TRANSFORMATION OF THE BIOCHEMICAL COMPOSITION OF *VACCINIUM CORYMBOSUM* FRUIT DURING STORAGE

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The biochemical composition of highbush blueberry fruit of 6 cultivars with different ripening periods, introduced in Belarus, is investigated on 14 indicators thrice a month at equal intervals. The nutrient and vitamin value of the fruit gradually decreased with the increasing amount of the dry matter, during storage, due to the depletion of free organic, ascorbic and phenol carboxylic acids, soluble sugars, pectic substances, tanning agents, anthocyanin pigments, catechins and flavonols. The transformation of the biochemical composition of the fruit, during storage, is strongly dependent on weather conditions of the season, rather than on their ripening period.

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Introduction

Currently, a special scientific and practical interest in cultivating highbush blueberry (*Vaccinium corymbosum*) is the matter related to its storage, , A short period limits possibilities of their selling and supplying to the market. The prolongation of the consumer properties of blueberry fruit by freezing and further storing at low sub-zero temperatures is ensuring, as some researchers state it, relative stability of their basic physical and chemical characteristics.

The results of the comparative study of the content of anthocyanins and levels of antioxidative activity of ethanolic extracts, made of frozen and dried blueberries after 3 months of storage with the similar extract from freshly harvested berries demonstrated no noticeable differences in the first case (frozen) and significant reduction of these characteristics in the second case (dried). [1]

The second study also demonstrated no significant changes in case of six-month storage of blueberries frozen at the temperatures of –18 ºC and –35 ºC with respect to both the quantitative content of phenolic compounds, including chlorogenic acid and anthocyanins, and the qualitative composition of such compounds. [2]

The third study incorporates the results of the comparative changes in the content of dry matters, titratable acids, vitamin C, soluble sugars and bioflavonoids of the fruits of several cultivars of highbush blueberry frozen at the temperature of –25 °C after 6- and 12-month storage.[3] The study demonstrated relative stability of the biochemical composition of the berries in general against expressed cultivar-specificity of the degree of the detected changes.

However, the inevitable trade loss of fruit after defrosting is significantly promoting the storage of blueberries in refrigeration units at low positive temperatures. Studies in this direction are of current interest, which will be demonstrated in the discussion of the results of this study.

This work establishes the essential regularities of the transformation of the biochemical composition of blueberry fruit during storage depending on plant genotypes and weather conditions of the season.

Material and Methods

Material

The research was carried out at the Central Botanical Garden of the National Academy of Sciences of Belarus in 2013 and 2014. The objects of the study were highbush blueberry fruits of 6 cultivars with different ripening periods: the first year – early ripening *Collins*, mid early ripening *Hardyblue* and mid ripening *Denise Blue*; the second year – early ripening *Bluetta*, mid ripening *Bluecrop* and late ripening *Elizabeth*. Berries were picked when in the state of economic maturity and immediately sent for storage. 400 ml disposable plastic food containers for berries and fruit with lids and holes were used as the storage containers. The berries had been stored for a month in the normal gas environment at the temperature of +3 or 4 ºC and relative humidity of 40-80 %.

Methods

Comprehensive analysis of the biochemical composition of blueberry fruit was performed three times throughout the entire storage period with a 10-day interval conditionally dividing the process into 3 stages. Correspondingly, the following contents were determined in the fresh averaged samples of blueberry fruit at the end of the 1st, 2nd and 3rd ten-day periods: dry matter – in accordance with the GOST (State Standard) 28561-90,[4] ascorbic acid (Vitamin C) – by the standard indophenol method,[5] titrate acids (total acidity) – by the volumetric method,[5] hydroxycinnamic acids (in

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terms of chlorogenic acid) – by the spectrophotometric method,[6] soluble sugars – by the accelerated semi-micro method,[7] pectin substances – by the calcium pectate method,[8] the amount of anthocyanin pigments – by the method of T. Swain and W. E. Hillis[9] with a calibration curve made for the crystalline cyanidin derived from black chokeberry fruit and purified by the method of Y. G. Skorikova and E. A. Shaftan,[10] true anthocyanins and the amount of catechins (using vanillin reagent) – by the photocolorimetric method,[5,11] the amount of flavonols (in terms of rutin) – by the spectrophotometric method[5]; tannins – by the titrimetric method of Leventhal.[12] All analytical determinations were made with a three-time biological repeatability. The data were statistically processed using the Excel software programme.

Description of weather conditions

The weather conditions during the ripening of blueberry fruit varied considerably throughout the research period. Thus, the maturing of the early ripening and mid ripening breeds in the 2nd decade of July of 2013 occurred under the temperature settings close to the multiannual norm with insufficient moisture, while the ripening period for the late ripening cultivars at the end of the 3rd decade of July and the 1st decade of August was characterized by very hot and extremely dry weather. In 2014, the maturing of the early ripening breeds in the 1st week of July was characterized by the temperatures typical of this period, but with abundant precipitation which replaced the drought lasting over the last ten days of June. The maturing of the mid ripening cultivars in the 2nd decade of July was accompanied by moderate temperature with sufficient moisture. The ripening period of the late ripening cultivars in 2014 turned out to be very extreme in terms of weather conditions, namely extremely hot and dry 3rd decade of July and partly the 1st decade of August that on the contrary was characterized by the abundance of rains.

Results and Discussion

Prior to the establishment of the experiments, cultivars of fruit blueberry differed significantly in terms of the amounts of the compounds determined in the dry fruit which varied within the ranges in 2013: free organic acids: 3.18-5.70%, ascorbic acid: 332.1-428.5 mg in 100 g, hydroxycinnamic acids: 970.2-1458.3 mg in 100 g, soluble sugars: 56-63 % with the sugar-acid index equal to 9.8-19.8, pectic substances: 4.48-5.61 % including amount of hydropectin: 1.35-1.77 %, protopectin: 3.04-4.09 %, bioflavonoids: 7008.7-9760.5 mg in 100 g including anthocyanin pigments: 4682.0-6981.0 mg in 100 g with true anthocyanins in the amount: 2030.0-3330.0 mg in 100 g, leucoanthocyanins: 2652.0-3987.0 mg in 100 g, catechins: 559.0-663.0 mg in 100 g, flavonols: 1408.3-2220.5 mg in 100 g, tannins: 1.87-2.29 %, amount of the dry matter varying from 12.5 to 15.4 %. In 2014 the same ranges for fruit of model highbush blueberry cultivars were as follows: free organic acids: 3.38-4.47 %, ascorbic acid: 192.7-312.7 mg in 100 g, hydroxycinnamic acids: 563.7-1915.1 mg in 100 g, soluble sugars: 54-60 % with the sugar-acid index equal to 9.8-19.8, pectic substances: 11.6-16.5 %, bioflavonoids: 11055.2-16755.7 mg in 100 g including anthocyanin pigments: 8762.0-12956.7 mg in 100 g with true anthocyanins in the amount of 4930.0-9088.3 mg in 100 g, leucoanthocyanins: 3697.2-5256.7 mg in 100 g, catechins: 767.0-1419.2 mg in 100 g, flavonols: 1526.2-2379.8 mg in 100 g, tannins: 2.87-4.12 % with the amount of the dry matter varying from 13.3 to 18.9 %. The considerable variation in the ranges gives evidence of the pronounced genotypic differences of the nutrient and vitamin value of fruit of the studied blueberry taxa.

Based on the research held in 2013 on the degree of the transformation of the biochemical composition of blueberry fruit of the *Collins*, *Hardyblue* and *Denise Blue* cultivars after a month-long storage period, the depletion of free organic and ascorbic acids (by 15-41 % and 42-45 %, respectively), soluble sugars (by 7-17 %), pectic substances and tanning agents (by 18-20 % and 7-17 %, respectively), anthocyanin pigments and flavonols (by 20-35 % and 4-13 % respectively) with the increase in the amount of the dry matter and of the sugar-acid index (by 14-28 % and 5-40 % respectively) without any significant changes in the amount of catechins and ambivalent changes in the amount of hydroxycinnamic acids (Table 1).

Similar tendencies in the transformation of the biochemical composition of blueberry fruit were observed in the same-type experiment carried out in 2014 with *Bluetta*, *Bluecrop* and *Elizabeth* cultivars. These tendencies manifested themselves in the depletion of free organic, ascorbic and hydroxycinnamic acids (31-51 %, 29-62 % and 10-48 % respectively), soluble sugars (5-9 %), pectic substances and tanning agents (22-33 % and 10-47 % respectively), anthocyanin pigments, catechins and flavonols (15-39 %, 7-35 % and 8-26 % respectively) compared with the initial values with the increase of the amount of the dry matter and the sugar-acid index (10-19 % and 34-89 % respectively). The reported decrease in the acidity level in blueberry fruits in the process of two-month storage at 3 °C[13] corresponds to our results. The study on the impact of allyl isothiocyanate (AITC) on the activity of scavenging enzymes, the content of flavonols and the quality of *Duke* cultivar blueberry fruits in the process of storage at 10 ºC also demonstrates the decrease in the content of phenolic acids, flavonols and anthocyans.[14] The studies devoted to changes in antioxidant activity of the fruits of a number of blueberry cultivars during storage at 1 and 8 °C also show the decrease of the named parameter by the end of the month storage, [15] which is an indirect proof of drop of the bioflavonoid level. The tendency of the growth of dry matter during storage in desiccator[16] is same as in our experiment.

The revealed loss of most organic compounds characterizing the decrease of the nutrient and vitamin value of blueberry fruit during storage was caused by their usage of these organic compounds as respiratory substrates, the relative amount of which was determined by their chemical nature, plant genotype and weather conditions of the season.

Table 1. Statistically valid relative differences as compared to the initial values of active substances contained in *Vaccinium corymbosum* fruit at different storage stages, (%)

Index	**1 stage**	**2 stage**	**3 stage**	**1 stage**	**2 stage**	**3 stage**	**1 stage**	**2 stage**	**3 stage**
2013	*Collins*			*Hardyblue*			*Denise Blue*		
Dry matter	-	-	**+14.1**	-	**+16.9**	**+27.9**	-	**+4.8**	**+19.2**
Free organic acids	-	-10.7	-15.4	-9.7	-20.8	-40.6	-	-14.5	-15.9
Ascorbic acid	-	-31.3	-41.8	-	-22.7	-44.6	-9.3	-19.1	-42.0
Hydroxycinnamic acid	-	-16.0	-9.1	**+28.2**	**+10.8**	-	**+35.9**	-	**+17.0**
Soluble sugars	-	-4.1	-6.6	-5.9	-14.8	-17.0	-5.8	-7.0	-7.0
Sugar-acid index	-	**+14.3**	**+5.1**	**+19.2**	-	**+39.4**	-	**+10.0**	-9.2
Pectic substances	-9.8	-13.4	-18.5	-8.2	-19.4	-19.4	-10.7	-11.9	-17.6
True anthocyanins	-	-11.4	-16.0	-17.1	-29.7	-41.4	**+3.0**	**+13.8**	**+24.1**
Leucoanthocyanins	-8.2	-9.5	-21.8	-15.9	-19.9	-29.5	-7.7	-25.1	-16.6
Amount of anthocyanin pigments	-5.9	-10.2	-19.6	-16.5	-24.6	-35.2	-3.3	-8.2	-
Catechins	-	-	-	-4.7	-7.0	-8.1	-	-	-
Flavonols	-3.7	-4.2	-4.2	-	-14.5	-13.0	**+2.8**	**+5.9**	**+15.4**
Amount of bioflavonoids	-5.1	-8.7	-16.0	-11.2	-21.3	-28.6	-1.9	-4.8	**+3.5**
Tannins	-	-10.9	-16.6	-	-3.6	-7.3	-3.2	-	**+13.4**
2014	*Bluetta*			*Bluecrop*			*Elizabeth*		
Dry matter	-	-	**+9.8**	-	-	-	**+9.5**	**+11.1**	**+18.5**
Free organic acids	-33.3	-43.8	-47.7	-26.3	-29.0	-30.5	-18.2	-44.8	-50.9
Ascorbic acid	-	-26.0	-29.2	-43.4	-44.9	-62.4	-15.1	-34.1	-45.8
Hydroxycinnamic acid	**+10.2**	**+17.9**	-10.2	**+11.6**	**+22.6**	**+14.3**	-47.8	-43.0	-48.2
Amount of soluble sugars	-	-5.6	-8.4	-	-3.7	-4.8	-	-4.6	-7.5
Sugar-acid index	**+48.1**	**+68.4**	**+74.4**	**+32.3**	**+38.5**	**+34.2**	**+20.9**	**+72.7**	**+89.2**
Amount of pectic substances	-15.8	-30.9	-32.7	-10.3	-17.2	-21.6	-	-13.7	-23.0
True anthocyanins	**+18.6**	**+17.1**	-34.5	-18.3	-18.3	-30.6	-22.8	-12.4	-39.7
Leucoanthocyanins	-	-	-	-13.8	-23.3	**+6.2**	-37.0	-11.4	-37.9
Amount of anthocyanin pigments	**+13.9**	**+12.9**	-25.7	-16.3	-20.5	-14.5	-28.6	-12.0	-39.0
Catechins	**+22.1**	**+17.7**	-	**+13.6**	-5.9	-17.0	-26.7	-21.2	-35.3
Flavonols	**+22.7**	**+15.0**	-7.7	**+5.6**	-6.4	**+6.4**	-31.7	-13.0	-25.5
Amount of bioflavonoids	**+15.4**	**+13.4**	-22.3	-11.2	-17.5	-11.8	-28.9	-12.9	-36.8
Tannins	-3.1	**+21.6**	**+5.9**	**+2.4**	-6.2	-9.8	-24.3	-22.3	-46.6

A dash (-) means that no statistically significant differences are observed at $p < 0.05$.

In order to identify the blueberry taxon with the lowest decrease in the amount of active substances in fruit, as well as to identify the storage stage at which the best preservation of the integral level of the nutrient and vitamin value of fruit is ensured, we used our own unique methods, based on comparing the tested objects, at various stages of storage with respect to their relative scales, amplitudes and correlations of statistically valid positive and negative deviations from the initial values of the studied parameters of the fruit biochemical composition.[17] The value of the overall amplitude of the detected deviations, regardless of their signs, made it possible to estimate the significance of the differences between the tested objects and the initial values of all the studied parameters at each stage of storage, which allowed ranging them according to the decrease of the degree of these differences. The correlation of the relative scales of the scopes of positive and negative differences as compared to the initial values of active substances contained in fruit served as a criterion for the estimation of the integral level of the nutrient and vitamin value of blueberry fruit in each cultivar at specific stages of storage.

The data presented in Table 2, characterizing the direction and the significance of the transformations in the biochemical composition of blueberry cultivars being tested, show the presence of significant genotypic and time-related differences in the direction and the extent of the above mentioned transformations testifying the marked specific character of different cultivars' responses in terms of exposure durations to low positive temperatures, based on the combination of all the parameters (dry matter, organic acids, carbohydrates and phenolic compounds) analyzed at different stages of storage compared to the initial values.

Table 2. The relative scale of the amplitude and the correlation of variously oriented differences with the initial values of active substances contained in fruit of the *Vaccinium corymbosum* cultivar at different storage stages

Cultivar	Storage stage	Relative scale of the shifts, %			
		+	-	amplitude	+/-
2013					
	1	1.0	53.0	54.0	0.02
Collins	2	14.3	158.2	172.5	0.09
	3	19.2	222.5	241.7	0.09
	1	47.4	105.1	152.5	0.45
Hardyblue	2	27.7	235.3	263.0	0.12
	3	67.3	324.6	391.9	0.21
	1	41.7	60.6	102.3	0.69
Denise Blue	2	34.5	114.7	149.2	0.30
	3	92.6	150.6	243.2	0.62
2014					
	1	151.0	52.2	203.2	2.89
Bluetta	2	184.0	62.5	246.5	2.94
	3	90.1	218.4	308.5	0.41
	1	65.5	139.6	205.1	0.47
Bluecrop	2	61.1	192.9	254.0	0.32
	3	61.1	203.0	264.1	0.30
	1	30.4	281.1	311.5	0.11
Elizabeth	2	83.8	245.4	329.2	0.34
	3	107.7	436.2	543.9	0.25

Thus, for the experiments held in 2013, in the setting of the gradual increase in the degree of the differences during storage compared to the initial values of the amount of active substances, the *Collins* and *Denise Blu*e cultivars were characterized by its smallest, but similar relative values – from 54.0 to 241.7 % and from 102.3 to 243.2 % respectively, while its largest value – from 152.5 to 391.9 % – became a characteristic of the *Hardyblue* cultivar indicating the most marked transformations of the biochemical composition of its fruit during storage. However, as shown above, alongside with the consumption of some organic compounds as respiratory substrates resulting in the decrease of their amount as compared to the initial values, their accumulation was also observed due to the inter-conversion of organic compounds, the cumulative relative effect of which at different stages of storage ranged from 1.0 to 19.2 % for the *Collins* cultivar, from 27.7 to 67.3 % for the *Hardyblue* cultivar and from 34.5 to 92.6 % for the *Denise Blue* cultivar; at the same time the two latter taxa showed the minimum values of this parameter at the second storage stage, whereas their maximum values, as well as for the *Collins* cultivar, were shown at the third stage of storage. To a certain degree, it corresponds to the data[18] on the dynamics of a number of biologically active compounds in the fruits of several cultivars, including highbush and lowbush blueberry, in the process of storage at different positive temperatures and have demonstrated no losses of ascorbic acid in 8 days of storage. There are no significant losses of phenolic compounds at a relative stability of antioxidative activity of blueberry fruits after 2-week storage at the temperature of +4 ^{o}C.[19]

In the experiments in 2014, like in the other one held in the year before, the smallest, but similar relative degree of differences related to the amount of the tested substances as compared to their initial values – within 203.2-308.5 % and 205.1-264.1 % – became characteristics of the early and mid ripening *Bluetta* and *Bluecrop* blueberry cultivars, whereas the biggest degree – from 311.5 to 543.9 % – became a characteristic of the late-ripening *Elizabeth* cultivar indicating the most marked transformations of the biochemical composition of its fruit during storage. Besides, the relative increase of the amount of some substances contained in fruit at different stages of storage varied from 90 to 184 % for the *Bluetta* cultivar, 61.1 to 65.5 % for the *Bluecrop* cultivar and 30.4 to 107.7 % for the *Elizabeth* cultivar. The studies of A. M. Connor et al.[20] also demonstrate expressed cultivar-specificity of transformation of the biochemical composition of blueberry fruits in the process of storage at low positive temperatures. In this case, same as in our studies, growth of the content of anthocyanins and the total quantity of bioflavonoids during the first three weeks of storage is noted.

The most objective view on the degree of transformations of the biochemical composition of blueberry fruit during storage can be formed with the help of the many-fold scale of the correlation of the relative values of the scopes of positive and negative deviations from the initial value of the analyzed parameters combination. It turned out also that in the experiment in 2013 all the tested objects at all stages of storage had it at a level significantly lower than 1.0, which indicated prevalence of negative transformations in the quality of blueberry fruit over positive ones.

At the same time, at all storage stages the minimum value of this correlation became a characteristic of the *Collins* cultivar, while its maximum value was attributed to the *Denise Blue* cultivar, with the *Hardyblue* cultivar taking an intermediate position. This means that the most significant excess of the loss of nutrients over their accumulation during storage became a characteristic of the early ripening cultivar, while the lowest excess was attributed to the late ripening breed, with the high-level expression of this effect shown by the *Hardyblue* and *Denise Blue* cultivars at the end of the second stage of storage and its marked weakening at the end of the experiment, especially for the last object.

Based on the comparison of the scale of the given correlation within the taxonomic range, the quantitative assessment was given with respect to the decrease of the integral level of the nutrient and vitamin value of blueberry fruit and, consequently, the deterioration of their consumer properties after a month of storage. The *Collins* cultivar was rated the highest, whereas the *Hardyblue* and *Denise Blue* cultivars were rated by 2.3 and 6.9 times lower, respectively.

In the experiment in 2014, the scale of the correlation of the relative values of the scopes of positive and negative deviations from the initial values of the tested substances contained in the mid and especially late ripening cultivars of blueberry at all stages of storage was 2 to 9 times lower than 1.0, which indicated prevalence of negative transformations in the biochemical composition of blueberry fruit over positive ones; the same as was observed in the experiment in 2013. In contrast to these taxa, the early ripening *Bluetta* cultivar during first two stages of storage lasting 20 days showed an improvement of its qualitative characteristics as compared to the initial values, which was confirmed by nearly three-fold excess of the scope of positive changes in their biochemical composition as opposed to negative ones; and only at the final stage of storage a seven-fold decrease of the integral level of the nutrient and vitamin value of blueberry fruit of this cultivar as compared to its initial values was observed. It should be noted that in the experiment of the same type in 2013, but on other cultivars of blueberry, the opposite results were observed: the highest excess of the loss of the related compounds over their accumulation at all storage stages became a characteristic of the early ripening cultivar, while the lowest excess was attributed to the late ripening breed. In our opinion, this may indicate a greater dependence of the revealed effects in the transformation of the biochemical composition of blueberry fruit during storage at low positive temperatures on weather conditions of the season rather than on plant genotype and fruit ripening periods.

Based on the comparison of the scale of the given correlation within the blueberry taxonomic range, the quantitative assessment was given with respect to the decrease of the integral level of the nutrient and vitamin values of blueberry fruit and, consequently, the deterioration of their consumer properties after a month of storage.

The *Elizabeth* cultivar was rated the highest, whereas the *Bluetta* and *Bluecrop* cultivars were rated by 1.6 and 1.2 times lower, respectively. This indicates the increase of the preservation of the quality of blueberry fruit harvested in 2014 at low positive temperatures in the range from the early to the late ripening cultivars, while in the experiment held in 2013 the opposite situation was observed.

References

[1]Lohachoompol, V., Srzednicki, G., Craske, J., *J. Biomed. Biotechnol.*, **2004**, *5*, 248-252.

[2]Scibisz, I., Mitek, M., *Acta Sci. Pol., Technol. Aliment*, **2007**, *6(4)*, 75-82.

[3]Skupien, K., *Acta Sci. Pol., Hortorum Cultus*, **2006,** *5(1)*, 19-25.

[4]*Methods for determination of the dry matter: GOST (State Standard)* 8756. 2-82. **1982**, p.

[5]*Methods for biochemical studies of plants*. (Ed.) Ermakova, A. I. **1987**, p. 430. Leningrad. Russia [In Russian].

[6]Marsov, N. G. *Phytochemical studies and biological activity of cowberries, cranberries and blueberries*. **2006**, pp. 99-101. Perm, Russia [In Russian].

[7]Pleshkov, B. P. *Workshop on plant biochemistry*. **1985**, pp. 110-112. Moscow. Russia [In Russian].

[8]Rupasova, Z. A., V. N. Reshetnikov, T. I. Vasilevskaya, N. P. Varavina, N. B. Krinickaiy, A. M. Bubnova, N. B. Pavlovsky, A. G. Pavlovskay, T. V. Kurlovich, Y. M. Pinchukova. *Orcharding*. **2014**; *26,* 413-426. Samokhvalovichy, Belarus [In Russian].

[9]Swain, T., Hillis, W. J. *Sci. Food Agric*. **1959**; *10(1)*, 63-68.

[10]Skorikova Y. G., Shaftan, E. A. *Method for determination of anthocyanins in fruit and berries*. In: *Proc. 3rd All-union Semin. Biol. Active (Therapeutic) Subst. Fruit and Berries.* **1968**, pp. 451-461. Sverdlovsk, Russia [In Russian].

[11]Andreev, V. Y., Kalinkina, G. I., Kolomiyets, N. E., Isaykina, N. V. *Pharmacy,* **2013,** *3(19)* AM-21.

[12]*State Pharmacopoeia of the USSR*. **1987,** *1*, 286-287. Moscow. Russia [In Russian].

[13]Chiabrano, V., Giacalone, G., *Int. J. Food Nutr*., **2015,** *66(3)*, 248-253.

[14]Wang, S. Y., Chen, C-T., *Food Chem.*, **2010,** *122(4)*, 1153-1158.

[15]Remberg, S. F., Haffner, K., Blomhoff, R., *Acta Hort*., **2003,** *600,* 595-598.

[16]Basiouny, M., Chen, Y., *Proc. Fla. State Hort. Soc*., **1988,** *101,* 281 – 284.

[17]Rupasova, Z. A., Reshetnikov, V. N., Yakovlev, A. P. *Method for ranking plant taxa*: Belarus Patent No. 17648 dated 08.07.**2013**. Minsk, Belarus [In Russian].

[18]Kalt, W., Forney, C. F., Martin, A., Ptior, R. L. *J. Agric. Food Chem*., **1999.** *47,* 4638-4644.

[19]Yuan, W., Zhou, L., Deng, G., Wang, P., Creech, D., Li, S., *Pharm. Crops,* **2011**, *2,* 11-23.

[20]Connor, A. M., Luby, J. J., Hancock, J. F., Berkheimer, S, Hanson, E. J., *J. Agric. Food Chem*., **2002,** *50(4)*, 893-898.

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