kanekoi (Reis *et al.*, 2008). Our data strongly support the first simple model and indicate that *D. kanekoi* and *D. ezoana* form one linage. Besides, it is clear that the speeds of nucleotide fixations in different species vary greatly. This may be due to the differences in the population size. Small island populations of *D. kanekoi* from Japan evolve much more rapidly than large continental populations of *D. littoralis*.

We found no difference between *kl-2* sequences of different laboratory lines of *D. virilis*. It is not surprising because *D. virilis* is a nearly monomorphic species (Mirol *et al.*, 2008). On the contrary *kl-2* sequences of two lines of *D. littoralis* from the South Caucasian population and from North European population (Andrianov *et al.*, 2010) are different. South population is ancestral in relation to North population. These data support a previously made proposal that South population of *D. littoralis* is a separate species.

We found no difference between *kl-2* sequences of Eastern *D. borealis* and *D. flavomontana*. Fly lines from Western population of *D. borealis* were unavailable to us. *D. borealis* comprises two different species, Eastern and Western. Eastern *D. borealis* is much more closely related to *D. flavomontana* (Morales-Hojas *et al.*, 2011). Our data support the hypothesis of hybrid origin of Eastern *D. borealis.* It may originate from hybridisation of *D. flavomontana* male with *D. borealis* female.

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A preliminary report and the frequency distribution of *Drosophila* species of Rabwah, Pakistan.

Tahir, Durr-e-Samin. Abdus Salam School of Science, Nusrat Jehan College, Rabwah, Pakistan; d.samin2@hotmail.com

To study the frequency distribution of some *Drosophila* species, a study was carried out in Rabwah, Pakistan (31° 32’ 59”N, 74° 20’ 37” E). Survey was made for the first time in this area and revealed seven species, which were already found before in Lahore, Pakistan. The collections were made twice, once during March-April and then during the month of September, 1992. It was observed that two *Drosophila* species, *D. immigrans* and *D. nepalensis*, were found only during the month of April. This observation is similar to the observation of Gupta (1973), who, while studying the fauna of Manipur, India, observed that these two species can be obtained only in the month of February. These collection data also reveal that all of these species are members of *melanogaster* and *immigrans* species groups of subgenera *Sophophora* and *Drosophila*, thus supporting the view of Bock and Wheeler (1973), who proposed that South East Asia is a fertile region for the rapid diversification and speciation of the members of the *immigrans* and the *melanogaster* species groups of *Drosophila*.

Table 1. Frequency distribution of the species.

Species	Subgenus	No. ♂	No. ♀
D. melanogaster	*Sophophora*	262	198
D. jambulina	"	9	7
D. malerkotliana	"	1	-
D. ananassae	"	1	-
D. takahashii	"	72	52
D. nepalensis	"	8	-
D. immigrans	*Drosophila*	39	77

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Intra-chromosomal association between allozyme loci in *Drosophila ananassae.*

Kumar, S., and A.K. Singh. Genetics Laboratory, Department of Zoology, Banaras Hindu University, Varanasi-221 005 India. Email: *aksbhu23@rediffmail.com*.

Non-random association between gene arrangements was referred to as linkage disequilibrium by Lewontin and Kojima (1960) and according to them this happens due to linkage between such arrangements. Chromosomal polymorphism in association with allozymes has been extensively studied in *Drosophila* (Rodriguez-Trelles, 2003; Iriarte *et al.*, 2002; Rodriguez *et al.*, 2000; Kamping and Delden, 1999; Zapata and Alvarez, 1992). Gametic disequilibrium studies have been done between second chromosome polymorphic arrangements and seven linked loci, in seven populations of *D. buzzatii* from Argentina, and a significant and consistent association for *Est-1, Est-2, Aldox* and *Xdh* has been reported (Rodriguez *et al.*, 2001). They explained that restriction of recombination in heterokaryotypes seems to be the best explanation for the linkage disequilibrium between inversion and enzyme loci located inside the rearranged segments.

Extensive work on allozyme-allozyme linkage disequilibrium in natural populations of *D. melanogaster* has been done involving 36 allozyme pairs (Langley *et al.*, 1974). Among all those, only three showed significant deviation from expectation and only one of them (*Odh-Ao*) could be established to show non-random association. In another study, Langley *et al.* (1977) found no linkage disequilibrium among even tightly linked loci which were nearly 3 cM distant from each other in natural populations of *D. melanogaster*.

D. ananassae, first described by Doleschall (1858), belongs to the *ananassae* species complex of the ananassae subgroup in the melanogaster species group of the subgenus Sophophora and is one of eight cosmopolitan species (Bock and Wheeler, 1972; Tobari, 1993). It occupies a unique status in genus *Drosophila* due to its several genetic peculiarities (Singh, 2010). One of its peculiar features is that of spontaneous crossing over in males, although at much lower frequencies than observed in females (Tobari, 1993; Singh and Singh, 1988). Recently we have started working on the enzyme polymorphism in this species to see genetic diversity in different natural populations derived from various parts of India (Kumar and Singh, 2012; Singh *et al.*, 2013). While studying enzyme polymorphism, we also planned to see association between those enzyme loci which are situated on the same chromosome. In this note we wish to report intra-chromosomal association among three