

Theses of the doctoral (PhD) dissertation

Ákos Ribárszki

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**EFFECT OF ACEROLA AND ASCORBIC ACID ON THE QUALITY OF
LARGE-SCALE ASEPTICALLY PRODUCED APPLE AND SOUR CHERRY
JUICE DURING STORAGE**

Ákos Ribárszki

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PhD School/Programme

Name: PhD School of Food Science

Field: Food Science

Head of School: Simonné Sarkadi Livia, DSc
Hungarian University of Agriculture and Life Science University
Institute of Food Science and Technology

Supervisor: Máté Mónika, PhD
Hungarian University of Agriculture and Life Science University
Institute of Food Science and Technology

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Signature of Head of School

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Signature of Supervisor

Introduction and objectives

Apples and cherries are the two main fresh and industrial fruits in Hungary. In the case of processed products, they are particularly popular in the form of juice obtained by aseptic preservation (not from concentrate (NFC)), which eliminates the seasonality caused by the short harvest period. An important step in manufacturing technology is color fixation, for which ascorbic acid is mainly used in industrial practice. However, there is a growing need to replace ascorbic acid, one possible solution being the addition of ascorbic acid-rich acerola juice.

My objectives were:

- Ascorbic acid treatment of crumbs and pressed juices is a popular color-fixing and color-preserving method in the fruit processing industry. There have been a number of attempts to induce ascorbic acid, of which acerola juice with high ascorbic acid is promising.
- My aim was to compare the color retaining effect of ascorbic acid and acerola juice in aseptically filled apple juice ('Idared') and sour cherry juice ('Érdi bőtermő') compared to control (without color fixation) samples during 12 months of storage in order to determine whether acerola juice may be a suitable substitute for ascorbic acid.
- In the case of apple juice, the preservation of the red color provided by the bright yellow color and the high anthocyanin content of the cherry juice is one of the most important goals of the applied aseptic technology. My goal was one of the most important parameters determining the quality of the product for both juices tracking of color and its characteristics without inhibiting color change, in case of ascorbic acid and acerola juice treated apple juice and sour cherry juice. My aim was to determine the positive and negative effects of treatments in industrial aseptic processing and storage compared to untreated samples.
- The biologically valuable components of fruits are phenolic compounds and their antioxidant activity. My aim was also to study the effect of the treatments during storage on the biologically valuable components of apple juice and sour cherry juice.

With my results, I would like to contribute to the development of modern semi-finished food production and increase its economic competitiveness.

Materials and methods

The apples and cherries used in the doctoral research were grown in Kiskőrös (N: 46°, 37'; E: 19° 17').

The processing of the raw materials for apples and cherries, as well as the filling of the aseptic sample bags, which were the subject of the experiments, took place under industrial conditions in the fruit and vegetable processing plant of Juice Products Zrt. in Kiskőrös.

Brief description of the technology: The fruit was processed immediately after harvest to avoid the effect of storage. The apple first went through an SRAML® JP6000 drum washer, then placed in a ProXES perforated disc fruit grinder, where the pre-crushing required for pressing was done (in the case of cherries, seeding and then two-phase passage). Pressing was performed with a DELLA TOFFOLA® PEC100 fruit press. The juice was then preheated to 40 °C with an FBR Elpo Mr49-6, then the dissolved and dispersed oxygen was removed with a deaerator (SRAML D-1000). Ascorbic acid and acerola juice were added to the juice immediately after compression. Heat treatment was performed with Della Toffola Priamo tube in a tube pasteurizer at 86 °C for 1 minute. The pasteurized juice was transferred to sterile 5 liter sample bags (SCHOLLE IPN®) after cooling in a closed system with an FBR ELPO® aseptic filling system.

Taking into account the color fixation treatments (ascorbic acid and acerola juice addition) and the planned storage time (12 months), 21-21 pieces 5 l sample bags per treatment were prepared for both fruit species under operating conditions. No color fixation treatment was performed during the preparation of the control samples. Ascorbic acid was added at 200 mg l⁻¹ in the preparation of ascorbic acid treated samples (apple (AAS), sour cherry (MAS)). In case of treatment of sour cherry with acerola juice was added with acerola juice which contain 200 mg l⁻¹ ascorbic acid (apple (AACE), sour cherry (MACE)). Dosing took place during storage in aseptic containers using its own dosing unit in less than one minute after pressing, so the degree of oxidation occurring during this is negligible. The coding of the samples is summarized in Table 1.

The 5 l sample bags were tested immediately after processing and every 2 months during 12 months of storage. At each test time point, 3-3 bags were analyzed for each sample. The samples were stored in the warehouse of Juice Products Joint Stock Company's vegetable and fruit processing plant in Kiskőrös at 20 °C.

Table 1. Coding of experimental apple juice and sour cherry juice samples

	Code of Sample	Applied heat treatment	
Apple Samples	AK	-	-
	AAS	ascorbic acid dosage	200 mg l ⁻¹
	AACE	acerola juice dosage	acerola juice contains 200 mg l ⁻¹ ascorbis acid (1.42 g l ⁻¹)*
Sour Cherry Samples	MK	-	-
	MAS	ascorbic acid dosage	200 mg l ⁻¹
	MACE	acerola juice dosage	acerola juice contains 200 mg l ⁻¹ ascorbis acid (1.42 g l ⁻¹)*

*Acerola concentrate: 65 ref%

In order to characterize the color of the samples during the storage time, the CIELab color coordinates (L*, a*, b*) were determined using a Konica-Minolta hand-held colorimeter, then the color difference (ΔE^*) from the data obtained, and tanning index (BI).

Total polyphenol content was determined using Folin-Ciocalteu reagent and sodium sulfate solution by the method of Singleton and Rossi (1965). Absorbance was measured with a Hitachi U-2900 spectrophotometer at 760 nm. Results are expressed in mgGE (gallic acid equivalent) l⁻¹.

Total monomeric anthocyanin content was determined using the method of Giusti and Wrolstad (1996). The difference in absorbance at 520 nm in pH 1.0 and pH 4.5 is proportional to the anthocyanin concentration given in mg l⁻¹ for cyanidin-3-glucoside.

Antioxidant capacity was determined by the method of Benzie and Starin (1995). The principle of the measurement is that ferric (Fe³⁺) ions are reduced to ferro (Fe²⁺) ions, which are complexed with tripyridyltriazine (TPTZ = 2,4,6 tripyridyl-S-triazine) at low pH to form colored products (ferrotripyridyltriazine). The blue color was measured with a spectrophotometer at 593 nm, the results are given for ascorbic acid in mmolAS (ascorbic acid equivalent) l⁻¹.

The ascorbic acid content and the quantification of polyphenolic compounds (catechin, chlorogenic acid, gallic acid) were determined using a Shimadzu high-performance liquid chromatography (HPLC) column with a reversed-phase C18 column (Supelco, 250 * 4.6 mm i.d., 5 μ m). The time required to record the chromatograms was 5 minutes for ascorbic acid and 40 minutes for polyphenol compounds, and the injected volume was 20 μ l for each sample at a flow rate of 1.5 ml / min (Székely et al., 2014; Székely et al. , 2019).

To determine the rheological properties, the Physica MCR51 (Anton Paar Hungary Kft.) Rotary viscometer was used. During the measurement, the deformation speed [1 / s] was increased from 600 1 / s to 1200 1 / s, and the shear stress [Pa] was recorded at a total of 20 measurement points. The slope of the flow curves thus determined corresponds to the viscosity of the samples [mPa.s].

The values of parameters were expressed as the mean \pm standard deviation (SD). The one-way analysis of variance (ANOVA) and Tukey post-test were performed at $p < 0.05$ to evaluate the significance of differences. Statistical analyses were conducted using SPSS version 27 Software.

Results and discussion

In the course of my doctoral experiments, the use of acerola juice as an antioxidant was investigated in the production and storage of apple juice and cherry juice for the treatment of ascorbic acid and its effect on the color and antioxidant compounds of fruit juices compared to untreated samples and whether it can be replaced by aceroleal ascorbic acid for color fixation.

Based on my results, it can be stated that the changes during storage took place in a multi-complex matrix, where the interaction and continuous transformation of different compounds prevailed.

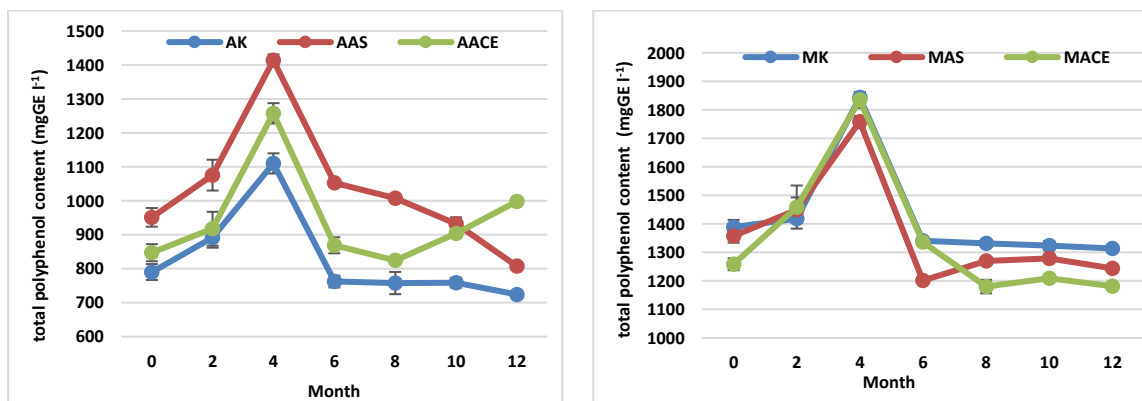


Figure 1. Change in total polyphenol content of apple juice and cherry juice samples during storage (mgGE l^{-1})

This is due, among other things, to the fact that in the case of apple juice and cherry juice samples, the total polyphenol content fluctuates during storage, showing a decreasing trend after growth and then stagnating (Fig. 1). Ascorbic acid treatment was more stable in the case of apple juice, while no difference could be made between treatments in the case of sour cherry juice. Of the polyphenolic compounds tested, catechin was also presented in the highest amounts in apple juice and sour cherry juice, followed by chlorogenic acid and then gallic acid. During storage of both fruits, the polyphenolic compounds were mostly stable until the 2nd month of the storage period, and then, following different dynamics, their amount decreased.

In terms of antioxidant capacity, treatment with acerola was clearly more effective in both juices (Fig. 2).

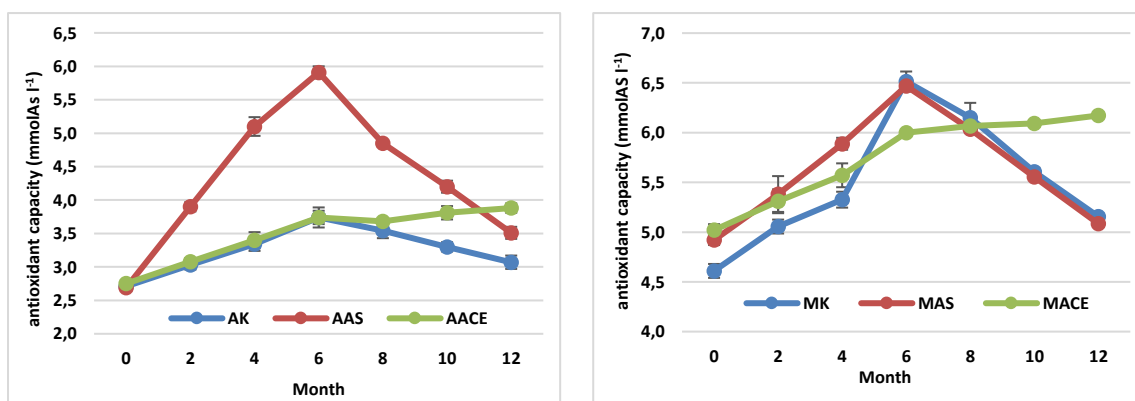


Figure 2. Change in antioxidant capacity (FRAP) of apple juice and cherry juice samples during storage (mmolAS l^{-1})

Regarding the total anthocyanin content of the sour cherry juice, a decreasing trend was found after a transient upward phase, with acerola treatment being clearly more effective in maintaining the anthocyanin content (Fig. 3).

Due to the copigmentation between the compounds, other components of acerola ascorbic acid are thought to play a role.

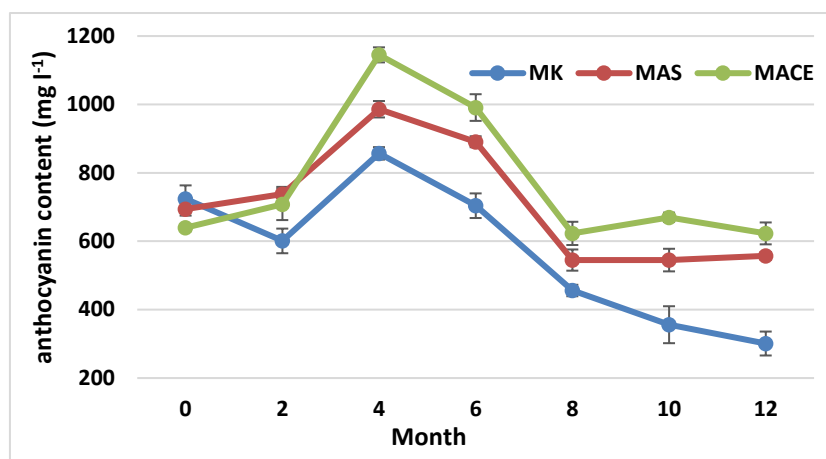


Figure 3. Change in total anthocyanin content of cherry juice samples during storage (mg l⁻¹)

During the analysis of the ascorbic acid content of the apple juice and sour cherry juice samples, there was a significant difference between the ascorbic acid treatment and the control sample (Fig. 4) as well as between the two treatments in favor of ascorbic acid treatment throughout the storage period.

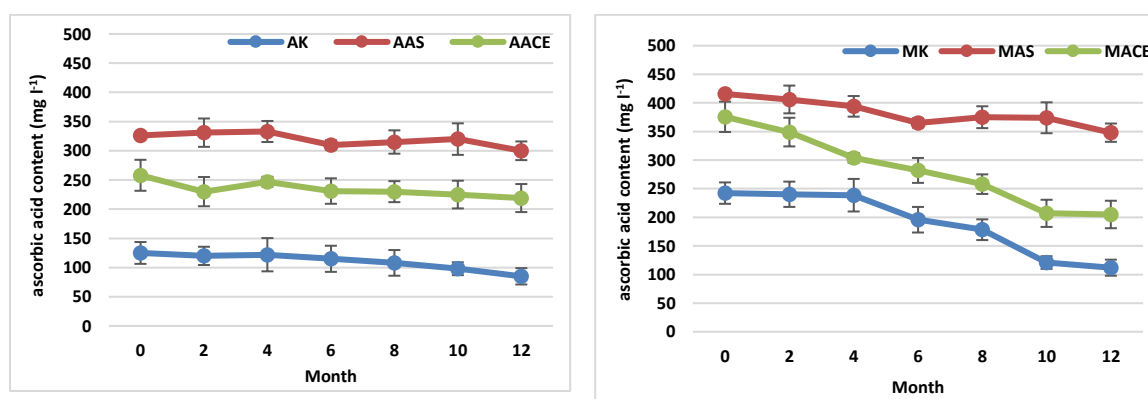


Figure 4. Change in ascorbic acid content of apple juice and sour cherry juice samples during storage (mg l⁻¹)

For the browning index (BI) used to summarize the change in color parameters, it was observed that the control showed the smallest change in the apple juice samples, while there was no significant difference between the ascorbic acid and acerola treatments when examining the entire storage period (Fig. 5). However, it can be observed that until the fourth month of storage, acerola treatment is better as good as ascorbic acid treatment.

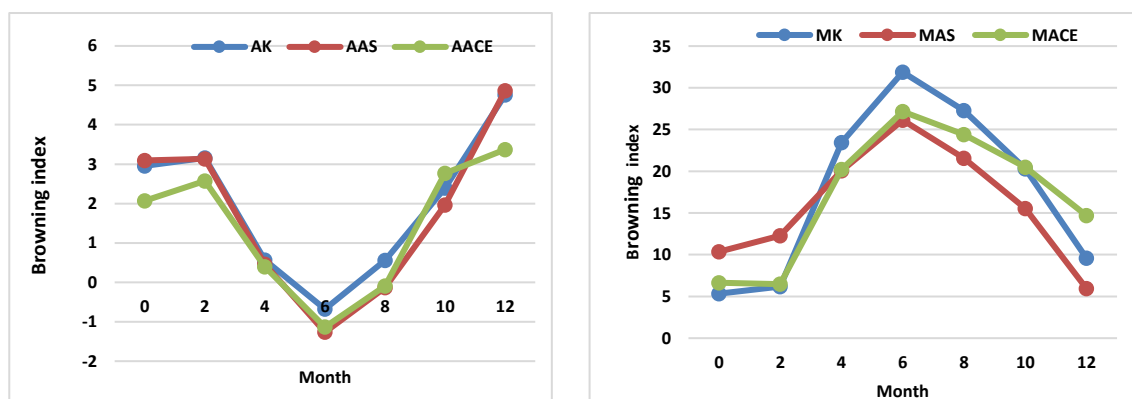


Figure 5. Change in browning index (BI) values of apple juice and cherry juice samples during storage

In the further stage of storage, ascorbic acid is more favorable, and at 12 months the difference is again negligible.

In the case of cherry juice, the browning index is the lowest for the ascorbic acid-treated sample when looking at the entire storage period. By 6 months, acerola treatment had been shown to be better or as good as ascorbic acid treatment, with ascorbic acid remaining better for the remainder of storage. Thus, in terms of color retention, the use of acerola juice is recommended for the 4-month storage period for apple juice and the 6-month storage period for cherry juice, but ascorbic acid is better for longer planned storage.

There was no difference in the viscosity of the samples in terms of treatments at the beginning of storage, as within each species, regardless of the treatment, the preparation operations and the pressing took place under the same conditions in terms of technology and duration. Accordingly, the initial values of the samples were similar. During the storage period, there was a 2.1-3.5% decrease in viscosity values for apple juices and 25.6-34.4% for seed cherries (Fig. 6). We fitted the Herschel-Bulkley model to the results, based on which $n = 1$, i.e., apple juice and sour cherry juice samples behave as Newtonian liquid regardless of storage time and treatments.

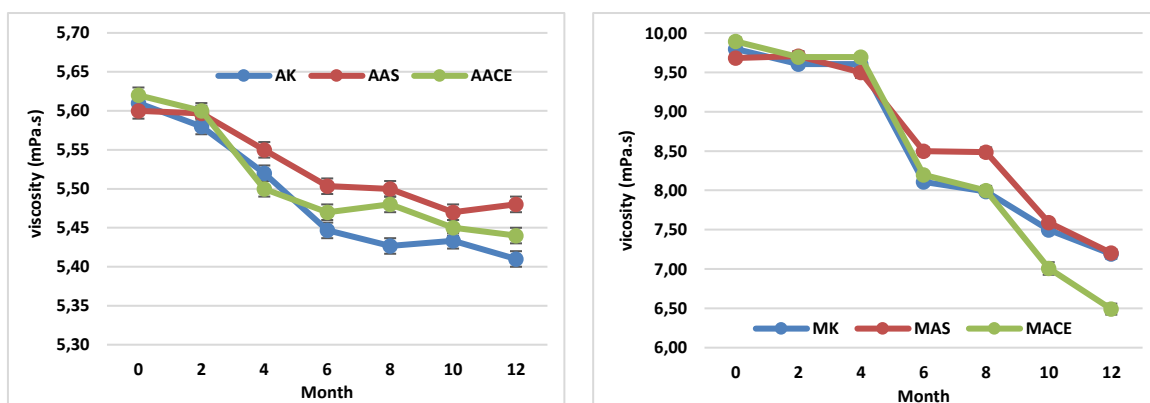


Figure 6. Change in viscosity of apple juice and sour cherry juice during storage (mPa.s)

If the preservation of antioxidant compounds is a priority for the product, acerola juice and ascorbic acid can be used as a possible alternative to improve the quality of aseptic cherry juice during storage.

Conclusions and recommendations

The results of my research do not clearly demonstrate that ascorbic acid can be effectively replaced by fortification of apple juice and sour cherry juice samples with acerola, but it can be concluded that its dosage is definitely more favorable compared to the control sample.

However, the present research also reveals that the amount of ascorbic acid commonly used in industrial practice (200 mg l^{-1}) proved to be too high for the 'Érdi bőtermő' sour cherry variety and did not create a sufficiently stable system. The amount of anthocyanin depends on the variety, so is the stability during heat treatment and storage. I recommend optimizing and determining the amount of ascorbic acid added per variety in order to maintain the content parameters of anthocyanin-containing raw materials.

Based on the results, it is recommended to determine the optimal amount of ascorbic acid addition during the technology, considering the modernization of processing machines and technologies, as well as the shortening of processing time, which affect biochemical processes, oxidation rate and thus the parameters of the juices I examined.

Continuing the topic, it is advisable to use an ultra-high-pressure screw centrifuge (variable pitch decanter) instead of a pneumatic drum press, as in this case the total moisture content in the fruit can be recovered, which would presumably result in a higher bioactive substance content in the juices.

New scientific results

1. Based on the browning index and the results of the color measurement, I found that in the case of aseptically packaged 'Idared' apple juice and 'Érdi bőtermő' sour cherry, stored aseptically, treatment with ascorbic acid (200 mg kg^{-1} dose) caused significantly less color change than acerola (equivalent to 200 mg kg^{-1} ascorbic acid). 'Idared' apple juice and 'Érdi bőtermő' cherries can be stored for 2 months without significant color change. Acerola juice is suitable for color fixation after 4 months of storage, but ascorbic acid treatment is recommended for 6-12 months of storage.
2. I found that in the case of 'Idared' apple juice aseptically packaged and stored for 12 months, acerola treatment (increase of 13.2% in control, 30.4% in ascorbic acid treatment, 41.0% in acerola treatment) was used to maintain total antioxidant capacity, while 6 Ascorbic acid treatment is recommended during one month of storage.
3. I found that the total antioxidant capacity (increase of 11.9% in the control, 3.45% in the case of ascorbic acid treatment, 22.9% in the case of acerola treatment) and the anthocyanin content were as high as possible in the case of aseptically packaged 'Érdi bőtermő' cherries (loss of 81.7% for control, 58.0% for ascorbic acid treatment, 21.1% for acerola treatment), color fixation with acerola was more effective and independent of storage time compared to the ascorbic acid-treated and control samples during the study period.
4. I found that the color characteristics (L^* , a^* , b^*) of aseptically packaged 'Idared' apple juice and 'Érdi bőtermő' cherry stored aseptically show a strong correlation with the antioxidant capacity of the control and ascorbic acid treated samples (L^* , $r = 0.941-0.959$, a^* , $r = -0.852$ for apple juice, $r = 0.941-0.962$ for cherry juice, b^* , $r = 0.766-0.977$).
5. I found that aseptically packaged and stored for 12 months 'Idared' apple juice and 'Érdi bőtermő' cherries juice had an effect of 4 months for catechin, 8 months for chlorogenic acid and 4 months for gallic acid acerol treatments, after which the storage time eliminates the effects of the treatments on phenolic acids. In the case of 'Idared' apple juice, ascorbic acid treatment has a significant protective effect on gallic acid after 6 months.
6. I found that in the case of 'Idared' apple juice and 'Érdi bőtermő' cherries produced with the same technological parameters, apple juice was more stable (viscosity reduction 2.1-3.5%) than cherries in terms of rheological properties during the 12-month storage period (viscosity reduction (25.6-34.4%).

Publications of the author related to the topic of the dissertation (in the grouping defined by the doctoral school)

Journal IF

Ribárszki, Á., Székely, D., Szabó-Nótin, B., Góczán, B., Friedrich, L., Nguyen, Q.D., M. Máté (2022): Effect of ascorbic acid and acerola juice on some quality properties of aseptic filling apple juice. *Acta Alimentaria* (accepted)

Ribárszki, Á., Székely, D., Szabó-Nótin, B., Máté, M. (2022): Changes in colour parameters and anthocyanin content of aseptically filled sour cherry juice during storage. *Progress in Agricultural Engineering Sciences* (accepted)

Székely, D., Vidák, K., Furulyás, D., **Ribárszki, Á.**, Stéger-Máté, M. (2019): Effect of drying methods on physicochemical parameters of different red beetroots (*Beta vulgaris* L.) *Periodica Polytechnica Chemical Engineering*, 63(3): pp. 485-490.

Conference full paper

Ribárszki, Á., Szabó-Nótin, B., Máté, M. (2021): Changes in color parameters and anthocyanin content of aseptically filled sour cherry juice during storage. Lippay János - Ormos Imre - Vas Károly (LOV) Tudományos Ülésszak. 2021. november 29. ISBN 978-615-01-3738-4 (pdf) <https://lov.uni-mate.hu/book-of-abstracts>

Gonelimali, F., Szabó-Nótin, B., Szalóki-Dorkó, L., **Ribárszki, Á.**, Máté, M. (2021): Evaluation of polyphenol extraction from apple pomace. Agrosym 2021. Jahorina, Bosnia and Herzegovina. 7-20 October 2021.

Gonelimali, F., **Ribárszki, Á.**, Székely, D., Pesti, Zs., Máté, M., Szabó-Nótin, B. (2021): Effect of apple pomace powder addition on millit flour based biscuits quality. International Conference on Agriculture & Food. Burgas, Bulgaria, 16-19 August 2021

Summary

Ribárszki, Á., Stéger-Máté, M. (2021): Changes in color parameters and polyphenol content of aseptically filled apple juice during storage. 6th International ISEKI-Food Conference 23-25 June 2021

Publication in Hungarian

Király, I., **Ribárszki, Á.**, Mihálka, V. (2017): Jégvédő háló hatása az alma gyümölcsének makroelem-tartalmára In: Kiss, Zoltán; Stefanovitsné, Bányai Éva (szerk.) 15. Magyar Magnézium Szimpózium: 15th Hungarian Magnesium Symposium: Program és összefoglalók Budapest, Magyarország: Magyar Kémikusok Egyesülete (MKE) pp. 29-30.

Ribárszki, Á., Kajtár Czinege, A. (2016): Jégeső elleni védőháló alkalmazása intenzív gyümölcsültetvényekben *AGROFÓRUM - A NÖVÉNYTERMESZTŐK ÉS NÖVÉNYVÉDŐK HAVILAPJA* 27(10): pp. 135-137.

Ribárszki, Á., Kajtár Czinege A. (2016): Jégvédő intenzív ültetvényekben *KERTÉSZET ÉS SZŐLÉSZET* 65(19): pp. 17-18.