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Fitness and Crossing Over Frequency of the Mutants *Drosophila melanogaster* in Dependence upon Age and Artificial Reorganizations of a Genotype

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Abstract: The objective of the given research is to study changes in crossing over frequency and state of main components of fitness at linear flies *D. Melanogaster* in dependence on age and experimentally created modifications in the genotype. Research techniques are traditional and well applied in genetics methods of revealing main components of fitness *D. melanogaster* in particular fertility, survival in extreme conditions (high temperature exposure and starvation), life expectancy etc., are used. Beside before mentioned physiological components of fitness genetic (defining crossing over frequency) and genetic biochemical (defining functional state of gene-enzymatic system Adh) characteristics of fitness are also applied. Crossing over frequency, activity and thermal resistance have been defined with the help of common methods. Being guided and published in special literature data we have used these characteristics to evaluate viability and general fitness of *D. melanogaster*. Reliability of the obtained data has been evaluated according to student criterion ($p<0.05$). The analysis result prove that fertility differs in mutants according to the age except the wild type which has more ability in fertility and unchangeable with age and all the mutants lines of *Drosophila melanogaster* (b, cn, cn and vg) have more fertility than the others. The data of showed that the mutant liens of *Drosophila maelanogaster* (b, cn, cn and vg) have more fertility than the other mutants. This fact should be taken into consideration while creating new genotypes through outbreeding, genetic engineering and while developing genetic theories of maturation and aging.

Key words: Fitness, crossing over, frequency of the mutants, *Drosophila melanogaster*, age and artificial reorganizations, a genotype

INTRODUCTION

Wide usage of induced mutagenesis and outbreeding in genetics, introduction of genetic engineering and other biotechnological techniques stirs interest towards the problem of studies in the field of adaptive abilities of gene modified genotypes. Studies into changes in fitness of genotypes in the process of aging which is also accompanied with changes in genetic balance caused by accumulation of mutant and other changes in genotype are in no case less important (Winkler *et al*., 2005).

In all cases of gene interaction in modified genetic environment, it may considerably influence realization of genetic information including establishment of such components of fitness as fertility, viability and life expectancy, etc. It should be mentioned that studying functions of isolated genes in modified genetic background favors deepening and detailed rendering of known theories and hypotheses about co-adapted genes, compensating and adapting gene complexes (Haber *et al*., 2005). All these theories and hypotheses, importance of which for genetics and selection cannot be overestimated require molecular-genetic and genetic-biochemical rendering. Unfortunately, researchers have scarce knowledge about influence of natural and artificial modification of genotypes upon before mentioned gene units and complexes (Garcia *et al*., 2010).

Understanding the properties of new mutations is critical to a broad range of evolutionary theory including models relating to the maintenance of genetic variation in the face of selection (Lande, 1975; Barton, 1986; Keightley and Halligan , 2009), the persistence of small populations (Lande, 1995) and the advantages of sexual reproduction (Agrawal and Chasnov, 2001; Haag and Roze, 2007; Salathe *et al*., 2006). Accordingly, spontaneous mutation has been the focus of numerous experimental studies (Keightley and Halligan, 2009; Halligan and Keightley, 2009; Keightley, 1996; Drake *et al*., 1998; Simmons and Crow, 1977) particularly with the fruit fly, *Drosophila melanogaster*. Mutation Accumulation (MA) experiments in which new mutations are allowed to fix by removing selection have typically

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measured changes in juvenile viability (egg-to-adult survival) as an indicator of total fitness. Adult survival and reproductive success (adult fitness) will often be important contributors to total fitness yet have been much less studied in MA experiments. Adult fitness is important to the understanding of mutation for several reasons. For many populations, it is thought that sexual selection is a stronger force than viability selection (Hoekstra *et al*., 2001). Moreover, variation in juvenile growth rather than survival can have carryover effects to adult size, condition and the realization of adult fitness.

This would imply that mutation pressure on total fitness could be much greater than studies examining juvenile fitness alone would imply. Because reproductively mature individuals typically express the most pronounced sex-differences in phenotype, implying divergence in selection pressures, the consequences of new mutations could be sex-specific. On the other hand, the expression of sexually selected traits may still share a common genetic basis between the sexes as their expression is thought to depend on the overall health and vigor of the individual (the genic capture hypothesis) (Tomkins *et al*., 2004; Rowe and Houle, 1996).

Thus, given facts prove that investigation into expressiveness of isolated structuring genes, crossing over frequency and phenological characteristics of fitness in genetically modified genotypes in dependence on age of individuals is a topical problem in genetics which is of general biological importance.

MATERIALS AND METHODS

Researchers are carried out on lines *D. melanogaster*, homozygotic in chromosome 2 where gene *Adh* and marking mutation b, cn and vg are localized. Single (b, cn and vg) double (b cn, cn and vg) and triple (b, cn and vg) mutants are studied as well as flies of wild type (C-S) first inbreeded within twenty generations. The genotype of the b, cn and vg triple mutant is changed in two directions; though saturation of this genotype with genes of line C-S with the help of saturating crosses and through replacement of mutant chromosome 1 into corresponding chromosome of flies of line C-S after the scheme by L.S. Luckinbill. Synthesized flies with modified genotypes are marked, respectively as b, cn and vg (C-S) and b, cn and vg (1 C-S).

Fitness of target mutants and flies with modified genotypes is evaluated defining main components of this feature; true fertility, life expectancy, ability to survive in adverse conditions and also level of combinatory activity at the site b, cn and vg and functioning state of gene enzymatic system Adh. Defining all these characteristics is carried out using common and well-probated methods. We estimate true fertility upon the number of descendents (imago) by one pair of flies, being in a flask (20 mL) for 3 days and nights. To define their ability to survive in high temperature conditions flies are heated for 15 min at 41°C (L_{t50} for the flies of wide type) after which ratio (percentage) of living flies to the number of all heated flies is calculated. Survival of investigated Drosophila in conditions of starvation is defined keeping flies of different age in flasks without any food and calculating the number of alive insects every 3 h. Results are shown in a number of hours necessary for death of 50% of flies. Life duration of insects is defined in days necessary for death of 50% of flies. Flies are kept in standard conditions with the addition of ethanol (10%).

Crossing over frequency is defined upon the number of combinatory descendents Fa by the test cross of triheterozygotes in marked genes *b*, *cn* and *vg* is expressed in percents. Activity Adh in extracts from Drisophila tissues is defined spectra photo metrically. Enzyme thermal resistance is determined after Chambers, Wilksand and Gibson. Obtained results are processed statistically with algorithms given by A.N. Plokhinskii and are estimated after student criterion.

RESULTS AND DISCUSSION

Marked mutations and fitness components at investigated flies of various ages: Following the data obtained experimentally investigated lines of flies can be divided into well adapted and badly adapted according to the state of certain fitness components (fertility, thermal resistance and life expectancy, etc.) Flies of wide type C-S have appeared to be best adapted.

Their fertility survival in high temperature conditions as well as life expectancy in standard conditions of keeping have been rather high and have been used in further investigations as controlling. Young cn mutants have turned to be close to the flies C-S upon their fitness characteristics. Fertility of cn females at young age (2-5 days) has appeared to be 90.5% of control level (C-S) but at mature age (6-9 days) before mentioned mutants have turned to be less fertile (57 % compared to C-S females at the same age). B and vg mutants have been even less fertile. Fertility of young females (2-5 days) of these lines is respectively 71 and 57% and at the age of 6-9 days, it is 54.6 and 31.3% of the level of C-S fly fertility at given age periods (Table 1). Young females (2-5 days) of double b and cn mutants have had the same low level of fertility as vg flies of the same age but fertility decrease with aging at b and cn flies was less significant (almost in 1.5 times). Thus, despite the fact that co-presence

Table 1: Fly fertility dependence on age of an individual, number of descendants by one pair n = 20-40 families

	Age (days)	
Lines	2-5	6-9
C-S	48.30±1.30	52.30±1.98
b	34.40±0.85⁺	28.60±1.66*⁺
cn	43.70±1.36⁺	30.20±0.80*⁺
vg	27.40±0.52⁺	16.40±0.46*⁺
b, cn	28.90±0.70⁺	24.10±0.46*⁺
b, vg	26.30±0.59⁺	20.70±0.43*⁺
cn, vg	36.00±0.39⁺	33.60±0.35*⁺
b, cn, vg	23.10±0.38⁺	19.90±0.43*⁺

*Differences are true in comparison with 2-5 days old flies. ⁺Differences are true in comparison with C-S flies

of b and cn mutations in the genotype shows more significant impact on fly fertility than single b and cn mutations. Impact of aging upon fertility of females of double b and cn mutants is less significant than upon fertility of single b, cn and vg mutants.

Mutants with vg mutation have had the lowest level of fertility. It refers to vg, b, vg and b, cn, vg mutants where beside low level of fertility we have observed more significant impact of aging upon this fitness component. On the contrary double cn and vg mutants have less significant pleiotrophic impact of mutations on fitness characteristics. The cn and vg fly fertility has been 1.3 times higher than that of single vg mutants and has decreased less intensively in dependence on the fly age. This higher fitness of cn and vg mutant in comparison with vg mutant can be explained with direct suppressing influence of the gene *cn* on the gene *cg* but mechanisms of such influence are still not clear. Significant impact of vg mutations on fitness of flies has been revealed not only in fertility. Flies with such mutation have appeared to be more sensitive to high temperatures have lived less under standard conditions of keeping and in the environment with ethanol. Impact of other marked mutations (b and cn) on before-mentioned characteristics either has not been revealed or has been less significant. It is worth mentioning that with the increasing age of investigated mutants interlinear differences of certain characteristics of fitness become more significant.

Thus for example, percentage of flies that survived under conditions of high temperatures has been almost the same (ranged between 60-65%) for investigated imago at the age of 1 day. As for flies at the age of 5 and 15 days statistically true interlinear differences have been revealed (Table 2). Ratio of vg and b, cn, vg flies that survived at high temperatures to those of type C-S has been respectively 74.7 and 85% for 5 day old flies. For the same types of flies at the age of 15 days this ratio is respectively almost 35.4 and 24.1%. Thus, young vg flies on the 1st day of life differ insignificantly from the flies of wild type C-S in the level of thermal resistance. With aging of genotypes this component of fitness decreases much faster at mutants than at C-S flies. Before mentioned

Table 2: Survival of linear flies at various ages under high temperature conditions (41°C and 15 min.), percentage of flies that survived n = 20

	Age (days)		
Lines	1	5	15
C-S	63.69±1.06*	50.08±1.32	15.56±1.40*
b	65.44±2.15*	52.17±1.29	18.20±1.90*
cn	63.30±1.42*	51.58±1.68	17.47±1.95*
vg	60.60±3.08*	37.42±2.04⁺	5.53±0.87*⁺
b, cn, vg	60.37±3.83*	42.95±1.80⁺	3.75±0.71*⁺

*Differences are true in compar is on with 2-5 days old flies. ⁺Differences are true in comparison with C-S flies

Table 3: Survival of linear flies of variousage underconditions of starvation (L_{150}) n = 20

	Age (days)		
Lines	1	5	15
C-S	50.21±3.18*	42.98±2.21	20.15±1.91*
b, cn, vg	45.41±2.21*	37.02±1.74⁺	11.45±0.97*⁺

*Differences are true in comparison with 5 days old flies. ⁺Differences are true in comparison with C-S flies

does not refer to single b and cn mutants whose survival has not been different from corresponding characteristic at C-S flies under conditions of high temperatures at all ages. Thus, studying into fertility and survival of flies at high temperatures allows to come to the conclusion that vg mutation in any genotype environment affects fitness more than other mutations and that intensity of this impact increases with aging. Analysis of survival for C-S and b, cn and vg flies at the age of 1, 5 and 15th days under conditions of starvation leads to the same conclusions (Table 3). There have not been any significant sexual differences as for survival of flies at high temperatures and their life expectancy in standard conditions of keeping. Only under conditions of starvation, we have observed better viability for female flies than for males. Under standard conditions of keeping life expectancy of individual vg, cn, vg and b, cn, vg flies has been significantly (almost in two times) less than that of C-S flies and of other mutants such as b, cn and b, cn. Presence of ethanol (10%) in the environment has led to increase in life expectancy for flies of all investigated lined except vg line. Generalizing data obtained while studying main phenological components of fitness, it should be pointed out that these characteristics depend not only on present in genotypes marked mutations but also on age, sex and conditions of keeping for investigated flies. Besides, vg mutation reveals the most significant impact on fitness components of flies both on its own and being the component of the genotype b, cn, vg and some other.

Gene-enzymatic system Adh at linear flies with various level of fitness: Taking into consideration, all known facts as for important role of locus Adh in viability and fitness of Drosophila, researchers have investigated state of gene enzymatic system Adh at well adapted line C-S and badly adapted line b, cn and vg. Allele control Adh is defined through estimation of three common criteria in particular, Adh activity, thermal resistance of this enzymeand its electrophoretic motility. The latter has been determined before. It has been found that C-S flies are homozygotes in Adh^S allele Adh and flies having *vg* gene contain Adh^f allele and also, F-alosyme of the enzyme. F-alosyme is considered to be more active and less resistant to high temperature. The research has shown that larvae, chrysalis and imago of well adapted flies C-S reveal significantly less activity of Adh than corresponding age groups of b, cn and vg flies. This fact proves the data as for presence of various alosymes Adh at these flies. In the course of maturation and aging (from larvae to 25 days old imago) Adh activity grows significantly at booth C-S and b, cn and vg flies (Table 4).

As for thermal resistance of the enzyme, this characteristic has not changed and has been almost the same at both C-S flies and at triple mutants which is probably connected with post translation Adh modification at mutants. Comparison of age dynamics for Adh activity at C-S and b, cn, vg flies with dynamics of studied earlier components of their fitness has shown that high Adh activity at aging and old flies is observed on the background of worsening phenological characteristics of fitness. Thus, high Adh activity as well as high thermal resistance of the enzyme is not obligatory condition of high fitness of individuals. Normal maturation of flies and necessary level of their fitness may be provided through certain optimal functioning of the gene-enzymatic system Adh that is in balance with all other enzymatic processes of cells. Changes in genotype which led to misbalance of the homeostatic enzyme cycle cause decrease in fitness and other undesirable consequences. Comparison of age dynamics for Adh activity at C-S and b, cn, vg flies with dynamics of studied earlier components of their fitness has shown that high Adh activity at aging and old flies is observed on the background of worsening phenological characteristics of fitness. Thus, high Adh activity as well as high thermal resistance of the enzyme is not obligatory condition of high fitness of individuals.

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Despite the absence of direct dependence between Adh activity and fitness components there is no doubt in importance of gene-enzymatic system Adh for processes of ontogenetic and phylogenetic fitness. The fact that certain alleles reveal selective dominance under certain conditions of existence proves presence of these alleles in adaptive and compensate complexes as well in co-adapted units of genes, common for considering mechanisms of phylogenetic fitness and heterosis.

Table 4: Activity of Adh in ontogenesis of linear flies, n/moll HADH/min•mg of protein n = 10-20

Stages of life cycle	Investigated lines: C-S	b, cn, vg
Larvae	50.49±4.20*	86.27±5.650*+
Chrysalis	48.66±3.18*	98.76±6.180*+
Imago (days)		
1	62.30±3.24*	104.53±8.160*+
3	88.51±4.24	135.16±9.820+
7	91.12±5.41	43.38±9.750+
10	96.45±7.21	203.01±10.46*+
15	99.05±6.15	251.30±10.80*+
25	102.33±9.25	242.10±10.95*+

*Differences are true in comparison with 3 days old imago of corresponding, +Differences are true in comparison with C-S flies

Crossing over frequency at b, cn and vg site in dependence on the age of flie: The idea that mainly recombination genesis provides perfection of adaptive ability of higher organisms in phylogenesis is well documented as been found that maximum combinatory activity is characteristic of young flies (1-3 days old). Following stages of life (4-6, 7-9, 10-12 days) are characterized by lower crossing over frequency and corresponds to data in genetic maps. However, old (23-26 days) flies show significant increase in crossing over frequency.

There are recombination genesis itself favors creating new complexes and units of co-adapted genes, i.e., new genotypes. Thus, there are reasons to believe that level of combinatory activity of chromosomes may serve not only as characteristic of genotype variability but also as characteristic of level of their fitness. Following these ideas, they have studied crossing over frequency at the site b, cn and vg at b, cn and vg mutants of different ages (Table 5).

Table 5: Crossing over frequency at the investigated site of chromosome 2 at b, cn and vg mutants of different age, n% = 15-20 families

Site of chromosome 2	Age of females (days): 1-3	4-6	7-9	10-12	23-26
b, cn	15.23±2.48*	7.42±1.210*+	7.91±1.110*+	8.63±1.04	12.20±1.50**
cn, vg	18.12±0.81*	12.55±1.38	10.52±1.25	13.88±1.26	16.71±1.08*
b, vg	33.44±3.84*	19.97±2.04	18.43±1.57	22.51±1.91	30.6±1.710*

*Differences are true incomparis on with corresponding characteristic at 7-9 days old flies. +Crossing over frequency at the site b and cn differs truly from that at site cn and vg

Crossing over frequency is measured in ratio of a number of single and double recombinants to all descendents of test crossing. Crossing over frequency at the site b, cn and cn, vg is calculated separately. It has reasons to think that high combinatory activity of young (1-3 days old) flies reveals high potential of combinatory variability of their descendents as well as better survival in changeable conditions of environment. On the contrary high crossing over frequency at old flies can reflect certain non-stability of genome and lower level of fitness of old individuals. Analysis of crossing over frequency at each investigated site of chromosome 2 has shown non-equal distribution of crossings over at the sites b, cn and cn, vg in particular at old flies (Table 5).

Lower crossing over frequency at the site b, cn can be explained by round center localization of this site as dependence of combinatory processes on structural peculiarities of different sites of chromosomes is well known. Nevertheless, degree of increasing combinatory activity at old (23-26 days old) flies at sites b, cn and cn, vg in comparison with those of chromosome 2 at 7-9 days old flies is almost the same (in about 1.5 times).

Combinatory activity, gene-enzymatic system of Adh and fitness of b, cn, vg mutant at genotype structural changes: Probable changes of gene balance while joining genes that are contrastive in fitness of genotypes are estimated upon the level of combinatory activity, functional state of locus Adh and on phenological characteristics of fitness at synthesized triple mutants b, cn, vg (C-S) and b, cn, vg (1 C-S). It has become clear that crossing over frequency at flies with genotypes modified truly increases in comparison with target b, cn and vg flies (Table 6).

Thus, crossing over frequency at 4-6 days old, b, cn and vg (C-S) flies has become 38% higher and at those of b, cn and vg of the same age, it has become 20% higher in comparison with b, cn and vg flies. Degree of increase in combinatory activity at flies with the genotype saturated with C-S genes has been different at flies of different age (Fig. 1). At young flies (1-3 days of life of imago) crossing over frequency has been high but practically similar to that of target b, cn, vg mutant and b, cn, vg (C-S) flies. Flies of middle age (4-6, 7-9, 10-12 days of life) have revealed lower level of combinatory activity, though at flies b, cn, vg (C-S), this level has been significantly higher than that of mutant b, cn, vg. At old flies (23-26 days of life) crossing over frequency has increased again and has been truly higher than at 7-9 days old flies. Degree of increasing combinatory activity at old b and cn and vg (C-S), flies has been truly higher than that of b, cn and vg flies of the same age. Character of split in descendents in trihybrid test crosses proves that

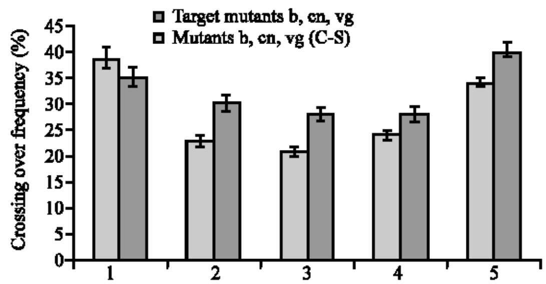

Fig. 1: Crossing over frequency (%) at the site b, cn and vg at Drosophila mutants b, cn, vg and b, cn, vg (C-S) of different age: 1, 1-3 days; 2, 4-6 days; 3, 7-9 days; 4, 10-12 days; 5, 23-26 days of life for flies, average among 15-20 experiments

Table 6: Crossing over frequency (%) at 5th day old mutant b, cn and vg at geno type changes *in vitro* n = 15-20 families

Site of chromosome 2	Investigated mutants		
	b, cn, vg	b, cn, vg (C-S)	b, cn, vg (1C-S)
b, cn	7.42±1.21	13.64±1.74*	11.53±1.00*
cn, vg	12.55±1.38	13.93±1.20	12.46±1.17
b, vg	19.97±1.44	27.57±1.85*	23.99±1.30*

*Differences are true incomparis on with line b, cn and vg

increase in frequency of genetic recombination at the site b, cn and vg at flies with modified genotypes is associated at first with increase in frequency of overlapping between genes *b* and *cn*. At 4-6 days old, b, cn, vg (C-S) females crossing over frequency at the site b and cn has increased in 1.81 times and at the females b, cn and vg (1C-S) of the same age has increased in 1.55 times in comparison with target b cn vg mutants. At the site cn and vg intensity of chromatide exchanges at mutants with modified genotypes and at target mutants has been practically the same.

Increase in crossing over intensity at old flies and at flies with modified genotypes can be considered as characteristic of certain non-stability of these genotypes because of changed genetic balance that is probable for introducing foreign genetic material into genotype. It is possible that under these conditions breaks in chromatides become more common and appear to happen more easily, especially on the places of localization of heterochromatin. It is interesting to compare frequency of recombination at investigated flies with activity of their Adh and state of fitness components.

Although, C-S and b, cn and vg flies differ in allele composition of locus Adh, it has been revealed that replacement of X-chromosome and saturation of b, cn and vg genotype with genes C-S does not practically lead to changes in activity and thermal resistance of Adh at mutants. Significant differences in activity and thermal resistance of Adh are absents at possible changes in allele control of this enzyme under conditions of saturating crosses. This fact can be explained by post translation modifications of alozymes. Activity and thermal resistance of Adh at b, cn and vg adults and at flies with modified genotypes differs insignificantly according to the results of investigation. As a result, it can be admitted that functioning state of gene-enzymatic system Adh under conditions of this research does not really reflect the level of genotype fitness. Reliable information about fitness of investigated flies has been obtained while studying other characteristics of fitness-fertility, life expectancy of individuals at various conditions of keeping and their survival at high temperatures, etc.

These data coincide with those about the dependence of Drosophila life expectancy on the allele position of gene *Adh* that are found in different sources of information. Saturation of b, cn and vg genotype with genes of line C-S is accompanied by increase in life expectancy. It increases up to the level of this characteristic at flies of wild type. Alongside with this both b, cn, vg (C-S) flies and C-S flies are characteristic of S-alozyme Adh. It is believed that replacement of Adh^F allele of b, cn and vg mutant onto allele Adh^S at saturating crosseswhich is characteristic of flies C-S is very important for normalization of life expectancy at mutants b, cn and vg. On the contrary replacement of chromosome 1 of a mutant onto the chromosome 1 of a wild type fly does not make the life period of mutant flies longer as replacement of chromosome 1could not have led to the change in allele control of Adh.

The data of Table 7 prove positive influence of genetic elements of wide genotype on survival of triple mutants under conditions of starvation. This characteristic at b, cn, vg (C-S) and b, cn, vg (1 C-S) flies has not differed from that of the flies of wild type and has been truly higher than at target b, cn and vg mutant. As for thermal resistance of flies this characteristic has appeared to be better only at replacement of chromosome 1; thermal resistance of b, cn, vg (C-S) has remained the same as at mutants b, cn, vg. Fertility of individuals that is the most important characteristic of fitness has turned out to be the least changeable under research conditions. Mutants with replaced chromosome as well as mutants with the genotype saturated with genes of wild type have had poor fertility as well as linear mutants b, cn and vg. If they suggest that the reason of poor fertility for triple mutants should be pleiotrophic influence of marked mutations, it becomes clear why significant impact on this characteristic of introduction into the mutant genotype of wild type is absent.

On the whole the data of Table 7 show that structural genotype recombination of b, cn and vg mutant through introduction of foreign genetic information does not worsen the fitness characteristics. It is possible that in spite of the poor co-fitness of genes for lines C-S and of genes for mutant b, cn and vg, their joining in one genotype can reveal positive influence on certain fitness components of mutant individuals due to high adaptive potential of wide type genes.

The results of the study show that structural and functional genotype changes that take place in ontogenesis and phylogenesis such as aging, mutagenesis, hybridizationand introduction into the genotype of side genetic material can significantly influence on genetic balance and fitness of organisms (Voro'eva, 2010). New information about pleiotropic interaction of mutant gene *b*, *cn* and *vg* at establishing fitness components at flies of various age that carry these mutations at different groupings.

Interaction of *b*, *cn* and *vg* genes and *Adh* locus genes has been first investigated for defining level of fitness for these genotypes on the example of double and triple mutants of Drosophila . It has been established that joined location of mutations in the genotype leads for both strengthening and weakening of their summary impact on certain fitness components. Summary effect of marked mutations depends on the age of flies, structural peculiarities of genotypeand other reasons (Olsson *et al*., 2010). Experiments on triple b, cn and vg mutants have shown that fly aging and interference withtheir genotyp saturating crosses, chromosome 1 replacement can increase Adh activity and recombination frequency at the site b, cn and vg of chromosome 2. Besides, strict dependence between certain fitness components, crossing over frequency and function of gene enzymatic system Adh has not always been revealed. Influence of mutations agingand genotype modifications *in vitro* on certain fitness components under conditions of experiment has been different in power and direction of

Table 7: Fitness components of target lines of Drosophila and flies with modified genotypes n = 20-80

Fitness components	Investigated genotypes: C-S	b, cn, vg	b, cn, vg (C-S)	b, cn, vg (1C-S)
Fertility, number of desc endents by one pair	49.01±3.21*	23.87±1.150	20.94±1.24	24.20±1.71
Life expectancy (L_{t50}) days	11.99±0.85*	6.31±0.570	11.89±0.82*	7.84±0.660
Life expectancy at the conditions of starvation (L_{t50}) (h)	42.98±2.21*	37.02±1.740	44.20±1.44*	43.31±1.67*
Thermal resistance at 5th day old flies (%)	50.08±1.32*	42.95±1.800	41.97±2.25	50.29±1.37*

*Differences are true incompar is on with line b, cn and vg changes in investigated characteristics (Showell *et al.*, 2011; Mortensen *et al.*, 2011). It should be mentioned that aging, structural genotype modifications (presence of marked mutations, saturating crosses, chromosome replacement) often lead to typical changes in certain fitness components, Adh activity and process of recombination.

In these cases, phenological characteristics of fitness, as a rule, get worse and Adh activity and crossing over frequency increase. These results coincide with existing theories of aging which associate this process with accumulation of mutations in a cell, intensive acidic macromolecule oxidation, lowering processes of repairing and marginotomy of chromosomes, etc. It cannot be excluded that there are similar mechanisms on the basis of decrease in genotype fitness which appears as a result of aging, mutationand structural modification *in vitro*. However, complete parallelism in changes of these characteristics in changes of characteristics, investigated at old flies and flies with modified genes is not observed. Some fitness components (fertility, life expectancy under conditions of starvation and survival at high temperature) have not changed or even improved up to the level of C-S line characteristics.

All this is true for b, cn and vg mutants after saturation their genotype with genes of C-S flies or replacement of chromosome 1 into chromosome of wild type (Maclellan *et al.*, 2011; Loeschcke *et al.*, 2011). But aging has always been accompanied with these components getting worse. Thus, genetic material of C-S flies introduced into b, cn and vg mutant genotype have revealed positive influence on certain characteristics of mutant fitness while aging in particular of those that carry marked mutations is accompanied with worsening all fitness components (Ueyama *et al.*, 2010).

In the summary, researchers should illustrate that we studied exchangeable influence of genes *b*, *cn* and *vg* in determining the level of adapting of the flies genotype. It is known in whole that the influence fore these mutants depends on the flies age, temperature and the diet conditionand the especial structure of genotype.

The experiment on triple mutants b, cn and vg proved that the aging of flies and artificial genotype changes could influence similarly and this by increasing crossing over frequency in the site b, cn and vg. The positive influence of the structural changes in the triple mutants genotype which is shown is improving the adapting indicators independently that is because for inserting the genes of wild type which have more adaptive ability than the mutants.

CONCLUSION

The changes which were found from new recombination while artificial reorganization of a genotype could be explained because of instability which appeared when we inserted foreign genetic material. Through these conditions, the general genetic balance may destroy, it is not strange that the destroyed genetic balance may happen in the old flies too.

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