Article

Desiccation-induced changes in recombination rate and interference in fruit flies: an evidence for fitness-dependent plasticity

Dau Dayal Aggarwal[1#], Sviatoslav R. Rybnikov[1#], Irit Cohen[1], Eugenia Rashkovetsky[1], Pawel Michalak[2], Abraham B. Korol[1*]

[1]Institute of Evolution and Department of Evolutionary and Environmental Biology, University of Haifa, Haifa, Israel

[2]Virginia Bioinformatics Institute, Washington Street, MC 0477, Blacksburg, VA 24061-0477, USA

[#]=equal contribution

*=corresponding author (korol@evo.haifa.ac.il)

ABSTRACT

Meiotic recombination is evolutionarily ambiguous, giving both benefits and costs to its bearers, with the resultant depending on a variety of conditions. The existing theoretical models explain why recombination does exist while the existence of its essential features remains underexplored. Here we test whether recombination plasticity and interference plasticity are fitness dependent. We compare effects of desiccation on recombination and interference in chromosome 3 in desiccation-sensitive and desiccation-tolerant *Drosophila* lines. We show that in desiccation-sensitive genotypes the treatment causes significant segment-specific increase in single- and double-crossover frequencies while in desiccation-tolerant genotypes changes are less pronounced, if seen at all. These results indicate that desiccation is a recombinogenic factor, and that desiccation-induced changes in both recombination and interference are fitness dependent, with a negative-feedback pattern implying the tendency of less fitted individuals to produce more variable progeny. Such dependence may play an important role in regulation of genetic variation in population experiencing environmental challenges.

Keywords: recombination, interference, plasticity, fitness dependence, desiccation, *Drosophila melanogaster*.

INTRODUCTION

Since the seminal experiments by Plough [1,2], meiotic recombination has been known to possess plasticity with respect to different ecological factors. Typically, its rates are low in optimal environment and rise as far as conditions become more stressful. To date, plasticity of recombination rate has been reported for many model species and numerous ecological factors (for recent reviews see [3–5]). Particularly, for fruit flies the phenomenon was observed with respect to heat [1,2,6–11], cold [1,8], and starvation [12]. Later on, Korol's group demonstrated in their experiments with tomato plants that cold-resistant genotypes rose chiasma number under high-temperature cultivation regime while heat-resistant ones – under low-temperature regime; the F_1 hybrids, tolerant across the examined range, showed no significant changes under any cultivation regimes [13]. Based on these results, the authors suggested that plasticity of recombination rate is modulated by genotype fitness in a negative-feedback way. However, such inter-genotype studies remain extremely limited. Particularly, in fruit flies plasticity of recombination rate was shown (more or less convincingly) to be fitness-dependent only with respect to heat [6,14] and specific chemicals associated with oxidative stress [15,16].

Obviously, recombination is expressed not only in terms of its rates. One of essential recombination features is interference, considered as mutual effects of crossover events when the resulting frequency of double crossovers may appear either lower (positive interference) or higher (negative interference) than that the expected under assumption of crossover independence. To date, the phenomenon attracts further more attention in light of its evolutionary role [17,18]. However, variation in interference between environments and genotypes is underexplored. Till now, plasticity of interference was reported only for fruit flies with respect to heat [2,10,19,20]. In all such experiments, the consistent pattern was an increase in double-crossover frequency under stress. Whether plasticity of interference is fitness-dependent has never been studied, to the best of our knowledge.

In this study, we aimed to test whether desiccation-induced changes in recombination rate and interference depend on flies' desiccation tolerance. As a precondition for such a test, we had first to check whether recombination rate and interference in fruit flies are affected by desiccation, which is a key ecological factor for this species [21,22]. The analysis was conducted on D. melanogaster lines that differed in desiccation tolerance. These lines resulted from our recent long-term experiments aimed to test whether directional selection for desiccation tolerance may cause indirect selection for recombination [23]. In that study we found, in line with theoretical predictions [24,25], that desiccation-tolerant lines showed segment-specific increase in recombination rate compared to the non-selected,

desiccation-sensitive ones. We also observed segment-specific relaxation of positive interference (and even appearance of negative interference) in the selected lines, and these changes were not necessarily coupled with changes in recombination rate. In the current study, we assumed and explicitly confirmed that the revealed difference between the selected and non-selected lines in terms of desiccation tolerance holds also for their F_1 hybrids with a standard multiple-marker strain. This allowed us to use the obtained F_1 heterozygous females for testing the hypothesis that desiccation-sensitive genotypes display higher plasticity of recombination rate and interference with respect to desiccation stress, compared to desiccation-tolerant ones, as expected by condition-dependent recombination concept.

MATERIAL AND METHODS

Flies and experimental arrangement

In our experiment we used two types of parental lines: (a) desiccation-sensitive (*S*), originating from flies collected in 2009 in Madhya Pradesh, Jabalpur, India; (b) desiccation-tolerant (*T*), originating from the same Jabalpur stock, though underwent long-term (48 generations) laboratory selection for desiccation tolerance [23]. Virgin females from the parental lines (three *S*- and three *T*-lines, each with three replicates; total 18 replicates) were mated with males from the marker stock *ru-cu-ca*, homozygous for six recessive mutations in chromosome 3: *ru, h, th, cu, sr,* and *e*. The mated females were then transferred to fresh food bottles for six hours to lay F_1 eggs. The third-instar F_1 larvae were either subjected to desiccation treatment or maintained as control (80 larvae per replicate). In *Drosophila*, changes in recombination can be induced by exogenous factors during a rather long period – from interphase till the end of pachytene [10,26]. Three-day old F_1 females obtained from both treated and non-treated F_1 larvae (20 females sampled from each replicate) were back-crossed with males from the same marker line (Figure 1). Recombination and interference were analyzed based on marker segregation in the obtained progeny. Recombination was measured as crossover frequency, while interference – as coefficient of coincidence, which is the ratio of the observed number of double crossovers to their number expected under assumption of independent recombination in adjacent intervals.

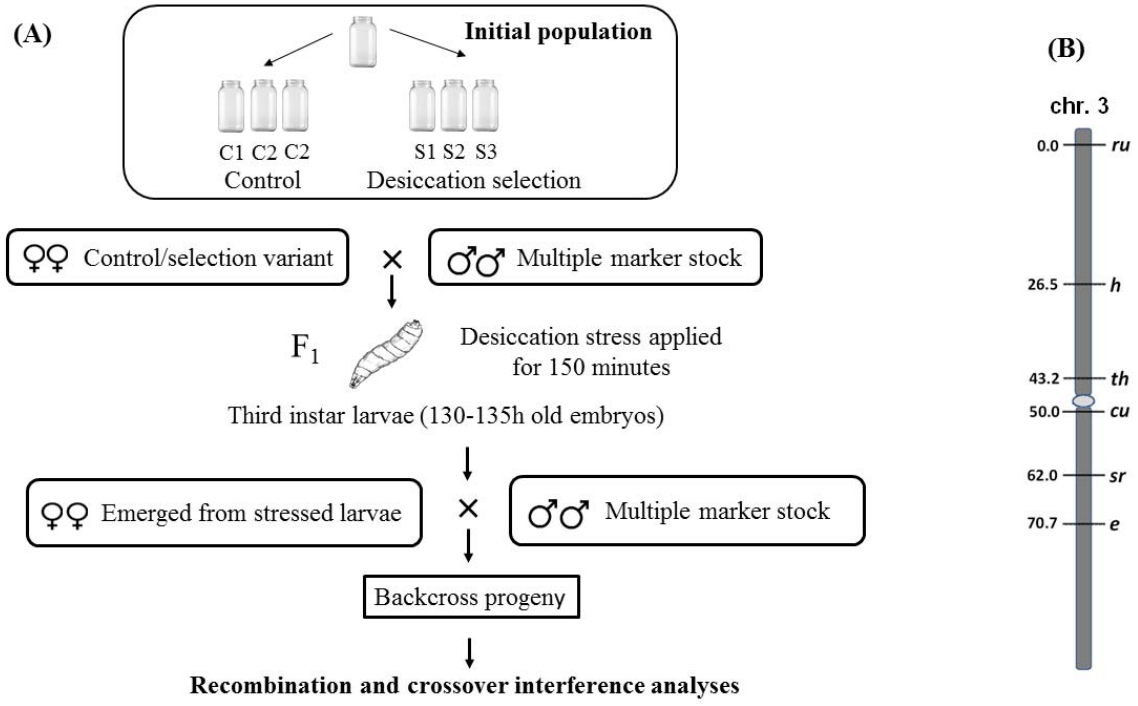

Figure 1: Experimental arrangement (A) and location of the markers in chromosome 3 (B)

Treatment and survival analysis

In each of 18 replicates involved in the experiment, 80 F_1 larvae were exposed to desiccation treatment and 80 F_1 larvae were maintained as control. For desiccation treatment, the larvae were divided into 16 groups, each of five larvae (in line with the method by Markow *et al.*, 2007). The blue indicating silica gel (2g) was placed into 16 dry plastic vials (20×90 mm). Groups of 5 larvae, gently isolated with a brush, rinsed, and air-dried, were placed into other 16 empty plastic vials, with their open ends covered with a muslin cloth to enable free air flow. Then, the larvae-containing vials were carefully placed above the silica gel-containing ones, and 16 obtained setups were made airtight by sealing the gaps between the two halves with multi-layers Parafilm. The larvae were treated during 150 min and then transferred into fresh food-containing vials. The F_1 females hatched from the desiccation-treated larvae (20 females per replicate) were further used in backcrosses. The control flies were reared under the same conditions, except the desiccation treatment. A similar experimental scheme was used in survival analysis of F_1 larvae. Groups of 10 larvae were treated by desiccation until death. Groups of the control larvae were left untreated. The number of immobile larvae was scored every 30 min.

Statistical analysis

For each pair of intervals, maximum likelihood analysis was performed to estimate the two recombination frequencies $r1$ and $r2$, and the coefficient of coincidence for non-treated and treated heterozygous females of tolerant or sensitive lines. Joint analysis of the progeny of non-treated and treated F_1 females included tolerant or sensitive lines to perform log-likelihood ratio test for significance of the stress effect on recombination parameters in stress-sensitive and stress-tolerant F_1 (supplementary text S1).

RESULTS

Adult flies from the *T*-lines were previously shown to have significantly higher desiccation tolerance compared to those from the parental *S*-lines [23,28]. We assumed that this difference holds also at the larval level, and passes through generations; however, these assumptions had to be tested explicitly. We found that larvae from the parental *T*-lines were significantly more tolerant compared to those of the parental *S*-lines: 1.56-fold increase for LT_{100} ($F_{1, 282}$=930.85, P<0.0001), and 1.57-fold increase for LT_{50} ($F_{1, 282}$=709.86, P<0.0001), where LT_{100} and LT_{50} are times to 100% and 50% levels of mortality under dry air, respectively (Figure 2, A). The F_1 larvae obtained from the parental lines in crosses with the standard multiple-marker strain *ru cu ca* demonstrated the same pattern: 1.42-fold increase for LT_{100} ($F_{1, 282}$=729.78, P<0.0001), and 1.34-fold for LT_{50} ($F_{1, 282}$=582.04, P<0.0001) (Figure 2, B). These results indicate that using the terms *S* and *T* is valid not only for the parental lines but also for their F_1 hybrids with the marker strain.

To test whether plasticity of recombination rate and interference shows fitness dependence, we compared the following hypotheses: (a) H_0 – the treatment does not affect both the *S*- and *T*-lines; (b) H_1 – the treatment affects the *S*- but not the *T*-lines; (c) H_2 – the treatment affects the *T*- but not the *S*-lines; and (d) H_3 – the treatment affects both the *S*- and *T*-lines (supplementary text S1).

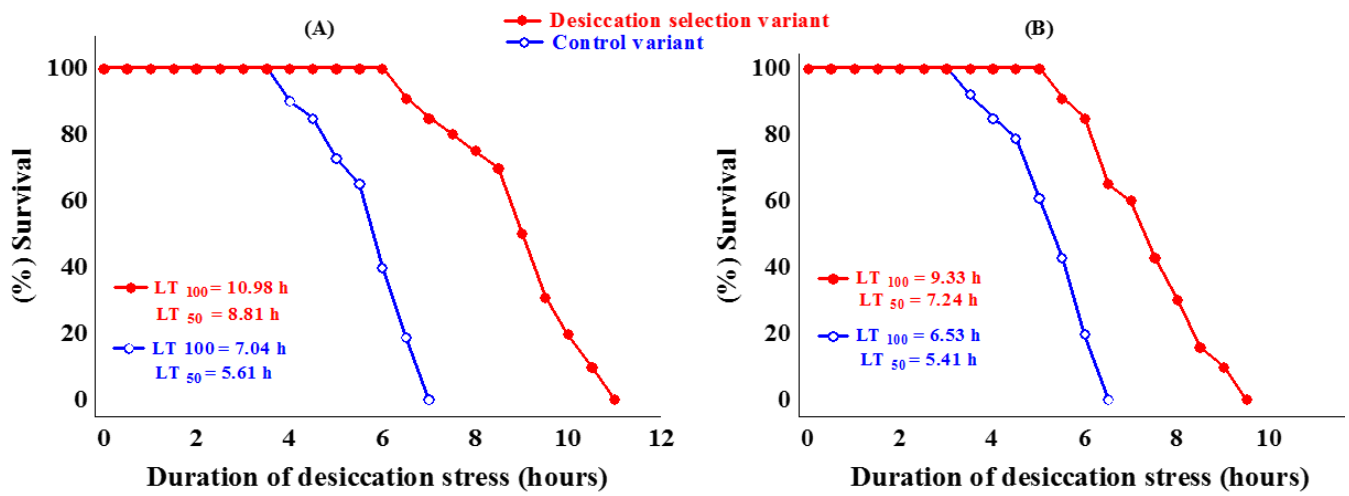

Figure 2: Larval desiccation tolerance in the parental lines (A), and in their F_1 hybrids with the *ru cu ca* marker strain (B)

With respect to plasticity of recombination rate, we found that for two out of five examined intervals (*h-th* and *cu-sr*), H_1 but not H_2 significantly differed from H_0 (P=1.45×10^{-3} and 6.51×10^{-3}, respectively; after correction for multiple tests, P=0.024 and 0.039, respectively). For three other intervals, neither H_1 nor H_2 significantly differed from H_0 (Table 1). With respect to plasticity of interference, the effect held for the 3L but not for the 3R arm: for two out of four examined pairs of adjacent intervals (*ru-h-th* and *h-th-cu*), H_1 but not H_2 significantly differed from H_0 (P=1.43×10^{-2} and 2.72×10^{-3}, respectively; after correction for multiple tests, P=0.064 and 0.024, respectively). For two other pairs, neither H_1 nor H_2 significantly differed from H_0 (Table 2). Given that two intervals (*th–cu* and *sr–e*) are small, the expected rates of double crossovers with their adjacent intervals are low. Thus, we additionally examined desiccation-induced changes in interference in some derivative intervals (supplementary table S2). In general, the noticed difference in interference reaction to treatment between the arms was quite robust. Interestingly, in pericentomeric regions desiccation relaxed positive interference in the S-lines (increase in double-crossover frequency) while aggravated it in the *T*-lines (decrease in double-crossover frequency); together, these two opposite effects sometimes result in highly significant difference between H_3 and H_0 (e.g. in case of *h–th–sr* pair of intervals).

The obtained results indicate that desiccation stress affects both recombination rate and interference. Furthermore, wherever recombination rate and interference are plastic, their plasticity tends to depend on fitness in a negative-feedback way, i.e. higher reactivity is associated with lower fitness.

Table 1: The effects of desiccation on recombination rates in chromosome 3 in desiccation-sensitive and desiccation-tolerant *Drosophila* lines

Interval	Crossover frequency, %				Competing hypotheses*								
	S-lines		*T*-lines		H_1 vs H_0			H_2 vs H_0			H_3 vs H_0		
	Non-treated	Treated	Non-treated	Treated	χ^2 df=3	*P*	*P*(fdr)	χ^2 df=3	*P*	*P*(fdr)	χ^2 df=6	*P*	*P*(fdr)
ru-h 1-2	18.84±0.82	20.13±0.85	20.76±0.85	23.16±0.89	3.48	0.323	0.484	4.53	0.209	0.363	8.01	0.237	0.395
h-th 2-3	14.49±0.74	18.67±0.82	21.20±0.86	22.40±0.88	15.48	1.45×10^{-3}	0.024	4.41	0.221	0.363	19.90	2.89×10^{-3}	8.7×10^{-3}
th-cu 3-4	5.64±0.49	4.89± 0.45	4.93± 0.46	4.31±0.43	1.59	0.662	0.794	1.45	0.694	0.794	3.03	0.805	0.805
cu-sr 4-5	8.22±0.58	11.16±0.66	9.42± 0.62	10.67±0.65	12.27	6.51×10^{-3}	0.039	7.17	0.067	0.200	19.44	3.48×10^{-3}	8.7×10^{-3}
sr-e 5-6	4.98±0.46	4.49±0.44	6.04± 0.50	5.16±0.47	0.91	0.823	0.823	4.39	0.222	0.363	5.30	0.481	0.601

***** H_0 = the treatment does not affect crossover rate in both the *S*- and *T*-lines; H_1 = the treatment affects crossover rate in the *S*- but not in the *T*-lines; H_2 = the treatment affects crossover rate in the *T*- but not in the *S*-lines; H_3 = the treatment affects crossover rate in both *S*- and *T*-lines

Table 2: The effect of desiccation on interference in chromosome 3 in desiccation-sensitive and desiccation-tolerant *Drosophila* lines

Interval	Coefficient of coincidence				Competing hypotheses*								
	S-lines		*T*-lines		H_1 vs H_0			H_2 vs H_0			H_3 vs H_0		
	Non-treated	Treated	Non-treated	Treated	χ^2 df=3	*P*	*P*(fdr)	χ^2 df=3	*P*	*P*(fdr)	χ^2 df=6	*P*	*P*(fdr)
ru-h-th 1-2-3	0.816±0.099	1.207±0.092	1.101±0.081	1.415±0.077	10.57	1.43×10^{-2}	0.064	9.08	2.82×10^{-2}	0.101	19.65	3.20×10^{-3}	0.013
h-th-cu 2-3-4	0.111±0.077	0.646±0.163	0.693±0.156	0.815±0.172	14.14	2.72×10^{-3}	0.024	0.92	0.821	0.823	15.06	1.98×10^{-2}	0.040
th-cu-sr 3-4-5	0.200±0.136	0.499±0.193	0.500±0.211	0.301±0.167	4.44	0.218	0.363	6.05	0.109	0.280	10.49	0.105	0.140
cu-sr-e 4-5-6	0.107±0.107	0.378±0.177	0.234±0.134	0.407±0.175	1.87	0.598	0.794	1.39	0.706	0.794	3.26	0.773	0.773

***** H_0 = the treatment does not affect crossover interference in both the *S*- and *T*-lines; H_1 = the treatment affects interference in the *S*- but not in the *T*-lines; H_2 = the treatment affects interference in the *T*- but not in the *S*-lines; H_3 = the treatment affects interference in both *S*- and *T*-lines

DISCUSSION

Desiccation as recombinogenic factor

In the *S*-lines, desiccation significantly raised crossover frequency in two out of the five examined intervals: *h-th* and *cu-sr*. Both intervals are close to the centromere; recombination in such regions is long known to be highly regulated, as well as highly reactive with respect to environmental stressors [2,10]. Our results indicate that desiccation is a recombinogenic factor; to our knowledge this has been shown earlier only for maize [29]. Biochemical processes behind the revealed recombinogenic effect of desiccation should obviously be a subject to further thorough investigations; general speculations may be the following.

A key damaging factor of desiccation is the oxidative stress caused by reactive oxygen species (ROS) emerging in low-water intracellular environment [30–33]. The ROS are commonly recognized to cause DNA damages by attacking both bases and deoxyribosyl backbone, either directly or via reactive intermediates of lipid peroxidation (reviewed in [34–36]). To a certain extent, ROS are scavenged with antioxidants, which are universal across life forms and include glutathione, ascorbic acid, and tocopherols [37–39]. Apparently, the capacity of this preventive mechanism has a limit, and the oxidative DNA damages do occur. Increase in DNA damages under desiccation is long known for prokaryotes [40–43]. Recently, analogous results have been reported for plants [44,45], rotifers [46], tardigrades [47,48], and even for Diptera [49]. The desiccation-induced DNA damage is subject to DNA repair. This process is shown to intensify under dehydration in different species [44,45]. In *D. melanogaster*, higher efficacy of DNA repair is observed in organisms with higher thermotolerance [50]. Finally, the intensified DNA repair, mediated, inter alia, by increased expression of *rad23*, and later on *rad51*, induces recombination [49,51].

Also, in the *S*-lines desiccation caused significant increase in double-crossover rates in two of the four examined pairs of intervals: *ru-h-th* and *h-th-cu*. This result is in line with the heat-induced relaxation of interference observed earlier in the X-chromosome [10,20], and in chromosomes 2 and 3 [10] of *Drosophila.* Whether or not changes in interference are necessarily coupled with those in recombination is a hot topic discussed for decades [10,23,52–56]. Indeed, higher recombination rates are often, but not always, associated with relaxation of positive interference and even appearance of negative interference (recently reviewed in Aggarwal *et al.*, 2015). Herein observed increase in double-crossover rates in the *S*-lines did not strictly coincide with increase in crossover frequencies in one or both adjacent intervals, in line with results of some earlier studies [10,52]. Thus, we do not

automatically derive the former from the latter. Yet, we still consider changes in recombination and interference as having the same nature, both being manifestations of meiosis deregulation, like in meiotic mutants which display much less regulated recombination, including more uniform distribution of crossover frequency along chromosomes and more relaxed positive crossover interference and crossover homeostasis [57–61]. However, this conclusion is based mainly on measurements of recombination/interference parameters and mechanistic considerations rather than on direct tests of changes in their variation.

Fitness dependence of desiccation-induced changes in recombination rate and interference

We consider the revealed difference between *S*- and *T*-lines in terms of their recombination/interference response to desiccation as an evidence for fitness-dependent plasticity. Earlier, fitness-dependent increase in recombination in fruit flies was reported in relation to other environmental stressors. First, Kilias *et al.* [15] observed higher crossover frequencies in a *Drosophila* line with low activity of alcohol dehydrogenase compared to another, high-activity line. The authors suggested that the former strain had lower fitness expressed as higher sensitivity to alcohol intoxication. However, given that alcohols are capable to cause DNA damages, it is impossible to distinguish between the stressor's effect on 'somatic' fitness and its direct effect on germline DNA metabolism. Later on, Korol *et al*. [6] revealed that heterozygous heat-sensitive *Drosophila* males showed, when heat-shocked, a several-fold higher increase in crossover frequency in chromosome 2 compared to heat-tolerant ones. Yet, recombination there could be both meiotic and pre-meiotic [62,63].

The first solid evidence for fitness dependence of stress-induced changes in meiotic recombination was provided by Zhong and Priest [14]. In their experiments, heat-treated *Drosophila* females showed increase in crossover frequency that negatively correlated with their heat-tolerance measured as productivity. Also, negative recombination-fitness correlation was recently reported by Jackson *et al*. [11]; however, they used the same genotype and watched, in fact, fitness variation within the reaction norm rather than plasticity variation across genotypes (which would be more relevant to the discussed problem of recombination-fitness association). Finally, Hunter *et al*. [16], in their exhausting analysis of recombination variation across ~200 Drosophila Genetic Reference Panel (DGRP) lines uncovered negative correlation between crossover rate in the *y–v* interval (X-chromosome) and flies' response to some chemicals. One of them, paraquat, is a model inducer of oxidative stress; thus, paraquat tolerance indeed seems an adequate measure for fitness. Yet, given that phenotype traits were

measured on the DGRP lines while recombination – on their F_1 hybrids with marker stocks, the correlation values may be biased. Remarkably, negative recombination-fitness correlation was reported in studies where fitness variation across genotypes came from their different genetic backgrounds rather than from different tolerance to an environmental stressor [64,65]. Similar pattern (negative association with fitness) was demonstrated for other processes affecting genetic variation, such as mutation [66] and segregation [67].

The revealed desiccation-induced increase of double crossovers in the *S*- but not in *T*-lines is the first, to the best of our knowledge, empirical evidence for fitness dependence of interference plasticity. An alternative explanation for the herein observed difference between the *S*- and *T*-lines in terms of their recombination and interference responses to desiccation is that high recombination rates, observed in the *T*-lines, reflect the values needed to ensure desiccation tolerance and, thus, could not be further increased. However, in the *S*-lines desiccation stress sometimes raised recombination rates even to higher values compared to the average level of the *T*-lines (e.g. in case of *cu–sr* interval). Due to the limited number of examined intervals, it would be reasonable to address the association between plasticity of recombination rate and interference, on one hand, and fitness, on the other, in further tests, with considerably higher number of intervals and genotypes.

Evolvability of fitness dependence

Fitness dependence of recombination can be considered as an evolvable feature. Its evolution can be driven by two forces. First, if fitness-dependent recombination is beneficial to the selected system, modifier alleles ensuring such strategy can be subjected to indirect selection, exactly as it happens in case of indirect selection for recombination rate [68,69]. Second, such modifier alleles can spread by hitchhiking, regardless of their effects on the selected system (the 'abandon-ship' mechanism) [70]. The latter mechanism seems to be inefficient in diploids. The authors of the 'abandon-ship' hypothesis demonstrated, by using an analytical model, that fitness-dependent recombination evolved in diploids due to selection for mean recombination rate rather than for recombination rate plasticity [70]. Yet, in our recent simulations analysis with diploid selection, fitness-dependent recombination was favored under different selection regimes, such as cyclical selection [71], Red Queen dynamics [72] and mutation-selection balance [73]. At that, the evolutionary advantage/disadvantage of fitness-dependent recombination is determined by trade-off between two opposite effects: benefits of protecting best allele combinations, and costs of shifting population's mean recombination rate [73]. Not rarely, the cost

overweight the benefits, which may be one of the reasons why several experimental-evolution studies reported no selection for recombination rate plasticity [74,75].

Fitness-dependent interference may be subject to indirect selection as well. Its evolvability is demonstrated in a modifier model by Goldstein *et al*. [76]. Recent theoretical models also support the evolvability of fitness dependence of other processes affecting genetic variation: sex [77], mutation [78], and dispersal [79]. This tempts us to conclude that evolvability of fitness dependence may be a widespread phenomenon in nature. The consistent evolutionary pattern is that generating *de novo* or releasing standing genetic variation is modulated by individual fitness in a negative-feedback way, i.e. lower individual fitness leads to higher genetic variation of the progeny [6]. Bauer put forward in a somewhat paradoxical form a similar idea that 'losers' rather than 'winners' in the struggle for existence provide the raw material for evolutionary change (cited according to [6]). The general reason for such an increased production of genetic variability can also be seen in the 'genomic stress' caused by external and internal factors [6,80–82]. How common is such a system of variability control, based on negative feedback, remains an open question deserving further studies.

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REFERENCES

1. Plough, H. H. 1917 The effect of temperature on crossingover in *Drosophila*. *J. Exp. Zool.* **24**, 147–209. (doi:10.1104/pp.4.2.281)
2. Plough, H. H. 1921 Further studies on the effect of temperature on crossing over. *J. Exp. Zool.* **32**, 187–202. (doi:10.1002/jez.1400320202)
3. Bomblies, K., Higgins, J. D. & Yant, L. 2015 Meiosis evolves: adaptation to external and internal environments. *New Phytol.* **208**, 306–323. (doi:0.1111/nph.13499)
4. Modliszewski, J. L. & Copenhaver, G. P. 2017 Meiotic recombination gets stressed out: CO frequency is plastic under pressure. *Curr. Opin. Plant Biol.* **36**, 95–102. (doi:10.1016/j.pbi.2016.11.019)

5. Stevison, L. S., Sefick, S., Rushton, C. & Graze, R. M. 2017 Recombination rate plasticity: revealing mechanisms by design. *Philos. Trans. R. Soc. Lond. B. Biol. Sci.* **372**, 20160459. (doi:10.1098/rstb.2016.0459)
6. Korol, A. B., Preygel, I. A. & Preygel, S. I. 1994 *Recombination Variability and Evolution*. London: Chapman & Hall.
7. Stern, C. 1926 An effect of temperature and age on crossing-over in the first chromosome of Drosophila melanogaster. *Proc. Natl. Acad. Sci. U. S. A.* **12**, 530–532.
8. Graubard, M. A. 1932 Inversion in Drosophila melanogaster. *Genetics* **17**, 81–105.
9. Politzer, O. 1940 Veränderungen der Crossoverhäufigkeit Durch Einwirkung von Temperatur und Alter (Versuche am III. Chromosom der Drosophila melanogaster). *Z. Indukt. Abstamm. Vererbungsl.* **78**, 129–147.
10. Grell, R. F. 1978 A comparison of heat and interchromosomal effects on recombination and interference in Drosophila melanogaster. *Genetics* **89**, 65–77.
11. Jackson, S., Nielsen, D. M. & Singh, N. D. 2015 Increased exposure to acute thermal stress is associated with a non-linear increase in recombination frequency and an independent linear decrease in fitness in Drosophila. *BMC Evol. Biol.* **15**, 175. (doi:10.1186/s12862-015-0452-8)
12. Neel, J. V 1941 A relation between larval nutrition and the frequency of crossing over in the third chromosome of Drosophila melanogaster. *Genetics* **26**, 506–516.
13. Zhuchenko, A. A., Korol, A. B., Gavrilenko, T. A. & Kibenko, T. Y. 1986 The relation between genotype adaptivity and reactivity of its recombination characteristics under temperature effect. *Genetika* **22**, 966–974 (in Russian).
14. Zhong, W. & Priest, N. K. 2011 Stress-induced recombination and the mechanism of evolvability. *Behav. Ecol. Sociobiol.* **65**, 493–502. (doi:10.1007/s00265-010-1117-7)
15. Kilias, G., Alahiotis, S. N. & Onoufriou, A. 1979 The alcohol dehydrogenase locus affects meiotic crossing-over in *Drosophila melanogaster*. *Genetica* **50**, 173–177.
16. Hunter, C. M., Huang, W., Mackay, T. F. C. & Singh, N. D. 2016 The genetic architecture of natural variation in recombination rate in *Drosophila melanogaster*. *PLoS Genet.* **12**, e1005951. (doi:10.1371/journal.pgen.1005951)
17. Segura, J. et al. 2013 Evolution of recombination in eutherian mammals: insights into mechanisms that affect recombination rates and crossover interference. *Proc. R. Soc. B Biol. Sci.* **280**, 1945. (doi:10.1098/rspb.2013.1945)

18. Bomblies, K., Jones, G., Franklin, C., Zickler, D. & Kleckner, N. 2016 The challenge of evolving stable polyploidy: could an increase in 'crossover interference distance' play a central role? *Chromosoma* **125**, 287–300. (doi:10.1007/s00412-015-0571-4)
19. Graubard, M. A. 1934 Temperature effect on interference and crossing over. *Genetics* **19**, 83–94.
20. Hayman, D. L. & Parsons, P. A. 1960 The effect of temperature, age and an inversion on recombination values and interference in the X-chromosome of Drosophila melanogaster. *Genetics* **32**, 74–88.
21. Matzkin, L. M., Watts, T. D. & Markow, T. A. 2009 Evolution of stress resistance in Drosophila: interspecific variation in tolerance to desiccation and starvation. *Funct. Ecol.* **23**, 521–527. (doi:10.1111/j.1365-2435.2008.01533.x)
22. Rajpurohit, S., Nedved, O. & Gibbs, A. G. 2013 Meta-analysis of geographical clines in desiccation tolerance of Indian drosophilids. *Comp. Biochem. Physiol. A Mol. Integr. Physiol.* **164**, 391–398. (doi:10.1016/j.cbpa.2012.11.013)
23. Aggarwal, D. D., Rashkovetsky, E., Michalak, P., Cohen, I., Ronin, Y. I., Zhou, D., Haddad, G. G. & Korol, A. B. 2015 Experimental evolution of recombination and crossover interference in *Drosophila* caused by directional selection for stress-related traits. *BMC Biol.* **13**, 101. (doi:10.1186/s12915-015-0206-5)
24. Charlesworth, B. 1993 Directional selection and the evolution of sex and recombination. *Genet. Res.* **61**, 205–224. (doi:10.1017/S0016672300031372)
25. Burt, A. 2000 Perspective: sex, recombination, and the efficacy of selection - was Weismann right? *Evolution (N. Y).* **54**, 337–351. (doi:10.1111/j.0014-3820.2000.tb00038.x)
26. Chandley, A. C. 1968 The effect of X-rays on female germ cells of *Drosophila melanogaster*. III. A comparison with heat-treatment on crossing-over in the X-chromosome. *Mutat. Res.* **5**, 93–107. (doi:10.1017/CBO9781107415324.004)
27. Markow, T. A., Matzkin, L. M. & Watts, T. D. 2007 Desiccation resistance in four *Drosophila* species: Sex and population effects. *Fly (Austin).* **1**, 268–273. (doi:5293 [pii])
28. Kang, L., Aggarwal, D. D., Rashkovetsky, E., Korol, A. B. & Michalak, P. 2016 Rapid genomic changes in Drosophila melanogaster adapting to desiccation stress in an experimental evolution system. *BMC Genomics* **17**, 233. (doi:10.1186/s12864-016-2556-y)
29. Verde, L. A. 2003 The Effect of Stress on Meiotic Recombination in Maize (*Zea mays* L.).
30. Finkel, T. & Holbrook, N. J. 2000 Oxidants, oxidative stress and the biology of ageing. *Nature*

408, 239–247. (doi:10.1038/35041687)

31. Walters, C., Farrant, J. M., Pammenter, N. W. & Berjak, P. 2002 Desiccation stress and damage. In *Desiccation and Survival in Plants: Drying Without Dying* (eds M. Black & H. W. Pritchard), pp. 263–292. Wallingford: CABI.
32. Kranner, I. & Birtić, S. 2005 A modulating role for antioxidants in desiccation tolerance. *Integr. Comp. Biol.* **45**, 734–740. (doi:10.1093/icb/45.5.734)
33. Caramelo, J. J. & Iusem, N. D. 2009 When cells lose water: lessons from biophysics and molecular biology. *Prog. Biophys. Mol. Biol.* **99**, 1–6. (doi:10.1016/j.pbiomolbio.2008.10.001)
34. Marnett, L. J. 2000 Oxyradicals and DNA damage. *Carcinogenesis* **21**, 361–370. (doi:10.1093/carcin/21.3.361)
35. Valko, M., Izakovic, M., Mazur, M., Rhodes, C. J. & Telser, J. 2004 Role of oxygen radicals in DNA damage and cancer incidence. *Mol. Cell. Biochem.* **266**, 37–56. (doi:10.1023/B:MCBI.0000049134.69131.89)
36. von Sonntag, C. 2006 *Free-Radical-Induced DNA Damage and Its Repair*. Berlin et al.: Springer. (doi:10.1007/3-540-30592-0)
37. Potts, M. 2001 Desiccation tolerance: a simple process? *Trends Microbiol.* **9**, 553–559.
38. Crowe, J. oh. H., Oliver, A. E. & Tablin, F. 2002 Is there a single biochemical adaptation to anhydrobiosis? *Integr. Comp. Biol.* **42**, 497–503. (doi:10.1093/icb/42.3.497)
39. Berjak, P. 2006 Unifying perspectives of some mechanisms basic to desiccation tolerance across life forms. *Seed Sci. Res.* **16**, 1–15. (doi:10.1079/SSR2005236)
40. Dose, K. & Gill, M. 1995 Dna stability and survival of bacillus subtilis spores in extreme dryness. *Orig. Life Evol. Biosph.* **25**, 277–293. (doi:10.1007/BF01581591)
41. Mattimore, V. & Battista, J. R. 1996 Radioresistance of Deinococcus radiodurans: Functions necessary to survive ionizing radiation are also necessary to survive prolonged desiccation. *J. Bacteriol.* **178**, 633–637. (doi:10.1111/j.1365-2958.1994.tb00397.x)
42. Zahradka, K., Slade, D., Bailone, A., Sommer, S., Averbeck, D., Petranovic, M., Lindner, A. B. & Radman, M. 2006 Reassembly of shattered chromosomes in Deinococcus radiodurans. *Nature* **443**, 569–73. (doi:10.1038/nature05160)
43. Pitcher, R. S., Green, A. J., Brzostek, A., Korycka-Machala, M., Dziadek, J. & Doherty, A. J. 2007 NHEJ protects mycobacteria in stationary phase against the harmful effects of desiccation. *DNA Repair (Amst).* **6**, 1271–1276. (doi:10.1016/j.dnarep.2007.02.009)

44. Boubriak, I. I., Grodzinsky, D. M., Polischuk, V. P., Naumenko, V. D., Gushcha, N. P., Micheev, A. N., McCready, S. J. & Osborne, D. J. 2008 Adaptation and impairment of DNA repair function in pollen of Betula verrucosa and seeds of Oenothera biennis from differently radionuclide-contaminated sites of Chernobyl. *Ann. Bot.* **101**, 267–276. (doi:10.1093/aob/mcm276)

45. Huang, Z., Boubriak, I., Osborne, D. J., Dong, M. & Gutterman, Y. 2008 Possible role of pectin-containing mucilage and dew in repairing embryo DNA of seeds adapted to desert conditions. *Ann. Bot.* **101**, 277–283. (doi:10.1093/aob/mcm089)

46. Gladyshev, E. A. & Arkhipova, I. R. 2010 Genome structure of bdelloid rotifers: Shaped by asexuality or desiccation? *J. Hered.* **101**, 85–93. (doi:10.1093/jhered/esq008)

47. Neumann, S., Reuner, A., Brümmer, F. & Schill, R. O. 2009 DNA damage in storage cells of anhydrobiotic tardigrades. *Comp. Biochem. Physiol. A Mol. Integr. Physiol.* **153**, 425–429. (doi:10.1016/j.cbpa.2009.04.611)

48. Rebecchi, L., Cesari, M., Altiero, T., Frigieri, A. & Guidetti, R. 2009 Survival and DNA degradation in anhydrobiotic tardigrades. *J. Exp. Biol.* **212**, 4033–4039. (doi:10.1242/jeb.033266)

49. Gusev, O. et al. 2010 Anhydrobiosis-associated nuclear DNA damage and repair in the sleeping chironomid: Linkage with radioresistance. *PLoS One* **5**, e14008. (doi:10.1371/journal.pone.0014008)

50. Lupu, A., Pechkovskaya, A., Rashkovetsky, E., Nevo, E. & Korol, A. 2004 DNA repair efficiency and thermotolerance in Drosophila melanogaster from 'Evolution Canyon'. *Mutagenesis* **19**, 383–390. (doi:10.1093/mutage/geh045)

51. Beltrán-Pardo, E. A., Jönsson, I., Wojcik, A., Haghdoost, S., Harms-Ringdahl, M., Bermúdez Cruz, R. M. & Bernal Villegas, J. E. 2013 Sequence analysis of the DNA-repair gene rad51 in the tardigrades Milnesium cf. tardigradum, Hypsibius dujardini and Macrobiotus cf. harmsworthi. *J. Limnol.* **72**, 80–91. (doi:10.4081/jlimnol.2013.s1.e10)

52. Denell, R. E. & Keppy, D. O. 1979 The nature of genetic recombination near the third chromosome centromere of *Drosophila melanogaster*. *Genetics* **93**, 117–130.

53. Foss, E., Lande, R., Stahl, F. W. & Steinberg, C. M. 1993 Chiasma interference as a function of genetic distance. *Genetics* **133**, 681–691.

54. Fujitani, Y., Mori, S. & Kobayashi, I. 2002 A reaction-diffusion model for interference in meiotic crossing over. *Genetics* **161**, 365–372.

55. Zhang, L., Liang, Z., Hutchinson, J. & Kleckner, N. 2014 Crossover patterning by the Beam-Film

model: analysis and implications. *PLoS Genet.* **10**. (doi:10.1371/journal.pgen.1004042)

56. Zickler, D. & Kleckner, N. 2016 A few of our favorite things: Pairing, the bouquet, crossover interference and evolution of meiosis. *Semin. Cell Dev. Biol.* **54**, 135–148. (doi:10.1016/j.semcdb.2016.02.024)
57. Baker, B. S. & Hall, J. C. 1976 Meiotic mutants: genie control of meiotic recombination and chromosome segregation. In *The Genetics and Biology of Drosophila* (eds M. Ashburner & E. Novitski), pp. 351–434. New York: Academic Press.
58. Szauter, P. 1984 An analysis of regional constraints on exchange in Drosophila melanogaster using recombination-defective meiotic mutants. *Genetics* **106**, 45–71.
59. Zhuchenko, A. A., Korol, A. B. & Kovtyukh, P. L. 1985 Change of the crossing-over frequency in Drosophila during selection for resistance to temperature fluctuations. *Genetica* **67**, 73–78. (doi:10.1007/BF02424463)
60. C, Z. M. & Rose, A. M. 1995 Mutant rec-1 eliminates the meiotic pattern of crossing over in Caenorhabditis elegans. *Genetics* **141**, 1339–1349.
61. Séguéla-Arnaud, M. et al. 2015 Multiple mechanisms limit meiotic crossovers: TOP3α and two BLM homologs antagonize crossovers in parallel to FANCM. *Proc. Natl. Acad. Sci. U. S. A.* **112**, 4713–4718. (doi:10.1073/pnas.1423107112)
62. Hiraizumi, Y. 1971 Spontaneous recombination in *Drosophila melanogaster* males. *Proc. Natl. Acad. Sci. U. S. A.* **68**, 268–270. (doi:10.1073/pnas.68.2.268)
63. Woodruff, R. C. & Thompson, J. N. 1977 An analysis of spontaneous recombination in Drosophila melanogaster males. *Heredity (Edinb).* **38**, 291–307. (doi:10.1038/hdy.1977.92)
64. Tucić, N., Ayala, F. J. & Marinković, D. 1981 Correlation between recombination frequency and fitness in Drosophila melanogaster. *Genetica* **56**, 61–69. (doi:10.1007/BF00126931)
65. Tedman-Aucoin, K. & Agrawal, A. F. 2012 The effect of deleterious mutations and age on recombination in *Drosophila melanogaster*. *Evolution (N. Y).* **66**, 575–585. (doi:10.1111/j.1558-5646.2011.01450.x)
66. Sharp, N. P. & Agrawal, A. F. 2012 Evidence for elevated mutation rates in low-quality genotypes. *Proc. Natl. Acad. Sci. U. S. A.* **109**, 6142–6146. (doi:10.1073/pnas.1118918109)
67. Singh, N. D., Criscoe, D. R., Skolfield, S., Kohl, K. P., Keebaugh, E. S. & Schlenke, T. A. 2015 Fruit flies diversify their offspring in response to parasite infection. *Science (80-.).* **349**, 747–750.
68. Kimura, M. 1956 A model of a genetic system which leads to closer linkage by natural selection.

Evolution (N. Y). **10**, 278–287. (doi:10.1111/j.1558-5646.1956.tb02852.x)

69. Nei, M. 1967 Modification of linkage intensity by natural selection. *Genetics* **57**, 625–641.
70. Agrawal, A. F., Hadany, L. & Otto, S. P. 2005 The evolution of plastic recombination. *Genetics* **171**, 803–812. (doi:10.1534/genetics.105.041301)
71. Rybnikov, S. R., Frenkel, Z. M. & Korol, A. B. 2017 What drives the evolution of condition-dependent recombination in diploids? Some insights from simulation modelling. *Philos. Trans. R. Soc. B* **372**, 20160460. (doi:10.1098/rstb.2016.0460)
72. Rybnikov, S. R., Frenkel, Z. M. & Korol, A. B. 2017 Recombination rate coevolution in a quantitative trait model with diploid antagonists. In *Mathematical Models in Ecology and Evolution (book of abstracts)*, pp. 82–83. London: City University.
73. Rybnikov, S. R., Frenkel, Z. M. & Korol, A. B. 2018 Fitness - dependent recombination can be evolutionarily advantageous in diploids□: a deterministic mutation-selection balance model. *bioRxiv* , 1–21. (doi:10.1101/381228)
74. Kerstes, N. A., Bérénos, C., Schmid-Hempel, P. & Wegner, K. M. 2012 Antagonistic experimental coevolution with a parasite increases host recombination frequency. *BMC Evol. Biol.* **12**, 18. (doi:10.1186/1471-2148-12-18)
75. Kohl, K. P. & Singh, N. D. 2018 Experimental evolution across different thermal regimes yields genetic divergence in recombination fraction but no divergence in temperature associated plastic recombination. *Evolution (N. Y).* **72**, 989–999. (doi:10.1111/evo.13454)
76. Goldstein, D. B., Bergman, A. & Feldman, M. W. 1993 The evolution of interference: reduction of recombination among three loci. *Theor. Popul. Biol.* **44**, 246–259.
77. Hadany, L. & Otto, S. P. 2007 The evolution of condition-dependent sex in the face of high costs. *Genetics* **176**, 1713–1727. (doi:10.1534/genetics.107.074203)
78. Shaw, F. H. & Baer, C. F. 2011 Fitness-dependent mutation rates in finite populations. *J. Evol. Biol.* **24**, 1677–1684. (doi:10.1111/j.1420-9101.2011.02320.x)
79. Gueijman, A., Ayali, A., Ram, Y. & Hadany, L. 2013 Dispersing away from bad genotypes: the evolution of fitness-associated dispersal (FAD) in homogeneous environments. *BMC Evol. Biol.* **13**, 125. (doi:10.1186/1471-2148-13-125)
80. McClintock, B. 1984 The significance of response of the genome to challenge. *Science (80-.).* **226**, 792–801. (doi:http://dx.doi.org/10.1126/science.15739260)
81. Hoffmann, A. A. & Parsons, P. A. 1991 *Evolutionary Genetics and Environmental Stres.* New

York: Oxford University Press.

82. Thaler, D. S. 1994 The evolution of genetic intelligence. *Science (80-.).* **264**, 224–225.