Inoculation Experiments with Heteroecious Species of the Japanese Rust Fungi

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By

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1. Uromyces Alopecuri SEYM.

In the spring of 1931, the writer noticed an abundance of aecidia on Ranunculus glaber (Lév.) MAK. (R. Vernyi Franch. et Sav. var. glaber Nakai) in the College farm, where there was grown a number of Alopecurus fulvus L. attacked by Uromyces Alopecuri. From these field observations and the record made by Arthur in North America, the writer assumed that there might be a genetic relationship between the aecidia on Ranunculus glaber and Uromyces Alopecuri, and he carried out this experiment in order to determine whether such a connection does exist.

As an inoculum, the aecidiospores on Ranunculus glaber which were collected in the neighbourhood of the College on April 20, 1933 were used. On the next day, inoculations with those aecidiospores were made on Alopecurus fulvus and Agropyrum semicostatum NEES. Uredosori began to appear on Alopecurus fulvus on May 2, and teleutosori on May 22, while no sign of infection of Agropyrum semicostatum occurred.

By examining the uredo- and teleutospores produced on Alopecurus fulvus by cultures, it is certain that an aecidium on Ranunculus glaber is the aecidial stage of Uromyces Alopecuri.

2. Puccinia Eulaliae BARCL.

In July 1930, an abundance of the uredostage of this Puccinia was found on Miscanthus sinensis ANDERS., following an aecidium on Plantago major L. var. asiatica DECNE. growing with it at a valley in Inabayama near Tottori. No other Aecidium or heteroecious species of Puccinia was to be found in the vicinity.

These indications, also in the light of results obtained by Transz-
The present writer to assume that there might be a relationship between the aecidia on Plantago and Puccinia Eulaliae on Miscanthus sinensis.

**Experiment 1**—The aecidiospores on Plantago major var. asiatica collected at Amedaki (Okaya-mura), the province of Inaba on June 20, 1932, were inoculated on leaves of Miscanthus sinensis and Phragmites communis Trin. on June 22. On the inoculated leaves of Miscanthus sinensis, uredosori of Puccinia Eulaliae appeared in abundance on July 4, and its teleutosori on August 14.

**Experiment 2**—In this experiment, inoculations with the aecidiospores on Plantago major var. asiatica which had been collected in the neighbourhood of the College on May 2, 1933 were made on Miscanthus sinensis and Imperata cylindrica Beauv. var. Koenigii Honda. Uredosori of Puccinia Eulaliae began to appear only on Miscanthus sinensis on May 15, as in Experiment 1, while no sign appeared on Imperata.

### 3. Puccinia Magnusiana Körn.

The writer collected numerous aecidia occurring on leaves, petioles and stems of Ranunculus polycephalus Mak. (R. Vernyi Franch. et Sav. var. japonicus Nakai) at Ochidani Park, Tottori on June 16, 1932. Inoculations with those aecidiospores were made on leaves of Alopecurus fulvus and Phragmites communis on the same day. Twelve days after inoculation, uredosori began to appear only on Phragmites communis, and after 7 weeks teleutosori occurred on the same plant, while on another plant inoculations were unsuccessful.

From the characters of the uredo- and teleutosporal stages produced on Phragmites communis by cultures, it is determined with certainty that an aecidium on Ranunculus polycephalus is the aecidial stage of Puccinia Magnusiana.

### 4. Puccinia Phragmitis (Schum.) Körn.

The writer attempted to inoculate with aecidiospores from aecidia on leaves of Rumex japonicus Meisn. (R. crispus var. japonicus Mak.) on Phragmites communis.

Fresh aecidiospores on Rumex japonicus which were collected beside a bush of Phragmites communis that was badly attacked by Puccinia Phragmitis the previous season at Omokage-mura near Tottori on May 4, 1932, were sown on Phragmites communis on the next day. Uredosori of Puccinia Phragmitis began to appear on the inoculated leaves of Phragmites on May 15, and teleutosori on June 30.

Ito\(^3\) in 1909 made the first cultures with Japanese material, proving that the aecidial stage on *Rumex domesticus* Hartm. is related to the uredo- and teleutostage on *Phragmites communis*.

5. **Puccinia angustata** Peck.

In the spring of 1931, the writer noticed that aecidia occurred abundantly on leaves and stems of *Lycopus lucidus* Turcz. var. genuina Herd. forming a pure association along a stream in Ochidani Park at Tottori, where there was grown a number of bushes of *Scirpus Cyperinus* Kuntz. var. concolor Mak. This indication induced him to assume that there might exist a genetic connection between aecidia on *Lycopus lucidus* var. *genuina* and *Puccinia* or *Uromyces* on *Scirpus*, and to perform inoculation experiments with the aecidiospores from *Lycopus* upon *Scirpus Cyperinus* var. *concolor*.

On June 16, 1932, a large amount of *Lycopus lucidus* var. *genuina* was collected and used as an inoculum. On the next day, the aecidiospores from *Lycopus* were sown on leaves of *Scirpus* which was potted in the laboratory. Two weeks after sowing numerous uredosori began to appear on the inoculated surface of leaves, and they developed abundantly day after day. Four weeks after, teleutospores of *Puccinia* appeared on the same leaves.

From the characters of the uredo- and teleutospores produced on *Scirpus Cyperinus* var. *concolor* by cultures, it is certain that an aecidial stage on *Lycopus lucidus* var. *genuina* is that of *Puccinia angustata*.

6. **Puccinia Agropyri** Ell. et Ev.

Dietel\(^2\), Hennings\(^3\), Ito\(^5\), Yoshinaga and the writer\(^5\) and others identified an aecidium on *Clematis paniculata* Thunb., *C. apiifolia* DC. and *C. fusca* Turcz. from our country as the aecidial stage of *Puccinia Agropyri*. But, the aecidual connection of this species has never been proved with Japanese material up to the present.

In May 1933, the writer attempted to make inoculations with aecidiospores from *Clematis paniculata* on *Agropyrum semicostatum*. The aecidiospores from aecidia on leaves and petioles of *Clematis paniculata* which had been collected at Tottori on May 9, 1933 were used as an inoculum. Two days later the aecidiospores were sown on leaves of *Agropyrum*

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2) Engl. bot. Jahrb. 27 (1899), (568); 37 (1905), 102. p. (568), 1899.
3) Ibid. XXXI, p. 731, 1901.
semicostatum. Eight days after inoculation, a number of uredosori began to appear on the inoculated leaves of Agropyrum, and after three weeks, teleutospores developed on the same portion of the leaves.

By examining these uredo- and teleutospores which were produced on Agropyrum by the experiments, it is clearly determined that an aecidial stage on Clematis paniculata is that of Puccinia Agropyri.

7. Puccinia Nolitangeris CORDA.

In the vicinity of Sapporo, an aecidium which is identical morphologically with the aecidial stage of Puccinia Nolitangeris is commonly found on Adoxa moschatellina L. wherever the host plant is present in the early spring to June.

In June 1925, the writer made this inoculation experiment in order to ascertain whether the aecidiospores from aecidia on Adoxa moschatellina in the neighbourhood of Sapporo is really the aecidial stage of this species.

For the inoculating materials, aecidia on Adoxa were collected by the writer in Mt. Moiwa near Sapporo on June 1, 1925. On the next day, the aecidiospores were sown on leaves of Impatiens Nolitangere L. which was potted in the laboratory. Twelve days after sowing of the spores, uredosori of this species appeared in abundance on the inoculated leaves of Impatiens.

8. Pucciniastrum Kusanoi DIET.

Experiment 1—Teleuto-material of this species on the fallen leaves of Clethra barbinervis Sieb. et Zucc. was collected at Misasa-mura, province of Hōki on April 27, 1933. This material was placed in a moist chamber, and the teleutospores germinated freely in a few days. The leaves bearing the germinating teleutospores were then suspended on needles of Abies Mayriana, Miyabe et Kudo, Larix Kaempferi Sarg. and Picea jezoensis Carr. on May 2. On a number of the inoculated needles of Abies Mayriana, spermogonia were produced on May 12, and peridermia began to appear on May 21. The peridermia developed rapidly and were mature in a few days. No sign of infection on Larix Kaempferi and Picea jezoensis occurred.

Experiment 2—This experiment was carried out in order to determine the return infection using the aecidiospores from Experiment 1 as an inoculum. Fresh aecidiospores from Experiment 1 were sown on leaves of Clethra barbinervis, Styrax japonica Sieb. et Zucc. and Viburnum furcatum Bl. on May 25. Ten days after sowing of the spores uredosori appeared on Clethra barbinervis, while on Styrax japonica and
Viburnum furcatum the inoculations were unsuccessful.

Remarks—From these experiments, it is quite certain that the present species is heteroecious and must have its aecidial stage on Abies spp. The aecidial stage of this species obtained from the experiments may be described as follows:

Spermogonia amphigenous, subcuticular, lenticular to flattened hemispherical, 90–150 μ across, 45–60 μ high, honey-yellow in colour; spermatia oblong, 3.5–5.5 × 1.2–2 μ, colourless, smooth.

Aecidia hypophyllous, in two rows, one on each side of midrib, on yellowish discoloured portions of affected needles, cylindrical, 0.24–0.32 mm across, 0.5–2 mm long; peridia colourless; peridial cells rhomboidal or hexagonal, usually elongated vertically, 50–69 × 12–18 μ, overlapping, with outer walls thin, smooth, with inner walls thicker, verrucose; aecidiospores globose, subglobose or ellipsoidal, 18–25 × 13.5–18 μ; epispor 1.5–2 μ thick, colourless, closely verrucose; contents yellow in colour.

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