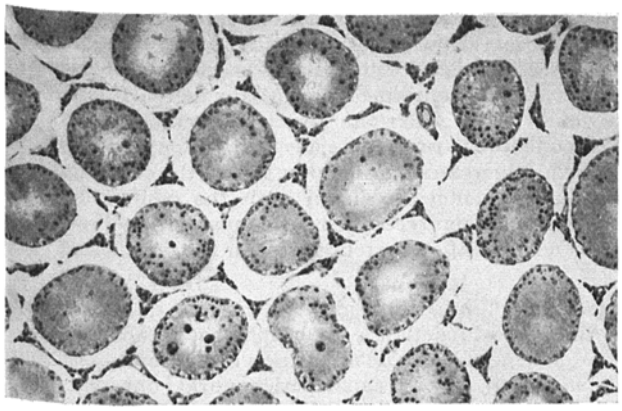

Fig. 2. Testis of rat after 30 days of treatment. The photograph shows a clear-cut decrease of the germinal epithelium and numerous giant cells as well as a marked atrophy of the seminal tubules ( × 230).

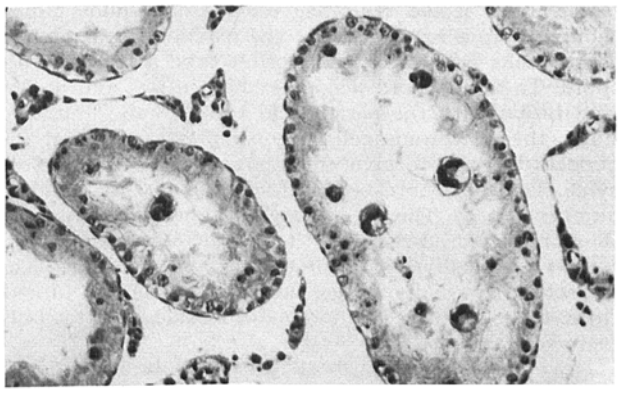

Fig. 3. Testis of rat after 30 days of treatment. A higher magnification of another section of the testicle of the same animal shows numerous pseudo giant cells ( × 585).

of degenerating spermatids. The behaviour of the intestinal mucosa, in which a normal number of mitoses was seen, shows that the FAA acts selectively on the testicular germinal epithelium and that its action is not strictly related to rapid cellular multiplication as is the case with radiomimetic substances. In the liver no notable proliferative phenomena were observed in the small bile-ducts. On the other hand, in our experiments the FAA acts firstly on the most mature elements of the germinal epithelium and not on the cells where the mitoses are more numerous. Thus, the FAA differs from other experimental conditions (radiation, etc.) which produce testicular atrophy, firstly damaging the germinative cells (spermatogonia). On the basis of those results it is clear that FAA produces a testicular atrophy by a selective action on the seminal epithelium.

Further study on the mechanism of action of this substance is currently under way.

*Riassunto.* Gli autori descrivono le lesioni del testicolo del ratto osservate nel corso di sperimentazioni con la fluoroacetamide. Tali lesioni consistono in alterazioni regressive interessanti elettivamente la linea seminale.

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## Hypochord in the Anurans

The hypochord in the anurans develops, according to some authors, below and parallel with the notochord[1] as a fibrous structure and later becomes connected with the perichordal tube by the fibrous tissue[2]. Shumway and Adamstone[3] opine that it develops from the lower surface of the chorda plate and its functions are unknown.

The present investigation deals with the analysis of the ontogeny of the hypochord in an anura (*Rana tigrina*) and its role in the development of the vertebral column with special reference to the localizations of alkaline phosphatase in the process. Fertilized eggs of *R. tigrina* from the midgastrula to the opercular stage (Shumway[4] stages 11 to 25), tadpoles at various stages of development and some adults were fixed in Zenker's fluid for histological study; for alkaline phosphatase, Gomori's modified technique[5] was followed.

*Observations.* The hypochord is formed out of endoderm when the notochord is already differentiated with covering sheaths, and the perichordal tube is formed round the notochord with an aggregation of mesenchymatous cells (stages 11 and 12). In the notochordal portion, the reaction for alkaline phosphatase is intense. In the hypochord, the reaction is less but greater than that of the perichordal tube region. Later (stages 13 to 15), the perichordal tube becomes well formed round the vacuolated notochord and the hypochord gradually separates from the endoderm to abut against the perichordal tube. The hypochord cells show two types of reaction: (a) cells in contact with the hypodermis show intense reaction and (b) cells in contact with the perichordal tube show less reaction. The mesenchyme cells round the perichordal tube and hypochord also show less reaction. The hypochord now starts vacuolations with less reaction for alkaline phosphatase (stages 16 and 17) and on its outer surface a layer similar to the elastic layer of the notochord is formed. The localizations for alkaline phosphatase become diminished in the perichordal region. In the successive stages of development (stages 18 to 25), the notochord becomes vacu-

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olated with feeble reactions for alkaline phosphatase though there is an increase of the reaction in the mesenchyme cells round the perichordal tube and the hypochord. The hypochord becomes small in size and undergoes fusion with the perichordal tube. In the limb bud stage, the sclerotome cells become plastered round the perichordal tube to give an intense reaction for the enzyme. The hypochord becomes very small with a limited number of cells. The outer regions of the cell membranes show an increased reaction though the cytoplasm of the cells is practically devoid of the enzymatic actions. The hypochord gradually merges with the perichordal tube of the notochord and is not detectable in an adult with fully regressed tail.

*Discussion.* The hypochord thus may be regarded as an accessory structure to the notochord and comes out of the hypodermis when chorda is fully differentiated and prior to the appearance of the perichordal tube. SPRATT[6] is also of the same opinion that it is hypodermal in origin. It remains as a transitory structure and the development is very much similar to that of the notochord, including sheath formations and vacuolations; gradually it becomes fused with the perichordal tube. Subsequently, the mesenchyme cells migrate beneath the hypochord. Thus, a very high order of contact specificity[7–9] develops amongst the cell surfaces of the perichordal tube and hypochord proper, resulting in a complete fusion of these two structures. The hypochord cells at first show less reaction for alkaline phosphatase, but gradually the reaction increases until the vacuolations start. With the commencement of chondrification, there is an increase in the reaction for alkaline phosphatase in the surrounding mesenchyme cells – a feature which is very similar to that of a bird[10]. Along with this there is an increase for the localization pattern of the enzyme, particularly in the intercellular regions. Later, when the structure becomes fused with the perichordal tube, the cells are without any reaction though it increases on the outer side of the cell membrane – a phenomenon associated with the ossification of a structure[11]. The hypochord thus may help in the formation of the ossified vertebral area in the anurans.

*Résumé.* L'hypocorde des Anoures est d'origine endodermique. Sa réaction à la phosphatase alcaline s'accroît graduellement jusqu'à l'apparition des vacuoles. Au cours de sa fusion avec le canal périchondral, une réaction à l'enzyme s'observe à la surface externe de la membrane cellulaire. L'hypocorde peut aussi contribuer à la formation des vertèbres.

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## Some Meiotic Consequences of Ethyl Methane Sulphonate and the Interaction of Copper or Zinc

The interaction of several cations on the chromosome breaking activity of EMS has been demonstrated[1,2]. Copper and zinc salts were found to enhance the primary effects of EMS in broad beans considerably. The effect of copper was confirmed for wheat[3].

It becomes evident that some ions can increase or decrease the activity of mutagenic compounds. Even low pH can modify the effects[4]. Modifications produced by various factors are not limited to monofunctional compounds of the mesyloxy group. It could be proved that the interaction still exists for bifunctional or trifunctional compounds of the same group, although less evident[5]. The mechanism by which ions can interact with alkylating agents is not yet clearly understood.

We reported that dry seed treatments with EMS can result in meiotic consequences which could, at least partially, explain the high sterility induced with this compound[6].

Continuing the investigations along this line of research, we intended to see if these meiotic consequences could be modified in some way by ions.

In the present experiments, barley dry seeds (piroline variety) were treated with EMS (0.3 g per 100 ml for 24 h) and with EMS solutions of the same concentration respectively added to $CuSO_4$ (0.03 mg per 100 ml) or $ZnSO_4$ (0.03 mg per 100 ml water), pH around 6.5.

In previous experiments, these concentrations were found to modify the action of EMS on mitotic chromosomes[1,2]. In the present experiments, spikes of the generation X1 were collected for meiosis examination. Chromosomes were stained with Feulgen and observed on squashes.

All kinds of aberrations reported after X-ray treatment could be detected in the slides. We mainly noted the following aberrations. At metaphase I they consist of rings of four, rings of six, chains of four and figures of eight, all these abnormalities arising from chromosome translocations and small chromosome fragments coming from deletions. At anaphase I, the aberrations consist of chromosome and chromatid bridges and lagging fragments of multiple origins.

At anaphase II, we noted lagging fragments, micronuclei and a few bridges. After treatment with solutions to which copper and zinc were added, the aberrations were qualitatively essentially similar as after treatment with solutions without one of these salts. Controls performed in the same experimental conditions show a low amount of chromosome fragments at anaphase I. No other abnormality could be seen.

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