SHORT COMMUNICATIONS

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VARIABILITY OF FLUCTUATING ASYMMETRY IN OVARIOLE NUMBER OF *DROSOPHILA SUBOBSCURA* CAUSED BY MICROCLIMATIC DIFFERENCE. **Marija Savić[1], Aleksandra Patenković[2], Marina Stamenković-Radak[1,2],** and **M. Anđelković[1,2]** . [1]*Institute of Zoology, Faculty of Biology, University of Belgrade,* 11000 Belgrade, Serbia; [2]*Siniša Stanković Institute for Biological Research,* 11060 Belgrade, Serbia

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Numerous morphometric traits in insects are often subject to natural selection and related to fitness in different degrees. Variation of morphometric traits can be studied as intra-population variability between individuals and traits (Anđelković, 1983; Imasheva, 1999) or as inter-population variability (Soule, 1982; Coyne and Beecham, 1987), which reflects the source of variation. Variable environmental conditions, particularly temperature, can cause change in the variability of some morphometric traits in *Drosophila* (Imasheva et al., 1998).

Ovariole number is an important life-history character related to reproductive success in *Drosophila*. Positive association with female fecundity has been found in *D. melanogaster* (Cohet and David, 1978; Boeletreau-Merle et al., 1982; Delpeuch et al., 1995). Ovariole number exhibits long-range latitudinal clines (Lemeunier et al., 1986.; Capy et al. 1993.; Delpeuch et al., 1995), which may constitute evidence for adaptivity of this trait. Variability in ovariole number is associated with abiotic environmental factors (such as temperature), as well as with some biotic factors (like nutrition).

Fluctuating asymmetry (FA) estimates the imprecision with which bilateral traits are determined during development. It is a specific aspect of phenotypic plasticity and a parameter of phenotypic variability. A number of studies argued that the variation between bilateral symmetrical traits may be a useful early indicator of the effect of genetic or environmental stress (Zakharov, 2004). Fluctuating asymmetry may also serve as a relevant and sensitive indicator of population gene pool adaptatation to new environmental challenges (McKenzie and Yen, 1995). Thus, it is of interest to investigate possible asymmetry differences which may be a result of microclimatic differentiation in habitat. Here we report the results of a study of variation in fluctuating asymmetry of ovariole number in two populations of *Drosophila subobscura*.

Flies were collected in 2006 in oak and beech woods on Mt. Goč in Serbia (NE exp. with alt. of 875 m, E exp. with alt. of 787 m) and isofemale lines were established in the laboratory. Ovarioles were counted in both ovaries on 8-day-old virgin females, one from each isofemale line, in the F1 generation. Dissections were done under a binocular microscope in Ringer`s solution for insects (130 M NaCl, 5 mM KCl, and 1x4 mM $CaCl_2$). The adult *Drosophila* female has paired ovaries, each with a variable number of ovarioles, and can be counted without any measurement error (Moreteau et al., 2000).

Fluctuating asymmetry (FA) was statistically analyzed according to Palmer and Strobeck (2001). The Kolmogorov-Smirnov and χ^2 non-parametric tests were used to test departures from normality. Non-significant signed right-left (R-L) size differences showed that directional assimetry (DA) is absent in our samples, which suggests that bilateral variations were mostly a consequence of fluctuating asymmetry. Multiple regression analysis showed no significant correlation between $|R-L|$ and $(R+L)/2$. The FA_4 index was used as the variance of the difference between right and left ovariole number. It is efficient in estimating between-side variations and is not biased by directional asymmetry (DA). The F-test was used to compare variabilities in the value of FA_4 between the populations.

For 38 females from the beech wood population the mean ovariole number was X±S.E.=53.34±4.18, while for 28 females from the oak wood, it was X±S.E.=54.79 ± 5.50.

This difference between the two populations was not significant ($t = -0.213$, $df = 64$, $P = 0.832$). There were no significant differences between variances ($F = 1.279$, $P = 0.482$). Inter-population analysis showed that females from the beech wood have a significantly higher FA index for ovariole number than those from the oak wood ($F = 3.147$, $p < 0.01$).

Our previous studies showed that *D. subobscura* lines derived from oak and beech woods differ with respect to their reaction to genetic stress, i.e., inbreeding (Rašić et al., 2007). The obtained inter-population differences in developmental stability of wing size as a response to inbreeding indicate the presence of some population specificities with respect to mechanisms that buffer perturbations in developmental processes. The results of our study indicate that similar mechanisms are involved in preserving asymmetry in ovariole number in these two populations of *D. suboscura.* The optimal laboratory temperature for *D. suboscura* is 18-19°C (Krimbas, 1993), and development is significantly slower below this temperature. This species generally prefers moist habitats around optimal temperatures. It seems that the influence of lower temperatures during development of flies is longer in the beech wood, although this habitat is characterized by less variable ecological conditions than in the oak habitat. In the literature, there is also some evidence for significant increase of FA at low temperatures in *D. melanogaster* (Delpeuch et al., 1995).

Stress response traits are important determinants of the survival capacity of species under changing global climate conditions and in the presence of localized pollution. Given that, monitoring microevolutionary change can be a way of studying the effects of global change, and using *Drosophila subobscura* as a model system seems promising (Rodriguez-Trelles et al., 1996, 1998; Stamenković-Radak et al., 1999).

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