

E₉ 58D/64D-68B/68C, E₁₂ 61C/61D-67A/67B. All individuals were dissected in Ringer *Drosophila* solution (pH 7.2). Salivary glands were fixed in ethyl alcohol: acetic acid (3:1), and were stained in lacto-aceto orcein (acetic orcein 80%, lactic acid 20%). All experiments, cultures and cytological preparations were carried out in a thermoregulated room at 19±1°C.

Several puffs show a similar pattern of activity in E_{st} and E₁₊₂₊₉₊₁₂ arrangements, for instance 68DE, 69B, 70A and 70BC. Other puffs show quantitative differences or differences in the timing of activity. Finally, a few puffs display strong differences. Among this group can be included the large puffs 67B and 61AC. In the E₁₊₂₊₉₊₁₂ arrangement 67B is active in third larvae, regresses by 0h prepupae and after puparium formation increases its activity throughout the prepupal period (Figure 1A). In spite of this, 67B is an occasional puff in the E_{st} chromosome. It only appears actively in the third instar and at the beginning of prepupation and always at low frequency (Figures 1B and 1C).

Another striking difference is the size of 67B in both chromosomes. In E₁₊₂₊₉₊₁₂ arrangement 67B is a large puff as can be seen in Figure 1A. The size of this puff varies little throughout its time of activity. In spite of this, 67B is a small puff in E_{st} chromosome. Its maximum size can be observed on Figure 1C. 61AC shows complementary behaviour (Fig. 2A and 2B). It is a large puff that maintains its activity throughout the prepupal period in E_{st}, and a small occasional puff in E₁₊₂₊₉₊₁₂ chromosome. It is important to emphasize the location of these puffs on the E chromosome. Both puffs are located at the boundaries of E₁₂ inversion. In the E_{st} chromosome (Fig. 1B and 2B), 67B is located between the active locus 66 and the occasional puff 67C/D, and 61AC is located between the occasional loci 60C/D and 61D-62A. In the E₁₊₂₊₉₊₁₂ chromosome 67B is located between 67CD and 61D-62A. It is interesting to note the behaviour of this last site. 61D-62A shows activity both in E_{st} and in E₁₊₂₊₉₊₁₂, whereas in E_{st} it is close to 61AC and is never very large (Fig. 2B), in E₁₊₂₊₉₊₁₂ it is close to 67B and is always very large (Fig. 1A). It is obvious that 67B and 61AC show the greatest differences in puffing activity between E_{st} and E₁₊₂₊₉₊₁₂ arrangements. Both are located at the boundaries of the E₁₂ inversion, and not only do they change their position in the chromosome but also the sites close to both are different in the two arrangements. It is possible that these differences in gene activity at puff level can be due to position effect.

References: Kunze-MUhl, E. & E.Müller 1958, Weiter Untersuchungen über die chromosomale Strukturtypen bei *Drosophila subobscura*, Coll.Z.indukt.Abstamm.-Vererb.Lehrer 87:65-84.

Di Pasquale Paladino, A. and P.Cavolina.
University of Palermo, Italy. Caffeine
effect on tumor manifestation in the
tu-pb stock of *D.melanogaster*.

Table 1. Complete development in medium containing various concentrations of caffeine.

	% ♀♀ tu	N.♀♀	% ♂♂ tu	N.♂♂
Control	32.82	1301	7.22	1149
Caffeine 500 ug/ml	29.13	1253	5.73	1238
Caffeine 1000 ug/ml	17.61*	1221	3.68*	1060
Caffeine 0.01 M	7.23*	166	1.45*	207

* P < 0.05 (compared to control)

We have now investigated the effect of caffeine added to the nutrient medium of developing tu-pb larvae. Statistical analysis of results obtained after complete development of larvae on medium containing various concentrations of caffeine (Table 1) demonstrates that a dose of 500 ug/ml does not exert any influence upon tumor incidence, while concentrations equal to 1000 ug/ml and 0.01 M (corresponding to 1984 ug/ml) exert a significant inhibitory effect upon tumor appearance in adult insects. 0.01M concentration was chosen for experiments involving exposition of larvae to caffeine medium for a limited period of time. Results are shown in Table 2.

Experiments involving egg deposition onto normal medium, followed by transfer onto caffeine medium: tumor incidence is significantly decreased, if compared to control, only in the 48h group. This finding suggests that caffeine is active only during the first stages of development.

Experiments involving egg deposition onto caffeine medium followed by transfer onto normal medium: tumor incidence was found to be lower in groups of larvae transferred from

caffeine to caffeine medium than in those transferred from caffeine to normal medium. The latter groups, however, do not significantly differ from larvae completing their development on normal medium. A significant decrease of tumor incidence can be therefore shown only if caffeine is present in medium since the beginning of development and for the whole larval life. Such decrease should not be ascribed to different survival rates, since survival patterns do not always correspond to tumor incidence patterns.

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Table 2. *($P < 0.05$) (compared to control)

Period of larval life treated		treated larvae	survival rate	% ♀♀ tu	N. ♀♀	% ♂♂ tu	N. ♂♂
Deposition onto normal medium:							
48 h	To normal	621	47.6	18.7	160	4.4	136
	To caffeine	1184	30.8	11.1*	252	0.7	276
72 h	To normal	272	53.6	13.5	74	2.7	72
	To caffeine	666	55.7	15.4	194	1.7	177
96 h	To normal	203	78.8	15.4	71	2.2	89
	To caffeine	401	62.1*	16.7	137	3.5	112
120h	To normal	426	66.9	21.3	145	1.4	140
	To caffeine	377	64.1	23.1	108	0.7	134
Deposition onto caffeine medium:							
24 h	To normal	1249	33.8*	23.3*	231	7.8	192
	To caffeine	1434	24.4	13.6	190	4.3	161
48 h	To normal	947	28.2*	21.5*	130	8.6	138
	To caffeine	1069	22.6	13.3	127	1.7	115
72 h	To normal	628	42.6*	35.9*	139	3.1	129
	To caffeine	716	18.8	9.2	76	0.0	59
96 h	To normal	402	64.1*	34.2*	105	6.5	153
	To caffeine	525	50.6	14.1	141	0.8	125
120h	To normal	480	52.5	29.7*	138	1.6	118
	To caffeine	540	50.3	16.5	121	0.0	151
Development in normal medium		986	43.7	27.4	215	6.4	216

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University of Palermo, Italy. Further investigations on the tu-pb melanotic tumor mutant of *D. melanogaster*.

In the search for understanding the mechanism of tumor manifestation in the tu-pb mutant, a peculiar case of melanotic tumor manifestation in *Drosophila melanogaster* (Di Pasquale Paladino & Cavolina 1982; Di Pasquale Paladino & Cavolina 1983), we have undertaken an analysis of factors that may in some way affect this character.

Results of temperature shift experiments, which are carried out in order to determine the temperature-sensitive period, are summarized in Fig. 1. Shift-down experiments show that tumor manifestation is inhibited when temperature is shifted during the early stages of larval development. Tumors appear, although with a very low frequency, when flies had been left at 23.5°C until the 72nd hr of development. Percent tumor incidence typical of the strain is attained when development is completed at 23.5°C. In shift-up experiments a detectable decrease of tumor incidence is found only when larvae are left at 18°C also during the late stages of development. Tumor incidence is found to decrease also when temperature shift corresponds to the 144th hr of development.