

## FRUIT PLANT ANTIOXIDANT SYSTEM ELEMENTS CHANGES AS A TEST-FACTOR OF ACID STRESS

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The pollution of air is caused basically oxides of sulfur and nitrogen, which turn in an atmosphere with formation of acid precipitation. Last can drop out for hundreds kilometers from a place of emission and damage to plants in agricultural areas, is especial fruit plants, recognized sensitive to pollution of air by gases. The plant is capable to some extent to resist to this influence by activity antioxidant system which can to break process free radicals oxidation of bimolecular. The important components of this system in plant are ascorbic acid (AA) and glutathione. However role of these matters and degree of their transformations in fruit plants tissues under influence of acid stress is insufficiently investigated. We studied changes of AA content, reduced glutathione (GSH) and total reduce activity (TRA). Stress modeled by spraying of a modeling branch of fruit-bearing peach trees by a solution of a sulfuric acid with pH = 2 during 2-3 hours. The control is no spread trees. 11 varieties of a peach on almond rootstock with the various contents AA in fruits and 9 varieties of apricot on apricot rootstock and are included in research by different reaction to acid influence investigated on external symptoms. The significant changes in the contents of the specified elements plant antioxidant system under influence of stress are established. The various reactions of peach and apricot varieties to this effect are shown. The relative parameters of a status antioxidant system are designed and on their basis the relative rating of peach and apricot varieties to this stress - factor is carried out. Most sensitive by this parameter have appeared the peach varieties of early term of maturing. Resistance of apricot trees to acid stress first of all depends on a genesis of variety. It is offered to use CRA as a test parameter for estimation of different varieties of fruit plants to acid stress.

### Introduction

Acid precipitations are one of air pollution manifestations by burning products and harmful technological ejection. Nonspecific symptoms of damaging and lasting in manifestation of toxicants' chronic dozes influence do not allow valuing the acid precipitation influence with high share of probability. The ability of the plant to resist acid stress depends on activity of the plant antioxidant system. The antioxidant system includes oxidizing-reducing ferments, glutathione, ascorbic acid (AA) etc. (1). AA and glutathione are the part of the cell protection against the effect of peroxide oxidation of lipids, they also regulate oxidizing-reduction processes (2). Glutathione protects SH-problems by keeping its reduction condition. It plays a specific part in reduction of oxidizing form of AA (3). Antioxidant system of fruit plants under acid stress is not yet investigated (4, 5). It is known only that fruit plants are sensitive or very sensitive to the air pollution by oxidants (6). It is significant to determine the degree of comparative resistance and sensitivity of the fruit plants to the acid stress under the condition of air pollution by combustion products and noxious acid waste products. It is important for using these fruit plants in commercial gardening and selection. In connection with the purpose of our research was to determine the effect of simulated acid rain (SAR) to the content of some components of peach antioxidant system and to develop diagnostic assessment of the peach varieties, stable under acid stress. To determine the degree of comparative resistance of the fruit plants to the acid stress.

### Methods

The research was carried on the group of peach varieties (*Persica vulgaris* Mill.), native variety (Nikita Botanical Gardens), USA and Hungary selection varieties, and the group of apricot varieties (*Armeniaca vulgaris* Hill) from different ecological-geographical groups of various age. The content of AA and total reducing activity (TRA) of the tissue was determined by the method off Pett modified by Prokoshev (7), the reduced glutathione (GSH) was calculated by difference. The leaves for analyses were selected at the stage of active growth of plant from the tops of annual shoots. In the course of experiment on effecting of SAR to the growth and productivity of plants, the plants were sprinkled by sulphuric acid dissolved in water distilled. The value of pH solutions is equal to 2. The frequency of sprinkling of rain is for a 3 hour period. The duration of the experiment – 1 vegetation period. Reiteration is quintuple. The research of the antioxidant system under the influence of acid stress was carried on model branches of fruit-bearing trees by the way of sprinkling by solution ( $H_2SO_4$ ) with pH

in the dose described above. The sprinkling was done every 2 weeks in the period of intensive shoots growth. The leaves were selected 3 day after the acid sprinkling. The control was the non-sprinkling trees which were under the effect of natural precipitations, the average pH of those, for the vegetation period was equal to 6,0. The reiteration of the last experiment was thrice – repeated. The data is processed by the statistics. The reliability is 5 % level of significance.

## Results

The content of AA in the leaves of the peach variety investigated in control trees was fluctuating from 0,70 to 1,51 mg/g fresh weight (FW) (fig. 1) and depended probably as on particular features of variety as its condition in the moment of exemption. The average concentration of it was 1,14 mg/g FW. The acid stress stimulated the reduction the content of AA in the leaves of all varieties, except for the variety 'Baby Gold' where it was increasing. Especially considerable reduction of AA content was found with varieties: 'Nebesniy tichochood', 'Ak-Sheftaliy', 'Pushistiy ranniy', 'Orechoviy', 'Red Haven'. It was 0,25-0,45 mg/g FW (fig. 1).

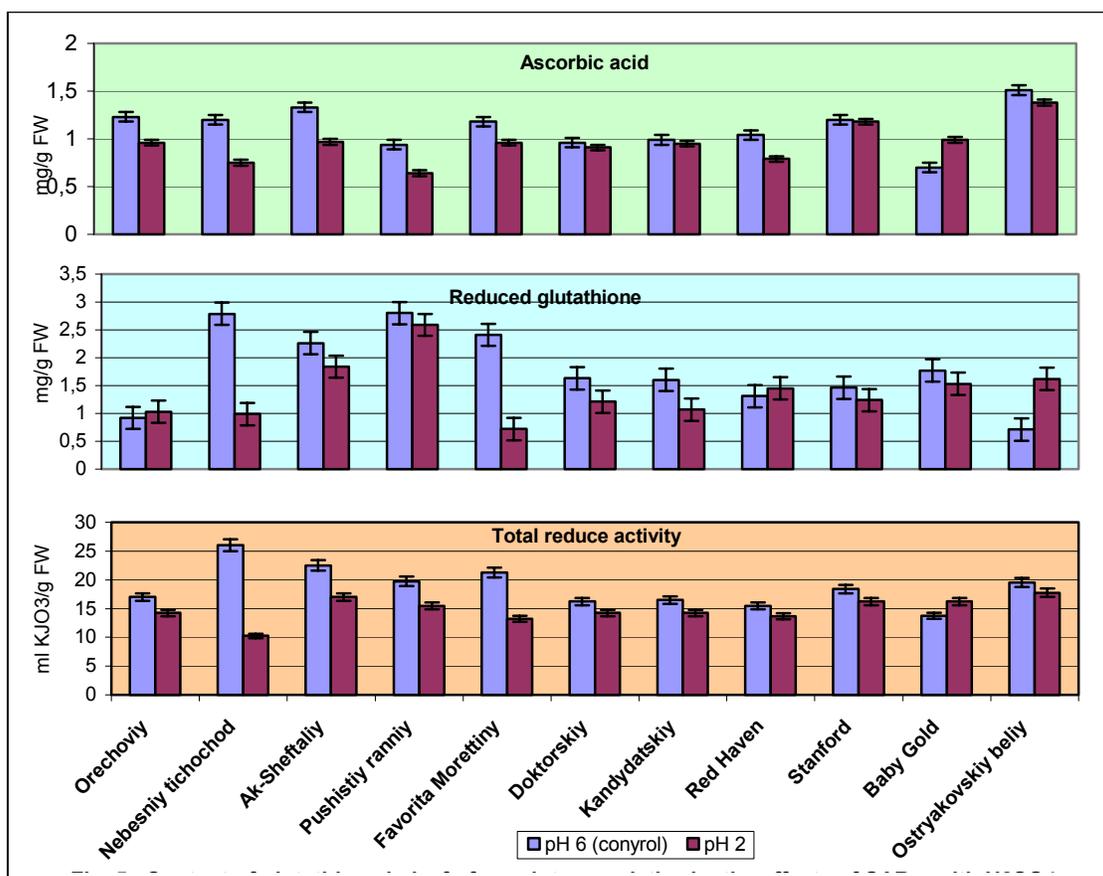


Fig. 1. The concentration of ascorbic acid, reduced GSH and total reduce activity of peach leaf tissue in condition of acid stress ( $H_2SO_4$ ), July 2001.

The concentration of GSH in peach leaves was fluctuating more – from 0,71 to 2,80 mg/g FW and it was 1,80 mg/g FW for variety. Most of varieties had content of GSH higher than AA besides the variety at earlier ripening contented it twice as much. And only variety 'Orechoviy' and 'Ostryakovskiy beliy' were contrary apposed (fig. 1). The content of GSH in leaves was found to be reduced with 8 varieties of 11 exanimete (under the conditions of acid stress). The most considerable reduction of GSH was found with the varieties of early ripening term: 'Nebesniy Tichochood' and 'Favorita Morettiny'. The other of three varieties: 'Orechoviy', 'Red Haven' and 'Ostryakovskiy beliy' had GSH increased for 0,11-0,91 mg/g FW, besides the 'Ostryakovskiy beliy' had more considerably increasing (fig. 1).

The value of TRA tissue, as integral sign of plant stability under the conditions of oxidizing stress had been found considerably reduced with the varieties of early ripening term (4,25-15,75 ml), and also

with variety 'Ak-Sheftaliu'. The variety 'Baby Gold', on the contrary, this indicator increased for 2,50 ml KJO<sub>3</sub> per gram of FW, evidently because of increasing of AA content. The other varieties investigated had TRA reduced from 1,75 to 2,75 ml/g. We calculated the degree of TRA reduction under acid stress and expressed it through the coefficient of antioxidant activity of tissue under acid stress (C aoa), which is equal to:

$$C\ aoa = \text{TRA under stress} / \text{TRA under optimal conditions} \times 100.$$

The more this coefficient is the stable antioxidant plant system under acid stress is. According to C aoa we subdivided conditionally all the peach varieties investigated on three groups of conditional stability (tabl. 1). The group of sensitive includes the varieties with C aoa up to 70 units, all of them are early ripening. Varieties 'Kandidatskiy' and 'Doctorskiy' have similar time of ripening and belong to the some dimension family: 'Laureate' x 'Sun Rise' and got to the group of low resistant with C aoa from 70 to 90. The varieties 'Ostryakovskiy belyi' and 'Baby Gold' are found out to be of middle resistant under stress, besides the 1<sup>st</sup> one increased the content of GSH, and 2<sup>nd</sup> - AA under stress.

Table 1. Comparative stability of peach varieties under acid stress

№	Variety	Term of ripening	C aoa	Group of resistance
1.	Nebesniy tichochod	3d ten days period of July	39	Sensitive
2.	Favorita Morettiny	1 <sup>st</sup> ten days period of July	62	-«-
3.	Pushistiy ranniy	2 <sup>nd</sup> ten days period of July	65	-«-
4.	Ak-Sheftaliy	2 <sup>nd</sup> ten days period of September	76	Low resistant
5.	Orechoviy	3d ten days period of July	84	-«-
6.	Kandidatskiy	1 <sup>st</sup> ten days period of August	86	-«-
7.	Doktorskiy	1 <sup>st</sup> ten days period of August	88	-«-
8.	Red Haven	2 <sup>nd</sup> ten days period of August	88	-«-
9.	Stanford	-«-	88	-«-
10.	Ostryakovskiy belyi	-«-	91	Middle resistant
11.	Baby gold	-«-	118	-«-

The concentration of AA in leaves of the apricot investigated varieties in control trees changed from 1,02 up to 1,81 mg/g FW (fig. 2). It was a little more then in the peach leaves tissue. The average concentration of it for all investigated varieties equaled 1,46 mg/g FW. It was higher then average for all apricot trees for varieties: 'Stark early Orange', 'Olymp', 'Tabu' and 'Henderson' and a little bit less for varieties 'Krimskiy amur', 'Kostinskiy' and 'Shalah'. The least concentration of ascorbat is found out for variety 'Vinosliviy' (1,02 mg/g FW). The acid stress caused decrease the ascorbat concentration in tissues of the majority investigated apricot varieties, most significant with varieties 'Stark early Orange' (0,58 mg/g FW of a blade). The concentration of this substance, on the contrary, grew, in leaves of varieties 'Krimskiy amur', 'Kostinskiy' and 'Shalah'. It was most significant in last two varieties.

The concentration of a GSH in leaves of an apricot trees changed from 0,05 up to 0,75 mg/g of a blade FW and the average on all apricot trees was 0,50 mg/g. A lot of varieties had the contents GSH close to average value in the control and only with varieties 'Stark early Orange' and 'Shalah' reached up to 0,75 and 0,71 mg/g accordingly. It was the lowest with varieties 'Tabu' and 'Henderson' (0,20 and 0,05 mg/g accordingly), fig. 2. In result of acid stress the GSH concentration could as be increased ('Stark early Orange', 'Henderson', 'Kostinskiy' and 'Shalah') and decrease. It was the most significant in hybrid varieties: 'Krimskiy medunec', 'Olymp' and 'Krimskiy amur'.

The concentration of TRA of apricot leaf tissue in the control was, as a rule, higher with the varieties of early ripening term: 'Stark early Orange', 'Olymp', 'Tabu', and 'Henderson' (fig. 2). In hybrid group varieties ('Krimskiy medunec', 'Kostinskiy'), where one of the parents was a variety 'Shalah', TRA had average value 16-17 ml KJO<sub>3</sub>/g, and variety 'Vinosliviy' has made only 12,1 ml KJO<sub>3</sub>/g of FW. The value of TRA in leaves tissues under the conditions of oxidizing stress had been found considerably reduced with the varieties of more early ripening term: 'Stark early Orange' and 'Olymp'. It was determined, basically, decreasing of ascorbat concentration by the stress (fig. 2). The varieties 'Kostinskiy' and 'Shalah' as a close relatives increased TRA of a blade tissue under stress by the significant increasing of AA and GSH concentration (fig. 2).

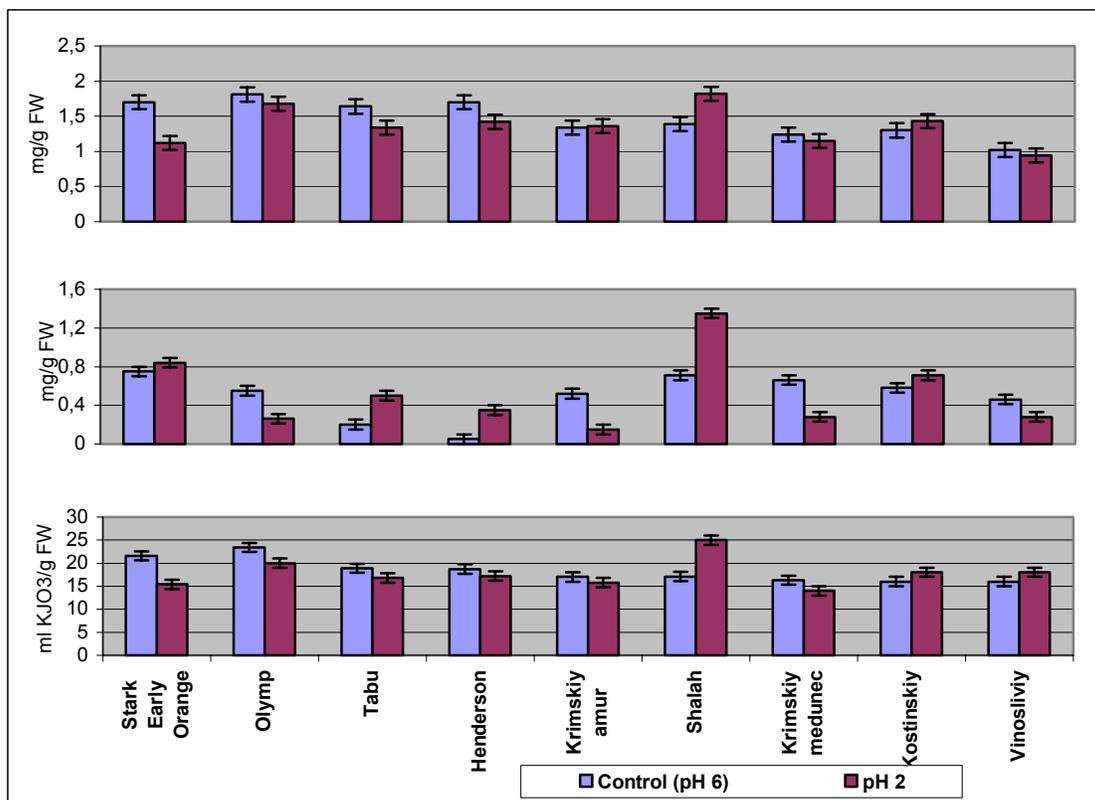


Fig. 2. The concentration of ascorbic acid, reduced GSH and total reduce activity of apricot leaf tissue in condition of acid stress ( $H_2SO_4$ ), July 2003.

As well as in a case with peach trees, we have calculated the coefficient of antioxidant activity of tissue under acid stress ( $C_1$  aoa) of the investigated apricot varieties and have arranged varieties in a growing number of relative stability to acid stress. We have divided all varieties of an apricot into 3 groups of relative stability (tab. 2).

Table 2. Comparative stability of apricot tree varieties under the acid stress.

№	Variety	Term of ripening	C aoa	Group of resistance
1.	'Stark Early Orange'	1st ten days period of July	71	Sensitive
2.	'Krimskiy medunec'	2nd ten days period of July	86	Low resistant
3.	'Olymp'	2nd ten days period of July	86	-->--
4.	'Tabu'	2-3 ten days period of July	89	-->--
5.	'Henderson'	3 ten days period of July	91	-->--
6.	'Krimskiy amur'	3 ten days period of July	93	-->--
7.	'Vinosliviy'	3 ten days period of July - 1st ten days period of August	97	-->--
8.	'Kostinskiy'	2nd ten days period of July	112	Middle resistant
9.	'Shalah'	1st ten days period of July	146	-->--

#### Discussion

The reaction of a plant to stress depends on activity of antioxidant system of plant, which is determined by many reasons among which origin of a plant, age, period of vegetation, presence of other stresses, adaptive opportunities of organism. Fruit plant as a whole are sensitive to pollution of air, but can differ on stability from each other. The way, chosen by us, of a relative rating of the large number of fruit plants varieties allows by simple means and at early stages of ontogenesis to estimate the reaction of a plant to acid stress. In limits of homogeneous group of fruit plant varieties lowest C aoa are inherent earlier ripening breeds of a peach and apricot trees. It confirms opinion that early

beginning and early finishing the vegetation or early ripening plants are more sensitive to stress (8, 9). It is known that the species of trees stable to the environmental pollution with heavy metals are able to accumulate AA in leaves (4). It is possible that in our situation, more stable varieties were accumulating as GSH ('Ostryakovskiy beliy'), as AA ('Baby Gold'). As for the other varieties which are more sensitive, the considerable part of antioxidants was spent to the reduction reactions and did not recover in post stress period (under this value of pH SAR). The varieties of apricot received from crossing of the representatives of various ecological-geographical groups, irrespective of ripening term, can inherit properties of this or that ancestor and considerably differ on stability to stress ('Kostinskiy', 'Olymp', 'Krimskiy medunec'). The variety 'Shalah' in our scale of relative stability has highest C<sub>aoa</sub> = 146. It is known, that this grade has high stability to fungous diseases (10) and to drought (11). It can be connected not only with this variety metabolism, but also with a structure of its leaf blade - larger and smooth (without rhizome, (12). Probably, the identical reaction of plants on different stresses has an identical physiological and genetic nature. Hybrids with participation of a breed 'Shalah' as the parent form reacted differently on acid stress. So, the breed 'Kostinskiy', was received from crossing varieties 'Vinosliviy' x 'Shalah' was closed to 'Shalah' for its reaction to acid stress and has received C<sub>aoa</sub> = 112. Other varieties of this hybrid family: 'Krimskiy medunec' ('Stepnjak' x 'Shalah') and 'Olymp' ('Vinosliviy' x 'Shalah') had weaker stability to stress and identical K<sub>aoa</sub> equal 86. Thus, the given test - factor will allow to establish its relative stability to acid stress on early stages of an individual development, which can directly be connected with drought-resistance, stability to fungous diseases and another biotic stresses.

#### Conclusions

1. All peach and apricot investigated varieties had different degree of antioxidant system modification under the acid stress. Some of them, mainly early ripening, had their AA, GSH and TRA reduced, others, increased.
2. It is proposed the coefficient of antioxidant tissue activity K<sub>aoa</sub> determines the degree of tissue reduce activity changing under the acid stress with comparison one in optimal conditions.
3. According to A<sub>aoa</sub> we subdivided conditionally all peach and apricot varieties investigated on three groups of conditional stability. They are sensitive, low resistance and middle resistance.
4. The coefficient of antioxidant activity may be used as a test of fruit plants sensitivity to the acid stress for the breeding process and arrangement more resistant of the plants in regions with unfavorable ecological conditions.

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