

supplying energy for growth and survival under saline condition and thereby inducing salinity resistant to crops.

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#### PRENATAL DETECTION OF FETAL SEX\*

IN 1956 Fuchs and Riis showed the possibility of fetal sex determination from amniotic fluid cells using Barr body technique. In 1971 Khudr and Benirschke used fluorescence studies to locate Y-chromosome (Y-body) in the nuclei of amniotic fluid cells in male cases.

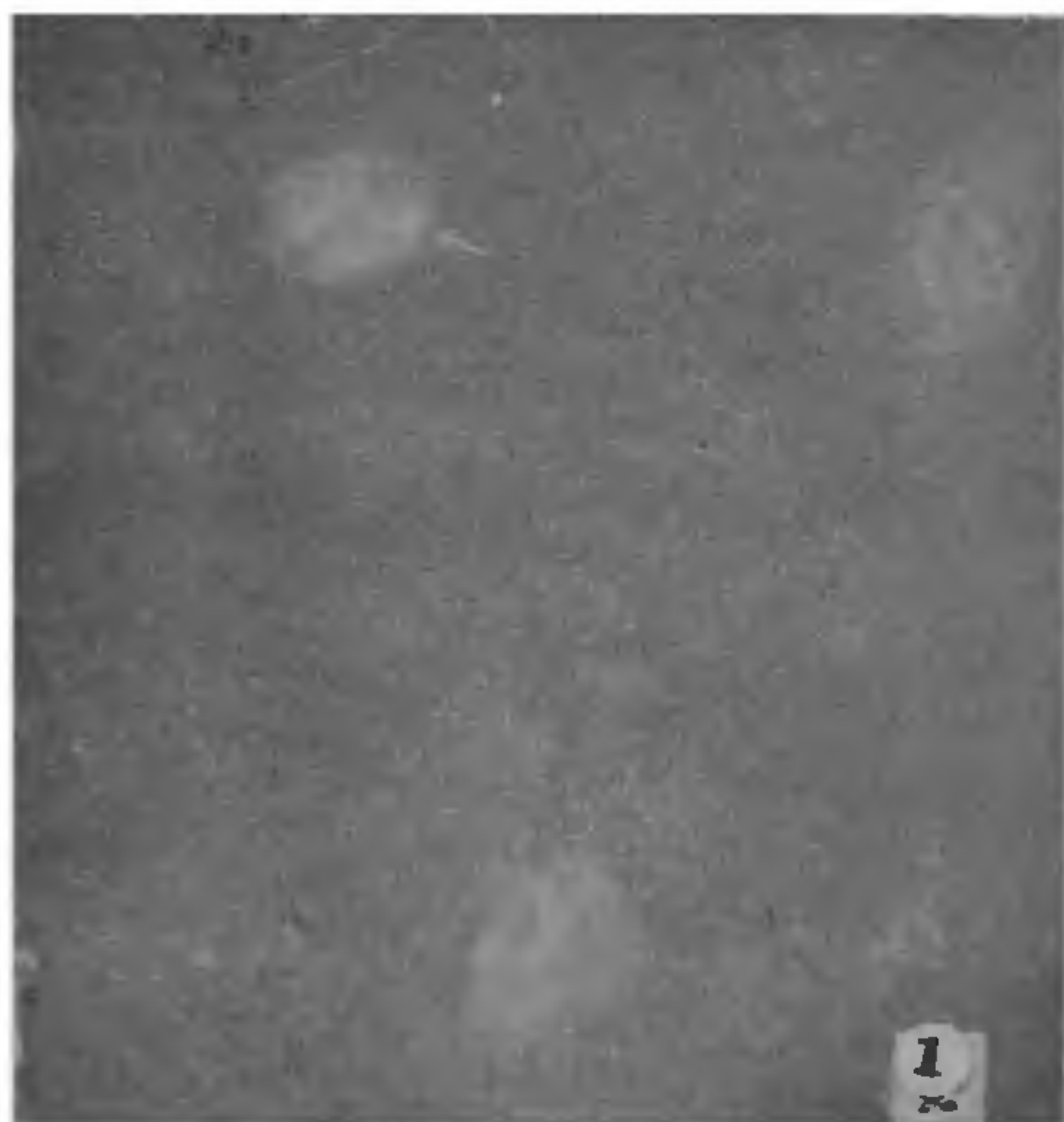
the family suffering from leukodystrophy. All abortions were done for psychiatric and health reasons.

The volume of amniotic fluid sample obtained varied from 5 ml to 12 ml depending on the month of pregnancy, and the technique used for abortion.

The sample was centrifuged at 1000 r.p.m. for 10 minutes. After discarding the supernatant, the cell pellet was resuspended and fixed in scetic acid alcohol (3 parts methanol + 1 acetic acid) for 20 minutes. The smears were prepared and some of the slides were stained in Giemsa stain for X-chromatin and the rest was stained in 0.5% quinacrine dihydrochloride (Atebrin) for fluorescence microscopy to study Y-body.

**Results and Comments.**—Results were confirmed by the gross examination of fetuses, mainly of external genitalia. The results agreed in all the cases except one, where the sample was not sufficient for the satisfactory preparation.

The amniotic fluid cells of male fetuses showed a single fluorescent body usually located peripherally sometimes accentrically within the nucleus (Fig. 1). In case of a female, amniotic fluid cells showed typical Barr body (Fig. 2) but no such fluorescent



FIGS. 1-2

The amniotic fluid cells are of fetal origin and derived mainly from fetal skin and amnion. The present paper deals with the use of simple and rapid techniques (Barr body and fluorescence method) available for prenatal sex determination.

**Material and Methods.**—The amniotic fluid samples were obtained from 15 patients undergoing therapeutic abortion by intrauterine prostaglandin or by saline injection between 10 to 18 weeks of gestation. One sample was obtained from a patient who had two sons and 8 other male members in

body. One hundred cells were scanned from each preparation and in the nuclei of male fetuses a typical fluorescent body was found in 20% to 30% of the cells and in the nuclei of female fetuses 20% to 60% of typical Barr body was found. The variation in number may be due to presence of different types of cells. In a case of leukodystrophy the amniotic fluid cells showed male fetus and the patient decided to continue pregnancy. She delivered a male child which confirmed our prediction of the sex.

This preliminary study shows that the method used may be of great help in determining the sex of the fetus at the early stage of pregnancy. It is a test which can be performed in less than one hour.

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#### RELATION OF SPORULATION TO FUSARIC ACID PRODUCTION IN MUSKMELON WILT PATHOGEN

FUSARIC acid (FA), a wilt toxin, is produced by many fusaria both *in vitro* and *in vivo*. From this laboratory Bhaskaran and Prasad<sup>1</sup> reported the *in vitro* production of FA by the muskmelon wilt pathogen [*Fusarium oxysporum* f. *melonis* (Leach and Currence) Snyd. and Hans.]. Interestingly the toxin production was more in media which favoured good sporulation<sup>2</sup>. Studies on the relation between sporulation and fusaric acid production were made and the results are reported here.

Hundred ml aliquot of sterilized Czapek's medium in 250 ml conical flasks were inoculated with 8 mm disc of the actively growing fungus and incubated at room temperature ( $28 \pm 2^\circ \text{C}$ ). At the end of incubation period, the flasks were shaken in a rotary shaker for 30 min and the spore load was estimated by Haemoagglometer counts. The content was passed through a double layered cheese cloth, washed with distilled water. The mycelial mat was separated from cheese cloth, blotted and weighed. The filtrate was centrifuged to separate spores from culture filtrate. The fusaric acid in culture filtrate, mycelium and spores was detected following the respective methods<sup>3,4</sup>. The presence of FA was confirmed by paper chromatography. The influence of certain media, viz., Czapek's, Coon's, Horne and Mittar's and Park's suggested for *Fusarium*<sup>5</sup> was tried for assessing growth, sporulation and FA level.

The effect of incubation periods on growth, sporulation and FA production is shown in Table I and the effect of different media on growth and FA production is presented in Table II. Among the media tested, Czapek's medium favoured relatively higher sporulation and FA production. Growth

TABLE I

Effect of incubation period on growth, sporulation and fusaric acid (FA)\* production by *Fusarium oxysporum* f. *melonis*, when grown in Czapek's medium

Incubation period in days	Mycelial dry weight mg/100 ml	FA in mycelium	Number of spores/ml	FA in spores	FA in culture filtrate
10	175	274.5	15,000	310.0	610.0
15	227	334.0	38,000	615.0	928.3
20	232	330.0	53,000	980.0	1240.0
25	240	332.0	52,000	950.0	1220.0
30	245	335.0	49,000	890.5	1195.0

\* FA in inhibition annules (mm<sup>2</sup>).

TABLE II

Effect of different media on growth, sporulation and FA\* production by *Fusarium oxysporum* f. *melonis*

Media used	Mycelial dry weight (mg/100 ml)	FA in mycelium	Number of spores/ml	FA in spores	FA in culture filtrate
Coon's medium	247	285.0	42,000	612.0	1130.2
Czapek's medium	238	280.0	51,000	845.0	1230.8
Horne and Mittar's medium	312	315.0	28,000	394.5	829.5
Park's medium	215	225.0	32,000	480.0	890.5

\* FA in inhibition annules (mm<sup>2</sup>).

studies in Czapek's broth indicated maximum sporulation and FA production on 20th day and growth on 30th day. Thus a parallelism existed between sporulation and FA production by this pathogen. Sandhu<sup>6</sup> demonstrated that FA is a product of active metabolism, probably ultimately connected with tricarboxylic acid cycle. The accelerated metabolism during sporulation may be the cause of more FA production. The toxin level in mycelium and conidia showed a marked variation. FA in mycelium and conidia was first reported<sup>2</sup> in *Fusarium oxysporum* f. *vasinfectum*. Toxin in mycelium and