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ESF – ThermAdapt – Final Report

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Stay

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„ Latitudinal variation and costs of thermal tolerance in the Yellow Dung Fly“

Project outline

Investigations concerning the response of ectotherm species to thermal stress have focused on survival rates and the impairment of locomotion. Yet, it is equally or even more important to assess the impact of thermal stress on reproductive traits as these are generally affected by less severe conditions and may therefore be evolutionarily even more relevant (Jørgensen et al. 2006). This may involve fitness

costs resulting from an increase in the expression of stress proteins such as heat shock proteins hsp (Silbermann & Tatar 2000; Sisodia & Singh 2006). In this project I investigated systematic variation in thermal tolerance, the associated expression of hsp, and fitness-related costs across latitudinal populations of the yellow dung fly (*Scatophaga stercoraria*: YDF), focusing on male reproductive success.

The project was aimed at answering the following questions:

(1) Is the ability of males to compete over access to females affected by exposure to high (sublethal) temperatures, and is this influenced by a previous hardening treatment?

(2) If so, do these effects differ between populations of different latitudinal origin?

(3) Is the expression of hsp70 affected by temperature stress and latitudinal origin? Is there a correlation between male heat shock response and their competitive ability?

Experimental design

After exposure to an acclimation temperature (i.e. hardening; 32°C for 1h) male flies originating from two Spanish and two Scandinavian populations were exposed to high temperatures (i.e. 35°C for 25 min, 16h after hardening), resulting in four treatment groups: (1) non-hardened and non-stressed, (2) non-hardened and stressed, (3) hardened and non-stressed, (4) hardened and stressed. Mating success was assessed in a mating competition experiment in which four males, one of each treatment group, were competing for one randomly chosen virgin female, starting one hour after the stress application to allow for recovery. Ten randomly chosen males of each treatment group (that did not enter the competition trial) were used for the parallel measurement of hsp70 expression and were frozen at –80°C 1h after the stress treatment (resulting in a total of 160 flies).

Main results

(1) There was no significant effect of the applied temperature stress or previous hardening on the copulation success of males (nominal logistic regression on binary data, all $p > 0.22$). Thus, short-term heat stress does not impair the ability of males to successfully compete for females.

(2) This was true for all populations, i.e. males of different populations did not differ in their ability to compete for females after heat stress.

(3) The expression of hsp70 did not differ between treatment groups or populations (3-way ANOVA on the effects of population, hardening and stress on hsp expression, all $p>0.17$). This may indicate that either hsp70 is not upregulated in response to heat stress (which seems unlikely), or that a recovery time of 1h after the stress was too short to allow for significant upregulation of the protein.

Aims of the visit

As hsp expression has so far never been measured in the YDF, we had to search for and establish appropriate antibodies for stress-inducible hsps (focusing on hsp70). The search for such antibodies depends greatly on experience, abundantly available at the Department of Genetics and Ecology at Aarhus University in Prof. Dr. Volker Loeschcke's group working on thermal adaptation. During my stay in Denmark, I successfully identified an appropriate antibody using Western blot and followed by an enzyme-linked immosorbent assay (ELISA, as described by Sørensen et al. 1999), based on previous experience on studying hsp expression in *Drosophila* in Aarhus. Currently, I am establishing the method in our lab at Zurich University and engaged in pilot experiments on the effects of different temperature stresses on survival and reproductive traits in the YDF. This line of research will be one of the focal parts of my research in the near future.

My visit in Aarhus gave me the opportunity to extend my technical skills, which will enhance my future career opportunities. The supervision and organisation by Prof. Dr. Volker Loeschcke and Dr. Jesper G. Sørensen was excellent and a warm thanks to Doth Andersen for her technical assistance. I am very grateful to ESF for the grant.