Experimental Plant Division

Head, Masatomo KOBAYASHI

Goal

Plant science is vital for food production and environment protection. The mission of this laboratory is to promote plant science through collection and distribution of bioresources. We participate with the National Bioresource Project and distribute *Arabidopsis* seeds, plant genetical materials and plant cultured cells to the international community of plant science. We also proceed to the development of novel technologies for the establishment and characterization of bioresources as well as to the conduction of technical training course.

Activities

1. Collection, preservation and distribution of plant resources
2. Development of novel techniques on the production, preservation and characterization of plant resources
3. Promotion of training course on the handling and advanced use of plant resources

Members

Head

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Senior Visiting Scientist

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Assistant

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Ishizuka, Sasaki, Kawamura, Takahashi, Shitomi, Mori
Uchida, Aso, Fujimura, Nemoto, Tarui
Kobayashi(S), Iuchi(A), Nakayama, Terakawa, Mori, Ishiyama
Iuchi, Kobayashi(M), Ota, Inaba, Sugawara

Specific aim **I. Collection, preservation and distribution of plant resources**

The principal resources that we preserve are the materials of *Arabidopsis thaliana*, the most popular model plant in the world. We extensively collect and distribute genomic materials such as seeds of insertion mutants and full-length cDNA clones that have lots of interests for plant research community. Global contribution is the important feature of our project. By the end of 2005, more than 1,100 laboratories and research groups over 39 countries and areas have registered as our user. Now we preserve approx. 360,000 accessions of *Arabidopsis* seeds, cDNA clones and plant cell lines. Since 2002, we have distributed more than 17,000 plant materials to the international research community. As we devote our efforts to control the quality of the resource, there have been very few complaints about the provided materials. Now this Division not only leads the plant resource project in Japan but also becomes one of the distinguished plant resource centers in the world.

1) Plant seeds

Arabidopsis transposon-tagged mutants deposited from RIKEN Genomic Sciences Center (GSC) as well as *Arabidopsis* activation-tagged mutants established by RIKEN Tsukuba Institute and RIKEN GSC are preserved and distributed. The transposon-tagged line is suitable for reverse genetics because the insertion site of transposable (Ds) element has been characterized, while the activation-tagged line is suitable for phenotype screening (forward genetics). In order to improve the value of transposon-tagged, we are going to establish seed pools that are homozygous for Ds insertion.

In addition to the mutant lines mentioned above, the Arabidopsis Information Service (AIS) collection of *Arabidopsis* seeds formerly preserved in The Sendai Arabidopsis Seed Stock Center (SASSC) has been deposited to us. The AIS collection is famous because AIS was the first organization that distributed seeds of wild-type Arabidopsis. Up to now, nearly 400 natural accessions of Arabidopsis that are useful for the

research on QTL are available from us.

2) Plant genetical materials

Full-length cDNA is important for the post-genome research, since it is useful to the production of transgenic plants and functional proteins. We preserve and distribute full-length cDNA clones of three plant species, namely, RIKEN Arabidopsis full-length cDNA (RAFL) clones (from RIKEN GSC), *Physcomitrella patens* (model moss) full-length cDNA clones (from National Institute of Basic Biology) and poplar (model tree) full-length cDNA clones (from Forestry and Forest Products Research Institute). The RAFL clone is regarded as world standard resource and used in hundreds of laboratories. As Japan has outstanding techniques to produce full-length cDNA clones, this category of resource will be established from some more plant species in near future. We intend to collect, preserve and distribute such clones to serve world research community.

3) Plant cultured cells

We preserve and distribute cultured cell lines of model plants such as Arabidopsis, rice, tobacco, and *Lotus japonicum*. Among them, tobacco BY-2 and Arabidopsis T87 cells are most popular cell lines. Request for the Arabidopsis T87 cell line is significantly increasing as we introduce this cell line to the Arabidopsis research community. We also provide technical information of this resource via website, e-mail and training program. Therefore our project is quite valuable for researchers who are going to use plant cultured cells in their research.

II. Development of technology

1) Development of technologies for the production, preservation and characterization of plant cultured cells

Cultured cells are important for the studies on signal transduction and cell development as well as production of useful materials. Recently, we have established the protocol for cryopreservation of tobacco BY-2 cells. We intend to apply the technique to the preservation of various cell lines.

2) Characterization of wild-type Arabidopsis

Wild-type Arabidopsis is useful for the research on biotic- and abiotic-stress, however, the phylogenetic information for this material is mostly unavailable. Approximately 400 natural accessions of Arabidopsis are preserved in RIKEN BRC, and we are now analyzing their phylogenetic background by establishing

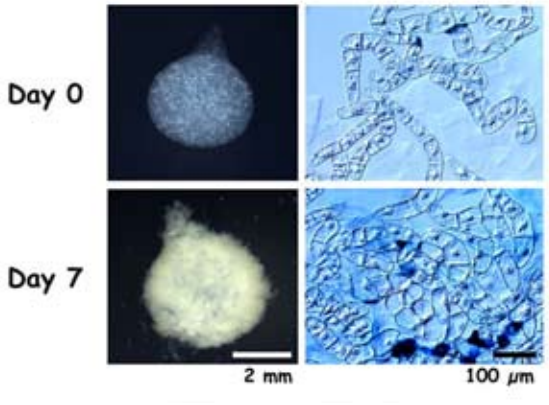
Experimental Plant Division was awarded the special prize from Botanical Society of Japan

maker system with simple sequence length polymorphism. We also characterize their response against various stresses to improve the value of resource.

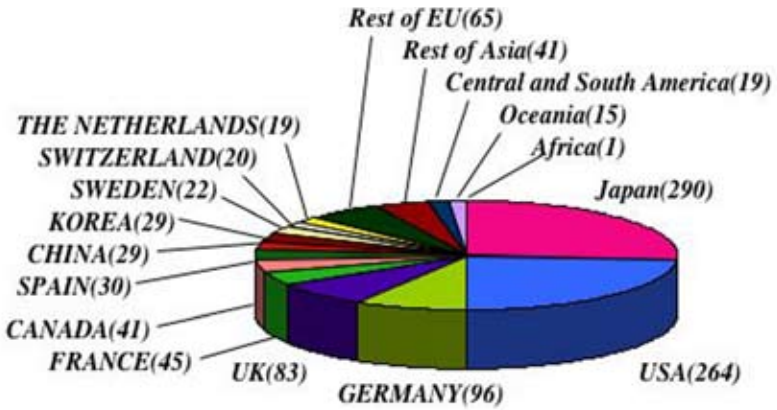
III. Training course

Since 2004, we have been giving training courses of basic and advanced techniques required for transformation and preservation of plant cultured cells. Total 23 researchers and students from universities, research institutes and private sectors have joined the program.

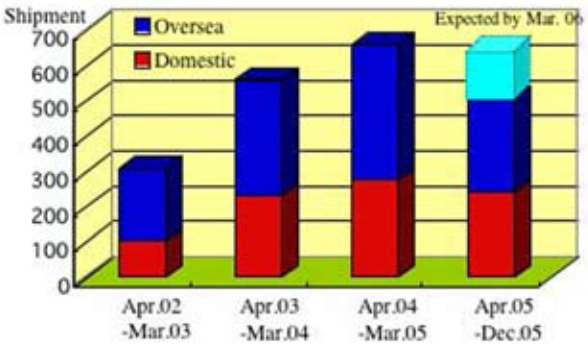
Researchers from various organization participate in the training course held by Experimental Plant Division



Growth of tobacco BY-2 cells in alginate beads for cryopreservation



Location of User's Laboratory



Destination of Shipment

Publications

Original Papers (* Peer reviewed journal)

1. Igarashi D., Miwa T., Seki M., Kobayashi M., Kato T., Tabata S., Shinozaki K. and Ohsumi C.: "Identification of photorespiratory glutamate:glyoxylate aminotransferase (GGAT) gene in *Arabidopsis*." Plant J. 33, 975-987 (2003).*

2. Sasaki A., Itoh H., Gomi K., Ueguchi-Tanaka M., Ishiyama K., Kobayashi M., Jeong D.-H., An G., Kitano H., Ashikari M. and Matsuoka M.: "Accumulation of phosphorylated repressor for gibberellin signaling in an F-box mutant." Science 299, 1896-1898 (2003).*

3. Abe H., Urao T., Ito T., Seki M., Shinozaki K. and Yamaguchi-Shinozaki K.: "Arabidopsis AtMYC2(bHLH) and AtMYB2(MYB) function as transcriptional activators in abscisic signaling." Plant Cell 15, 63-78 (2003).*

4. Narusaka Y., Narusaka M., Seki M., Ishida J., Nakashima M., Kamiya A., Enju A., Sakurai T., Satoh M., Kobayashi M., Tosa Y., Park P. and Shinozaki K.: "The cDNA microarray analysis using an Arabidopsis pad3 mutant reveals the expression profile and classification of genes induced by *Alternaria brassicicola* attack." Plant Cell Physiol. 44, 377-387 (2003).

5. Sakai M., Sakamoto T., Saito T., Matsuoka M., Tanaka H. and Kobayashi M.: "Expression of novel rice gibberellin 2-oxidase gene is under homeostatic regulation by biologically active gibberellins." J. Plant Research 116, 161-164 (2003).

6. Motohashi R., Ito T., Kobayashi M., Taji T., Nagata N., Asami T., Yoshida S., Yamaguchi-Shinozaki K. and Shinozaki K.: "Functional analysis of the 37-kDa inner envelope membrane polypeptide in chloroplast biogenesis using a Ds-tagged Arabidopsis pale-green mutant." Plant J. 34, 719-731 (2003).

7. Sakamoto T., Morinaka Y., Ishiyama K., Kobayashi M., Itoh H., Kayano T., Iwahori S., Matsuoka M. and Tanaka H.: "Genetic manipulation of gibberellin metabolism in transgenic rice." Nature Biotech. 21(8): 900-913 (2003).

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10. Narusaka Y., Narusaka M., Seki M., Fujita M., Ishida J., Nakashima M., Enju A., Sakurai T., Satou M., Kamiya A., Park P., Kobayashi M. and Shinozaki K.: "Expression profiles of Arabidopsis phospholipase A IIA gene in response to biotic and abiotic stresses." Plant Cell Physiology 44, 1246-1252 (2003)*.

11. Zhao L., Nakazawa M., Takase T., Manabe K., Kobayashi M., Seki M., Shinozaki K. and Matsui M.: "Overexpression of LSH1, a member of an uncharacterised gene family, causes enhanced light regulation of seedling development." Plant J. 37, 694-706 (2004).*

12. Higuchi M., Pischke MS., Mahonen AP., Miyawaki K., Hashimoto Y., Seki M., Kobayashi M., Shinozaki K., Kato T., Tabata S., Helariutta Y., Sussman MR. and Kakimoto T.: "In planta functions of the Arabidopsis cytokinin receptor family." PNAS 101, 8821-8826 (2004).*

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"A new lead compound for abscisic acid biosynthesis inhibitors targeting 9-*cis*-epoxycarotenoid dioxygenase." Bioorg. Med. Chem. Lett. 14, 3033-3036 (2004).*

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20. Kobayashi T., Niino T. and Kobayashi M.: "Simple cryopreservation protocol with an encapsulation technique for tobacco BY-2 supension cell cultures." Plant Biotech. 22(2), 105-112 (2005).*

21. Itoh H., Sasaki A., Ueguchi-Tanaka M., Ishiyama K., Kobayashi M., Hasegawa Y., Minami E., Ashikari M. and Matsuoka M.: "Dissection of the phosphorylation of rice DELLA protein, SLENDER RICE1." Plant Cell Physiol. 46(8), 1392-1399 (2005).*

22. Suzuki H., Ishiyama K., Kobayashi M. and Ogawa T.: "Specific expression of the gibberellin 3β-hydroxylase gene, *HvGA3ox2*, in the epithelium is important for *Amy*1 expression in germinating barley seeds." Plant Biotech. 22(3), 195-200 (2005).*

23. Ueguchi-Tanaka M., Ashikari M., Nakajima M., Itoh H., Katoh E., Kobayashi M., Chow T.-Y., Hsing Y.-I.C., Kitano H., Yamaguchi I. and Matsuoka M.: "GIBBERELLIN INSENSITIVE DWARF1 encodes a soluble receptor for gibberellin." Nature. 437(29), 693-698 (2005).*

24. Katagiri T., Ishiyama K., Kato T., Tabata S., Kobayashi M. and Shinozaki K.: "An important role of phosphatidic acid in ABA signaling during germination in *Arabidopsis thaliana*." The Plant J. 43, 107-117 (2005).*

25. Osakabe K., Abe K., Yamanouchi H., Takyuu T., Yoshioka T., Ito Y., Kato T., Tabata S., Kurei S., Yoshioka Y., Machida Y., Seki M., Kobayashi M., Shinozaki K., Ichikawa H. and Toki S.: "*Arabidopsis* Rad51B is important for double-strand DNA breaks repair in somatic cells." Plant Mol. Biol. 57, 819-833 (2005).*

Oral Presentations

1. Motohashi R., Myouga F., Yamazaki T., Ito T., Kuromori T., Seki M., Kobayashi M., and Shinozaki K.: "Large-scale analysis of albino and pale green mutants using *Ac/Ds* transposon system in Arabidopsis." 14th International Conference on Arabidopsis Research, Madison, Jun. (2003).

2. Myouga F., Motohashi-Takahashi R., Yamazaki T., Ito T., Kuromori T., Hirayama T., Seki M., Kobayashi M., Nagata N., Yoshida S., and Shinozaki K.: "Large-scale genetic analysis of albino and pale green (*apg*) mutants using *Ac/Ds* transposon system in *Arabidopsis*." Plant Biology 2003, Honolulu, Jul. (2003).

3. Hobo T., : "The expression and its role of *AtNCED2* gene, encoding 9-cis-epoxycarotenoid dioxigenase, a key enzyme in ABA biosynthesis in Arabidopsis." Plant Biology 2003, Honolulu, Jul. (2003).

4. Taji T., : "Functional genomics in Arabidopsis-related halophyte, *Thellungiella halophila* using Arabidopsis microarray." Plant Biology 2003, Honolulu, Jul. (2003).

5. Ito Y., Katsura K., Maruyama K., Taji T., Kobayashi M., Seki M., Shinozaki K. and Yamaguchi-Shinozaki K.: "Overexpression of the DREB/CBF gene family improves stress tolerance to

drought, high salt and low temperature in rice." Keystone Symposia on Plant Response to Abiotic Stress, Santa Fe, Feb. (2004).

6. Ito Y., Katsura K., Maruyama K., Taji T., Kobayashi M., Seki M., Shinozaki K. and Yamaguchi-Shinozaki K.: "Overexpression of the DREB/CBF gene family improves stress tolerance to drought, high salt and low temperature in rice." Gordon Research Conference on Salt and Water Stress in Plants, Hong Kong, Jun. (2004).

7. Taji T., Seki M., Satou M., Sakurai T., Kobayashi M., Ishiyama K., Narusaka Y., Narusaka M., Zhu J.-K. and Shinozaki K.: "Comparative genomics in salt tolerance between Arabidopsis thaliana and Arabidopsis-related halophyte, Thellungiella halophila using Arabidopsis microarray.", Gordon Research Conference on Salt and Water Stress in Plants, Hong Kong, Jun. (2004).

8. Katagiri T., Kobayashi M. and Shinozaki K.: "Functional analysis of a phosphatidic acid in ABA signaling during germination." 15th International Conference on Arabidopsis Research, Berlin, Jul. (2004).

9. Higuchi M ., Pischke MS., Mahonen AP., Miyawaki K., Hashimoto Y., Seki M., Kobayashi M., Shinozaki K., Kato T., Tabata S., Helariutta Y., Sussman MR. and Kakimoto T.: In planta functions of the Arabidopsis cytokinin receptor family." 15th International Conference on Arabidopsis Research, Berlin, Jul. (2004).

10. Abe H., Urao T., Seki M., Ito T., Kobayashi M., Shinozaki K. and Yamaguchi-Shinozaki K.: "Arabidopsis MYC(bHLH) and MYB proteins function as transcriptional activators in abscisic acid signaling." 15th International Conferince on Arabidopsis Research, Berlin, Jul. (2004).

11. Kobayashi M., Abe H., Iuchi S. and Kobayashi T.: "Report of resource project in RIKEN BRC."15th International Conference on Arabidopsis Research, Berlin, Jul. (2004).

12. Kobayashi T., Niino T. and Kobayashi M.: "Cryopreservation of tobacco BY-2 by encapsulation simple prefreezing method." International Symposium on Cell and Molecular Biology of Tobacco BY-2 Cells, Yokohama, Sep. (2004).

13. Asami T., Han S.‐Y., Kitahata N., Saito T., Kobayashi M., Nakashima K., Yamaguchi-Shinozaki K., Shinozaki K. and Yoshida S.: "Abscisic acid biosynthesis inhibitor targeting 9-cis-epoxycarotenoid dioxygenase." 18th International Conference on Plant Growth Substances, Canberra, Sep. (2004).

14. Kakimoto T., Higuchi M., Pischke MS., Mahonen AP., Miyawaki K., Hashimoto Y., Seki M., Kobayashi M., Shinozaki K., Kato T., Tabata S., Sussman M. and Helariutta Y.: "Functions of the three cytokinin receptors in Arabidopsis." 18th International Conference on Plant Growth Substances, Canberra, Sep. (2004).

15. Iuchi S. and Kobayashi M.: "Screening of *Arabidopsis thaliana* ecotype in abiotic stress conditions." 18th International Conference on Plant Growth Substances, Canberra, Sep. (2004).

16. Suzuki H., Ishiyama K., Kobayashi M. and Ogawa T.: "Specific expression of the gibberellin 3-beta-hydroxylase gene in the epithelium of germinating barley seeds." 18th International Conference on Plant Growth Substances, Canberra, Sep. (2004).

17. Seki M., Ishida J., Nakajima M., Enju A., Sakurai T., Iida K., Satou M., Akiyama K., Oono Y., Fujita M., Mizukado S., Kamei A., Narusaka M., Narusaka Y., Yamaguchi-Shinozaki K., Ecker J., Kobayashi M., Kohara Y., Sawasaki T., Hayashizaki Y., Yokoyama S., Toyoda T., Endo Y. and Shinozaki K.: "Functional genomics using RIKEN Arabidopsis full-length (RAFL) cDNAs." Cold Spring Harbor Laboratory Meeting on Plant Genomes: From Sequence to Phenome, Cold Spring Harbor, Dec. (2004).

18. Niino T., Tanaka D. and Kobayashi T.: "Utilization of plant cryopreservation in Japan." Congreso Internacional Biotecnologia y Agricultura Bioveg 2005, Cuba, Feb. (2005).

19. Kobayashi M., Abe H., Iuchi S. and Kobayashi T.: "Report on resource project in RIKEN BRC." 16th International Conference on Arabidopsis Research, Madison, Jun. (2005).

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