Sexual conflict, sexual selection, and genetic variance in fitness

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Abstract

Understanding sex-specific genetic variance for fitness is of fundamental importance to our understanding of evolution. This thesis presents the findings of empirical investigations into sex-specific genetic variance in fitness. The findings are discussed in terms of their implications for our understanding of the classic evolutionary paradoxes of what maintains genetic variance in fitness and what maintains sexual reproduction, as well as more specific implications regarding adaptation and population viability. Males and females reproduce and accrue fitness in fundamentally different ways, which inevitably comes at a detriment to the fitness of individuals of the opposite sex. This is known as sexual conflict, and because males and females use largely the same genome to develop, grow and reproduce, a genetic tug-of-war ensues. Alternative alleles at sexually antagonistic (SA) genes have opposing fitness effects in males and females. The consequence of this genetic tug-of-war is that alternative allelic variants at SA loci can be maintained in the population. Such SA genetic variation can therefore maintain genetic variance for fitness. Variance in fitness can also be maintained by a constant influx of mutations with weakly deleterious effects and weak selection against them, in what is referred to as mutation-selection balance. Because the average deleterious mutation will be detrimental to both sexes, this source of genetic variance in fitness will have predominantly sexually concordant (SC) effects. This thesis uses a wild-caught population of the seed beetle *Callosobruchus maculatus* to investigate these two mechanisms of maintaining genetic variance in fitness, as well as the consequences they bear on adaptation, population viability, and the maintenance of sexual reproduction. Results largely support much of the theoretical expectations for sexual conflict, sexual selection and maintenance of genetic variance in fitness, as well as stimulate new thoughts and hypotheses about the nature of SA genetic variation and its interaction with weakly deleterious partially recessive mutations.

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“Nothing in genetics makes sense except in light of genomic conflict"

-Bill Rice
List of Papers

This thesis is based on the following papers, which are referred to in the text by their Roman numerals.


IV  Grieshop, K. and Arnoqvist, G. Sex-specific genetic variance for fitness characterized by sex-specific dominance and epistasis. *Manuscript*.

V  Grieshop, K., Arnoqvist, G. and Berger, D. Sexual selection in males, but not females, purges the standing genetic load in a seed beetle. *Manuscript*.

* These authors contributed equally to the study.

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The following were written during the course of my doctoral studies but are not part of the present dissertation:

Berger, D., Stångberg, J., Grieshop, K., Martinossi-Allibert, I. and Arnqvist, G. Life-History Trade-Offs, Germline Maintenance and Mutation Rates under Climate Warming. *In review*


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Introduction

The maintenance of genetic variance in fitness

One of evolutionary biology’s most longstanding debates is over which mechanism(s) maintain genetic variance in fitness (Lewontin 1974, Barton and Turelli 1989, Charlesworth and Hughes 2000, Barton and Keightley 2002, Mitchell-Olfs et al. 2007). Fitness is, by definition, under directional selection, which should drive genetic variation at each of the loci underlying variance in fitness toward fixation for whichever alleles render the organism nearest to its fitness optimum, thereby exhausting the raw material of evolution (Fisher 1930, Wright 1935, Kimura 1958, Lewontin 1958, 1974, Charlesworth 1987, Hartl and Clark 1997). However, observations of paradoxically abundant genetic variance in fitness and life history traits (e.g. Charlesworth 1987, Houle 1992) indicate widespread genetic polymorphisms at the underlying loci. What maintains polymorphism at these loci?

Hypotheses for the maintenance of genetic variation can be broadly categorized into mutation-selection balance and balancing selection (Dobzhansky 1955, Lewontin 1974), where genetic polymorphisms are maintained at low and intermediate equilibrium frequencies, respectively. Under mutation-selection balance, selection against a constant influx of mutations with weakly deleterious partially recessive effects provides a constant source of variance in fitness attributable to variation across genotypes in their relative share of the population’s mutation load (Haldane 1927, Lande 1975, Crow 1993, Zhang et al. 2004). Under balancing selection, various mechanisms of selection maintain alternative alleles at loci that underlie fitness without invoking a constant mutational input. These can be subdivided into ‘overdominance’ and ‘balancing selection’, in keeping with contemporary terminology. Overdominance is simply heterozygote advantage (i.e. selection at the underlying loci is actually not directional, see above), where heterozygous individuals exhibit relatively greater fitness than either homozygote (Fisher 1922). As I describe later, such strict overdominance has been ruled out as a generally plausible mechanism for maintaining genetic variance in fitness and life history traits (although the term ‘overdominance’ is still sometimes used to refer loosely to other form of balancing selection). In contrast, balancing selection takes many forms, for example antagonistic pleiotropy (Rose 1982, 1985), genotype-environment interactions (Hedrick 1986, Gillespie and Turelli 1989), spatial or temporal variation in selection (Ellner
and Hairston 1994), and negative frequency dependent selection (Cockerham et al. 1972, Hedrick 1972, Clarke 1979, Clarke et al. 1988, Asmussen and Basnayake 1990), all of which have theoretical and empirical support (see below) and can generate a population-level net heterozygote advantage at underlying loci (Charlesworth and Hughes 2000, Barton and Keightley 2002, and Mitchell-Olds et al. 2007).

In addition to these mechanisms, specific types of gene action—e.g. dominance, epistasis and pleiotropy—can also contribute to maintaining genetic variance for fitness and life history traits by expanding the range of conditions under which other mechanisms maintain stable polymorphisms. For example, variation in a trait can be maintained as a pleiotropic side effect of polymorphisms that are maintained for other reasons (Barton and Turelli 1989, Barton and Keightley 2002). This seems especially plausible considering that most loci inevitably affect multiple traits (Caspari 1952, Wright 1968, Lynch and Walsh 1998). Epistasis, i.e. interactions between loci, can also contribute to maintaining polymorphisms for fitness (Gimelfarb 1989, Barton and Keightley 2002, Arnqvist et al. 2014). Again, epistasis is quite likely involved in some way since it is an emergent property of many loci having additive effects when fitness functions are approximately Gaussian (Brodie 2000, Arnqvist et al. 2014; and see Paper IV). As a final example, environment-specific dominance (i.e. dominance reversal) drastically increases the capacity (or is perhaps even required) for antagonistic pleiotropy to maintain polymorphisms for fitness (Rose 1982, 1985, Hoekstra et al. 1985, Curtsinger et al. 1994). Again, this is expected to be the case for the average locus underlying two overlapping Gaussian fitness functions under antagonistic selection (Fry 2010; and see Paper IV).

Some of the above mechanisms and modes of gene action are more generally plausible than others (Barton and Turelli 1989, Charlesworth and Hughes 2000, Barton and Keightley 2002, Mitchell-Olds et al. 2007). Mutation-selection balance is clearly ubiquitous, but simple mutation-selection balance arguments alone cannot completely explain observed patterns of genetic variation in fitness and life history traits (Charlesworth and Hughes 2000, Kelly and Willis 2001, Kelly 2003, Charlesworth et al. 2007, Mitchell-Olds et al. 2007). This is perhaps because mutations tend to have pleiotropic effects on many traits (Barton and Keightley 2002, Estes et al. 2005), thus multivariate purifying selection against mutations may overwhelm per-locus mutation rates, stifling that source of genetic variance (Turelli 1985, Wagner 1989, Barton 1990, Barton and Keightley 2002). As mentioned, strict over-dominance (i.e. unconditional heterozygote advantage) is not a robust explanation for the stable maintenance of genetic polymorphisms for fitness (Clarke 1979, Charlesworth and Hughes 2000, Barton and Keightley 2002). Theory suggests that such unconditional heterozygote advantage should be a transient phenomenon, where a mutant allele causing heterozygote advantage is initially promoted, but after the polymorphism is established it
destabilizes with the increasing frequency of homozygous mutants (Clarke 1979). Even if a polymorphism reaches stable equilibrium via overdominance, gene duplication is predicted to enable all individuals to exhibit both alleles, resulting in so called “fixed heterozygosity” (Clarke 1979, Mitchell-Olds et al. 2007). Lastly, many studies that thoroughly investigate the mechanism(s) underlying balanced polymorphisms reveal something other than simple overdominance, for example, genotype-environment interactions (Mukai 1988, Li et al. 2003, Weinig et al. 2003), spatial or temporal variation in selection (Schemske and Bierzychudek 2001, 2007, Schmidt and Conde 2006), negative frequency dependent selection (Subramaniam and Rausher 2000, Gigord et. al 2001, Fitzpatrick et al. 2007, Kazancıoğlu and Arnqvist 2014, Baris et al. 2017), or antagonistic pleiotropy (Barson et al. 2015). Contemporarily, therefore, the question of what maintains genetic variance in fitness and life history traits concerns the relative contributions of mutation-selection balance and balancing selection (Barton and Keightley 2002, Mitchell-Olds et al. 2007).

With mutation-selection balance being ubiquitous but unable to account for all of the observed genetic variance, the debate for many evolutionary biologists is over which of the many mechanisms of balancing selection is/are the most plausible or most widespread. For reasons beyond the scope of this thesis, negative frequency dependent selection (where rare alleles have a fitness advantage), seems to have a limited capacity to maintain many genetic polymorphisms across the genome (Barton and Turelli 1989, van Doorn and Dieckmann 2006), unless via maintaining certain loci or “super-genes” (i.e. polygenic haplotypes) with epistatic effects throughout the genome (e.g. Kazancıoğlu and Arnqvist 2014, Baris et al. 2017). This leaves genotype-environment interactions, spatially or temporally varying selection, and antagonistic pleiotropy. All of these have a common feature: selection favors different alleles in different contexts. However, antagonistic pleiotropy is a particularly powerful version of that, representing a genetic tradeoff between alternative allelic variants in alternative contexts (see Paper IV). Indeed, antagonistic forms of balancing selection show a greater capacity to maintain genetic variance than non-antagonistic balancing selection (Connal lon and Clark 2013). I explain in more detail in Paper IV why antagonistic pleiotropy is likely the most widespread form of balancing selection among multicellular sexually reproducing Eukaryotes, as well as present evidence in favor of that argument.

This thesis will, therefore, focus on the evolutionary implications of genetic variance in fitness generated by mutation-selection balance and antagonistic pleiotropy. Specifically, in the context of having separate sexes, I suggest that these are likely the two most common mechanisms for maintaining genetic variance in fitness, and therefore represent important facets of our understanding of evolution. Briefly, the fact that the sexes reproduce and accrue fitness in fundamentally different ways (see next section) means that
they form two different genetic “environments” with different optimal trait values for the life history traits that make up fitness. Yet, males and females produce these traits using (largely) the same genes, engendering many loci across the genome to a specific type of antagonistic pleiotropy called intralocus sexual conflict (ISC). As discuss below, ISC therefore has important implications for our understanding of the maintenance of genetic variance in fitness (see Paper IV), as well as adaptation (see Paper I) and tolerance to inbreeding (see Paper III). Then, as discussed more generally in the context of mutation-selection balance so far, strong directional sexual selection would erode genetic variance for fitness if it were not supplied by a constant mutational input. Thus, variation among individuals in competitive lifetime reproductive success should reflect their relative load of mutations with weakly deleterious partially recessive effects on female/population offspring production, depending, of course, on the extent to which sexual selection favors SA genetic variation (see below). As discussed below, sexual selection therefore also has important implications for our understanding of the maintenance of genetic variance in fitness, as well as the value of males to females and prevalence of sexual reproduction (see Papers II and V).

Intralocus sexual conflict

Of the many various contexts over which selection could vary, having separate sexes must be one of the most widespread, since sexual reproduction is ubiquitous among Eukaryotes (Speijer et al. 2015). As long as there are separate sexes there will be sex-specific selection, which will operate with disregard for—and inevitably to the detriment of—the fitness of the opposite sex (Kokko and Jennions 2014). When the outcome of selection in one sex is detrimental to the fitness of individuals of the opposite sex, selection is said to be sexually antagonistic (SA), generating a scenario referred to as sexual conflict (Chapman et al. 2003, Arnqvist and Rowe 2005). Because the sexes share most of the genome (aside from the sex-chromosomes) and because sexual conflict is an emergent property of having separate sexes, SA genetic polymorphisms for fitness are inevitable (Connallon and Clark 2014), and a population’s genetic variance for fitness is expected to be predominantly SA (Connallon and Clark 2012; see Paper IV). This sexual conflict over the alternative allelic variants of particular genetic loci is known as intralocus sexual conflict (ISC; Lande 1980, Arnqvist and Rowe 2005, Bonduriansky and Chenoweth 2009, van Doorn 2009). As a brief illustrative example, female fitness tends to increase with larger body size in insects (within certain physiological limits) because it enables greater fecundity (Fairbairn et al. 2007). Thus, females benefit from genotypes that cause them to invest limiting resources into growth and development. Male fitness, by contrast, tends to increase with fertilization success, and does not typically depend on body
size in the same way as female fitness (Fairbairn et al. 2007). Thus, males would tend not to benefit from a genotype causing them to allocate resources toward developing and/or maintaining a large body size, especially if it comes at the expense of investment into traits that increase fertilization success. Thus, males and females maximize fitness via different phenotypic values for body size, a highly polygenic trait attributable to many genes across the genome (Fairbairn et al. 2007), yet they use the same genome during development. It is therefore easy to imagine that sex differences in the phenotypic optima for body size and developmental rate would generate ISC throughout the genome (Fairbairn et al. 2007).

ISC is effectively cross-sex antagonistic pleiotropy (Rose 1982, 1985), where alternative alleles pleiotropically affect two traits: male fitness and female fitness. Indeed, in much the same way that trait-specific dominance plays an important role in enabling antagonistic pleiotropy to maintain polymorphisms for fitness (Hoekstra et al. 1985, Curtsinger et al. 1994), sex-specific dominance is likewise effective or perhaps even required for the stable maintenance of SA polymorphisms (Kidwell et al. 1977, Prout 2000), and is expected for the average SA locus (Fry 2010; described above and in Paper IV). Interestingly, the most definitive identification of a specific SA genetic polymorphism in a natural population was shown to exhibit at least partial sex-specific dominance; that is, which allele was dominant over the other in heterozygotes depended on the sex of the individual (Barson et al. 2015). In their study of Atlantic salmon, Barson et al. (2015) identified a gene that explains 39% of the observed variation in age at sexual maturity, a SA life history trait that is very closely tied to fitness, where the allele benefiting male fitness was dominant in heterozygous males, and the allele benefiting female fitness was dominant in heterozygous females. This partial resolution of the ISC over age at maturity has likely contributed to the maintenance of this polymorphism in this population (Barson et al. 2015), which contributes to some portion of the genetic variance in fitness.

Sex-specific dominance is not the only means by which to promote the stable maintenance of SA polymorphisms for fitness. First, SA polymorphisms will reach a stable equilibrium if selection is nearly equivalent between the two sexes (Kidwell et al. 1977; Prout 2000). But even in the absence of this rather stringent criterion, many other mechanisms besides dominance reversal act to expand the parameter space for protected polymorphism. For example, X-linkage (Rice 1984, Patten & Haig 2009), linkage (disequilibrium) between SA loci (Patten et al. 2010, Úbeda et al. 2010), assortative mating for fitness (Arnqvist 2011), linkage (disequilibrium) with parentally antagonistic loci (Patten et al. 2013), epistasis between SA loci (Arnqvist et al. 2014), and linkage (disequilibrium) with meiotic drive loci (Patten 2014), have all been shown to increase the stable maintenance of SA polymorphisms for fitness. In addition, as in the case of partial resolution via dominance reversal (see above), partial resolution of ISC by parental effects
(Day and Bonduriansky 2004, Bedhomme and Chippindale 2007) and sex-biased gene expression (Bedhomme and Chippindale 2007) can also contribute to maintenance of SA genetic variance for fitness (see Paper IV). These are all, independently, quite plausible mechanisms, with some being expected or perhaps even inevitable. For example, as discussed above (and more thoroughly in Paper IV), SA epistasis and dominance reversal are emergent properties of polygenic additivity and (overlapping) Gaussian fitness functions. Likewise, linkage disequilibrium between SA loci happens simply due to recurrent admixture (i.e. fertilization) between sperm and egg sub-populations each generation (Úbeda et al. 2010). And the concept of selection favoring physical linkage between SA loci, and sex-linkage of SA loci (due to the fitness benefits of co-inheriting alleles with congruent sex-specific effects), is so widely excepted that the process is expected to play a key role in the origin and evolution of sex-chromosomes (Bull 1983, Rice 1996, van Doorn and Kirkpatrick 2007, Jordan and Charlesworth 2012, Blaser et al. 2013), which are nearly ubiquitous among Eukaryotes. Thus, ISC would seem more of a rule than an exception. Indeed, a growing body of empirical evidence supports this notion (e.g. Chippindale et al. 2001, Fedorka and Mousseau 2004, Pisschedda and Chippindale 2006, Brommer et al. 2007, Foerster et al. 2007, Arnvist and Tuda 2009, Bilde et al. 2009, Mainguy et al. 2009, Svensson et al. 2009, Conallon et al. 2010, Cox and Calsbeek 2010, Innocenti and Morrow 2010, Delph et al. 2011, Lewis et al. 2011, Tarka et al. 2013, Berger et al. 2014a,b).

ISC has other evolutionary implications beyond maintaining genetic variance for fitness. For example, it can pose a severe gender load on the population (Chippindale et al. 2001, Rice and Chippindale 2001, Pisschedda and Chippindale 2006, Arnvist and Tuda 2009). That is, if SA genetic variation largely underlies sex-specific reproductive fitness, then the most reproductively successful males will be those carrying alleles that detriment female fitness, and therefore the population’s offspring production. Because female fitness (i.e. offspring production) is the limiting factor to a population’s growth rate, the gender load on the population has the potential to generate a “tragedy of the commons” scenario (Kokko and Brooks 2003, Berger et al. 2016). If a population exhibits mostly SA genetic variance for fitness (see above), and if selection is stronger in males than in females (see next section), then the selfish fitness benefits posed to the most reproductively successful males will causes the population to become enriched for male-benefit allelic variation that detriments female/population offspring production (Berger et al. 2016). In the most extreme case, this can drive populations toward a downward spiral of dwindling population size, inbreeding and extinction (Kokko and Brooks 2003, Martinez-Ruiz and Knell 2017). In addition to the gender load on population growth rate, I show in Paper III that male-benefit/female-detriment allelic variation carries an inherently greater risk of extinction upon inbreeding, which could exacerbate the gender load
and accelerate a population’s path toward extinction under the above scenario (Grieshop et al. 2017).

At first, the gender load imposed by ISC may seem to inhibit populations from even persisting, much less adapting, to novel environments (Chippindale et al. 2001, Rice and Chippindale 2001, Pischedda and Chippindale 2006, Arnqvist and Tuda 2009). However, the stress of novel environmental conditions can cause the expression of conditionally deleterious alleles (Hoffmann and Merilä 1999, Martin and Lenormand 2006), which may decrease the extent to which SA loci contribute to variance in sex-specific fitness (Whitlock and Agrawal 2009, Long et al. 2012). Furthermore, since male-specific reproductive traits seem especially sensitive to stress (Sheldon et al. 1998; Brooks 2000; Tomkins et al. 2004; Bussiere et al. 2008; Kwan et al. 2008; Bellamy et al. 2013; Sharp and Agrawal 2013), one might predict that male-benefit genetic variation might be poorly suited for buffering stress. Because females typically maximize their fitness by having long reproductively active lives, classic life history theory would predict a female-benefit genotype to promote a life history strategy that devotes resources to longevity, somatic tissue maintenance and immune system function (Stearns 1992, Fairbairn et al. 2007), which make a typical female-benefit genotype well suited for buffering stress. By contrast, since males typically maximize their fitness by competing intensely for fertilizations (Janicke et al. 2016), a typical male-benefit SA genotype would promote a sort of ‘live-fast/die-young’ life history strategy characterized by high metabolic rate, fast development and disproportionate investment into reproductive traits, potentially at the expense of longevity, somatic tissue maintenance and immune system function (Stearns 1992, Fairbairn et al. 2007). Thus, male-benefit genotypes should, indeed, be poorly suited to buffer stress, which may require that males with male-benefit genotypes that are exposed to stress adjust resource allocation toward basic survival at the expense of reproductive competitiveness (Stearns 1992). This, in combination with the expression of conditionally deleterious alleles (see above), means that male-benefit genotypes will likely not be the most reproductively successful under stress—in fact, perhaps the opposite. A novel environment requiring that organisms buffer stress may render males with female-benefit (stress-tolerant) genotypes more reproductively competitive, where males with ‘live-fast/die-young’ genotypes may struggle to even survive, much less compete for fertilizations. Thus, regardless of the extent to which SA genetic variation underlies fitness variance, upon stress males and females may benefit from the same alleles, nullifying the gender load imposed by ISC, enabling populations adapt to novel environments (Berger et al. 2014a; see Paper I).
Sexual selection

Sexual selection, though not necessarily an inevitable outcome of having separate sexes (Kokko and Jennions 2014), has received much more attention than sexual conflict. This is perhaps because it is responsible for some of the most conspicuous, exaggerated and bizarre traits ever observed (Andersson 1994), begging explanation and grabbing the attention of (arguably) the founder of evolutionary biology—Charles Darwin (Darwin 1871). Sexual selection can and has been studied from many angles, with perhaps the broadest evolutionary implications coming from our understanding of what maintains genetic variance for sexually selected traits in the face of strong directional sexual selection. This question bears obvious resemblance to the less specific version of that question, regarding the maintenance of genetic variance for fitness (discussed above). In the context of female choice for male secondary sexual traits, this concept has been awarded its own title (the “lek paradox”), where the concept of “genic capture” has been invoked to account for this paradox (Rowe and Houle 1996, Tomkins et al. 2004). Genic capture is a useful concept that is applicable, by analogy, to more general questions (Tomkins et al. 2004). Under genic capture, sexually selected traits are theorized to be so highly polygenic (reflecting allelic variation throughout the genome) that they effectively reveal individuals’ genome-wide genetic quality, which bears consequence to general organismal condition (Rowe and Houle 1996, Hunt et al. 2004, Tomkins et al. 2004, Radwan 2008).


Such a process, in which only those males carrying relatively few mutations contribute to each generation’s offspring production, could promote population mean fitness if acting via males, but not via females (Whitlock and Agrawal 2009). This is because non-birth-giving males contribute so little to population growth rate that there would be little or no demographic consequence to having most of the population’s males lose the competition for mates each generation (Whitlock and Agrawal 2009). This additional population-level benefit to sex could help compensate for what is commonly referred to as the “two-fold cost of sex” (Lehtonen et al. 2012). Classically, this refers to the fact that sexually reproducing females contribute to population growth at half the rate of asexual females (Maynard Smith 1971, Hartfield and Keightley 2012, Lehtonen et al. 2012). The cost of sex (Lehtonen
et al. 2012) therefore begs the question of why sexual reproduction is so common. How can sexual females compete with asexuals? What prevents an asexual mutant lineage from taking over in a sexually reproducing population? The population-level benefits of strong sexual selection in males may contribute to the maintenance and prevalence of sex (Whitlock and Agrawal 2009).

Of course, this requires that the allelic variation that is being selectively filtered out of the population via males is that which is also detrimental to female fitness—i.e. that the mutation load has sexually concordant (SC) effects (Whitlock and Agrawal 2009). However, due to the effectiveness of purifying selection at fixing/eliminating alleles with beneficial/detrimental effects in both sexes, a population in mutation-selection balance should be left with predominantly SA genetic variance for fitness (Connallon and Clark 2012). Thus, it is possible that sexual selection acts primarily on SA genetic variation (Rice and Chippindale 2001, Pischedda and Chippindale 2006, Radwan 2008, Prokop et al. 2012). In accord, empirical evidence for sexual selection in males revealing/purging SC mutations comes mostly from studies of induced mutations (e.g. Radwan 2004, Hollis et al. 2009, Grieshop et al. 2016, see Paper II), with the process and/or its detection seemingly limited in the context of the standing genetic variation for fitness (e.g. Chenoweth et al. 2015, but see Paper V). Indeed, whether sexual selection will have a net positive or negative effect on population mean fitness depends largely on the extent to which it acts on SA versus SC allelic variation (Whitlock and Agrawal 2009).

Study system

The research presented in this thesis was conducted using the seed beetle species Callosobruchus maculatus (Coleoptera: Bruchidae; Figure 1). This beetle is a pest of leguminous crops that has colonized most of the tropical and subtropical regions of the world (Southgate 1979). Thus, laboratory conditions (see Papers) fairly closely resemble the grain storage facilities and crop fields they have inhabited since the early Holocene, making our laboratory assays of competitive lifetime reproductive success (i.e. fitness) relevant to their natural ecology. Females lay eggs on the surface of dry beans and hatched larvae bore into the beans, where they complete their life cycle, emerging from the beans as reproductively mature adults (Southgate 1979). This species is facultatively aphagous (requiring neither food nor water to reproduce successfully), exhibits a polyandrous mating system (Miyatake and Matsumura 2004) and has X/Y sex-determination (Angus et al. 2011). These features, together with a generation time of only ~3 weeks, render C. maculatus an ideal laboratory model system.
Figure 1: Photograph of a copulating pair of *C. maculatus*. Females (left) are ~5 mm in length, and males (right) are smaller.

Photo by Ivain Martinossi-Allibert
Summary of papers

Intralocus sexual conflict and environmental stress (Paper I)

A measure of the degree of SA genetic variance for fitness in a population is the intersexual genetic correlation ($r_{MF}$) for fitness, where a negative $r_{MF}$ reveals allelic variation with opposing fitness effects in the sexes (i.e. SA genetic variation). Paper I investigates the change in $r_{MF}$ between benign/adapted and novel/stressful environmental conditions, in the context of the predictions described above regarding SA genetic variation and stress. In two populations, one exhibiting mostly SA genetic variance for fitness and one not, $r_{MF}$ became more positive under stress (though only significantly so the former, Figure 2). In both populations, males and females from genotypes that benefit males under benign conditions were less tolerant to stress than those with typical female-benefit genotypes, supporting the predictions outlined above (Figure 3).

Figure 2. Lome (A) and Ofuya (B) populations’ $r_{MF}$s at benign (29° c) and stressful (36° c) environmental conditions.
Figure 3. Demonstration of the mechanism underlying the effect displayed in Figure 2. Males and females (white and black dots, respectively, of panels C and D), with male-benefit genotypes at benign conditions are less tolerant to the stress in both Lome (left) and Ofuya (right) populations.
Strong sexual selection in males against a mutation load that reduces offspring production in seed beetles (Paper II)

Paper II uses 4 randomly chosen genotypes from the Lome population (above) to investigate the potential for sexual selection in males and females to purge mutations with deleterious effects on female/population productivity. Mutations were induced with ionizing radiation in F₀ grandparents, and found to have SC deleterious effects F₁ monogamous-pair productivity (i.e. female offspring production) and F₂ male competitive fitness (Figure 4). Sexual selection against these induced mutations was consistently strong in males (regardless of genetic background or in which sex the mutations were induced), but was non-significant in females (Figure 5). These findings support the theory described above regarding sexual selection tending to be stronger in males and therefore enabling SC deleterious mutations to be purged from the population via males, but not females.

Figure 4. Mutations induced with ionizing radiation via F₀ grandfathers (black) or grandmothers (grey) have SC detrimental effects on F₁ population productivity and F₂ male competitive fitness, whereas male fitness does not reflect the standing genetic load on the population in non-mutated controls (dashed).
Figure 5. Regardless of whether mutations were induced via grandfathers or grandmothers (a), and regardless of genetic background (b), sexual selection against induced mutations with deleterious effects on population productivity was strong in males (black), and non-significant in females (white).
Male-benefit sexually antagonistic genotypes show elevated vulnerability to inbreeding (Paper III)

Paper III documents an inbreeding experiment, in which replicated lineages of genotypes from the Lome population (above) were subject to 10 consecutive generations of single-pair, full-sib inbreeding. The genetic stress induced by genome-wide homozygosity was measured as lineage extinction per genotype, the genotypes having been characterized in Paper I according to their relative SA and SC fitness. Thus, variation in extinction from inbreeding reflects both the relative amount of deleterious mutation load carried by each genotype via the effect of Concordance on Extinction risk, as well as the relative effect of male- versus female-benefit SA genetic variation on extinction via the effect of Antagonism on Extinction risk (Figure 6). Concordance had a marginally non-significant effect (owing to limited genetic variance in that dimension) on extinction risk (Figure 7b), supporting the theory that mutation load causes inbreeding depression, and supporting the logic that extinction risk reflects the stress of genome-wide homozygosity. Congruent with the findings of Paper I, male-benefit genotypes were significantly less tolerant to the stress of inbreeding/homozygosity (Figure 7a). The implications of this finding are discussed in terms of the interaction between the gender load and mutation load (see above) upon inbreeding, and the potential threat to population viability posed by SA genetic variation.

Figure 6. The intersexual genetic correlation for raw log-transformed data from Paper I, demonstrating the derived variables Antagonism and Concordance.
Figure 7. Extinction due to homozygosity was greater for male-benefit and generally low fitness (i.e. low-male/low-female) genotypes (though latter effect was marginally non-significant).
Sex-specific genetic variance for fitness characterized by sex-specific dominance and epistasis (Paper IV)

Paper IV presents the results from a diallel partitioning of variance in sex-specific competitive fitness for the Lome population (above) into its various different contributing inheritance classes (e.g. additivity, dominance and epistasis). This paper therefore investigates the potential different modes of gene action and patterns of inheritance to contribute to the maintenance of genetic variance in fitness via maintaining SA polymorphisms throughout the genome. Bayesian (Figure 8a) and restricted maximum likelihood (Figure 8b) approaches both revealed the genetic architecture for fitness in this population to exhibit pronounced dominance, epistasis, and sex-specific versions thereof, supporting the prediction that sex-specific dominance reversal and epistasis likely contribute to the maintenance of SA genetic variance for fitness (see above).

Figure 8. Diallel variance partitioning of sex-specific fitness via Bayesian (a) and restricted maximum likelihood (b) estimation, revealing pronounced contribution to fitness variance in this population stemming from dominance, epistasis and sex-specific dominance and epistasis.
Sexual selection in males, but not females, purges the standing genetic load in a seed beetle (Paper V)

Paper V used the data from Paper IV to address the question of whether or not strong sexual selection in males can purge the deleterious mutation load on the population (see above). It focuses on modeling the male/female-inbred/outbred (co)variance matrix for fitness, which enabled the quantification of the standing genetic load on sex-specific fitness in this population by subtracting inbred ($i$) breeding values from outbred ($o$) breeding values for males and females separately. Male and female mutation loads were highly significantly correlated ($r_{oM-iM_oF-iF} = 0.83$, $(0.67-0.96)$, $P < 0.0001$), meaning this reflects the SC mutation load on the population, and also enabling cross-sex correlations that avoid bias due to shared measurement error. Male outbred fitness was significantly negatively correlated to the mutation load on female fitness ($r_{oM_oF-iF} = -0.52$, $(-0.72, -0.13)$, $P = 0.012$), but the reverse was not true ($r_{oF_oM-iM} = -0.02$, $(-0.37, 0.26)$, $P = 0.876$), demonstrating that sexual selection in males, but not females, reveals the SC mutation load on population mean fitness in this population. This rare demonstration of genic capture of the population’s standing genetic load on fitness via males, but not females, supports the theory that strong sexual selection in males may contribute to the maintenance, and hence prevalence, of sexual reproduction.
Thesis conclusions

This thesis presents results that are largely congruent with the theoretical expectations for sexual conflict, sexual selection and maintenance of genetic variance in fitness, as well as stimulates new thoughts and hypotheses about the nature of SA genetic variation and its interaction with weakly deleterious partially recessive mutations. The seed beetle populations used exhibit abundant SA genetic variance for fitness (Papers I and IV), which is expected for populations at mutation-selection balance. Male-benefit SA genotypes are less tolerant to stress (Papers I and III), which is predicted from classic life history theory. This SA genetic variation may pose a gender load on population viability (Paper III), but the population nevertheless still exhibits some SC genetic variance for fitness (Papers III and IV), expected to be maintained at least in part by mutation-selection balance. Sexual selection acts more strongly in males than females against novel mutations (Paper II), and with enough statistical power this purging of SC mutations is also seen to continue purifying the standing genetic load on the population (Paper V), supporting a role for strong sexual selection in males contributing to the maintenance of sexual reproduction. Lastly, the SA genetic variation underlying much of the sex-specific fitness variance in this population is characterized by pronounced sex-specific dominance reversal and SA epistasis (Paper IV), which are theorized to promote, or even be necessary for, the maintenance of genetic variance in fitness via sexual conflict.


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Svensk sammanfattning

Vår kunskap om könsspecifik selektion och genetisk variation för fitness är central för förståelsen av evolutionära processer. I den här avhandlingen presenteras resultaten av empiriska undersökningar av just könsspecifik genetisk variation för fitness. Resultaten diskuteras med fokus på deras betydelse för de klassiska evolutionära paradoxerna angående vad som bibehåller genetisk variation i fitness och varför organismer som förökar sig sexuellt är så vanliga, men även mer specifika konsekvenser för en populations anpassningsförmåga och livskraftighet avhandlas. Evolutionen har ofta gynnat olika reproduktiva strategier hos hannar och honor, och dessa strategier kan medföra kostnader för det motsatta könet. Den könskonflikt som uppstår på grund av detta kan också inbegripa en genetisk dragkamp eftersom könen delar genetisk arvsmassa men gynnas av olika anpassningar. Konsekvensen är att alternativa varianter av gener gynnas hos honor och hannar, vilket resulterar i en form av balanserande selektion som kan bibehålla genetisk variation i en population. Genetisk variation i fitness kan även upprätthållas genom en jämvikt mellan ett konstant inflöde av genetisk variation via mutatorer med svagt negativ effekt och svag selektion mot dessa mutationer. Eftersom en negativ mutation normalt kommer vara skadlig för båda könen kommer den här typen av källa till genetisk variation i fitness ha liknande effekt hos könen. I arbetet med denna avhandling har jag använt en vilt infångad population av fröbaggaen *Callosobruchus maculatus* för att undersöka dessa två underliggande mekanismer bakom upprätthållandet av genetisk variation för fitness, samt vilka potentiella konsekvenser de kan ha för en populations anpassningsförmåga och för bibehållandet av sexuell reproduktion. Resultaten i denna avhandling stödjer i stort många av de antaganden som ligger till grund för teorin om könskonflikter, sexuell selektion och vad som upprätthåller genetisk variation för fitness. Resultaten ger också upphov till nya idéer och hypoteser angående genetisk variation med könspecifika effekter och dess interaktion med partiellt recessiva negativa mutationer.
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A doctoral dissertation from the Faculty of Science and Technology, Uppsala University, is usually a summary of a number of papers. A few copies of the complete dissertation are kept at major Swedish research libraries, while the summary alone is distributed internationally through the series Digital Comprehensive Summaries of Uppsala Dissertations from the Faculty of Science and Technology. (Prior to January, 2005, the series was published under the title “Comprehensive Summaries of Uppsala Dissertations from the Faculty of Science and Technology”.)