論　　説

生きている花粉からの有機溶媒による脂質の抽出

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Extraction of lipid from *Lilium auratum* pollen without losing the viability by soaking in organic solvent

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Iwanami (1971, 1972a, 1972b, 1973a, 1974) reported that the pollen grains and seeds of various higher plants and resting eggs of brine-shrimp retained the viability for many days in organic solvents such as diethyl ether, acetone, pyridine and chloroform. Since these organic solvents are known as extracting solvent of lipids, next important problem is whether the solvents diffuse into pollen grain through cell membrane or not. To check up this point, *Lilium auratum* pollen stored in dry condition and stored in organic solvents were studied electron microscopically. During the observations, the authors' attention was directed to alteration of lipid vesicles of the pollen grains.

1) Material and method

Pollen grains of *Lilium auratum* were collected from freshly opened anthers at anthesis and stored in following three conditions.

1) in a plastic bottle with silica gel at −15°C.

2) in acetone in a corked test tube at −15°C.

3) in diethyl ether in a corked test tube at −15°C.

After 1 year, pollen grains stored in organic solvents were filtered and dried by means of an aspirator. These powdered pollens and pollens stored in dry condition were fixed with 3% glutaraldehyde (3hr), rinsed in water (12hr) and postfixed with 1% osmium tetroxyde (20hr). After fixation and dehydration, the materials were embedded in methacrylate : epon (1 : 1). They were not satined with any reagent because fine structures of cytoplasm such as golgi apparatus and mitochondria come to be clear, however, black color of lipid vesicle is torned down, by the treatment. Ultrathin sections were cut by using a Reichert-urtramicrotome with a glass knife and examined with a JEM-100B electron microscope. To test the viability of these pollen grains, pollens were sown on culture medium (sucrose 10%, agar 1%, boric acid 100 ppm) in a straight line as reported in previous paper (Iwanami 1959). After culture for 24 hours at 28°C, germination rate and pollen tube length were measured by using universal projector

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(Olympus UP-560). Each treatment was duplicated three times and mean values were recorded.

2) Results and discussion

In pollen grain stored in dry condition for 1 year at −15°C, numerous large lipid vesicles were distributed over the entire volume of pollen grain and very small lipid vesicles were distributed along cell

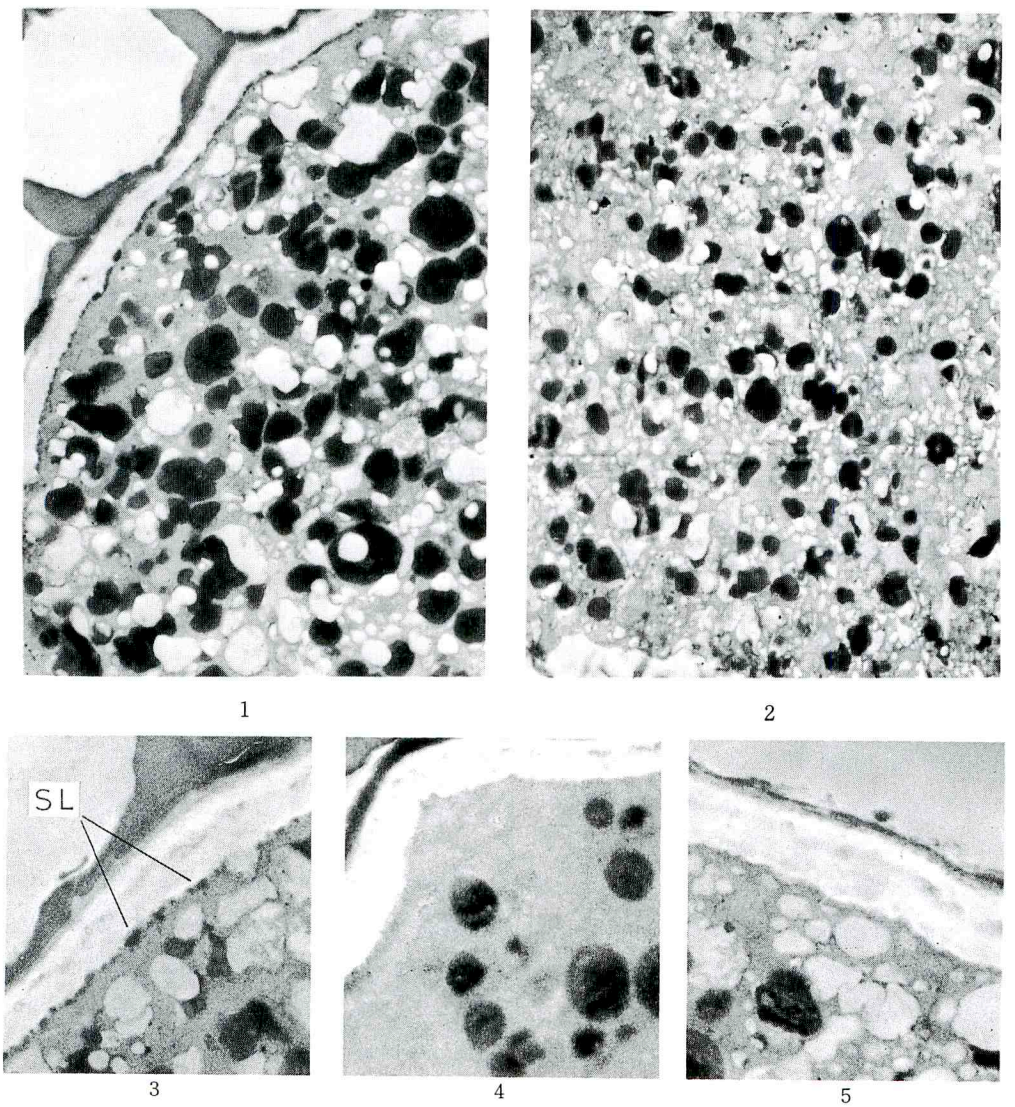

Fig. 1 Alteration of lipid vesicles in *Lilium auratum* pollen by soaking in organic solvents. There are numerous large lipid vesicles in plasma and very small vesicles along cell membrane in pollen stored in dry condition. Against this, there are small lipid vesicles in plasma and no vesicles along cell membrane in pollen soaked in acetone and diethyl ether.

1. ··· pollen stored in dry condition for 1 year at −15°C (X4.000), 2. ··· pollen soaked in diethyl ether for 1 year at −15°C (X4.000), 3. ··· same as 1 (X10.000), 4. ··· pollen soaked in acetone for 1 year at −15°C (X10.000), 5. ··· same as 2 (X10.000), SL. ··· small lipid vesicles.

membrane (Fig. 1-1, 3). Against this, there were small vesicles in plasma and no lipid vesicles along cell membrane in the pollen grains that stored in acetone and diethyl ether (Fig. 1-4,5) Further, white vesicles that may be consist of polysaccharide disappered when pollens were soaked in acetone.

As shown in Table 1, pollen grains stored in dry condition germinated well and grew long pollen tubes and pollen grains stored in acetone and diethyl ether grew longer pollen tube than unsoaked pollens. As previously reported (1972a, 1973c), thus soaked pollens of *Camellia* and *Lilium* in various organic solvents grew longer pollen tubes than unsoaked ones and flower pollinated by the soaked pollens produced normal seeds.

Table 1 Growth of *Lilium auratum* pollens which had been stored in various conditions for 1 year at −15°C.

Condition	Germination rate (%)	Pollen tube length (mm)
in dry condition (unsoaked)	84.1	7.2
in acetone	68.4	8.5
in diethyl ether	84.8	8.6

It can be concluded from these observations that organic solvents diffuse into plasma of pollen grain through cell membrane. And, it is assumed that the organic solvents take out some free materials such as lipid vesicle from pollen grain, however, essencial materials for germination and pollen tube growth hard to be extracted.

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雑　　録

日本地質学第81年総会より

徳永重元幹事によれば、昭和49年9月1日～3日、北海道大学にて行われた同会で、下記の花粉学的発表があった。

飯田祥子・南関東における更新統の花粉分析

山形理他5名・琵琶湖底質の花粉学的研究

古市光信他2名・三豊層下部の花粉化石

佐藤誠司・花粉分析による北海道雨竜地域の第三系の層序の検討

黒田登美雄他1名・堆積環境と花粉組成の関係

新刊紹介「塚田松雄：花粉は語る（1974）」

（岩波新書・青版定価230円）

著者は現在第一線で国際的に活躍している花粉学者である。花粉分析を高知大学中村教授につき学び、さらに大阪市立大学大学院（修士・博士）で、その基礎となる花粉形態を私が指導した。ドイツのルドルフ教授のマツ属の二葉松亜属と五葉松亜属の区別（1934）とは別の私の区別法（発芽溝のソバカス文様のあるのが五葉松型、無いのが二葉松型）をミネソタ大学でライト教授を前にして説明した話など私には学者冥利の思い出も書いてくれてある。大阪市大を出てアメリカに渡り、アリゾナ大学・エール大学などをへて、現在ワシントン大学教授である。

花粉を主役・脇役として理解しやすく書いてある。そのスタートは花粉形態で、ゴールは花粉分析である。イネの歴史ではイネ科花粉の発見が、いつの間にかイネ花粉となり、さらにイネ栽培に変るのは形態的には物足りない。もっと花粉同定についてのキメテとか苦心も書いてほしい。しかしトウモロコシ（アメリカ原産）の歴史では、果実も出土するので理解しやすいが、アメリカインディアンのナバオ族がその花粉も食べる話も書いてほしかった。この原稿を書いている最中に、来日して東京にいる彼から私に電話をしてくれた。この話をすると彼もナバオの話は知っていたので、なお残念であった。その他糞や犯罪やマンモスと花粉の話など興味ぶかく述べている。

日本・ヨーロッパ・ユーラシア大陸などの先史時代からの話を花粉分析を中心として気候変化の歴史、農耕文化の発達などを分りやすく述べてあり、花粉学の入門書としても一読をすすめたい。副題は人間と植生の歴史である。（1974. 12. 25 第1版）。

（上野実朗）