AUTOCYN IN PIOPHILA CASEI (DIPTERA, PIOPHILIDAE)

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Abstract. Two long-adapted laboratory strains of Piophila casei originating from Prague were found to be autogenous. However, protein-deprived females compared to protein-fed ones showed lower fecundity and fertility, and somewhat retarded oviposition extended over a longer period.

The cheese skipper, Piophila casei (Linne, 1761) is a fly endowed with numerous remarkable adaptations. Most of them probably either pre-determined its pronouncedly synanthropic way of life, or developed in the evolution of its synanthropy. Among them are properties favouring very early reproduction and thus contributing substantially to the rate of population increase: the nearly mature eggs in the ovaria at the moment of adult emergence (Bennettová and Zuska, unpublished), the ability of the adults to mate and oviposit soon after emergence (Bachmann 1918), and the preference of mating to feeding in very young males (Zuska, unpublished).

The aim of the present paper is to show that this complex of adaptations is augmented by another property: the autogeny of females, which secures early production of fertile eggs without dependence on protein in food of adults.

MATERIAL AND METHODS

Adult flies used in the experiments were obtained from our continuous laboratory cultures, which are kept in darkness at 25 ± 1 °C and about 80 % r.h., in discrete generations, and are fed on lean beef (Zuska, 1975a). Two highly inbred strains (Nos. 3 and 5) were used in each derived from a single fertilized female captured in a slaughterhouse in Prague and maintained in the laboratory since 1969 and 1970 respectively. Unmated and unfed adults emerged from isolated puparia were placed in shell vials (90 by 15 mm). Each vial, with one male and one female, contained about 1 ml of experimental food poured in a warm state onto the bottom and consisting of 40 % (w) D-glucose, 1.5 % agar, and either 58.5 % distilled water (food for protein-deprived flies—PD) or 1 % sodium caseinate and 57.5 % distilled water (food for protein-fed flies—PF). The environment within the vials was standardized by keeping these in glass containers with a saturated solution of KCl on the bottom, at 25 ± 1 °C, in darkness. If the male died sooner than the female, he was replaced by another, unmated male of the same age previously fed with the same food.

In experiment 1, a comparison was made of the percentage of females laying eggs, and laying fertile eggs, and between PD and PF flies. The test was planned so that two different strains of flies were also compared. Six groups of fly pairs were used. Groups 1 and 2 originated from strain No. 3, laboratory generation 117, groups 3 and 4 from strain No. 5, generation 102, and groups 5 and 6 (which were also the object of experiment 2 and were treated slightly differently — see below) from strain No. 3, generation 121. Odd groups were fed with PD food and even groups with PF food. Freshly emerged females were placed in the vials during the first day of their imaginal life and paired immediately with males of the same age; exceptionally, males one day older were introduced on the first day or males one day younger were added next day. After three days, the pairs were transferred into clean vials with fresh food of the same kind and the transfer was then repeated every seven days. The old vial was searched for eggs and, three days after transfer, for hatched eggs. A female was classified as "fecund" if at least one egg was found, and "fertile", when at least one of her eggs...
Table 1. Percentage of fecund and fertile females as influenced by strain and protein-content in food (Experiment 1)

<table>
<thead>
<tr>
<th>Group No.</th>
<th>Strain</th>
<th>Diet</th>
<th>Females (% of group) primiparous at age</th>
<th>Total of females (% of group)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>0-3 days</td>
<td>3-10 days</td>
</tr>
<tr>
<td>1</td>
<td>3</td>
<td>PD</td>
<td>9 (9) **</td>
<td>59 (55)</td>
</tr>
<tr>
<td>2</td>
<td>3</td>
<td>PF</td>
<td>35 (35)</td>
<td>62 (60)</td>
</tr>
<tr>
<td>3</td>
<td>5</td>
<td>PD</td>
<td>10 (10)</td>
<td>46 (45)</td>
</tr>
<tr>
<td>4</td>
<td>5</td>
<td>PF</td>
<td>28 (28)</td>
<td>60 (60)</td>
</tr>
<tr>
<td>5***</td>
<td>3</td>
<td>PD</td>
<td>10 (5)</td>
<td>35 (35)</td>
</tr>
<tr>
<td>6***</td>
<td>3</td>
<td>PF</td>
<td>15 (15)</td>
<td>85 (85)</td>
</tr>
</tbody>
</table>

* No females primiparous later. ** Percentage of fertile flies in parentheses. *** See also Table 2. **** One female fertile at consequent oviposition.

Table 2. Number of eggs and fertile eggs produced by females fed with protein-deficient and protein-containing diet (Experiment 2)

<table>
<thead>
<tr>
<th>Group No.</th>
<th>Diet</th>
<th>Egg-laying females (% of group)</th>
<th>No. of eggs produced per egg-laying female*</th>
<th>Total No. of eggs produced per egg-laying female</th>
<th>Fertile eggs: total eggs ratio</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>0-3 days</td>
<td>3-6.5 days</td>
<td>6.5-10 days</td>
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<tr>
<td>5</td>
<td>PD</td>
<td></td>
<td>10</td>
<td>30</td>
<td>15</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>3 (2)</td>
<td>86 (85)</td>
<td>97 (62)</td>
</tr>
<tr>
<td>6</td>
<td>PF</td>
<td></td>
<td>15</td>
<td>85</td>
<td>40</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>50 (50)</td>
<td>98 (91)</td>
<td>47 (29)</td>
</tr>
</tbody>
</table>

* Average No. of fertile eggs in parentheses, ** No eggs produced later
hatched. The pre-oviposition/pre-fertile-oviposition period of each female was noted and those flies which were once found fertile were then discarded. Those flies which laid sterile eggs continued to be observed (but, with one exception, never became fertile later).

Experiment 2 was arranged similarly, but its aim was to test possible differences in egg production between PD and PF flies. Freshly emerged females were introduced into the vials within the first day of their imaginal life, together with males always of the same age (groups 5 and 6). The pairs were transferred into clean vials with fresh food after three days and then every 3.5 days, and the eggs produced were counted; three days after the transfer the number of hatched eggs was recorded.

Both experiments were carried out under xenic conditions. D-glucose (pharmaceutic grade) was supplied by Lachema, Brno; agar (Japanese fibrous, Kobe No. 1) by Koospol, Prague; sodium caseinate was prepared at this institute at pH = 7 from casein acc. to Hammarsten supplied by Reanal, Budapest; distilled water was also prepared at this institute in glass apparatus.

RESULTS AND DISCUSSION

From the results of experiment 1 (Table 1) it is apparent that autogeny is a general property of the examined strains of *P. casei*, though differences between PD and PF females in egg production certainly exist. Thus the proportion of non-fecund females seems to be invariably higher in PD than in PF females. Fecund sterile females also occur more frequently in PD than in PF females, though this difference does not seem to be so evident. The most conspicuous dissimilarity between the two groups is that among PF females the occurrence of first oviposition starts, culminates and ends much earlier than in PD females. No significant differences were found between the two strains used.

The data obtained in experiment 2 (Table 2) suggest analogical conclusions. It is obvious that egg-laying is not only earlier in PF than in PD females, but also more productive within the first days. The average number of all eggs laid by a female, and the number of fertile eggs produced, is distinctly larger within the first week, the percentage of sterile eggs being very close to zero. In PD females egg-laying is more retarded and therefore less intensive, but the proportion of sterile eggs is also negligible within this period.

To summarize, it is obvious that even a relatively small amount of protein content in food distinctly influences the time and intensity of oviposition. However, this may not be due to the nutrient contribution of protein. The above mentioned advanced development of ovaria in a freshly emerged female suggests that the protein stimulates oviposition rather than egg production, this is an adaptation ensuring that a medium of adequate for the development of larvae is selected by the female. Nevertheless, though protein-deficient food causes fewer females to produce eggs and fertile eggs, and egg-laying females to produce fewer eggs and fertile eggs, the experiments show unequivocally that most females (and also males) of *P. casei* are able to reproduce successfully if deprived of protein in adult stage. In fact, the PD females appear to lay fertile eggs produced long after emergence. Thus, e.g. one PD female resumed fertile oviposition after one week’s interruption.

The decrease of fecundity and fertility in the ageing females of *P. casei*, which is clearly demonstrated by experiment 2, seems to be quite general in insects (Rockstein and Miquel 1973). However, in the females (at least in PF group) of *P. casei*, egg production is reduced abruptly after about the first ten days of imaginal life, which is hardly a consequence of mere ageing, but rather an indication of that the ovaries are genetically and hormonally rigorously directed toward early reproduction.

Of course, it must not be overlooked that autogeny is obviously at least partly controlled genetically (references in Engelmann 1970) and therefore liable to selection. The flies tested in experiments 1 and 2 originate from long-adapted laboratory strains and their autogeny may only be a feature of the general increase in
fitness due to the selective pressure of harsh laboratory conditions, just as it is or may be with increased radioreistance (Zuska 1973). It should be pointed out here that an exploratory experiment performed earlier (strain 5, laboratory generation 19) gave lower autogeny rates.

The lack of protein in the food of adults was in clear correlation not only with the retardation of oviposition, but also with the increase of life-span, which was observed in both experiments 1 and 2 and is known from other experimental situations (Zuska, unpublished).

The relatively short adult life-span of *P. casei* (Bachmann 1918, Simmons, 1927, Zuska 1973, 1975b, 1976), its ability to survive on purely glycicic diets and the strong dependence upon the concentration of the nutrient saccharide for longevity (Zuska 1975b), the ability to mate and lay fertile eggs very soon after emergence (Bachmann 1918, Zuska, present paper and unpublished), an abrupt decrease of fecundity after the first decade of the imaginal life, low production of viable eggs later in adulthood, little dependence of reproduction upon nutrition of adults including autogeny (present paper), and the tendency of the female toward monogamy (Grigolo et al. 1974), suggest the existence of evolutionary trends towards the following state:

(a) the vegetative functions of the adult are largely suppressed;
(b) reproduction begins and culminates soon after emergence;
(c) only the first gonotrophic cycle remains fully functional.

The occurrence of autogeny in *P. casei* is also quite interesting from the comparative physiology viewpoint. Autogeny often occurs among Diptera. However, not many muscid flies are known to be autogenous and only very few acalyptrate flies were found to possess this ability.

**CONCLUSIONS**

1. Most females (of two long-adapted laboratory strains) of *P. casei* are autogenous.
2. Protein-deprived females show lower fecundity and fertility than protein-fed females: fewer females are fecund and fertile; fecund females lay fewer eggs and the proportion of sterile eggs is higher.
3. Protein-deprived females compared to protein-fed ones show somewhat retarded oviposition extended over a longer period.
4. Males fed with food devoid of protein as adults are fertile.
5. The nutrition of adults is probably of only very limited importance for reproduction and the protein contained in their food may stimulate oviposition rather than support egg formation.

**Acknowledgement.** The extensive technical help of Mrs. E. Čadková is gratefully acknowledged.

**АВТОГЕНЕЗ У PIOPHILA CASEI (DIPTERA, PIOPHILIDAE)**

Я. Зуска.

Резюме. Был установлен автогенез у двух штаммов *Piophilica casei*, происходящих из Праги и держащих продолжительное время в лаборатории. Однако, самки, кормленные пищей без белков, откладывали более низкое количество яиц и более низкое количество оплодотворенных яиц. Их откладка происходила позднее и продолжалась более долгое время, чем у самок, кормленных пищей с белками.
REFERENCES


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A NEW CESTODE ENIOCHOBOTHRIUM TRYGONIS SP.N. FROM TRYGON SEPHER

Four specimens of a new cestode belonging to the genus Eniochobothrium Shipley et Hornell, 1906 were recovered from the spiral valve of Trygon sepheph at Ratnagiri, India. It is identified as Eniochobothrium trygonis sp.n.

Eniochobothrium trygonis sp.n.
Host: Trygon sepheph. Location: Spiral valve. Locality: Ratnagiri, Maharashtra, India.
Type specimens: Collected on 16th April 1973, deposited in Cestodology Laboratory, Department of Zoology, Marathwada University, Aurangabad. All measurements are given in millimeters unless otherwise mentioned.
Description: (based on 4 specimens). Worms very small, not visible with naked eye, measuring 1.56 in length, divisible in four parts: scolex, anterior segments, middle segments and posterior segments. Total number of segments in the described specimen is 29. External segmentation quite distinct.
Scolex elongated, oval with four very movable, rounded suckers, each 0.09 in diameter and a rostellum slightly conical at apex, 0.06 in diameter, without any crown or circle of hooks. A short but wide neck is present.
Second part of 12 segments, gradually increasing in breadth with length almost constant. Posterior margin of each segment projects to form an acute cone in the middle.
Nine segments constitute third part with constant width and slight increase in length. Segments measure 0.03 in length and 0.27 in width. Last two to three segments slightly smaller. Segments of imbricate types with posterior concave borders. Imbrication less in posterior segments.
Posterior fourth part with remaining eight segments. Length of segments increasing rapidly. Last segment measures 0.28 in length and 0.13 in width.
Eight testes 0.02 in diameter, arranged in circle in the centre of the segment. The two circular bodies which are situated behind testes resemble ovary. No isthmus observed. These are smaller in size compared to testes measuring 0.009 × 0.008 and 0.01 × 0.008. Other parts are not seen.
The vitelline follicles are small, rounded bodies, externally bounded by lateral margins of segments and internally by excretory ducts. Clear two excretory ducts present running longitudinally.
Eniochobothrium trygonis sp.n. differs from E. gracile (Shipley et Hornell, 1906) in total length, and in number of segments of second, third and fourth regions (1.56 against 5.12) (12, 9 and 8 against 18, 18 and 8). The third part of E. gracile is very narrow, differing in width from E. trygonis. Suckers of E. trygonis are very large in size compared to E. gracile. The last