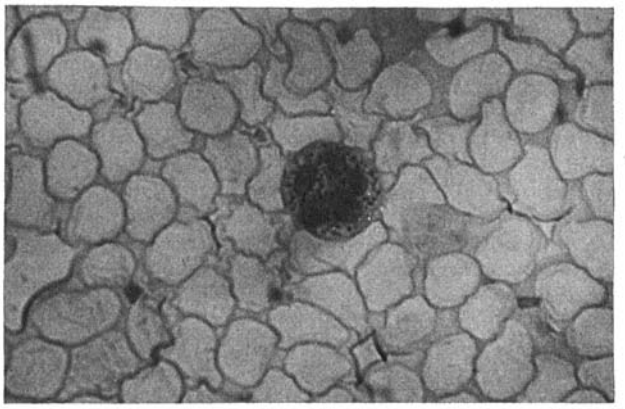

Fig. 1. Ribonuclease activity of polymorphonuclear leucocytes from hyperimmunized rabbit. The enzyme-positive granules are scattered throughout the whole extent of the cytoplasm. × 800.

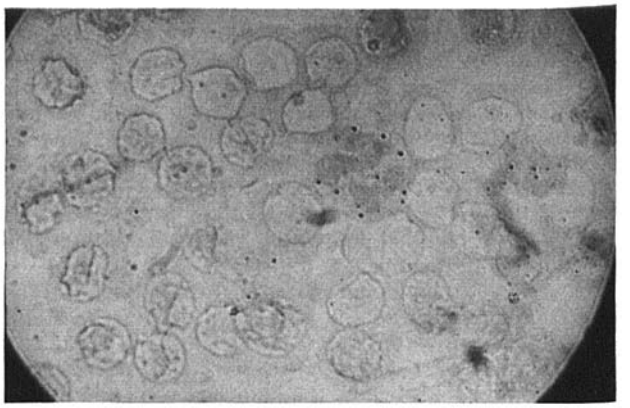

Fig. 2. Ribonuclease activity of polymorphonuclear leucocytes from normal rabbit. Note a few positive granules in the perinuclear zone. × 800.

particles of antigen is a well-known concept[9]. There are proposals that neutrophils engulf and perhaps digest particulates as chylomicrons in the blood, thus serving a function in clearing lipemic serum and metabolizing lipids[10]. The PMN leucocytes perhaps function as agents involved in the alteration of the antigen and transfer of information to immunological competent cells, which in this case are probably circulating cells. Such a role has been associated with the macrophage system[9]. COHN[11] suggests the possibility of macrophage ribonucleic acid in some way transferring information to competent lymphoid cells. HULLIGER and SORKIN[12], while investigating antibody formation in the circulation, showed changes in the composition of peripheral blood cells of hyperimmunized rabbits with a high number of PMN leucocytes at a stage when antibody synthesis in blood and thoracic duct cells was fairly active.

In a recent report CLINE[13] indicated a profound effect of phagocytosis on the several aspects of RNA metabolism. He postulates that particle ingestion induces an accelerated rate of destruction of pre-existing RNA and an increased rate of RNA synthesis. It is possible then, that the circulating PMN leucocytes of rabbit in the event of an infection are involved in the engulfing of the antigen. And during the process of phagocytosis the induced destruction of pre-existing RNA and increased RNA metabolism would need the increased concentration of intracellularly located ribonuclease to degrade this RNA. It may also be speculated that the rapidly proliferating PMN leucocytes contain much less ribonuclease inhibitor than the normal animal cells. UTSUNOMIYA[7] conceives of such a possibility of lower concentration of ribonuclease inhibitor in hepatic proliferating cells.

More extensive work is required to understand the dynamics of leucocytic enzymes in a wide variety of infections, especially before such conclusions are established.

Résumé. Les leucocytes du sang périphérique de lapins hyper-immunisés contre *Coxiella burneti* ont été comparé avec des leucocytes de lapins normaux. Par la méthode cytochimique on a pu constater chez les animaux hyper-immunisés un accroissement du taux des leucocytes polynucléaires, l'enzyme ayant une activité accrue ce qui s'accorde avec un titre élevé d'anticorps dans le plasma. L'auteur cherche à déterminer la signification de cette augmentation de l'enzyme chez les animaux hyper-immunisés et de son rapport avec la réaction immunogène.

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Effects of the Inhibitor of Xanthine Dehydrogenase, 4-Hydroxypyrazolo(3,4 d)pyrimidine (or HPP) on the Red Eye Pigments of *Drosophila melanogaster*

The mutants *ry* and *ma-l* of *D. melanogaster* are deficient in xanthine dehydrogenase (XDH)[1-3]. In consequence they do not form the reaction products isoxanthopterin and uric acid, and accumulate the corresponding substrates 2-amino-4-hydroxypteridine and hypoxanthine[4-6]. There is also a partial loss of the red pteridines of the eye; the eye colours being a dark red-brown (yellow-orange in the double mutant *st ry* or *ma-l;st*) in contrast to the bright red of normal flies[7,8].

Phenocopies of the *ma-l* and *ry* – or, correspondingly, of the *st ry* and *ma-l;st* – eye colour mutants have been obtained when wild-type or *st* flies were grown on a medium containing the inhibitor HPP[9-11].

Because at least 3 substances (neo-, iso- and drosopterin) contribute to the red colour of the eye in *Drosophila*[12,13], we proceeded to examine their modifications in the phenocopies produced by HPP as compared to the normal flies.

Wild-type (*Oregon R*) and *st* mutant strains were grown, according to KELLER and GLASSMAN[11], on different concentrations of HPP: 0.0, 0.01, 0.02, 0.03, 0.04, 0.05, 0.06, 0.08 and 0.10 g%. Adult flies were taken 2–3 days after they emerged and placed in boiling water for 1 min; their heads were dissected and squashed on Whatman No. 1 filter paper. The chromatograms, developed by ascending chromatography in *n*-propanol/2% ammonium acetate (1:1) in water, according to VISCONTINI et al.[12], were observed in the visible light and under an UV-lamp, emitting mainly at 365 nm. An arbitrary visual grading system (from '0' to '4' units) was applied to quantitate the results.

The concentration of HPP required to obtain 50% of adult flies with phenocopy eye colours was about 0.08% in the case of *Oregon R* and about 0.03% for *st*.

As the most evident modification produced by HPP, the chromatograms revealed a partial loss of the red eye pigments roughly proportionate to the concentration of the inhibitor. However, the relative amounts of individual drosopterins were not affected to the same degree: neodrosopterin appeared to be the most susceptible component, i.e. its loss was much more prominent than the loss of drosopterin and isodrosopterin, so that the latter substances accounted for a larger proportion of the total red pigments in the flies raised on effective concentrations of HPP as compared to the normal flies. Moreover, no observable change of the yellow eye pigments (sepia- and isosepiapterin) accompanied the loss of the drosopterins.

It seems, therefore, that the phenocopy eye colours, produced by HPP, can be ascribed to the marked drop of the reddest neodrosopterin and to the relative prevalence of the orange coloured drosopterin and isodrosopterin and of the yellow sepia- and isosepiapterins.

Riassunto. Esemplari di *D. melanogaster* di tipo selvaggio o del mutante *st*, allevati su terreno contenente l'inibitore della xantino-deidrogenasi 4-idrossipirazolo-(3,4 d)pirimidina (HPP), presentano l'occhio colorato in arancione scuro o in giallo. Cromatograficamente è stato possibile precisare che siffatte modificazioni dipendono dalla diminuzione dei pigmenti rossi contenuti nell'occhio, diminuzione che risulta molto più accentuata nel caso della neodrosopterina, il cui colore proprio è più rosso di quello degli altri pigmenti.

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Fungistatic Action of Aflatoxin B_1

BURMEISTER and HESSELTINE[1] demonstrated that growth of some bacteria is inhibited by aflatoxin. The toxicity of aflatoxin towards microorganisms has been investigated further by examination of the effect of aflatoxin B_1 on *Flavobacterium aurantiacum*[2]. Although the molecular site of inhibitory action has not been established, it has been shown that aflatoxin contains a coumarin nucleus[3]. KNYPL's[4] report that coumarin inhibits germination and growth of certain fungal spores prompted experiments designed to measure the fungistatic capacity of aflatoxin B_1 towards certain organisms.

2 nutrient media were employed. One (A) a modified Czapek's broth containing glucose, 45 g; $NaNO_3$, 4.5 g; KH_2PO_4, 3.0 g; KCl, 0.75 g; $MgSO_4 \cdot 7H_2O$, 0.75 g; $FeSO_4$, 0.02 g; tap water to 1 l. The other (B) contained the same constituents as (A) except that 5.0 g/l of yeast extract was substituted for the $NaNO_3$.

The fungi were grown by inoculating $2.5 \cdot 10^6$ fungal spores into 25 ml of culture medium A or B in 300 ml Erlenmeyer flasks. Incubation was carried out for a specific time on a rotary shaker at 30 °C. Dry weights were measured by harvesting the flask contents and drying them for 12 h at 110 °C.

Growth of several species of *Aspergillus* and *Penicillium* was inhibited by aflatoxin B_1 (Table) when grown in medium A. It is noteworthy that mycelial growth of strains of *A. flavus* was inhibited by B_1, since these organisms produce aflatoxin. Strain NRRL 3239 of *A. flavus* is a poor aflatoxin producer whereas strains NRRL 3000, NRRL 2999, and NRRL 3240 elaborate significant quantities of the toxin. Since growth of both the poor aflatoxin-producing strain and the actively producing strains is inhibited by B_1, it appears that capacity for toxin production is not directly linked to aflatoxin sensitivity.

Growth of the following organisms was not significantly inhibited by aflatoxin B_1: *A. terreus* NRRL 1967, *A. ochraceus* NRRL 408, *A. niger* NRRL 3, *A. clavatus* NRRL 1.

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