Issue Number 4 March 2008

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Hideko Urushihara, Professor,
Graduate School of Life & Environmental Sciences, University of Tsukuba

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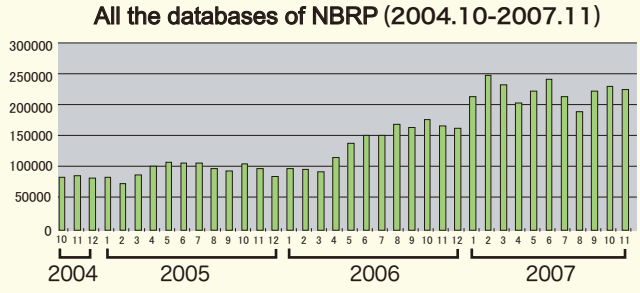
Transition of the Number of Monthly Database Users



Introduction to Resource Center No. 22

National BioResource Project "Cellular Slime Molds"

Hideko URUSHIBARA, Professor, Graduate School of Life and Environmental Sciences, University of Tsukuba

1 What are cellular slime molds?

[What are cellular slime molds?]

Cellular slime molds are referred to as dictyostelid, a group of microorganism that inhabits relatively wet surfaces of soil, such as underneath fallen leaves. A feature of cellular slime molds is that under starvation stress, unicellular amoebas spread multicellularly, sporulate, and form carpophores that are composed of a cluster of spores and a supporting stalk (Fig. 1). At the mention of "slime molds", we often think of myxomycetes with colorful carpophores on a plasmodium, which grow in sizes in the order of square meters (these myxomycetes are well known from the research of Kumagusu Minakata). However, these myxomycetes are true slime molds consisting of multinuclear cytomegalic cells and are different from cellular slime molds. Carpophores of cellular slime molds are mononuclear and multicellular and grow to approximately 1–5 mm in height, which can be barely identified by naked eyes. The so-called pseudoplasmodium is a slug-like structure that is formed in the process of carpophore formation; it is motile in most species and is called "slug" in English and "mobile body" in Japanese (Fig. 2). Although carpophores develop asexually, sexual reproduction leading to the formation of macrocysts is also noted. Cellular slime molds are classified into 3 genera—Dictyostelium, which forms a typical carpophore in which a spore cluster is supported by a single steady stalk; Polysphondylium, which has a branched stalk; and Acytostelium, which develops a fragile noncellular stalk (Fig. 3).

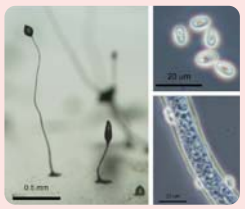


Fig.1 : Enlarged images of carpophores (left), spores (top right), and a stalk (bottom right) of cellular slime molds.

Fig. 2 :
A scanning microscopic image that helps to understand the life cycle of *Dictyostelium discoideum* at a glance (reprinted from the image collection of dictyBase; a slug is shown on the bottom left).

Dictyostelium
Polysphondilium (a branched stalk)
Acytostelium (noncellular stalks)

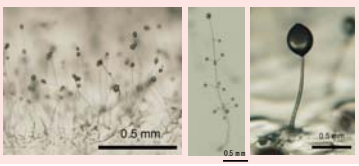
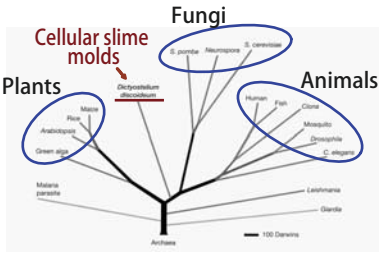


Fig.3 : Carpophores of the 3 genera of cellular slime molds. The images correspond to the names on the phylogenetic tree shown above.

[Position of Cellular Slime Molds on the Phylogenetic Tree]

Although cellular slime molds exhibit amoeboid movement or possess a slug body at a certain stage of development, they form a stalk composed of cellulose and vacuolized stalk cells; this characteristic of stalk formation appears to be similar to that of plants. On the other hand, as symbolized by the Japanese name "Kiirotamahokorikabi" (yellow spherical dust fungus), carpophores remind us of fungi. However, recent molecular phylogenetic analysis and genome com- parison have revealed that cellular slime molds diverged after the evolution of plants and before the evolution of fungi on the phylogenetic tree of eukaryotes (Fig. 4). I usually answer a frequently asked question, "Are cellular slime molds animals or plants?" by saying, "They are unique organisms that are neither animals nor plants."



(Eichinger et al., Nature 435:43-57, 2005)

Fig.4: Phylogenetic tree showing the evolutionary position of cellular slime molds.

[Usefulness of Cellular Slime Molds as a Model Organism]

Cellular slime molds feed on bacteria in soil and agricultural environment. It is possible to isolate a standard strain of *D. discoideum* in a pure culture (Fig. 5) and to easily apply various biochemical and molecular biological approaches and genetic manipulation techniques. Cellular slime molds are frequently used as model organisms in various fields of basic science such as cell biology, developmental biology, biophysics, and mathematical biology because of their active proliferation and movement, adoption of a simple multicellular system in which cells differentiate into only 2 forms—spores and stalk cells—within a period of 24 hours, and flexible adjustment of the ratio between cell types. In contrast, it is known that cellular slime molds harbor numerous genes that are pathogenic for humans and can serve as hosts for pathogenic microorganisms (Fig. 6); hence, they are listed as important model organisms of medical science by NIH (http://www.nih.gov/science/models/).

Fig.5: Culturing of cellular slime molds.The optimum temperature to culture cellular slime molds is 20°C–25°C.Although usually a simple incubator, as shown in the image (left) is used for culturing cellular slime molds, it is also possible to culture at room temperature, except in summer. Double culturing and development by using round and square dishes and shake-culturing in liquid media are illustrated. In addition, a microscope (top right) and a clean bench are shown.

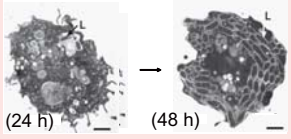


Fig.6 : Infection of *Legionella* bacterium (indicated by L), a causative microorganism of pneumonia (left). Although *Legionella* is incorporated into a food vacuole, it avoids fusion with lysosome and continues to proliferate, thereby completely filling the cell in a single day.
Farbrother et al,m 2006, Cell Microbiol. 8:438-456.

↳To the next page

In addition, cellular slime molds are reported to secrete secondary metabolites that show antiproliferative effects on cancer cells; thus, cellular slime molds are considered to be promising microbiological resources for drug design. The genome and cDNA analyses of cellular slime molds were completed in 2005; thus, substantial genetic information is now available.

2 Aim to Develop as a Core Facility of Cellular Slime Molds

[Implementation Structure]

The collection, preservation, and distribution of cellular slime mold resources are managed by the University of Tsukuba, the core facility dealing with gene clones of *D. discoideum*, and the National Institute of Advanced Industrial Science and Technology (AIST), a sub-institute that handles strains. These institutes cooperate with external administrative committee members who are selected from the workshop of cellular slime molds (Fig. 7).

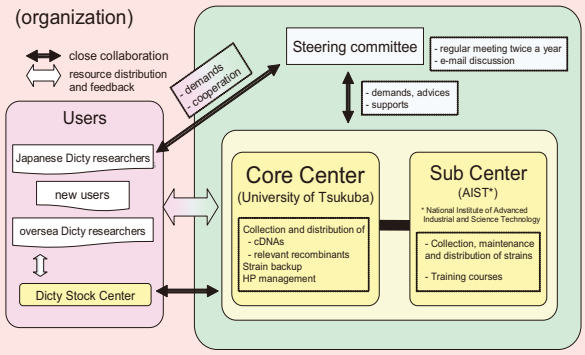


Fig. 7: Organizational chart for resource consolidation

[Genetic Resources]

Non-redundant sets of cDNA clones preserved in the University of Tsukuba, which were primarily used for the EST analysis will be rearranged and preserved as genetic resources. Among 12,500 genes predicted, currently, approximately 3,000 full-length clones are arrayed on 96-well plates. All the non-redundant clones will be consolidated in the future. Since cellular slime molds are convenient to handle for post-genome analysis, research using gene sets as a whole has been initiated.

[Strain Resources]

The Dicty Stock Center (DSC) established in Columbia University, US, is supported by NIH and preserves cellular slime molds. However, it is difficult to convey this information to Japanese researchers who have not used cellular slime molds as a research resource; furthermore, it is problematic to export cellular slime mold cultures from US to Japan, especially recombinant strains. Therefore, our project aimed to collect, preserve, and distribute resources with a focus on Japanese researchers. We closely cooperate with DSC and have established a framework to mutually complement each other so that the stock of resources at DSC and NBRP can serve as a global standard of cellular slime mold resources. Moreover, we hope to receive feedback from local and international gene clone users as we strive to consolidate both genes and recombinant strains as high quality resources. We also offer consultations to inexperienced researchers regarding which strain to use and culturing methods. We hope that all the researchers would feel free to use our resources as secondary materials.

A Request to Researchers

We have a request to researchers who subscribe to our bioresources.
When researchers publish research achievements such as journal articles, please ensure that the information of the resources used is included in Materials & Methods or Acknowledgement. In addition, please contact the facility from where the resources were provided.
NBRP has opened a registration website for achievement articles. The information can be easily registered using the address provided following.
http://rrc.nbrp.jp/

Coming up in the next issue!
The special topic on resources discussed in the next month's issue will be "Fowls and Quails."

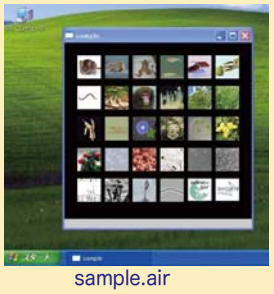
10 minutes Information Technology - 30 -

Development of Desktop Applications by Flex

A sample web application developed by Flex and Flex Builder was introduced in the previous issue (http://www.shigen.nig.ac.jp/shigen/news/n_letter/2008/2/sample.html). In Flex, not only web applications but also desktop applications can be developed.

Desktop applications can be developed in the same manner as web applications by Flex Builder using simple manipulations such as drag-and-drop.

As an example, I have developed "sample.air", a simple desktop application which opens a website when you click on an image.


sample.air

This application has an ".air" extension and you can run it on an environment where Adobe Integrated Runtime (AIR) is installed. The air files will run on the environments stated below.

Windows	Mac OS X
Intel Pentium 1GHz or faster processor Microsoft Windows 2000 with Service Pack 4; Windows XP with Service Pack 2; or Windows Vista Home Premium, Business, Ultimate, or Enterprise 512MB RAM	PowerPC G4 1GHz or faster processor or Intel Core Duo 1.83GHz or faster processor Mac OS X v10.4.910 or 10.5.1 (PowerPC); Mac OS X v10.4.9 or later, 10.5.1 (Intel) 512MB RAM

Developers can provide users with an installation option called seamless installation to install AIR and AIR applications together (this method may not work on some types and versions of browsers).

■ **Here is the link to seamless installation.**

http://www.shigen.nig.ac.jp/shigen/news/n_letter/2008/3/index.html

※ If the seamless installation does not work properly, please download the applications one-by-one from the link given below. Start by installing AIR, followed by "sample.air".
(If the file name of "sample.air" is changed to "sample.zip" after downloading, please change it back to "sample.air.")

■ **AIR (Software to run "air" applications)**

http://labs.adobe.com/downloads/air.html

■ **sample.air ("air" application developed as a sample)**

http://www.shigen.nig.ac.jp/shigen/news/n_letter/2008/3/sample.air

You can download various AIR applications from the Adobe website (http://www.adobe.com/jp/devnet/air/gallery/).

There is also an AIR contest (http://www.adobe.com/jp/special/air/contest/) going on so please try to submit your original AIR application.

(Shingo SAKANIWA)

Editor's Note Prof. Urushihara provided us the topic on cellular slime molds, which were newly adopted in the second National BioResource Project. I was impressed by a movie that she had presented at the panel exhibition site of the Biochemistry and Molecular Biology (BMB) conference 2007; this movie showed aggregation of single cells. Cellular slime molds are definitely a fascinating creature. According to Prof. Urushihara, it is not very difficult to culture cellular slime molds; therefore, in the near future, cellular slime molds may become as popular as Escherichia coli among researchers. I sincerely appreciate the contribution of Prof. Urushihara. (Y.Y.)

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