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| Title:      | Of War and Love: A Study of Sexual Conflict and Sexual Selection Using <i>Drosophila melanogaster</i> Laboratory System |
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**Abstract:** Intersexual conflict is defined as the conflict between the evolutionary interests of the two sexes of a given species. It is an outcome of the differences in investment in reproduction by the two sexes. Intersexual conflict can happen in two different ways – Intra-locus and Inter-locus. Interlocus conflict is characterized by males seeking as many mates as possible, often coercing females to mating, whereas females trying to minimize harmful male interactions and increasing their life-time progeny production. This can potentially maintain an open ended co-evolutionary arms' race, perpetuated by male adaptations and female counter-adaptations. Such conflict has been shown to be pervasive, affecting a wide range of species and expected to affect many more. However, the evolutionary outcome of interlocus conflict, in terms of changes in reproductive behaviour of the two sexes and life-history traits, is poorly understood. In this thesis, I present the results of a laboratory experimental evolution study, where replicate populations of *Drosophila melanogaster* were subjected to altered levels of interlocus conflict. The level of conflict was varied by altering the operational sex ratio. Three sex ratio regimes were adopted – M, male biased (3:1), C, equal sex ratio and F, female biased (1:3). While male biased regime represents increased male-male competition and intersexual conflict, female biased condition represents the opposite. Equal sex ratio represents the ancestral conditions. Three replicate populations per regime were maintained. I present results of assays done between 8-60 generations of selection. After only 8 to 12 generations of selection I observed interesting evolutionary trends. Fitness (progeny production under competitive condition) of males from the male biased and female biased regime was found to have diverged significantly. While males from the male biased (M) regime were found to be more competitive, males from the female biased (F) regime were found to be poor at competition. M-males courted females more often compared to males of the other regimes. M-females had higher fitness compared to F-females when both were competed against the ancestral females. In a separate experiment, I also found a significant effect of the selection regime on mating latency between ancestral males and selected females, indicating M-females evolved increased resistance to mating attempts. If this is true, then it is likely that M-females had also evolved to be less affected by harmful male interactions, potentially explaining the difference in fitness of the females. After 40-50 generations of altered levels of interlocus conflict in my study, I found the evolution of males' ability to harm females and the consequent evolution in their life-history. Males from the male biased (high conflict) regime evolved higher activity and courtship frequency. This was associated with their greater mate harming ability in terms of increased mortality of females mated to them. Consistent with the theories of life-history evolution, I observed an increase in rates of aging and decline in mean lifespan of males from male-biased populations. Males from the female biased regime did not evolve in terms of courtship frequency and locomotor activity. However, they were found to be less harming (less damaging to female lifetime fitness) and relatively long lived. This is the first empirical evidence, clearly showing the evolution of male reproductive traits under intersexual conflict and its life-history consequences in terms of changes in life-span and rate of aging (Nandy et al. *Evolution*, in press). After 55-60 generations of selection, I observed evolution of sperm competitive ability (sperm defence-P1, offence-P2) in the above mentioned selection experiment. While male biased operational sex ratio is thought to generate increased male-male competition compared to the equal sex ratio condition, females biased sex ratio is expected to generate the opposite condition. Males from populations with female biased operational sex ratio evolved reduced P1 and P2, without any measurable change in the male reproductive behaviour. Males in the male-biased regime evolved increased P1, but there was no significant change in P2. Increase in P1 was associated with an increase in copulation duration, possibly indicating greater ejaculate investment by these males. This study is one of the very few empirical evidence for the evolution of sperm competitive ability of males under different operational sex ratios (and hence different levels of male-male competition) and is an important contribution to our understanding of post-copulatory sexual selection (Nandy et al. 2013, *Evolution*). The assays done after 40-50 generations of selection, suggested that females from populations experiencing higher level of intersexual conflict evolved increased resistance to mate harm, in terms of both longevity and fitness (Nandy et al. *BMC Evolutionary Biology*, in press). Females from the populations with low conflict were significantly more susceptible to mate harm, suffering greater fitness depression upon continuous exposure to the males. However, these females produced more progeny upon single mating and had significantly higher longevity in absence of any male exposure – an indication of trade-off between resistance related traits and other life-history traits, such as longevity and fecundity. Alternatively, increase in body size of these females can lead to such observations as increased body size also represents increase in the available resources. I also report two additional findings which are novel and of great interest. I found tentative evidence of an increased male cost

of interacting with more resistant females, a novel finding, which was hitherto expected theoretically but lacked empirical support. In addition, I found females of the M-regime to be more active. However, at this point it is difficult to predict whether this evolved as a direct response to the selection on females or is a correlated response of the selection on male-activity. After 40-55 generations of selection I quantified the effect of the adaptation to sexual antagonism on three important life-history traits – development time, larval survivorship and starvation resistance (Nandy et al. under preparation). While I did not find a significant effect of selection on larval survivorship, the other two traits were affected by the selection. Flies from the F-regime had significantly higher pre-adult development time. I found starvation cost of reproduction (decline in starvation resistance due to reproductive activity) to have decreased in F-males possibly due to their reduced investment in reproductive behaviour and/or physiology. The same cost was found to have increased in F-females, possibly due to their increased susceptibility to male-induced harm. In this thesis I also addressed theories of sexual selection predicting the evolution of male reproductive strategies. Theories suggest that when cost of reproduction is high, males should show adaptive mate choice and prudent ejaculate investment strategies. I tested this prediction using “laboratory island analysis” on outbred populations of *D. melanogaster* which have been adapted to the laboratory conditions for hundreds of generations. I have shown that sperm limited males preferentially mated with young and/or well fed females (Nandy et al. 2012, Scientific Reports). The most striking finding was the strong positive correlation between the degree of mating bias showed by the males and the variance in the fitness of the females. In a separate experiment (Nandy and Prasad 2011, Journal of Insect Science), I found that early life experience, in terms of co-inhabitant numbers, significantly affected male mating behavior (copulation duration) and at least one fitness component (sperm defense ability). These results are very important empirical support for the theories of sperm competition. In addition, the nonlinear trait response observed in this study was a novel finding pointing to the incompleteness of the theories.

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