Longevity in the *Drosophila virilis* Species Group. II. The *D. montana* Phylad[1]

EDWARD J. DURBIN and JONG S. YOON, Department of Biological Sciences, Bowling Green State University, Bowling Green, OH 43403-0212

ABSTRACT. Longevity differences due to species, strain, and sex were examined for members of the *Drosophila montana* phylad of the *D. virilis* species group: *D. ezoana, D. kanekoi, D. littoralis, D. borealis, D. flavomontana, D. lacicola, D. montana,* and a new species (A) from British Columbia. Longevity of newly eclosed adults of each of these species on standard cornmeal medium was analyzed in order to discover intraspecific, interspecific, and sexual differences. *Drosophila flavomontana* Colorado males lived the longest (52 days), whereas *D. borealis* Minnesota males survived only 24 days. Female *D. kanekoi* lived almost 49 days, but females of species A died after 23 days. Strains of the other species had longevities between these extremes. This species phylad is divided into three subgroups based on cytological and biochemical characteristics. The results of the longevity study mirrored this phylogeny with two exceptions. Females lived longer than males for over one-half of the species. The results suggest that genetic similarity may contribute to adult longevity, although environmental interactions probably are also a factor.

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INTRODUCTION

Species of the insect family Drosophilidae have been favorite tools for studies on longevity and aging (Lamb 1978). The major focus of most of these experiments has been the role of various environmental factors in determining the adult longevity of *Drosophila* (David et al. 1983, Hollingsworth 1969, Felix and Ramirez 1967, Miller and Thomas 1958). Other authors have chosen to study longevity as affected by species, sex, and mating status (Maynard Smith 1958, 1959, Spiess et al. 1952). Recently, an approach combining environmental differences with phylogenetic similarity has been suggested by Schnebel and Grossfield (1983). They studied related species from different habitats and looked at both intraspecific and interspecific variation in length of adult life. As far as is known, however, no one has made an extensive study of longevity in relation to phylogeny.

This study is the second in a series of experiments designed to yield information regarding the role of genetic similarity in determining adult longevity of *Drosophila*. In the first study (Durbin and Yoon 1986), the *D. virilis* species phylad was examined for species and sex differences in longevity. The *D. virilis* species group is one of the most well studied groups of any organism. Throckmorton (1982) has written an excellent review on this group. The group is divided into two phylads: *virilis* and *montana*. The *montana* phylad consists now of at least eight species: *D. ezoana, D. littoralis, D. kanekoi, D. borealis, D. flavomontana, D. lacicola, D. montana,* and an as yet unnamed species (A) from Smithers, British Columbia.

The purpose of this study was to determine the adult longevity of both sexes for species of the *montana* phylad. The results enhance our knowledge of the basic biology of these species and will contribute to the understanding of evolution in *Drosophila*.

METHODS AND MATERIALS

The eight species (fifteen strains) in the *montana* phylad of the *D. virilis* species group were studied (Table 1). These species were obtained from the National *Drosophila* Species Resource Center, Bowling Green State University (BGSU). These strains were chosen because they are being used in cytological studies. Stock vials of each species contained approximately equal numbers of flies, and were maintained under the same conditions as the experimental vials.

The experimental procedure was the same as in the previous study (Durbin and Yoon 1986). Longevity was measured as the difference between eclosion date and date of death. The results were analyzed by species, strain, and sex with Tukey's Studentized Range Test (Zar 1984). Results from both sexes were pooled for determination of significant species differences.

RESULTS AND DISCUSSION

Adult longevity in the *D. montana* species phylad varied with species, strain, and sex (Table 1). The mean longevities were less, however, than those reported by Durbin and Yoon (1986) for the *virilis* phylad of this same species group. In that study, significant differences were also observed owing to strain and sex. Mean adult longevity in the *montana* phylad ranged from 23 days for *D. borealis* Minnesota males to 52 days for *D. flavomontana* Colorado males, and from 24 days for established species A females to almost 49 days for *D. kanekoi* females (Table 1).

The *montana* phylad contains three loosely-defined subgroups based on preliminary chromosomal studies (L. H. Throckmorton, unpubl. data). The first subgroup consists of *D. ezoana, D. littoralis, D. kanekoi,* and species A, whereas the second subgroup is made up of *D. borealis* and *D. flavomontana*. *Drosophila lacicola* and *D. montana* comprise the third subgroup. This phylogeny is mirrored somewhat by the results of this study, with two exceptions (Table 1). In the first subgroup, species A does not fit in with the other three species. None of the other three species were significantly different from each other, but species A is quite dissimilar. There is no apparent reason why this difference was so great. The results from the second subgroup (*D. borealis* and *D. flavomontana*) did not follow directly from the chromosomal studies. *Drosophila flavomontana* had a significantly ($P \leq 0.05$) higher longevity than *D. borealis* (Table 1). One possible reason for this difference may be the ecology of these species. Throckmorton (1982) reported that even though there are areas of sympatry, *D. flavomontana* is most different ecologically from the group containing *D. borealis, D. flavomontana, D. lacicola,* and *D. montana*. The species of the third subgroup yielded the expected

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TABLE 1

Adult longevity of the D. montana *phylad. Values are given as* $\bar{x} \pm SE$.

Species	Stock no.	Strain	Sex	*N*	Longevity (days)	Range
D. ezoana	0971	Japan	♂	22	37.4 ± 4.1 BC*	2-63
			♀	40	41.4 ± 1.9	3-64
D. ezoana	0971.2	Finland	♂	32	40.2 ± 2.1 BC	8-67
			♀	29	43.5 ± 1.8	11-69
D. littoralis	1001.6	U.S.S.R.	♂	30	43.4 ± 2.7 ABC	17-66
			♀	32	41.3 ± 2.3	16-69
D. littoralis	1001.7	Finland	♂	33	36.2 ± 3.1 BC	12-59
			♀	36	40.1 ± 1.4	9-62
Species A	1091	British Columbia	♂	25	31.7 ± 2.9 E	7-53
			♀	27	23.8 ± 2.8	7-55
D. kanekoi	1061	Japan	♂	18	45.3 ± 3.5 AB	9-58
			♀	26	48.5 ± 2.7	12-76
D. borealis	0961	Minnesota	♂	32	23.1 ± 2.5 DE**	3-38
			♀	33	38.8 ± 2.6	4-61
D. borealis	0961.6	Manitoba	♂	27	31.3 ± 3.2 DE	6-57
			♀	31	34.1 ± 2.9	5-56
D. flavomontana	0981.1	Craig Co., Colorado	♂	30	52.3 ± 2.7 A	28-78
			♀	23	45.9 ± 2.3	11-64
D. flavomontana	0981.0	Chester, Idaho	♂	38	39.4 ± 4.2 AB	10-58
			♀	35	48.1 ± 3.2	18-62
D. lacicola	0991	Saranac, New York	♂	34	30.9 ± 1.8 CDE**	17-57
			♀	28	42.8 ± 2.8	2-68
D. lacicola	0991.14	Brule, Wisconsin	♂	29	32.6 ± 3.0 DE	5-54
			♀	27	36.7 ± 2.4	5-62
D. montana montana	1021.14	Cottonwood, Utah	♂	29	30.2 ± 2.9 DE	1-52
			♀	20	32.3 ± 3.9	3-59
D. montana montana	1021.22	Anchorage, Alaska	♂	19	26.4 ± 2.1 CD**	11-39
			♀	34	43.6 ± 2.4	20-62
D. montana ovivororum	1071	Karesuando, Sweden	♂	28	40.8 ± 2.7 ABC	20-60
			♀	21	44.6 ± 3.7	1-65

*Species with the same letter are not significantly different at $P \leq 0.05$.
**Significant difference between sexes at $P \leq 0.05$.

results, if longevity is based on genetic similarity. *Drosophila lacicola* and *D. montana* were not significantly different from each other (Table 1).

No significant ($P \leq 0.05$) strain differences were observed in this study. This result was quite surprising since some intraspecific variation has been found in chromosomal studies (E. J. Durbin, unpubl. data). Survival curves for these species are typical. The survival of males of this phylad is shown in Figure 1. For most species there was a gradual decline in percent survival starting around days 10 to 20. Two species, *D. borealis* and *D. montana* Anchorage had no survivors past day 40, whereas no species had any survivors past day 80.

For females the survival curves were similar (Fig. 2). However, the curve for females was shifted to the right, indicating the increased longevity of females. Unlike the males, all of the species had females that survived past day 50. Since all females were dead by day 80, there was a large increase in mortality between days 50 and 80. Only one female (*D. kanekoi*) survived beyond 70 days.

The differences in longevity between male and female *Drosophila* have never been determined. Various authors have reported that females live longer; others have noted that the males are longer lived (Gonzales 1923, Maynard Smith 1959). In our previous study we found mixed results in the *D. virilis* species phylad (Durbin and Yoon 1986). In general, females outlived males by one to two weeks, but in some the results were reversed. For the *D. montana* phylad, the results again were mixed (Table 1). In over 60% of the species, females lived longer than

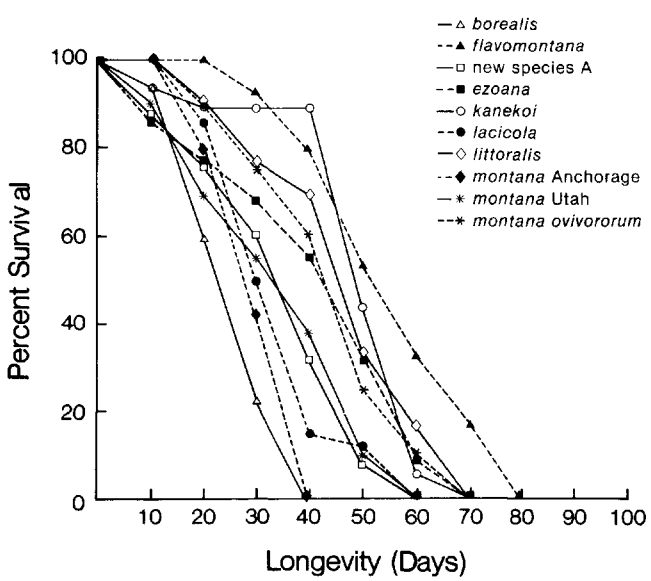

FIGURE 1. Longevity of males of the *Drosophila montana* phylad.

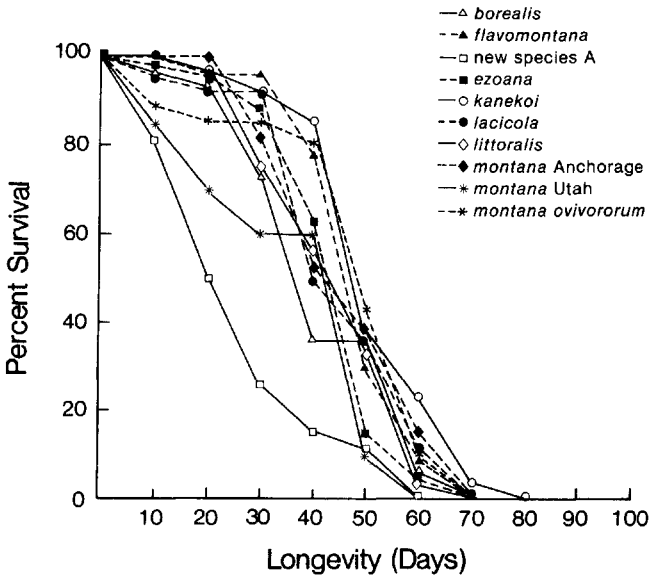

FIGURE 2. Longevity of females of the *Drosophila montana* phylad.

males, but in only three of these species was this difference significant ($P \leq 0.05$, Table 1). In only one species (species A) did males live significantly ($P \leq 0.05$) longer than females. Species A females and *D. borealis* males had over one-third mortality before day 20. Other observations (E. J. Durbin and J. S. Yoon, unpubl. data) indicate that these two cases of early deaths were not due to the etherization. The problem of etherization in the *D. virilis* group was discussed by Durbin and Yoon (1986).

Attempts have been made to attribute differences in adult longevity in *Drosophila* to genetic differences, or ecological differences, or both. The relationship between phylogeny and longevity has not been established in *Drosophila*. We looked at the differences in the latitudes from where these species were first collected. There were more dissimilarities, when species of nearby latitudes were compared, than when phylogenetic similarities were considered (E. J. Durbin and J. S. Yoon, unpubl. data). However, latitude is not irrelevant since differences in habitat occur even at identical latitudes and most likely contribute to species longevity. What we suggest is the idea proposed earlier by Schnebel and Grossfield (1983) that the most useful approach for studying longevity in *Drosophila* is one which incorporates analyses of both phylogenetic and ecological diversity.

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