



Changes in chromosomal polymorphism and global warming: The case of *Drosophila subobscura* from Apatin (Serbia)

Goran Zivanovic¹ and Francesc Mestres²

¹Department of Genetics, Institute for Biological Research "Sinisa Stankovic", University of Belgrade, Belgrade, Serbia.

²Departament de Genètica, Universitat de Barcelona, Barcelona, Spain.

Abstract

In this study, chromosomal inversion polymorphism data for a natural population of *Drosophila subobscura* from a swampy region near the town of Apatin (Serbia) were compared with data for the same population collected approximately 15 years earlier. The pattern of chromosomal inversion polymorphism changed over time. There were significant increases in the frequency of characteristic southern latitude ("warm" adapted) chromosomal arrangements and significant decreases in the frequency of characteristic northern latitude ("cold" adapted) chromosomal arrangements in the O and U chromosomes. The chromosomal arrangements O_{3+4} and O_{3+4+22} (derived from the O_{3+4} arrangement) showed significant increases in 2008 and 2009 with regard to the 1994 sample. There was also a significant increase (~50%) in the U_{1+2} arrangement, while U_{1+8+2} (a typical southern arrangement) was detected for the first time. Since the Apatin swampy population of *D. subobscura* has existed for a long time in a stable habitat with high humidity that has not been changed by man our results indicate that natural selection has produced chromosomal changes in response to the increase in temperature that has occurred in the Balkan Peninsula of central southeastern European.

Key words: chromosomal inversions, *Drosophila subobscura*, global warming, karyotypes.

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Introduction

The climate change of recent decades has led to heritable genetic changes in animal species as diverse as birds, squirrels and mosquitoes (Bradshaw and Holzapfel, 2006). Long-term changes in chromosomal inversion polymorphisms in *Drosophila* species suggest that the genetic constitution of populations is responding to climate change (Van Heerwaarden and Hoffmann, 2007). This suggestion is strongly supported by the fact that genes located within inversions are associated with a variety of traits including those involved in climate adaptation (Hoffmann and Rieseberg, 2008). This characteristic was observed in some species of *Drosophila*, such as *D. melanogaster* in Australia (Anderson *et al.*, 2005; Umina *et al.*, 2005), *D. robusta* in North America (Levitan and Etges, 2005) and *D. subobscura* in Europe (Rodríguez-Trelles and Rodríguez, 1998; Solé *et al.*, 2002; Balanyà *et al.*, 2004, 2006, 2009) and America (Balanyà *et al.*, 2006, 2009).

In all *D. subobscura* populations from southwestern and central Europe studied to date an increase in the fre-

quency of "warm"-adapted chromosomal arrangements typical of southern latitudes, and a decrease in the frequency of "cold"-adapted chromosomal inversions typical of northern latitudes, has been observed (Rodríguez-Trelles and Rodríguez, 1998; Solé *et al.*, 2002; Balanyà *et al.*, 2004, 2006, 2009). This species is an excellent model organism for such studies because of its extensive polymorphism for chromosomal inversions: 67 inversions distributed in 93 chromosomal arrangements have been identified, with the frequencies of most of these varying clinally with latitude (Krimbas and Loukas, 1980; Prevosti *et al.*, 1988; Menozzi and Krimbas, 1992; Krimbas, 1993).

Of the five acrocentric chromosomes in *D. subobscura*, the O chromosome (homologous to the 3R chromosomal arm of *D. melanogaster* and second chromosome of *D. pseudoobscura*) is the longest and most polymorphic, with about 40 natural chromosomal arrangements (Krimbas, 1993). Since the 1960s more than 150 natural populations have been studied throughout the Palearctic distribution of this species. However, few long-term analyses of changes in inversion polymorphism have been done in southeastern Europe or the Balkan Peninsula. For this reason, the main aim of this work was to study a population of *D. subobscura* from Apatin (Serbia). This population is

Send correspondence to Goran Zivanovic. Department of Genetics, Institute for Biological Research "Sinisa Stankovic", University of Belgrade, Bulevar Despota Stefana 142, 11000 Belgrade, Serbia. E-mail: goranziv@ibiss.bg.ac.rs.

located in a swampy region on the left bank of the Danube river in the lowlands of the Pannonian plain. This area, which is midway along the Danube river about 1366–1433 km from the mouth of this river in the Black sea, is deliberately left to be flooded permanently or during spring across an area of at least 3500 ha. Consequently, the Apatin *D. subobscura* population has existed in a very humid habitat that has never been changed by man. This is probably the main reason for the large differences observed between the Apatin and other *D. subobscura* populations from the Balkan Peninsula (Zivanovic *et al.*, 2002; Zivanovic and Marinkovic, 2003; Zivanovic, 2007; Zivanovic and Mestres, 2010a,b).

Specifically, we analyzed inversion polymorphisms in *D. subobscura* from Apatin 14 and 15 years after the initial studies, and used this information to assess variation in composition. Our intention was to compare samples from June 2008 and 2009 with the sample from June 1994 at exactly the same site, paying special attention to calendar and climatology data. The results may be especially interesting because during this period there were several heat waves, for example the first evidence of climate change in the region was recorded in summer 2003 (Schar and Jendritzky, 2004). The study should tell us whether long-term chromosomal inversion polymorphism changes, if they exist, are characteristic of all or only of specific chromosomes and their arrangements. Finally, the analysis of possible changes in frequency of some chromosomes in the O_{3+4} group (O_{3+4+1} , O_{3+4+22} and so on) correlated to climate changes is of particular interest, because some genes involved in thermal adaptation are located in the O_{3+4} region (Quintana and Prevosti, 1991; Moltó *et al.*, 1992; Laayouni *et al.*, 2007).

Material and Methods

Drosophila subobscura flies were collected from a poplar (*Populus alba*) wood in a swampy region located on the left bank of the river Danube, near the town of Apatin (45°40' N, 19°00' E, approximately 200 km NW of Belgrade). This region is included in the Ramsar List, which lists wetlands of international importance, and is known as the Upper Danube wetland area. Meteorological data for the site were obtained from the Serbian Republic Hydrometeorological Service. Samples were collected twice from exactly the same place in June 2008 and June 2009. To allow comparison with the June 1994 sample, the 2008 and 2009 samples were collected 2.5 days earlier per decade because spring/summer has advanced an average of 2.5 days per decade in Europe (Menzel *et al.*, 2006). Unfortunately, the need to trap on precise days and Danube river flooding limited the sample size obtained. Only wild males (June 2008, June 2009) and the sons of wild females (June 2009) were used to assess chromosomal inversion polymorphism. The males were crossed individually with virgin females of the Kussnacht strain that were homokaryotypic for standard chromosomal arrangements in all five chromosomes.

Polytene chromosomes were stained and squashed in aceto-orcein solution. At least eight larvae from the progeny of each cross were examined. The chromosomal map of Kunze-Mühl and Müller (1958) was used for cytological analysis of the chromosomal arrangements and the nomenclature for these arrangements followed that of Kunze-Mühl and Sperlich (1955). Departure of the observed frequencies of chromosomal arrangements and chromosomal karyotypes from expectations was tested using χ^2 test contingency tables. The degree of chromosomal inversion polymorphism in the population analyzed was assessed using the index of free recombination (IFR) (Carson, 1955). Chromosomal inversion polymorphism patterns were obtained by computing Nei's genetic distances (Nei, 1972) between Apatin 2008+2009 and Apatin 1994 and another five previously analyzed Balkan populations (Kamariste, June 1996; Djerdap, June 2001; Petnica, June 1995; Jastrebac, June 1993; Zanjic, June 1997) (Zivanovic *et al.*, 2002; Zivanovic and Marinkovic, 2003; Zivanovic, 2007).

Results

The frequencies of chromosomal arrangements in the Apatin population from June 2008 and June 2009, together with those previously obtained for this population in June 1994 (Zivanovic *et al.*, 2002), are shown in Table 1. A total of 18 chromosomal arrangements were found in the old and new samples. Some of the chromosomal arrangements found earlier (U_1 , E_{1+2} , O_6 , O_{3+4+2} , in 1994) were not observed in 2008 and 2009, whereas some chromosomal arrangements, such as U_{1+8+2} , $E_{1+2+9+12}$, O_{22} and O_{3+4+22} that were not found 15 years earlier were observed in the 2009 samples (Table 1). There were significant differences in the frequencies of the U chromosomal arrangements between the June 1994 and June 2009 samples ($\chi^2 = 21.2$, *d.f.* = 4, $p < 0.001$). Similarly, the 1994 and 2008+2009 samples also differed significantly in the frequencies of five U chromosomal arrangements ($\chi^2 = 17.4$, *d.f.* = 4, $p < 0.01$). There was a significant decrease in the frequency of the U_{st} arrangement in the two comparisons ($\chi^2 = 9.5$, *d.f.* = 1, $p < 0.01$ and $\chi^2 = 6.9$, *d.f.* = 1, $p < 0.01$, for the first and second comparison, respectively). Over the same period of time, U_{1+2} showed a significant increase in frequency ($\chi^2 = 8.0$, *d.f.* = 1, $p < 0.01$, for the first case and $\chi^2 = 8.1$, *d.f.* = 1, $p < 0.01$, for second). The U_{1+8+2} arrangement was detected for the first time in the 2008 and 2009 samples.

There were also significant differences among seven O-chromosomal arrangement frequencies between the 1994 and 2009 samples (first case) ($\chi^2 = 25.1$, *d.f.* = 6, $p < 0.001$). This significant difference was also observed in the second case (comparing the 1994 and 2008+2009 samples: $\chi^2 = 23.8$, *d.f.* = 6, $p < 0.001$). The O_{st} arrangement showed a significant decrease in frequency ($\chi^2 = 4.2$, *d.f.* = 1, $p < 0.05$ and $\chi^2 = 5.1$, *d.f.* = 1, $p < 0.05$, for the first

Table 1 - Frequencies of chromosomal arrangements for the Apatin natural population of *Drosophila subobscura*.

Chromosomal arrangement	June 1994		June 2008		June 2009		June 2008+2009	
	n	p (%)	n	p (%)	n	p (%)	n	p (%)
A _{st}	25	50.0	3	60.0	7	46.7	10	50.0
A ₁	21	42.0	2	40.0	7	46.7	9	45.0
A ₂	4	8.0	/	/	1	6.6	1	5.0
Total	50		5		15		20	
J _{st}	39	39.0	4	40.0	11	36.7	15	37.5
J ₁	61	61.0	6	60.0	19	63.3	25	62.5
Total	100		10		30		40	
U _{st}	52	52.0	5	50.0	6	20.0	11	27.5
U ₁	1	1.0	/	/	/	/	/	/
U ₁₊₂	23	23.0	4	40.0	15	50.0	19	47.5
U ₁₊₂₊₆	24	24.0	1	10.0	6	20.0	7	17.5
U ₁₊₈₊₂	/	/	/	/	3	10.0	3	7.5
Total	100		10		30		40	
E _{st}	70	70.0	7	70.0	18	60.0	25	62.5
E ₁₊₂	2	2.0	/	/	/	/	/	/
E ₁₊₂₊₉	11	11.0	/	/	6	20.0	6	15.0
E ₁₊₂₊₉₊₁₂	/	/	/	/	2	6.6	2	5.0
E ₈	17	17.0	3	30.0	4	13.4	7	17.5
Total	100		10		30		40	
O _{st}	61	61.0	4	40.0	12	40.0	16	40.0
O ₆	6	6.0	/	/	/	/	/	/
O ₂₂	/	/	/	/	1	3.3	1	2.5
O ₃₊₄	23	23.0	6	60.0	10	33.3	16	40.0
O ₃₊₄₊₁	8	8.0	/	/	2	6.6	2	5.0
O ₃₊₄₊₂	2	2.0	/	/	/	/	/	/
O ₃₊₄₊₂₂	/	/	/	/	5	16.7	5	12.5
Total	100		10		30		40	

and second case, respectively), while the O₃₊₄₊₂₂ arrangement was observed for the first time in the 2009 sample. The O₃₊₄ arrangement increased in frequency, but there were no significant differences ($\chi^2 = 1.3$, $d.f. = 1$, $p > 0.05$ and $\chi^2 = 3.3$, $d.f. = 1$, $p > 0.05$, for the first and second case, respectively). However, O₃₊₄ and the other O_{3+4+x} chromosomal arrangements derived from O₃₊₄ (O₃₊₄₊₁, O₃₊₄₊₂ and O₃₊₄₊₂₂) showed significant increases in frequency ($\chi^2 = 5.5$, $d.f. = 1$, $p < 0.05$ and $\chi^2 = 7.1$, $d.f. = 1$, $p < 0.01$ for the two cases, respectively). Finally, there was no significant difference in frequency for any chromosomal arrangement between the 2008 and 2009 samples.

The frequencies of the chromosomal karyotypes obtained for the Apatin population are shown in Table 2. The 1994 and 2008+2009 samples had 28 and 23 chromosomal karyotypes, respectively. Sixteen chromosomal karyotypes were found in both the old and new samples. Twelve chromosomal karyotypes previously observed were not found

in the new samples, while seven chromosomal karyotypes present in the new samples were not detected in 1994. There were significant differences in the frequency of the U karyotypes ($\chi^2 = 19.6$, $d.f. = 8$, $p < 0.05$) between the 1994 and 2008+2009 samples. The U_{st}/U_{st} karyotype showed a dramatic decrease (not seen in the 2008 and 2009 samples), whereas U_{st}/U₁₊₈₊₂ appeared for the first time in the 2009 sample. There were also significant differences in the frequencies of all 14 O-chromosome karyotypes for the two samples analyzed (1994 versus 2008+2009) ($\chi^2 = 29.2$, $d.f. = 13$, $p < 0.01$). O_{st}/O_{st} showed a significant decrease in frequency ($\chi^2 = 7.3$, $d.f. = 1$, $p < 0.01$), while O_{st}/O₃₊₄ showed a significant increase ($\chi^2 = 7.8$, $d.f. = 1$, $p < 0.01$) and O₃₊₄/O₃₊₄₊₂₂ was detected for the first time in the 2009 sample. A similar result was found for all nine E chromosome karyotypes ($\chi^2 = 16.8$, $d.f. = 8$, $p < 0.05$). E_{st}/E_{st} showed a significant decrease in frequency ($\chi^2 = 4.5$,

$d.f. = 1, p < .05$), while for E_{st}/E_8 there was a significant increase ($\chi^2 = 5.1, d.f. = 1, p < 0.05$) and $E_{st}/E_{1+2+9+12}$ was observed for the first time in the 2009 sample.

The variations in chromosomal polymorphisms were compared with the meteorological data shown in Table 3.

The combined average for the maximum, minimum and mean temperatures (26.4 °C, 14.4 °C and 20.6 °C, respectively) for 2008 and 2009 were higher than in 1994. A year-by-year analysis from 1994 onwards revealed several heat waves such as the well documented heatwave of 2003

Table 2 - Frequencies of chromosomal karyotypes for the Apatin natural population of *Drosophila subobscura*.

Chromosomal karyotype	June 1994		June 2008		June 2009		June 2008+2009	
	n	p (%)	n	p (%)	n	p (%)	n	p (%)
J_{st}/J_{st}	9	18.0	1	20.0	1	6.7	2	10.0
J_{st}/J_1	21	42.0	2	40.0	9	60.0	11	55.0
J_1/J_1	20	40.0	2	40.0	5	33.3	7	35.0
Total	50		5		15		20	
U_{st}/U_{st}	17	34.0	/	/	/	/	/	/
U_{st}/U_{1+2}	10	20.0	4	80.0	3	20.0	7	35.0
U_{st}/U_{1+2+6}	8	16.0	1	20.0	1	6.7	2	10.0
U_{st}/U_{1+8+2}	/	/	/	/	2	13.3	2	10.0
U_1/U_{1+2}	1	2.0	/	/	/	/	/	/
U_{1+2}/U_{1+2}	3	6.0	/	/	4	26.7	4	20.0
U_{1+2}/U_{1+2+6}	6	12.0	/	/	3	20.0	3	15.0
U_{1+2}/U_{1+8+2}	/	/	/	/	1	6.7	1	5.0
U_{1+2+6}/U_{1+2+6}	5	10.0	/	/	1	6.7	1	5.0
Total	50		5		15		20	
E_{st}/E_{st}	29	58.0	3	60.0	3	20.0	6	30.0
E_{st}/E_{1+2}	1	2.0	/	/	/	/	/	/
E_{st}/E_{1+2+9}	8	16.0	/	/	6	40.0	6	30.0
$E_{st}/E_{1+2+9+12}$	/	/	/	/	2	13.3	2	10.0
E_{st}/E_8	3	6.0	1	20.0	4	26.7	5	25.0
E_{1+2+9}/E_{1+2+9}	2	4.0	/	/	/	/	/	/
E_8/E_8	1	2.0	1	20.0	/	/	1	5.0
E_8/E_{1+2}	1	2.0	/	/	/	/	/	/
E_8/E_{1+2+9}	5	10.0	/	/	/	/	/	/
Total	50		5		15		20	
O_{st}/O_{st}	22	44.0	/	/	2	13.3	2	10.0
O_{st}/O_6	4	8.0	/	/	/	/	/	/
O_{st}/O_{22}	/	/	/	/	1	6.7	1	5.0
O_{st}/O_{3+4}	7	14.0	4	80.0	5	33.3	9	45.0
O_{st}/O_{3+4+1}	4	8.0	/	/	1	6.7	1	5.0
O_{st}/O_{3+4+2}	2	4.0	/	/	/	/	/	/
O_{st}/O_{3+4+22}	/	/	/	/	1	6.7	1	5.0
O_6/O_{3+4}	1	2.0	/	/	/	/	/	/
O_6/O_{3+4+1}	1	2.0	/	/	/	/	/	/
O_{3+4}/O_{3+4}	7	14.0	1	20.0	1	6.7	2	10.0
O_{3+4}/O_{3+4+1}	1	2.0	/	/	/	/	/	/
O_{3+4}/O_{3+4+22}	/	/	/	/	3	20.0	3	15.0
O_{3+4+1}/O_{3+4+1}	1	2.0	/	/	/	/	/	/
O_{3+4+1}/O_{3+4+22}	/	/	/	/	1	6.7	1	5.0
Total	50		5		15		20	

Table 3 - Meteorological data for the Apatin region for the month of June from 1994 to 2009.

Year	Temperature (°C)			Rainfall (mm)
	Maximum	Minimum	Mean	
1994	26.3	13.8	20.3	55.8
1995	25.2	13.5	19.1	97.8
1996	27.5	13.6	21.0	22.9
1997	26.7	13.6	20.4	81.0
1998	27.9	15.1	21.7	69.4
1999	26.0	13.9	20.1	124.1
2000	29.4	13.4	22.6	9.8
2001	23.7	12.3	18.1	231.0
2002	28.5	14.6	21.9	37.7
2003	31.6	16.5	24.6	23.0
2004	25.7	13.6	19.5	71.6
2005	25.3	13.3	19.8	77.0
2006	25.7	14.3	20.0	100.6
2007	28.8	15.4	22.3	49.5
2008	27.3	15.7	21.8	91.2
2009	25.5	13.0	19.4	107.6

Max. T, Min. T and Mean T stand for maximum, minimum and mean temperatures, respectively.

(Schar and Jendritzky, 2004) and less drastic ones in 2000, 2002 and 2007. These data suggest a gradual increase in temperature at the locality studied. However, desiccation does not seem to affect the Apatin habitat: although the rainfall pattern is irregular, a general decrease has not been observed. Indeed, data from 2006 onwards suggest that there has been a tendency for rainfall to increase, in agreement with global warming predictions (Houghton, 2005; Wentz *et al.*, 2007).

The IFR values obtained for the Apatin population were: 75.60 ± 1.88 (June 2008) and 77.13 ± 2.11 (2009), with a mean of 76.75 ± 1.63 . These values were only slightly lower than that for 1994 (84.62 ± 1.55). Table 4 shows Nei's genetic distances between the Apatin and other Balkan populations. This table also provides older genetic distances between Apatin (1994) and these Balkan populations (Zivanovic *et al.*, 2002; Zivanovic, 2007). The

new genetic distances were lower than those obtained previously.

Discussion

The world's climate is changing rapidly, with a global temperature increase of 0.6 °C in the past three decades, and 0.8 °C in the past century (Hansen *et al.*, 2006; Van Heerwaarden and Hoffmann, 2007). Many animals and plants adapt rapidly to variations in environmental conditions (Endler, 1986). In particular, widespread generalist species such as *D. melanogaster* and *D. subobscura*, which have short generation times and rapidly form phenotypic and genotypic clines, are excellent sensitive indicators of such global warming (Van Heerwaarden and Hoffmann, 2007; Balanyà *et al.*, 2009). The results of long-term studies indicate that there have been changes in the frequency of chromosomal arrangements in *D. subobscura*. In recent decades, a significant decrease in the frequency of standard chromosomes ("cold" adapted and abundant in northern latitudes) and an increase in some non-standard chromosomal arrangements ("warm" adapted and characteristic of southern latitudes) have been described in natural *D. subobscura* populations from southwestern and central Europe (de Frutos and Prevosti, 1984; Gosteli, 1990; Orengo and Prevosti, 1996; Rodríguez-Trelles *et al.*, 1996; Solé *et al.*, 2002; Balanyà *et al.*, 2004, 2006, 2009) and in American colonizing populations (Balanyà *et al.*, 2006, 2009). These changes have been attributed to the adaptation of chromosomal inversion polymorphism to a warmer climate in Europe and America.

The effect of global warming in the Apatin population is reflected as variation in chromosomal polymorphism. Specifically, there was a significant increase in the frequency of some chromosomal arrangements characteristic of southern latitudes ("warm" adapted) and a significant decrease in the frequency of some standard inversions ("cold" adapted) in the O and U chromosomes. Thus, O_{3+4} showed a significant increase when considered together with O_{3+4+x} arrangements (~40% on average) in 2008 and 2009 compared to 1994. Similar results have been reported for other southwestern European populations (Orengo and Prevosti, 1996; Rodríguez-Trelles and Rodríguez, 1998). Interestingly, associations between heat-tolerance and the O_{3+4} arrangement have been described previously for *D.*

Table 4 - Nei's genetic distances calculated between the *D. subobscura* population from Apatin (2008+2009), the same population in 1994, and another five Balkan populations.

	Apatin	Kamariste	Djerdap	Petnica	Jastrebac	Zanjic
	45°40' N	45°29' N	44°37' N	44°16' N	43°26' N	42°24' N
	June 1994	June 1996	June 2001	June 1995	June 1993	June 1997
Apatin June 08+09	0.058	0.145	0.053	0.119	0.092	0.094
Apatin June 1994	0.000	0.223	0.151	0.198	0.198	0.210

subobscura (Quintana and Prevosti, 1991). Furthermore, several genes that code for heat stress proteins (Moltó *et al.*, 1992) and others involved in thermal adaptation (Laayouni *et al.*, 2007) are located in this arrangement. In addition, the O_{3+4} and O_{3+4+x} frequencies in the old Apatin population (June 1994) were 53% and 59% lower, respectively, than in all other southern Balkan populations (Jastrebac, June 1993; Petnica, June 1995; Kamariste, June 1996; Zanjic, June 1997; Djerdap, June 2001; Avala, June 2004) (Zivanovic *et al.*, 2002; Zivanovic and Marinkovic, 2003; Zivanovic, 2007; Zivanovic and Mestres, 2010a,b). In contrast, the frequencies of the O_{3+4} and O_{3+4+x} arrangements in the new Apatin population (2008 + 2009 sample) were no much lower (by 19% and 28%, respectively) than those previously reported for the southern Balkan populations. Data for the O-chromosome suggest that the Apatin population is evolving towards a more “southern” population. Similarly, for the U chromosome, a significant increase in the U_{1+2} frequency (~50%) during the period studied, as well as the presence of the U_{1+8+2} arrangement which was not previously observed, indicate evolution towards a “southern” population.

Our results agree with those from southwestern and central European populations (Rodríguez-Trelles and Rodríguez, 1998; Solé *et al.*, 2002; Balanyà *et al.*, 2004, 2006, 2009). The relatively low IFR values obtained in this study (not previously observed in 1994) are characteristic of ecologically central European populations of *D. subobscura* (Krimbas, 1993). This result, and the smaller genetic distances between the new Apatin population (2008+2009) and all other Balkan populations (Table 4) compared to the distances previously found, provide further evidence that the Apatin population is becoming more “southern”. A similar decrease in genetic distances over time has been observed in Mediterranean, Atlantic and Central European populations of *D. subobscura* (Balanyà *et al.*, 2004).

The absence of frequency changes for chromosomal polymorphism in the remaining chromosomes (A, J and E) suggests that our results were most probably not a consequence of gene flow from the south, where the frequencies of all the arrangements in these three chromosomes are completely different in all southern *D. subobscura* populations analyzed (Zivanovic *et al.*, 2002; Zivanovic and Marinkovic, 2003; Zivanovic, 2007; Zivanovic and Mestres, 2010a,b). Together, these observations suggest that we have detected the effect of selection on chromosomal polymorphism composition (see also the discussion of this topic in Hoffman and Rieseberg, 2008). Some form of natural selection appears to be acting as a reaction to the increase in temperature associated with many heat waves that have markedly affected the Balkan Peninsula; random processes and genetic drift do not appear to be involved. The latter conclusion is supported by the high migration rate among *D. subobscura* populations from the Balkan Peninsula (Zi-

vanovic *et al.*, 2007) since migration among populations efficiently purges most effects of genetic drift.

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