

Gerasimova, T.I.¹ and Yu.V. Ilyin.^{2 1} Institute of Molecular Genetics, ²Institute of Molecular Biology, USSR Academy of Sciences. Transpositions of various mobile elements and their relation of unstable mutagenesis in *Drosophila melanogaster*.

It has been shown earlier that the instability of ct^{MR2} is due to the integration of the mobile element $mdg4$ in the 7B region, i.e., the region of the cut locus (Gerasimova 1981). The formation of stable ct^+ revertants in a homozygous ct^{MR2} stock was often accompanied by new unstable mutations in other X-chromosome loci: y , w , cm , sn , g , m , with a frequency of $1-8 \times 10^{-4}$. While $mdg4$ left the region of

the cut locus, it was not detected elsewhere in the X-chromosome. Hence the emergence of new unstable mutations involves the integration of other mobile elements in the loci concerned. This was tested for the P-element. To that end, the polytene chromosomes of ct^{MR2} larvae and stable revertants carrying new mutations were subjected to in situ hybridization with the 3H -DNA of plasmids containing $mdg4$ and the P-element obtained from D. Rubin (Rubin et al. 1982). The standard hybridization procedure was used (Ilyin et al. 1978). The results are listed in Table 1. The X-chromosome of the ct^{MR2} mutant has a hybridization site for the P-element only in 17C. Yet, the 17C region does not contain the P-element itself, only the flanking sequences cloned together with the P-element (Rubin et al. 1982). The formation of stable revertants involves the removal of $mdg4$ from the 7B region and the appearance of new copies of the P-element in the X-chromosome. Some new mutations are associated with the integration of the P-element in the loci concerned. For instance, the revertants carrying mutations in the sn locus, $ct^{+sn^{MR2}}$ and $ct^{+sn^{110}}$, were characterized by the appearance of the P-element in 2-4 new sites, including the 7D region which corresponds to the sn locus (Fig. 1). For a revertant with a w^1 mutation, the P-element appeared in the 3C region, again corresponding to the locus concerned. Thus, the excision of $mdg4$ from the 7B region in the case of stable ct^+ revertants is often accompanied by a mobilization of the P-element. Its integration in the regions of the w and sn loci is the cause of these unstable mutations. However, in the case of other unstable mutations: y , cm , g , m , the P-element was not detected in the relevant regions. The emergence of these mutations is probably associated with the transposition of other mobile elements that are activated at the same time as $mdg4$ and the P-element.

Table 1. Results of in situ hybridization of 3H -DNA of the P-element and $mdg4$ with X-chromosomes of the ct^{MR2} mutant and its derivatives.

Mutations	Hybridization sites	
	for the P-element	for $mdg4$
ct^{MR2}	17C	17B*
$ct^{+sn^{MR110}}$	3A; 7D; 17C	-
$ct^{+sn^{+}}$	4F, 6F, 9B, 17C	-
$ct^{+sn^{MR2+}}$	2F, 7D, 9CD, 10F, 17C	-
$ct^{w^{MR1}}$	1B, 1DC, 3C, 5F, 17C	-
$cm^{MR1} ct^{MRpN1}$	17C	7B
$cm^{+} ct^{+} sn^{17}$	3A, 7D, 9B, 13D, 17C	

* = 3C, 7B, 7D = sites of localization w , ct and sn loci.



Fig. 1. Results of in situ hybridization with polytene chromosome $cm^{+}ct^{+}sn^{17}$ mutant. 7D = the site of the localization of sn locus.

Earlier a stock was described (a derivative of ct^{MR2}) carrying two unstable alleles, $cm^{MR1} ct^{MRpN1}$, and characterized by a 70% simultaneous reversion of both alleles at the carmine and cut loci (Gerasimova 1983). Almost half of the double $cm^{+}ct^{+}$ revertants carried sn mutations. This was accompanied by the excision of $mdg4$ from the 7B region and the integration of the P-element in 7D (the region of the sn locus). The nature of the mobile element in the cm locus (GE) is unknown, but it is neither $mdg4$ nor the P-element.

Thus at least three mobile elements are simultaneously mobilized in this case. Different mobile elements seem to have different locus specificities: *mdg4* prefers the cut locus, the P-element prefers *w* and *sn*.

The above results show that the activation of mobile elements conforms to the "all-or-none" principle. Various mobile elements are mobilized with a frequency of 10^{-3} - 10^{-4} , leading to the reversion of some mutations and to mutagenesis in other genes. This is often accompanied by the appearance of new copies of the P-element in the X-chromosome.

What triggers off these transposition processes? Hardly the P-element, for it is present in all cells, while transposition occurs in one out of 1000-10000 cells. The processes may be genome-controlled, as suggested by the *cm^{MR1} ct^{MRpN1}* stock, where the transposition events involving a number of mobile elements are enhanced by an order of magnitude as compared with the *ct^{MR2}* stock. The activation of mobile genetic elements is certainly important in evolution, since it is capable of causing spontaneous changes in the genome and ensuring its rapid rearrangement.

The authors would like to thank Prof. N.F.Maysoedov for his interest and support, Prof. G.P.Georgiev for a discussion of the results, Mrs. N.V.Knizhnikova for technical assistance.

References: Gerasimova, T.I. 1982, *Molec.Gen.Gent.* 184:544; Gerasimova, T.I. 1983, *DIS* 59:37-38; Ilyin, Y.V., N.A.Tchurikov, E.V.Ananiev, A.P.Ryskov, G.N.Yenikolopov, S.A.Limborska, N.E.Maleeva, V.A.Gvozdev & G.P.Georgiev 1978, *Cold Spring Harbor Symp.Quant.Biol.* 42:959; Rubin, G.M., M.G.Kidwell & P.M.Bingham 1982, *Cell* 29:987.

Ghosh, M. University of Calcutta, India.
Nucleolar chromatin thread in different species of *Drosophila*.

It has been reported earlier (Ghosh & Mukherjee 1982) that the nucleolus of *Drosophila* salivary glands exhibit a variation in morphological conformations of nucleolar chromatin thread (NCT) in different cytological preparations.

That such intranucleolar structures are DNA materials have also been confirmed by Feulgen staining, Acridine Orange and Hoechst 33258 fluorescence. Furthermore, these NCT structures have been classified into four principal types (Ghosh & Mukherjee 1982).

Results on the treatment of nucleolar chromatin thread with various chemical agents viz. NaOH, HCl, DNase, 2,4 Dinitrophenol, heat treatment followed by acridine orange (AO) fluorescence revealed that both chromosome and NCT exhibit identical AO fluorescence. These findings suggest that the general organization of both chromosome and nucleolar chromatin is similar (Ghosh & Mukherjee 1982).

In the present investigation it has been revealed that in all the species (altogether 14 species reported in this issue) of *Drosophila* studied, 4 main types of NCT are manifested as has been reported earlier (Ghosh & Mukherjee 1982). In addition, some interesting features of the NCT have also been noted. These features are as follows.

In some preparations NCT appears as puff. These puffs resemble the puffs of chromosome (Fig. 1). Sometimes specific chromosome ends as a large puff, i.e., nucleolus with NCT (Fig. 2). The nucleolus itself also appears as a puff from the terminal portion of a chromosome (Fig. 3), showing the nucleolus with Type 2 NCT. It is also interesting enough to note that in *Drosophila melanogaster* the X-chromosome of some nuclei ends at its terminal (proximal) portion as a large puff within which a clear doublet is present. The terminal large puff appears as a nucleolus with NCT (doublet band). The doublet of the NCT is similar to that with some doublets of chromosome (Fig. 4). The banding pattern, i.e., bands, interbands are also observed in the NCT (Fig. 5).

The NCT sometimes appears as a small puff, i.e., chromosomal small band ends in a small puff in the nucleolus (Fig. 6). The NCT of *Drosophila* sometimes appears as banded structures i.e., NCT with regular bands and interbands as they are found in the chromosomes. Such regions can be clearly distinguished as dark and light bands and interbands of NCT. In many nuclei of different species of *Drosophila* a clear connection has been marked between the chromosomal band(s) and the nucleolus, i.e., NCT is continuous with the chromosomal band(s) (Figs. 7 & 8).