

Semeshin, V.F., I.F.Zhimulev, A.G.Shilov, E.S.Belyaeva & E.M.Baricheva. Institute of Cytology and Genetics, Novosibirsk, USSR. RNP-bodies in puffs of *Drosophila melanogaster*.

Round electron-dense bodies were observed in the 71CE puff under the electron microscope (EM). The occurrence frequency of these bodies is 0.5-1% in 0-1 h. prepupae under normal conditions but more frequently in 71CE of prepupae transferred from 14°C to 25°C. Their number in the

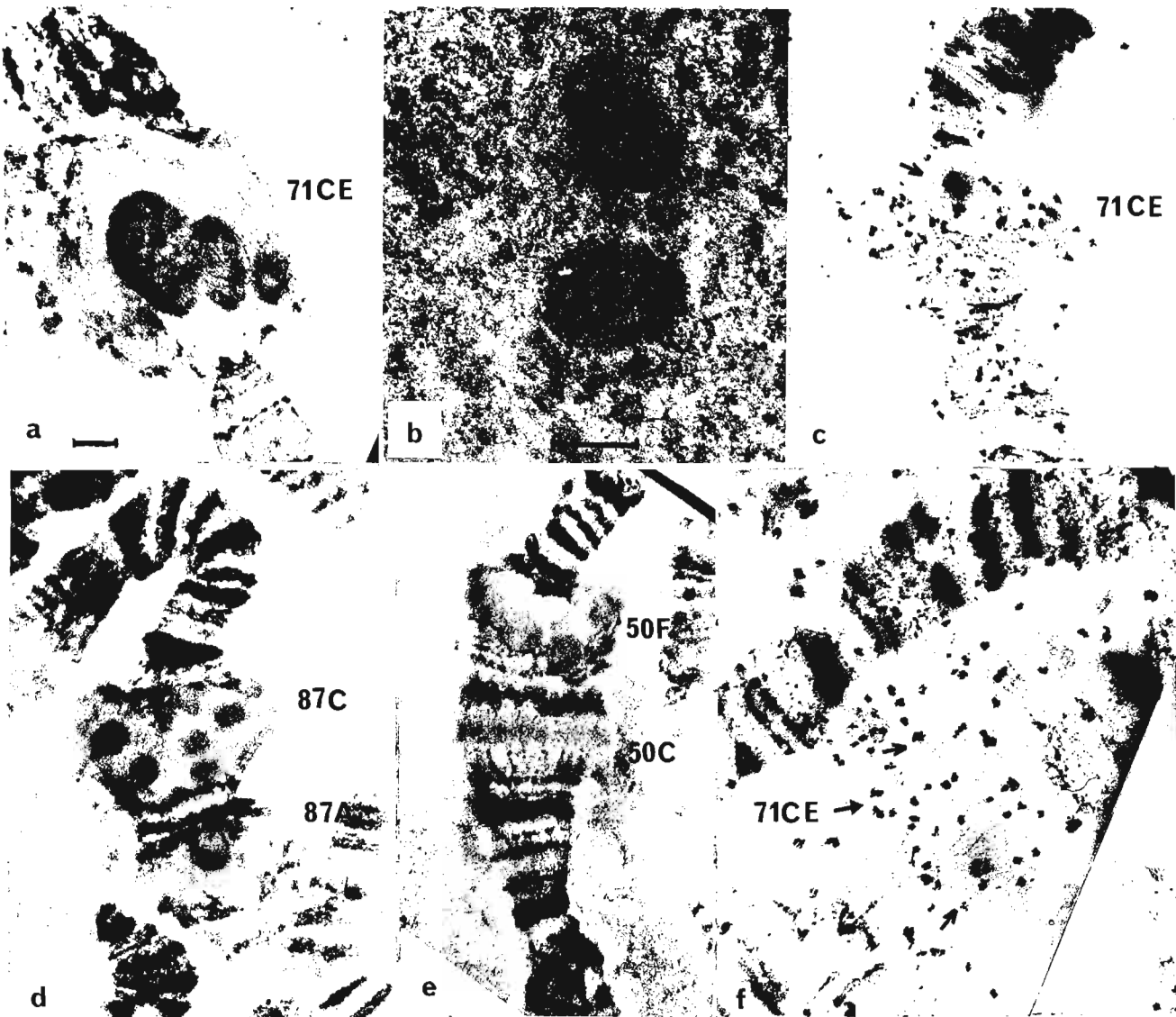


Figure 1 a-f. RNP-bodies in puffs of 0-1 hr prepupae *Drosophila melanogaster* polytene chromosomes. (a) The 71CE puff, salivary glands were fixed in ethanol : acetic acid (3:1) mixture and squashed in 45% acetic acid. Preparing of squashed chromosomes for EM is described in more detail in Semeshin et al. 1979. (b) RNP-bodies after double fixation with glutaraldehyde and osmium tetroxide. The method used was that described by Yamamoto (1970). (c) and (f) EM autoradiography of well developed 71CE puff. Salivary glands were incubated with  $^3\text{H}$ -uridine during 15 min 3 hr after the temperature shift from 14°C to 25°C (1000  $\mu\text{Ci/ml}$ , spec. activity 24 Ci/mM). (d) 87A and 87C heat shock puffs and (e) 50C and 50F puffs. The conditions for the induction of RNP-bodies were as follows: Larvae were kept 24 hr at 14°C, subsequently shocked during 30 min at 37°C and transferred to 25°C. Salivary glands were fixed 30 min after the heat shock period and prepared for EM as (a). Bars in (a) and (b) are equal to 1  $\mu$ . Magnification in (c-f) is the same as in (a). Arrows in (f) indicate the RNP-bodies.

puff varies from 1 to 8 and their size 1-5 $\mu$  in diameter (Fig. 1a). In salivary gland chromosomes fixed in glutaraldehyde, these bodies are spherical and they consist of densely packed fibrillar material surrounded by granules about 200 Å in diameter (Fig. 1b). Morphologically these bodies are similar to the nucleolus. Silver grains after  $^3\text{H}$ -uridine incorporation are homogeneously distributed over the bodies and other parts of the 71CE puff (Fig. 1c,f).

The RNP-bodies appear in the 71CE puff with a frequency about 10% 3 hr after a temperature shift from 14°C to 25°C. They are particularly abundant 5 hr after the shift (about 70%). A 30 min heat shock (25°C→37°C→25°C) also induces the appearance of the RNP-bodies in the 71CE puff, but in lesser extent (about 105%, 1 hr after cessation of the heat shock and about 25% after 25 hr).

The following experiment was performed to stimulate the formation of the RNP-bodies. Shortly before puparium formation larvae were collected at 14°C and transferred promptly to 37°C. Only individuals which formed puparium 30 min at 37°C were taken. The 0 hr prepupae were transferred to 25°C, their salivary glands were dissected and fixed after 15, 30, 60 and 120 min. The 71CE puff appears quite small after the heat shock but during subsequent 30 min its size increases and 2 hr later it becomes as large as in a normal 2 hr prepupa. The sizes of the 87A and 87C heat shock puffs concomitantly decrease and these puffs were also found to contain RNP bodies (Fig. 1d). Other puffs containing RNP-bodies were 38B, 50F, 66B, 78D, 82F. They all well developed in 0-1 hr prepupae (Ashburner 1972), but the RNP-bodies do not make their appearance in all the puffs (the 50C puff in Fig. 1e, for example). Figure 2 shows the variations in the frequency of RNP-bodies in the 71CE puff after heat shock recovery and in the 87A and 87C puffs as they regress.

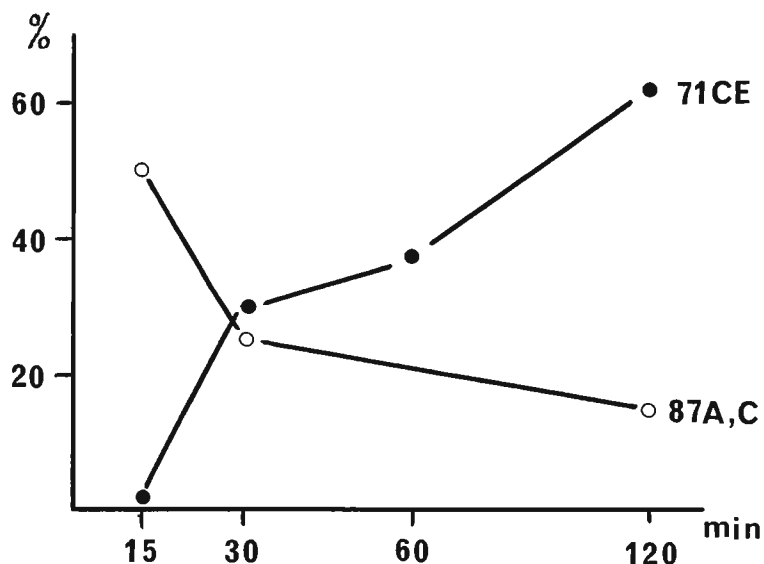


Figure 2. Kinetic of RNP-bodies in the 71CE puff (closed circles) and in the 87A and 87C puffs (open circles). Ordinate: Frequency in %. Abscissa: Heat shock, min of recovery at 25°C after 30 min at 37°C.

The RNP-bodies observed are very similar to the "RNP-droplets" in puffs of salivary gland chromosomes of Chironomidae (see for references Stockert & Diez 1979). These "RNP-droplets" can be induced by cold and heat shock and also cycloheximide (an inhibitor of protein synthesis). The "RNP-droplets" are inducible by dimethylsulfoxide under a normal temperature condition (18°C) with a frequency of 70-90% (Sass 1981). According to Dr. Sass these "RNP-droplets" are formed as a result of a delay in the packaging of transcripts during the stimulation of the puff.

It may be regarded as if RNP-bodies could be an additional characteristic of the cell response to the heat shock. Their appearance is presumably connected with impairment of transcriptional processes and transport newly synthesized product from the nucleus.

References: Ashburner, M. 1972 in Results and Problems in Cell Differentiation 4:101-151;

Sass, H. 1981, Chromosoma 83:619-643; Semeshin, V.F., I.F. Zhimulev & E.S. Belyaeva 1979, Chromosoma 73:163-177; Stockert, J.C. & J.L. Diez 1979, Chromosoma 74:83-103; Yamamoto, H. 1970, Chromosoma 32:171-190.