Esterase polymorphism in several populations of the two-spotted spider mite, Tetranychus urticae Koch

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Summary. Esterases of 7 field populations and 2 laboratory strains of the spider mite were analyzed by means of disc electrophoresis on polyacrylamide gel. Several different esterase patterns were found among these populations, and in some representative populations and strains high intrapopulation variability was also revealed. One of the laboratory strains was found to be almost homogeneous in this respect.

The two-spotted spider mite, Tetranychus urticae, an economically important pest, has haplo-diploid sex determina-tion (arrhenotoky). Several authors 1-3 have postulated that low genetic variability should result from arrhenotoky in comparison with ordinary diploid sex determination. A relatively low level of biochemical variability has indeed been reported for the honey bee, a classical example of arrhenotoky⁴. For *T. urticae*, high interpopulation but low intrapopulation variability was deduced^{3,5,6}. Nevertheless, there are some indications that significant intrapopulation variability actually exists. The variability in the induction of diapause⁷ and in the sex ratio⁸ can be adduced as examples. Also, although selection for insecticide resistance should theoretically be slow9, a high adaptability to pesticides being used in the field does not confirm this supposition⁶. This discrepancy has been noticed by several authors^{6,10}, but detailed analysis has not been done yet. The aim of this work was to study inter- and intrapopulation variability of nonspecific esterases in several field populations and laboratory strains of the spider mite by means of gel electrophoresis. Esterases are supposed to be directly connected with insecticide resistance. Moreover, their activity was sufficiently high for analyses of single females. No other enzymes were studied.

Experimental. Active adult females were used in the experiments. They originated from 7 local populations from southern (CB1, CB2, CB3) and western Bohemia (PR1, PR2, PR3, PR4). Mites were collected from Lamium album (CB2, PR1, PR3, PR4), Urtica dioica (CB1), Malva neglecta (PR2) and Phaseolus vulgaris (CB3). Both laboratory strains MH and ST were derived from populations collected from hops in central Bohemia 2 years ago and then reared on beans (16/8 L/D period at 24 °C). The mites of other local

populations were also transferred to beans. The samples for study of interpopulation variability were prepared from 30 homogenized females; for intrapopulation variability the homogenates from single females were used. The mites were homogenized in 40% (w/v) sucrose with 1% Triton X-100. The electrophoreses were performed by the method of Williams and Reisfield¹¹. The concentration of acrylamide in gels was 5.5%. The gels contained Triton X-100 (concentration 0.2%). Spacer gel was omitted. 50 µl of the homogenate of one female or its aliquot was applied on each gel. The current density of 2 mA/gel was used. Esterases were visualized by staining for 40 min with 0.56% (w/v) solution of 1-naphthylacetate in 0.2 M phosphate buffer, pH 6.0, which contained 0.2% (w/v) Fast blue RR salt and 2% acetone. Gels were then scanned at 480 nm with a densitometer (accessory of Pye Unicam SP8-100 spectrophotometer). The esterases, which appeared as peaks on electropherograms, were further investigated.

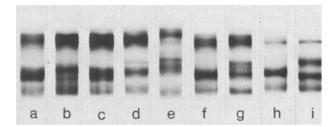


Figure 1. The esterase patterns of spider mites of field populations (a-g) and laboratory strains (h, i). a, PR1; b, PR2;, c, PR3; d, PR4; e, CB1; f, CB2; g, CB3; h, MH; i, ST.

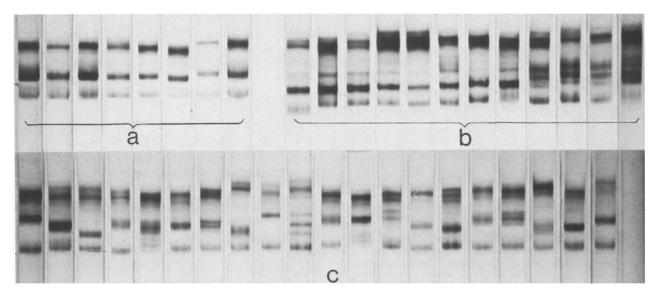


Figure 2. Some typical esterase patterns of single spider mite females of laboratory strains MH (a) and ST (b) and field population CB1 (c).

Results and discussion. Characteristic esterase patterns of mites of several local populations and strains can be seen in figure 1. These patterns are typical for each population, i.e. they are the same for groups of females of a population living on various parts of the plant. The influence of the plant on the esterase pattern of mites seems to be unimportant. The data presented give evidence of different esterase patterns in various populations of spider mites¹². They also confirm the assumption by other authors that interpopulation variability is high^{3,5,6}. In order to verify their second assumption - low intrapopulation variability - single females of the following populations and strains were examined (number of females in brackets): CB1 (90), CB2 (34), MH (21) and ST (45). 14 esterase zones of different mobilities were found. They were marked E₁-E₁₄ (E₁ possessed the highest mobility). The relative mobilities of esterase zones were related to E₁. The maximum and minimum number of esterases that we observed was 7 and 3, respectively. All females of the strain MH had only 3 esterase zones. In CB1, females with 3 esterase zones were found in 51% and in CB2 and ST in 15% and 18%, respectively. Females with 4 esterase zones were the most numerous in ST (47%), then in CB2 (24%) and CB1 (20%). Females with 5 esterase zones were present in CB2, ST and CB1 by 50%, 36% and 23%, respectively. The gels with esterases from single females of MH, ST and CB1 are shown in figure 2. The populations CB1 and CB2 were very heterogeneous - only a few females had identical esterase patterns. The strain ST was far less heterogeneous and the strain MH was almost homogeneous in this respect. The esterase heterogeneity of spider mite populations was examined in September and October when the mites were present in large numbers on plants and the highest variability was supposed to occur¹³. The results presented here argue against conceptions of intrapopulation uniformity of spider mites. The importance of genetic heterogeneity is clear - it guarantees high adaptability to changing

conditions of the environment¹⁴. And actual heterogeneity, as here observed, correlates well with practical experience as regards the ability of spider mites to develop resistance to almost any pesticide. In accordance with the data of some authors 10 we found by contrast a low variability in our laboratory strains of spider mites. But on the whole our findings suggest that even in the presence of a restrictive mechanism such as arrhenotoky, a high level of genetic heterogeneity is somehow maintained, as exemplified by the case of two-spotted spider mites. While esterase polymorphism is possibly related to insecticide resistance, this model may nevertheless become informative as to the maintenance of genetic variability under restrictive conditions of reproduction, like parthenogenesis.

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Effect of reserpine, 5-hydroxytryptamine and endocrinological manipulations on ovarian maturation of a marine

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Summary. An increase was observed in the ovarian index in Scylla serrata following administration of reserpine (a neuroleptic), but there was no change after 5-hydroxytryptamine administration. The releasing and inhibiting mechanisms for reproductive hormones of S. serrata are discussed.

The presence of catecholaminergic and serotonergic neurons has been reported in the eyestalk and other ganglia of crustaceans^{1,2}. The biogenic amines are known to trigger the release of neurohormones in the vertebrate hypothalamo-hypophysial system³ in insects⁴ and in the crustaceans^{1,5}. Fingerman et al. have shown that in the crab, *Uca* pugilator 5-hydroxytryptamine (5-HT) releases red-pigment-dispersing hormone from the eyestalk. Recently Farooqui⁶ reported that reserpine (RSP) (a monoamine depleter) brings about ovarian maturation in the sexually quiescent crab, Scylla serrata Forskal (Crustacea, Decapoda) probably by either inhibiting and/or stimulating the release of hormones via depletion of monoamines from their storage sites. The present study describes the effect of RSP (a monoamine depleter) and 5-HT on female S. serrata subjected to different endocrinological manipulations,

with the object of understanding the stimulatory/inhibitory mechanisms of a gonad-inhibiting hormone (GIH) from the eyestalks and a gonad-stimulatory hormone (GSH) from the brain and the thoracic ganglion.

Material and methods. Female crabs of the species Scylla serrata were collected from the Karla backwaters, Ratnagiri, West coast of India. They were acclimated to the laboratory conditions for about 1 week before the start of the experiment. Crabs of approximately equal weight (110-115 g) and size (70-72 mm carapace width) were selected for the study. Besides weight and size, the ovarian color was recorded by making a hole in the carapace at the start of the experiment. The holes were sealed after applying antibiotics with araldite adhesive (Ciba, India) as described earlier⁶. In S. serrata the immature ovary is white colored, whereas the mature ovary is orange-red in color⁶.