ported in a variety of tissues including oyster mantle[15], kidney[16], turtle bladder epithelium[17], and villus cavity cells of the chorioallantoic membrane[18]. This laboratory has also found that purified chicken and salmon erythrocyte carbonic anhydrases are stabilized in the face of rising temperature and pH when lipid vesicles are present[19]. The membrane-enzyme association may also be important in orienting the enzyme in an appropriate way for it to function, possibly in the secretion of hydrogen ions.

1 Acknowledgments. This work was supported by NIH grants DE04345 and AM20374. Dr Cao was a visiting scientist from the Department of Biology, East China Normal University, Shanghai, The Peoples Republic of China. We thank Kathryn Majarwitz for assisting with the morphometric analysis. The PTH was provided by the National Institute of Arthritis, Diabetes, Digestive and Kidney Diseases of the National Institutes of Health, US Department of Health and Human Services.

2 Anderson, R.E., Schraer, H., and Gay, C.V., Anat. Rec. *204* (1982) 9.
3 Sternberger, L.A., Hardy, P.H. Jr, Cuculis, J.J., and Meyer, H.G., J. Histochem. Cytochem. *18* (1970) 315.
4 Willingham, M.C., and Yamada, S.S., J. Histochem. Cytochem. *27* (1979) 947.
5 Väänänen, H.K., and Parvinen, E.K., Histochemie *78* (1983) 481.
6 Chambers, T.J., and Magnus, C.T., J. Path. *136* (1982) 27.
7 Bernstein, R.S., and Schraer, R., J. biol. Chem. *247* (1972) 1306.
8 Gay, C.V., Faleski, E.J., Schraer, H., and Schraer, R., J. Histochem. Cytochem. *22* (1974) 819.
9 Warshawsky, H., Goltzman, M.F., Rouleau, M.F., and Bergeron, J.J.M., J. Cell Biol. *85* (1980) 682.
10 Vaes, G., in: Collagenase in Normal and Pathological Connective Tissues, p. 185. Eds D.F. Woolley and J.M. Evanston. Wiley & Sons, New York 1980.
11 Lucht, U., Z. Zellforsch. *145* (1973) 75.
12 Taylor, T.G., and Lewis, P.E., J. Endocr. *52* (1972) xlvii.
13 Stoeckel, M.E., and Porte, A., Z. Zellforsch. mikrosk. Anat. *94* (1969) 495.
14 McWhinnie, D.J., Comp. Biochem. Physiol. *50A* (1975) 169.
15 Wheeler, A.P., Am. Zool. *19* (1979) 995.
16 Sanyal, G., Pessah, N.I., and Maren, T.H., Biochim. biophys. Acta *657* (1981) 128.
17 Schwartz, J.H., Rosen, S., and Steinmetz, P.R., J. clin. Invest. *51* (1972) 2653.
18 Anderson, R.E., Gay, C.V., and Schraer, H., J. Histochem. Cytochem. *29* (1981) 1121.
19 Kim, J.-S., Gay, C.V., and Schraer, R., Comp. Biochem. Physiol. *74B* (1983) 295.

0014-4754/85/111472-03$1.50 + 0.20/0

Mobile gene localization and viability in *Drosophila melanogaster*

C. Biemont, E.S. Belyaeva, E.G. Pasyukova and G. Kogan

Population Biology Laboratory, University Lyon 1, F-69622 Villeurbanne (France), and Institute of Molecular Genetics, USSR Academy of Sciences, Kurchatov Sq. 46, Moscow 123183 (USSR), 8 November 1984

Summary. The location of the mobile element mdg-1 was determined by in situ hybridization in salivary gland chromosomes of *Drosophila melanogaster*. The locations of mdg-1 are nonrandom and some 'hot spots' exist. Moreover, the spectra of mdg-1 locations vary with the viability values of the families from which the larvae originated. This suggests that particular frequency spectra are associated with lethality resulting from inbreeding.
Key words. Mobile elements; inbreeding; in situ hybridization; *Drosophila melanogaster*.

The genome is now known to contain many reiterated transposable elements that may be the cause of genetic instability of structural genes through activation, inactivation or regulation[1-3]. These elements may appear at different chromosomal locations in different strains and in different individuals from the same laboratory stock or the same natural population[4-9]. Only a little information about the amount of polymorphism of such elements in natural populations is available, though such information could shed light on their origin, their effects on the organisms carrying them and their possible role in the adaptation of populations. Indeed, though the molecular structure of some of these elements is now well known, we have little idea whether they do or do not provide benefit to the organisms. However, some transposable elements in bacteria do procure a higher adaptability to their carriers[10,11] and the number and location of mobile dispersed genes (mdg) in *Drosophila melanogaster* may be associated with fitness[5,12]. Here, we study the location of the element mdg-1 in the chromosomes of individuals from a natural population of *Drosophila melanogaster* in connection with fitness after inbreeding.

A laboratory population of *Drosophila melanogaster* was established from flies captured in Azerbaidjan (USSR) recently. The population so formed was maintained in the laboratory by mass culture at 25 °C for three generations before the experiment was started. 58 sib pairs, offspring of 58 pairs of unrelated flies of the stock, were set up and allowed to lay eggs. Egg hatchability and egg-to-adult survival were then determined in their offspring that developed at 25 °C[13]. Other subsequent samples of eggs laid by the same sib pairs were allowed to develop at 18 °C in order to lower the developmental rate and to obtain large larvae with chromosomes suitable for in situ hybridization[4]. The number and location of the mdg-1 element were then analyzed in the salivary gland chromosomes of these inbred larvae. 17 sib progenies were chosen for the cytological study according to their viability values, which ranged from 0.32 to 1 (see the distribution of viability values in figure 1). Low overall viability values are due to either a high embryonic mortality rate or a high larvo-pupal mortality, or to both[13]. We thus distinguished three classes of inbred progenies characterized by 1) low egg hatchability (overall viability values ranging from 0.38 to 0.61; five progenies), 2) low larval viability (with overall viability values ranging from 0.32 to 0.72; seven progenies), 3) high egg-to-adult survival rate (viabilities higher than 0.90; five progenies). We have also

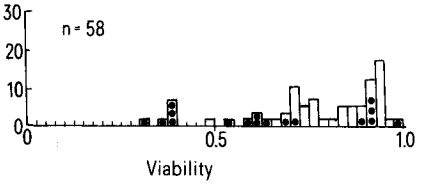

Figure 1. Distribution of the proportion of the fertilized eggs developing to the adult stage in F2 inbred progenies from sib pairs. ●indicates the families from which the larvae were taken. N: number of pairs.

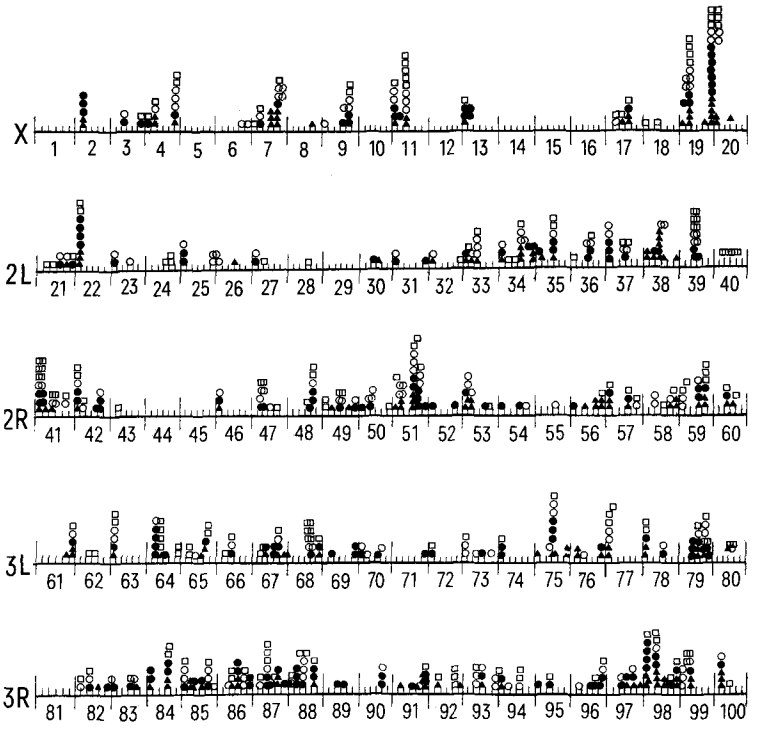

Figure 2. Location and number of mdg-1 insertions (frequency spectra) found in the 23 genomes. The region numbers correspond to Bridges' subdivision of the chromosomes. Inbred families with ▲ low egg hatchability, ● low overall egg-to-adult survival, ○ high overall egg-to-adult survival; □ initial natural population. Polytene chromosomes were prepared for 3HDNA hybridization following the procedure of Ananiev et al.[4].

analyzed mdg-1 in six larvae whose parents were taken from the initial Azerbaidjan stock and crossed randomly. The egg-to-adult survival of these larvae was higher than 0.90. As a total 23 genomes were scored.

Figure 2 shows the distribution pattern of mdg-1 along the chromosomes for all the larvae analyzed. In total 623 insertions were detected on 250 cytologically distinguishable sites. The overall distribution is non-uniform. There are clearly some regions in the chromosomes in which more insertions were found than in others. This confirms and extents other results from X chromosomes[7] and laboratory populations[4,14]. Many of the sites of insertion (19B, 19F, 22A, 35CD, 39DE, 42A, 98C) correspond to regions of intercalary heterochromatin[5,12,14], whose principal features (e.g. weak points, late replication, ectopic pairing) are supposed to be associated with moderate tandem repeats[15] or mobile genetic elements[16]. Mobile elements may insert only at specific 'accessible' parts of the DNA sequences of the chromosomes.

To test the distribution of mdg-1 along the chromosomes for randomness we assumed a truncated Poisson distribution[17] following the formula

$$P(X=K) = e^{-\lambda}\frac{\lambda^k}{k!}\frac{1}{1-e^{-\lambda}}.$$

The parameter λ was estimated as the limit value of the recurrence formula $\lambda_{n+1} = \bar{x}(1-e^{-\lambda n})$. λ is the mean of the usual Poisson distribution and $\bar{x}$ is the mean of the truncated Poisson curve. A theoretical distribution was then obtained. Figure 3 shows that the observed curve fits the truncated Poisson distribution ($\chi^2 = 8.05$; d.f. = 5). From this the number of sites with no insertion is then estimated to be 28 and the total number of sites where an insertion of mdg-1 may be expected should be

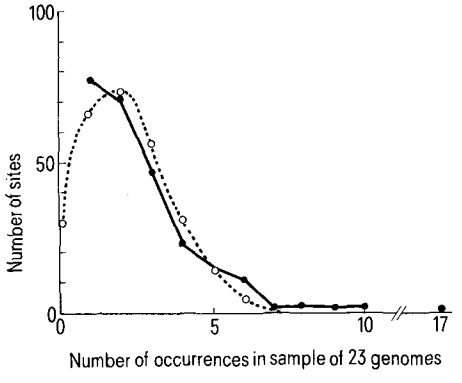

Figure 3. Distribution of the mdg-1 element in 23 genomes (all chromosomes confounded). ●—● observed frequencies, ○—○ theoretical frequencies of the Poisson distribution.

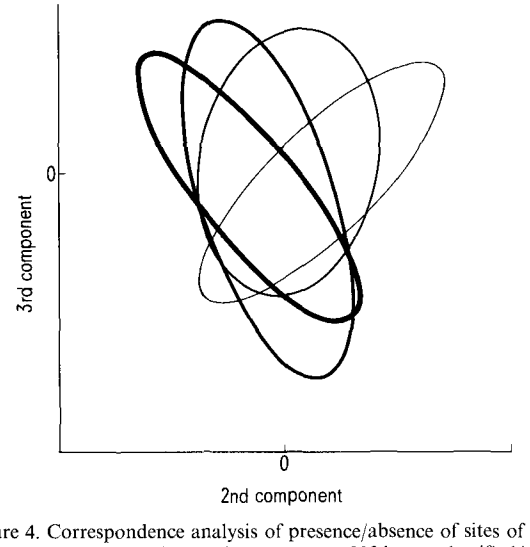

Figure 4. Correspondence analysis of presence/absence of sites of insertion of mdg-1 on the polytene chromosomes of 23 larvae classified in four classes of viability (see text). Only the 5% confidence ellipses are represented. —— low egg hatchability families, —— low egg-to-adult survival, —— high viability, —— initial natural population.

around 280. However, some bands seem at first sight to be 'hot spots' of mdg-1 insertion (see fig. 2 and the tail of the distribution in fig. 3). This is particularly striking for the bands 19F, 39CD, 42A, 48E, 51D, 98C. Some of them were also shown in other experiments to be sites of insertions or hot spots for transposition[5,12,14]. The number of these hot spots, however, is small and thus the distribution in figure 3 does not deviate significantly from a Poisson distribution. The hot spots may be clusters of mobile elements[15,18,19] and their location may characterize a population. This hypothesis is reinforced by the observation that among the sites that hybridized most frequently in our experiment with a natural population are some that also hybridized most frequently in previous experiments with laboratory populations[14].

To test whether the different distribution spectra of insertions are correlated with different classes of viabilities, we analyzed the rough data by a factorial correspondence analysis[20]. Naturally, we had only a contingency table with the values 0 and 1 (absence or presence of an insertion site in a bands), but factorial correspondence analysis is the best and most powerful technique that can be used to analyze such a table. The aim of this method is to represent the data in a new space with a reduced number of dimensions. Figure 4 represents the projection of the values on the plan of the second and third components of the analysis. We choose these two plans as they lead to a better discrimination of the different families; the larvae from the natural population are dispersed on the second and third axes of the analysis but not on the first one. As seen from figure 4, the different classes of viability progenies have distinct frequency spectra of mdg-1 location. For example the correspondence analysis reveals that a pattern with insertions in bands 7D, 38C, 51B, 56E, 67C, 75F, 76B, 85B, is seen only in families from the low egg hatchability class (that is also the class with the lower values of egg-to-adult survival).

It is well known that differences in the location of mobile elements occur among individuals from the same population. Our data indicate in addition that the pattern of distribution of mdg-1 may correlate with the viability value of the family from which the larva comes. However, in situ hybridization was done on chromosomes of inbred larvae that survived. These larvae, as seen from figure 4, may have a particular pattern of mdg-1 locations. It is possible that the embryos and larvae that dies had another particular distribution spectrum of mdg-1 locations connected with lethality. This hypothesis is reinforced by recent results showing that a specific mobile element, the L factor, may be involved in the lethality associated with an unstable X chromosome[21].

Many experiments will be necessary to be able to assess the role of mobile elements in natural populations. Our results indicate that the search for relationships between location of elements and viability or other characteristics associated with fitness should be a promising way of investigation.

Acknowledgments. We thank V. Gvozdev for useful discussion, M. Ashburner for critically reading an earlier draft of the manuscript, L. Konovalchouc for valuable technical assistance, P. Fouillet, E. Wajnberg and D. Pontier for help with statistics and computerization. This research was supported by the USSR Academy of Sciences and the Centre National de la Recherche Scientifique (Laboratory No. 243).

1 Spradling, A.C., and Rubin, G.M., A. Rev. Genet. *15* (1981) 219.
2 Calos, M.P., and Miller, J.H., Cell *20* (1980) 579.
3 Davidson, E.H., and Britten, R.J., Science *204* (1979) 1052.
4 Ananiev, E.V., Gvozdev, V.A., Ilyin, Y.U., Tchurikov, N.A., and Georgiev, G.P., Chromosoma *70* (1978) 1.
5 Gvozdev, V.A., Belyaeva, E.S., Ilyin, Y.U., Amosova, I.S., and Kaidanov, L.Z., Cold Spring Harb. Symp. quant. Biol. *45* (1981) 673.
6 Ising, G., and Block, K., Cold Spring Harb. Symp. quant. Biol. *45* (1981) 527.
7 Montgomery, E.A., and Langley, C.H., Genetics *104* (1983) 473.
8 Potter, S.S., Brorein, W.J., Dunsmuir, P., and Rubin, G.M., Cell *17* (1979) 415.
9 Young, M.W., and Schwartz, H.E., Cold Spring Harbor Symp. quant. Biol. *45* (1981) 635.
10 Chao, L., Vargas, C., Spear, B.B., and Cox, E.C., Nature *303* (1983) 633.
11 Biel, S.W., and Hartl, D.L., Genetics *103* (1983) 581.
12 Belyaeva, E.S., Pasyukova, E.G., Gvozdev, V.A., Ilyin, Y.O., and Kaidanov, L.Z., Molec. Gen. Genet. *185* (1982) 324.
13 Biemont, C., Mech. Ageing Dev. *8* (1978) 21.
14 Belyaeva, E.S., Ananiev, E.V., and Gvozdev, V.A., Chromosoma *90* (1984) 16.
15 Zhimulev, I.F., Semeshin, V.F., Kulichkov, V.A., and Belyaeva, E.S., Chromosoma *87* (1982) 197.
16 Gvozdev, V.A., Proc. XIV int. Congr. Genet., vol. 3, p. 257. MIR Publ., Moscow 1981.
17 Johnson, N.I., and Kotz, S., Discrete distributions. Houghton Mifflin Company, Boston 1969.
18 Wensink, P.C., Tabata, S., and Pachl, C., Cell *18* (1979) 1231.
19 Balakiveva, M.D., Belyaeva, E.S., Kogan, G.L., Pasyukova, E.G., Alatortsev, E.V., and Gvozdev, V.A., 16th FEBS meeting (Abstr.) Moscow.
20 Benzecri, J.P., L'analyse des données. II. L'analyse des correspondances. Dunot, Paris 1976.
21 Lim, J.K., Simmons, M.J., Raymond, J.D., Cox, N.M., Doll, R.F., and Culbert, T.P., Proc. natn. Acad. Sci. USA *80* (1983) 6624.

0014-4754/85/111475-03$1.50 + 0.20/0

Occurrence of ricinoleic acid in submerged cultures of various *Claviceps* sp.

V. Křen, T. Řezanka and Z. Řeháček[1]

Institute of Microbiology, Czechoslovak Academy of Sciences, Videňská 1083, 142 20 Prague 4 (Czechoslovakia), 2 November 1984

Summary. Ricinoleic acid was found in different *Claviceps* sp., indicating that it is neither specific chemotaxonomic marker of *Claviceps purpurea,* nor a specific indicator of peptide alkaloid synthesis.
Key words. Claviceps; ricinoleic acid; ergot alkaloids; fatty acids.

Mycelial differentiation of the fungus *Claviceps* involves morphological and biochemical changes. In parasitic cultures a conspicuous feature of differentiation is the formation of sclerotium and alkaloid synthesis. An analogous pattern is found also in saprophytic *Claviceps* strains. In these strains the production of alkaloids under conditions of submerged cultivation is accompanied by sporulation[2] and differentiation of the culture into the sclerotial type of mycelium[3]. Sclerotial cells are assumed to be the dominant producers of alkaloids[3,4]. Another indicator of differentiation of the culture into the sclerotial type is abundant production of lipids (triglycerides)[4–6].

In sclerotial cells lipids are found in substantially higher