Russian Entomol. J. 10 (4): 433–436 © RUSSIAN ENTOMOLOGICAL JOURNAL, 2001

Population homeostasis and sexual behavior of *Archips podana* (Scop.) (Lepidoptera: Tortricidae)

Гомеостаз популяции и половое поведение у *Archips podana* (Scop.) (Lepidoptera: Tortricidae)

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KEY WORDS: polymorphism, reproductive behavior, homeostasis of population, leafrollers.

КЛЮЧЕВЫЕ СЛОВА: полиморфизм, репродуктивное поведение, гомеостаз популяции, листовертки.

ABSTRACT: At the population level the polymorphic species *Archips podana* Scop. (Lepidoptera : Tortricidae) maintains homeostasis between insects-specialists and generalists by the reproductive behavior. Inbreeding leads to a drop in the quantity of the fertilized females. Males of only one, ecologically more specialized genotype P^2P^2 (phenotypes B and 0) appear in F_1. Females begin to attract some number of males without spines on the aedeagus (penotype 0). However the sexual behavior of males of phenotype 0 is close to the behavior of males of the opposed genotype P^1P^1 (phenotype A). As a result, at the population level, the probability of fertilization of females by males of the opposed genotype rises. In the filial generation the quota of heterozygous individuals (P^1P^2) which characterize insects-generalists increases.

РЕЗЮМЕ: На популяционном уровне полиморфный вид *Archips podana* Scop. (Lepidoptera: Tortricidae) поддерживает гомеостаз между насекомыми- специалистами и генералистами с помощью репродуктивного поведения. Близкородственное скрещивание приводит к снижению количества оплодотворенных самок. В F_1 появляются самцы только одного, экологически более специализированого генотипа P^2P^2 (фенотипы Б и 0). Самки начинают привлекать некоторое количество самцов без зубцов на эдеагусе (фенотип 0). Однако половое поведение самцов фенотипа 0 близко к поведению самцов противоположного генотипа P^1P^1 (фенотип А), в результате чего на уровне популяции повышается вероятность оплодотворения самок самцами противоположного генотипа. В следующем поколении возрастает доля гетерозиготных особей (генотип P^1P^2), характеризующих насекомых-генералистов.

Introduction

In articles on population phenetics the question of the homeostasis of polymorphic populations and mechanisms of its support often arises [Imasheva, 1998; Novozhenov & Mikhailov, 1998]. As it is affirmed in R. Robinson's survey [1971], nonpolymorphic species of moths do not exist. It may also be connected with the fact that polymorphic populations use the environment more fully than monomorphic ones [Novozhenov, 1997].

Numerous investigations on the model polymorphic species, the apple surface eating tortricid *Archips podana* Scop. (Lepidoptera : Tortricidae), have led to the identification of four morphological forms of the sex apparatus for both males and females [Safonkin & Kulikov, 2001]. Males differ in the number and location of spines on the aedeagus (symbols: A — a spine is on the end of the aedeagus, B — a spine is on the side, A+B — both spines are on the aedeagus, 0 — no spines). The gene, which controls this trait, is in one of the autosomal linkage groups. Homozygous males with genotype P^1P^1 have one spine A, P^2P^2 — one spine B, while heterozygous ones (P^1P^2) have both spines. Morphological type 0 is characterized by the absence of the spine in phenotype B. The connection has been shown between the gene, responsible for the phenotypic display of the structural trait of the sex apparatus, with a complex of characteristics which include ecology and sexual behavior of individuals. For females, the presence of cuticular outgrowths on the external side of the antrum has been noted [Safonkin, 2000a]. The outgrowths differ in location and number (symbols: C — an outgrowth is in the center, L — an outgrowth is on the side, D — two outgrowths, Z — absence of outgrowths). Possibly, this trait is under the polygenic control.

The purpose of the present investigation was to study one of the possible mechanisms of maintaining polymorphism of populations at the level of the reproductive behavior.

Methods

The study was conducted in the suburbs of Moscow within 1997–2000. At the beginning the larvae of *Ar-*

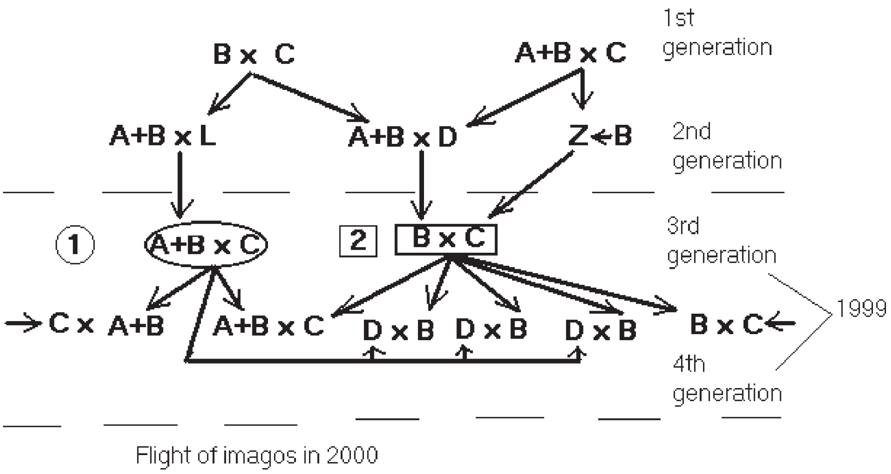

Fig. 1. Scheme of imago crossings of different phenotypes in the two lines of *Archips podana* Scop. under the experiments of 1997–2000. B, A+B, C, L, D, Z — phenotypes of males and females; 1 and 2 — numbers of the lines under study; arrows — imagos-progenies from the stated type of crossing. The remaining explanations are in the text.

Рис. 1. Схема скрещивания имаго разных фенотипов в двух линиях по экспериментам 1997–2000 гг. Б, А+Б, C, L, D, Z — фенотипы самцов и самок, 1 и 2 — номера исследуемых линий; стрелки — имаго-потомки от данного типа скрещивания. Остальные объяснения в тексте.

chips podana and those of filial generations, gathered in an apple orchard, were fed on branches of different fruit trees. Imagos, after emergence, were put into Petri dishes in pairs and kept there till their death. The larvae, which emerged in the same Petri dishes, were put on the branches of fruit trees in capron cages. Not less than 3-5 leaves of a branch were allotted per larva of the 2nd instar. The posterity from each imaginal pair was reared separately. For wintering the diapausing larvae were transferred in the capron cages to the level of the ground litter in the orchard.

The attractiveness of virgin females was determined by placing them in the apple garden. For this, each female was at once put into a cylinder cage of 4 cm di and 6 cm h, with a cell of 2x2 mm. Each cage with moistened cotton wool on its bottom was, in each turn, put into a standard sticky trap "Attracon—A". Collection of the attracted males was made within the first three days of the female's life, after which it was transferred into a Petri dish for coupling with some male.

Preparations of genitalia for determining a morphological type were made from all imagos after their death.

Results

I. The scheme of conducting crossings

The scheme of experimental crossings is given in Fig. 1. The scheme is based on the supposition mentioned earlier that the complex of the adaptive traits under consideration and connected with a phenotype is transmitted through the male genotype [Safonkin & Kulikov, 2001]. It is seen that in line 1, contrary to line 2, all crossings of females were made with the heterozygous genotype of males (P^1P^2). In line 2 in 1998, with crossing a male with phenotype A+B and a female with phenotype D (from a heterozygous male) in the F_3 generation, a male of genotype P^2P^2 was selected and crossed with a near-related female. In the 4th generation the crossings of males and females of lines 1 and 2, as well as of females from other lines, were carried out.

II. The emergence of imago of different lines

Since *Archips podana* can have two generations a year in the region near Moscow, its development to the imago stage depends much on the duration of the photoperiod. Without additional daylight at the beginning of July, most larvae of the spring-summer generation (3rd in Fig. 1) go into a diapause in the third instar, passes the winter, finishes eating in the spring of the following year, and the flight of the imago takes place at the beginning of June. The larvae of the autumn generation (4th in Fig. 1), which emerge in August, also reach the third larval instar quickly and leave for wintering. So, the attraction of females of the 3rd and 4th generations in the experiment conducted was studied in the summer of 2000.

III. Characteristics of female attraction

The analysis of the female attraction of the 4th generation has shown that the ratio of phenotypes among the males trapped does not differ. For the 3rd generation, with an external similarity of the pattern of catches, one can see principal differences in the female attraction of the two lines. Criterion χ^2 shows a trustworthy difference in the ratio of male phenotypes ($\chi^2 = 11.29$, df=3, $p<0.05$). In the second line females began to attract males of phenotype 0, therefore the quota of the genotype P^2P^2 (phenotypes B and 0) does not differ from the average one. In the first line, unfortunately, only one

Table 1. Attractiveness of females of the 3rd and 4th generations of the two lines in the field experiment of 2000.

Таблица 1. Аттрактивность самок 3-го и 4-го поколений двух линий в полевом эксперименте 2000 г.

Line No., generation	Number of males per female	Phenotypes of males, %			
		A	B	0	A+B
1					
3rd gen. (1)	52	55.77	17.31	0.0	26.92
4th gen. (4)	69.33±24.68	36.06	19.71	0.0	44.23
2					
3rd gen. (7)	54.28±15.19	42.57	15.26	3.21	38.95
4th gen. (10)	49.00±15.41	35.58	18.61	0.0	45.81

The number of females is shown in parentheses.

Table 2. Phenotypic diversity of the two investigated lines of *Archips podana* (according to the results of 1999–2000).

Таблица 2. Фенотипическое разнообразие двух исследуемых линий всеядной листовертки (по результатам 1999–2000 гг.).

Line No.	Quantity of imagos in the filial generation							
	Male phenotypes				Female phenotypes			
	A	B	0	A+B	C	L	D	Z
1	8	1	0	6	10	0	6	0
2	0	32	1	0	15	7	13	0

female survived after the diapause. It is, therefore, not possible to make a trustworthy comparison between the lines. However it has been noted that an increase in the quota of males type A takes place at the expense of a decrease in the quota of males of type A+B.

IV. Analysis of the posterity from the 3rd generation

Data on the structure of the filial generation from males of the first and second lines in the 3rd generation are stated in Table 2. The table contains the summary quantities of phenotypes of imago, i.e. emerged both in the autumn of 1999 and in the spring of 2000.

The peculiarity of the inbred posterity from crossing a phenotype B male and a female C of the second line, besides the presence of phenotype B males only, is that there were also one phenotype 0 male and one hermaphroditic pupa perished at a later stage of the development.

V. Sexual activity of imago

The activity of males of *Archips podana* was analyzed in an earlier work [Safonkin & Kulikov, 2001].

Table 3. Success of the reproductive behavior of males and females of *Archips podana* Scop. under the experiments of 1999–2000 in crossings between different lines and inside one line.

Таблица 3. Успех репродуктивного поведения самцов и самок всеядной листовертки по экспериментам 1999–2000 гг. в скрещиваниях между разными линиями и внутри одной линии.

Male phenotype	Crossing success	Between the lines				Inside one line			
		Phenotype of a female *							
		C	L	D	K_1	C	L	D	K_2
B	+	5	2	4	2.7	1	1	0	0.4
	–	3	0	1		4	0	1	
A+B	+	2	3	0	1.7	1	1	1	0.7
	–	1	2	0		1	1	2	

* phenotype Z in the posterity of these lines has not been discovered. K1 — quota of the fertilized females in crossings between the lines, K2 — quota of the fertilized females in crossings inside the line.

Males of phenotype B have a higher activity, therefore the number of successful couplings of this phenotype (by the presence of spermatophore in the copulatory pouch of the female), to the unfertilized, is 2.6, while A+B has 1.9 and A — only 0.6.

The success in coupling males and females between the lines in the experiment is close to the average observations of many years, while this process between individuals inside one line is reduced sharply (Table 3). The estimation of the compliance of the success in the above process between individuals of different lines and inside one line by criterion χ^2 shows a difference both as a whole between successful and unsuccessful couplings of individuals in two versions of the experiment and for males of phenotype B ($\chi^2 \geq 7.61$, df = 1, $p<0.01$).

Discussion

As it was shown by us earlier [Safonkin & Kulikov, 2001], males of phenotype 0 are constantly met in the investigated populations over the area in the quantity of 1-2% out of the caught males. Males of this phenotype lack the morphological trait of phenotype B but have the same genotype P^2P^2. However, as to behavioral characteristics, namely the flight to synthetic analogues of the pheromone of a female, males of phenotype 0 are closer to the behavior of phenotype A males. It is characterized by the fact that males of phenotypes A and 0 were caught in a greater number by traps in which, as a lure, an attractant with cis-11-tetradecenol in amount of 0.03–0.2 mg per 1 mg of the main components of cis-11/trans-11-tetradecenyl acetates was used. At the same time, males of phenotype B flew better to the attractant in which cis-11-tetradecenol amounted only to 0.005 mg per 1 mg of the main components. It may mean that males with a reduced morphological trait in the structure of genitalia compete less with males of their own phenotype B. So it is known that *Anagasta kuhniella* [Robinson, 1971] has a gene which lowers the viability but, possibly, changes the short-distance behavior of individuals. Because of differences in the rate of development, imagos with the changed genotype prevail at the beginning and at the end of the flight period of the generation. At the population level it means that a selective coupling of imagos will show itself to a definite level. As to *Archips podana*, males of phenotype A are less capable of a successful coupling with females of all phenotypes. Therefore the flight of a small quantity of more active males of phenotype 0 to the pheromone, which to a greater degree attracts males of phenotype A, raises the probability of leaving posterity because of lessening the competition between individuals.

The analysis of the distribution of the phenotypes in the filial generation in the two investigated lines of the leafroller has shown that, as a result of the near-related crossing, the third generation of line 2 has become more homozygous, as to the trait inherited, than the posterity of line 1. It has been confirmed by two facts: firstly, the appearance of a phenotype 0 male, besides those of phenotype B, and a hermaphroditic individual in the posterity; secondly, the attraction of males without the lateral spine on the aedeagus (phenotype 0) by females. Moreover, with the inbreeding a significant decrease in the number of successful couplings takes place. So, the inbred crossing in this species leads to negative consequences, especially for the following generation.

There is an optimum of the genetic diversity in natural populations, the deviation from which to any side leads to unfavorable biological consequences [Altukhov et al., 1996; Altukhov & Blank, 1999]. As to *Archips podana*, it is known that females of different phenotypes may differ in the composition of the pheromone secreted [Safonkin, 2000a]. With the inbreeding, a paradoxical situation occurs. The appearance of phenotype 0 males in traps with females shows that females of all phenotypes began to attract, to a greater degree, males of genotype P^1P^1 opposed to the paternal genotype P^2P^2. As a result, there arises the probability of appearance of the heterozygous genotype P^1P^2 in the posterity of all individuals of the population.

We also wish to note another aspect of this problem. In the population phenetics the conception of the adaptive behavior is known, which postulates that within one species there can be both groups of individuals-generalists and those of individuals-specialists [Sergievsky, 1988; Via, 1990]. For *Archips podana*, phenotypes A and B mark individuals-specialists, while A+B marks individuals-generalists [Safonkin, 2000b]. The ratio between these groups depends on environmental factors and, as to time, is rather stable, the heterozygous phenotype A+B being a buffer system of the population.

At all stages of the vital activity in the posterity of individuals of *Archips podana* one can discover competition between different forms: in imago, in the process of the reproductive behavior; at an egg stage, the imprinting may take place since individuals whose morphology corresponds to the paternal phenotype, particularly for genotype P^2P^2, prevail among the larvae emerged; and, indirectly, in larvae in the process of feeding on different plants [Safonkin & Kulikov, 2001]. Consequently, there is a tendency to a constant appearance of homozygous genotypes P^1P^1 or P^2P^2, i.e. of insects-specialists. But S. Kelley and B. Farrell (1998) put a question: "Is specialization a dead end?" Investigating the population of *Archips podana* from this position, we wish to note that the main function of imago is to leave a posterity. For moths, including Tortricidae, this function is mainly defined by a success in the reproductive behavior on the basis of sex pheromones. Therefore, the base line for homeostasis of a polymorphic population is introduced just through the mechanism of selecting a sex partner.

So, for the population of the leafroller under study the answer to the question put in the scientific literature is affirmative, since at the population level "the support" of generalists takes place.

ACKNOWLEDGEMENTS. This work is granted by the Russian Fund for the Fundamental (Basic) Researches (RFFI Grant 00-04-48756).

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