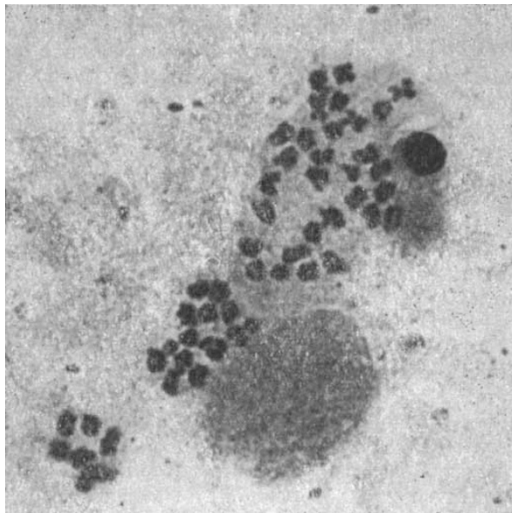

Fig. 2. Dividing cell in primary culture from *Clupea harengus* gonads. Aceto-orcein (×1,100).

to that of the growth medium, giving a final colchicine concentration of about 0·5 μg/ml. After exposure to the colchicine for 12–16 h, aceto-orcein preparations were made for chromosome counts. In addition, some coverslips were stained with haematoxylin and eosin.

Complete monolayers were not obtained during the course of this experiment, but considerable attachment and proliferation occurred. The cultured cells appeared to be fibroblasts (Fig. 1). The cells were highly vacuolated, in sharp contrast to the appearance of freshwater fish cells in culture. This may have been a consequence of the hypotonicity of the growth medium. Numerous dividing cells were seen (Fig. 2); fourteen metaphase chromosome groupings were counted, ten of which gave counts of fifty-two. One cell gave a count of fifty chromosomes, and three cells gave a count of fifty-three. Metacentric, acrocentric, and possible telocentric chromosomes were visible, and varying chromosome sizes were represented among the karyotype.

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Technique for Investigation of Sex Chromatin in Amniotic Membrane of Rat Foetuses

SEX chromatin in the amniotic cells of rat foetuses is useful for sex diagnosis before the development of the gonads, especially in view of the fact that sexual dimorphism is not conspicuous in other tissues[1,3]. To be a simple and accurate technique, however, the preparation of amnion for the examination of sex chromatin requires some special precautions. Absolute or 95 per cent alcohol has been habitually used as the fixative[3], but we feel that it is troublesome because it shrinks the cells and coagulates the mucopolysaccharides. Cells of rat foetuses have several heterochromatic bodies[1,3], and consequently sex chromatin is not easy to distinguish when the cells shrink. Furthermore, the amnion in young rat foetuses has a considerable content of mucopolysaccharides which when coagulated hinder fixation and make the cells difficult to see. To overcome these difficulties we have used 50 per cent glacial acetic acid as the fixative, and have obtained with it good fixation and swelling of cells, which makes it easier to differentiate sex chromatin from chromocentres.

The following procedure was adopted. The uterus wall was cut and foetuses were removed together with their enveloping membranes; the chorion was peeled off and the amnion was opened and separated from the foetus by cutting the umbilical cord; the amnion was immersed immediately in 50 per cent glacial acetic acid for 30 min at room temperature; it was extended over an albuminized slide with care to avoid folding of the amnion; later it was scratched gently with a razor blade and spread over the slide; histological preparations were dried at room temperature, hydrolysed in normal hydrochloric acid for 10 min at 60° C and stained with carbol and fuchsin[2].

Sex chromatin was found in 10 per cent to 35 per cent of amniotic cells of female foetuses while male foetuses only showed 0 per cent to 4 per cent of cells with a sex chromatin-like body (Fig. 1).

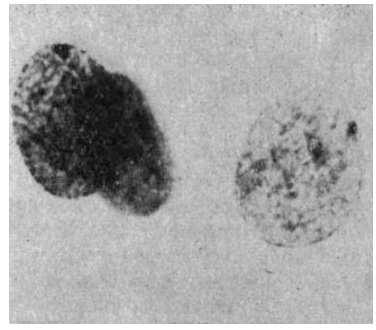

Fig. 1. Sex chromatin in cells from a 14 day old rat foetus.

This technique was consistently used to prepare amniotic cells from rat foetuses. It was, however, also successful in the few cases in which it was used to determine the sex of mouse foetuses.

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RADIOBIOLOGY

Possible New Approach to the Evaluation of Radiation Injury of Bone Marrow

IT is well established that injury to bone marrow plays a main part in death from radiation. Fluorescence microscopy with acridine orange was explored by Meisel *et al.*[1] as a technique for evaluating such injury in cell populations. With the same technique Breivis[2] also quantified cellular damage induced in rat marrow by chloroethylamines and X-rays. This technique enabled them to determine the number of damaged cells in the