CHAPTER 14

GENETIC ASPECTS OF POLLUTANT ACCUMULATION IN PLANTS

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Abstract: The object of our research was to study interspecies and intervariety variability of pollutant (nitrates, heavy metals, radionuclides) accumulation in productive parts of vegetable crops, as well as the character of inheritance of nitrate and heavy metal accumulation in tomato fruits in the open ground. We investigated the genetical basis of cadmium, lead and nitrate accumulation using the diallel analysis method. Genotype variation between tomato varieties and hybrids in diallel crosses in cadmium (12.8-15.5-fold) and lead accumulation (7.5–14.1-fold) was established. It gives the possibility to select tomato varieties for growing in the contaminated area, as well as to use them in breeding for development of new varieties and hybrids with minimum accumulation of pollutants in fruits. An independent type of inheritance of cadmium and lead accumulation was revealed. Genotype variation between tomato varieties and hybrids in diallel crosses in nitrate accumulation (3.8-8.3fold) was revealed. The general type of heavy metal and nitrate accumulation inheritance in tomato fruits - overdominance towards reduction of pollutant value. To make use of heterosis breeding as a method of development of high-yielding hybrids of tomato for open ground with minimal accumulation of pollutant is proposed. Study of uptake of ¹³⁷Cs and ⁹⁰Sr in vegetables in Gomel region (Chernobyl zone) revealed the interspecies genetic variation in 137 Cs (tomato – 3.1; cabbage –3.3; carrot – 3.0; onion – 0.8-fold) and 90 Sr accumulation (tomato -1.8; cabbage -2.6; carrot -1.5; onion -2.3-fold). It gives a possibility to select the genotypes with low content of radionuclides.

Keywords: nitrates; heavy metals; radionuclides; vegetable crops; interspecies genetic variation; hybrids; diallel crosses

Introduction

At present the quality of farm produce is determined not only by the content of useful substances (proteins, fats, carbohydrates, vitamins, etc.), but also by the pollutant accumulation (nitrates, heavy metals, radionuclides, pesticides, etc.). All high nutrition value of any farm produce can become useless if it contains some toxic substances exceeding hygienic norms.

The problem of pollution is connected with anthropogenic contamination of agrolandscape (industry, transport, agriculture) and with Chernobyl catastrophe. In Belarus about 20% of the territory is contaminated with radionuclides. Considerable regions of the Ukraine and Russia are contaminated as well. Vegetables can actively accumulate some pollutants in the productive part. Problems of pollutants in vegetable growing in Belarus are complicated by the fact that more than 80% of vegetables are grown on households including those situated in pollutant contaminated areas.

The process of pollutant accumulation in farm produce depends on three main factors (Kilchevsky and Khotylyova, 1997): (i) genetical (crop and variety characters, deterring the input, transport, accumulation and detoxication of the pollutant); (ii) environmental (proximity to the source of pollution and the intensity of pollution, abiotic and biotic factors of environment, landscape etc); (iii) agrotechnical (doses and terms of fertilizer and pesticide application, control of pollutant intake by plants from the soil using agrotechnical methods, etc.).

Andryushchenko (1981), Gamzikova (1992) and Kilchevsky and Khotylyova (1997) think that the most radical and the cheapest way of lowering pollutant accumulation in produce is plant breeding. This way is possible in determining interspecies and intervariety genotype variability in pollutant accumulation, finding out their genetic determination, donors with minimum pollutant accumulation, the development of breeding strategy in these traits.

So the object of our research is to study interspecies and intervariety variability of pollutant accumulation in productive parts of vegetable crops, as well as the character of inheritance of nitrate and heavy metal accumulation in tomato fruits in the open ground.

Materials and Methods

NITRATES

To study nitrate content inheritance in tomato fruits by diallel analysis method we tested in 1991–1992 in open ground in Gorky Mogilev region 28 hybrid tomato combinations between 8 initial forms using two agricultural back-grounds of mineral food (normal and higher than normal). Parent material was *Talalihin*(1), *Dohodny*(2), P-7(3), *Beta*(4), *Sub-arctic mini*(5), *Line-7*(6), *L pimpinellifolium*(7), *Torosa*(8). The nitrate content in fruits (mg kg⁻¹)

was analysed by the potentiometric method. The combining ability of samples was evaluated in method 2 of Griffing (1956), genetic inheritance parameters – in Hayman's method (1954).

HEAVY METALS

In experiment were studied parents *Talalihin*(1), *Dohodny*(2), *Sprint*(3), *Line* -7(4), *Opus*(5), *Povarek*(6), *Radek*(7) and the hybrids between them in the open ground in 1995. During the vegetative period plants were sprayed with the salt solution of cadmium and lead in doses of 0.25 of MPC (maximum permissible concentration) in the soil. Heavy metal content was evaluated in mg kg⁻¹ of dry matter using the method of atomic absorption. The combining ability of samples was evaluated in method 2 of Griffing (1956), genetic inheritance parameters – in Hayman's method (1954).

RADIONUCLIDES

Studies were made of the five varieties of four vegetable crops (tomato, cabbage, carrot, onion) in the open ground in Bragin district, Gomel region in 1996–1998 at the density of the soil pollution of 10Ci km⁻² (¹³⁷Cs) and 1 Ci km⁻² (⁹⁰Sr). ¹³⁷Cs and ⁹⁰Sr content was determined in productive organs of vegetables by spectrometrical and radiochemical methods respectively.

Coefficient of accumulation (CA), being equal to the ratio of the specific radionuclides activity per unit plant sample (Bg|kg) to the specific soil activity (Bg|kg), was used as a parameter evaluating radionuclides migration in the soil-plant system.

Results and Discussion

NITRATES

Nitrate content in tomato varieties and hybrids on the experimental plot (control background) ranges from 14.2 to 55.0 mg kg⁻¹ in 1991, from 10.0 to 37.5 mg kg⁻¹ in 1992 (Table 1). On the higher background nitrate content in 1991 changed from 13.5 to 13.3 mg kg⁻¹, in 1992 – from 10 to 55.3 mg kg⁻¹ (Table 1). The lowest effect GCA had *L. pimpinellifolium* and Torosa, the highest – Beta, Line - 7 and P-7. The MPC of nitrate content in tomato fruits is 100 mg kg⁻¹.

Heterosis effect can be evaluated according to the level of dominance Hp (Table 2). In the majority of cases (39.3-46.4%) the effect of negative superdominance manifested itself that is heterosis towards decreasing nitrate content in fruits. There have been frequent cases of intermediate inheritance (32.1-46.4%). Positive superdominance manifests itself seldom (10.7-21.4%).

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Years	No.	1	2	3	4	5	6	7	8	Effect	Variance
	Genotypes									GCA	SCA
	1	22.9	27.4	35.6	17.4	19.7	23.3	15.5	17.7	-4.2	21.2
	2	24.6		19.3	16.4	31.1	39.0	19.5	14.7	-2.8	50.4
1991*	3			24.9	29.5	25.5	69.4	19.5	33.9	3.8	173.4
	4				113.3	22.0	31.8	16.1	16.3	13.2	751.9
	5					27.4	25.5	18.5	23.3	-2.4	6.6
	6						40.6	17.0	23.2	6.6	155.7
	7							24.2	18.6	-7.1	25.0
	8								13.5	-7.0	11.8
	1	32.7	30.3	35.7	20.3	32.3	26.8	15.3	10.0	0.3	39.9
	2	25.3		18.5	21.0	18.5	55.3	14.5	24.3	0	89.7
	3			29.2	27.8	25.8	50.7	17.7	22.8	2.4	40.5
1992*	4				49.2	27.7	25.0	18.8	20.0	2.6	75.9
	5					31.0	34.7	16.7	26.8	1.1	11.2
	6						37.8	19.8	29.5	8.4	116.1
	7							14.8	13.7	-8.7	2.3
	8								11.3	-6.3	13.0

TABLE 1. Contents of nitrates in tomato fruits at the high background of mineral nutrition

*Significantly at p < 0.05

TABLE 2. Degree of dominance at the character "nitrate content in tomato fruits"

Year	Background	Hybrids	Hp < −1	-1 < Hp < 1	Hp > 1
1991	Control	Quantity	11	10	7
		%	39.3	35.7	25.0
	High	Quantity	13	12	3
	-	%	46.4	42.9	10.7
1992	Control	Quantity	13	9	6
		%	46.4	32.1	21.4
	High	Quantity	12	13	3
		%	42.9	46.4	10.7

The average level of dominance H/D ranged from 0.95 to 2.74 (Table 3) taking into account testing medium, it also confirms the manifestation of dominance and superdominance. On the higher agricultural backgrounds the frequency of dominant genes increases because the parameters $[\sqrt{(4DH2)+F}]$ / $[\sqrt{(4DH2)-F}]$ overreach 2.17–5.24%.

Correlation between trait meaning and the sum variance and covariance is more often positive which testifies the prevalence of dominance of minimum nitrate content in fruits. Meanings of fully dominant D_{\max} and fully recessive R_{\max} parent prove it. The studied varieties differ in one group of genes.

Thus nitrate accumulation is more often inherited by heterosis type towards their minimum content in fruits. In this connection we may consider heterosis breeding for producing hybrid F_1 , as a method of decreasing nitrate accumulation in the produce.

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Background	Year		$\sqrt{H}_{\rm l}/_{\rm D}$		H_2	√4DH7 +		r		$D_{ m max}$		$R_{ m max}$		h^2	
					$4H_1$		ц							H_2	
						$\sqrt{4DH_2}$ -									
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Control	1991	1992	0.95	2.74	0.27	1.30		0.65	-0.37	14.96	18.53	34.31	13.60	0.21	0.06
					0.22	1.45									
High	1991	1992	1.10	1.08	0.12	5.24		0.98	0.89	16.87	12.96	123.13	61.02	0.69	0.90
					0.19	2.17									

TABLE 3. Parameters of Hayman at character "nitrate content in tomato fruits"

HEAVY METALS

The study of cadmium accumulation in tomato fruit revealed the difference between parents by a factor of 5.3 in 1995 and 4.1 in 1996 (Table 4).

All parents and hybrids accumulated cadmium against the contaminated backgrounds more than MPC (0.03 mg/kg). It may be a reflection of poor mechanism of cadmium detoxication in plants and a high potential danger of cadmium under air pollution of agrocenosis. The difference between hybrids in cadmium accumulation was 12.9 in 1995 and 8.6 in 1996; with a whole diallel scheme – 12.8 in 1995 and 15.5 in 1996. Varieties *Talalikhin* 186 and *Dokhodny* may be used as donors of

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2 0.13 0.36 0.13 0.52 0.60 0.32 -0.09 0.04 3 0.53 0.23 0.21 0.22 0.19 -0.12 0.04	
3 0.53 0.23 0.21 0.22 0.19 -0.12 0.04	
4 0.51 1.67 0.28 0.61 0.09 0.21	
5 0.69 0.26 0.13 0.14 0.22	
6 0.57 0.33 0.01 0.06	
7 0.042 -0.08 0.03	
Content of Cadmium, 1996 (LSD $_{05} = 0.12$)	
1 0.36 0.22 0.30 0.29 0.20 0.30 0.30 0.02 0.00	
2 0.15 0.17 0.14 0.38 0.25 0.20 -0.05 0.01	
3 0.54 0.43 0.22 0.06 0.04 0.02 0.03	
4 0.46 0.25 0.24 0.20 0.04 0.01	
5 0.55 0.09 0.09 0.02 0.03	
6 0.62 0.05 0.01 0.04	
7 0.37 -0.06 0.02	
Content of Lead, 1995 (LSD $_{05} = 0.41$)	
1 2.03 0.37 0.39 1.14 0.48 1.13 1.08 0.09 0.28	
2 1.25 0.43 1.24 0.88 0.53 0.63 -0.14 0.19	
3 2.58 0.43 0.53 0.31 1.16 0.06 0.72	
4 2.97 0.21 0.41 0.93 0.27 0.71	
5 0.89 0.34 0.46 -0.35 0.20	
6 1.74 1.88 0.02 0.45	
7 1.08 0.05 0.15	
Content of Lead, 1996 (LSD $_{05} = 0.26$)	
1 2.16 0.44 0.93 0.37 0.59 0.55 1.11 0.12 0.37	
2 2.05 0.45 1.31 1.88 0.51 0.49 0.22 0.56	
3 0.79 0.31 0.75 0.75 0.92 -0.17 0.05	
4 0.68 0.39 0.40 0.57 -0.28 0.10	
5 0.94 0.37 0.52 -0.10 0.22	
6 1.77 1.80 0.08 0.35	

TABLE 4. Content of cadmium and lead in tomato fruits of diallel crosses (mg/kg)

*Talalikhin 186 (1), Dokhodny (2), Sprint (3), Line 7 (4), Opus (5), Povarek (6), Radek (7).

low cadmium accumulation. *Opus* and *Povarek* had the highest content of cadmium.

Parents differed in accumulation of lead against the contaminated background by a factor of 3.3 (1995) and 3.2 (1996), hybrids – 9.0 and 6.1, all genotypes – 14.1 and 7.0 respectively. Variety *Opus* can be used as a donor of minimum accumulation of lead. Varieties *Talalikhin* 186 and *Povarek* accumulated a maximum level of lead in fruits.

Analysis of dominance direction for the studied traits showed that a general type of inheritance of cadmium and lead accumulation in 1995–1996 was an overdominance (negative heterosis) to the decrease of heavy metal accumulation in fruits (Table 5).

Hayman's parameters supported our findings about type of inheritance of heavy metal accumulation in tomato fruits (Table 6).

The average degree of dominance $\sqrt{(H_i/D)}$ was more than one that showed an exhibition of overdominance.

The relationship of dominant and recessive genes was from 1.72 to 1.97 indicating the prevalence of dominant alleles. Correlation coefficient r between trait value and sum of variances and covariances moved to 1. It shows that maximum trait value will be in the genotypes with full recessive

Type of contamination	Parameters	$H_{\rm p} > 1$	$-1 \le H_p \le 1$	$H_{\rm p} < -1$
1995				
Cadmium	Quantity genotypes	6	6	9
	%	28.6	28.6	42.8
Lead	Quantity genotypes	1	1	19
	%	4.8	4.8	90.4
1996				
Cadmium	Quantity genotypes	0	5	16
	%	0	23.8	76.2
Lead	Quantity genotypes	1	4	16
	%	4.8	19	76.2

TABLE 5. Degree of dominance Hp at the Cadmium and Lead accumulation in tomato fruits

TABLE 6. Hayman parameters at the Cadmium and Lead accumulation in tomato fruits

				$\sqrt{4DH_1+F}$				
Trait	Years	$\sqrt{H_1/D}$	$H_{2}/4H_{1}$	$\sqrt{4}DH_1 - F$	r	$D_{\rm max}$	R _{max}	$h^2/4H_1$
Cadmium	1995	1.82	0.21	1.72	0.82	0.24	0.98	0.08
accumulation	1996	1.84	0.21	1.97	0.66	0.25	0.72	1.49
Lead	1995	1.34	0.22	1.96	0.98	0.902	3.09	1.85
accumulation	1996	1.15	0.21	1.85	0.96	0.72	2.12	1.96

allele. This information was supported by the value of full dominant D_{max} and full recessive R_{max} parents.

Variation in cadmium and lead accumulation was controlled by one-two loci because the parameter $h^2/4H_1$ changed from 0.08 to 1.96.

Analysis of the adaptive ability and ecological stability of genotypes for traits of heavy metal accumulation was evaluated by parameter b_i (regression coefficient; Eberhart and Russell, 1966) and parameter Sg_i (relative genotype stability; Kilchevsky and Khotylyova, 1997).

It was shown that tomato hybrids on the average accumulate less cadmium and lead in fruits in comparison with parents and differ in the greater stability of accumulation traits (values of b_i and Sg_i were lower).

RADIONUCLIDES

In experiment using four vegetable crops, we established differences between varieties in ¹³⁷Cs coefficient of accumulation in productive organs of all crops in all years of fruits (Table 7). Maximum differences between varieties were:

			Years	5	
Crops	Varieties	1996	1997	1998	Average
Tomato	Sprint	0.0047	0.0045	0.0054	0.0049
	Peramoga 165	0.0093	0.0067	0.0071	0.0077
	Kalinka	0.0129	0.0108	0.0078	0.0105
	Dohodny	0.0158	0.0153	0.0142	0.0151
	Rusha	0.0089	0.0078	0.0095	0.0087
	LSD	0.0037	0.0025	0.0030	0.0025
Cabbage	¹ 1 Gribovsky 147	0.0222	0.0263	0.0106	0.0197
	Belorusskaya 85	0.0073	0.0045	0.0060	0.0060
	Rusinovka	0.0207	0.0149	0.0127	0.0161
	Amager 611	0.0127	0.0123	0.0078	0.0109
	Turkiz	0.0192	0.0235	0.0151	0.0193
	LSD	0.0047	0.0068	0.0049	0.0068
Carrot	Nantskaya	0.0067	0.0047	0.0065	0.0060
	Losinoostrovskaya	0.0084	0.0134	0.0117	0.0112
	Vitaminnaya	0.0162	0.0101	0.0114	0.0126
	NIIOH 336	0.0192	0.0190	0.0151	0.0178
	Shantene	0.0222	0.0179	0.0142	0.0181
	LSD	0.0035	0.0029	0.0072	0.0051
Onion	Vetraz	0.0246	0.0214	0.0170	0.0210
	Shutgarten rizen	0.0257	0.0194	0.0173	0.0208
	Iantarny	0.0258	0.0235	0.0218	0.0237
	Klivitcky ruzhovy	0.0263	0.0201	0.0158	0.0207
	Skvirsky	0.0231	0.0170	0.0192	0.0198
	LSD	0.0075	0.0039	0.0056	0.0034

TABLE 7. Coefficient of ¹³⁷Cs accumulation in the varieties of vegetables

tomato -3.1, cabbage -3.3, carrot -3, onion -0.8 times. Tomato varieties which accumulated least of all of ¹³⁷Cs were Sprint; carrot - Nantskaya, cabbage - Belarusskaya 85, Amager 611.

Marked differences between varieties in coefficient of ⁹⁰Sr accumulation in productive organs (Table 8) were found. Maximum difference between varieties was: tomato-1.8; cabbage-2.6; carrot-1.5; onion-2.3 times.

In their ability to accumulate ¹³⁷Cs vegetables can be placed in the order of priority in the following way: onion, cabbage, carrot, tomato. The same line can be made for vegetables in their ability to accumulate ⁹⁰Sr: onion, carrot, cabbage, tomato.

Revealed differences in radionuclide accumulation in varieties allow selecting varieties for lower radionuclide intake together with vegetables 2–3 times. The presence of genetic variability makes it possible to carry out breeding aimed at lowering radionuclide accumulation in vegetables.

			Years		
Crops	Varieties	1996	1997	1998	Average
Tomato	Sprint	0.0302	0.0191	0.0204	0.0232
	Peramoga 165	0.0596	0.0369	0.0304	0.0423
	Kalinka	0.0498	0.0229	0.0227	0.0318
	Dohodny	0.0445	0.0160	0.0199	0.0268
	Rusha	0.0325	0.0195	0.0176	0.0232
	LSD	0.0179	0.0062	0.0068	0.0094
Cabbage	¹ 1 Gribovsky 147	0.0901	0.0934	0.0756	0.0863
	Belorusskaya 85	0.1942	0.2017	0.1931	0.1963
	Rusinovka	0.1971	0.1896	0.1805	0.1891
	Amager 611	0.1621	0.1776	0.1700	0.1699
	Turkiz	0.2469	0.02200	0.2120	0.2263
	LSD	0.0283	0.0231	0.0284	0.0181
Carrot	Nantskaya	0.1959	0.1612	0.1616	0.1729
	Losinoostrovskaya	0.2171	0.1990	0.2036	0.2066
	Vitaminnaya	0.2093	0.1759	0.1721	0.1858
	NIIOH 336	0.2779	0.2305	0.2267	0.2450
	Shantene	0.2903	0.2580	0.2435	0.2640
	LSD	0.0477	0.0276	0.0234	0.0148
Onion	Vetraz	0.4786	0.4098	0.4062	0.4315
	Shutgarten rizen	0.7925	1.1206	1.0633	0.9921
	Lantarny	0.6359	0.5345	0.5210	0.5638
	Klivitcky ruzhovy	0.5525	0.5506	0.5456	0.5496
	Skvirsky	0.6529	0.5510	0.5420	0.5820
	LSD	0.0953	0.0759	0.0931	0.1876

TABLE 8. Coefficient of ⁹⁰Sr accumulation in the varieties of vegetables

Conclusion

Analysis of literature and results of carried out experiments show that intervariety variability in pollutant accumulation in vegetables are quite enough to single out genotypes lowering their intake with food 2–5 times. Analysis of inheritance of nitrate and heavy metal accumulation in tomato fruits shows that the main type of inheritance in conditions of produce contamination is superdominance towards decrease of pollutant content. It allows us to admit that heterosis breeding is the method of lowering pollutant accumulation in tomatoes.

In our opinion (Kilchevsky and Khotylyova, 1997) general strategy of breeding towards lowering pollutant accumulation in agricultural produce should include three main stages:

- 1. Evaluation of the initial material corresponding to a number of useful traits and pollutant accumulation in contaminated areas, selection of initial forms for hybridization which correspond to tasks of breeding.
- 2. Selection in early generations (F_2-F_5) according to the traits of usefulness as well as to traits of correlation associated with accumulation of pollutants in contaminated areas.
- 3. Carrying out of competitive and ecological tests on the polluted and clean territory to evaluate selection results.

High price of carrying out analysis of pollutant content will hardly permit us to control their accumulation in many samples in F_2 - F_5 . Hence it is very important to study correlation of these traits with other morphobiological and physiological traits for indirect selection. Breeding aimed at lowering pollutant accumulation in agricultural produce is the most radical, cheap and economically based means of lowering pollutant intake with food.

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