Rev. Chilena Ent.
2001, *28*: 17 - 24

PREADULTS INTERACTIONS BETWEEN THE COLONIZING *DROSOPHILA SUBOBSCURA* AND THE ESTABLISHED SPECIES *D. HYDEI, D. IMMIGRANS* AND *D. MELANOGASTER* (DIPTERA: DROSOPHILIDAE) IN CHILE USING NATURAL SUBSTRATES.

ZAID ALAMIRI[1]

ABSTRACT

The Palaearctic species *Drosophila subobscura* recently colonised Chile successfully. The results showed that the presence of the well-established species did not interfere with *D. subobscura* oviposition behaviour. When it competed on limited food resources with *D. immigrans* and *D. melanogaster;* its developmental time and viability were significantly affected. On the other hand, the development time *D. hydei* and *D. immigrans* only showed considerable reduction; however, all of these three species did not suffer changes in their viability. A double edge mechanism could explain the results reported here. The larval wastes of *D. subobsucra* facilitate the development of these established species while the metabolic wastes of the latter, except *D. hydei,* affect that of *D. subobscura.* An interesting thing is that competitive interactions had no significant influence on *D. subobscura* body size. This, in the context of the reproductive fitness, could explain its colonisation ability in spite of being poor competitor.

Key words: colonisation, competitive interactions, *D. subobscura* and larval metabolic wastes.

RESUMEN

Recientemente, la especie paleártica *D. subobscura* ha colonizado a Chile exóticamente. La presencia de la otras especies bien establecidas, como han mostrado los resultados de este trabajo, no intervino con la conducta de oviposición de este especie. Sin embargo, cuando estuvo (*D. subobscura*) en competencia, sobre un recurso limitado de alimento, con *D. immigrans* y *D. melanogaster,* tanto su tiempo de desarrollo como su viabilidad fueron afectados significativamente. Por el otro lado, el tiempo de desarrollo de solamente *D. hydei* y *D. immigrans* fue disminuido considerablemente. En cuanto a la viabilidad de esas tres especies, ninguno mostró cualquier cambio. Los resultados obtenidos podrían ser interpretados en base de un mecanismo de doble sentido que se trata de que los desechos larvales de la especie colonizadora han facilitado el crecimiento y el desarrollo de las tres especies con quienes *D. subobscura* coexiste en la naturaleza; Sin embargo ella misma sufrió de los desechos metabólicos de las últimas, con excepción de los de *D. hydei.* Una cosa interesante es que la interacción competitiva con las tres especies no tuvo influencia sobre su masa corporal. Mirándolo dentro del contexto de su eficiencia reproductiva, eso podría explicar su habilidad colonizadora a pesar de ser mala competidora.

Palabras clave: colonización, interacciones competitivas, *D. subobscura* y desechos metabólicos larvales.

INTRODUCTION

Drosophila subobscura Collin is a typically Palaearctic species distributed all over Europe, North Africa and Asia Minor, with the exception of the northern parts of Scandinavia and Finland (Lákoova & Saura, 1982). It was detected in Chile for the first time in Puerto Montt (Lat. 41° 60´S) in 1978 (Brncic & Budnik, 1980). Few years later, it expanded its distribution range, and thus occupying along North - South slope so ecologically contrasting zones where it is hot & semi- arid in the North, and windy, wet and cold in the South. It was found far southern as Punta Arenas (Lat. 54° 40´S9), associated with native wild flora such as *Berberis*

---

[1]Department of Entomology, University of California, Riverside, CA, 92521. E-mail: zalamir@hotmail.com

(Recibido: 11 de mayo de 2000. Aceptado: 6 de octubre de 2001)

buxifolia, Maytenus magellanica, Embothrium coccineum and different species of *Nothofagus* (Brncic *et al.*, 1981).

It has also crossed the Andes mountain range and was detected in the Pacific coast of North America, where it has now become established, as Prevosti *et al.*, (1982) argued that its rapid extension could be attributed to its high reproductive potential associated with the environmental conditions in its new territory which are similar to that ones found in its origin places; secondly, its rapid genetic microdifferentiation in terms of chromosomal and enzymatic polymorphism. To put it in another words, the ecological versatility expressed by the species is related with its double edge genetic strategy of being semi-rigid as well as semi-flexible, capable of sharing the advantage's of both types of genetic adaptive strategies, characterised by its great colonizing ability which has transformed it to a quasi-cosmopolitan one, able to coexist not only with domestic species of the genus in many man made environments but also with the local *Drosophila* fauna in wild habitats when colonizing a new territory (Budnik & Brncic, 1987).

In regards to its competitive ability Budnik *et al.* (1982,1983,1989,1993) in series of competition experiments using the technique of seeding a certain number of eggs in culture medium, confirmed that its pre-adult fitness in terms of viability and egg-to-adult development time, was negatively affected when being in competition with some of species such as *D. simulans, D. immigrans, D. melanogaster* and *D. pavani*. The present experiment is more representative of nature in the sense of leaving the adult females to deposit their eggs together; secondly, instead of culture medium; pieces of apple fruits were used as a natural substrate. Thus, the aim of the present study was to examine four components of the fitness, namely the fecundity, development time, viability and body size of *D. subobscura* when competes on limited food sources with *D. hydei; D. immigrans* and *D. melano*gaster.

MATERIALS AND METHODS

The four species populations used in the study were come from flies captured in the university experimental station located at the north of Valdivia province (39° 45´S; 73° 14´W). They were maintained during the experimental period in 250-cm glass bottles containing 50 cc culture medium (Burdik, 1954). The experiment consisted of a combinations of 5 adult females pairs at their maximum fecundity age of *D. subobscura* with each of *D. melanogaster*; *D. immigrans* and *D. hydei;* as they were left to oviposit for 24 hr in 2.5cmx7.5 cm glass vial containing sterilized moisten sand river at the bottom with 6.72 gr. of apple (*Malus sylvestris*, CV. Granny). The following day, the insects were retired to count the number of eggs laid by the combination of each two species. Then the vials were kept in a controlled room of 22 ±±1 °C, 60% R.H. and 12L: 12D(08:00 – 20:00).

Flies were collected once daily (09.00-10.00 AM), as it continued for one week following the last adults emergence, as separated by species and kept in alcohol 75% in small plastic vials for analysis required. The fecundity, mean thorax length, egg-to-adult development time, and viability were measured. Adult thorax length was measured to the nearest 0.02 mm with a binocular stereoscope fitted with an ocular micrometer, from the anterior margin of the thorax to the posterior tip of the scutellum, as viewed from the side. Egg-to-adult development time was estimated as the average number of days from experiment initiation to adult emergence, where all females & males counted at a particular scoring were taken as having emerged at the midpoint in time between that scoring and the previous one. Ten replicates for each combination were used.

RESULTS

In view of the definition of competition adopted in this paper, no direct comparisons were made between species. Instead, the reproductive fitness of each species in the mixed (experiment) population was compared with that of conspecific reared in the pure (control) populations.

The first noticeable observation as Table 1 shows is that *D. subobscura* displayed almost identical fecundities in the pure and in all its combinations in the mixed cultures. This indicates that the presence of the other species didn't affect its oviposition behaviour. The same holds true for *D. hydei* and *D. melanogaster*. However, *D. immigrans* reduced its fecundity significantly when it was in combination with *D. subobscura* (T=3.86; 9 df; P<0.004).

TABLE 1
FECUNDITY OF THE SPECIES IN THE PURE AND MIXED CULTURES EXPRESSED AS MEAN EGGS NUMBER ±± S.E/VIAL.

Combination	species			
D.subobscura+ D.hydei	D. subobscura		D. hydei	
	pure	mixed	pure	mixed
	25.4±4.4	25.3±3.3	113.1±17.8	78.5±9.7
D.subobscura+D.immigrans	D. subobscura	D. immigrans		
	pure	mixed	pure	mixed
	25.4±4.4	29.9±4.3	105.0±8.1	56.4±5.2**
D.subobscura+D.melanogaster	D. subobscura	D. melanogaster		
	pure	mixed	pure	mixed
	25.4±4.4	23.3±4.3	72.0±8.2	56.0±5.7

Table 2 discloses that *D. subobscura* preadults lengthened significantly the time(in days) needed to reach the adult stage when they compete with the other species(T=4.5, 9d.f;p<0.004 with *D. melanogaster* and T=6.83, 9 d.f; P<0.000 with *D. immigrans*), with the exception of *D. hydei* (T=1.65, 9 d.f; P<0.11)where no such differences between pure and mixed culture were detected. It could be said that some mechanism helped facilitate its growth; however, these species in turn, showed a significant reduction in their period of time (T=9.5, 9 d.f; P<0.000 and T= 7.1, 9 d.f; P<0.000 for *D. hydei* and *D. immigrans;* respectively); meanwhile *D. melanogaster* increased its developmental time significantly (T=10.7, 9 d.f; P<0.000).

With regard to viability, it is clearly evident as table 3 demonstrates that the presence of *D. melanogaster* and *D. immigrans* preadults only, had negatively affected *D. subobscura* viability (T=2.2; 9d.f; P<0.05 and T=2.5, 9 d.f; P<0.03, respectively). On the other hand, the presence of the *D. subobscura* preadults had no effect on the viability of the all three other species.

It is obvious from the graphics that the survival percent of *D. subobscura* emerging daily and hence its total viability was significantly high in the presence of *D. hydei* as compared to that of its combinations with *D. immigrans* and *D. melanogaster* and even its control. This means that some kind of facilitation mechanism occurred when it was accompanied with *D. hydei* .

It is interesting to note from the graphics also that *D. subobscura* presence with other species had a positive effect on time of emergence or survival

TABLE 2
EGG-TO-ADULT TIME OF DEVELOPMENT IN DAYS IN THE PURE AND MIXED CULTURES (MEANS ± S.E)

Combination	species			
D.subobscura+ D.hydei	D. subobscura		D. hydei	
	pure	mixed	pure	mixed
	21.7±0.5	24.2±1.6	27.5±1.3	19.2±1.4***
D.subobscura+D.immigrans	D. subobscura		D. immigrans	
	pure	mixed	pure	mixed
	21.7±0.5	28.2±2.1***	22.9±0.9	16.9±2.5***
D.subobscura+D.melanogaster	D. subobscura		D. melanogaster	
	pure	mixed	pure	mixed
	21.7±0.5	28.2±1.3***	16.4±0.87	19.9±1.2***

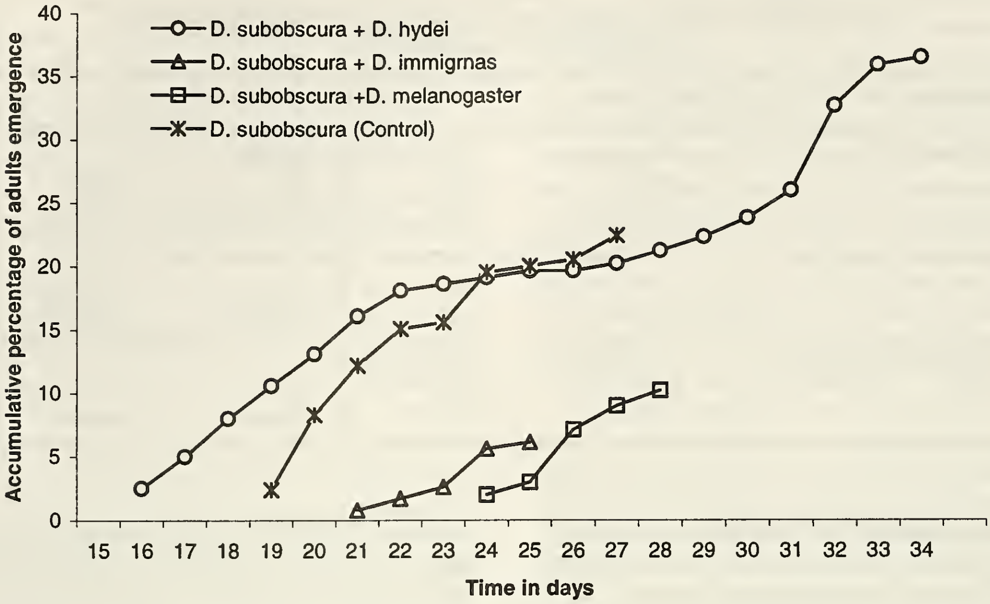

Figure 1. *D. suboscura* accumulated emergence and viability when it was competing with *D. hydei,* as compared to other species combinations.

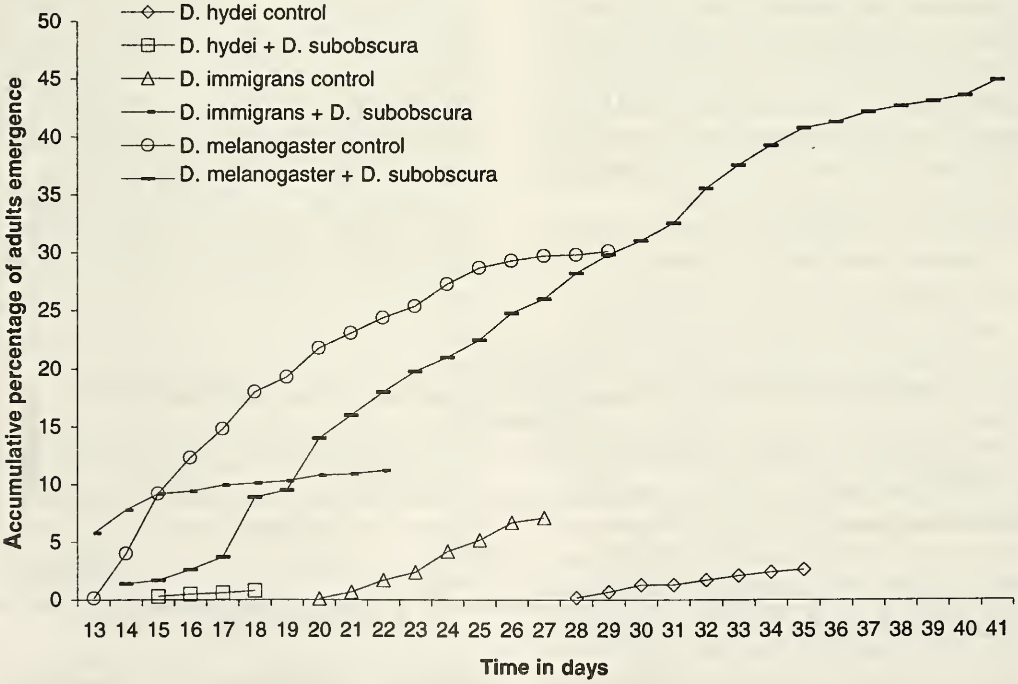

Figure 2. *D. suboscura* larvae facilitation effect on the developmental time of each of *D. hydei*, *D. immigrans* and *D. melanogaster.*

TABLE 3
TOTAL VIABILITY IN THE PURE AND MIXED CULTURES
(MEANS ± S.E)

Combination	species			
D.subobscura+ D.hydei	D. subobscura		D. hydei	
	pure	mixed	pure	mixed
	30.1±5.4	34.3±2.4	20.2±3.3	18.0±1.2
D.subobscura+D.immigrans	D. subobscura		D. immigrans	
	pure	mixed	pure	mixed
	30.1±5.4	13.7±2.6*	23.4±3.9	23.9±5.4
D.subobscura+D.melanogaster	D. subobscura		D. melanogaster	
	pure	mixed	pure	mixed
	30.1±5.4	15.2±2.9*	45.9±5.6	53.3±5.0

or on both. For example, the early emergence of *D. hydei*; the high survival of *D. melanogaster;* the time of emergence or as is the case of *D. immigrans* both survival and time of emergence were positively affected. Survival is high.

BODY SIZE OR THORAX LENGTH

The thorax lengths of *D. subobscura* females (Table 4) did not indicate or present divergence from the control when coexisting with the three species. This is very interesting because of its direct relation with the reproductive fitness and hence the descendants left by the females. Since it implies females' ability to withstand the negative effects that might result from competition. the other species also maintained their body size with exception of *D. immigrans*, which had increased it significantly (T=9.1, 9d.f; $P<0.000$).

DISCUSSION

In this paper, "interspecific competition" is applied to any interaction between two species sharing a common resource such as the presence of one species reduces the fitness of the second species (Birch, 1957; Ayala, 1970).

There has been increasing interest about *D. subobscura*, since its detection for first time in Puerto Montt in 1978, to accomplish genetic and ecological studies (centered on the mechanisms adopted by this species that disclose the sp. colonization ability) orientated to explore the mechanisms adopted by this species which stands

TABLE 4
FEMALES BODY SIZE IN THE PURE AND MIXED CULTURE
(MEANS ± S.E/vial).

Combination	species			
D.subobscura+ D.hydei	D. subobscura		D. hydei	
	pure	mixed	pure	mixed
	0.771±0.01	0.761±0.01	0.916±0.009	1.895±0.01
D.subobscura+D.immigrans	D. subobscura		D. immigrans	
	pure	mixed	pure	mixed
	0.771±0.01	0.771±0.01	0.832±0.009	1.11±0.08**
D.subobscura+D.melanogaster	D. subobscura		D. melanogaster	
	pure	mixed	pure	mixed
	0.771±0.01	0.792±0.1	0.626±0.091	0.618±0.007

behind its very successful colonization which helped colonise in very short time its new territory). This experiment goes in this context to analyse the competitive ability of *D. subobscura* in relation to the other species with which it shares breeding sites and consequently the food resources in the southern Chile. The results obtained here confirm in their general trend those reached by the other authors. However, when *D. subobscura* females were left with females of each of the three species mentioned in the study, it was clearly evident that their presence as adults did not affect or influence the *D. subobscura* female's oviposition behaviour even on limited space and food. A competitive situation for the site might disturb species from laying eggs as had been seen by Barker (1971) who found that in the mixed culture of *D. melanogaster* and *D. simulans,* the latter selected the medium surface centre laying more eggs there; while the former put little eggs on the border. In this study, the two species combinations laid more eggs randomly all over the medium surface without showing any apparent disturbance or competition for the site. Similar observations were found by Fellows & Heed (1972) who showed that the fecundities of *D. arizonensis* and *D. mojavensis* in mixed populations of 35 of the former and 5 of the later, on Cina (*Rathbunia alamosnesis*) and Agria (*Machaerocereus gummosus*) cactus were statistically similar, but not on Organ Pipe cactus (*Lemaireocereus thurberi*), in which *D. arizonensis* suffered a great than 50% fecundity reduction in the mixed culture. In the same study, it was seen that the presence of only 5 *D. nigrospiracula* reduced the expected net fecundity of both 35 *D. arizonensis* and 35 *D. mojavensis* by about 50%, on Saguaro cactus (*Carnegiea gigantea*). In this study, *D. immigrans* reduced its net fecundity about 50%. This observation is important as it can partially explain that the species conduct was not impeded by the occurrence of other species. Quietly , as it is the case in the nature, where the available resources are shared by different species together. And hence the females succeed in leaving descendants even if they would be subjected to competition. This was confirmed by Brncic (1987) who sustained that *D. subobscura* had emerged from the first three rotten fruits of *Eriobotrya japonica; Morus alba; Prunus avium* and *Vitis vinicola* collected in a suburban locality near Santiago; together with *D. immigrans, D. melanogaster* and *D. simulans.* This means that *D. subobscura* had used these fruits as a breeding sites together with the other species.

With regard to the time of development and viability, our results are largely in agreement with that of Budnik & Brncic (1982,1983); Budnik & Cifuents(1989) and Brncic (1987), who found that the development time and viability of *D. subobscura* were severely affected when it was in competition with *D. melanogaster, D. immigrans, D. simulans,* and *D. pavani,* probably by the effect of larval metabolic wastes which they are arm of double edge, it means in the case of *D. subobscura,* the metabolic wastes of other species produced this result. Botella *et al.* (1985) postulated that both urea and uric acid are responsible for such reduction in viability and length of development time. In contrast, regarding *D. hydei,* the results reported here which constitute the first empirical evidence, establish that this species does not interferes with *D. subobscura,* probably due to a process of mechanical facilitation produced by the larval tunneling of the medium. Arthur & Cassey (1992), declared that *D. hydei* tend to be deeper-feeding, and thus enhancing the coexistence, making it more easier and liquid for *D. subobscura* on one hand and on the other the metabolic larval wastes of both species had produced such mutual facilitation in which the presence of one species increase the fitness of others(Bos *et al.,* 1977).

As the results showed, all three species did not suffer significant changes in their viability; meanwhile the development time of *D. hydei* and *D. immigrans* was decreased significantly indicating that some kind of facilitation produced by the larval wastes had favoured them to shorten their time of development. Such observation was found also in these species on sharing common resources with *D. subobscura* (Budnik *et al.,*1982,1983,1989) or in another species (Budnik & Brncic, 1974, 1976; Budnik, 1980; Dawood & Strickberger,1969). *D. melanogaster,* in contrast to the other species and to results reported by Budnik *et al.,* (1982,1983,1989), when it was accompanied by *D. subobscura*, it lengthened its time of development significantly. This could be explained on the basis of crowding suffered by larvae which tend in such situation as Ménsua & Moya (1983) argued, to arrest their development in third instar, delaying thus their pupation considerably (up to 20 days at 25°C).

Besides, if we know that the number of *D. melanogaster* emerged from the mixed culture is twice the control number. To put it in another words, the time required for development, with increased crowding, is prolonged and there is tendency for emergence to be spread throughout a greater period(Miller, 1964). Furthermore, such events would participate also in increasing the pupal mortality of *D. subobscura* that was observed in this experiment, due to the larval activity of the other species especially that of *D. melanogaster* which lead to drowning in the medium (Moth & Barker,1976).

Finally, it is clear that there is a good relationship between body size and fitness which had in the case of *D. subobscura* an evolutionary significance in the sense that females maintained its body size would potentially leave descendants and hence help the species get its successful survival. The species evolutionary history may have a trade-off between the decreased viability and time of development and maintaining the body size in order to leave its offspring, and thus compensating for bad or negative effects suffered by the species when compete with others (Santos *et al.*,1994).

It is evident from the foregoing discussion that *D. subobscura* is competitively poor even if in low larval density (Table 1) as compared to that used in seeding eggs experiments. however, it disclosed a high colonisation ability since its detection. This indicates that to maintain its frequency in the nature, it should adopt some mechanisms to such as escaping competition by using breeding sites not used by other species (Alamiri, 2000) or rather being the first in colonise or use the resources before the other come (Shorrocks & Bingley,1994) which could among many other mechanisms account for its successful colonisation.

REFERENCES

Alamiri,Z. 2000. Oviposition behaviour in four species of Drosophila. Gayana.*64*(2):123-129.

Arthur,W. & S.Cassey.1992.Variability of a niche difference between *Drosophila hydei* and *D melanogaster*. Ecol. Entomol.*17*:354-358.

Ayala, F.J. 1970. Competition, coexistence and evolution, p. 121-158. In M.K.Hecht and W.C. Steere (ed) Essay in evolution and genetics in honor of Theodosius Dobzhansky, Appleton-Century-crofts, New York.

Barker, J.S.F. 1971. Ecological differences and competition interaction between *Drosophila melanogaster* and *Drosophila simulans* in small laboratory populations. Oecologia. *8*: 139-156.

Birch, L.C. 1957. The meaning of competition. Am. Nat. *91*: 5-18.

Bos,M.; B. Burrent, R. Farrow & R.A. Woods. 1977. Mutual facilitation between larvae of the sibling species *Drosophila melanogaster* and *D. simulans*. Evolution. *31*: 824-828.

Botella, L.M & J.L. Mensua.1986.Larval arrest in crowded cultures of *Tribolium castaneum* (Coleoptera). Environ. Entomol. *15*: 1264-1267.

Brncic, D.1987. Coexistencia de diferentes especies de *Drosophila* en frutas fermentadas naturalmente. Medio Ambiente *8*: 3-9.

——— & M. Budnik.1980. Colonization of *Drosophila subobscura* Collin in Chile. Drosophila Information Service *55*:20.

——&———.1987. Chromosomal polymorphism in *Drosophila subobscura* at different elevations in central Chile. Genetica. *75*: 161-166.

———., A. Prevosti, M. Budnik, M. Monclus & Ocaña. J. 1981. Colonization *of Drosophila subobscura* in Chile. 1.First population and cytogenic studies. Genetica *56*:3-9.

Budnik, M.1980. Alteration of the preadult viability induced by larval metabolic wastes in different species of *Drosophila*. Ciencia E Cultura *32*: 1391-1394.

——— & D. Brncic. 1974. Preadult competition between *Drosophila melanogaster, Drosophila simulans* and *Drosophila willistoni*. Ecology *55*: 657-661.

——— & ———. 1975. Effects of larval biotic residues on viability in four species of *Drosophila*. Evolution *29*:777-780.

——— & ———. 1982. Colonización de *Drosophila subobscura* Collin en Chile. Acta V Congr. Latinoam. Genetica. 177-188.

——— & ———.1983. Preadult competition between colonizing populations of *Drosophila subobscura* and established populations of *Drosophila simulans* in Chile.Oecologia *58*:137- 140.

——— & L. Cifuentes. 1989. Larval interactions between a colonizing populations 0f *Drosophila subobscura* and their established species of *Drosophila* in Chile. Rev. Brasil. Genet. *12*: 499-504.

——— & ———. 1993. Effects of larval biotic products on preadult viability in European and Chilean stocks of *Drosophila subobscura*. Evolución Biologica *7*: 303-311.

Burdick, A.B. 1954. New medium of reproductive quality stable at room temperature. Drosophila Information Service *28*: 170.

Dawood, M.M. & M.W. Strickberger. 1969. The effect of larval interaction on viability in *Drosophila melanogaster*. III. Effects of biotic residues. Genetics. *63*: 213-220.

Fellows, D.P. & W.B. Heed. 1972. Factors affecting host plant selection in desert-adapted catiphilic *Drosophila*. Ecology *53*: 850-858.

Lakovaara, S. & A. Saura. 1982. Evolution and speciation in the *Drosophila obscura* group. in: M. Ashburner, H.L. Carson y J.N.Thompson, JR.(eds). The genetics and biology of *Drosophila*. Academic Press, London. vol. 3b PP. 2-60.

Mensua, J.L. & A. Moya. 1983. Stopped development in crowded cultures of *Drosophila melanogaster.* Heredity *51*: 347-352.

Miller, R.S. 1964. Larval competition in *Drosophila melanogaster* and *D. simulans*. Ecology. *45*: 132-148.

Moth, J.J. & J.S.F. Barker. 1976. Interspecific competition between *Drosophila melanogaster* and *Drosophila simulans.* Oecologia. *23*: 151-164.

Prevosti, A., G. Ribo, M.P. Garcia, E. Sagarra, M. Aguada, L. Serra & M. Monclus. 1982. Los polimorfismos cromosómicos y aloenzimáticos en las poblaciones de *D. subobscura* colonizadoras de Chile. Actas V Congr. Latinoam. Genética 189-197.

Santos, M.; K. Fowler & L. Partridge. 1994. Gene-environment interaction for body size and larval density in *Drosophila melanogaster:* an investigation of effects on development time, thorax length and adult sex ratio. Heredity: *72*: 515-521.

Shorroks, B. & M. Bingley. 1994. Priority effects and species coexistence: Experiments with fungal - breeding Drosophila. J. Anim. Ecol. *63*: 799-806.