



Japan Society for Bioscience,
Biotechnology and Agrochemistry (JSBBA)

Program of the

JSBBA West

2nd Student

Forum

November 30th, 2019

The Day of Issue : November 25th, 2019

Fukuoka City Science Museum
Science Hall

Organizer:

JSBBA WEST Student Committee

Sponsors:

JSBBA WEST

Kyushu University

Fukuoka City Science Museum

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Preface

This second student forum could only be held with the cooperation of everyone. We would like to sincerely thank everyone who participated in this forum, especially all the professors and staff of Kyushu University who supported the preparation of this forum. This organization was established as a subordinate organization of the West Japan Branch of the *Japan Society for Bioscience, Biotechnology and Agrochemistry* for the purpose of international exchange among students. We hope that your experience here will prove useful in the future as demand for globalization continue to rise. While this event might be considered a success, we recognize that there is much room for improvement. We apologize for any inconveniences we may have caused or failed to foresee. We hope to be able to address these problems and organize a better conference next year. We look forward to seeing you again in the next forum.

Executive Chairperson Akira BABA

Vice-Chairperson HeeYung WOO

Kota WATANABE

ご案内

(1) 参加者のみなさまへ

■会場・受付

福岡市科学館 6階 サイエンスホール

受付：6階 ホワイエ

(会場には専用駐車場はございませんので公共交通機関をご利用ください。)

■受付時間

11月30日(土) 9:45~12:30

■参加登録手続き

(事前登録された方へ)

- 受付にて、ネームカードをお受け取り下さい。
- 懇親会ご参加の方は受付にて会費 1000 円をお支払いください。
(すでにお支払い済みの方は必要ありません。)

(当日参加される方)

- 受付にて当日参加登録後、ネームカードをお受け取り下さい。
- 懇親会ご参加の方は受付にて会費 1000 円をお支払いください。

(参加者の皆様へお願い)

- 会場では、ご所属とご氏名を記入したネームカードの着用をお願いします。
- ネームカードホルダーは、お帰りの際、受付にご返却をお願いします。

■懇親会のご案内

日時： 11月30日(土) 17:30~18:30

場所：福岡市科学館 6階 サイエンスホール

会費：1,000 円

※会場の都合上アルコールなしでの懇親会となります。予めご了承ください。

■一般的な注意事項

- 携帯電話はマナーモードにしてください、会場内での通話をご遠慮下さい。
- 服装はインフォーマルなもの(ノーネクタイなど)をお願いします。
- クロークは設けておりません。予めご了承ください。

(2) 発表者の皆様へ

- 発表は全て英語で行ってまいります。
- 口頭発表者および一部ポスター発表者を対象とした**優秀発表賞**を選考します。
(表彰式は懇親会で行います。)

■口演発表の方法・受付

- スライドは標準サイズ(4:3)で準備してください。
- 発表時間は10分、質疑は3分で行います。
- **11月28日(木)**までに PowerPoint ファイルを jsbbawssf2018@gmail.com
(担当:馬場)までお送りください。
- PCは事務局で用意します。
- 念のため、USBメモリー等でデータをお持ちください。

■ポスター発表の方法・受付

- ポスター発表に先立ち、ポスター内容を口頭で簡潔に説明するショートプレゼンテーションを行ってまいります。プレゼンテーションは**1分以内**、スライドは標準サイズ(4:3)、2~3枚程度で準備してください。**11月28日(木)**までに PowerPoint ファイルを jsbbawssf2018@gmail.com (担当:馬場)までお送りください。
- ポスターボードのサイズは縦180cm×横120cmです。
- ポスターの貼付は、11:00~12:30の休憩時間にお済ませください。
貼付番号は要旨集に記載しています。
- ポスター発表の示説時間は60分間です。
- ポスターは、閉会式後に必ず撤去して下さい。
(未回収分は事務局で処分します。)
- 優秀発表賞にエントリーしている学生は審査員の先生に5分以内で概要を説明してください。

Guidance

(1) To all participants

■ Location and Reception

Fukuoka City Science Museum 6th floor Science Hall

Reception: 6th Floor

(There is no private parking so please use public transportation.)

■ Reception hours

November 30 (Sat.) 9: 45 ~ 12: 30

■ Registration Procedure

(To preregistered people)

- Please pick up your name card at the reception desk.
- If you are planning to join the award ceremony reception, please pay 1000 yen at the desk.

(Those who have already paid are not necessary.)

(To people who have **not yet** pre-registered)

- At the reception desk, please register and pick up your name card.
- If you wish to join the award ceremony reception, please pay 1000 yen at the desk

(Requests to all participants)

- At the venue, please wear your name card with your affiliation and name.
- After the symposium, please return the name card holder to the reception desk.

■ Information on the party

Date and time: November 30 (Sat.), 17: 30-18: 30

Location: Fukuoka City Science Museum 6th Floor Science Hall

Fee: 1,000 yen

※ Alcohol will not be provided

■ Other precautions

- Mobile phone should be in silent mode, please refrain from calling in the venue.
- Dress code: semi-formal (tie is not necessary)
- No cloak service is provided.

(2) To all presenters

- All presentations will be made in English.
- The Outstanding Presentation Award recipients will be selected.

(The awarding ceremony will be held during the award ceremony reception.)

■ Submission method of oral presentation

- Please prepare slides in standard size (4: 3).
- Total presentation time is 13 minutes :10 min for the actual presentation and 3 min for Q&A
- Please send the PowerPoint file to jsbbawfsf2018@gmail.com (responsible: Baba) by **November 28 (Thu.)**.
- The PC will be prepared by secretariat.
- Please bring your data in USB memory as a back-up just in case of emergency.

■ Method and acceptance of poster presentation

- A short presentation within 1 minute will be allotted to each poster presenter before the poster session to explain about the poster. Please prepare 2~3 slides in standard size (4: 3) and send the PowerPoint file to jsbbawfsf2018@gmail.com (responsible: Baba) by **November 28 (Thu.)**.
- Size of poster board: 180 cm length and 120 cm width.
- Please put up your posters from 11: 30 to 12: 30 (lunch break). Then, confirm your poster board number which is noted on the conference website.
- The poster presentation time is 60 minutes.
- Please be sure to take your poster down after the closing ceremony.
(Any uncollected posters will be disposed by the secretariat.)
- For presenters who have applied for the award of excellence in poster presentation, please explain your poster within 5 minutes to the judge evaluating your presentation.

プログラム **Program**

9:45 ~ 受付開始 **Reception starts**

10:00 ~ 開会挨拶 **Opening speech**

10:15 ~ 11:00 特別講演 **Keynote speech**

「グローバルコミュニケーションの第一歩 一言語、文化、宗教を超えて」

“The first step to global communications: going beyond language, culture and religions”

糀 広大 先生

(株式会社シンカクシヨンリサーチ代表取締役

九州 SDGs アクションサミット実行委員会代表)

昼休憩・ポスター貼付 **Lunch break & poster preparation**

12:30 ~ 13:30 口頭発表 **Oral session (O-01 ~ 04)**

O-01 12:30 ~ 12:45

Studies on new control strategy for meta-lactic acid fermentation driving with mixed culture system

○SIYUAN Y, TASHIRO Y, SAKAI K

(Grad. Sch. of Biosci. Biotechnol. Sci., Kyushu Univ.)

O-02 12:45 ~ 13:00

Bdellovibrio bacteriovorus can be inhibited at less than pH 6.6

○MORIYA T, YOSHIMURA J, HOSHIKO Y, MAEDA T

(Kyutech・Department of Biological Functions Engineering)

O-03 13:00 ~ 13:15

Comparison of the gut microbiota of Mongolian with five Asian countries

○SHINODA A¹⁾, DEMBEREL S²⁾, MISHIMA R¹⁾, NAKAYAMA J¹⁾

(¹⁾ Department of Bioscience and Biotechnology, Faculty of Agriculture, Kyushu Univ.

²⁾ Mongolian University of life Sciences)

O-04 13:15 ~ 13:30

Production of γ -aminobutyric acid from waste biomass using *Halomonas* as cell factory

○ZOU Z, NAKAYAMA H

(Fisheries & Environmental Sciences, Grad. Sch., Nagasaki Univ.)

小休憩 **Short break**

13:45 ~ 14:15 ショートプレゼンテーション (ポスター奇数番号)
Short presentation (Poster of odd number)
14:15 ~ 14:45 ショートプレゼンテーション (ポスター偶数番号)
Short presentation (Poster of even number)

小休憩 Short break

15:00 ~ 16:00 ポスター発表 (奇数番号) Poster session (Odd number)
16:00 ~ 17:00 ポスター発表 (偶数番号) Poster session (Even number)
17:00 ~ 閉会挨拶 Closing speech
17:30 ~ 18:30 懇親会・表彰式 Award ceremony reception

特別講演 Keynote speech

糀 広大 先生

株式会社シンカクションリサーチ代表取締役

九州 SDGs アクションサミット実行委員会代表

「グローバルコミュニケーションの第一歩 一言語、文化、宗教を超えて」

“The first step to global communications: going beyond language, culture and religions”

アジア・日本の玄関口である福岡では、学校や職場・地域において外国人と接する機会が増えてきています。

でも、いざ外国人と接する時に、「言葉ができないし・・・」と躊躇してしまうことが多くありませんか？

うまくコミュニケーションがとれないのは、「言葉が通じない」が原因なのでしょうか？

もっと別の理由ではないでしょうか。

青年海外協力隊員として海外で実際に活動をしてきた後、JICA 九州の国際協力推進員として福岡の国際理解進展のため小学校から大学、各種団体を対象に異文化理解や、国際協力をテーマにした講演やワークショップ活動し、現在も同様の活動を続ける糀広大（こうじ こうだい）氏をむかえ、言語、非言語双方のコミュニケーションの手法を一緒に考えてみましょう。

福岡市科学館 HP より

<https://www.fukuokacity-kagakukan.jp/activity/2019/11/1130.html>

O-01	<p>Studies on new control strategy for meta-lactic acid fermentation driving with mixed culture system</p> <p style="text-align: right;">○SIYUAN Y, TASHIRO Y, SAKAI K (Grad. Sch. of Biosci. Biotechnol. Sci., Kyushu Univ.)</p>
<p>【Introduction】 The meta-fermentation is a method driving with complexed microbial system to produce valuable chemicals. We previously succeeded in producing L-lactic acid with 100% optical purity from model kitchen refuse (MKR), simply using the combination of <i>Bacillus coagulans</i> MN-07, <i>B. thermoamylovorans</i> MN55-6, and <i>B. hisashii</i> N-11. In this case, higher concentration of lactic acid was accumulated than that using the original MAR-compost. The purpose of this study is to improve meta-fermentation performance further by controlling pH.</p> <p>【Methods and Results】 MKR after homogenization and sterilization was inoculated of the 3 strains mentioned above. The meta-fermentation was performed at pH 6, pH 6.5, pH 7, pH 7.5, and pH 8 by constantly control for 72 hours under anaerobic condition. At 72h, the concentration of lactic acid was 22.2 g/L, 24.5 g/L, 24.0 g/L, 19.9 g/L and 14.5 g/L, respectively. The selectivity of lactic acid was 95.7%, 95.0%, 84.3%, 88.7% and 89.0%, respectively. Overall, the meta-fermentation was optimized by constant control at pH 6.5.</p>	

O-02	<p><i>Bdellovibrio bacteriovorus</i> can be inhibited at less than pH 6.6</p> <p style="text-align: right;">○MORIYA T, YOSHIMURA J, HOSHIKO Y, MAEDA T (Department of Biological Functions Engineering, Kyutech)</p>
<p>【Introduction】 <i>Bdellovibrio bacteriovorus</i> is a Gram-negative predator bacterium which is known to exist in various environmental samples. In recent years, anti-microbial resistance ineffective against antibiotics has currently been recognized as a serious issue all over the world. To solve the issue, <i>B. bacteriovorus</i> is worthy of attention because it has a unique life cycle by which prey cells will be killed; therefore, it may be used as a new-type antibiotic. However, the nature of <i>B. bacteriovorus</i> is still unknown, and no research has been conducted focusing on changes in predatory effects under pH conditions, hence we investigated the effect of pH on the predation of <i>B. bacteriovorus</i> 109J.</p> <p>【Methods and Results】 Co-cultivation of <i>B. bacteriovorus</i> 109J and <i>E. coli</i> BW25113 as a prey in HEPES buffer solution from pH 6.6 to pH 7.6 for 10 days showed a decrease in turbidity under the pH conditions higher than pH 6.7; however, no predatory activity was found at pH 6.6. Plaque forming unit of <i>B. bacteriovorus</i> was compared at 5 pH conditions by using a two-layer plating method. As a result, a low number of <i>B. bacteriovorus</i> cells were detected at lower pH conditions. Furthermore, the assay of metabolic activity showed that <i>B. bacteriovorus</i> 109J has a low metabolic activity at lower pH conditions. In conclusion, the predation by <i>B. bacteriovorus</i> is active at an alkaline condition rather than an acidic condition.</p>	

O-03	Comparison of the gut microbiota of Mongolian with five Asian countries ○SHINODA A ¹⁾ , DEMBEREL S ²⁾ , MISHIMA R ¹⁾ , NAKAYAMA J ¹⁾ (¹⁾ Department of Bioscience and Biotechnology, Faculty of Agriculture, Kyushu Univ. ²⁾ Mongolian University of life Sciences)
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【Introduction】 Metabolic diseases, e.g., obesity and type-2 diabetes (T2D), are seriously increasing in Asian countries under the influence of dietary westernization. Among the Asian countries, Mongolian has higher BMI with unique dietary habit with high consumption of meat and dairy product and less vegetable. To address the notion on how their gut microbiota associates with their metabolic mellitus, we analyzed fecal microbiota of healthy (n=114), obese (n=49), T2D (n=47), and obese + T2D (n=40) subjects, in comparison with 317 healthy subjects from Japan, China, Indonesia, Korea, and Thailand.	
【Methods and Results】 To investigate the bacterial composition of Mongolian and five Asian countries, we collected stool samples from 567 participants aged from 24 to 90 years old. The V3-V4 region of the 16S rRNA gene was amplified from stool samples and subjected to the amplicon sequencing. A principal coordinate analysis using the weighted Unifrac distances among samples showed two enterotype-like clusters, each driven by genus <i>Prevotella</i> and other genera, respectively. In contrast to Japanese samples, Mongolian samples, with or without metabolic mellitus, were mostly included in <i>Prevotella</i> -type cluster, suggesting robustness of Mongolian gut microbial community, built up under the unique diets.	

O-04	Production of γ-aminobutyric acid from waste biomass using <i>Halomonas</i> as cell factory ○ZOU Z, NAKAYAMA H (Fisheries & Environmental Sciences, Grad. Sch., Nagasaki Univ.)
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【Introduction】 The moderate halophile, <i>Halomonas elongata</i> , is known to biosynthesize ectoine as major compatible solute to adapt high-salinity environment and assimilate a variety of sugars and amino acids derived from biomass as a carbon and nitrogen sources. Thus, <i>H. elongata</i> could be a practical cell factory for bioproduction of fine chemicals such as ectoine. In this study, we aim to develop a novel cell factory, which can produce GABA using salinity waste-biomass as a feedstock.	
【Methods and Results】 A recombinant <i>H. elongata</i> GOP-Gad strain was generated by introducing a codon-optimized glutamate decarboxylase gene (<i>HeGadBmut</i>) into <i>H. elongata</i> GOP strain's genome. Crude glycerin provided as a by-product of BDF produced in Unzen City, Nagasaki, was added as a sole carbon source to the modified M63 minimal liquid medium containing (NH ₄) ₂ SO ₄ or NaNO ₃ as a sole nitrogen source. The <i>H. elongata</i> GOP-Gad strain was cultured in the modified M63 liquid media under high-salinity conditions corresponding to 3-6% NaCl, then the production of GABA was confirmed by HPLC analysis.	

P-01	<p>Elucidation of the metabolic mechanism of D-BCAA produced by <i>Lactobacillus otakiensis</i></p> <p style="text-align: right;">○KOJIMA K, GODA A, MAEDA J, FUJINO Y, DOI K (Bioscience and Biotechnology, Kyushu Univ.)</p>
<p>【Introduction】 <i>Lactobacillus otakiensis</i> is a D-branched chain amino acid (D-BCAA, such as D-Val, D-Leu, and D-allo-Ile) producer and possesses a novel BCAA metabolizing gene; BCAA racemase gene (<i>bcaa-R</i>). Marked accumulation of D-BCAAs was observed only in <i>L. otakiensis</i>, however, the details of the production mechanism of D-BCAAs remain unclear. To clarify the role of D-BCAAs, localization and content of D-BCAAs were measured. Gene expression levels of D-BCAA related genes were also analyzed by DNA microarray and qRT-PCR along with the different stage of cell growth.</p> <p>【Methods and Results】 In several stages of cell growth, cell components were extracted and fractionated. The amount of D-BCAAs was determined by UPLC. Whereas D-BCAAs contents in extracellular fraction were increased in parallel with the cell growth, the contents in intercellular fraction decreased at stationary phase. Moreover, the transcriptional levels of <i>bcaa-R</i> and putative amino acid transporter (<i>ctrA</i>) increased at stationary phase. The increase of D-BCAAs in peptidoglycan was also observed at stationary phase, indicating that produced D-BCAAs were transferred into cell walls. <i>L. otakiensis</i> might utilize D-BCAAs for their morphological robustness.</p>	

P-02	<p>Functional analysis of Receptor Binding Protein of <i>Lactococcus</i> phage Q1 isolated from lactic acid fermentation plant</p> <p style="text-align: right;">○YOSHIDA N , YAMASAKO A , FUJINO Y , DOI K (Faculty of Agriculture, Kyushu Univ.)</p>
<p>【Introduction】</p> <p>Bacteriophage contamination sometimes causes severe damage to dairy industry. Previously, we have isolated a new bacteriophage named ϕQ1, which infects <i>Lactococcus lactis</i> subsp. <i>cremoris</i> Q1, an important industrial strain being used in yogurt fermentation. ϕQ1 belongs to <i>Siphoviridae</i> according to its morphology and shows narrow host range. To elucidate the infection mechanism of ϕQ1, whole genome sequence of this bacteriophage was determined and putative genes that are involved in host recognition were investigated.</p> <p>【Methods and Results】</p> <p>The phage particles were collected by ultracentrifugation before phage DNA was extracted by phenol/chloroform based method. Phage genome was then analyzed by PacBio RS II and putative open reading frames were estimated by MiGAP (Microbial Genome Annotation Pipeline). ϕQ1 genome contained 86 ORFs and 1 putative receptor-binding protein (Q1-RBP) was found. Q1-RBP showed high similarity with RBP of ϕAM7 and ϕP087 (99% and 95%, respectively). To clarify the function of the protein, Q1-RBP was recombinantly expressed as fusion protein with AcGFP in <i>E.coli</i>. Purified proteins were subjected to binding assay against several lactic acid bacteria.</p>	

P-03	<p>Elucidation of control factors for selective production of L-lactic acid by meta-fermentation with complex microbial systems</p> <p style="text-align: center;">○SEKIYA K , ISHIDA Y , KIMURA T , TASHIRO Y , SAKAI K (Grad. Sch. Biosci. Biotechnol. Sci., Kyushu Univ.)</p>
<p>【 Introduction 】 We defined a new concept, meta-fermentation as the fermentation production process of various useful organic acids and alcohols by using complex microbial system. Although the meta-fermentation shows several merits when compared with pure-sterilized fermentation process, factors that affect meta-fermentation are still unclear. Therefore, in this study, we focused on inoculum, temperature, and culture medium as meta-fermentation factors, and investigated the dynamics of meta-fermentation by analyzing the bacterial flora and measuring fermentation products.</p> <p>【 Methods and Results 】 As a fermentation factor for meta-fermentation, the temperature (30, 37, 40, 45, 50, 55, 60, 65°C) and inoculum (digested liquid fertilizer, compost A, compost B, compost C) were changed. In each batch fermentation transition of the bacterial flora and fermentation products were analyzed. As a result, the fermentation product and the bacterial flora were highly simplified at 50°C, regardless of the inoculum. Based on this result, batch fermentation was performed at 50°C in order to examine the influence of the medium (semi-synthetic medium and garbage medium) as fermentation factors. The most dominant bacteria were the same in the both culture medium, and no difference was found in the main fermentation product.</p>	

P-04	<p>Isolation of uncultured thermophilic bacteria from volcanic ash using fluorescent micromanipulator</p> <p style="text-align: center;">○OKIMURA M, FUJIMOTO R, OKUGAWA Y, TASHIRO Y, SAKAI K (Grad. Sch. Biosci. Biotechnol. Sci., Kyushu Univ.)</p>
<p>【 Introduction 】 Our previous study suggested that predominant extreme thermophilic bacteria in several volcanic ashes would be unculturable and difficult to isolate with the conventional technique. This study aims to establish a new isolation method to obtain unculturable extreme thermophilic bacteria from volcanic ashes of Mt. Sinabung, Mt. Mayon and Mt. Sakurajima, after optimizing enrichment condition for extremely thermophilic bacteria belong to genus <i>Calditerricola</i>.</p> <p>【 Methods and Results 】 Volcanic ash samples or the type culture strain <i>Calditerricola satsumensis</i> YMO81^T were enriched with CYS medium at 75°C, and the culture volume (200 mL) and shaking speed (150 rpm) were optimized by monitoring turbidity. The main bacteria enriched in all ash samples were assigned to <i>Calditerricola</i> spp. (Homology 99% or higher) by direct sequence analysis of the cultures. These suggested that the single bacterial species were enriched with high purity. Isolation trials not only by fluorescent micromanipulator but also by conventional solid gellan plating failed to obtain pure viable cells. On the other hand when while gellan plating method was done under high temperature all samples form colonies successfully.</p>	

P-05	<p>Elucidation of cold-tolerance mechanism of <i>Calditerricola</i> spp. isolated from hyperthermal compost and digested sludge</p> <p style="text-align: right;">○OJIMA M, MAEDA K, TASHIRO Y, SAKAI K (Grad. Sch. Biosci. Biotechnol. Sci., Kyushu Univ.)</p>
<p>【Introduction】 In Kagoshima city, sewage treatment using activated sludge method is being carried out and the residual sludge is further converted via hyperthermal compost. So far <i>Calditerricola satsumensis</i> YMO81^T and <i>C. yamamurae</i> YMO722^T (compost isolates) were isolated from hyperthermal compost. In addition, we have isolated <i>C. satsumensis</i> DD2 and D3 (sludge isolates) from digested sludge (20°C). Previous studies suggested that these isolates showed different cold responses, which contributed to their survival of each ecological niches. In this study, we examined the survival rate and fatty acid composition of each strain at low and high temperature.</p> <p>【Methods and Results】 After pre-cultured at 75°C using CYS liquid medium, each strain was further incubated at 20°C, and then change in their CFU were monitored to determine the survival rate. Fatty acid analysis using GC was performed after extraction and methylation membrane lipid. By measuring the survival rate at 20°C, the compost isolates quickly died in 1 day, while the sludge isolates survived for a long time (>230 days). Therefore, it is considered that only sludge isolates acquired cold tolerance. Membrane fatty acid analysis showed that their composition of sludge isolates changed by incubation at low and high temperatures. These results suggest that gene regulatory mechanism may be involved in change in their fatty acid composition of cell membrane, resulting cold tolerance.</p>	

P-06	<p>Dynamics of physicochemical and bacterial characteristics in the autothermal thermophilic aerobic digestion process equipped with two types of aeration systems</p> <p style="text-align: right;">○MIN Z, TASHIRO Y, SAKAI K (Grad. Sch. Biosci. Biotechnol. Sci., Kyushu Univ.)</p>
<p>【Introduction】 A unique full-scale autothermal thermophilic aerobic digestion (ATAD) equipped with gas-inducing (GI) agitator has been used to convert human excreta to high nitrogen liquid fertilizer in Chikujo Town, Japan. However, the precise mechanism of nitrogen conservation is still not clear. Thus, the effect of aeration system on physicochemical and bacterial community characteristics during the lab-scale ATAD was investigated. This study might be of special interest for the illustration of nitrogen conservation mechanisms in the unique ATAD process.</p> <p>【Methods and Results】 Two types of agitators were applied in the study: a disk turbine (DT) and a gas-inducing (GI). Two systems were performed under the same conditions. To achieve the same K_{La} value, DT system required much higher air flowrate, suggesting the GI system has higher oxygen transfer rate and could reduce heat loss. Lower dissolved oxygen and higher copies number in the first day of GI system, indicated that it is more beneficial than DT system for bacterial proliferation. GI system had a higher carbon removal and accumulated higher nitrogen with more advantages for fertilizer production. Moreover, the difference of beta-diversity structures and bacterial community structure between these two systems suggested that GI agitator is unique and showing great potential to be applied to ATAD system for liquid fertilizer production.</p>	

P-07	<p>Study on the association of gut microbiota and its metabolites with obesity in Filipino adults</p> <p style="text-align: center;">○WATANABE M¹, SIANOYA AC², ELEGADO FB³, DALMACIO LM², NAKAYAMA J¹,</p> <p style="text-align: center;">(¹Department of Bioscience and Biotechnology, Faculty of Agriculture, Kyushu University, ²Univeristy of Philippines Manila, ³University of Philippines Los Banos)</p>
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【Introduction】 Obesity and obesity-related diseases are now one of the biggest health concerns in the world and the gut microbiota and its metabolites are thought to be relevant to development of these metabolic disorders. In the Philippines, it is also true that the number of adults who are overweight and obesity has been gradually increasing; 23.6 % and 5.1% respectively according to a report released by the Asia Roundtable on Food Innovation for Improved Nutrition. The aim of this study is to investigate the association of gut microbiota and its metabolites with obesity in Filipino adults.

【Methods and Results】

In this study, we collected 30 Filipino fecal samples and classified into healthy (n=11), overweight (n=11), obese (n=8) in accordance with body mass index (BMI). The V3-V4 region of 16S rRNA gene was amplified and subjected to the Illumina MiSeq. Partial least square discriminant analysis (PLS-DA) at genus level showed different distribution of each group, therein obese group was significantly correlated with fecal butyrate level. Spearman rank correlation showed significant negative correlation between BMI and *Roseburia* known as saccharolytic butyrate/propionate producer. These results suggest link of gut microbiota with obesity through the alteration of its metabolic activity.

P-08	<p>Effect of self-inducing aeration and stirring device on the growth of major bacteria found in the ATAD process</p> <p style="text-align: center;">○SAKAMOTO S¹, MISHIMA K², TASHIRO Y¹, SAKAI K¹</p> <p style="text-align: center;">(¹ Grad. Sch. Biosci. Biotechnol. Sci., Kyushu Univ., ²Fukuoka Univ.)</p>
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【Introduction】 In Chikujo town, Fukuoka Prefecture, human excreta have been converting to liquid fertilizer by unique autothermal thermophilic aerobic digestion (ATAD). The unique ATAD adopts a self-inducing aeration and stirring device that performs aeration and agitation simultaneously (self-inducing aeration system). We confirmed stable generation of nanobubbles by the self-inducing system while not by a conventional system. In this study, we compared the growth behavior of the major bacteria (*Acinetobacter towneri*) isolated from the ATAD process in the lab-scale fermenter equipped with the self-inducing and the conventional type.

【Methods and Results】 CASO liquid medium (1 L) was prepared in the lab-scale fermenter equipped with the self-inducing system or the conventional system, inoculated with *A. towneri*, and incubated at 30 °C for 48 hours under similar oxygen supply condition (K_{La} , about 45 h⁻¹). The cell density was measured every 6 hours using the turbidity method and the colony counting method, and the specific growth rate was calculated. The maximum cell density with the conventional system was much higher than that with the self-inducing system. The maximum specific growth rate with the conventional system was two-times higher than that with the self-inducing system.

P-09	<p>Functional analysis of the vacuolar protein sorting gene SPBC1709.03 unique to fission yeast.</p> <p style="text-align: right;">○INAGAWA T, OKUBO K, HIGUCHI Y, TAKEGAWA K (Bioscience and Biotechnology, Kyushu Univ.)</p>
<p>【Introduction】 Genetic selections and genome-wide screens in <i>Saccharomyces cerevisiae</i> have resulted in the identification of a large number of genes required for delivery of proteins to the vacuole. We found that <i>Schizosaccharomyces pombe</i> contains many genes that are homologous to Vps proteins of <i>S. cerevisiae</i>. We recently found that the deletion of a nonessential gene (SPBC1709.03) resulted in severe vacuolar sorting defect in <i>S. pombe</i> cells. In this study, we analyzed the phenotype and function of this gene.</p> <p>【Methods and Results】 The predicted SPBC1709.03 gene product consists of 428 amino acids, and hydropathy analysis indicates that the deduced product has a signal peptide at the N-terminus and one transmembrane domain at the C-terminus. When this gene was deleted, the disruptants showed pleiotropic phenotypes including vacuolar sorting defect, abnormal endocytosis and cold-sensitive growth. Because the budding yeasts do not have homologous genes of SPBC1709.03, the fission yeast may have a unique transport mechanism to the vacuole, in which SPBC1709.03 protein is involved.</p>	

P-10	<p>Identification of bacteriocins produced by newly isolated lactic acid bacteria belonging to <i>Enterococcus</i></p> <p style="text-align: right;">○NORIDOMI K, GONG X, MASUNAGA R, HOSHI Y, SONOMOTO K, ZENDO T (Department of Bioscience and Biotechnology, Faculty of Agriculture, Kyushu Univ.)</p>
<p>【Introduction】 Bacteriocins are ribosomally synthesized antimicrobial peptides produced by bacteria. Among them, circular bacteriocins have high stability caused by circular structure. However, details of the biosynthetic mechanism including circularization have not been unveiled. Previously, three enterococcal isolates were found to show antimicrobial activity and to have the gene encoding a circular bacteriocin, enterocin NKR-5-3B (Ent53B) precursor, but no strain has been reported to produce Ent53B alone. In this study, to elucidate the biosynthetic mechanism of Ent53B, the three isolates were analyzed for bacteriocin production and genes for the bacteriocin biosynthesis.</p> <p>【Methods and Results】 Previously, bacteriocin-producing strains, <i>Enterococcus</i> sp. 6674, 69R5 and 7771 isolated from miso or bran bed were found to possess the gene (<i>enkB</i>) encoding the Ent53B precursor. Following Ent53B biosynthetic genes of the isolates were amplified by PCR, and the products were sequenced. As a result, although missense mutations were confirmed, no significant changes in nature of amino acids were observed in the genes except a frame shift mutation found in the <i>enkB2</i> region of 69R5 and 7771 strains. LC/MS analysis and cross-immunity assay proved that all the three isolates produced Ent53B and possessed self-immunity to Ent53B despite the mutations.</p>	

P-11	<p>Analysis of substrate specificity of α-L-arabinofuranosidase candidate enzymes in <i>Aspergillus nidulans</i></p> <p style="text-align: right;">○YAMADA H , MATSUNAGA E , HIGUCHI Y , TAKEGAWA K (Bioscience and Biotechnology, Kyushu Univ.)</p>
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【Introduction】

Cell walls of some filamentous fungi, including *Aspergillus* sp., contain galactofuranose (Galf). Galf has been found in many pathogenic bacteria and filamentous fungi, but not in yeasts, plants, or mammals. β -D-Galactofuranosidase (Galf-ase) hydrolyzes the glycosidic linkage of Galf. We found that Galf-specific Galf-ase genes AN2395 and AN3200 from *A. nidulans*. However, Galf-ase activity was still detected in the culture medium and cells in double-deletion strains, suggesting that Galf-ase was produced in addition to both genes. In this study, we analyzed the possibility that some of α -L-arabinofuranosidases (Araf-ases) might exhibit the Galf-ase activity.

【Methods and Results】

The genome of *A. nidulans* contains nine putative Araf-ase genes belonging to the GH3, 43, 51, 54, 62 family. To investigate the Galf-ase activity, these genes were amplified and cloned into pET vectors. The enzyme activity was measured by the expression and purification in *E. coli*. As a result, all of the genes had weak Galf-ase activity, and Araf-ases from different GH families possess a distinctive Galf-ase activity. Therefore, we demonstrated the presence of unidentified Galf-ase(s) in *A. nidulans*.

P-12	<p>Analysis on the relationship between early endosome dynamics and other organelles in <i>Aspergillus oryzae</i></p> <p style="text-align: right;">○TAKATA A, TAKEGAWA K, HIGUCHI Y (Bioscience and Biotechnology, Kyushu Univ.)</p>
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【Introduction】 Recent years, it has been clarified that the protein secretory ability of *Aspergillus oryzae* is involved not only in exocytosis but also in endocytosis. We focused on an organelle called early endosome (EE) in endocytic pathway, which sorts substances taken up into cells. We identified AoHok1, which functions as a linker protein for the EE and motor protein. In the Δ *Aohok1* cells, it was revealed that with the cessation of EE dynamics, the intracellular distribution of secretory vesicles was abnormal, and the amount of α -amylase secretion was reduced. The purpose of this study was to further clarify the physiological function of EE dynamics in the interaction with each organelle by analyzing the influence of *Aohok1* disruption on other organelles.

【Methods and Results】 In order to observe the intracellular distribution of each organelle, a construct expressed by fusing EGFP with a marker protein specifically localized in each organelle was introduced into control and Δ *Aohok1* strains, and analyzed with a fluorescence microscope. AoErg6, AoFbh2, AoRab7, and AoVam3 were used as marker proteins for lipid droplets (LDs), mitochondria, late endosomes (LEs), and vacuoles, respectively. As a result, although LDs, mitochondria and LEs were not significantly affected, the cessation of EE dynamics resulted in the abnormal localization of vacuoles, suggesting that EE motility maintains vacuolar distribution.

P-13	<p>Maternal protein restriction causes alteration in hippocampal monoamine neurotransmitter system.</p> <p style="text-align: right;">○KABASHIMA N¹, KITANO G¹, FURUYA S^{1,2}. (¹Grad. Sch. Bioenv. Sci, ²Innovative Bio-Arch. Center, Kyushu Univ.)</p>
<p>【Introduction】 Schizophrenia (SCZ) is a chronic and serious psychiatric disorder characterized by combination of hallucinations, delusions, and extremely disordered thinking and behavior, and is thought to be caused by combination of genetic and environmental factors. Epidemiologic studies suggest that maternal starvation is one of such environmental factors. It has been shown that protein restriction during fetal and neonatal periods leads to behavioral abnormalities reminiscent of SCZ in rats and mice. To understand how maternal protein restriction affects brain functions at molecular level, I investigated whether maternal protein restriction affects the monoamine neurotransmitter systems in the adult brain of F1 generation.</p> <p>【Methods and Results】 Pregnant DBA / 2J was divided into two groups: normal diet group (AIN-93G: 20% casein) and restriction diet group (AIN-93G: 10% casein). After weaning, F1 generation mice fed a normal diet and were dissected at 10 weeks of age. Quantification of monoamines demonstrated that serotonin was altered and 5-HIAA/5-HT turnover was increased in the hippocampus of the restriction group. A decrease in Htr1a and increases in Htr2a and 2c were also observed in the restriction group. These results suggest that the maternal protein malnutrition leads to dysregulation of the hippocampal serotonin system, which could be involved in behavioral deficits.</p>	

P-14	<p>Analysis of the mating-type switching mechanism in <i>Ogataea polymorpha</i></p> <p style="text-align: right;">○KAI N, TAKEGAWA K, MAEKAWA H (Bioscience and Biotechnology, Kyushu Univ.)</p>
<p>【Introduction】 The mating type switching mechanism of methanol-utilizing yeast, <i>Ogataea polymorpha</i>, have been investigated recently. The mating type switching is caused by homologous recombination of chromosomes between inverted repeats (IR) adjacent to the two MAT loci proximal to the centromere region, which occurs only under nitrogen starvation conditions. It is not known whether chromosomal position of MATs and/or their surrounding sequences, apart from IRs, are important for the mating type switching.</p> <p>【Methods and Results】 To identify DNA sequence that are important for the mating type switching, we deleted one of IRs from its original position and inserted at a different position within the MAT intervening region. Mating type switching was observed in such strains, suggesting that neither chromosomal position of IRs nor surrounding DNA sequences affect the mating type switching and IR sequence may be necessary and sufficient for a recombination reaction between two IRs. We hypothesize that a DNA lesion in one of IRs under starvation conditions triggers the mating type switching. We will investigate if artificial introduction of DSB in one of IRs can induce the mating type switching by CRISPR/Cas9 system.</p>	

P-15	<p>Involvement of AoCdc48 in useful material productivity in <i>Aspergillus oryzae</i>.</p> <p style="text-align: center;">○MORITA Y, KIKUMASTU F, TAKEGAWA K, HIGUCHI Y (Bioscience and Biotechnology, Kyushu Univ.)</p>
<p>【Introduction】 In budding yeast, Cdc48 has functions of recognizing ubiquitinated misfolding proteins and transporting them to the 20S proteasome in endoplasmic reticulum-associated degradation (ERAD), inner nuclear membrane-associated degradation (INMAD), mitochondria-associated degradation (MAD) and endosome and Golgi-associated degradation (EGAD). In addition, Cdc48 is an essential protein for growth and is a cause of ER stress. However, it is not unclear about the function and involvement in useful material productivity in filamentous fungi; therefore, in this study, we analyzed AoCdc48, an ortholog of Cdc48 in <i>Aspergillus oryzae</i>, to reveal its function.</p> <p>【Methods and Results】 We observed hyphal morphology by microscope in <i>Aocdc48</i>-repressed strain. We found that <i>Aocdc48</i> repression might cause abnormal hyphal form by aberrant location of organelles. In addition, we analyzed phenotype and productivity of useful materials in strains of <i>Aocdc48</i> over expression and point mutation of AAA ATPase domain. We found that overexpression of <i>Aocdc48</i> might not affect productivity of α-amylase and kojic acid, and that AAA ATPase D1 domain might be important for the function of AoCdc48. Currently, we are analyzing degradation of ERAD substrate protein and production of heterologous protein in the involvement of AoCdc48 function.</p>	

P-16	<p>Studies on Sustainable Plant Production System Using PGPB, Biochar and Co-compost from Palm Plantation</p> <p style="text-align: center;">○SALMAN Z, TASHIRO Y, SAKAI K (Bioscience and Biotechnology, Kyushu Univ.)</p>
<p>【Introduction】 In contribution to establish a zero-emission palm oil industry in Malaysia; co-compost was produced with shredded empty fruit bunch and anaerobic sludge from palm oil mill effluent. A hundred strains were isolated from the produced co-compost, screened and grouped into three groups according to their functions: plant growth promoting (PGPB) traits, bio-controlling, and composting. In this study, four PGPB strains, <i>Citrobacter sedlakii</i> CESi7, <i>Citrobacter sedlakii</i> CE9, <i>Enterobacter cloacae</i> B3 and <i>Enterobacter cloacae</i> subsp. <i>dissolvens</i>, were selected. The strains were investigated for their efficiency as PGPB inoculants in screening and cultivation tests.</p> <p>【Methods and Results】 The selected strains were screened in a plate assay for their abilities in nitrogen fixation, and solubilization of phosphate, potassium and silicate. The production of Indole-3-acetic acid (IAA) was quantified using the spectrophotometric assay. The plant growth promotion of the strains as inoculants was evaluated with <i>Brassica rapa</i> (Komatsuna) in a cultivation test. As a result, CESi7 and CE9 had remarkably high potassium solubilizing ability, and moderate phosphate solubilizing ability. CESi7 had the highest IAA production among the other strains. In the cultivation test all inoculated plants had a germination rate between 93%-100%, and plants inoculated with CESi7 had a 100% germination rate.</p>	

P-17	<p>Elucidation of target genes controlled by autoinducing peptide, AIP_{Cb}, of <i>Clostridium butyricum</i> MIYAIRI 588</p> <p style="text-align: right;">○SHIGEOKA E , YOUSUF B , NAKAYAMA J (Bioscience and Biotechnology, Kyushu Univ.)</p>
<p>【Introduction】 Quorum sensing (QS) is microbial cell density-dependent regulatory system which controls the expression of certain target genes. Some gram-positive bacteria secrete autoinducing peptide (AIP) as a QS signal to control virulence. <i>Clostridium butyricum</i> MIYAIRI 588, which is an intestine conditioning probiotics, produces AIP_{Cb}, whose target is yet to be elucidated, notably in terms of its probiotic functionality.</p> <p>【Methods and Results】 A protected linear peptide, Z-CFWAH (100 mg), was chemically synthesized by Fmoc solid phase synthesis strategy and then cyclized to form thioester intramolecularly, thereafter Z-group was deprotected, resulting in 320 µg of AIP_{Cb}. The synthetic AIP_{Cb} was used to investigate its target genes by an RNA-seq experiment, therein MIYAIRI 588 was cultured to early-log and late-log phases in BHI medium with or without 0.3 µM of AIP_{Cb} and RNA was extracted for RNA-seq experiment. The result of RNA-seq analysis suggested that the transcription of phosphotransferase operons for some sugar transportation were induced by AIP_{Cb}, although it is yet to be confirmed by RT-qPCR experiment.</p>	

P-18	<p>Altered neuronal signaling and transmission in the brain-specific <i>Phgdh</i> KO mice.</p> <p style="text-align: right;">○UTSUMI S¹, HAMANO M³, FURUYA S^{1,2} (¹Grad. Sch. Bioenv. Sci., ²Innov. Bio-Arch., Kyushu Univ., ³Kyushu Inst. Tech.)</p>
<p>【Introduction】 L-Serine, a nonessential amino acid, is synthesized <i>de novo</i> from a glycolytic intermediate, 3-phosphoglycerate, through three catalytic steps known as phosphorylated pathway. The first step in this pathway is catalyzed by D-3-phosphoglycerate dehydrogenase (Phgdh). Patients of L-serine deficiency caused by mutations in <i>PHGDH</i> exhibit severe neurodevelopmental symptoms, including microcephaly, psychomotor retardation, and intellectual disabilities. To understand the molecular mechanism underlying these severe neurological symptoms, we generated brain-specific <i>Phgdh</i> knockout mice (BKO) and have investigated alterations in neuronal signaling cascades and structural elements.</p> <p>【Methods and Results】 We analyzed protein expression profiles of signaling components involved in neurotransmission in the brain of BKO. Western blot analysis demonstrated that the protein level of calcium/calmodulin-dependent protein kinase II (CaMKII) was markedly decreased in the cerebral cortex, cerebellum and hippocampus of BKO. Further, myelin basic protein, a structural element of myelin sheath, was significantly decreased in the cerebellum. These observations strongly suggest that L-serine deficiency leads to diminishments in neuronal Ca⁺⁺ signaling and axonal transmission.</p>	

P-19	<p>Elucidation of biosynthetic mechanisms of a circular bacteriocin, leucocyclicin Q</p> <p>○HOSHI Y, SADO S, YOSHIMURA K, IKEDA S, SONOMOTO K, ZENDO T (Department of Bioscience and Biotechnology, Faculty of Agriculture, Kyushu Univ.)</p>
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【Introduction】 Bacteriocins are ribosomally synthesized bioactive peptides that are usually active against related bacterial strains. Among them, circular bacteriocins have unique head-to-tail structure. Their biosynthetic mechanisms are being studied to elucidate how the linear precursor peptides are cyclized and secreted. In addition, self-immunity, a mechanism that protects producer bacteria from their own bacteriocin, has not been elucidated. In this study, biosynthetic mechanisms of leucocyclicinQ (LcyQ), a circular bacteriocin produced by *Leuconostoc mesenteroides* TK41401, are being characterized.

【Methods and Results】 The upstream gene region, *lcyQBCDXI* in the LcyQ biosynthetic gene cluster was found to be essential to produce LcyQ with both secretory and self-immunity mechanisms, while the function of the downstream gene region *lcyEFGH* was not clarified. Heterologous strains expressing *lcyEFGH* alone and with *lcyQBCDXI* were constructed and examined for secretion of and self-immunity to LcyQ. As a result, *lcyQ~H* strain secreted more LcyQ than *lcyQ~I* strain. This indicated that *lcyE~H* had a function of enhancing the secretion of LcyQ. In contrast, *lcyE~H* strain showed no enhanced tolerance to LcyQ. This suggested that *lcyE~H* did not contribute to self-immunity to LcyQ, whereas the upstream region is involved in both secretion and self-immunity.

P-20	<p>Analysis of spindle orientation checkpoint mechanism in methylotrophic yeast <i>Ogataea polymorpha</i></p> <p>○FUKUYAMA N, TAKEGAWA K, MAEKAWA H (Bioscience and Biotechnology, Kyushu Univ.)</p>
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【Introduction】 During the mitotic phase, accurate segregation of replicated chromosomes and cell nuclei is essential, and if there is mis-orientation of the chromosome segregation device called spindle, the nucleus cannot be properly segregated. The budding yeast *Sacchomyces cerevisiae* is known to have a spindle orientation checkpoint (SPOC) that temporarily halts cell cycle progression when cells sense spindle mis-orientation until it is corrected. Considerable detail of the molecular mechanism has been elucidated in *S. cerevisiae* which is the most studied model system in basic research. On the other hand, SPOC has not been investigated in other budding yeast species. In this study, we clarify the SPOC mechanism in *Ogataea polymorpha*.

【Methods and Results】 We identified the homologous genes of the SPOC components in *O. polymorpha* based on amino acid sequences. Identified SPOC components are *BUB2*, *BFA1* and *FRK1*. We examined the phenotypes of these gene mutants at the time of spindle mis-orientation, and obtained a data that suggested the existence of the SPOC. Currently, we are investigating double mutants of the three SPOC genes to determine whether they work in the same signaling pathway.

P-21	Chronological variation of bacterial community structure on human scalp hairs ○YAMADA A, WATANABE K, TASHIRO Y, SAKAI K (Bioscience and Biotechnology, Kyushu Univ.)
<p>【Introduction】</p> <p>Bacterial communities on various part of the human body are distinct. Recently, we found that bacteria are also present on human hair and the main bacteria would originate from hair roots (Watanabe et al. M&E, 2019). However, it is still unclear about their interactions between hair, bacteria. Thus, we first investigate chronological variation of the bacteria and a possibility of nutrients supply from hair.</p> <p>【Methods and Results】</p> <p>Hair was collected from 5 volunteers every 3 months for 1 year. In addition, hairs collected from 2 volunteers were stored at room temperature to make it be non-nutritive and compared their bacterial community structure. Bacterial DNAs were extracted from all the samples and 16S rRNA gene copy number per 1cm hair by qPCR. After that, 16S amplicons sequencing (MiSeq, Illumina) were analyzed. As a result, stable phylum occupancy was shown throughout the year. Long-preserved hair showed a correlation between change in the phylum / genus occupancy and the time of preservation, regardless of nutrient condition.</p>	

P-22	Possible interference of host complement system by quorum peptide of <i>Clostridium perfringens</i> ○HONDA K, NAKAYAMA J (Bioscience and Biotechnology, Kyushu Univ.)
<p>【Introduction】</p> <p>Quorum sensing (QS) is the system to control gene expression using signal molecules which represent cell density of bacteria. <i>Clostridium perfringens</i> secretes autoinducing peptide (AIP_{Cp}) as the QS signal that trigger the expression of toxin genes. AIP_{Cp} contains thiolactone structure, which is known in a complement protein (C3b), which plays a role in innate immunity. The thiolactone structure of C3b are involved in the binding of C3b to bacteria cell surface, which triggers bacteriolysis. Therefore, it is hypothesized that AIP_{Cp} protects <i>C. perfringens</i> from the attack of complement system and is examined in this study.</p> <p>【Methods and Results】</p> <p><i>C. perfringens</i> str. 13 that is a wild strain and str. TS230 that is an AIP_{Cp}-negative strain were cultured in mouse serum. 100 μL of <i>C. perfringens</i> culture was mixed with 300 μL of mouse serum and was then incubated for 1 h at 37°C. Then, the number of viable cells were analyzed by dilution plate culture. As a result, the viable counts of str. TS230 were decreased to 0.016 ± 0.034 %, but that of str. 13 was 11 ± 8.3 % to the control. The difference suggests that AIP_{Cp} contributes to the protection of <i>C. perfringens</i> cell from the attack of complement system, although further studies are required.</p>	

P-23	<p>L-Serine deficiency leads to apoptosis in <i>Phgdh</i>-deficient MEF cells.</p> <p style="text-align: center;">○OSAKI Y¹, MATSUO Y¹, HARAGUCHI Y¹, HAMANO M³, SAYANO T⁴, FURUYA S^{1,2}</p> <p style="text-align: center;">(¹ Kyushu Univ., ²Innov. Bio-Arch., Kyushu Univ., ³Kyushu Inst. Tech., ⁴Keio Univ.)</p>
<p>【Introduction】 Although depletion of L-serine (Ser) causes cell death in mouse embryonic fibroblasts deficient in 3-phosphoglycerate dehydrogenase (Phgdh), its form and cascade executing cell death remain unexplored. We previously showed that Ser deficiency results in caspase 3-dependent apoptosis via p53 and Bax activation in Phgdh-deficient MEF (KO-MEFs) under Ser deprived condition. However, the Ser-restricted conditions that have been used before have raised the possibility of shortage of other nutrients. In this study, we reconstructed a more stringent Ser-deprived condition and elucidated the cell death pathway in KO-MEFs.</p> <p>【Methods and Results】 Western-blot analysis demonstrated that under the novel Ser-deprived condition p53 phosphorylation at Ser15, which is induced by DNA damage, was upregulated in KO-MEFs, which was accompanied by subsequent fragmentation of caspase-3. Quantitative real-time PCR analysis demonstrated that expression of Bax, a pro-apoptotic factor, was remarkably upregulated. Also, we observed that expression of Bcl2, which is an apoptosis inhibitor, was downregulated under Ser-deprived condition for 24 hours. These results suggest that reduced availability of Ser triggers DNA damage, and then induces apoptosis mediated by mitochondria/caspase-dependent pathway.</p>	

P-24	<p>Functional analysis of putative glycosyl transferase Omh6p localized to the nucleus and cytoplasm in fission yeast</p> <p style="text-align: center;">○SHIMOMURA K , MAEKAWA H, TAKEGAWA K (Bioscience and Biotechnology, Kyushu Univ.)</p>
<p>【Introduction】 In order to clarify the specific functions of genes encoding putative α-1,2-mannosyltransferases involved in the elongation step of O-linked sugar chains, we isolated and characterized six <i>Schizosaccharomyces pombe</i> genes <i>omh1-omh6</i> that share significant amino acid similarity to the <i>Saccharomyces cerevisiae</i> KTR family. The Omh1-5 proteins are localized in the Golgi. In contrast, we found that Omh6p is localized to the nucleus and cytoplasm. In order to clarify the specific functions of Omh6p, we isolated and characterized the <i>omh6</i> Δ mutants.</p> <p>【Methods and Results】 The growth of the fission yeast <i>omh6</i> Δ strain was slower than that of the wild strain, and cells with multiple septa were observed only in the <i>omh6</i> Δ strain. The <i>omh6</i> Δ cells had a round cell morphology as compared to the wild type cells. In addition, when the <i>omh6</i> gene was overexpressed, the cells tended to be larger than the wild type cells. These results suggest that Omh6p may have the mannosyltransferase activity in the nucleus and cytoplasm, and affect the cell cycle and cell morphology of fission yeast cells.</p>	

P-25

Production of human DNaseI using the fission yeast expression system.

○IWAI R, MURAI K, MAEKAWA H, HIGUCHI Y, TAKEGAWA K
(Bioscience and Biotechnology, Kyushu Univ.)

【Introduction】

In recent years, glycoprotein production is highly demanded in pharmaceutical industry. There has been a rapid increase in the number and demand for approved biopharmaceuticals produced from animal cell culture processes over the last few years. However, animal cell production systems have problems such as high cost and reduction of its physiological activity with its heterogeneous glycosylation. In this study, we examined the production of human DNaseI in *Schizosaccharomyces pombe*.

【Methods and Results】

The wild type strain of *S. pombe* harboring the plasmid expressing human DNaseI under the control of *hCMV* promoter was grown up to mid-log phase at 30°C. We detected the expression of Strep-tagged DNaseI with its specific antibody by western blotting. Most of DNaseI was not secreted into the culture medium. However, we found that the produced DNaseI was glycosylated by the analysis of endoglycosidase treatment. We are investigating the optimal production of DNaseI in *S. pombe*.



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