c.249+450T>C ( $P = 0.007$ ), in the Korean population. However, the *IFITM3* c.-23G>A (rs34481144) polymorphism was not found in the Korean population. In addition, we found significantly different haplotype distributions of the *IFITM3* gene between those of H1N1 influenza 2009 pandemic patients and healthy controls.

**F024*****In Silico* Evaluation of Acetylation Mimics in the 27 Lysine Residues of Human tau Protein**Yong-Chan Kim<sup>1,2</sup> and Byung-Hoon Jeong<sup>1,2\*</sup><sup>1</sup>Department of Bioactive Material Sciences, Jeonbuk National University, <sup>2</sup>Korea Zoonosis Research Institute, Jeonbuk National University

Various neurodegenerative diseases, including Alzheimer's disease (AD), are related to abnormal hyperphosphorylated microtubule-associated protein tau accumulation in brain lesions. Recent studies have focused on toxicity caused by another post-translational modification (PTM), acetylation of the lysine (K) residues of tau protein. Because there are numerous acetylation sites, several studies have introduced mimics of tau acetylation using amino acid substitutions from lysine to glutamine (Q). However, human tau protein contains over 20 acetylation sites; thus, investigation of the effects of acetylated tau is difficult. Here, we *in silico* evaluated acetylation effects using SIFT, PolyPhen-2 and PROVEAN which can estimate the effects of amino acid substitutions based on the sequence homology or protein structure in tau isoforms. In addition, we also investigated 27 acetylation effects on amyloid formation of tau protein using Waltz. 15 acetylation mimics were estimated to be most detrimental, which indicates that there may be novel pathogenic acetylation sites in human tau protein.

**F025**

**Probiotics Roles (CACC517) on the Modulation of Gut Microbiota for the Healthy and IBD Dogs**

Donghyun Shin and Seungwoo Son\*

*The Animal Molecular Genetics & Breeding Center, Jeonbuk National University*

The importance of probiotics in Canine gut health is widely acknowledged as crucial. Especially, Canine inflammatory bowel disease (IBD) is believed to be caused by a complex interaction of genetic, immunologic, and microbial factors. However, gaps still remain in the exact roles played by probiotics in modulation of gut microbiota and health. This study determined the probiotic roles of *Bifidobacterium longum* (CACC517) isolated from health canine feces in gut microbiota modulation using 16S RNA sequencing. We collected feces samples from healthy dogs and dogs with IBD as assessed by a clinical disease activity index. Dogs with Intestinal inflammation was associated with significant differences in the composition of the intestinal microbiota when compared to healthy dogs. Metagenome analysis showed that microbial diversity and richness in dogs fed probiotics. Proportions of Bacteroidaceae and Fusobacteriaceae were significantly less in healthy dogs after CACC517 treatment. In IBD dogs, Enterococcaceae was significantly less and Lachnospiraceae was more abundant after probiotics treatment. Our study suggest that CACC517 increased the diversity and richness in the microbial community of healthy dogs and IBD dogs, which indicated alterations in microbial groups.

[This research was supported by a grant from the Strategic Initiative for Microbiomes in Agriculture and Food, Ministry of Agriculture, Food and Rural Affairs, Republic of Korea (Project No. 918002-04)]

**F026**

**A Putative Signal Peptidase AfSec11 is Involved in Caspofungin Resistance in *Aspergillus fumigatus***

Eunji Jang, Yong-Sung Park, Suzie Kang, Hyewon Seo, and Cheol-Won Yun\*

*School of Life Sciences and Biotechnology, Korea University*

AfSec11 is a putative signal peptidase of *Aspergillus fumigatus*, and it has high homology with yeast Sec11 on the amino acid level. Deletion of promoter region of AfSec11 resulted in decrease of AfSec11 transcription and slower growth than wild type cells. And showed higher sensitivity to the caspofungin which inhibits the synthesis of beta-glucan of cell wall than wild type cells. However, no difference was found from wild type cells in the sensitivity to the amphotericin B and fluconazole which inhibit sterol function of cell wall. We also found that deletion of promoter region of AfSec11 resulted in failure of post-translation modification of FksA which is the target of caspofungin. Finally, we found that the deletion mutant of promoter region of AfSec11 showed decrease of virulence in the murine model. Taken these results together, we suggest that AfSec11 regulates the post-translational modification of Fks1 and is involved in caspofungin resistance.

[Supported by the Cooperative Research Program for Agriculture Science and Technology Development (Project No: PJ01368101), Rural Development Administration, Republic of Korea.]

**F027****Genome-wide Analysis Reveals the Role of VadA in *Aspergillus nidulans* Conidia**

Ye-Eun Son, Seo-Yeong Jang, and Hee-Soo Park\*

*School of Food Science and Biotechnology, Institute of Agricultural Science and Technology, Kyungpook National University*

Conidium is a asexual spore that mainly propagates in the *Aspergillus* species. Previous study demonstrated that VadA is a conidial specific gene that is required for conidial viability, spore wall integrity, sterigmatocystin production, and fungal development in the model fungus *Aspergillus nidulans*. In this study, *vadA* transcriptomic analysis revealed that VadA affects mRNA expression of a variety of genes in *A. nidulans* conidia. Among the affected genes, those in the category of trehalose biosynthesis, cell-wall integrity, stress response, and secondary metabolism, were mainly affected in conidia. Interestingly, deletion of *vadA* resulted in alteration of genes associated with UV stress response in conidia, and the *vadA* deletion mutant conidia were more sensitive against UV stress, suggesting that VadA is required for proper UV response in *A. nidulans*. In addition, most genes in the certain secondary metabolism gene clusters for sterigmatocystin, asperfuranone, monodictyphenone, and asperthecin were upregulated in the *vadA* deletion mutant conidia. Overall, these results propose that VadA is a conidial specific regulator, controlling conidial wall integrity, stress response, and secondary metabolism in *A. nidulans* conidia.

**F028****The Homeobox Domain Protein HbxB Plays a Key Role in Governing Development and Secondary Metabolism in *Aspergillus nidulans***

Sung-Hun Son, He-Jin Cho, and Hee-Soo Park\*

*School of Food Science and Biotechnology, Institute of Agricultural Science and Technology, Kyungpook National University*

Homeobox domain proteins are conserved transcription factors in eukaryotic organisms and play an important role in the development and secondary metabolism in fungi and other eukaryotes. In the present study, we identified eight homeobox domain proteins in the model filamentous fungus *Aspergillus nidulans* by genomic analysis and conducted phenotype analysis for each deletion mutants to examine the role of each gene. The result of development phenotypic analyses found that HbxB is affected the development and secondary metabolism. In development, the *hbxB* deletion mutant strains exhibit reduced conidia formation but increased sexual structure cleistothecia production and germination rate. Overexpression of *hbxB* led to the induced formation of conidiophore and decreased production of cleistothecia structure. In asexual spores, the absence of *hbxB* reduced trehalose biogenesis and also decreased resistance of conidiophore to thermal, oxidation, and UV radiation stresses. In addition, *hbxB* deletion changes mRNA levels of genes associated with the trehalose biosynthesis. Moreover, the *hbxB* deletion mutant strains shown a reduction of sterigmatocystin production. These results demonstrated that HbxB is a regulator of development and play crucial roles in tolerance of asexual spore, secondary metabolism in *A. nidulans*.

**F029**

**The Complete Genome Sequence of Fowlpox Virus Isolated in South Korea**

Si-Hyeon Kim, Il Jang, Yong-Kuk Kwon, and Hye-Ryoung Kim\*

*Avian Disease Division, Animal and Plant Quarantine Agency*

Fowlpox is important disease in poultry industry, because it can cause drop in egg production, slow growth and mortality. Fowlpox have occurred in Korean poultry farm. But there is no complete genome of Korean isolate. Illumina Miseq instrument was used for whole genome sequencing for the strain. Using *de novo* assembly, 278,446 bp contig is obtain. Although its average coverage was high, 5' end and 3' end parts were not assembled, because the Inverted terminal repeat regions were identical. These parts were restored by mapping to a complete genome sequence of an American fowlpox virus strain (AF198100). The obtained genome size of Korean strain was 297,482 bp containing 256 Open Reading Frames. The percent identity with the reference strain was 98.04 %. This is the first study of complete genome sequence of Korean fowlpox virus. If more Korean strains complete sequence were obtained from this established method, genetic characterization of the Korean fowlpox viruses would be determined.

**F030**

**Phosphorylation of Replication Protein A Modulates Meiotic Recombination in Budding Yeast**

Hyung Seok Choi and Keun Pil Kim

*Department of Life Sciences, Chung-Ang University*

RPA (Replication Protein A) is essential for DNA replication and promotes DNA repair and recombination by inhibiting secondary structure of DNA. RPA complex, composed with Rfa1, Rfa2, and Rfa3, exhibits strong binding affinity for single strand DNA (ssDNA) and support Rad51 (RecA homolog)-mediated repair or recombination processes. During meiosis, RPA involves in two main processes, meiotic replication and recombination by protecting ssDNA tail of DSB ends generated by the resection. Further, it has been reported that ATR<sup>Mec1</sup> phosphorylates RPA, so that this modification modulates RPA's dual functions: preventing secondary structure of ssDNA binding and recruiting diverse accessory factors. Here, we demonstrate that the phosphorylation of yeast Rfa2 involves in regulating meiotic recombinant. To understand the functional importance of RPA phosphorylation in yeast meiosis, we tested recombinational activity of Rfa2 serine phosphomimetic mutation S27A, S122A and S122D: Rfa2-S27 is Ime2-dependent phosphorylation region and Rfa2-S122 is Mec1-dependent phosphorylation region. We observed that these mutations, rfa2-S27A, rfa2-S122A, rfa2-S122D, exhibit normal double-strand break formation, but defects in joint molecule formation and recombination outcomes. I will discuss more detail about recombinational role of Rfa2 in the presentation.

**F031****Complete Genome Sequence of *Lactobacillus paraplantarum* CK401**Hyun-Myung Oh<sup>1\*</sup> and Dongil Jang<sup>2</sup><sup>1</sup>*Institute of Liberal Arts Education, Pukyong National University,* <sup>2</sup>*Cotde, Inc.*

Here we report complete genome sequence of Gram-positive, rod-shaped and chemoheterophilic *Lactobacillus paraplantarum* strain CK401 with anti-avian flu activity. Circular chromosome of 3,164,408 bp with 43.99% G+C content and five circular plasmids: pCK401A (15,102 bp with 37.33% G+C content), pCK401B (15,825 bp with 40.58% G+C content), pCK401C (12,380 bp with 36.03% G+C content), pCK401D (40,656 bp with 42.00% G+C content) and pCK401E (41,380 bp with 40.12% G+C content). Total genes of 3,118 in number were predicted: 3,029 are protein coding genes and 2,061 of protein coding genes could be functionally assigned. The chromosome harbors 5 operons of 16 rRNAs and 68 tRNA genes. No CRISPR locus was found in this genome. Canonical glycolytic and pentose phosphate pathways plus lactate/alcohol/butanediol dehydrogenases and acetate kinase suggested that the strain may have a heterofermentative metabolism. Diverse glycosyl hydrolases (GH) were encoded in 47 genes in 17 GH families: alpha-amylases, alpha-/beta-galactosidases, beta-glucosidases, a beta-glucuronidase, a galactanase, and an invertase. Some bacteriocin genes and confirmed hyaluronic acid gene showed that the antibiotic-resistance-free strain CK401 will be a promising probiotic candidates for animals.

**F032****Complete Genome Sequence of *Altibacter* sp. Strain ALE3EI Isolated from Jeju Island**Hyun-Myung Oh<sup>1\*</sup> and Dongil Jang<sup>2</sup><sup>1</sup>*Institute of Liberal Arts Education, Pukyong National University,* <sup>2</sup>*Cotde, Inc.*

Here we report complete genome sequence of Gram-negative, rod-shaped and chemoheterophilic aerobic bacterial strain ALE3EI. The 16S rRNA gene sequence analysis revealed that strain ALE3EI showed high similarity with the *Altibacter lentus* JLT2010<sup>T</sup> (96.7%). Genomic DNA was extracted using i-genomic BYF mini kit (iNtRON Biotechnology, Republic of Korea) following manufacturer's protocols. Genome sequencing of the strain ALE3EI was performed using PacBio RS II single-molecule real-time (SMRT) sequencing technology (Pacific Biosciences, Menlo Park, CA). A standard PacBio library with an average of 20 kb inserts were prepared and were sequenced, yielding > 352.84 x average genome coverage. De novo assembly of the 112,619 reads with 14,158 nucleotides on the average (1,594,478,562 bp in total) was conducted using the hierarchical genome-assembly process (HGAP) pipeline of the SMRT Analysis v2.3.0.

Circular chromosome of 2,987,299 bp with 40.38% G+C content. Total genes of 2,729 in number were predicted: 2,654 are protein coding genes and 2 rRNA operons plus 37 tRNA genes could be assigned. Canonical glycolytic and pentose phosphate pathways were present but there were no genes to produce NRPS/PKS and no prophage regions identified. The genome harbors the genes for synthesis and degradation of cyanophycin, which is a nitrogen-storage polymer composed of a polyaspartate backbone with arginine side chains and is of biomedical interest as a biodegradable substitute for synthetic polyacrylates.

**F033**

**Changes in Fecal Microbiota Composition by Age in Korean Calves**

**Yeonsu Oh<sup>1</sup>, Dohyeon Yu<sup>2</sup>, Hak-Jong Choi<sup>3</sup>, and Jinho Park<sup>4\*</sup>**

*<sup>1</sup>College of Veterinary Medicine and Institute of Veterinary Science, Kangwon National University, <sup>2</sup>College of Veterinary Medicine, Gyeongsang National University, <sup>3</sup>Microbiology and Functionality Research Group, World Institute of Kimchi, <sup>4</sup>College of Veterinary Science, Chonbuk National University*

Within 10 days of age (Day 1-10, n=8), 11-20 days (Day 11-20, n=10), 21-30 days (Day 20-30, n=11) feces of normal calves were collected and analyzed. After extracting the collected fecal DNA, the V3-V4 region of the 16S rRNA gene was amplified and analyzed through MiSeq equipment of Illumina, and a total of 1,202,745 reads were obtained (average of 41,474 reads per head). As a result of confirming the rarefaction curve showing the increase rate of the number of Operational Taxonomic Units (OTUs) according to the number of acquired sequences, a sufficient number of OTUs required for analysis was secured. Through the results of alpha- and beta-diversity, taxonomic composition analysis, and LEfSe calculation, it was confirmed that the intestinal microbial species, diversity, and 6 of characteristic microorganisms increased in Korean calves by age. This is considered to be related to the settlement of microorganisms in the intestine as the feed and breeding environment changes according to the calf growth stage.

[This research was supported by Technology Development Program (Project No. 1116043-1) for Bio-industry, Ministry for Agriculture, Food and Rural Affairs, Republic of Korea.]

**G001****Microbial Production Regulates the Activity of Insect Olfactory Receptor**

Junho Lee

*Chonnam National University*

The olfactory nervous system recognizes and distinguishes many different chemicals in the general living environment. Insects have evolved a group of odorant-gated ion channels composed of highly-developed olfactory receptors capable of distinguishing and distinguishing between various chemicals with symbolic or evasive specificities. Recently, aphid genomes related to olfaction, including olfactory receptors and proteins, have been identified and olfactory receptors have been reported that are differentially differentiated from *Drosophila*. The genome of the olfactory receptor has a very conservative sequence and a systematic signaling system. A representative receptor, odorant-gated ion channels comprised of a highly conserved co-receptor (Orco) has a homotetramer channel structure with four subunits arranged symmetrically around the central hole. It has a very similar structure to the 7-transmembrane receptor present in the human body and has a very similar structural form and gating mechanism to receptors of neurotransmitters. Therefore, it is possible to identify attractant or repellent substance using the olfactory receptor activity regulating system of insects. Through this study, MZ01 shows the attracting phenomenon by activating insect receptor OR85b, the results of the scientific analysis of the performance of the extracts are presented.

**G002****Utilization of Jerusalem Artichoke and Lipid Extracted Algae Hydrolysate for Zero Waste DHA Production Using *Aurantiochytrium* sp. KRS 101**Sung-Woon Heo<sup>1</sup>, Hye-won Yang<sup>2</sup>, Young Taek Oh<sup>2</sup>, and Bongsoo Lee<sup>3\*</sup><sup>1</sup>*Department of Chemical and Biomolecular Engineering, Korea Advanced Institute of Science & Technology,*<sup>2</sup>*Nakdonggang National Institute of Biological Resources,* <sup>3</sup>*Department of Microbiology and Resources, College of Science and Technology, Mokwon University*

The current study investigated the potential of Jerusalem artichoke and lipid extracted algae (LEA) hydrolysates as inexpensive carbon and other nutrient sources for DHA production using *Aurantiochytrium* sp. KRS101. Response surface methodology (RSM) with central composite design (CCD) was used to determine the maximum point of DHA productivity under 51.9 g/L of fructose and 4.9 g/L of yeast extract. Hydrolysis of 76.6 g/L Jerusalem artichoke powder with 0.25 N of sulfuric acid at 70°C were able to produce desired concentration of fructose for use as the medium. Cultivation of the microalgae using the Jerusalem artichoke hydrolysate (JAH) and JAH+LEA (JLH-1) resulted in 27.8% and 31.3% improvements in biomass productivity over basal medium control, respectively. In addition, it was found that simultaneous one-step hydrolysis of Jerusalem artichoke and LEA (JLH-2) resulted in 7.2% higher biomass and 47% higher DHA yields than hydrolyzing the two components separately (JLH-1). These results suggest that a strategy using hydrolysate of Jerusalem artichoke combined with recycled LEA is not only able to replace the expensive nutrient sources, but also produce efficient and eco-friendly DHA from *Aurantiochytrium* sp.

[This work was supported by grant from the Advanced Biomass R&D Center (ABC) of the Global Frontier Project (ABC-2010-0029728) and the Nakdonggang National Institute of Biological Resources (NNIBR; NNIBR202002101)]

**G003****Twelve Quick Steps for Genome Assembly and Annotation in the Classroom**Hyungtaek Jung<sup>1</sup>, Min-seung Jeon<sup>2</sup>, and Seong-il Eyun<sup>2,3\*</sup><sup>1</sup>Centre for Agriculture and Biocommodities, Queensland University of Technology, Brisbane, Australia, <sup>2</sup>Department of Life Science, Chung-Ang University, <sup>3</sup>EyunZen Bio Inc.

Although third-generation long-read DNA sequencing technologies are increasingly used, generating high-quality genome assemblies and annotations for many species still presents significant challenges due to their large genome sizes, complexity, and high chromosome numbers. Indeed, selecting the most appropriate sequencing and software platforms and annotation pipelines for a new genome project can be daunting because tools often only work in limited contexts. Herein, we state 12 steps to help researchers get started in genome projects by presenting guidelines that are broadly applicable, sustainable over time, and cover all aspects of genome assembly and annotation projects. We review some commonly used approaches, including practical methods to extract high-quality DNA, choices for the best sequencing platforms and library preparations and the range of potential bioinformatics pipelines. Also, we include information on how to build a wide community for a genome project, the importance of data management, and how to make the data and results Findable, Accessible, Interoperable, and Reusable (FAIR) by submitting them to a public repository and sharing them with the research community.

[Supported by National Research Foundation of Korea Grants (2018R1C1B3001650 and 2018R1A5A1025077) and the Korean Ministry of Agriculture, Food, and Rural Affairs (918010042HD030)]

**G004****Scale-up Study on the Fed-batch Fermentation of *Ralstonia eutropha* for the Production of Polyhydroxyalkanoates (PHAs)**Jong-Min Jeon<sup>1</sup>, Yung-Hun Yang<sup>2</sup>, and Jeong-Jun Yoon<sup>1\*</sup><sup>1</sup>Green & Sustainable Materials R&D Department, Korea Institute of Industrial Technology (KITECH), <sup>2</sup>Department of Biological Engineering, College of Engineering, Konkuk University

Polyhydroxyalkanoate (PHA) is bio-degradable materials to replace petroleum based plastics, and which made from renewable carbon sources has interest. In this study, one-pot fed-batch based on C/N ratio balance regulation was used throughout all scales for PHA production. *Ralstonia eutropha* was cultured to produce PHA with the gluconate as major carbon source to increase of biomass and PHA production. As a result, C/N ratio of feeding solution (total w/w: 3% sodium gluconate, 0.2% ammonium chloride) for biomass production was determined as 15:1, result in 13.5 g/L of biomass production in maximum. For PHA production, C/N ratio of feeding solution has changed 150:1, which was added after maximum biomass production, result in 50.6% of PHA content and 0.23 of PHA yield (PHA g/sodium gluconate g). In 50 L scale fermentation, biomass has reached on 11.5 g/L, 15% lesser than 1 L scale, while PHA content has increased up to 57%. In addition, pH, dissolved oxygen, agitation and organic acids were monitored during all period, figured out that dissolved oxygen ratio has much effect on cell growth and PHA accumulation in 50 L scale fermentation. These results suggest that application of one-pot feeding strategy could enhance the productivity of PHA and reduce additional cost by two-phase fermentation.

[This study was supported by National Research Foundation of Korea (NRF) grant funded by the Korea government (Ministry of Science and ICT, MSIT) (NRF-2017R1E1A1A01073690)]

**G005****Exploring Associations between Body Weight and Gut Microbiota Composition with Analysis of Opportunistic Pathogen Contamination from Post-weaning to Finishing Pigs**

Jae Hyoung Cho<sup>1</sup>, Hyeri Kim<sup>1</sup>, Sheena Kim<sup>1</sup>, Eun Sol Kim<sup>1</sup>, Jo Eun Kim<sup>2</sup>, Hyunjung Jung<sup>2</sup>, Tai-Young Hur<sup>2</sup>, Jin Ho Cho<sup>3</sup>, Minho Song<sup>4</sup>, Ju-Hoon Lee<sup>5</sup>, and Hyeun Bum Kim<sup>1\*</sup>

<sup>1</sup>Department of Animal Resources Science, Dankook University, <sup>2</sup>National Institute of Animal Science, Rural Development Administration, <sup>3</sup>Department of Food and Animal Science, Chungbuk National University, <sup>4</sup>Division of Animal and Dairy Science, Chungnam National University, <sup>5</sup>Department of Food Science and Biotechnology, Institute of Life Science and Resources, Kyung Hee University

In this study, the association between gut microbial community and body weights of pigs were investigated across different growth stages starting from pre-weaning, post-weaning to finishing at the age of 2, 8, 14 and 19 weeks respectively in addition to investigating opportunistic pathogens. A total of 64 fecal samples from heavy (n=8) and light (n=8) weight pigs were selected for cross-sectional and longitudinal studies of microbial communities. All pigs were fed a standard diet based on corn and soybean meal. The barcoded 16S rRNA gene amplicons from hypervariable regions (V5 to V6) of fresh fecal total DNA were sequenced using the Illumina MiSeq platform. Beta-diversity analysis revealed that microbial community structures were significantly different among the four age groups. At the genus level, at week 2, population of *Bacillus* was elevated in the light group; at week 8, higher abundance CF231 and Eubacterium was observed in the heavy group; at the week 14, *Turicibacter* and *Oscillospira* were observed as significantly high in heavy group; and at the week 19, *Sutterella*, *Faecalibacterium* and *Clostridium* showed higher abundance in heavy group. For analysis of pathogens, *Campylobacter*, which can cause watery diarrhea with mucous and occasional blood, was observed as significantly high in light group at week 14. Our results may provide insights into swine gut microbiome succession related to body weight gain at different growth stages.

[Supported by grants from MFDS and NRF]

**G006****Development of *Leuconostoc mesenteroides* EFEL Strain-specific Barcodes PCR and Its Application for Rapid and Accurate Identification and Quantification in Kimchi**

Seul-Ah Kim and Nam Soo Han\*

Brain Korea 21 Center for Bio-Resource Development, Division of Animal, Horticultural, and Food Sciences, Chungbuk National University

*Leuconostoc mesenteroides* has been considered as kimchi starter to manufacture commercial products with good quality in Korea. Identification of the starter and their enumeration during kimchi fermentation is of great importance for maintaining the quality. For this, in this study, *L. mesenteroides*-EFEL, used as commercial kimchi starter, strain-specific barcodes PCR method was developed by comparative pan-genome analysis and applied to rapid and accurate identification and quantification during kimchi fermentation. As result, three gene sequences were selected as targets for specific barcode primers for identification of *L. mesenteroides* EFEL. PCR analysis against genomic DNA of various lactic acid bacteria showed the primer set makes a specific binding with genomic DNA of *L. mesenteroides* EFEL strains. By using the primer set, a standard curve for qPCR analysis of *L. mesenteroides* was established and no significant difference ( $p < 0.05$ ) were found in the recovery yield of genomic DNA from MRS medium and kimchi. In addition, the qPCR method was proved to be used to enumerate *L. mesenteroides* EFEL during kimchi fermentation. In conclusion, the newly developed PCR method in this study can be applied to rapidly identify and to accurately *L. mesenteroides* EFEL population change during kimchi fermentation process.

**G008****Isolation of 2'-Fucosyllactose-utilizing *Bifidobacterium* Species from Feces of Breast-feeding Infants**

Dong Hyeon Lee and Nam Soo Han\*

*Brain Korea 21 Center for Bio-Resource Development, Division of Animal, Horticultural, and Food Sciences, Chungbuk National University*

Human milk oligosaccharides (HMO) reach to distal colon, collectively modify the gut microbiota, and show various health functions. However, the utilization of HMO by individual gut bacteria was not known well. The aim of this study was to isolate various *Bifidobacterium* spp. that can use 2-fucosyllactose (2'-FL) as carbon source from fecal samples of breast-feeding infants. For classification of isolates, a multiplex PCR using species-specific primers based on 16S rRNA through 23S rRNA region was used, and for identification, 16S-rRNA gene sequencing was conducted. Cell growth of each isolates was measured in semi-modified MRS medium containing 1% 2'-FL by optical density (O.D 600 nm), TLC, and HPLC. As results, 11 *Bifidobacterium* spp. were isolated from the infant feces. Among them, seven isolates were identified as *B. infantis* and two isolates were *B. bifidum* and one isolate was *B. breve*. All isolates could utilize 2'-FL for their cell growth and a strain of *B. bifidum* showed the highest cell growth (O.D 1.15). Notably, *B. infantis* and *B. breve* metabolized fucose residue completely during cell growth, while *B. bifidum* remained it in the medium. In conclusion, we isolated *B. bifidum*, *B. infantis* and *B. breve* utilizing 2'-FL as carbon source from breast-feeding infant.

**G009****Enzymatic Characterization and Action Modes of Arabinan-specific *exo*- $\alpha$ -(1,2)-L-Arabinofuranosidase GH43 from *Bacteroides intestinalis***

Jeong-Rok Song and Tae-Jip Kim\*

*Brain Korea 21 Center for Bio-Resource Development, and Division of Animal, Horticultural and Food Sciences, Graduate School of Chungbuk National University*

The gene encoding an *exo*- $\alpha$ -(1,2)-L-arabinofuranosidase GH43 (BtinAF<sub>2</sub>) was cloned from *Bacteroides intestinalis* DSM 17393, and was expressed in *Escherichia coli* MC1061. Its open reading frame consists of 981 nucleotides encoding 326 amino acids (34.4 kDa). The corresponding gene with C-terminal six histidines was cloned into the constitutive expression vector of pHCXHD, which was designated as pHBtinAF<sub>2</sub>. Recombinant BtinAF<sub>2</sub> showed the highest activity against sugar beet arabinan in 50 mM sodium acetate buffer (pH 6.0) at 50°C. However, it could hardly hydrolyze debranched (linear) arabinan and arabinooligosaccharides (AOS) as well as *p*-nitrophenyl arabinofuranoside. The thin layer chromatography analyses revealed that BtinAF<sub>2</sub> can exclusively hydrolyze  $\alpha$ -(1,2)-L-arabinofuranosidic branches of single- and double-substituted (branched) AOS to produce only L-arabinose and linear AOS. On the contrary, it could hardly hydrolyze  $\alpha$ -(1,3)-L-arabinofuranosidic branched AOS and  $\alpha$ -(1,2)- and/or  $\alpha$ -(1,3)-branched arabinooligosaccharides. BtinAF<sub>2</sub> shares 65.4% of amino acid sequence identity with the other known *exo*- $\alpha$ -(1,2)-L-arabinofuranosidase from *Cellvibrio japonicus*. Accordingly, BtinAF<sub>2</sub> is categorized as an *exo*-acting hydrolase being highly specific for  $\alpha$ -(1,2)-L-arabinofuranosidic branches of arabinose-containing substrates. [Supported by the National Research Foundation: NRF-2020R1F1A1076947]

**G010****Functional Expression and Enzymatic Characterization of Two Eukaryotic  $\alpha$ -L-Arabinofuranosidases GH51 from *Saccharomycopsis fibuligera* KJJ81**Tae Hyeon Park<sup>1</sup>, Chang-Yun Choi<sup>1</sup>, Hyun Ah Kang<sup>2</sup>, and Tae-Jip Kim<sup>1\*</sup><sup>1</sup>Brain Korea 21 Center for Bio-Resource Development, and Division of Animal, Horticultural and Food Sciences, Graduate School of Chungbuk National University, <sup>2</sup>Department of Life Science, Chung-Ang University

$\alpha$ -L-Arabinofuranosidases (AFs, EC 3.2.1.55) are *exo*-acting arabinosyl hydrolases producing L-arabinose, which have been mainly studied from bacteria and fungi. In this study, two putative AF GH51 (SfAF-A<sub>a</sub> and A<sub>b</sub>) genes were cloned from a yeast, *Saccharomycopsis fibuligera*, and were expressed in *Escherichia coli*. Both SfAF-A<sub>a</sub> and SfAF-A<sub>b</sub> are composed of 517 amino acid residues, and they share 92% of amino acid sequence identity with each other. Fungal AF from *Penicillium chrysogenum*. and bacterial AF from *Thermotoga maritima*. showed 48 and 41% of amino acid sequence identities with those of SfAF-As, respectively. Each gene encoding SfAF-A with C-terminal six histidines was expressed in *E. coli* BL21(DE3). SfAF-A<sub>a</sub> showed the highest activity on *p*-nitrophenyl arabinofuranoside (*p*NPAf) in 50 mM sodium phosphate (pH 7.0) and 40°C, whereas SfAF-A<sub>b</sub> has its optimum at pH 6.0 and 50°C. These AFs can hydrolyze  $\alpha$ -(1,2)-,  $\alpha$ -(1,3)-, and  $\alpha$ -(1,5)-arabinofuranosidic linkages in single-substituted and/or short-chain arabinoxylooligosaccharides and arabinooligosaccharides, except for double-substituted substrates. On the contrary, both SfAF-As can hardly hydrolyze polymeric substrates such as arabinans and arabinoxylans.

[Supported by the National Research Foundation: NRF-2017M3C1B5019292]

**G011****Enzymatic Characterization of  $\gamma$ -Glutamyl Hydrolase from *Bacillus licheniformis* DSM 13 Heterologously Expressed in *Bacillus subtilis***

Damee Park, Ji-Eun Park, and Tae-jip Kim\*

Brain Korea 21 Center for Bio-Resource Development, and Division of Animal, Horticultural and Food Sciences, Graduate School of Chungbuk National University

$\gamma$ -Glutamyl hydrolases (GGHs, EC3.4.19.9) are *endo*-acting hydrolase which cleaves the  $\gamma$ -glutamyl bonds between D- and/or L-glutamate of poly- $\gamma$ -glutamate ( $\gamma$ -PGA). In the present study, the gene encoding a  $\gamma$ -glutamyl hydrolase (BIGGH13) from *Bacillus licheniformis* DSM 13 was cloned into the constitutive expression vector, pUBRT-A. The open reading frame of BIGGH13 (1,245 nucleotides) encodes 414 amino acids with the predicted molecular mass of 45.6 kDa. BIGGH13 shares 58% of amino acid sequence identity with a GGH from *B. subtilis*. Recombinant BIGGH13 with C-terminal six histidines was extracellularly expressed, concentrated, and purified from *Bacillus subtilis* KCTC 3135 harboring pUABIGGH via N-NTA affinity chromatography. BIGGH13 showed the highest enzyme activity against  $\gamma$ -PGA in 50 mM sodium acetate buffer (pH 6.0) at 55°C. The SDS-PAGE analysis revealed that *endo*-acting BIGGH13 can produce low molecular weight  $\gamma$ -PGA and  $\gamma$ -glutamyl peptides from  $\gamma$ -PGA polymer. As  $\gamma$ -glutamyl peptides are known to enhance the 'Kokumi' taste, the highly active GGH-producing microorganisms can be the promising starter candidates for the soybean-fermentation.

[Supported by the National Research Foundation: NRF-2017M3C1B5019292]

## G012

### Development of Strain-specific Primers for Identification of *Lactobacillus plantarum* AMT-74419

Jae Woong Choi, Nho-Eul Song, Somang Jung, Sung Hun Yi, and Young-Kyung Rhee\*

*Korea Food Research Institute*

*Lactobacillus plantarum* AMT-74419 which was isolated from Kimchi has many properties for using probiotics. For commercialization and quality assurance, strain-specific primer sets were developed. To figure out specific region of *L. plantarum* AMT-74419, we performed comparative genomics with other 19 *L. plantarum* strains. 13 Singleton genes were screened and AMT74419\_02068 which has not been found in the other bacteria was selected. As a result, PCR product was confirmed in only AMT-74419. Primer sensitivity was analyzed by serial dilutions of purified genomic DNA from 50 ng/μl to 1.56 ng/μl concentration. From this result, these primer could be applied for identifying specific strain in probiotics products.

## G013

### Sunscreen Boosting Effect by Solid Lipid Nanoparticles-loaded Fucoxanthin Formulation

Yong-Jik Lee<sup>1</sup>, Seok-Cheol Cho<sup>2</sup>, Jae Hak Shon<sup>3</sup>, Han-Seung Lee<sup>3</sup>, Sang-Jae Lee<sup>3</sup>, and Gae-Won Nam<sup>1\*</sup>

<sup>1</sup>Department of Bio-cosmetics, Seowon University, <sup>2</sup>Food Science & Engineering Major, Seowon University, <sup>3</sup>Major in Food Biotechnology and Research Center for Extremophiles & Marine Microbiology, Silla University

Fucoxanthin is a bioactive compound that is a kind of natural carotenoid. Fucoxanthin is known to protect against UV-B-induced cell damage in hairless mice, even though it is physicochemically unstable to heat and acid due to its polyunsaturated structure, indicating that fucoxanthin possesses a low bioavailability, and this disadvantage limits its application in the cosmetic industry. Solid lipid nanoparticle (SLN) systems are known to be suitable as carriers for sunscreen agents. In this research work, the sunscreen-boosting effect of SLN, as a deliverer of functional ingredient, especially fucoxanthin, has been developed and evaluated by comparing the sunburn protection factors (SPF) of macroemulsion (cream and lotion type) and an SLN formula containing various kinds of sunscreen agents, respectively. Several results such as stability test, particle size, DSC analysis, and X-ray analysis show that the SLN formula loading fucoxanthin has the possibility of being a stable and high-functioning ingredient delivery system. Moreover, the SLN formula has shown a higher SPF value than others, meaning that the SLN formula exhibits a good sunscreen-boosting effect. This study indicates that the use of SLN as a carrier enhanced the bioavailability of fucoxanthin and shows that SLN could be a promising carrier for the production of sunscreen products by allowing the scaling-up of production.

**G014****Elucidating the Regulatory Elements for Transcription Termination and Post-transcriptional Processing in the *Streptomyces clavuligerus* Genome**Soonkyu Hwang<sup>1,2</sup>, Namil Lee<sup>1,2</sup>, Donghui Choe<sup>1,2</sup>, Yongjae Lee<sup>1,2</sup>, Woori Kim<sup>1,2</sup>, Yujin Jeong<sup>1,2</sup>, Suhyung Cho<sup>1,2</sup>, Bernhard Palsson<sup>3,4,5</sup>, and Byung-Kwan Cho<sup>1,2,6\*</sup>

<sup>1</sup>Department of Biological Sciences, Korea Advanced Institute of Science and Technology, <sup>2</sup>KAIST Institute for the BioCentury, Korea Advanced Institute of Science and Technology, <sup>3</sup>Department of Bioengineering, University of California San Diego, La Jolla, CA 92093, USA, <sup>4</sup>Department of Pediatrics, University of California San Diego, La Jolla, CA 92093, USA, <sup>5</sup>Novo Nordisk Foundation Center for Biosustainability, 2800 Kongens Lyngby, Denmark, <sup>6</sup>Intelligent Synthetic Biology Center

Identification of transcriptional regulatory elements in the GC-rich *Streptomyces* genome is essential for the production of novel biochemicals from secondary metabolite biosynthetic gene clusters. In this study, we identified the transcriptional regulatory elements in  $\beta$ -lactam antibiotic-producing *Streptomyces clavuligerus* ATCC 27064 by determining a total of 1,427 transcript 3'-end positions (TEPs) using term-seq method. The data integration with dRNA-seq and RNA-seq data generated 1,648 transcription units (TUs) and 910 transcription unit clusters (TUCs). TU architecture showed that the transcript abundance in TU isoforms of a TUC was potentially affected by the sequence context of their TEPs, suggesting that the regulatory elements of TEPs could control the transcription level in additional layers. We also found that 189 non-coding TUs, contained potential *cis*- and *trans*-regulatory elements that played a major role in regulating the 5'- and 3'-UTR. These findings highlight the role of transcriptional regulatory elements in transcription termination and post-transcriptional processing in *Streptomyces*.

[This work was supported by the Novo Nordisk Foundation (NNF10CC1016517 to B.O.P.), the Intelligent Synthetic Biology Center of Global Frontier Project (2011-0031957 to B.-K.C.), and the Bio & Medical Technology Development Program (2018M3A9F3079664 to B.-K.C.) of the National Research Foundation funded by the Korean government (MSIT).]

**G015****Iron Competition Triggers Antibiotic Biosynthesis in *Streptomyces coelicolor* during Coculture with *Myxococcus xanthus***

Namil Lee<sup>1</sup>, Woori Kim<sup>1</sup>, Yongjae Lee<sup>1</sup>, Suhyung Cho<sup>1</sup>, Kyoung-Soon Jang<sup>2,3</sup>, Sun Chang Kim<sup>1,4</sup>, Bernhard Palsson<sup>5,6,7</sup>, and Byung-Kwan Cho<sup>1,4,7\*</sup>

<sup>1</sup>Department of Biological Sciences and KI for the BioCentury, Korea Advanced Institute of Science and Technology, <sup>2</sup>Biomedical Omics Group, Korea Basic Science Institute, <sup>3</sup>Division of Bio-Analytical Science, University of Science and Technology, <sup>4</sup>Intelligent Synthetic Biology Center, <sup>5</sup>Department of Bioengineering, University of California San Diego, La Jolla, CA 92093, USA, <sup>6</sup>Department of Pediatrics, University of California San Diego, La Jolla, CA 92093, USA, <sup>7</sup>Novo Nordisk Foundation Center for Biosustainability, Technical University of Denmark, Lyngby 2800, Denmark

Microbial coculture to mimic the ecological habitat has been suggested as an approach to elucidate the effect of microbial interaction on secondary metabolite biosynthesis of *Streptomyces*. Here, we found that iron competition triggered antibiotic biosynthesis in *Streptomyces coelicolor* during coculture with *Myxococcus xanthus*. During coculture, *M. xanthus* enhanced the production of a siderophore, myxochelin, leading *M. xanthus* to dominate iron scavenging and *S. coelicolor* to experience iron restricted conditions. This chemical competition activated the actinorhodin biosynthetic gene cluster (BGC) and the branched-chain amino acid degradation pathway which implies the potential to produce precursors, along with activation of a novel actinorhodin export system. Furthermore, we found that iron restriction increased the expression of 21 secondary metabolite BGCs (smBGCs) in other *Streptomyces* species. These findings suggested that the availability for key ions stimulates specific smBGCs, which had the potential to enhance secondary metabolite biosynthesis in *Streptomyces*.

[This work was supported by a grant from the Novo Nordisk Foundation (NNF10CC1016517), ISBC of the Global Frontier Project (2011-0031957), the Basic Science Research Program (2018R1A1A3A04079196), the Basic Core Technology Development Program for the Oceans and the Polar Regions (2016M1A5A1027458), and Bio & Medical Technology Development Program (2018079664) through the NRF.]

**G016****Delivery of Protein Cargos for Biotechnological Engineering in Prokaryotes**

Hyang-Mi Lee, Jun Ren, Kha Mong Tran, Hyunjoo Kim, and Dokyun Na\*

Department of Biomedical Engineering, Chung-Ang University

Here, we developed a method to deliver protein cargos into living bacteria using cell-penetrating peptides (CPP). We evaluated the cell penetration efficiency of 98 CPPs with our optimized delivery method. We found that cell penetration efficiency was enhanced for the CPP-tagged proteins up to 12-fold in *E. coli* compared with untagged protein. To demonstrate the applicability of the CPP-conjugated strategy for protein delivery, we used meganuclease I-SceI for plasmid removal in *E. coli* harboring different plasmid copy numbers. CPP-conjugated I-SceI successfully eliminated low-to-high copy number plasmids. In addition, we developed a marker gene excision method based on a CPP-conjugated Cre recombinase/loxP system. Our method showed high efficiencies in markerless gene editing for metabolic engineering and ease of conducting experiments in *Methylomonas* sp. DH-1. Taken together, the CPPs can be used to deliver other proteins as well for microbial engineering.

**G017****De Novo Biosynthesis Pathway of Acrylic Acid in Metabolically Engineered *Escherichia coli***Hyeon Yu<sup>1</sup>, Yoo-Sung Ko<sup>1,2</sup>, Je Woong Kim<sup>1,2</sup>, Tong Un Chae<sup>1</sup>, Chan Woo Song<sup>1</sup>, and Sang Yup Lee<sup>1,2,3\*</sup>

<sup>1</sup>Metabolic and Biomolecular Engineering National Research Laboratory, Systems Metabolic Engineering and Systems Healthcare (SMESH) Cross Generation Collaborative Laboratory, Department of Chemical and Biomolecular Engineering (BK21 Plus Program), Institute for the BioCentury, Korea Advanced Institute of Science and Technology (KAIST), <sup>2</sup>Systems Metabolic Engineering and Systems Healthcare Cross-Generation Collaborative Laboratory, KAIST, <sup>3</sup>BioProcess Engineering Research Center, KAIST

Acrylic acid (AA) serves as important industrial chemical for various applications including acrylate esters and superabsorbent polymers. Here, we report the development of a new biosynthetic pathway through the  $\beta$ -alanine (BA) route for the production of AA in metabolically engineered *Escherichia coli* from glucose. We first validated the operation of the downstream pathway, and then constructed the downstream pathway by introducing efficient enzymes; such as Act, Acl2 AND YciA. For the enhanced production of AA we introduced the downstream pathway into the BA producing *E. coli* strain for the direct fermentative production of AA from glucose. AA production was also enhanced through the expression of the upstream genes (*panD* and *aspA*) under the constitutive BBa\_J23100 promoter. Replacement of the native promoter of the *acsGen* with the BBa\_J23100 promoter in the genome increased AA production to 55.7 mg/L in flask. Resulting engineered strain was able to produce the 237 mg/L of AA in fed-batch fermentation as a highest titer to date.

[This work was supported by the Technology Development Program to Solve Climate Changes on Systems Metabolic Engineering for Biorefineries (NRF-2012M1A2A2026556 and NRF-2012M1A2A2026557). This work was further supported by Hanwha]

**G018****Production of Aromatic Polyesters from Renewable Resource by Metabolically Engineered *Escherichia coli***Hyeon Yu<sup>1</sup>, Jung Eun Yang<sup>1</sup>, Si Jae Park<sup>2</sup>, Won Jun Kim<sup>1</sup>, Hyeon Jun Kim<sup>1</sup>, Bumjoon J. Kim<sup>1</sup>, Hyuk Lee<sup>3</sup>, Jihoon Shin<sup>4</sup>, and Sang Yup Lee<sup>1\*</sup>

<sup>1</sup>Metabolic and Biomolecular Engineering National Research Laboratory, Department of Chemical and Biomolecular Engineering (BK21 Plus Program), BioProcess Engineering Research Center, Center for Systems and Synthetic Biotechnology, and Institute for the BioCentury, Korea Advanced Institute of Science and Technology (KAIST), <sup>2</sup>Division of Chemical Engineering and Materials Science, Ewha Womans University, <sup>3</sup>Division of Drug Discovery Research, Korea Research Institute of Chemical Technology, <sup>4</sup>Center for Bio-based Chemistry, Green Chemistry & Engineering Division, Korea Research Institute of Chemical Technology

Aromatic polyesters are widely used indispensable plastics currently produced from petroleum. A D-phenyllactate producing strain that overexpresses the *Clostridium difficile* isocaprolyl-CoA:2-hydroxyisocaproate CoA-transferase (HadA) and evolved polyhydroxyalkanoate synthase genes can produce poly(52.3 mol% 3-hydroxybutyrate(3HB)-co-47.7 mol% D-phenyllactate) from glucose and sodium 3HB. Various poly(3HB-co-D-phenyllactate) polymers are produced from glucose, a renewable carbon source, by heterologous expression of *Cupriavidus necator*  $\beta$ -ketothiolase (*phaA*) and reductase (*phaB*) genes. One step fermentation of this metabolically engineered strain produces poly(61.9 mol% 3HB-co-38.1 mol% D-phenyllactate) in 13.9 g/L. The engineered microbial system should be useful for the production of aromatic polyesters from renewable carbon source.

[This work was supported by the Intelligent Synthetic Biology Center through the Global Frontier Project (2011-0031963) and also by the Technology Development Program to Solve Climate Changes on Systems Metabolic Engineering for Biorefineries (NRF2012M1A2A2026556 and NRF-2012M1A2A2026557) from the Ministry of Science and ICT through the National Research Foundation of Korea.]

**G019****Biosynthesis of 4-Amino 1-Butanol from Metabolically Engineered *Corynebacterium glutamicum***Hyeeun Yu<sup>1</sup>, Cindy Pricilia Surya Prabowo<sup>1,3</sup>, Jae Ho Shin<sup>1,3</sup>, Jae Sung Cho<sup>1,3</sup>, Tong Un Chae<sup>1,3</sup>, and Sang Yup Lee<sup>1,2,3\*</sup><sup>1</sup>*Metabolic and Biomolecular Engineering National Research Laboratory, Systems Metabolic Engineering and Systems Healthcare (SMESH) Cross Generation Collaborative Laboratory, Department of Chemical and Biomolecular Engineering (BK21 Plus Program), Institute for the BioCentury, Korea Advanced Institute of Science and Technology (KAIST),* <sup>2</sup>*BioInformatics Research Center, KAIST,* <sup>3</sup>*BioProcess Engineering Research Center, KAIST*

4-Amino-1-butanol (4AB) plays a role as important intermediate compound for drugs and a precursor of biodegradable polymers used for gene and protein delivery. Since there has been no report on the bio-based production from renewable resources. Now, we report for the fermentative biosynthesis of 4AB from renewable resource glucose through newly designed pathway consisting of a putrescine aminotransferase (encoded by *ygjG*) and an aldehyde dehydrogenase (encoded by *yqhD*) from *Escherichia coli*, which convert putrescine to 4AB by the metabolically engineered *Corynebacterium glutamicum*. To enhance further 4AB production, several metabolic engineering strategies such as optimizing culture condition, eliminating competing pathways, and fine-tuning of the expression levels of *ygjG* and *yqhD* were applied. The finally engineered *C. glutamicum* strain produced 24.7 g/L of 4AB in a fed batch culture. The strategies reported here should be useful for developing other microbial strains capable of efficiently producing primary amino alcohols.

[This work was supported by the Technology Development Program to Solve Climate Changes (Systems Metabolic Engineering for Biorefineries) from the Ministry of Science and ICT through the National Research Foundation (NRF) of Korea (NRF 2012M1A2A2026556 and NRF 201 2M1A2A2026557).]

**G020****Microbial Production of Biodegradable Polymer, Poly(D-Lactate-co-Glycolate-co-4-Hydroxybutyrate) for New Biomedical Application**Hyeeun Yu<sup>1</sup>, So Young Choi<sup>1</sup>, Tong Un Chae<sup>1,2</sup>, Jihoon Shin<sup>3</sup>, Jung Ae Im<sup>1,4</sup>, and Sang Yup Lee<sup>1,2,4\*</sup><sup>1</sup>*Metabolic and Biomolecular Engineering National Research Laboratory, Systems Metabolic Engineering and Systems Healthcare Cross-Generation Collaborative Laboratory, Department of Chemical and Biomolecular Engineering (BK21 Plus Program), Institute for the BioCentury, Korea Advanced Institute of Science and Technology (KAIST),* <sup>2</sup>*Applied Science Research Institute, KAIST,* <sup>3</sup>*Center for Bio-based Chemistry, Green Chemistry and Engineering Division, Korea Research Institute of Chemical Technology,* <sup>4</sup>*BioProcess Engineering Research Center and Bioinformatics Research Center, KAIST*

Poly(D-lactate-co-glycolate-co-4-hydroxybutyrate) [poly(D-LA-co-GA-co-4HB)] and poly(D-lactate-co-glycolate-co-4-hydroxybutyrate-co-D-2-hydroxybutyrate) [poly(D-LA-co-GA-co-4HB-co-D-2HB)] have attracted attention as potential biomedical polymers. Enhanced production of such polymers was performed on genetically engineered *Escherichia coli*. To validate the polymer properties, engineered *E. coli* strain expressing *xylBC* from *Caulobacter crescentus*, evolved PHA synthase from *Pseudomonas* sp. MBEL 6-19 (*phaC1437*), and evolved propionyl-CoA transferase from *Clostridium propionicum* (*pct540*) in a defined medium supplemented with sodium 4-hydroxybutyrate (4HB) at various concentrations. The 4HB biosynthetic pathway was additionally constructed by expressing *Clostridium kluyveri* *sucD* and *4hbD* for the production of polymer without 4HB feeding. Resulting strain produced poly(D-LA-co-GA-co-4HB-co-D-2HB) and poly(D-LA-co-GA-co-4HB) from glucose and xylose. Through modulating the expression levels of the heterologous genes and performing fed-batch cultures, the polymer content and titer could be increased to 65.76 wt% and 6.19 g/L, respectively, while the monomer fractions in the polymers could be altered as desired. The polymers produced, in particular, the 4HB-rich polymers showed viscous and sticky properties suggesting that they might be used in medical applications.

[This study was supported by the Technology Development Program to Solve Climate Changes (Systems Metabolic Engineering for Biorefineries) from the Ministry of Science and ICT through the National Research Foundation (NRF) of Korea]

**G021****Metabolic Engineering of *Escherichia coli* for High-level 3-Hydroxypropionic Acid Production Using Crude Glycerol**Ji Hye Hyun<sup>1</sup>, Je Woong Kim<sup>1,3</sup>, Yoo-Sung Ko<sup>1,3</sup>, Tong Un Chae<sup>1,3</sup>, and Sang Yup Lee<sup>1,2,3\*</sup>

<sup>1</sup>Metabolic and Biomolecular Engineering National Research Laboratory, Systems Metabolic Engineering and Systems Healthcare Laboratory, Department of Chemical and Biomolecular Engineering (BK21 Plus Program), Institute for the BioCentury, Korea Advanced Institute of Science and Technology (KAIST), <sup>2</sup>BioProcess Engineering Research Center, KAIST, <sup>3</sup>Bioinformatics Research Center, KAIST

3-Hydroxypropionic acid (3-HP), which is an industrially important C3 chemical, is over-produced in a high level from glycerol/crude glycerol as a sole carbon source using metabolic engineered *Escherichia coli* for the potential of the industrial scale production. Glycerol-dependent 3-HP biosynthetic pathway is constructed with the introduction of heterologous genes and subsequent screening of these genes combinations is performed. By fed-batch fermentation, 49.9 g/L of 3-HP is produced. Additional improvement in 3-HP production is successful through manipulating fermentation condition leading to produce 71.9 g/L of 3-HP yielding 0.466 g 3-HP/g glycerol with the productivity of 1.94 g/L. The strain also produce 61.0 g/L of 3-HP yielding 0.594 g 3-HP/g crude glycerol and with the productivity of 2.28 g/L/h when taking crude glycerol as its sole carbon source.

[The work on was supported by the Technology Development Program to Solve Climate Changes on Systems Metabolic Engineering for Biorefineries from the Ministry of Science, ICT and Future Planning (MSIP) through the National Research Foundation (NRF) of Korea (NRF-2012M1A2A2026556 and NRF-2012M1A2A2026557) and by Hanwha Chemical].

**G022****Reconstruction of Formic Acid and CO<sub>2</sub> Assimilation Pathways in *Escherichia coli***Ji Hye Hyun<sup>1,2</sup>, Junho Bang<sup>1,2</sup>, and Sang Yup Lee<sup>1,2,3\*</sup>

<sup>1</sup>Metabolic and Biomolecular Engineering National Research Laboratory, Department of Chemical and Biomolecular Engineering, Korea Advanced Institute of Science and Technology (KAIST), <sup>2</sup>Systems Metabolic Engineering and Systems Healthcare Cross-Generation Collaborative Laboratory, KAIST, <sup>3</sup>Bioinformatics Research Center and BioProcess Engineering Research Center, KAIST

Bioconversion of gaseous one-carbon (C1) compounds are arising as vital issue to solve environmental problems. The bioconversion technologies have a number of advantages such as environmental friendliness and low energy requirements. Here, efficient assimilation of Formic acid (FA) and CO<sub>2</sub> in *Escherichia coli* is performed by reconstructing C1 assimilation pathway. First, the synthetic C1 assimilation pathway is introduced. Moreover, the glycine cleavage system is reversed by knock-out of *gcvR* gene and overexpression of *gcvTHP* genes. By knock-out of unnecessary genes and overexpression of the reconstructed THF cycle, *gcvTHP*, and *lpd* genes in one vector, the pyruvate-forming flux from FA and CO<sub>2</sub> is increased to 14.9%. Additionally, by introducing the *Candida boidinii* formate dehydrogenase (Fdh) gene, there has been reduction of glucose usage and the redox supplying became possible. The resulting strain shows specific glucose, FA, and CO<sub>2</sub> consumption rates of 370.2, 145.6, and 14.9 mg/g dry cell weight (DCW)<sup>-1</sup>·h<sup>-1</sup>, respectively. The C1 assimilation pathway consume 21.3 wt% of FA. Importantly, cells sustain slight growth using only FA and CO<sub>2</sub> after the depletion of glucose. This result suggest that the sustainable cell growth solely on C1 compounds without additional carbon source such as glucose, will be realized.

[This work was supported by the C1 Gas Refinery Program through the National Research Foundation of Korea (NRF) funded by the Ministry of Science and ICT Grant (NRF-2016M3D3A1A01913250)]

**G023****Metabolic Engineering of *Rhodococcus opacus* for High-level Fatty Acid and Fuel Production**Ji Hye Hyun<sup>1</sup>, Hye Mi Kim<sup>1</sup>, Tong Un Chae<sup>1</sup>, So Young Choi<sup>1</sup>, Won Jun Kim<sup>1</sup>, and Sang Yup Lee<sup>1,2\*</sup>

<sup>1</sup>Metabolic and Biomolecular Engineering National Research Laboratory, Systems Metabolic Engineering and Systems Healthcare Cross-Generation Collaborative Laboratory, Department of Chemical and Biomolecular Engineering (BK21 Plus program), Center for Systems and Synthetic Biotechnology, Institute for BioCentury, Korea Advanced Institute of Science and Technology (KAIST), <sup>2</sup>Bioinformatics Research Center

Production of free fatty acids (FFAs) and the derivatives from renewable non-food biomass by microbial fermentation is in great interest. Here, we report the development of metabolically engineered *Rhodococcus opacus* strains producing FFAs, fatty acid ethyl esters (FAEEs) and long-chain hydrocarbons (LCHCs). The optimization of culture condition is performed to produce 82.9 g/L of triacylglycerols from glucose. An engineered strain with the deletion of acyl-CoA synthetases, overexpressing three lipases with lipase specific-foldase produces 50.2 g/L of FFAs. Another engineered strain with the deletion of acyl-CoA dehydrogenases, overexpressing lipases, foldase, acyl-CoA synthetase, and heterologous aldehyde/alcohol dehydrogenase and wax ester synthase produces 21.3 g/L of FAEEs. A third engineered strain with the deletion of acyl-CoA dehydrogenases and alkane-1 monooxygenase, overexpressing lipases, foldase, acyl-CoA synthetase, and heterologous acyl-CoA reductase, acyl-ACP reductase and aldehyde deformylating oxygenase produces 5.2 g/L of LCHCs. Metabolic engineering strategies and engineered strains developed here will help to establish oleaginous biorefinery platform for the sustainable production of chemicals and fuels.

[This work was supported by the Technology Development Program to Solve Climate Changes on Systems Metabolic Engineering for Biorefineries from the Ministry of Science and ICT through the National Research Foundation (NRF) of Korea (NRF-2012M1A2A2026556 and NRF-2012M1A2A2026557)]

**G024****Biosynthesis of Methyl-anthranilate, a Grape Flavor Compound by Metabolically Engineered Microbes**Ji Hye Hyun<sup>1</sup>, Zi Wei Luo<sup>1</sup>, Jae Sung Cho<sup>1</sup>, and Sang Yup Lee<sup>1,2\*</sup>

<sup>1</sup>Metabolic and Biomolecular Engineering National Research Laboratory, Systems Metabolic Engineering and Systems Healthcare Cross-Generation Collaborative Laboratory, Department of Chemical and Biomolecular Engineering (BK21 Plus program), Center for Systems and Synthetic Biotechnology, Institute for BioCentury, Korea Advanced Institute of Science and Technology (KAIST), <sup>2</sup>Bioinformatics Research Center

Methyl anthranilate (MANT) is a widely used compound that has grape scent and flavor, but it is currently produced by petroleum-based processes. Here we report the direct fermentative production of MANT from glucose by metabolic engineered *Escherichia coli* and *Corynebacterium glutamicum* strains harboring a synthetic plant-derived metabolic pathway. With the optimization of the key enzyme anthranilic acid (ANT) methyltransferase1 (AAMT1) expression, the increasement of the direct precursor ANT supply, and the enhancement of the intracellular availability and salvage of the cofactor S-adenosyl-L-methionine required by AAMT1, the production of MANT has been improved in both engineered microorganisms. Furthermore, *in situ* two-phase extractive fermentation using tributyrin as an extractant is built up to overcome the toxicity of MANT. Fed-batch cultures of the final engineered *E. coli* and *C. glutamicum* strains in two-phase cultivation mode lead to the production of 4.47 g/L and 5.74 g/L MANT, in minimal media containing glucose. The metabolic engineering strategies built here will be useful for the production of volatile aromatic esters including MANT.

[This work was supported by the Technology Development Program to Solve Climate Changes on Systems Metabolic Engineering for Biorefineries from the Ministry of Science, ICT and Future Planning (MSIP) through the National Research Foundation (NRF) of Korea (NRF-2012M1A2A2026556 and NRF-2012M1A2A2026557)]

**H001****Molecular Study of Microbiome-regulated Insect Odorant Receptors**

Junho Lee

*Chonnam National University*

The highly specialized olfactory receptor neurons (ORNs) on the antennae of male moths can recognize blends of several pheromone components. In present study, a total of six candidate pheromone compounds are found and functionally characterized in the electrophysiological study. In here, we report on novel candidate pheromone compounds in the same species. The olfactory receptor is analyzed revealed that finding compounds are specifically affect in on olfactory receptor. *In silico* study revealed that odorant-gated ion channels comprised of a highly conserved co-receptor and our chemicals are binding at extra cellular site. Functional analyses on the odorant-gated ion channels comprised of a highly conserved co-receptor were then performed using the heterologous expression system of *Xenopus* oocytes. pheromone components did not respond to any tested pheromone components and analogs. These results may contribute to clarifying how pheromone detection works in odorant-gated ion channels comprised of a highly conserved co-receptor.

**H002****Stochastic Processes Dominate Nematode Community Structure in Mountain Ecosystems in Northeast Asia**Shuqi Zou<sup>1</sup>, Ke Dong<sup>1</sup>, Rongrong Lin<sup>2</sup>, and Sangseob Lee<sup>1\*</sup><sup>1</sup>*Kyonggi University*, <sup>2</sup>*Kookmin University*

Nematode occupies an important position in ecosystems, but there are few studies on nematodes in term of the assembly processes of their communities. Aiming to reveal the assembly process of the nematode communities in mountain ecosystems and to identify the potential influencing factors of the assembly processes, we collected soil samples on three mountains in the Northeast Asia region at different elevations and classified the nematode communities by sequencing the amplicons of partial 18S rRNA gene of nematodes.  $\beta$ -Nearest taxon index was calculated to infer the community assembly processes. We found that regardless of which mountain, the stochastic process dominates the assembly process of the nematode community, although the changing trends along the gradient were different between mountains. Elevation itself showed a significant impact on the community assembly processes, while other environmental factors also contributed. However, the impact of environmental factors was not consistent between mountains. Our results suggested that soil pH, NO<sub>3</sub><sup>-</sup> and annual average temperature were the most important factors affecting the community assembly processes. We further compared the assembly processes of different nematode functional guilds, and found the bacterial-feeding and omnivorous nematode exhibited similar patterns on the community assembly processes with the whole nematode community.

[This work was supported by a research grant from NRF under NRF-2018R1C1B6007755.]

### H003

#### **Elevational Variation in Soil Nematode Diversity and the Influencing Factors**

Zhi Yu<sup>1</sup>, Ke Dong<sup>1</sup>, Rongrong Lin<sup>2</sup>, and Sangseob Lee<sup>1\*</sup>

<sup>1</sup>*Kyonggi University*, <sup>2</sup>*Kookmin University*

The elevational distribution of soil nematodes has been widely investigated with no general patterns discovered in term of their diversity pattern. Here, we investigated the soil nematode communities on three mountains to understand their diversity patterns and to look for the best explanatory factors of them. High throughput sequencing approach targeting the 18S rRNA gene was used to study the nematode communities from different elevational transects. Our results indicated that the diversity of the whole soil nematodes showed a high correlation with elevation, and maximized in mid elevations on Mt. Halla and Mt. Norikura, whereas no significant elevational difference was found on Mt. Jiri. We found that climatic factors were the most important factors influencing the diversity on Mt. Halla and Mt. Norikura, whereas edaphic parameters had the largest impact on Mt. Jiri. Moreover, multiple regression model showed that the overall explanatory proportion of measured environmental parameters did not change dramatically after removing the mid-domain effect. We also tested the assumptions of Rapoport's rule by comparing the average elevational distribution range of the community members. We found that the ranges changed significantly with the increasing elevation on Mt. Norikura, but not on Mt. Halla and Mt. Jiri.

[This work was supported by a research grant from National Research Foundation of Korea under the number of NRF-2018R1C1B6007755.]

### H004

#### **Anti-bacterial Activities Measurement of Radioisotopes Using a New Laboratory Instrument**

Jang Guen Park and Eun-Ha Cho\*

*Korea Atomic Energy Research Institute*

Radioisotopes have a half-life that is specific to each radioisotope and emit radiation of an endemic energy range. Physical energy of the emitted radiation destroys a cell membrane in vivo. These properties of radioisotope are usually used in the cancer treatment research. However, the radiation of radioisotopes also has the effect of killing bacteria. In this study, therefore, we developed a new instrument for evaluating an anti-bacterial activity and measured the radiation level and the anti-bacterial activity of radioisotope I-131 on new laboratory instrument. It is intended to start the evaluation studies of the antibacterial activity specific radioisotopes. Moreover, it is possible to develop the research to overcome the disease caused by bacteria in a future.

**H005****Melanogenesis Inhibitory Effect of Ethanol Extract from *Eleocharis ussuriensis* G. Zinserl**

Yong Tae Jeong, Buyng Su Hwang, Young-Kyung Lee, Hwang Yong, Chul Hwan Kim, Hyeon Ju Nam, Young Taek Oh, Hye-Won Yang, Seok Won Jang, and Pyo Yun Cho\*

*Nakdonggang National Institute of Biological Resources*

The purpose of this study was to investigate melanogenesis inhibitory activity of ethanol extract from *Eleocharis ussuriensis* G. Zinserl. Whole plant, Aerial part, and subterranean part were extracted from *E. ussuriensis* G. Zinserl with 70% ethanol and hot water. All extracts except ethanol extract of subterranean part (EU-SE) did not showed anti-melanogenesis effect. EU-SE were 26.9% inhibited the melanogenesis compared with control at 50 µg/ml in B16F10 cell. EU-SE inhibited melanogenesis of B16F10 cell in a dose-dependent manner. It is noteworthy that the expression of three key proteins, tyrosinase, tyrosinase-related protein-1 (TRP-1), and TRP-2 which is involved in melanogenesis, are significantly decreased by EU-SE. These results suggest that ethanol extract from *E. ussuriensis* G. Zinserl. may be an effective whitening agent.

[This work was supported by a grant from the Nakdonggang National Institute of Biological Resources (NNIBR) funded by the Ministry of Environment (MOE) of Republic of Korea (NNIBR202002101).]

**H006****Glucose Uptake Increased by *Polygonum maackianum* Regel Ethanol Extract in Skeletal Muscle Cells**

Pyo Yun Cho, Buyng Su Hwang, Young-Kyung Lee, Hwang Yong, Chul Hwan Kim, Hyeon Ju Nam, Young Taek Oh, Hye-Won Yang, Seok Won Jang, and Yong Tae Jeong\*

*Nakdonggang National Institute of Biological Resources*

Increase of glucose uptake in L6 myotubes were studied with ethanol extract of *Polygonum maackianum* Regel. Whole plant was extracted from *P. maackianum* Regel with 70% ethanol and hot water. Ethanol extract was only increased glucose uptake compared with control. Ethanol extract stimulated glucose uptake in L6 myotubes in a dose-dependent manner. Maximum stimulation was seen with 100 µg/ml of ethanol extract (140.3 ± 12.8% of control). Organic solvent fractions of n-hexane (PM-H), chloroform (PM-C), ethyl acetate (PM-E), butanol (PM-B) and water layer (PM-W) were obtained from the 70% ethanol extract of *P. maackianum* Regel. PM-H, PM-C, and PM-B was increased the glucose uptake compared with control. Especially, PM-B was showed significantly increased the glucose uptake among the compounds at the concentration of 50 µg/ml (118.1 ± 5.2% of control). PM-B stimulated glucose uptake in L6 myotubes in a dose-dependent manner.

[This work was supported by a grant from the Nakdonggang National Institute of Biological Resources (NNIBR) funded by the Ministry of Environment (MOE) of Republic of Korea (NNIBR202002101).]

## H009

### **Complete Genome Sequence of the Marine Bacterium *Falsirhodobacter* sp. PG104**

Yong Min Kwon and Youngik Yang\*

*National Marine Biodiversity Institute of Korea*

A novel marine bacterium, designated strain PG104, belonging to the family *Rhodobacteraceae* was isolated from the red algae collected in the coastal region of the Pohang, South Korean and its complete genome determined using a hybrid approach combining Oxford Nanopore GridION and Illumina NovaSeq platforms. The genome consists of a one circular chromosome of 2,279,458 bp with 66.68 mol% G + C content and five plasmids, named pL104-1 to pL104-5 with length ranging from 44,911 bp to 175,961 bp. The genome contains 2643 protein-coding genes (CDSs), 51 tRNA genes, as well as 9 rRNA operons as 16S-23S-5S rRNA.

[This work was supported by the MABIK in-house program (2020M00500).]

## H010

### **The Characteristics of Saemangeum Soil Microbial Community with Soil Management Method**

Yang-Yeol Oh<sup>1\*</sup>, Gye-Ryeong Bak<sup>2\*</sup>, Hee-Kyoung Ock<sup>1</sup>, Jin-Hee Ryu<sup>1</sup>, Su-Hwan Lee<sup>1</sup>, Kang-Ho Jung<sup>1</sup>, and Bang-Hun Kang<sup>1</sup>

<sup>1</sup>*National Institute of Crop Science, RDA*, <sup>2</sup>*Highland Agriculture Research Institute, National Institute of Crop Science, RDA*

Soil biodiversity is worsening due to climate change. Saemangeum reclaimed land needs proper soil management because there is only one-tenth organic matter content compared with conventional soil. The objective of this study was to evaluate characteristics of soil microbial community in Saemangeum reclaimed land. The analysis was carried out using the illumine Mseq system for DNA sequencing of the bacterial 16S rRNA genes in soil. Soil management of Saemangeum reclaimed land was found to have significant influences on productivity of maize and bacterial community diversity and composition. Diversity index of Saemangeum reclaimed land showed highest community richness in 1 Chemical Fertilizer (CF) treatment. Predominant Class in soils were *Alphaproteobacteria*, *Betaproteobacteria*, *Deltaproteobacteria*. In particular, the compost+Chemical Fertilizer (CFC) treatment tended to increase *Gammaproterobacteria*. The productivity of maize showed significant differences between soil management method. Our results indicated that differences observed in bacterial diversity and composition were related to soil management methods.

**H011****H5 Highly Pathogenic Avian Influenza Virus in Vietnamese Domestic Poultry between 2015 and 2017**

Min-Ji Park<sup>1</sup>, Soo-Jeong Kye<sup>1</sup>, Yu-Na Lee<sup>1</sup>, Yoon-Gi Baek<sup>1</sup>, Mi-Ja Park<sup>1</sup>, Myoung-Heon Lee<sup>1</sup>, Dang Nguyen Tho<sup>2</sup>, Long To Thanh<sup>2</sup>, and Youn-Jeong Lee<sup>1\*</sup>

<sup>1</sup>Avian Influenza Research & Diagnostic Division, Animal and Plant Quarantine Agency, <sup>2</sup>National Center for Veterinary Diagnosis, Department of Animal Health

Highly pathogenic avian influenza (HPAI) has caused substantial economic losses to the poultry industry of Vietnam. To estimate the prevalence of avian influenza virus (AIV) in Vietnam, genetic analysis was conducted on the samples introduced from Vietnam.

A total of 36 samples, collected from 10 provinces in Vietnam between 2015 and 2017, were tested for HPAI. Twenty-six and 10 samples were confirmed for H5N1 and H5N6 HPAI, respectively. Among these samples, 16 samples were chosen further genetic analysis based on the clustering patterns and geographical distribution. Phylogenetic analysis of the AIVs revealed 3 HPAI clades (2.3.2.1C H5N1, 2.3.4.4C and 2.3.4.4D H5N6) were circulating in Vietnam. Clade 2.3.2.1C H5N1 was divided into 4 different genotypes. Genotype VN1 was identified in the southern region whereas VN2 and VN8 were identified in the northern region. Genotype VN5 was predominant in both regions. Clade 2.3.4.4C H5N6, divided into 4 different genotypes, was prevalent in the northern part of Vietnam, which might have reflected the predominant viral change in China from H5N1 to H5N6 starting in 2014. In the southern-central region of Vietnam, clade 2.3.4.4D H5N6 of single genotype was identified during 2015. Given that the route of entry and spread of HPAI in Vietnam remain to be elucidated, enhanced poultry import control and effective surveillance may help curb the emergence of new HPAI viruses.

[Supported by grants from Animal and Plant Quarantine Agency]

**H012****Pathogenicity of Clade 2.3.4.4 B H5N6 Highly Pathogenic Avian Influenza Viruses Isolated during 2017/2018 Winter Season in South Korea**

Min-Ji Park, Soo-Jeong Kye, Yu-Na Lee, Mi-Ja Park, Yoon-Gi Baek, Myoung-Heon Lee, and Youn-Jeong Lee\*  
Avian Influenza Research & Diagnostic Division, Animal and Plant Quarantine Agency

The latest outbreak of the highly pathogenic avian influenza occurred due to the novel H5N6 viruses classified into clade 2.3.4.4 B during 2017/2018 winter season in South Korea. In this study, we performed the *in-vivo* experiment to understand the pathogenic features of 2 genetically distinguished viruses, A/duck/Korea/HD1/2017(H5N6)(HD1) and A/duck/Korea/H214/2018(H5N6)(H214).

In SPF chickens, HD1 and H214 yielded the LD<sub>50</sub> (median lethal dose) of 10<sup>3.4</sup> and 10<sup>4.7</sup> EID<sub>50</sub>/0.1ml, respectively, with the mean death times of 2.2 and 3.1 days for each virus at the titer of 10<sup>6</sup> EID<sub>50</sub>/0.1ml. The transmissibility of both viruses was 67%. Both viruses shed virus from 1 dpi via OP route in the inoculation group and the viral titers were higher in HD1 than H214, recording the highest on 2 and 3 dpi, respectively. In the contact group of HD1, viral shedding started on 3 dpi, in contrast to H214 which shed from 6 dpi. In the experiment conducted at ducks using HD1, no clinical signs were observed. However, HD1 transmitted from infected to naïve ducks by direct contact, showing the prolonged period of viral shedding.

Taken together, both viruses are highly pathogenic in SPF chickens. However, HD1 replicated more rapidly than H214, which might be attributable to the intrinsic characteristics of each virus including LD<sub>50</sub>. Meanwhile, HD1 could play a role in transmission in ducks without exhibiting any clinical symptoms.

[Supported by grants from Animal and Plant Quarantine Agency]

**H016****Diminishing Returns Epistasis during the Experimental Evolution of Two *Escherichia coli* Strains with Different Replication Properties**Kitae Kim<sup>1</sup>, Soon-Kyeong Kwon<sup>1,2</sup>, and Jihyun F. Kim<sup>1\*</sup><sup>1</sup>Department of Systems Biology, Yonsei University, <sup>2</sup>Division of Life Science, Gyeongsang National University

The fitness advantage of mutation depends on the background fitness of the ancestor. The beneficial effects of mutation tend to be lower in the fitter genotype. This phenomenon is called diminishing returns epistasis. In this study, we conducted an experimental evolution to investigate the causes and effects of diminishing returns epistasis throughout the evolutionary process. When two genetically different ancestors were parallelly evolved for similar generations, fitness gain was lower in the ancestor that had higher background fitness. To identify the mechanism of diminishing returns epistasis, we scrutinized the tempo and mode of evolution by phenome, genome, transcriptome analysis at the populational level. We found that the replication rate of the chromosome is not a crucial factor for fitness increase during the evolutionary process. However, the dissimilarity between the ancestral and evolved transcriptome was significantly proportional to populational fitness gain. These findings suggest that mutations selected during the early stages of evolution can dramatically change global transcription patterns, but later stage mutations manifest diminishing returns epistasis because they cannot significantly change the fine-tuned transcription patterns. Our research may provide insight into understanding the populational evolution by uncovering the patterns and mechanisms of diminishing returns epistasis.

**H018****Finding Genetic Factors of the Plant-probiotic Rhizobacterium TRM1 Contributing to Bacterial Wilt Resistance in Tomato**

Boyoung Lee, Hyein Park, Ju Yeon Song, and Jihyun F. Kim\*

Yonsei University

Tomato is one of the most in demand vegetables in the world, and bacterial wilt caused by the soil-borne pathogen *Ralstonia solanacearum* is a devastating lethal disease in solanaceous crops. As reported in 2018, a flavobacterium TRM1 was found to be enriched in the rhizosphere microbiome of the bacterial wilt-resistant tomato variety Hawaii 7996 relative to the susceptible cultivar MoneyMaker and also be able to suppress disease development in the susceptible plant. Although TRM1 was shown to reduce bacterial wilt in tomato and its functions were deduced from the genome information, which genetic factors of this plant probiotic are responsible for disease suppression or how the factors affect the plant host or the wilt pathogen remains elusive. As the first attempt to investigate how TRM1 could endow the disease resistance, we examined if TRM1-10 affects the growth of *R. solanacearum* SL341 by an *in vitro* co-cultivation method. For the next step to figure out the genetic factors of TRM1 involved in reducing the tomato wilt disease, we employed transposon mutagenesis and massive sequencing (Tn-seq) expecting to disclose genes contributing to the molecular interactions between the plant-probiotic microbe and the wilt pathogen and also between the plant probiotic and the tomato plant in the rhizosphere, and the findings will be presented.

[This work is supported by the Basic Science Research Program (2018R1A6A1A03025607), National Research Foundation, Ministry of Education.]

**H019****Establishment of a Gastric Culture Collection and Screening Microbes for Antagonistic Activity against *Fusobacterium nucleatum***

Lae-Guen Jang<sup>1</sup>, Jaekyung Yoon<sup>1</sup>, HyeonGwon Lee<sup>1</sup>, Jongseok Kim<sup>1</sup>, Sungeun Lee<sup>1</sup>, Soon-Kyeong Kwon<sup>2</sup>, Yong Chan Lee<sup>3</sup>, and Jihyun F. Kim<sup>1\*</sup>

<sup>1</sup>Department of Systems Biology and Division of Life Sciences, Yonsei University, <sup>2</sup>Division of Life Science, Gyeongsang National University, <sup>3</sup>Department of Internal Medicine and Institute of Gastroenterology, Severance Hospital, Yonsei University College of Medicine

*Fusobacterium nucleatum* is regarded as an opportunistic pathogen since it has been frequently found from clinical specimen in a variety of diseases, including colorectal cancer and stillbirth. In a recent study, gastric microbiota of patients with gastric cancer in Taiwan has shown that *F. nucleatum* exhibited a gastric cancer-specific bacterial signature. Therefore, elimination efforts of this bacterium in a digestive tract may be prerequisite to prevent oral and extraoral diseases. In general, *F. nucleatum* isolated from clinical samples is most sensitive to a number of antibiotics. However, owing to concerns about antibiotic resistance and dysbiosis, microbial-interaction-mediated eradication may be more promising. In this study, a bacterial culture collection from stomach biopsies has been established to find bacteria having antagonistic effects against *F. nucleatum*, and over 900 isolates have been successfully recovered followed by taxonomic identification. Subsequently, a soft-agar overlay test against *F. nucleatum* was been performed by screening out 476 isolates. Antagonistic effect was observed from *Streptococcus* isolates, and further antagonistic tests confirmed that *F. nucleatum* inhibition effect is obvious when only grown on solid or semi-solid medium. In conclusion, we identified potent bacteria for the removal of *F. nucleatum* from human gastric tissue, and further identification of molecules associated with antagonist effect against *F. nucleatum* is to be pursued.

**H020****Metacaspase Genes Responses in the Harmful Dinoflagellates *Cochlodinium polykrikoides* and *Prorocentrum minimum* during Cell Death**

Hui Wang, Hansol Kim, Sofia Abassi, Hye Jeong Choi, Bui Thi Nhu Quynh, and Jang-Seu Ki\*

Department of Life Science, Sangmyung University

Metacaspases (MCAs) are cysteine proteases that share sequence homology with caspases, and may play roles in programmed cell death (PCD). In the present study, we identified the novel MCA genes (designated as *CpMCA* and *PmMCA*) from the red tide-causing dinoflagellates *Cochlodinium polykrikoides* and *Prorocentrum minimum*, and examined their molecular characteristics and gene expression in response to algicide-induced cell death. Both MCA genes contained the dinoflagellate spliced leader sequence (dinoSL) and a poly (A) tail. Putative *CpMCA* (293 aa, 32.4 kDa) and *PmMCA* (288 aa, 32.7 kDa) proteins had conserved MCA family motifs, and genomic comparison revealed that no intron present in *CpMCA*, but two introns were found in *PmMCA*. Phylogenetic analysis showed that *C. polykrikoides* and *P. minimum* may have acquired the MCA gene from bacteria by means of horizontal gene transfer (HGT). In addition, expressions of *CpMCA* significantly increased following exposure to the common algicides copper sulfate and oxidizing chlorine, which trigger cell death in dinoflagellates, suggesting that *CpMCA* may be involved in cell death. Further experiments (e.g., TUNEL apoptosis assay) are required to elucidate the exact role of MCA genes in the activation, regulation, and execution of PCD in response to environmental stressors and nutrient limitation.

**H021****16S rRNA Metagenomics Reveals Specific Bacteria Association in the Growth Stages of Harmful Dinoflagellates**

Hansol Kim, Yoseph Seo, Taehee Kim, Yeon Su Lee, Buhari Lawan Muhammad, Hyunjun Park, and Jang-Seu Ki\*

*Department of Life Science, Sangmyung University*

Dinoflagellates are unicellular protists which exhibit a great diversity of form. Most are marine plankton, but they also are common in freshwater habitats. In addition, they are the most important primary producer in aquatic environments. However, some species can form harmful algal blooms (HABs; referred to red tide), and even contain biotoxins that affect humans and many other organisms. Recent studies have shown that HAB-forming dinoflagellates may grow in association with co-occurring bacteria as ectosymbiotic, endosymbiotic, and/or free-living forms. Even, many genes coded in the dinoflagellate nuclear genomes were acquired from symbiotic bacteria via horizontal gene transfer (HGT) and/or lateral gene transfer (HGT). In the present study, we examined the bacterial community structures of both free-living bacteria (FLB) and particle-associated bacteria (PAB) from different growth stages of HAB-forming dinoflagellates using pyrosequencing. These metagenomics showed that Roseobacter clades are intimately associated with host dinoflagellate in culture stages and field samples, suggesting they may strongly influence on the host cell growth and HABs in environments.

**H023****Whole Genome Sequencing of MERS-CoV and Related Virus Using Universal Primers**Kwanwoo Kim<sup>1</sup>, Seil Kim<sup>2,3</sup>, and Hana Yi<sup>1,4\*</sup>*<sup>1</sup>Department of Public Health Sciences, Graduate School, Korea University, <sup>2</sup>Korea Research Institute of Standards and Science, <sup>3</sup>Center for Convergent Research of Emerging Virus Infection, <sup>4</sup>School of Biosystems and Biomedical Science, Korea University*

Full genome sequencing is essential to verify any variations of viral pathogens. Due to the low viral titers in clinical samples, universal primers are needed that allows amplification of a tiny amount of viral nucleic acid to be deep-sequenced. In this study, we aimed to produce a universal primer set for the genomic amplification of MERS-CoV. As a result, 13 pairs of primers were designed to produce amplicons (length of 2.5–3 kb) stretched over the entire genome and the experimental procedure was optimized. The selected primer set was applied to MERS-CoV and NeoCoV and produced amplicons with expected size. Further experiments with animal samples were also completed using the same methods. The mixture of amplicons was successfully sequenced using MinION, Miseq, Pacbio RSII, and Sanger sequencing. The sequencing result demonstrated that primer sets developed in this study have universality and specificity to MERS viruses.

[This research was supported by a grant of the Korea Health Technology R&D Project through the Korea Health Industry Development Institute (KHIDI), funded by the Ministry of Health & Welfare, Republic of Korea (grant number : HI20C0558).]