

for kindly going through the manuscript and for his keen interest.

17 September 1983

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ELICITATION OF MOMILACTONE BY GIBBERELLINE IN RICE

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INVOLVEMENT of momilactone in disease resistance of rice cultivars has been previously reported¹⁻³. In this communication, elicitation of momilactone by gibberelline in healthy coleoptiles and leaf sheaths of rice and stimulation of momilactone biosynthesis in GA₃-treated and untreated infected tissues is reported.

The selection of the rice cultivar for the study was based on its responses to *Acrocyndrium oryzae*, the causal fungus of sheath rot disease. Earlier pathogenicity test of *A. oryzae* on different rice cvs. revealed that tall cvs. particularly Mahsuri, Rupsail, Badkalamkati were resistant while semidwarf ones viz Jaya, IR-8, CR-126-42-1 were susceptible to sheath rot⁴. A susceptible cv. (Jaya) was, therefore, chosen to induce resistance by chemical activation of host defense mechanism. Different concentrations (0.1, 1, 10 and 100 ppm) of GA₃ solution were sprayed on 9-week old rice plants (cv. Jaya) grown in earthen pots containing soil compost, twice at an interval of 4 days and inoculated with spore suspension (8×10^6 spores/

ml, 1 ml/leaf sheath) after 3 days of the second spray. Control plants were sprayed with sterile distilled water. The replicate pots (4 plants/pot) were taken for each treatment and the disease intensity was assessed 21 days after inoculation following the method of Raychoudhuri and Purkayastha⁵. The results showed that susceptibility of plants decreased when treated with GA₃ (10 and 100 ppm). The disease indices (DI/leaf sheath) were 6.50 and 1.57 for control and treated (100 ppm) plants respectively. Again, when 10 ppm of GA₃ solution was sprayed on dark grown coleoptiles and inoculated with *A. oryzae*, the roots became brown after 48 hr of incubation while the control (uninoculated, treated) roots remained white.

Mimilactone was extracted from both treated and untreated coleoptiles following the method of Cartwright *et al*² with modifications¹. The fractions obtained from Sephadex LH-20 column were evaporated to dryness and the residue in each case was dissolved in 1 ml. of 95% ethanol. Aliquots of each fraction were applied separately on TLC plates (silica gel G, BDH), developed in chloroform-ethanol (97:3) solvent system, dried and sprayed with a mixture of vanillin-H₂SO₄. The R_f value of the authentic sample (momilactone A) was compared with the isolated momilactone A. Spectral analyses of samples were also carried out for quantification of momilactone. For extraction of momilactone untreated leaf sheaths and GA₃ (100 ppm)-treated leaf sheaths (cv. Jaya) were excised 3 days after second spray, inoculated with spore suspension and incubated for 48 hr. Two hundred grams of infected leaf sheaths (both treated and untreated) were extracted for momilactone following the method as described. The fractions containing momilactone (detected by chemical method) obtained from Sephadex LH-20 column was dried on to activated celite 545 (600 mg) and applied to a column of silica gel in hexane. The

Table 1 Effect of GA₃ on momilactone 'A' level in rice cv. Jaya

Treatment	Plant part	Concentration of momilactone (as μg momilactone A/g fresh wt. of tissue)	
		Healthy	Infected
Untreated	Coleoptile	0	5.59
Treated (10 ppm GA ₃)	Coleoptile	13.20	16.70
Untreated	Leaf sheath	0	8.64
Treated (100 ppm GA ₃)	Leaf sheath	14.54	19.90

fractions were eluted with a gradient of 20, 50, 70 and 85% of chloroform in hexane and finally 95% chloroform in hexane. The fractions were evaporated to dryness in a rotary evaporator, the residue dissolved in 10% ethanol in chloroform and chemically tested for momilactone as described. GA₃ induced momilactone synthesis in healthy dark grown coleoptiles as well as in leaf sheaths and markedly stimulated momilactone biosynthesis in treated, infected leaf sheaths and coleoptiles.

Gibberellic acid (GA₃), which is a degraded diterpene⁶, may act as a precursor or gibberellin-mediated enzyme (associated with momilactone biosynthesis) production may account for the elicitation of momilactone synthesis in GA₃-treated, non-inoculated tissues. The mechanism involved in the induction of momilactone synthesis by gibberellin requires further study.

The authors thank CSIR for financial assistance. Thanks are also due to Smt. Saswati Biswas for her help.

2 May 1983; Revised 16 December 1983

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A HISTOCHEMICAL STUDY OF THE EFFECT OF RMI 12,936 ON THE STEROIDOGENIC ACTIVITY IN THE OVARY OF THE GERBIL, *TATERA INDICA*

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17 β -HYDROXY-7 α -methylandroster-5-en-3-one (RMI 12,936) is believed to be antiprogesterone in its

activity. It inhibits ovulation and causes atrophy of the ovary of the rats and disrupts estrous cycle and induces luteolysis in hamsters¹⁻⁴. In the present study the drug's effect on ovarian steroidogenesis in the gerbil, *Tatera indica hardwickei* (Gray) is examined by histochemical analysis.

Female nonpregnant gerbils, freshly collected around Mysore City, were maintained in the laboratory for a few days. The drug RMI 12,936 (kindly courtesy gift to Dr L. L. Albrecht, Morrell Research Centre, USA) was administered subcutaneously at a dosage of 2 mg/animal/day in 0.2 ml olive oil for 15 days. Another group of gerbils received only the vehicle (olive oil) for the same duration. All these animals were kept under laboratory conditions and were sacrificed 24 hr after the last injection. Ovaries were dissected out free from fat and immediately were frozen at -20°C in a cryostat (IEC). Frozen, air-dried sections were taken at 16 μ m thickness. Δ^5 3 β - and 17 β -hydroxysteroid dehydrogenases (HSDHs) were localized as described by Baillie *et al*⁵. Parallel cryostat sections were incubated for glucose-6-phosphate dehydrogenase (G-6-PDH) and NADH-diaphorase as described by Pearse⁶. Sudan Black B was employed to localize lipids⁶. The pattern of distribution and intensity of enzyme activity were visually assessed on a 4-point scale ranging from nil (-) to intense (+++). (table 1).

In the control group, HSDHs activity was localized in the theca interna and granulosa cells of the developing follicles, corpus luteum, atretic follicles, ovarian stroma of the ovary (figure 1). The presence of NADH-diaphorase and G-6-PDH was also noted which showed intense reaction in the above sites (table 1). Lipid deposition was observed to be intense in ovarian stroma and moderate in other areas (figure 2). But in the RMI-treated ovary, HSDHs activity was very faint in all the above identified sites of steroidogenic activity (figure 3). NADH-diaphorase and G-6-PDH were moderately active in these regions (table 1). However, intense lipid accumulation was detected in all the regions of the ovary (figure 4).

The histochemical detection of HSDHs indicates the probable sites of steroidogenesis⁵. Likewise the occurrence of G-6-PDH activity in the cells which contain HSDHs provides evidence, though indirect, of the steroidogenic potentiality of these cells, since G-6-PDH is known to provide the NADPH that is needed for hydroxylation during steroidogenesis⁷. Further, the presence of the diaphorase is essential in the localization of HSDHs, as the formazan is deposited by the action of this enzyme at the site of activity. Therefore,