Dasmohapatra, D.P., N.K. Tripathy and C.C. Das. Berhampur University, Orissa, India. Distribution of different species of Drosophila in Khallikote Ghats, Ganjam District, Orissa, India.

The genus Drosophila has a wide range of distribution covering entire India. The available data on field collection cover most parts of the country, but there still remain large regions lacking dependable data on the Drosophila fauna. In this short communication we wish to report the Drosophila fauna from the Khallikote Ghats, Orissa, India, which are about 60 km to the north-east of Berhampur at 19°15' and 19°5' N latitude and 84°20' and 85°15' E longitude. This mountain range has woody plants at its foot while teak plantation and thick bushy vegetation occur in its upper ranges. The table gives the different species of Drosophila collected on banana bait during several collection trips conducted between the months of January and March, 1980. The average temperature during this period was 27°C. A total of 745 flies were collected which included eight different species belonging to two subgenera.

Species	No. of flies collected			Percentage
	Male	Female	Total	
Subgenus: Sophophora				
D. malerkotliana	245	189	434	58.25
D. kikkawai	78	50	128	15.83
D. takahashii	23	55	78	10.45
D. rajasekari	8	31	39	5.23
D. bipectinata	8	27	35	4.69
D. melanogaster	4	14	18	2.41
D. suzukii	1	1	2	0.26
Subgenus: Scaptodrosophila				
D. nigra	3	8	11	1.46

The dominant species in the collection belonged to melanogaster species group (especially D. malerkotliana and D. kikkawai) with males outnumbering the females; the sex ratio, however, was reversed in the case of D. takahashii, D. rajasekari, D. bipectinata and D. melanogaster.

Gilbert, D.G. Indiana University, Bloomington, Indiana. Effects of CO_2 vs. ether on two mating behavior components of D. melanogaster.

Various effects of two anesthetics, carbon dioxide and ethyl ether, on Drosophila have been reviewed by Ashburner and Thompson (1978). These authors indicate that carbon dioxide treatment can markedly reduce survival and fertility of adults if administered up to 3 hours post-eclosion, but shows no toxic effect if used 5 or more hours after eclosion. Light ether treatment does not produce similar toxic effects. Bingo (1971) found ether to have slighter effects on behavior of D. grimshawi than cold or carbon dioxide when flies were tested a few hours after anesthetization. To determine whether the type of anesthesia used in virgin collection had any long-term effects on reproductive behavior in D. melanogaster, virgin males and females were collected with carbon dioxide or ether and were paired 3 days later in a 2 x 2 factorial experiment. Latency to mounting and copula durations were measured.

Table 1. Analysis of variance in mating behavior components due to female and male anesthetic treatment 3 days previously.

	Mounting latency			Copula duration		
Term	Df	Ms	F	Df	Ms	F
Female treatment	1	0.6022	4.65*	1	0.00883	1.99
Male treatment	1	0.3554	2.74	1	0.00222	0.50
Interaction	1	0.0906	0.70	1	0.00047	0.11
Error	56	0.1294		49	0.00444	

* $p < 0.05$

The D. melanogaster stock tested was a strain homozygous for esterase 6 Slow derived from flies trapped in Bloomington, Indiana, and free of extreme CO_2 sensitivity associated with viruses. The stock was maintained in half-pint bottles of well yeasted cornmeal-molasses-agar media at 25±1°C, 60±10% humidity, on a 12:12 hour light/dark cycle. Eight hours after clearing the stock bottles of adults, newly eclosed flies were sexed and separated by first shaking flies into a transfer bottle. They were then either anesthetized on a CO_2 diffusion pad for the duration of sexing, up to 5 minutes, or anesthetized with ether until their surface clinging response was lost, about 30 seconds. Twenty males or females were housed per vial for 64 to 76 hours at 25°C.

All collected flies were alive and appeared vigorous at this time, 3 days after the ether or CO_2 anesthetization. Male and female pairs were aspirated from their holding vials into observation vials containing media seeded with liquid yeast two days previously. A block of pairings consisted of an ether-treated male with an ether-treated female, an ether male with a CO_2 female, a CO_2 male with an ether female, and a CO_2 male with a CO_2 female. Two blocks were started at each observation period by adding all males, then all females within 5 min of each other, or the reverse order. The time of initial pairing, time of male mounting female and time of dismounting were recorded for each pair to the nearest half-minute.

The factorial analysis of variance for latency to mounting and copula duration are presented in Table 1. These two measures were transformed to their common logarithms for analysis to reduce the correlation of group means with variances. Within group variances are homogeneous, as determined by an F_{max} test (F_{max} = 1.45, Df = 4,14 and F_{max} = 2.26, Df = 4,13 for mounting latency and copula duration, respectively). Female anesthetic treatment significantly affected latency to mounting, and had the largest, but nonsignificant effect on copula duration. Male treatment and the interaction of treatments are nonsignificant components of variance.

Table 2. Mean effects of female anesthetic treatment on mating components.

Treatment	Mounting latency	Copula duration
Ether	11.5 min	22.21 min
CO_2	18.2 min	20.93 min

The effect of carbon dioxide treatment on females is to increase latency to mounting and decrease copula duration relative to ether treatment, as indicated in Table 2. These effects suggest that carbon dioxide use in virgin collecting may have a long-term effect on reproductive responses of females.

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References: Ashburner and Thompson 1978, in: The Genetics and Biology of Drosophila, v. 2a (Ashburner and Wright, eds.), pp. 1-109; Ringo 1971, DIS 47: 118.

Gilbert, D.G. Indiana University, Bloomington, Indiana. Sperm counts and initial sperm storage in D. melanogaster.

In the course of investigating reproductive functions of the male anterior ejaculatory duct enzyme esterase 6 (Richmond et al. 1980), I have examined the number of sperm initially stored by D. melanogaster females from ejaculates of males differing in their esterase 6 genotype. This note describes the methods used for counting sperm and the major results for 47 matings of 3 to 5 day virgins of the Oregon-R strain.

The dissection methods reported here are modified from those described by Fowler (1973) in two important respects. Female reproductive tracts are dissected directly in aceto-orcein stain rather than in Ringer's saline, avoiding a saline-stain reaction which destroys the specimen within a week. Specimens dissected in the stain and sealed under coverslips preserve for several months. Secondly, the spermathecae are dissected from the uterus, pared of their surrounding fat which inhibits staining, and squashed under a separate coverslip. With this method, sperm heads in the densely packed mass of spermathecal sperm stain deeply enough to count the preparations accurately.

Materials used in dissections are two fine forceps, two tungsten dissecting needles, a dissecting microscope, slides and coverslips, and nail polish for sealing slides. The orcein stain used is the salivary chromosome "dissecting" solution described by Strickberger (1962). Viewing specimens with phase optics at 1000X, the stained 10 micron long sperm heads of D. melanogaster can be readily counted with a hand held counter.

The uterus, with attached ventral receptacle, dorsal spermathecae and parovaria, along with the lower portion of the common oviduct, are simply dissected from the female. A mated, etherized female is placed in a drop of orcein stain on a slide. Squeezing the abdomen with the left forceps, the extruded ovipositor is grasped with the right forceps and pulled posteriorly until the reproductive tract is out of the abdomen. Any exterior chitin and digestive tract are dissected away. To obtain clear counts of spermathecal sperm, these paired organs are dissected from the uterus by severing the spermathecal ducts. The fat is dissected away, and the spermathecae are transferred to a second drop of orcein stain on the slide. After applying coverslips to both spermathecal and uteral preparations, the spermathecae are squashed with a hard pressure that expels the sperm mass entirely from its opaque capsule. The uterus-receptacle is squashed gently to flatten it for phase optics without disrupting receptacle integrity.