can be bound to a protein molecule has been completely developed by Klotz (7). This method is based on the law of mass action and assumes that binding occurs in a stepwise fashion, with the first mole bound being held the most firmly. The expression relating moles of small molecules bound per mole of copper-protein complex, r, with concentration of unbound ion (A) is given by

$$r = \frac{m(A)}{K + (A)}$$

Here, K is the intrinsic dissociation constant for the system, and m is the maximum number of bound ions per molecule. In order to evaluate m and K, the equation is rearranged to

$$\frac{1}{r} = \frac{K}{m}\frac{1}{(A)} + \frac{1}{m}$$

A graph of $1/r$ versus $1/(A)$ will be a straight line with the intercept on the $1/r$ axis equal to $1/m$ and the slope of the line equal to K/m.

The results of the binding studies involving histamine at *p*H values of 6.95 and 8.90 and Antistine at a *p*H of 6.95 are shown in Table 1. It was found that these binding data obeyed the law of mass action; that is, the binding increased with an increase in concentration of unbound ligand in equilibrium with the Cu(II)-proteinate.

Extrapolation of the linear plot of the reciprocals of the amount bound versus the concentration of unbound ligand yielded values for the maximum moles of ligand bound per mole of proteinate. These values were 2.75 and 20.0 for histamine at *p*H values of 6.95 and 8.90, respectively. The Cu(II)-proteinate was capable of binding a maximum of 1.74 moles of Antistine per mole of proteinate at a *p*H of 6.95.

The equilibrium constants for the first mole of ligand bound, obtained from the slope of the linear plot, were utilized to determine the free energy change for the formation of the ligand-proteinate complex. These values were -0.871 kcal and -0.910 kcal per mole for histamine at *p*H values of 6.95 and 8.90, respectively, while the corresponding value of Antistine was -0.868 kcal per mole at *p*H 6.95.

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References and Notes

1. Supported in part by grant No. E-1354 from the National Institutes of Health, U.S. Public Health Service.
2. Antistine is the commercial name for 2(N-phenyl-N-benzyl-aminomethyl)-2-imidazoline hydrochloride, which was kindly donated by the Ciba Pharmaceutical Company, Summit, N.J.
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Hereditary Ovarian Tumors in Drosophila melanogaster

A recent study has been published which describes normal oogenesis in the fruit fly, *Drosophila melanogaster* (*1*). During this study ovarian tumors were observed. However, the incidence was extremely low (two tumorous chambers among 39,900 developing eggs). It was shown subsequently that ionizing radiation (4000 r of Co^{60} gamma rays) increased the incidence of tumors by 26 times.

In *Drosophila* a developing egg consists of a chamber containing 16 cells. Fifteen of the cells function as nurse cells and nourish the 16th cell (the primary oocyte). All 16 cells arise from a single cell in the germarium, which undergoes four consecutive divisions. It was postulated therefore that, in the region of the germarium where 16 cell cysts are formed, an interaction takes place between cytoplasmic substances localized in this region and the genome of the cells. The stimulated genetic material is thought next to manufacture a substance which inhibits further cytokinesis in a precise fashion. Radiation might occasionally inactivate that portion of the genome of an oogonium responsible for the production of the inhibitor. This mutation would then be passed to the progeny of the oogonium. These cells would now divide in an uncontrolled fashion and produce the observed tumorous chambers which contain hundreds to thousands of mitotically active cells.

We set out to detect mutant genes which would cause such uncontrolled cell division; but we recognized that such genes would be difficult to obtain, since they would generally produce consequences which would be lethal at an early stage in the life-cycle. From our knowledge of oogenesis, it seemed reasonable to predict that some of the genes we were looking for might be found among the 60 or so nonallelic, recessive female sterile mutants of *Drosophila melanogaster,* because uncontrolled division in egg chambers would convert developing eggs to tumors and so sterilize the fly.

We therefore obtained approximately 20 female sterile mutants and proceeded to make Feulgen whole mounts of ovaries of females homozygous for the various female sterile genes (*2*). To our amazement, the first female sterile mutant examined turned out to be a case in point, and the first ovarian preparation contained more tumors than the total we had observed from all sources up to that time. The incidence of tumors was found to increase with the age of the female. In this strain, adjacent chambers in an ovariole often fuse together. If one such chamber is tumorous and the other is normal, there will be produced a compound chamber containing normal and tumorous cells. The actively dividing tumorous cells will subsequently invade the normal tissue of the compound chamber.

This mutant which produces tumors of one tissue at one particular stage in the life-cycle is *fused* (*fu*), discovered by C. B. Bridges in 1912. It is located at 59.5 on the X-chromosome. The allele in question is spontaneous in origin, but alleles induced by x-rays or chemicals have been frequently observed. In our stock (which was obtained from the Yale collection) *fu* is balanced over M5. Females heterozygous for *fu* show no tumors. On the other hand, females homozygous for fu^{ff} (an allele of *fu* induced by formalin treatment) also show ovarian tumors. It appears that *fu,* in addition to its many other bizarre effects (*3*), produces ovarian tumors and therefore represents excellent material for further studies of the mechanism of tumorous growth.

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Fluorescence of Ethylenediamine Derivatives of Epinephrine and Norepinephrine

In 1952, Weil-Malherbe and Bone introduced a method for the chemical determination of total "epinephrinelike" substances in blood, which included separation of the catechol amines from other plasma components by adsorption chromatography and measurement of the separated fraction by fluorometry (*1*). Shortly thereafter, Persky and Ros-