SEX-HORMONES IN NEUROSPORA CRASSA
FURTHER STUDIES ON ITS BIOLOGICAL PROPERTIES¹)

by

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ABSTRACT

Sexual hormones, isolated from the single strains (Em A, Em a) and cross (Em A × Em a) cultures of Neurospora crassa have been found to restore the fertility of a completely sterile strain obtained from the “selfed” progeny of Em A. In an attempt to see the effects of different concentrations (dilutions) of the “cross-extract” on Em A and also on the cross Em A × Em a, different threshold values were indicated.

We (ISLAM & WEIJER, 1969) reported the isolation of sexual hormones (sex and fertility inducing substances) from the single strain culture - Emerson-A (Em A) and Emerson-a (Em a) as well as established cross, Em A × Em a, of Neurospora crassa. A brief description of isolation, biological and biochemical characterizations of the hormonal substances was given in this paper. Subsequently, in another paper we (VIGFUSSON et al., 1971) have dealt at length with the biological properties; vis-a-vis its effects on the UV-induced sterility mutants, wild type strains (Em A and Em a) and also on the wild type cross Em A × Em a. This paper describes the further biological characterizations of the hormonal substances, viz: a) The effects of the extract of the single strains (Em A, Em a) and of the cross (Em A × Em a) on the sterile isolate I-5. b) The effects of the different concentrations (dilutions) of the extract from the cross-Em A × Em a (referred to as the “Cross-extract”) on the single strain culture (Em A) and also on the wild cross (Em A × Em a).

MATERIALS AND METHODS

Strain I-5 (perithecium No. 1, random spore No. 5) was an isolate from the homokaryotic fruiting of Em A which was subsequently proved to be completely sterile (ISLAM & WEIJER, sent for publica-

¹) Part of the work reported here is from the Ph. D. dissertation of the author and the remaining portion is from his graduate work, previously not reported elsewhere.

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tion elsewhere). This sterile isolate and the wild type strains - Em A and Em a were utilized in these experiments.

WESTERGAARD'S crossing medium (WESTERGAARD & MITCHELL, 1947) was used throughout the experimentation and the method for the characterizations of the extracts were as previously reported by us (0.1 ml/plate). To see the effects of different concentrations of the ‘cross-extract’ initially 5 mg of the extracted residue was dissolved in 5 ml of chloroform (100 \( \mu \)g/0.1 ml) and from this appropriate dilutions were made so that 0.1 ml of the extract given to each plate contained 20, 10, 5 or 2 \( \mu \)g. Each experiment was repeated at least twice with a minimum of 4 replica plates in each case.

**Experiments and Results**

(a) The effects of the extracts of Em A, Em a and Em A x Em a on the sterile isolate I-5.

The sterile isolate I-5 was crossed to both Em A and Em a with and without the addition of the extracts. Ten crossing plates were treated with the extracts (Em A, Em a and Em A \( \times \) Em a) in each case whereas an additional 10 plates (without extracts) were kept as controls. At seven day intervals the plates were examined for a possible effect of the extracts. It was found that the cross I-5 \( \times \) Em a treated with the extracts, developed perithecia after two weeks of incubation. The control plates of both the cross (I-5 \( \times \) Em a, I-5 \( \times \) Em A) and also the extract treated plates of the cross I-5 \( \times \) Em A showed no development of perithecia (occasionally one or two ill-developed perithecia, without any spores were found in some plates). The perithecia in the cross I-5 \( \times \) Em a, developed under the influence of the extracts, showed spore-shed after 4 weeks. The experiment (I-5 \( \times \) Em a) was repeated several times and similar results were obtained. From the results obtained it showed that the ‘cross-extract’ and the single strain extract of Em a are more potent in their biological effects than the extract of Em A, which gave rise to very few perithecia (0—15) per plate (Plate 1—4). In addition, a mixture of the extracts of Em A and Em a in the ratio 1:1 was made and tested for their combined activity. It was found to be less potent than that of the ‘cross-extract’.

(b) The effects of the different concentrations of the ‘cross-extract’ (Em A \( \times \) Em a) on single strain culture (Em A) and on wild type cross (Em A \( \times \) Em a).

The effects of different concentrations (2, 5, 10, 20 and 100 \( \mu \)g/plate) of the ‘cross-extract’ were studied later. The study was limited to the protoperithecia development in single strain culture of Em A and fertility of the wild type cross Em A \( \times \) Em a. The results indicated that with the increase in the dose of the extract from 2 \( \mu \)g/plate to 20 \( \mu \)g/plate there was an increase in the number