



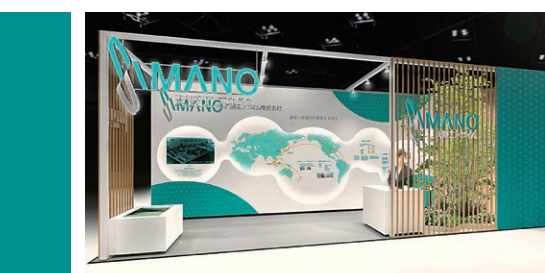
Volume
21

Courtesy of Yoshikazu Suematsu "Karakuri Puppet Float Festivals"

Enzyme Wave

2018





Enzyme Wave vol.21

CONTENTS

Trend	02
Vegans and the vegan market	

Column	03
Manufacturing which Learns from “Karakuri”	

Report	05
Expectations to be met for application of maltotriose transferase to food processing	

Symposium	07
Fourth Sino-Japan Joint Symposium on Enzyme Technology	

Information	08
Exhibition information	

Information	09
The Prof. Jokichi Takamine Research Foundation (NPO)	

Conference presentation	10
-------------------------	-----------

The ultimate vegetarian

Recently, more and more people are choosing and practicing diets from a variety of options that suit their lifestyles and beliefs. Vegetarians are one such choice.

There are many types when you look inside the vegetarians: lacto-vegetarian eat dairy products; ovo-vegetarian eat eggs, a lacto-ovo vegetarian eat both dairies and eggs. Vegans, those who avoid consuming animal-derived products, is a term proposed by six people including Donald Watson in 1944 when vegetarianism campaign was established. It combines the first and last word of “vegetarian”. Their principles were based on “emancipation of animals from exploitation by man” and “maintaining environmental sustainability.” Due to these reasons, many vegans are conscious that such eating habits lead to animal welfare and environmental protections, not just being healthy (1*). Vegans do not even eat any animal-derived products (such as honey, beeswax, and gelatin). Not only that, stricter so-called ethical vegans do not even wear silk, leather, or wool (*2).

Sales of vegan-labeled products are increasing 3% annually in 2015. For US millennials (people born between 1980s and early 2000s), the label of “Vegan” is an important factor when purchasing products. In addition, more than half of US consumers consider veganism healthy (*3).

Protein products are the most important ingredients for vegans. A variety of meat substitutes are being developed using soybeans, wheat gluten, peas, quorn (mycoprotein), and so on. Among these, consumers tend to prefer those meat alternatives made with less additives and more natural ingredients (*4).

Amano Enzyme’s non-animal-derived products will contribute to the development of vegan food market with delicious and accessible products.



A vegan hamburger made with seitan

《Types of vegetarians》

Type	Meat and fish	Dairy products	Eggs
Vegan	×	×	×
Ovo-vegetarian	×	×	○
Lacto-vegetarian	×	○	×
Lacto-ovo vegetarian	×	○	○

*1) The Vegan Society: <https://www.vegansociety.com/>

*2) Vegetarian Nation: <http://www.vegetarian-nation.com/>

*3) Statista: Sales growth of the vegan market between 2015 and 2020 worldwide, by country

*4) Mintel: Germany hosted the highest number of vegan launches worldwide in 2016

Column

Manufacturing which Learns from “Karakuri”

Author profile

Yoshikazu Suematsu

Professor Emeritus at Nagoya University, President of Ninth Generation Tamaya Shobei Appreciation Society
 Visiting Professor at Aichi Institute of Technology’s Research Institute for Industrial Technology



Also recently engaged in scientific research on topics including *Karakuri* puppets and manufacturing in Edo.

Held “*Karakuri* Lectures” (overseas cultural exchange projects) in 20 cities in 12 countries.

Received a Science and Technology Award presented by the Minister of Education (category of promoting understanding) in April 2006.

Manufacturing which Learns from “*Karakuri*”

In Japan, mechanical devices dating back to ancient times are known as “*Karakuri*.” Initiatives aimed at developing *Karakuri* with powered movement (power looms) led to the growth of the automobile industry, the leading industry of Aichi prefecture and the Chubu region. Here, we will discuss the connections between industrial technology and the *Karakuri* puppet float festivals which have been passed down from generation to generation in those regions.

Aichi Prefecture, #1 in Japan for Manufacturing

For 40 years, Aichi prefecture has been the highest-ranking prefecture in Japan for monetary amounts of manufactured product shipments.

Figure 1 shows the top 10 highest-ranking prefectures in Japan for monetary amounts of manufactured product shipments at the end of 2014, with a significant and clearly visible difference between Aichi and other prefectures.

Figure 2 shows the distribution of *Karakuri* puppet

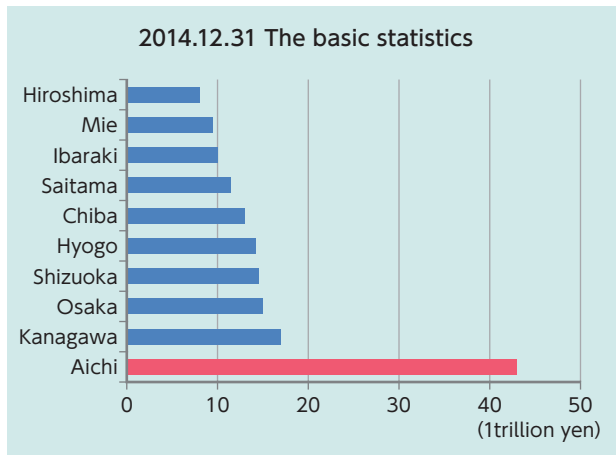


Figure 1 : Shipments of manufactured products in 2014 (top 10 highest-ranking prefectures)

float festivals in Japan. There are nearly 80 locations in Japan where these festivals are held, but 80% of them are concentrated in the Chubu region (Aichi, Gifu, Mie, Shiga). The majority of these are in turn located in Aichi prefecture.

We will now describe how these *Karakuri* puppet float festivals have become a source of great ingenuity for the people of Japan, and how *Karakuri* themselves have led to the birth of Japan’s unique production systems.



Figure 2 : Distribution of areas throughout Japan holding *Karakuri* puppet float festivals

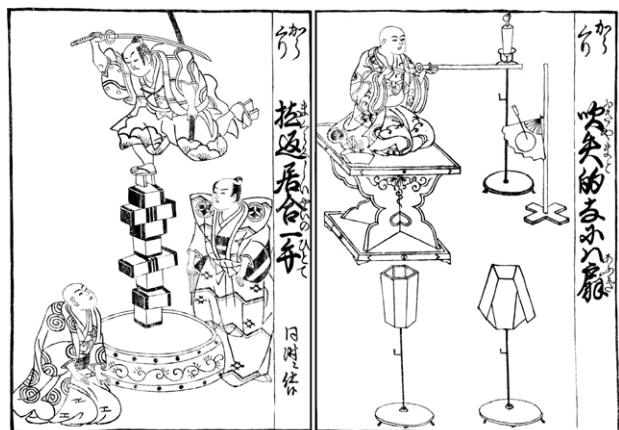


Figure 3 : Programs for performances of the Takeda Karakuri Theatre

Uniquely Japanese Karakuri

The term *Karakuri* began to come into wide use from around the 17th century. In a broad sense, it refers to mechanical devices, mechanisms, clocks and looms, mechanical gimmicks and structures, contrivances, and trick devices, but there is no equivalent word for it in other languages. Japan's unique word "*Karakuri*" created a distinctive culture in Japan, strongly represented by *Karakuri* puppets.

Karakuri Puppets: from Stages to Festival Floats

The amusement and enjoyment of *Karakuri* puppets were deeply ingrained into the minds of the common people by the "Takeda *Karakuri* Theatre" which was performed in 1662, in Osaka prefecture (Figure 3). Showing over 10 *Karakuri* puppet acts daily, it was met with great public acclaim. It was held in various areas for 100 years, presenting the wonder of *Karakuri* puppets to the citizens of Japan. In time, festivals featuring *Karakuri* puppets came to be held in the Owari and Nagoya areas of Aichi prefecture.

In 1619, *Karakuri* puppets were placed on festival floats for the first time in Nagoya as part of the Nagoya Toshogu Festival, and in 1707 they were included on all 9 floats used in the festival. By around 1830, the festival processions numbered 7000 people, and the success of these events gave rise to *Karakuri* puppet float festivals in other areas which spread throughout the Chubu region.

In addition to those used in the acts of Takeda *Karakuri*, many original skills and techniques of manipulating *Karakuri* puppets were also displayed. As shown in Figure 4, such programs included "*Men-kaburi*", in which the faces of



Figure 4 : *Karakuri* techniques specific to Aichi

puppets would change near-instantly, and "*Aya-watari*", in which puppets would leap from stick to stick while in mid-air.

Karakuri Puppet Float Festivals and Manufacturing

Karakuri puppet float festivals, in which floats were lined up before many spectators and puppet performances were presented one after the other, brought effects to local areas and tourists comparable to those brought by auto shows. Furthermore, they made it possible for these annual festivals to be continued for several hundred years.

It was precisely because the Japanese people came to realize the amusement and enjoyment of *Karakuri* puppets, which could also in a sense be considered wooden robots, that they developed a love for robots themselves, leading to the rapid introduction of robots for industrial use. *Karakuri* puppet float festivals became a source of human creativity and ingenuity, and various ideas for improvements. One prominent example of a "*Karakuri* improvement" was an unpowered carrier vehicle invented by a certain automobile parts manufacturer, with hints taken from tea-serving dolls which its employees had seen as children. It is precisely in our modern age where computers have infinite capabilities, that *Karakuri* puppet float festivals, which can foster interest in *Karakuri* puppet mechanisms, are seen as growing in value to society even further.

To learn more about *Karakuri*,
please visit the "*Karakuri* Frontier" website.
<http://karafro.com/>

Expectations to be met for application of maltotriosyl transferase to food processing

Amano Enzyme discovered a unique enzyme potentially applicable to food processing (maltotriosyl transferase) from its in-house microbial strain and turned it into a product known as Glyco Transferase "Amano." The present article describes its characteristics and some examples of its application.

What is Glyco Transferase "Amano"?

Hardening of cooked rice and bread during storage is ascribable to retrogradation of starch, which is a fundamental component of these foods. Accordingly, inhibition of starch retrogradation is a major challenge for the food industry. Glyco Transferase "Amano" acts on starch and catalyzes a "maltotriosyl transferring reaction," i.e., transglycosidation of triose units, as illustrated in Figure 1. This enzymatic reaction presumably converts starch to a glucose

polymer with a highly branched structure resistant to retrogradation. Another feature of Glyco Transferase "Amano" is its excellent heat stability. Since this enzyme is stable at temperatures higher than the gelatinization temperatures of starches from various plant sources, it acts on starch effectively and is expected to be widely applicable to food processing.

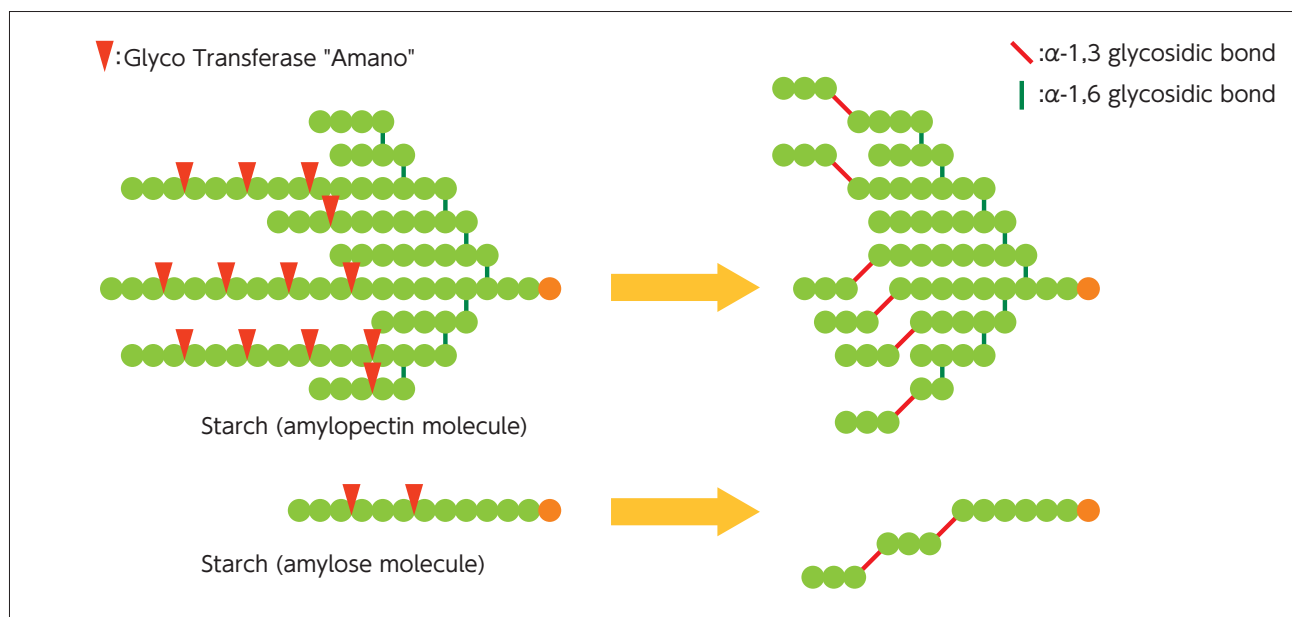


Figure 1. Changes in starch structure caused by Glyco Transferase(deduced scheme)

Labor saving in the manufacture of rice-cake sweets involving enzyme addition

In the industrial process of manufacturing rice-cake sweets and dumplings, soybean β -amylase was conventionally used for inhibition of starch retrogradation and maintenance of soft food texture. Due to the limited thermostability of the soybean β

-amylase used, cooling down of the pre-steamed dough to a temperature $\leq 75^{\circ}\text{C}$ was required prior to enzyme addition and made this process cumbersome. The thermostability of Glyco Transferase "Amano" is superior to that of soybean β -amylase and thus

allows pre-mixing of this enzyme with other raw materials (e.g., rice flour) before starting the steaming process. Figure 2 compares the effects of enzymes added prior to the steaming process on maintenance of the soft texture of rice cakes. In contrast to soybean β -amylase, Glyco Transferase "Amano" added prior to the steaming process successfully maintains the soft texture of the resulting rice cakes during storage.

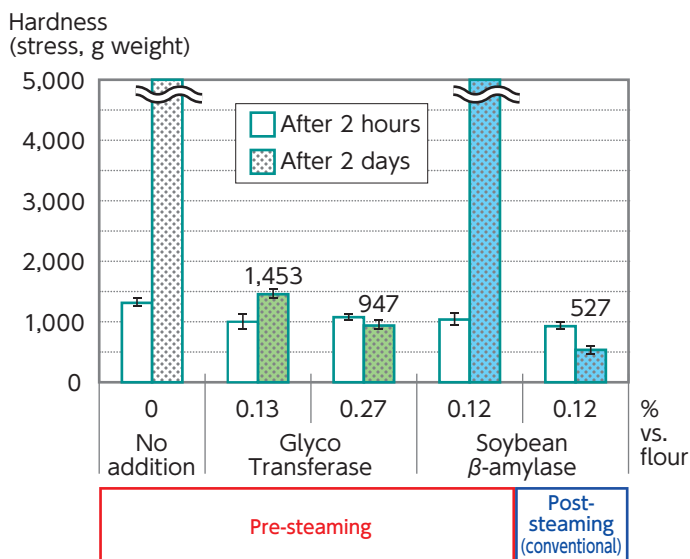


Figure 2. Effect on maintenance of soft texture of rice cake during storage (after storage of the rice cake dough at pH 5.3 and 5°C)

Maintenance of clarity of liquid dextrin

Liquid dextrin is produced by hydrolysis of starch. In recent years, there has been a growing demand for liquid dextrin with a low dextrose equivalent (DE) for use as a material in foods including liquid diets. However, liquid dextrin with the lower DE tends to be the more susceptible to white clouding due to retrogradation after storage for a certain period of time. For example, a solution of liquid dextrin with a low degree of degradation not enzymatically treated becomes insolubilized due to retrogradation and precipitates after storage for 5 days (Figure 3 (1)). In contrast, clarity is successfully maintained in the same solution treated with Glyco Transferase "Amano" (Figure 3 (2)).

As described above, Glyco Transferase "Amano" is expected to serve as a new technological option in food processing.



Figure 3. Maintenance of clarity of liquid dextrin
 (1): Control (dextrin)
 (2): Glyco Transferase-treated
 Both preparations were stored at 5°C for 5 days.



Author profile

Masamichi Okada

Manager, Food & Industrial Enzyme Department, Marketing Division

[Personal history]

Joined Amano Pharmaceutical Co., Ltd. (current Amano Enzyme Inc.) in 1995.

Has been serving as the Manager since 2018.

Fourth Sino-Japan Joint Symposium on Enzyme Technology



Amano Enzyme holds the Sino-Japan Joint Symposium on Enzyme Technology every two years with the aim of promoting the application research of enzymes in Asia through interpersonal relationships between Japanese and Chinese researchers who study enzymes. This year marks the fourth symposium; it was held on Saturday, October 21st, 2017 at Jiangnan University in Wuxi, Jiangsu, China. The symposium was well attended with a total of about 100 attendees involved in enzyme-related work from Chinese universities and companies.

Amano Enzyme and China share a long, close history. It started in 1939, when Amano Enzyme began selling pharmaceuticals in China. In 1979, an Enzyme Technology Friendship Exchange Delegation was dispatched. And in 2007, Amano Enzyme China was established, followed by Amano

Enzyme Manufacturing (China) in 2009. This symposium began in 2011 co-hosted by Jiangnan University and Amano Enzyme, born out of their friendly ties.

At the fourth symposium Sakayu Shimizu (Professor Emeritus, Kyoto University) and Yasuhisa Asano (Professor, Toyama Prefectural University) were invited from Japan to give lectures. A total of seven lectures were made by individuals from Japan and China. Below is a list of all the lecturers and the titles of their talks (in the order given).

Amano Enzyme is committed to strengthening technology exchanges between Japan and China and encouraging development of the enzyme industry in both countries.

① Chen Shao Jun

(Chairman of the China Association of Fragrance Flavor and Cosmetic Industries),
Domestic economic trends and the cosmetics industry in the new normal economy

② Yasuhisa Asano

Enzymes, activation, and molecules: Screening screening studies

③ Sakayu Shimizu

Production of bio-based chemical products: the present and future prospects

④ Yang Sheng

(Researcher, Shanghai Institutes for Biological Sciences, Chinese Academy of Sciences),
The special creation of biocatalysts and the application of an enzyme library

⑤ Zhou Jiahai

(Researcher, Shanghai Institute of Organic Chemistry, Chinese Academy of Sciences),
A study on the catalysis and control mechanisms of enzymes consisting of multiple modules

⑥ Lou Wen Yong

(Professor, School of Food Science and Engineering, South China University of Technology),
The immobilization of enzymes and its applications

⑦ Kang Zhen

(Associate Professor, School of Biotechnology, Jiangnan University),
Genetic mining of cosmetic enzymes and expression by microorganisms



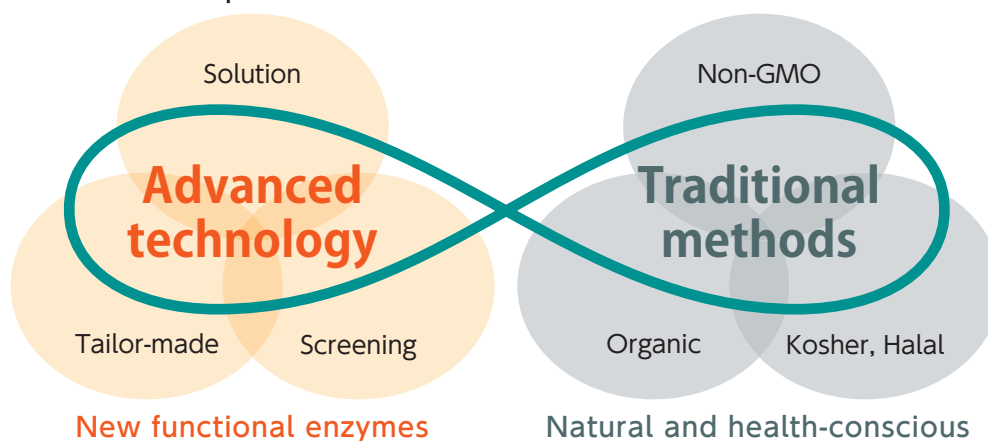
from right

Jiangnan University, President Chen
Professor Emeritus, Shimizu
Professor, Asano
Motoyuki Amano

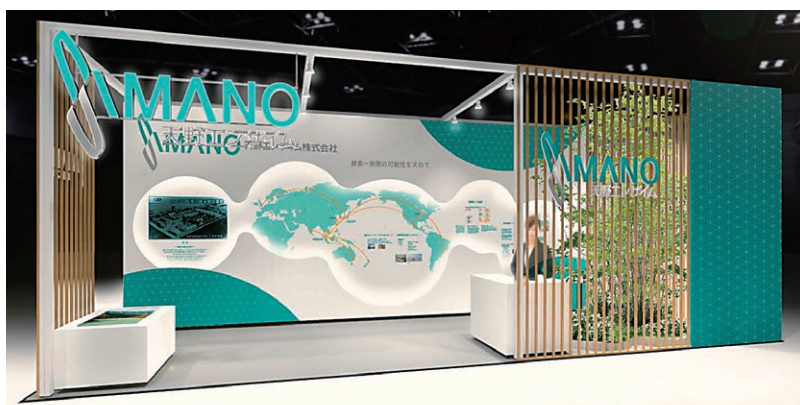
Exhibition information

Amano Enzyme exhibits at various exhibitions around the world to promote our specialty enzymes and examples of their use to a large number of people. Last year, we redesigned our exhibition booths and began introducing new products and the latest trends and information on markets.

〈Product concept〉



Amano Enzyme will work to meet the needs of all customers and continue to provide value to society. Please stop by our booth when you visit an exhibition.



Booth design

〈2018 Exhibitions〉

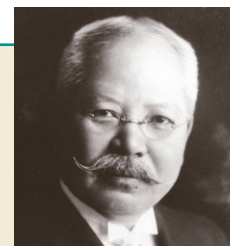
Date	Exhibition	Location
15-17, May	Vitafoods Europe 2018	Geneva (Switzerland)
16-18, May	ifia Japan 2018	Tokyo (Japan)
20-22, Jun.	CPhI China 2018	Shanghai(China)
15-18, Jul.	IFT 18	Chicago (USA)
3-5, Oct.	Fi Asia 2018	Jakarta (Indonesia)
9-11, Oct.	CPhI Worldwide 2018	Madrid (Spain)
4-6, Dec.	CPhI India	Mumbai (India)
13-15, Dec.	16th IFATS	Las Vegas (USA)



The Prof. Jokichi Takamine Research Foundation (NPO)

Professor Jokichi Takamine

Living in the turbulent times of the late 19th and early 20th centuries, Jokichi Takamine left his mark not only as a scientist and entrepreneur but also in international diplomacy. He is known as the father of modern biotechnology due to his research and development of microbially-derived starch degrading enzymes centered around Takadiastase.



Dr. Jokichi Takamine
(photo courtesy Great People of Kanazawa Memorial Museum)

The Prof. Jokichi Takamine Research Foundation (NPO)

The Prof. Jokichi Takamine Research Foundation (NPO) celebrates its 10th anniversary in 2018. We would like to take this opportunity to thank all of our members, whose support has allowed us to reach this great milestone. We will continue to expand our education-oriented activities, such as issuing newsletters and holding lectures aimed at raising awareness about Dr. Jokichi Takamine, who made a significant contribution to the development and commercialization of science and technology in modern Japan and to the friendship between Japan and the United States.

Main activities

In fiscal 2017, we held a total of seven lectures in Tokyo, Saitama, Chiba, Aichi, Ishikawa, and Toyama.

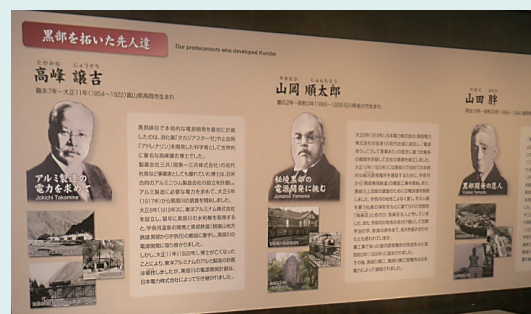
In addition to junior high schools and universities in Takaoka and Kanazawa, lectures were also held at academic conferences, events, and seminars. These saw participants from a wide range of ages and backgrounds take part. In addition, the number of inquiries from newspapers, books, television, etc. has gradually increased, and we are providing materials and information. Opportunities to hear opinions and impressions have increased. We will continue to devote ourselves to such activities in the future and kindly ask for your ongoing support and cooperation.

Topics

2017 marks the 100th anniversary of the establishment of RIKEN and the Kurobe Hydropower Plant. Both were proposed by Dr. Takamine, plans that were realized with great results over time.



Special lecture at the 2017 the Japanese Society for History of Pharmacy (JSHP)



The pioneers of Kurobe
(Jokichi Takamine, Juntaro Yamaoka, and Yutaka Yamada)

Recruitment of new members

Our research foundation widely recruits people who agree with our goals. All members receive publications related to Professor Takamine and a regularly issued journal, along with invitations to various talks and events and other newly obtained information. If you would like to join, please apply by letter including your name (in the case of corporations, your company name and department name), address with postcode, telephone number (landline only), profession, age and gender. We will reply with a payment slip for your initial membership fee and annual membership fee.

The Secretariat

The Prof. Jokichi Takamine Research Foundation

5F Daini-Meiwa Building, 1-15-11 Toranomon, Minato-ku, Tokyo 105-0001

*For further details, visit our website : <http://www.npo-takamine.org/ask.html>

Conference presentation

In 2017 Amano Enzyme has decided academic presentations such as the following.
Please look forward to the activities of the future of Amano Enzyme.

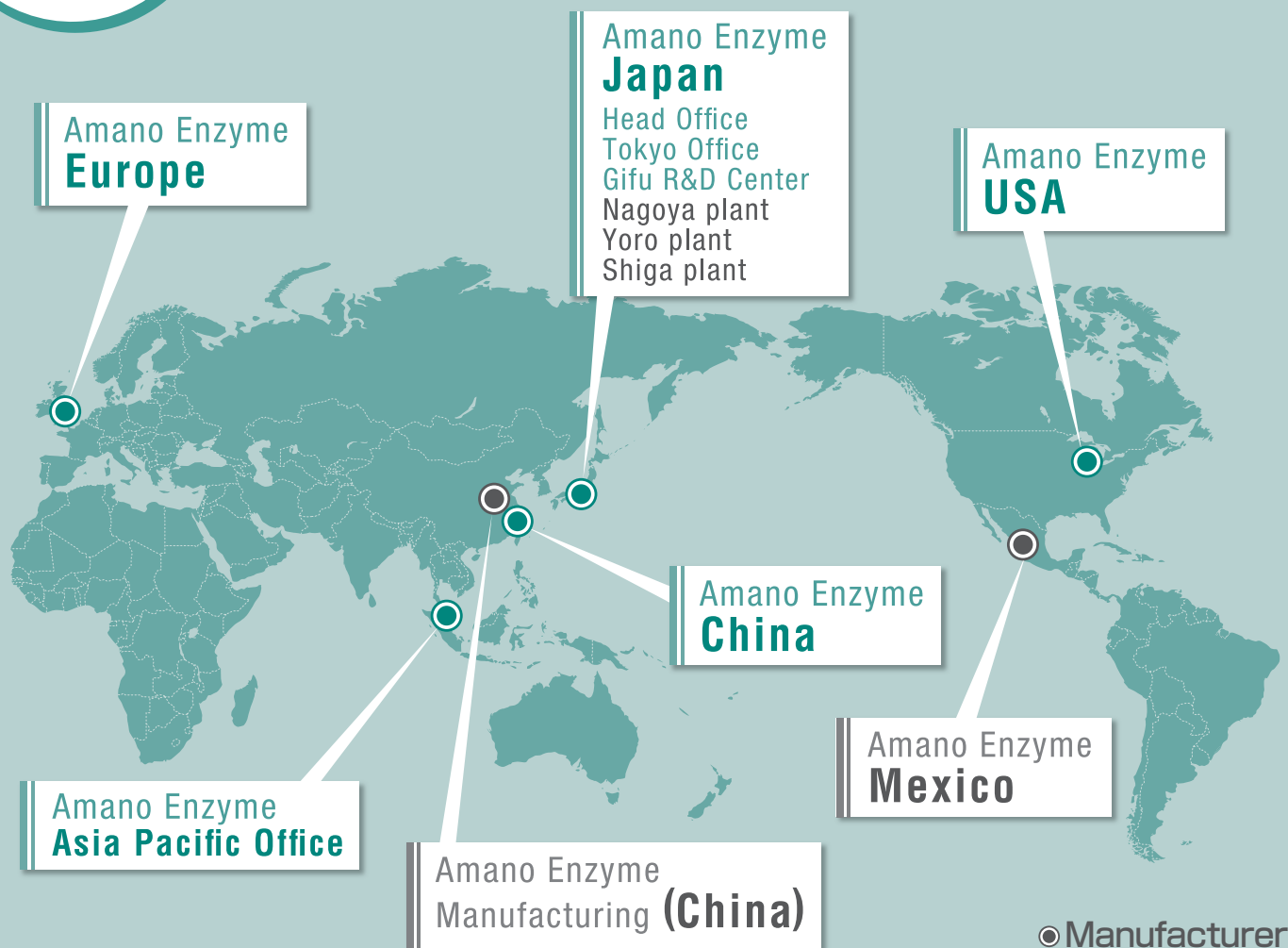
Conference/Meeting	Date	Title	Speaker
The 16th Congress of the Japanese Society for Regenerative Medicine	Mar. 7th, 2017 (Miyagi, Japan)	Enzyme preparation supporting regenerative medicine (Luncheon seminar sponsored by Amano Enzyme)	Goto, M., Yoshimura, K. Chaired by Goto, M.
The 65th the Annual Meeting of the Chubu branch of the Japanese Society of Applied Glycoscience	Mar. 10th, 2017 (Aichi, Japan)	Characteristics and utilization of Industrial enzymes ~Focusing on aroma-forming enzymes~	Okada, M.
48th Annual Meeting of Japan Pancreas Society	Jul. 14th, 2017 (Kyoto, Japan)	Correct usage of digestive enzymes based on latest research (Luncheon seminar sponsored by Amano Enzyme)	Ko, S. Chaired by Okazaki, K.
Environmental-microbiology GODO symposium 2017	Aug. 29th- 31st, 2017 (Sendai, Japan)	Application of microbial lipases.	Yoshida, K., Koikeda, S.
The 11th Symposium on Biorelevant Chemistry, The Chemical Society of Japan	Sep. 7th-9th, 2017 (Tokyo, Japan)	Creation of lipase mutants with extended substrate specificity.	Yoshida, K., Ono, M., Yamamoto, T., Utsumi, T., Koikeda, S., Ema, T.
19th Japanese-German Workshop on Enzyme Technology 2017	Sep. 21st-22nd, 2017 (Rostock, Germany)	Protein engineering of <i>Candida rugosa</i> lipase for improving thermostability and altering substrate specificity.	Ishihara, S., Yoshida, K., Takahashi, T., Koikeda, S., Ishikawa, K.
Enzyme Engineering XXIV	Sep. 24th-28th, 2017 (Toulouse, France)	Protein engineering of <i>Candida rugosa</i> lipase.	Ishihara, S., Yoshida, K., Takahashi, T., Koikeda, S., Ishikawa, K.
25th Annual Meeting of Japan Digestive Disease Week (JDDW2017)	Oct. 12th, 2017 (Fukuoka, Japan)	Comparison of digestive activity of digestive enzyme formulas with the digestive tract model.	Kuroda, M., Ko, S.
Symposium [Frontiers of fermented food research]	Nov. 18th, 2017 (Hiroshima, Japan)	Use of enzymes derived from <i>Aspergillus oryzae</i> in healthcare field.	Kuroda, M., Yamaguchi, S.
15th Annual Meeting of International Federation for Adipose Therapeutics and Science (IFATS2017)	Dec. 1st, 2017 (Miami, USA)	Optimization of enzymatic digestion for SVF isolation from human lipoaspirates.	Asahi, R., Shirado, T., Sato, N., Furukawa, K., Yoshimura, K.
Forum on personal care product & cosmetics technology	Dec. 10th, 2017 (Dongguan, China)	Technology of enzyme use in cosmetics field	Kuroda, M. Interpreter: Wang, Y.
The Second International Symposium on Biofunctional Chemistry, The Chemical Society of Japan	Dec. 14th-16th, 2017 (Kyoto, Japan)	Synthetically useful variants of industrial Lipase from <i>Burkholderia cepacia</i> and <i>Pseudomonas fluorescens</i> .	Yoshida, K., Ono, M., Yamamoto, T., Utsumi, T., Koikeda, S., Ema, T.
The 19th Symposium on the Chemistry of Biocatalysis, The Society of Biocatalysis Japan	Dec. 21st-22nd, 2017 (Nagasaki, Japan)	Creation of industrial lipase mutants with extended substrate specificity.	Yoshida, K., Ono, M., Yamamoto, T., Utsumi, T., Koikeda, S., Ema, T.

Journal/Book	Date	Title	Author
Journal of Applied Glycoscience 2017 Vol.7 p.181	Apr. 2017	A foreword Screening - Looking for the possibility of microorganisms -	Mori, S.
Kagaku to Seibutsu (Japan Society for Bioscience, Biotechnology, and Agrochemistry)2017, Vol.55, No.5 p.303-305	May, 2017	Monobody-mediated alteration of enzyme specificity.	Tanaka, S., Koide, S.
Nutrition Research 2017, Vol.44, p.60-66	Aug, 2017	The consumption of an acid protease derived from <i>Aspergillus oryzae</i> elevates colon <i>Bifidobacterium</i> levels in rats fed a high-fat diet.	Yang, Y., Kato, N., Kuroda, M., Yamaguchi, S.
Manual for diagnostics of exocrine pancreatic insufficiency p.53-56	Oct, 2017	Treatment of exocrine pancreatic insufficiency [Digestive enzymes medicine]	Kuroda, M., Ko, S.
Organic & Biomolecular Chemistry (Royal Society of Chemistry) 2017, Vol.15, p. 8713-8719	Nov, 2017	Synthetically useful variants of industrial Lipase from <i>Burkholderia cepacia</i> and <i>Pseudomonas fluorescens</i> .	Yoshida, K., Koikeda, S., Ema T.

Volume
21

Amano Enzyme

World Network



● Manufacturer



Enzyme – Explore Unlimited Possibilities

<http://www.amano-enzyme.co.jp/>

AMANO ENZYME INC. (Publisher)

Head Office:

2-7, 1-chome,
Nishiki, Naka-Ku, Nagoya,
460-8630 Japan

Tel: +81-(0) 52-211-3032

Fax: +81-(0) 52-211-3054

E-mail: www-info@amano-enzyme.com

Tokyo Office:

1-1, 1-chome, Uchisaiwai-cho,
Chiyoda-ku, Tokyo,
100-0011 Japan

Tel: +81-(0) 3-3597-0521

Fax: +81-(0) 3-3597-0527

AMANO ENZYME U.S.A. CO., LTD.

1415 Madeline Lane, Elgin, IL 60124 U.S.A.

Tel: +1-847-649-0101

Fax: +1-847-649-0205

E-mail: aeu.sales@amano-enzyme.com

AMANO ENZYME EUROPE LTD.

Roundway House, Cromwell Park,
Chipping Norton, Oxfordshire, OX7 5SR, U.K.

Tel: +44-(0) 1608-644677

Fax: +44-(0) 1608-644336

E-mail: aeu.sales@amano-enzyme.com

AMANO ENZYME CHINA LTD.

C3-5F "800SHOW", No.800,
ChangDe Road, Shanghai 200040, P.R.China

Tel: +86-(0) 21-6249-0810

Fax: +86-(0) 21-6248-7026

E-mail: shanghai@amano-enzyme.com

AMANO ENZYME ASIA PACIFIC OFFICE

Suite 710, 7th Floor, Block B, Kelana Square,
17, Jalan SS7/26, Kelana Jaya, 47301
Petaling Jaya, Selangor, Malaysia

Tel: +60-3-7887-2351

Fax: +60-3-7887-0351