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Reduction of the Crop Plant Allergenicity Due to Soil Treatment with Bacillus oligonitrophilus *KU-1 Strain*

Key words. Food allergy, crop plants, *Bacillus oligonitrophilus* KU-1, soil, genome rejuvenation.

1. INTRODUCTION

Environmental contamination with agricultural chemicals, heavy metals and other pollutants results in the plant stress and synthesis of various defense molecules (Thi and De Blic [2005]). These plant-synthesized substances may be active allergens. Taking into consideration the enhancement of food allergy in humans all over the world (Uguz *et al.* [2005]), development of methods for reduction of plant allergenicity remains an actual problem.

Here we present a simple method for attenuation of the plant allergenic potency using the soil treatment with silicate-breaking bacteria (*Bacillus oligonitrophilus* KU-1).

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2. MATERIALS AND METHODS

Preparation of bacterial culture. *B. oligonitrophilus* KU-1 strain was isolated from soil of Kazan, Russia. Identification of *B. oligonitrophilus* KU-1 strain was made as has been described by Krasilnikov (Krasilnikov [1949]). The isolated bacteria were cultivated in modified liquid medium of Alexandrov (Alexandrov and Zak [1950]) with potassium (g/L: Na_2HPO_4 – 10, $(NH_4)_2SO_4$ – 2, $MgSO_4$ – 0.5, SiO_2 – 0.15, $CaCO_3$ – 0.05, pH 8.0) at 20 °C without shaking during 2 days.

Soil treatment. Stationary phase *B. oligonitrophilus* KU-1 culture ($1.0x10^9$ cells per mL) was used for soil treatment. For perfusion of 1 square meter, we used 1 L of the bacterial culture. Soil enhancement with *B. oligonitrophilus* KU-1 was performed weekly during May and June. Various crop cultures (strawberry, apple, tomato and beetroot) were grown on the treated soil. In control, the same crops were grown on non-treated soil. Other environmental conditions (soil constitution, irrigation regime, light regime, etc.) were identical in the control and experiment.

Estimation of allergenicity. The following allergic tests were used: oral provocation test (OPT), skin incision test (SIT), and skin prick test (SPT). Reaction of the mast cell destruction (RMCD test) in rats was also investigated. The estimation of allergenicity was performed in two steps. Initially, patients were studied using the usual allergens of the same crop plants. Patients with the revealed hypersensitivity reaction were then selected for assessment of hypersensitivity to crops grown on the *B. oligonitrophilus* KU-1-treated soil.

Statistics. Data were analyzed using paired Student *t* test. A p value <0.05 was considered to indicate significance.

3. RESULTS AND DISCUSSION

The results of various allergy tests are given in Table 1. Significant reductions of allergenic potency were detected as follows: in OPT (strawberry, tomato and beetroot), in SIT (strawberry, apple and tomato), in SPT (all crops), in RMCD test (strawberry and apple).

Table 1 – Values of allergy tests in patients with food allergy

Type of test	Control values			Experimental values			Value of *t*-test
	Number of patients	Number of hypersensitive patients	Mean± SD	Number of patients	Number of hypersensitive patients	Mean± SD	
	strawberry						
SIT	136	136	100±0	133	98	73.7±3.8	6.8*
SPT	137	120	87.6±2.8	136	89	65.4±4.1	4.5*
RMCD	137	50	36.5±4.1	123	14	11.4±2.9	5*
OPT	137	42	30.7±3.9	135	13	9.6±2.5	4.5*
	apple						
SIT	30	28	93.3±4.6	30	12	40±8.9	5.3*
SPT	30	22	73.3±8.1	30	11	36.7±8.8	3.1*
RMCD	24	18	75±8.8	25	10	40±9.8	2.7*
OPT	30	14	46.7±9.1	30	8	26.7±8.9	1.3 (NS)
	tomato						
SIT	54	43	79.6±5.5	54	31	57.4±6.7	2.6*
SPT	54	43	79.6±5.5	54	26	48.2±6.8	3.6*
RMCD	54	18	33.3±6.4	52	14	26.9±6.1	0.7 (NS)
OPT	54	26	48.2±6.8	54	9	16.7±5.1	3.7*
	beetroot						
SIT	154	72	46.8±4	146	73	50±4.1	0.6 (NS)
SPT	154	89	57.8±3.9	145	62	42.8±4.1	2.6*
RMCD	154	89	57.8±3.9	143	69	48.3±4.2	1.6 (NS)
OPT	154	91	59.1±4	149	7	4.7±1.7	12.6*

Abbreviations. OPT=oral provocation test, SIT=skin incision test, SPT=skin prick test, RMCD=reaction of the mast cell destruction, NS=non significant, SD=standard deviation.

We consider that reduction of allergenicity might be connected with lowering of allergen expression. Our suggestion is confirmed, firstly, by the fact that blood samples showed more reduced values of RMCD test for strawberry and apple grown on the *B. oligonitrophilus* KU-1-treated soil, and, secondly, by more pronounced reduction of allergenicity in OPT in comparison with SIT and SPT.

We are not aimed at determining what kind of plant allergens reduced due to treatment of soil with *B. oligonitrophilus* KU-1. We would like to suggest an original hypothesis of genome rejuvenation for alternative explanation of the phenomenon observed. Namely, we suggest that decrease in the vitality of plant organism is the possible reason for synthesis of allergens. In this connection, promotion of survivability is the necessary condition for reduction of allergenicity. Bioavailable silicon released by *B. oligonitrophilus* KU-1 is possibly the essential element for promotion of plant survivability. We showed previously that *B. oligonitrophilus* KU-1 (donor of bioavailable silicon) being administered orally reduced significantly malignant growth in patients with cancer disease due to stimulation of vitality (Malkov *et al.* [2005]).

Anyway, soil treatment with *B. oligonitrophilus* KU-1 is a simple and inexpensive method for reduction of allergenic capacity of crops.

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