GENETICS

Antimutagenic Effect against Colchicine of Kinetin in Black Currant Seedlings

It is generally known that the juvenility of plant organs can be prolonged by treatment with kinetin; moreover, senescent tissues can be rejuvenated¹, and several authors have shown increased mutability of older tissues². Treatment of plant leaves by kinetin appreciably inhibits the multiplication of viruses^{3,4}, although the virus contains RNA. After incorporation into the host cells, the virus may cause information disturbances by inducing the synthesis of foreign RNA. In this interpretation, the changes brought about by the virus are in the nature of mutations.

On this basis we have supposed that kinetin has an antimutagenic activity, or, in other words, that it takes part in the protection of the information-system controlling heredity. In order to test this idea, we attempted to neutralize the genome mutation-inducing effect of colchicine by pre-treatment of the plant with kinetin.

We chose black currant ('Altayskaya Desernaya' × 'Boskoop Giant' F_1 seedlings) as a test plant. The treatment of the shoots of the seedlings by kinetin (50 p.p.m.) or by kinetin (50 p.p.m.) + colchicine (0.8 per cent) was started at the 3-4 leaf age in the greenhouse, and was continued for 24 days, repeating the treatment daily. Following this, a solution of colchicine (0.8 per cent) and gibberellic acid (GA; 50 p.p.m.) was applied to the shoots of the plants for a period of 21 days, twice a day using the 'cotton cup' method. Colchicine was added to gibberellic acid because in earlier experiments we obtained a much higher percentage of genome mutation by applying this technique than by the use of the classical colchicine treatment⁵. Evaluation was made by the 'squash' technique, using fixed shoots 5 days after finishing the treatment.

Table 1. ANTAGONIZING EFFECT OF PRETREATMENT BY KINETIN ON THE GENOME MUTATION OF BLACK CURRANT SEEDLINGS INDUCED BY COLCHICINE

Pretreatment	Mutagenic treatment	No. plants	Mixoploids No.	%
—	Colchicine + GA	44	12	27.3
Kinetin	Colchicine + GA	55	0	0.0
Kinetin + colchicine	Colchicine + GA	48	13	27.0

The pre-treatment with kinetin provided a complete protection against the mutagenic treatment, without influencing the growth of the seedlings. It seems, however, that a longer time is needed for the realization or the stabilizing effect of kinetin, since colchicine applied together with kinetin—as a pre-treatment—has inhibited the stabilization.

In the past, a number of publications have reported the natural occurrence of kinetin-like substances^{6,7}. Thus, it is possible that these substances play a part in stabilizing the transfer of genetic information. This view is supported by the results of Matthews⁸, who has shown the presence of RNA-attached purine derivatives, and attributed a stabilizing role to these compounds.

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mental conditions were unchanged except that the plants were grown (during the summer) under net shades with natural lighting, until 24 h before inoculation when they were placed in the dark at 25° C; the plants then received a light intensity of less than 10 ft.-candles for 1.5 h before they were inoculated. Lesions were counted immediately after leaves were gathered. For U_1 and U_2 the patterns for the numbers of macroscopic and starch lesions which developed at 20°, 25° and 30° C and 640 ft.-candles (Table 1) resembled those at 4,000 ft.-candles (Fig. 1); for VM, no macroscopic lesions were formed and starch lesions were not identified consistently. In these experiments lesions of all strains were smaller and contained less starch than those in the previous experiments. This made lesions of U_2 and VM more difficult to identify than in the previous experiments, especially when the inoculated plants were incubated at 30° C. In a previous paper, it was shown that the ratio of the numbers of macroscopic lesions to the numbers of starch lesions could be used as a measure of lesion size². In the present experiments, in plants inoculated with U_1 and U_2 and incubated at 25° C and 640 ft.-candles, this ratio was 36.0 per cent and 0.2 per cent, respectively, whereas in plants incubated at 25° C and 4,000 ft.-candles it was 68.4 per cent and 4.7 per cent respectively. This difference between the size of lesions in the two sets of experiments is considered to be due primarily to the light intensity used for the incubation of inoculated plants. It may be due in part to the light intensity the plants received immediately before inoculation.

Table 1. EFFECT OF TEMPERATURE AND LOW LIGHT INTENSITY (640 FT.-CANDLES) ON NUMBERS OF LESIONS FORMED BY STRAINS OF TMV ON PINTO BEAN LEAVES*

Virus strain	Class of lesions	Temperature of incubation (°C)		
		20	25	30
U_1 †	Macroscopic	100	230	327
	Starch	263	699	747
U_2 ‡	Macroscopic	100	2	0
	Starch	693	1,160	995
VM‡	Macroscopic	0	0	0
	Starch	§	§	§

* Numbers of lesions were adjusted in the same way as those in Fig. 1.

† Mean data for 2 experiments.

‡ Mean data for 3 experiments.

§ Occasional lesions recognized.

The experiments show that the patterns of number and size of lesions, in relation to post-inoculation temperature, vary with different virus strains and that the size of lesions is affected by light intensity. In several investigations made to examine the phenomena of interference and heat activation, U_2 and VM were not observed to form lesions on Pinto bean plants given environmental conditions, suitable for the development of lesions of U_1 (refs. 6-9). The temperature used for the incubation of inoculated plants was 30° C and the light intensity was 400 or 500 ft.-candles. The present results indicate that although these conditions are favourable for the development and identification of lesions of U_1 , they are relatively unfavourable for the development and identification of lesions of U_2 and VM.

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