ROLE OF SURFACE WAX OF CHILLI FRUITS IN INDUCING RESISTANCE TO FRUIT ROT PATHOGENS *COLLETOTRICHUM CAPSICI* AND *HELMINTHOSPORIUM ROSTRATUM*

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FRUIT rot of chilli (*Capsicum annuum*) is one of the important diseases that affects red ripe fruits. The pathogens *Colletotrichum capsici* and *Helminthosporium rostratum* were isolated from the majority of fruit samples showing fruit rot. The role of surface wax in the resistance of green fruits to fungal infection was studied. Surface wax was found to confer resistance to *Gloeosporium limetticola* in *Citrus aurantifolia*[1,2] and to *Erysiphe polygoni* f. sp. *hordei* in barley[3].

Healthy fruits of three different ages, viz. 5, 15 and 25 days, free from visible injury, and uncontaminated by spray chemicals were taken from plants. Surface wax was extracted from 5 g of fruit skin and estimated following the procedure of Martin and Batt[3]. The fruit skins were dipped in ether and gently agitated for 15–30 sec. This was repeated in four successive beakers containing ether. The ether extracts were made up to 25 ml. The volume of ether extract was reduced to 10 ml and the wax present in the ether extract was precipitated by adding excess of acetone. The solvents were then allowed to evaporate and the precipitate dried. The difference between the final and initial weight of the beakers gave the wax content of fruits. It was expressed as mg/g fresh weight and mg/g dry weight. Seven samples were used for each age group.

The surface wax extracted from the fruits was coated on a glass slide by a method slightly modified from Blakeman and Sztejnberg[5]. The wax (2 mg) from each fruit sample was dissolved in 10 ml of acetone and the solution was covered with a plastic cover. A wick of cotton thread was placed in contact with the wax solution. The other end of the wick was passed through a glass tube placed over the plastic cover and then allowed to touch a cavity slide. The wax solution rose through the wick and got deposited uniformly inside the cavity slide. This process was allowed for 48 h and the cavity slide was removed and placed in a ventilated container for seven days. To this cavity slide, 0.1 ml of spore suspension ($\sim 10^6$/ml) of *C. capsici* and *H. rostratum* containing the wetting agent Tween-20 was added. The slide was incubated in a moist chamber for 8 h and the number of germinating spores was counted. One hundred spores were examined per sample and four samples were used for the study. Spores placed in a drop of water served as control.

Entry of fungi into fruits of different ages was studied by the method of Shipton and Brown[6]. Chilli fruits of different ages, viz. 5, 15 and 25 days, were surface-sterilized with 0.1% mercuric chloride, washed repeatedly in sterile water, sprayed with spore suspension ($\sim 10^6$/ml), and incubated in a moist chamber for 48 h. The skin of the fruits was then cut into bits and immersed immediately in 10–15 ml of alcoholic cotton blue. They were boiled for 90 sec and then allowed to remain in the stain for 48 h at room temperature (28 ± 2°C). After rinsing with water, they were placed in chloral hydrate solution for 30 min, mounted in 50% glycerine, and examined under an oil immersion lens for conidial germination and entry.

Surface wax content of chilli fruits of different ages is presented in table 1. An interesting trend was noticed when the quantity of surface wax was expressed on the basis of two factors. While there was no significant difference between fruits of the three ages when wax content was expressed on fresh weight basis, it was highest in 5-day-old fruits when expressed on the basis of the dry weight of the fruits. In the experiment to study the possible role of wax in inhibiting spore germination, no significant difference was seen in the germination of spores in the presence of surface wax extracted from fruits of different ages (table 2). The tissue clearing technique revealed that the germination of spores and formation of appressoria were noted in fruits of all the ages. However, when sections were examined, entry of fungus was seen only in 25-day-old red ripe fruits.

Table 1 *Surface wax content of chilli fruits at different ages*

Age of fruits (days)	Surface wax	
	(mg/g fresh weight)	(mg/g dry weight)
5	2.90	21.84
15	3.00	21.74
25	3.40	15.40
C.D. ($P=0.05$)	NS	2.99

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Table 2 *Effect of surface wax on germination of spores of Colletotrichum capsici and Helminthosporium rostratum*

Treatment	*C. capsici* (% germination)	*H. rostratum* (% germination)
5-Day old fruit wax	93.63 (80.02)	96.63 (79.73)
15-Day-old fruit wax	94.50 (76.73)	95.00 (77.77)
25-Day old fruit wax	96.25 (79.57)	95.75 (79.74)
Control (sterile water)	99.13 (87.27)	99.76 (88.57)

Figures in parentheses are transformed values.

	C.D. ($P=0.05$)
Fungus	NS
Treatment	4.55
Fungus × treatment	NS

In the present study, the amount of wax was higher in resistant green fruits than in susceptible ripe fruits, when expressed on dry weight basis. The chief components of cuticle are cutin and wax. Cutin forms the framework of the cuticular membrane which carries the wax on its surface and embedded within it[7]. A difference in quantity of wax between susceptible and resistant tissues has been reported in leaves of *Citrus aurantifolia* with reference to infection by *Gloeosporium limetticola*[1,2].

Dickinson[8] discussed the possibility that the waxy surface may present the first barrier by repelling the water film required by the pathogen for germination. Nutmen and Roberts[9] ascribed differences in susceptibility of coffee varieties to berry disease caused by *Colletotrichum coffeanum* to some physical and chemical differences in the cuticle, which made penetration of the resistant variety more difficult.

The possible mechanisms of interrupting infection are: (i) thickness of natural waxy layer in barley against *Erysiphe polygoni* f. sp. *hordei*[3], (ii) repelling of film of water on the leaf surface in sorghum against *Peronosclerospora sorghi*[10], and (iii) chemical substance in the cuticle of chrysanthemum against *Botrytis cinerca*[11]. After a critical examination of the role of wax in disease resistance, Royle[12] suggested that there was some evidence that cuticle provided protection against pathogens.

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MANGIFERINE-INDUCED CHROMOSOME ABERRATION IN ROOT-TIP CELLS OF *SOLANUM INCANUM* L.

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ROOT-TIPS from mangiferine-treated seeds of *Solanum incanum* showed several kinds of chromosome aberrations. The frequency of aberration increased with concentration of mangiferine. These observations are suggestive of its mutagenic property. The correlation coefficient between the aberrations and concentration is significant at the 1% level.

Mangiferine, a naturally occurring glucosylxanthone, is widely distributed in higher plants[1] such as in the members of the families Gentianaceae[2] and Anacardiaceae[3]. It has been found to work as an antimitotic and antifertility agent in animals, particularly mammals[4,5]. These effects had not been known in plants. If this drug can cause antifertility in plants, it may be of use in plant breeding programmes, specially in those cases where fruits and seeds are not required. The present paper reports some initial observations on mitotic chromosome