Oryzanone, an Attractant of the Rice Stem Borer

Sir:

It has been reported that after the 2,4-D (2,4-dichlorophenoxyacetic acid) treatment of crops there arises an increase in injurious insects¹⁾. This is also verified in case of the rice stem borer (*Chilo suppressalis* Walker) in Japan^{2,3)}.

To solve the above problem the authors propose to investigate the possibility of developing a repellent for the rice stem borer by the application of a repellent mixed with 2,4-D.

However, in order to find a repellent it was first necessary to find a good attractant which could then be used as a standard. If a suitable good attractant is to be found and standardized, consequently no repellent can be based against the standardized attractant.

This report concerns a study of a new naturally occurring rice stem borer attractant which the authors isolated and named Oryzanone.

FIG. 1. Odor Trap Apparatus.

The extracting procedure from rice plants was established by a adopting the following assay method. An odor trap method consisting of four small glass tubes was used (Fig. 1).

Small pieces of absorbent cotton wool containing an appropriate amount of the sample to be tested were placed in two test tubes. The other two tubes also containing small pieces of absorbent cotton but not containing the test samples were used as controls. All the above substances were placed in a petri dish as shown in Figure 1. The test samples dissolved in methanol were placed in the tubes and allowed to dry at 90°C for 30 minutes. Ten insects grown on sterilized artificial media⁴⁾ were set free in the centre of the dish and placed in the dark at 25°C for 24 hours. The insects found inside the test sample tubes and the control tubes or outside the tubes were recorded as positive (+), negative (−) and indifferent (±), respectively. Each test was repeated five times, using a total of fifty insects.

The results of fractionation and characterization of the borer attracting component obtained from rice plants showed that the substance was soluble in alcohol and ether. It was neutral and distillable at 100°–140°C, at 3 mmHg. The chromatogram with silicic acid and hexane⁵⁾ showed that this substance was chromatographically polar. Hydrolysis with 2N alcoholic potash demonstrated that this substance is contained in the unsaponifiable fraction. After rechromatographing the unsaponifiable oil with silicic acid and hexane, the effluent with hexane containing 1% ethanol was distilled. The distillate obtained was about 50 mg from one Kg of air-dried rice plants.

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The threshold dosage for the test of the above distillate was found to be 1.25 microgram per test tube (Table I).

After examination of this component by

TABLE I.
ATTRACTANT TEST WITH ORYZANONE

No. of insects	No. of test	Dosage per tube (microgram)	Response		
			+	-	±
50	5	500	19	4	27
50	5	25	19	4	27
50	5	1.25	13	7	30
50	5	0.0625	4	3	43
50	5	0.0031	2	3	45
50	5	0.0001	4	3	43

FIG. 2. 2,4-dinitrophenylhydrazone of Oryzanone.

chromatography and the chromatostrip method⁶⁾, it was found separable into two components, of which the component more active as a borer attractant, was shown to be an aliphatic unsaturated ketone compound by ultraviolet (max. 251m μ) and infrared spectra examination. By the decomposition of its 2,4-dinitrophenylhydrazone (m. p. 249°C) (Fig. 2) the original compound was recovered and identified by the physiological test and absorption spectra. It was named Oryzanone after the rice plant *Oryza sativa* L., from which it was isolated. The authors wish to thank Prof. Y. Sumiki of the University of Tokyo and Prof. T. Tamura and Prof. K. Iyatomi of Nagoya University for their constant advice and encouragement.

Katsura MUNAKATA
Tetsuo SAITO
Shigeru OGAWA
Shoziro ISHII*

Faculty of Agriculture,
Nagoya University,
Anjō, Japan

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* National Institute of Agricultural Science, Nishigahara, Tokyo, Japan.

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Hydroxyaspergillic Acid as Growth Inhibitant against Hiochi-bacteria (Studies on Growth Inhibitor of Hiochi-bacteria, Specific Saprophytes of Saké. Part 4)

Sir:

The authors searched for antibiotics active against hiochi-bacteria using the broth filtrates of the shaking cultures among 314 strains of *Aspergillus oryzae*. The culture filtrates of 18 strains showed inhibition against the growth of hiochi-lactobacillus H-34 and true hiochi-

bacillus H-1¹⁾. Recently, we have succeeded in isolating an antibiotic, as pale yellow crystals of m. p. 152°C from culture filtrates of these strains. This communication deals with this antibiotic.

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