Research Article

Asymmetry of frontal bristles and postocular setae in species and hybrids of the *Anastrepha fraterculus* complex (Diptera, Tephritidae)

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Abstract

Asymmetry of the frontal bristles and postocular setae was studied in samples from natural populations and laboratory colonies of *Anastrepha* sp. 1 *aff. fraterculus*, of *A.* sp. 2 *aff. fraterculus*, and in F1 hybrids obtained from laboratory reciprocal crosses. Natural populations were sampled in a zone of sympatry and in two geographically distant regions with different climatic conditions. Asymmetry was scored as the differences between the number of bristles and of setae on the right and left sides of the head, males and females analyzed independently. The two traits exhibited variability according to the model of fluctuating asymmetry (FA). No significant differences among samples were found in the FA of frontal bristles. A significant FA was observed for the postocular setae of *A.* sp. 1 males from a southern population (Vacaria, RS) as compared to the asymmetry exhibited by males and females of some other samples. No significant differences in FA were observed among the interspecific hybrids and the laboratory samples of both parental species. The higher FA found in the males from Vacaria was attributed to climatic conditions prevailing in that region. The absence of a higher FA in hybrids may be related to the relatively recent evolutionary history of the two species.

Key words: developmental instability, fluctuating asymmetry, fruit flies, insect.

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Introduction

Fluctuating asymmetry (FA) is interpreted as the result of small disturbances during development, occurring at random on either side of a bilaterally symmetrical morphologic trait (van Valen, 1962, Palmer and Strobeck, 2003). Asymmetry is thought to reflect the developmental stability of an organism and, as such, thought to be under the influence of genetic and environmental stresses (Zakharov, 1992). Several biotic and abiotic stressors have been reported as being responsible for environmental influences on the symmetry of bilateral traits in different organisms (Leary and Allendorf, 1989, Parsons, 1990, Polak, 1993, Woods *et al.*, 1999, Indrasamy *et al.*, 2000), although other studies reported contradictory results (rev. Hoffman and Woods, 2003, Leamy and Klingenberg, 2005). One source of inconstancy may reside in the choice of traits to be used, since the FA response seems to be charac-

ter-specific (Woods *et al.*, 1999, Indrasamy *et al.*, 2000, Hasson and Rössler, 2002). Analysis of relationships between genetic stresses and levels of FA has also yielded contradictory results. Meta-analyses and recent findings, for example, do not support either a general association of heterozygosity with FA (Vøllestad *et al.*, 1999, Leamy and Klingenberg, 2005) or an association of FA with fitness components (Lens *et al.*, 2002a, Kruuk *et al.*, 2003, Leamy and Klingenberg, 2005). However, in some instances, clear-cut differences in FA were found. In the tephritid fruit fly *Bactrocera (Dacus) dorsalis*, a significant increase in FA was observed during the period that preceded its eradication from Japan, as a result of the decrease in population size and loss of genetic diversity (Tsubaki, 1998). Another example is the bird *Turdus helleri*, in which variation in FA was correlated with the degree of habitat disturbance (Lens *et al.*, 2002b). In zones of hybridization between two species, a decrease in developmental stability has been predicted, but conflicting results have been obtained (Markow and Ricker, 1991). In some hybridization zones, there was no excess of FA, while in others there was a positive correlation between FA and introgression (*e.g.*, Jackson, 1973,

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Graham and Felley, 1985). The two contrasting results could be related to the relative age of the hybridization zones, as in more recent hybridizations there may not have been enough time for coadaptation of the two parental genomes (Graham and Felley, 1985).

Developmental stability is a crucial parameter that has to be assessed in programs of environmental monitoring and conservation (Leary and Allendorf, 1989), as well as in mass production of insects for use in the control of insect pests (Clarke and Mackenzie, 1992). Among the insect pests, the tephritid fruit flies represent one of the most important groups of worldwide distribution (White and Elson-Harris, 1992), and it has been suggested that FA may also be used as an additional indicator of the "quality" of individuals produced in biofabrics (Hunt *et al.*, 1998).

Asymmetric patterns have been described for the head bristles in the fruit flies *Ceratitis capitata* (Hunt *et al.*, 1998, Pires *et al.*, 2004) and *Anastrepha grandis* (Pires *et al.*, 2004) and shown to be cases of FA. However, there are no studies on the degree of fluctuating asymmetry in fruit flies species submitted to different environmental conditions or genetic stresses. In this sense, it would be of interest to study asymmetry in species of fruit flies that present a wide geographic distribution and of which hybrids can be obtained in the laboratory. The *A. fraterculus* complex of cryptic species offers this opportunity. These species infest more than 67 host fruits of 17 different families (Zucchi, 2000) and are found from the southern United States through the Caribbean islands and down to central Argentina (White and Elson-Harris, 1992). Three species of the complex have been found in Brazil, and temporarily named *A.* sp. 1 *aff. fraterculus*, *A.* sp. 2 *aff. fraterculus*, and *A.* sp. 3 *aff. fraterculus.* They were found in zones of both allopatry and sympatry, where no hybrids were found as yet. However, hybrids can be obtained in the laboratory, but hybridization induces a reduction in egg hatching and in the survival of both males and females. Since the males are more affected, it causes a deviation in the adult sex ratio according to Haldane's rule (Selivon and Perondini, 1998, Selivon *et al.*, 1999, 2004, 2005). Studies on reproductive isolation among samples of *A. fraterculus* from other regions of South America also showed reduced viability associated with Haldane's rule (Vera *et al.*, 2006).

The present study was set to describe the levels of asymmetry of some characters in samples of *A.* sp. 1 *aff. fraterculus* and *A.* sp. 2 *aff. fraterculus* from populations either in sympatry or found in allopatry in areas with diverse climatic regimens, and to compare the asymmetry levels of these samples of natural populations with the asymmetry of flies from colonies maintained under controlled conditions in the laboratory. Another objective was to test whether or not hybridization of these two related species would change the level of asymmetry of some specific characters.

Material and Methods

Anastrepha species

The analysed species were *A.* sp. 1 *aff. fraterculus* and *A.* sp. 2 *aff. fraterculus*, herein shortly referred to as *A.* sp. 1 and *A.* sp. 2. They are closely related species of the *Anastrepha fraterculus* complex of cryptic species, hardly distinguishable from one another by usual taxonomic characteristics like the aculeus of the ovopositor. Clear distinction can however be achieved, based on conspicuous differences in the sex chromosomes, in four diagnostic loci of isozymes, in morphometric parameters of the wings and also the aculeus, and in eggshell morphology (Selivon and Perondini, 1998, Selivon *et al.*, 2005). Two populations of each species were sampled in different regions. In a zone of sympatry, the valley of the Paraíba River (Selivon *et al.*, 2005), *A.* sp. 1 was collected in guavas (*Psidium guajava*) and *A.* sp. 2 in oranges (*Citrus* spp), in the locality of Santa Isabel (State of São Paulo, 23º18' S, 46º13' W, altitude 554 m). The other *A.* sp. 1 sample was obtained from guavas collected in Vacaria (State of Rio Grande do Sul, 28°30' S, 50º54' W, altitude 950 m), and the other *A.* sp. 2 in Natal (State of Rio Grande do Norte, 05º48' S, 35º13' W, sea level). The *A.* sp. 1 samples were coded as SIg (Sta. Isabel, guavas) and Vac (Vacaria), and the *A.* sp. 2 samples as SIo (Sta. Isabel, oranges) and Nat (Natal). The infested fruits were brought to the laboratory, and samples of the emerged adults were separated by sex and preserved in 70% ethanol. Other adults that emerged from the Sta. Isabel samples were used in the interspecific crossing experiments.

Additionally, an *A.* sp. 1 (Lab1) and an *A.* sp. 2 (Lab2) sample were taken from laboratory colonies that have been maintained in the laboratory for 29 generations, and preserved in 70% ethanol. These colonies derived from flies collected in Santa Isabel, in guavas and oranges, respectively.

Crosses

Virgin adults of *A.* sp. 1 and *A.* sp. 2 emerged in the laboratory were used for the interspecific reciprocal crosses. The hybrid offspring of these crosses were coded as H12 (sp. 1 females X sp. 2 males) and H21 (sp. 2 females X sp. 1 males), respectively. The interspecific crosses were made according to Selivon *et al.* (1999, 2005). Briefly, five mating pairs were maintained in small population cages and fed corn meal protein hydrolysate, sugar and water. Guavas were furnished as substrate for larval development. The cages were maintained in a room with controlled temperature (~25 ºC), humidity (~70%), and a natural photoperiod regimen. Emerged F1 adult flies were collected and preserved in 70% ethanol.

Analyzed structures

Figure 1 shows the location of the two bilateral structures examined on the head of the flies, the frontal bristles and the postocular setae (Norrbom *et al.*, 1999). The frontal bristles are arranged in two rows, one on each side of the frons, parallel to the eyes, while the postocular setae are arranged in one row at the posterior rim of each eye, from the top toward the ventral side of the head. The specimens presented in Figure 1 were prepared for scanning electron microscopy according to Selivon and Perondini (1998), and examined under a Zeiss DMS 940 electron microscope.

Asymmetry measurements

The adult heads were examined under a stereomicroscope, and the number of specific bristles and setae on each side of male and female heads were scored separately. Sample size is given under Results for clarity. Asymmetry scores were calculated as the differences between the number of bristles and setae on right and left side (R-L), and asymmetry expressed by the modules, |R-L|, of the scores for each trait (Palmer and Strobeck, 2003). The samples were scored twice at different times by one of the authors (JMGAS), the two scores confronted, and discrepancies corrected by reexamining the involved specimens.

Statistical analysis

It is often observed that the degree of asymmetry is positively correlated to the magnitude of the trait measured. This scaling problem is usually corrected by dividing the asymmetry score of each individual (R - L) by the average magnitude of the trait measure for that individual on each

Figure 1 - Scanning electron micrographs of *A.* sp. 1 heads: (A) frontolateral view of the head; (B) dorsal view showing the postocular setae (arrow); frontal bristles (arrows) of an asymmetric (C) and a symmetric (D) individual. Scales in (A) and (B) = 200 μm; (C) and (D) = 100 μm.

side of the body (R + L/2) (Palmer and Strobeck, 2003). In order to detect possible scaling problems, correlations between the asymmetry value for each trait and the average number of bristles and setae on each side of the head were calculated (Palmer and Strobeck, 2003). Two tests were employed to evaluate the type of asymmetry, fluctuating asymmetry (FA, defined as random variation on each side of the bilateral traits), directional asymmetry (DA, greater trait development on a certain side of the body), or antisymmetry (AS, deviation from symmetry, forming a platykurtic or bimodal distribution) (van Valen, 1962), presented by the traits. First, the number of bristles and setae on each side of the head was compared by a paired *t*-test that indicates if a distribution has or not a mean value equal to zero. Secondly, the means, skewness and kurtosis of the signed asymmetry distributions were calculated, since - for traits exhibiting FA - the values should be normally distributed around zero (van Valen, 1962, Palmer and Strobeck, 2003). The differences in the level of unsigned asymmetry for each trait among the different samples were evaluated by two-factor ANOVA (sex and sample) and Newman-Keuls tests, preceded by the Bartlet test for homogeneity of variances (Zar, 1999). An overall, holistic asymmetry for each sample was calculated by adding the asymmetry of the two traits for each individual, after having standardized the signed asymmetry values for each trait to a mean equal to zero and a variance equal to one (Indrasamy *et al.*, 2000). The pooled asymmetry was tested by ANOVA, as mentioned above. The statistical tests were performed with the Statistica 6.0 (Statsoft Inc., 1997) software.

Results

Asymmetry for each trait was scored independently for male and female individuals, and the mean values are shown in Table 1. Regression of the individual asymmetry scores for the average number of frontal bristles and postocular setae on each side of each individual showed no significant correlations in the samples (data not shown). Hence, as indicated by Palmer and Strobeck (2003), no correction for scaling was necessary for the asymmetry scores of bristles and setae in the analyzed samples.

The results of the paired *t*-test applied to values of the right and left side of individuals from the different samples showed that the distributions of the asymmetry scores obtained for the two bilateral structures had mean values equal to zero (data not shown). Only in two cases, the frontal bristles of Nat and H21 females, the distributions presented a slight departure from the mean zero. These distributions, however, showed opposite results, a skewing to the left in the Nat sample and to the right in H21, suggesting that they do not really represent cases of DA. Indeed, the signed asymmetry variation for both traits in all samples must represent cases of true FA, since no skewness nor kurtosis were found for the distributions of R - L values, as shown in Table 1.
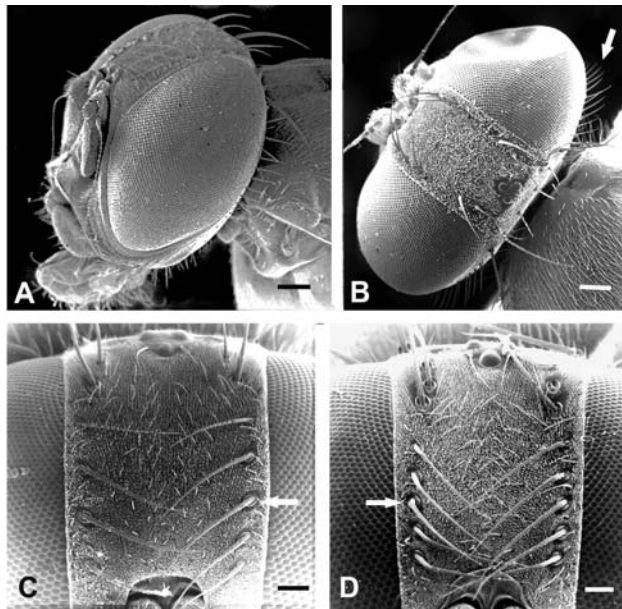
Table 1 - Means, skewness and kurtosis of signed asymmetry distributions and unsigned asymmetry of frontal setae and postocular bristles in samples of *A*. sp.1 (Vac , SIg, Lab1), *A*. sp.2 (Nat, SIo, Lab2) and hybrids (H21, H12) from reciprocal laboratory crosses.

Setae/ bristles	Gender	Samples	N	Asymmetry (R-L)				Unsigned asymmetry	
				Mean	s.d.	Skewness	Kurtosis	Mean	s.d.
Frontal	Males	Nat	100	-0.06	0.763	0.086 ns	0.895 ns	0.46	0.610
		SIo	99	0.08	0.817	-0.151 ns	0.272 ns	0.55	0.611
		Vac	79	-0.03	0.891	-0.396 ns	-0.230 ns	0.71	0.535
		SIg	100	-0.08	0.8	0.146 ns	-0.161 ns	0.56	0.574
		Lab1	50	-0.02	0.769	0.315 ns	-0.457 ns	0.54	0.542
		Lab2	50	-0.14	1.049	-0.039 ns	-0.243 ns	0.74	0.751
		H21	99	-0.01	0.776	-0.273 ns	0.893 ns	0.48	0.612
		H12	66	0.11	0.636	0.281 ns	0.504 ns	0.38	0.519
	Females	Nat	103	-0.18	0.772	0.183 ns	-0.325 ns	0.56	0.554
		SIo	103	0.00	0.779	-0.126 ns	-0.332 ns	0.54	0.556
		Vac	100	0.06	0.763	-0.380 ns	-0.412 ns	0.54	0.540
		SIg	96	-0.03	0.76	-0.241 ns	0.308 ns	0.49	0.580
		Lab1	50	-0.04	0.781	-0.463 ns	0.027 ns	0.52	0.579
		Lab2	50	-0.16	0.817	0.308 ns	-0.192 ns	0.60	0.571
		H21	75	0.16	0.678	-0.207 ns	0.861 ns	0.43	0.550
		H12	100	0.11	0.777	0.201 ns	-0.485 ns	0.55	0.557
Postocular	Males	Nat	100	-0.15	1.919	-0.394 ns	0.028 ns	1.51	1.185
		SIo	99	-0.26	1.601	0.426 ns	0.403 ns	1.27	0.998
		Vac	79	-001	2.091	-0.104 ns	0.029 ns	1.68	1.225
		SIg	100	0.01	1.418	-0.191 ns	-0.200 ns	1.05	0.947
		Lab1	50	0.22	1.787	-0.011 ns	0.403 ns	1.34	1.188
		Lab2	50	-0.14	1.927	0.417 ns	-0.185 ns	1.52	0.931
		H21	99	0.17	1.803	-0.244 ns	0.604 ns	1.42	1.204
		H12	66	-0.14	1.796	-0.216 ns	-0.654 ns	1.44	1.069
	Females	Nat	103	0.04	1.633	-0.160 ns	0.113 ns	1.24	1.052
		SIo	103	-0.23	1.682	-0.027 ns	-0.349 ns	1.32	1.059
		Vac	100	0.12	1.559	0.091 ns	-0.059 ns	1.14	1.064
		SIg	96	0.16	1.475	-0.175 ns	-0.236 ns	1.18	0.894
		Lab1	50	0.06	1.531	-0.463 ns	0.027 ns	1.14	1.010
		Lab2	50	-0.22	1.753	0.308 ns	-0.192 ns	1.34	1.359
		H21	75	0.16	1.669	0.152 ns	-1.062 ns	1.39	0.928
		H12	100	-0.04	1.803	-0.055 ns	-0.715 ns	1.42	1.027

The mean values of the unsigned asymmetry scores, |R-L|, of each trait for males and females from different samples are shown in Table 1. Differences in asymmetry values were compared individually for each trait. The ANOVA test showed distinct patterns of variation for the two traits scored (Table 2). For the frontal bristles, no significant differences in FA were found either between the sexes, or the samples, within or between the species *A*. sp. 1 and *A*. sp. 2, or between their reciprocal hybrids. Similar results were found when the FA of postocular setae was compared between samples from laboratory colonies and the reciprocal hybrids (Table 2). However, significant interactions between sex and sample in the FA levels of postocular

setae were found among the samples of natural populations, and in the comparisons of hybrids either to laboratory samples or to samples from natural populations (Table 2). The Newman-Keuls test showed the following results: (1) among the samples of natural populations of the two species, *A*. sp. 1 males from Vac showed a higher FA than females of the same population (p = 0.011), than conspecific SIg males (p = 0.002) and females (p = 0.019), and than *A*. sp. 2 males from SIo (p = 0.048); (2) among the natural populations and laboratory samples, FA was higher in the males from Vac than in their conspecific females (p = 0.027), and than in both males (p = 0.004) and females (p = 0.043) from SIg; (3) comparisons among samples of

Table 2 - Two-factor ANOVA (sex and sample) for the unsigned fluctuating asymmetry values of frontal bristles and postocular setae in samples from natural and laboratory populations of *A.* sp. 1 and *A.* sp. 2 and their hybrids.

	Frontal						Postocular					
Source	df effect	MS effect	df error	MS error	F	p-level	df effect	MS effect	df error	MS error	F	p-level
	Natural populations						Natural populations					
Sex	1	0.230	3	0.623	0.370	0.586	1	4.899	3	4.422	1.108	0.370
Samples	3	0.469	3	0.623	0.753	0.590	3	3.399	3	4.422	0.769	0.583
Sex*samples	3	0.623	772	0.326	1.909	0.127	3	4.422	772	1.111	3.979	**0.008***
	Natural and laboratory populations						Natural and laboratory populations					
Sex	1	0.546	5	0.421	1.297	0.306	1	6.361	5	2.714	2.344	0.186
Samples	5	0.566	5	0.421	1.344	0.377	5	2.422	5	2.714	0.892	0.548
Sex*samples	5	0.421	968	0.337	1.247	0.285	5	2.714	968	1.118	2.426	**0.034***
	Laboratory populations and hybrids						Laboratory populations and hybrids					
Sex	1	0.011	3	0.587	0.018	0.902	1	1.485	3	0.273	5.430	0.102
Samples	3	1.177	3	0.587	2.006	0.291	3	0.898	3	0.273	3.283	0.178
Sex*samples	3	0.587	532	0.342	1.716	0.163	3	0.273	532	1.146	0.239	0.869
	Natural populations and hybrids						Natural populations and hybrids					
Sex	1	0.230	3	0.608	0.379	0.582	1	3.671	5	2.867	1.281	0.309
Samples	3	0.479	3	0.608	0.788	0.575	5	2.691	5	2.867	0.939	0.527
Sex*samples	3	0.608	772	0.327	1.861	0.135	5	2.867	1108	1.121	2.557	**0.026***

(*) significant at 0.05.

natural populations and the laboratory hybrids showed similar results, Vac males exhibiting a higher FA than their conspecific females ($p = 0.027$), and than males ($p = 0.004$) and females ($p = 0.05$) from SIg, although the difference with the latter one was at the limit of significance.

Comparisons of the pooled standardized FA values of the two traits among samples by 2-way ANOVA showed no significant differences in FA levels (data not shown), even for the cases of Vac males which had shown higher FA levels of the postocular setae than some of the samples, as described above.

Discussion

The results of the analysis reported here showed, as previously observed for the frontal bristles of *Anastrepha grandis* and *Ceratitis capitata* (Pires *et al.*, 2004), that frontal bristles and postocular setae of *A.* sp. 1 and *A.* sp. 2 consistently exhibit a FA pattern of variation. They also indicate that the two traits may show different levels of disturbance. The frontal bristles, for example, showed the same level of FA in the two species, *A.* sp. 1 and *A.* sp. 2, regardless of sex and whether the flies derived from a natural population, laboratory colonies or were hybrids obtained in the laboratory. The same was found for the postocular setae of *A.* sp. 2 and some samples of *A.* sp. 1 (SIg population). However, *A.* sp. 1 males from the Vac population exhibited a significantly higher FA of the postocular setae when compared to conspecific individuals. These findings are in line to other studies indicating that an asymmetry response

seems to be related to noises in the development of specific characters, rather than to an overall developmental instability of the individuals (Woods *et al.*, 1999, Indrasamy *et al.*, 2000, Hasson and Rössler, 2002, Leamy and Klingenberg, 2005). Indeed, the pooled asymmetry of the two traits indicated no significant differences among the samples, a result that masks the significant FA of postocular setae in the Vac males.

Variation at the genotypic level as related to FA levels is controversial, and has not been confirmed in meta-analysis and recent studies (Vøllestad *et al.*, 1999, Lens *et al.*, 2002a, Kruuk *et al.*, 2003, Leamy and Klingenberg, 2005). In line with these arguments, analysis of 19 enzymatic loci indicated that the heterozygosity of the Vac population does not differ significantly from those observed for other populations of the same species (*A.* sp. 1), and is similar to that of *A.* sp. 2 (Selivon *et al.*, 2005). Hence, the higher FA of Vac males cannot be attributed to a lower level of heterozygosity.

The higher FA of Vac males may be related to environmental stress. Stressful conditions have been associated to increased FA in some studies, for example, in the bird *Turdus helleri* (Lens *et al.*, 2002b) and in the fruit flies *Bactrocera* (*Dacus*) *dorsalis* (Tsubaki, 1998) and *Drosophila melanogaster* (Woods *et al.*, 1999). Temperature is an abiotic factor that has been shown to be related to phenotypic variation. In *Drosophila melanogaster*, an increase in the length variation of the body (David *et al.*, 1983) and wings (Tantawy and Mallah, 1961) was observed at the extremes of the experimental temperatures

employed. In *D. melanogaster* and *D. buzatti*, an increase in the FA of sternopleural bristles, number of branches in the arista, and in the wing length was also found at the extremes of the experimental temperature range (Imasheva *et al.*, 1997). In the aphid *Myzus persicae*, a temperature-related phenotypic plasticity was observed, but there was no linear relationship between the temperature values and the degree of FA (Kanegae and Lomônaco, 2003). Of all the localities where samples were taken for the present analysis, harsher conditions certainly prevail in Vacaria, due to its southern latitude and higher altitude. Thus, one may consider that the higher FA of *A.* sp. 1 males from Vacaria may be related to the more severe climatic conditions of that region. A similar effect, however, was not observed in the SIo and Nat samples of *A.* sp. 2, taken from localities at expressive latitudinal distance. It is therefore reasonable to consider that the specific climatic conditions of the two regions may not differ enough so as to interfere with the developmental processes of these flies, or that *A.* sp. 2 shows a different response to stress, exhibiting a distinctive level of buffering in its development.

It is relevant to note that no differences in asymmetry were observed in F1 hybrids between *A.* sp. 1 and *A.* sp. 2. These results are in line to the data on other species, in which hybridization did not increase the phenotypic variation in the F1 generation either (Jackson, 1973). On the other hand, there are hybridized populations that show higher levels of developmental instability (Leary *et al.*, 1985, Graham and Felley, 1985). In a study on hybridization between *D. melanogaster* and *D. simulans*, a significant increase in the FA of sternopleural and fronto-orbital bristles and of wing length was found in F1 females, but not in males (Markow and Ricker, 1991). The contrasting asymmetry response to hybridization was interpreted as related to the degree of isolation and genomic incompatibility between the two species (Graham and Felley, 1985). Since it is estimated that the species of the *fraterculus* complex have a recent evolutionary origin (Morgante *et al.*, 1980, Smith-Caldas *et al.*, 2001, Selivon *et al.*, 2004, 2005), it could be assumed that genomic coadaptation is still at such a level that hybridization was not sufficient to induce alterations in the developmental stability of the first generation of hybrids.

However, an alternative hypothesis to explain the fact that FA was not increased in the F1 hybrids is based on the observation that hybridization between the two species of *Anastrepha* causes a decrease in viability of both sexes and an alteration in the F1 progenies according to Haldane's rule (Selivon *et al.*, 1999, 2005). Differential mortality among the offspring may be masking the level of asymmetry, since the nonviable individuals might have exhibited high levels of FA, if they had survived. Thus, following hybridization, the survivors would exhibit levels of FA similar to those of their parents (Campbell *et al.*, 1998).

Bristle asymmetry may also be implicated in reducing the mating success of males of some dipteran species. It was observed that *Drosophila pseudoobscura* males which had mated exhibited less asymmetry than males which did not copulate (Markow and Ricker, 1992). Similarly, asymmetric scorpion fly (*Panorpa japonica*) males showed less mating success than the symmetric ones (Tornhill, 1992). In the tephritid *C. capitata*, symmetric males also had greater mating success than the asymmetric ones (Hunt *et al.*, 1998), observations that have been confirmed in natural populations (Hunt *et al.*, 2004).

Additional analyses are still required to ascertain whether or not the asymmetric variation of the frontal or other head bristles has any implication in the mating success in *Anastrepha* species, and to evaluate the robustness of these traits as indicators of stressful conditions.

Acknowledgments

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