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Research Article

Anthocyanin and bioactivity properties of *berberis crategina* DC. In buffer system and apple juice: impact of temperature, time, and pH; Prediction using artificial neural network

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ABSTRACT

Our work contributes to investigate and estimate the degradation, altered bioactivity and color of *Berberis crategina* anthocyanins in different buffer systems and apple juice. Anthocyanins are glycosides of anthocyanidins, a subclass of flavonoids. These pigments impart red to blue coloration to fruits and flowers. Anthocyanins have antioxidant properties due to the positively charged oxygen atoms they contain. Chemical structure, enzymes, temperature, light, pH, oxygen, ascorbic acid, sugars, metals, sulfur dioxide, and copigmentation affect the stability of anthocyanins. In this study, it was primarily aimed to investigate the effects of temperature, time and pH on total anthocyanin content (TAC), total phenolic content (TPC), antioxidant activity (AA) and color of *Berberis crataegina.* Another aim was to estimate the TAC, TPC, AA, and color of *Berberis* based on temperature, time, and pH with ANN modeling. An artificial neural network (ANN) was used to predict the relationship between TAC, TPC, AA and color of *Berberis crataegina* and temperature, time, and pH for both apple juice and buffer solution. It was found that high temperature and low acidity increased anthocyanin degradation, while total phenolic content and antioxidant activity decreased. L^* and $h°$ were found to decrease and C^* to increase due to anthocyanin degradation. The results indicate that pH is the most effective factor (73%) in prediction and that ANN performs better than a buffer solution for apple juice. The sum of square errors of the validation samples was 7.89 for buffer solution and 1.26 for apple juice. This study showed that the parameters studied can be successfully estimated using ANN.

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INTRODUCTION

Berberis crataegina DC. is a wild species of *Berberis* that is encountered in the Central Anatolia region of Turkey. This plant is known as 'karamuk' in local places in Turkey. The fruit of *Berberis crataegina* DC. varies from dark purple to black and oval [1]. The *B. crataegina* has high vitamin C and anthocyanin contents, organic acids, and tannins. Depending on these properties, they provide antifungal activity, anti-inflammatory and antioxidant activity [2]. The fruits and leaves of *Berberis* have high antioxidant capacity [3,4]. Due to its antioxidant properties, it has some beneficial effects on health like anti-allergic, anti-inflammatory, vasodilator, antimicrobial, and antithrombotic [5]. Anthocyanins are glycosides of anthocyanidins, which are a subset of flavonoids. They are natural pigments that can be water-soluble [6]. These pigments handle colors ranging from red to blue in the fruit and flowers [7]. The positively charged oxygen atom possessed by anthocyanins provides them with antioxidant properties [8]. The stability of anthocyanin is impressed by its chemical structure, enzymes, temperature, light, pH, oxygen, ascorbic acid, sugars, metals, sulfur dioxide, and copigmentation [9].

ANN is an effective nonlinear modeling tool that mimics the learning concept of the human brain. The learning activity of ANN is classified as supervised and unsupervised learning. If we have any information about the outputs of the system, this is called supervised learning, while it is described as unsupervised learning when we have no information about the output of the system. In the supervised learning procedure, ANN constructs the nonlinear relationship between input/inputs and output/outputs, therefore it can be used for prediction, classification, pattern recognition, etc. ANN generally structures from an input layer, one or more hidden layer(s), and an output layer. Each layer consists of neurons, and all neurons connect by weights. Furthermore, the activation function of neurons processes the information which comes from other layers. Each layer has a special mission in an ANN structure. The input layer provides information for the network while the hidden layer is responsible for learning activity. On the other hand, the output layer is a supervisor by gives information about forecasts [10]. ANN has been used in various studies in different fields such as medicine, environmental science, economy, industry, construction, etc. On the other hand, there are many applications of ANN in the field of food science [11,12]. Huang et al. [13] indicated that advantages of ANN such as flexible learning algorithm, diverse network topology, fast learning algorithm, and high error tolerance make the ANN a powerful analytical tool in food science, particularly in sensory analysis, analytical chemistry, and process control.

Our study has two aims. The first aim is to show the degradation, varying bioactivity, and color of *Berberis* anthocyanin at different buffer systems. Anthocyanin stability is affected by minor components. For this reason, also apple juice was used to color. Another aim is to estimate the TAC, TPC, AA, and color of *Berberis* based on temperature, time, and pH with ANN modeling.

This paper is structured as follows: The first section gives a brief overview of *Berberis crataegina* DC. and the literature of related works. The second section presents the material and methods of the paper and related solutions of the study are given in Section 3. Our conclusions are drawn in the final section, Section 4, with the future direction of studies.

MATERIALS AND METHODS

Fruit Materials and Chemicals

B. crataegina DC. fruits considering in this study were collected in August from the mountainous area of Yıldızeli district in Sivas, Turkey. After the fruits were separated from the undesirable substances, they were washed with tap water. Fruits were placed in polyethylene pouches and were stored at -18 °C (Vestel Freezer, Turkey) until the sample preparation stage. Apple juice (Cappy, Turkey) was obtained from the local market. Gallic acid standard and 1,1- diphenyl-2-picrylhydrazyl (DPPH) were purchased from Sigma Aldrich and Folin-Ciocalteau reagent from Merck. Solvents and reagents were of analytical grade.

Preparation of *B. crataegina* DC. Concentrate

After squeezing the fruits with a blender, they were filtered through cheesecloth. So, *B. crataegina* DC. juice was got. This juice was centrifuged at 8000 rpm for 5 minutes at 4 °C. After this stage, the supernatant was collected and concentrated to 68.2 °Brix in a rotary evaporator (Heidolph Hei-Vap Value G1, Germany) at 45 °C, 200 rpm, and 150 Pa. The obtained *B. crataegina* concentrate (BCC) was used to color buffer solutions and apple juice.

Preparation of Buffer Systems

Citrate-phosphate buffer solutions were prepared at six different pH levels (2.0, 3.0, 4.0, 5.0, 6.0, and 7.0) according to [14]. The pH values of the buffer systems were controlled using a pH meter (Ohaus Corporations, USA) before use.

Degradation Studies

Anthocyanin analysis was performed in the concentrate to investigate the effect of temperature and pH on *B. crataegina* DC. anthocyanin. According to the result obtained, 100 mL buffer solutions and apple juice were colored with 0.46 g concentrate, as 4 mg anthocyanin/100 mL. Ekici et al. [15] reported that 100 mL of a solution containing 4 mg of anthocyanin would be enough. They were divided into 10 ml portions into Pyrex glass tubes and exposed to heat treatment in a thermostatic water bath (Nüve St30, Turkey) at 70, 80, and 90 °C for 5 hours. Samples were taken at one-hour intervals from the thermostatic water bath and cooled with tap water. The determination of TAC, TPC, AA, and color analysis were made in these samples.

Determination of Total Anthocyanin Content

TAC of the samples was analyzed according to the pH differential method [16]. The absorbance of the sample was determined using a spectrophotometer (Shimadzu UV-1800 spectrometer, Japan). TAC in the sample was expressed as cyanidin-3-glucoside (cyd-3-glc) equivalents and calculated with the following equations:

The principle of the photo-thermal energy conversion is converting the energy of the incident radiation to thermal energy, solar thermal systems collectors are one of the examples. The energy equation for the solar system collector, accounting for the volumetric heat release, can be written as:

$$A = (A_{\lambda max} - A_{700})pH_{1.0} - (A_{\lambda max} - A_{700})pH_{4.5} \quad (1)$$

$$C(mg/kg) = \frac{(A \times MW \times DF \times 1000 \times d)}{\varepsilon M} \quad (2)$$

where A is the absorbance value of the samples, C is the concentration of monomeric anthocyanin, MW is molecular weight (449.2 g/mol for cyd-3-glc); εM is molar extinction coefficient (26,900 L/mol/cm for cyd-3-glc), DF is dilution factor, d is the path length of the cuvette (1 cm).

Determination of Total Phenolic Content

The TPC of the samples was determined by Folin-Ciocalteu colorimetric method with some modifications [17]. Folin-Ciocalteu reagent was diluted 10-fold with distilled water. Na_2CO_3 (7%) was prepared with distilled water to pH 10. Sample (30 µL), diluted Folin-Ciocalteu reagent (150 µL), and Na_2CO_3 (120 µL) were added to the 96-microwell plate. After 60 min incubation at room temperature (20 °C) in the dark, absorbance was measured at 750 nm by a microplate reader (Multiscan FC, Thermo Fisher, USA). Distilled water 30 µL and Folin-Ciocalteu reagent (150 µL) and Na_2CO_3 (120 µL) were mixed for blank, and TPC was expressed as gallic acid equivalents (mg GAE/mL) with the help of the calibration curve that was generated as using gallic acid standard.

Determination of Antioxidant Activity

The radical scavenging activity of DPPH (1, 1-diphenyl-2-picrylhydrazyl) was measured according to the method described by Orhan et al. [18] with some modification. 6x10-5 M DPPH was prepared with pure methanol. 30 µL sample and 270 µL DPPH were added to the 96-microwell plate. The plate was stood in the dark for an hour, after that the absorbance was measured at 520 nm by a microplate reader (Multiscan FC, Thermo Fisher, USA). 30 µL distilled water and 270 µL methanol were prepared for blank and 30 µL distilled water and 270 µL DPPH were prepared for control. DPPH radical scavenging activity was expressed as percent inhibition with the following equation:

$$\%\ \text{Inhibiton} = 1 - \frac{\text{Absorbance of sample}}{\text{Absorbance of control}} \times 100 \quad (3)$$

Determination of Color

The color of samples was identified with the color determination device (Konica Minolta Chromameter Cr-5, Japan). The results were explained according to L^*, $h°$, C^* system. Lightness (L^*) represents brightness while chroma (C^*) represent color saturation or color intensity, it is calculated by $C^* = (a^*2 + b^*2)$ ½. The Hue angle ($h°$) relates to color tone and is determined by $h° = (b^*/a^*)$ equation [19].

Artificial Neural Network to Predict TAC, TPC, AA, and color of *Berberis*

In this study, we have conducted a total of 648 experiments to predict TAC, TPC, AA, and color by using ANN. Inputs of the model were defined as temperature (70, 80, 90 °C), time (5 hours- one-hour intervals), pH (six different pH levels: 2.0, 3.0, 4.0, 5.0, 6.0, and 7.0). Each categorical value of input variables was accepted as a different neuron. By the way, we had 15 neurons at the input layer. On the other hand, the output layer consisted of 6 neurons. Outputs of the model were TAC, TPC, AA, and color values (L^*, $h°$, C^*).

Moreover, 108 experiments with a fixed PH value of 3.04 belonging to apple juice were modeled to predict TAC, TPC, AA, and color values (L^*, $h°$, C^*). We had 14 input neurons and 6 output neurons for apple juice. And same modeling parameters of buffer were used for apple juice.

The data mining module of the STATISTICA 10.0 software package was used for ANN analyses. Multilayer Perceptron (MLP) which is a special type of feed-forward neural network was used in this study. An MLP generally contains an input layer, one or more hidden layers, and an outputs layer. Training algorithms are very important for MLP networks. We prefered to use Broyden-Fletcher-Goldfarb-Shanno (BFGS) training algorithm which is very efficient at nonlinear least squares [20]. 500 different network architectures contain different numbers of neurons in the hidden layer and different kinds of activation functions in neurons were tried. Networks were trained during 200 cycles and the training algorithm was stopped when 0.0000001 change in the sum of square error was occupied. On the other hand, we have 648 experiments for modeling the forecasting system. The number of experiments is sufficient for the generalization of a network. The data set was divided into three samples as 70% training, 15% testing, and 15% validation.

Statistical Analysis

All data represented averages from two times in triplicate. All results were reported as mean ± standard deviation. Shapiro-Wilk test was used to evaluate the normality of the data. Levene test was used to test variance homogeneity. One-way analysis of variance (ANOVA) was performed to analyze the differences between normal data. Tukey test was used for multiple comparisons of homogeneous data. Dunnet T3 test was used to compare inhomogeneous data. The difference between the data that did not show normal

distribution was evaluated with the Kruskal Wallis test. Pairwise comparisons of these data were tested using the Mann-Whitney U test. All transactions were analyzed for significant differences comparing the means at a 5% significance level ($p<0.05$) using SPSS software (IBM, 2018).

RESULTS AND DISCUSSION

Degradation of Anthocyanins

The degradation results for the *B. crataegina* anthocyanin are shown in Table S1 for buffer solutions and apple juice. There was a significant difference between 70, 80, and 90 °C for anthocyanin content of *B. crataegina*. Also, pH levels were significantly effective on *B. crataegina* anthocyanins (statically data not presented for pH). In addition, the statistical analysis presented a significant decrease in anthocyanin content during the heating period ($p<0.05$). Degradation of anthocyanin because of thermal effect is linked with cleavage into a chalcone structure and further transformation into a coumarin glucoside derivative with a loss of the B-ring. Also, anthocyanin degradation may be caused by the fission of covalent bonds, or improved oxidation reactions because of thermal processing. The rate of degradation of anthocyanins increases as the temperature rises during processing and storage [21]. As expected, anthocyanin degradation increased while the temperature raised for all pH values for five hours.

This increase in degradation concurs well with Farhadi Chitgar et al. [22] who researched degradation of *Berberis vulgaris* anthocyanin at 90°C. Another study reported that anthocyanin degradation of *Morus nigra* L. increased by 56.02%, 83.74%, and 91.67%, respectively, at 60, 70, and 80 ° C after 10 hours [23]. Reversible changes occur in the chemical structure of anthocyanins in aqueous acidic solutions. Because the ionic structure of anthocyanins changes depending on the pH value of the environment. While anthocyanins show high stability in acidic environments, they are less stable in alkaline environments [24].

As seen in Table S1, anthocyanin degradation increased with rising pH from 2 to 7. Anthocyanins were most stable in pH 2 buffer solution at all processing temperatures. Anthocyanin degradation was found to be 28.22%, 52.58%, and 83.25%, respectively, for 70, 80, and 90 °C in the pH 2 buffer system colored with BCC. Our results agree with the previous study by Hou et al. [25] who studied at 100 °C heating temperature and in the range pH 1-6 with black rice anthocyanins.

Temperature, pH, and heating time have a significant effect on anthocyanin stability in apple juice ($p<0.05$). Anthocyanin content decreased over time as the temperature increased in apple juice. The pH value of apple juice used in the study is 3.04. Therefore, anthocyanin degradation was compared for pH 3 buffer solution and apple juice. Anthocyanins degraded more in apple juice at 70 °C compared to the pH 3 buffer solution. In contrast, the anthocyanins in apple juice at 80 and 90 °C degraded less than the buffer solution. Compared to the buffer solution, the higher stability of anthocyanin may be attributed to the detractive effect of copigmentation on anthocyanin degradation. Molecules showing copigment behavior such as flavonoids, alkaloids, and organic acids are colorless molecules, and when they are added to the anthocyanin solution, they increase and stabilize the color of this solution [26]. Also, primary sucrose content in fruit juice can be minimized anthocyanin degradation [27]. Apple is one of the major sources of phenolic compounds, flavonoids [28] and the sucrose concentration is ranged from 8.5–55.10 (g/l) in apple juice [29].

Li et al. [30] investigated the thermal stability of purple-fleshed sweet potato anthocyanin in aqueous solutions with various pH (2, 3, 4, 5, and 6) and fruit juices at 80, 90, and 100 °C, and reported that higher stability of anthocyanins was obtained in aqueous solutions with pH 3 and 4 and in apple and pear juices.

Total Phenolic Content and Antioxidant Activity

Table S2 shows that temperature and pH levels (statically data not shown for pH) have a significant effect ($p<0.05$) on TPC and AA of BCC, but heating process time has no significant effect ($p>0.05$). TPC was higher at 90 °C, within the first 3 hours at pH 2. With the increase in temperature after the 3rd hour, TPC decreased as time passed. TPC had the highest value in the heat treatment at 70 °C applied to the pH 4 buffer solution. These values were 1.66, 1.59, 1.57, 1.59, 1.57, and 1.54 mg GAE/mL respectively in the 0-5h range. In the current study, while TPC of buffer systems colored with BCC increases in the pH range 2-4; TPC decreased in the pH range 5-7 for all temperatures. The increase in TPC at 90 °C can be due either to the breakage of esterified or glycosylated linkages or because of the Maillard reaction [31]. Our results are in good agreement with those reported by Sólyom et al. [32] who observed an increase in TPC from 81.32 to 117.1 mg GA/g DW after 4 h of thermal treatment at 150 °C. In apple juice, an increase in TPC was found because of heating as depicted in Table S2. TPC is higher in apple juice than in all buffer solutions. This may be because of the high phenolic content of apple juice [28].

In all buffer solutions and apple juice, AA decreased with increasing temperature and duration of treatment. Minimum and maximum AA were in the pH 7 and pH 4, respectively, for all heating temperatures. The AA of apple juice with a pH of 3.04 used in the study was lower than that of pH 3 and pH 4 during the five-hour treatment process. A previous study showed that anthocyanins, extracted from purple potato, exhibited similar AA under thermal treatments of 100–150 °C for 60 min [33]. In a study like ours conducted by Turturicə et al. [34], it was reported that heat treatment reduced DPPH free radical scavenging activity, and this decrement is eliminated by increasing heating time. This may result from the loss or degradation of

certain types of phenolic compounds or other compounds responsible for AA during heating. The AA of the BCC buffer systems decreased as the acidity of the system decreased. In contrast to our study, Sui et al. [35] have reported that there was an increasing trend of antioxidant capacity as the pH was increased. Since anthocyanins may have different structures under different pH conditions, their quinoidal base, pseudo base, and chalcone forms which are predominant forms at higher pH conditions may be more effective in TPC and AA.

Color Values

The heat treatment temperature, heating time, and pH (statically data not shown for pH) were found important for the change in L^*, $h°$, and C^* values statistically ($p<0.05$) as seen in Table S3. L^* is a measure of color on the light-dark axis, so an increase in the L^* value indicates that the samples become bright [36]. Our findings showed that with the increase in heating temperature and time, BCC became clear for all buffer solutions and apple juice. For all pH values and apple juice during five hours of heat treatment, the $h°$ took a value between 12.61-71.15. This means that as the heat treatment time increased, there was a color change from a red hue to a yellow hue. h° is an attribute related to color tone or color [37]. The results regarding L^* and $h°$ value were like a study that reported the stability of anthocyanins in pomegranate juices and model solutions by Fischer et al. [38]. The C^* value relates to the color saturation or color intensity [37]. According to the results in Table S3, up to pH 5, the C^* value decreased with increasing process temperature and elapsed time. On the contrary, in buffer solutions with a pH value of 5, 6, and 7, the C^* value increased as the heating temperature raised, and the treatment time lengthens The color became less intense and clearer because of the increase in the C^* value. It was found that from pH 2 to pH 6, L^* and $h°$ increased by reducing the acidity of buffer solutions. Also, C^* increased when the environment was more alkaline. When the color of BCC in apple juice and pH 3 buffer system was compared, it was found that L^* and $h°$ were higher in apple juice at all temperatures. At C^*, a result like that of anthocyanin degradation was obtained. It was reported that the C^* value is closely related to anthocyanin degradation [39].

Prediction Results for Buffer System

The best network was selected depending on the performance parameters related to the forecasting capability and the specifications of the best network are given in Table 1. The selected network contains 15 neurons in the input layer, a hidden layer with 10 neurons, and 6 neurons in the output layer. The activation function of neurons in the hidden layer is selected as a hyperbolic tangent function and the activation function of neurons in the output layer is selected as a logistic function. When a network overfits, generally correlation coefficient of the training sample is higher than the correlation coefficients of testing and validation samples. As seen in Table 1, an overfitting problem was not observed.

The architecture of ANN in this study is given in Figure 1. There are several ways to produce information about the relative importance of the variables used in a neural network. In sensitivity analysis, it is tested how the neural network forecasts and, indirectly, the error rates would increase or decrease if each of its input variables were to change. During the analysis, if an important variable is undergone a change, the error will increase a great deal; if an unimportant variable is removed, the error will not increase very much [40]. Sensitivity analysis indicates the error change of the network when the input parameters are omitted. Values of sensitivity analysis of buffer solution were found as 240.81, 50.34, and 36.85 for pH, time, and temperature, respectively. The most important parameter depending on the error change is pH as seen in Figure 2. Prediction accuracy is most affected by pH. Furthermore, the Pearson correlation coefficient for the train, test, and validation samples were inspected, and a perfect fit was observed for all output parameters. The values of Pearson correlation coefficients vary between 0.96- 0.99.

Figure 3 represents the three-dimensional relations between the most important input parameter pH, predicted values of output parameters, and target values of output parameters. Figure 3 clearly indicates that pH, as the most important input parameter, is capable to explain all output parameters sufficiently by MLP 15-10-6. On the other hand, the selected network has a very low sum of square error values. In addition, we have seen that pH, temperature, and time have a significant effect on all these output

Table 1. Best network specifications for buffer solutions and apple juice

Network name	Training perf.	Testing perf.	Validation perf.	Hidden layer Activation function	Output layer Activation function	SSE Error for Training	SSE Error for Testing	SSE Error for Validation
MLP 15-10-6 (buffer solutions)	0.99	0.98	0.98	Hyperbolic tangent function	Logistic function	5.65	7.35	7.89
MLP 9-9-6 (apple juice)	0.94	0.94	0.95	Logistic	Logistic	2.29	1.41	1.26

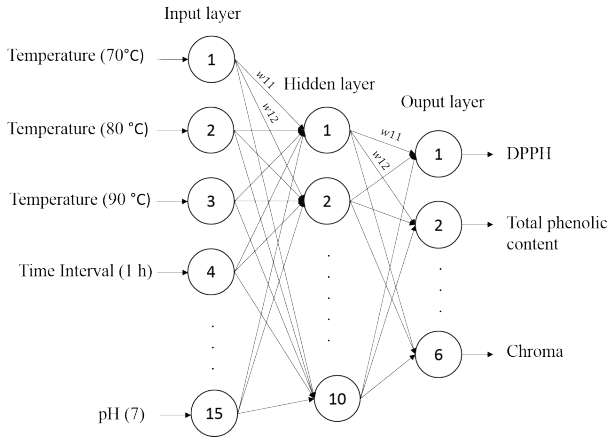

Figure 1. The architecture of the artificial neural network.

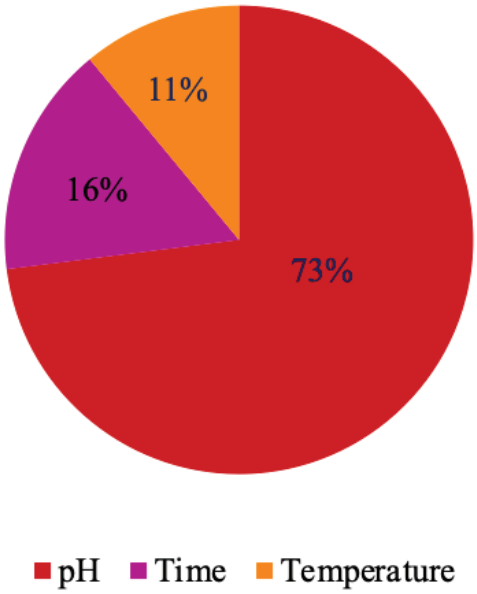

Figure 2. Importance of parameters due to the ANN.

variables. It is possible to estimate all selected outputs for the given combination of pH, temperature, and time.

Prediction Results for Apple Juice

To understand the functionality of the system in the food environment, another set of experiments was conducted. Apple juice with a fixed pH value of 3.04 was used. Same ANN modeling steps were applied to the buffer solution for a fair comparison. The pH value of apple juice is a fixed value (3.04), so it was not included in the input variable of ANN. Temperature with three categories and time with six categories were selected as inputs of the model and the same six outputs were predicted. The results are as follows.

As seen in Table 1, when the apple juice is used as a buffer, the performance of the model is decreased. On the other hand, the sum of square errors of training, testing, and validation samples are decreased and lower than handled with buffer solution as depicted in Table 1. This means that the food environment is more proper for predicting TAC.

Pearson correlation is a statistical parameter that measures the relationship between inputs and outputs. The Pearson correlation coefficients of training, testing, and validation samples are given in Table 1. As seen in Table 1, the performance of ANN increases when the pH is included in the analysis as an input parameter because of the interaction between pH and output parameters. On the other hand, while pH is 3.04, the sum of square errors related to training, testing and validation sets decreases. This result is a clear proof that when apple juice is used instead of buffer solution, ANN forecasts better.

Global sensitivity analysis was applied to detect which input is more important for the ANN model. Sensitivity values were 94.12 for time and 80.58 for the temperature of the apple juice. When the pH is 3.04, time becomes more critical for an accurate prediction. Although time is important, the temperature is almost as necessary as the time parameter for an accurate estimate.

In the literature, many studies try to predict the total AA and TPC of foods by ANN. Cabrera and Prieto [41] used ANN in their study to predict the antioxidant capacities of essential oils. They concluded that ANN was able to predict the antioxidant capacities of essential oils in both the DPPH and linoleic acid assays with high accuracy. Another study tried to predict the AA of teas by ANN. They concluded that prediction capability was very good in the testing data set since 0.4% relative errors were found [42]. In addition to these studies, Hosu et al. [43] trained ANNs to predict the AA of different wines. The relationships between the concentration of total phenolic, anthocyanins, tannins content, and flavonoids, associated with the AA, and the wine distinctive classes were predicted by ANN. They concluded that ANN was able to provide accurate predictions. Furthermore, ANN was compared with ANFIS for estimating AA and anthocyanin content at different ripening stages of sweet cherries. Researchers indicated that ANN outperforms ANFIS for this study [44]. Cheok et al. [45] optimized TPC extracted from mangosteen hull powder by using ANN and Response Surface Method (RSM). They concluded that ANN is a better modeling technique than RSM depending on the performance indicators. Besides these researches, the effect of thermal treatment, different acidity levels, and time on the anthocyanin content and degradation of grape skin, black carrot, and red cabbage were inspected by Ekici et al. [15]. They predict the relations by ANN and ANFIS and concluded that ANFIS performed better than ANN.

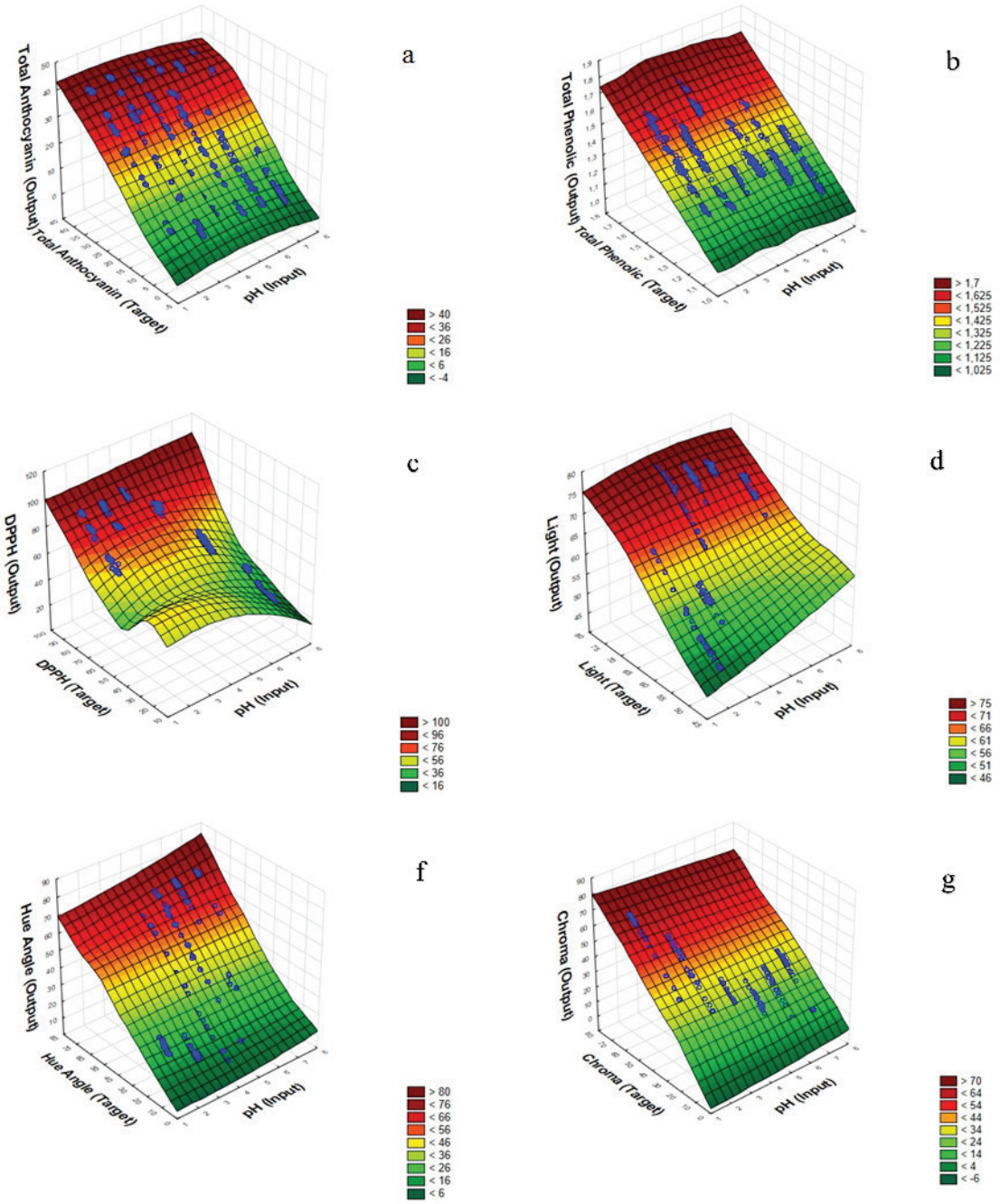

Figure 3. 3D Graphs for ph-outputs and targets relations (a: Total Anthocyanin- pH; b: Total Phenolic-pH; c: DPPH-pH; d: Light-pH, e: Hue Angle-pH; e: Hue Angle-pH, f: Chroma-pH).

CONCLUSION

Anthocyanin's stability is affected by its chemical structure, enzymes, temperature, light, pH, oxygen, ascorbic acid, sugars, metals, sulfur dioxide, and copigmentation. In this research, we inspected the effect of thermal treatment, different acidity levels, and time on TAC, TPC, AA, and color. Also, ANN was used to estimate the relation between TAC, TPC, AA, and color of *Berberis crataegina* and temperature, time, and pH for both apple juice and buffer solution. The results of this study showed that the stability of *Berberis* creteagina anthocyanin and color depended on temperature, heating time, and pH. Anthocyanin degradation was 83.25-94.5% at 90 °C in the range of pH 2-7. The evidence from this study points toward the idea that *Berberis* crategina juice can be exposed to loss of high anthocyanin and discoloration in a heat process that uses high temperatures. Also, the results about TPC and AA showed heat treatment applied to different pH systems can affect the bioactive

properties of *Berberis* crategina. Anthocyanins of *Berberis* crategina are more resistant to heat treatment in apple juice, suggesting that this fruit can be used as a colorant in fruit juices such as apple juice. The error values of the training, testing, and validation subsets of the ANN model are low, and the Pearson correlation coefficients (R) are quite high for our ANN model. On the other hand, the prediction accuracy of the ANN model for the food environment (apple juice with a pH = 3.04) is higher than the buffer solution. This indicated that pH is around 3 gives more suitable results for TAC, TPC, AA, and color. The performance of the ANN model in this study coincides with the performance of the studies in the literature. According to results of this research, ANN can also be further used to predict the changing bioactivity, anthocyanin, and color of other *Berberis* species under different conditions.

AUTHORSHIP CONTRIBUTIONS

Authors equally contributed to this work.

DATA AVAILABILITY STATEMENT

The authors confirm that the data that supports the findings of this study are available within the article. Raw data that support the finding of this study are available from the corresponding author, upon reasonable request.

CONFLICT OF INTEREST

The author declared no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

ETHICS

There are no ethical issues with the publication of this manuscript.

REFERENCES

[1] Baytop T. Türkiye'nin Tibbi ve Zehirli Bitkileri. Istanbul, Turkey; Istanbul Üniversitesi Yayınları; 1963.

[2] Demirci M, Tomas M, Tekin-Cakmak ZH, Karasu S. *Berberis crataegina* DC. as a novel natural food colorant source: ultrasound-assisted extraction optimization using response surface methodology and thermal stability studies. Food Sci Technol 2021;2061:1–8. [CrossRef]

[3] El-Merahbi R, Liu YN, Eid A, Daoud G, Hosry L, Monzer A, et al. *Berberis libanotica* ehrenb extract shows anti-neoplastic effects on prostate cancer stem/progenitor cells. PLoS One 2014;9:1–10. [CrossRef]

[4] Singh J, Kakkar P. Antihyperglycemic and antioxidant effect of *Berberis aristata* root extract and its role in regulating carbohydrate metabolism in diabetic rats. J Ethnopharmacol 2009;123:22–26. [CrossRef]

[5] Balasundram N, Sundram K, Samman S. Phenolic compounds in plants and agri-industrial by-products: Antioxidant activity, occurrence, and potential uses. Food Chem 2006;99:191–203. [CrossRef]

[6] Ongkowijoyo P, Luna-Vital DA, Gonzalez de Mejia E. Extraction techniques and analysis of anthocyanins from food sources by mass spectrometry: An update. Food Chem 2018;250:113–126. [CrossRef]

[7] Longo L, Vasapollo G. Extraction and identification of anthocyanins from Smilax aspera L. berries. Food Chem 2006;94:226–231. [CrossRef]

[8] Kong JM, Chia LS, Goh NK, Chia TF, Brouillard R. Analysis and biological activities of anthocyanins. Phytochemistry 2003;64:923–933. [CrossRef]

[9] Sun J, Cao X, Bai weibin, Liao X, Hu X. Comparative analyses of copigmentation of cyanidin 3-glucoside and cyanidin 3-sophoroside from red raspberry fruits. Food Chem 2010;120:1131–1137. [CrossRef]

[10] Hill T, Marquez L, O'Connor M, Remus W. Artificial neural network models for forecasting and decision making. Int J Forecast 1994;10:5–15. [CrossRef]

[11] Goñi SM, Oddone S, Segura JA, Mascheroni RH, Salvadori VO. Prediction of foods freezing and thawing times: Artificial neural networks and genetic algorithm approach. J Food Eng 2008;84:164–178. [CrossRef]

[12] Movagharnejad K, Nikzad M. Modeling of tomato drying using artificial neural network. Comput Electron Agric 2007;59:78–85. [CrossRef]

[13] Huang Y, Kangas LJ, Rasco BA. Applications of Artificial Neural Networks (ANNs) in food science. Crit Rev Food Sci Nutr 2007;47:113–126. [CrossRef]

[14] Balıkesir Univerity. Preparing buffer systems. Available at: http:// lisanskimya.balikesir.edu.tr/~f10501/tam.html 2008 Accessed on March 08, 2024.

[15] Ekici L, Simsek Z, Ozturk I, Sagdic O, Yetim H. Effects of temperature, time, and pH on the stability of anthocyanin extracts: Prediction of total anthocyanin content using nonlinear models. Food Anal Methods 2014;7:1328–1336. [CrossRef]

[16] Mónica Giusti M, Wrolstad RE. Characterization and Measurement of Anthocyanins by UV-visible Spectroscopy. Handb Food Anal Chem 2005;2:19–31.

[17] Hayta M, İşçimen EM. Optimization of ultrasound-assisted antioxidant compounds extraction from germinated chickpea using response surface methodology. LWT - Food Sci Technol 2017;77:208–216. [CrossRef]

[18] Orhan I, Kartal M, Naz Q, Ejaz A, Yilmaz G, Kan Y, et al. Antioxidant and anticholinesterase evaluation of selected Turkish Salvia species. Food Chem 2007;103:1247–1254. [CrossRef]

[19] Ekici L, Kafadar AD, Albayrak S. Physicochemical, sensory, and bioactive properties of some traditional Turkish sorbets. J Food Process Preserv 2018;42(6):e13664. [CrossRef]

[20] Nawi NM, Ransing MR, Ransing RS. An improved learning algorithm based on the Broyden-Fletcher-GoldfarbShanno (BFGS) method for back propagation neural networks. Proc - ISDA 2006 Sixth Int Conf Intell Syst Des Appl 2006;1:152–157. [CrossRef]

[21] Patras A, Brunton NP, O'Donnell C, Tiwari BK. Effect of thermal processing on anthocyanin stability in foods; mechanisms and kinetics of degradation. Trends Food Sci Technol 2010;21:3–11. [CrossRef]

[22] Farhadi Chitgar M, Aalami M, Maghsoudlou Y, Milani E. Comparative Study on the Effect of Heat Treatment and Sonication on the Quality of Barberry (*Berberis* Vulgaris) Juice. J Food Process Preserv 2017;41. [CrossRef]

[23] Kara Ş, Erçelebi EA. Thermal degradation kinetics of anthocyanins and visual colour of Urmu mulberry (Morus nigra L.). J Food Eng 2013;116:541–547. [CrossRef]

[24] Aşkın B, Küçüköner E. Factors Affecting the Stability of Anthocyanins. Turkish J Agric - Food Sci Technol 2019;7:1759–65. [CrossRef]

[25] Hou Z, Qin P, Zhang Y, Cui S, Ren G. Identification of anthocyanins isolated from black rice (Oryza sativa L.) and their degradation kinetics. Food Res Int 2013;50:691–697. [CrossRef]

[26] Gradinaru G, Biliaderis CG, Kallithraka S, Kefalas P, Garcia-Viguera C. Thermal stability of Hibiscus sabdariffa L. anthocyanins in solution and in solid state: Effects of copigmentation and glass transition. Food Chem 2003;83:423–436. [CrossRef]

[27] Nikkhah E, Khayamy M, Heidari R, Jamee R. Effect of sugar treatment on stability of anthocyanin pigments in berries. J Biol Sci 2007;7:1412–1417. [CrossRef]

[28] Boyer J, Liu RH. Apple phytochemicals and their health benefits. Nutr J 2004;3:1–15. [CrossRef]

[29] Karadeniz F, Ekşi A. Sugar composition of apple juices. Eur Food Res Technol 2002;215:145–148. [CrossRef]

[30] Li J, Li XD, Zhang Y, Zheng ZD, Qu ZY, Liu M, et al. Identification and thermal stability of purple-fleshed sweet potato anthocyanins in aqueous solutions with various pH values and fruit juices. Food Chem 2013;136:1429–1434. [CrossRef]

[31] Maillard MN, Soum MH, Boivin P, Berset C. Antioxidant activity of barley and malt: Relationship with phenolic content. LWT - Food Sci Technol 1996;29:238–244. [CrossRef]

[32] Sólyom K, Solá R, Cocero MJ, Mato RB. Thermal degradation of grape marc polyphenols. Food Chem 2014;159:361–366. [CrossRef]

[33] Nayak B, Berrios JDJ, Powers JR, Tang J. Thermal degradation of anthocyanins from purple potato (Cv. Purple Majesty) and impact on antioxidant capacity. J Agric Food Chem 2011;59:11040–11049. [CrossRef]

[34] Turturicə M, Stənciuc N, Bahrim G, Râpeanu G. Effect of thermal treatment on phenolic compounds from plum (prunus domestica) extracts - A kinetic study. J Food Eng 2016;171:200–207. [CrossRef]

[35] Sui X, Dong X, Zhou W. Combined effect of pH and high temperature on the stability and antioxidant capacity of two anthocyanins in aqueous solution. Food Chem 2014;163:163–170. [CrossRef]

[36] Lozano JE, Ibarz A. Colour changes in concentrated fruit pulp during heating at high temperatures. J Food Eng 1997;31:365–373. [CrossRef]

[37] Pathare PB, Opara UL, Al-Said FAJ. Colour measurement and analysis in fresh and processed foods: A review. Food Bioprocess Technol 2013;6:36–60. [CrossRef]

[38] Fischer UA, Carle R, Kammerer DR. Thermal stability of anthocyanins and colourless phenolics in pomegranate (Punica granatum L.) juices and model solutions. Food Chem 2013;138:1800–1809. [CrossRef]

[39] Kammerer DR, Schillmöller S, Maier O, Schieber A, Carle R. Colour stability of canned strawberries using black carrot and elderberry juice concentrates as natural colourants. Eur Food Res Technol 2007;224:667–679. [CrossRef]

[40] Homma T, Saltelli A. Importance measures in global sensitivity analysis of model output. Reliab Eng Sys Saf 1996;52:1–17. [CrossRef]

[41] Cabrera C, Lloris F, Giménez R, Olalla M, López MC. Mineral content in legumes and nuts: Contribution to the Spanish dietary intake. Sci Total Environ 2003;308:1–14. [CrossRef]

[42] Cimpoiu C, Cristea VM, Hosu A, Sandru M, Seserman L. Antioxidant activity prediction and classification of some teas using artificial neural networks. Food Chem 2011;127:1323–1328. [CrossRef]

[43] Hosu A, Cristea VM, Cimpoiu C. Analysis of total phenolic, flavonoids, anthocyanins and tannins content in Romanian red wines: Prediction of antioxidant activities and classification of wines using artificial neural networks. Food Chem 2014;150:113–118. [CrossRef]

[44] Taghadomi-Saberi S, Omid M, Emam-Djomeh Z, Ahmadi H. Evaluating the potential of artificial neural network and neuro-fuzzy techniques for estimating antioxidant activity and anthocyanin content of sweet cherry during ripening by using image processing. J Sci Food Agric 2014;94:95–101. [CrossRef]

[45] Cheok CY, Chin NL, Yusof YA, Talib RA, Law CL. Optimization of total phenolic content extracted from Garcinia mangostana Linn. hull using response surface methodology versus artificial neural network. Ind Crops Prod 2012;40:247–253. [CrossRef]

Table S1. Thermal degradation of *Berberis crataegina* DC. anthocyanins

pH	Time (h)	Anthocyanin (mg cyd-3-glc/L)		
		70°C	**80°C**	**90°C**
2	**0**	40.51 ± 0.82^{Aa}	39.12 ± 0.64^{Ba}	39.90 ± 0.56^{ABa}
	1	38.27 ± 0.39^{Ab}	33.30 ± 0.24^{Bb}	24.59 ± 0.31^{Cb}
	2	32.68 ± 1.19^{Ac}	29.55 ± 0.38^{Bc}	16.87 ± 0.38^{Cc}
	3	32.77 ± 0.59^{Ac}	25.13 ± 0.65^{Bd}	11.90 ± 0.45^{Cd}
	4	31.59 ± 0.54^{Ad}	21.75 ± 0.19^{Be}	8.69 ± 0.14^{Ce}
	5	29.08 ± 0.37^{Ae}	18.55 ± 0.15^{Bf}	6.69 ± 0.39^{Cf}
	Degradation (%)	**28.22**	**52.58**	**83.25**
3	**0**	39.99 ± 0.36^{Ba}	40.99 ± 0.21^{Aa}	40.17 ± 0.50^{Bab}
	1	36.06 ± 0.27^{Ab}	31.47 ± 0.35^{Bb}	23.22 ± 1.87^{Cbc}
	2	30.65 ± 1.91^{Ac}	26.52 ± 0.34^{Bc}	11.79 ± 0.29^{Cab}
	3	29.92 ± 0.40^{Acd}	20.49 ± 0.37^{Bd}	4.60 ± 1.03^{Ca}
	4	31.02 ± 1.04^{Ac}	15.97 ± 0.91^{Be}	3.20 ± 0.48^{Cc}
	5	28.35 ± 1.69^{Ad}	11.54 ± 0.13^{Bf}	2.21 ± 0.41^{Cd}
	Degradation (%)	**29.13**	**71.85**	**94.51**
4	**0**	39.82 ± 0.36^{Ba}	36.88 ± 0.27^{Ca}	40.44 ± 0.07^{Aa}
	1	34.16 ± 1.46^{Ab}	29.71 ± 0.36^{Bb}	24.08 ± 0.27^{Cb}
	2	31.47 ± 0.99^{Bc}	25.33 ± 0.23^{Bc}	16.15 ± 0.66^{Cc}
	3	29.35 ± 0.67^{Ad}	21.34 ± 0.38^{Bd}	10.36 ± 0.28^{Cd}
	4	26.91 ± 0.37^{Ae}	18.40 ± 0.08^{Be}	6.68 ± 0.10^{Ce}
	5	23.64 ± 0.36^{Aef}	15.66 ± 0.22^{Bf}	5.32 ± 0.26^{Cf}
	Degradation (%)	**40.63**	**57.54**	**86.85**
5	**0**	41.17 ± 0.59^{Ca}	37.20 ± 0.22^{Ba}	42.80 ± 0.49^{Aa}
	1	31.40 ± 1.34^{Ab}	27.12 ± 0.45^{Bb}	19.39 ± 0.52^{Cb}
	2	28.66 ± 0.21^{Ac}	21.51 ± 0.38^{Bc}	10.70 ± 0.60^{Cc}
	3	27.41 ± 0.65^{Ad}	18.08 ± 0.31^{Bd}	6.16 ± 0.37^{Cd}
	4	25.17 ± 0.61^{Ae}	14.70 ± 0.45^{Be}	3.85 ± 0.29^{Ce}
	5	22.91 ± 0.11^{Af}	13.08 ± 0.45^{Bf}	2.63 ± 0.58^{Cf}
	Degradation (%)	**44.36**	**64.84**	**93.85**
6	**0**	37.91 ± 0.61^{Ba}	36.56 ± 0.32^{Ca}	40.48 ± 0.69^{Aa}
	1	25.45 ± 0.28^{Ab}	19.54 ± 0.55^{Bb}	12.70 ± 0.38^{Cb}
	2	20.20 ± 0.30^{Ac}	14.63 ± 0.17^{Bc}	7.36 ± 0.20^{Cc}
	3	17.99 ± 0.34^{Ad}	11.07 ± 0.76^{Bd}	4.05 ± 0.43^{Cd}
	4	16.04 ± 0.23^{Ae}	8.46 ± 0.57^{Be}	2.92 ± 0.23^{Ce}
	5	14.14 ± 0.12^{Af}	5.81 ± 0.14^{Bf}	2.60 ± 0.17^{Cf}
	Degradation (%)	**62.7**	**84.11**	**93.58**
7	**0**	40.92 ± 1.00^{Aa}	34.80 ± 0.23^{Ca}	32.11 ± 0.28^{Ba}
	1	12.99 ± 0.22^{Ab}	9.57 ± 0.11^{Bb}	5.86 ± 0.97^{Cb}
	2	9.36 ± 0.26^{Ac}	6.20 ± 0.07Bc	3.22 ± 0.55^{Cc}
	3	6.53 ± 0.15^{Ad}	4.66 ± 0.25^{Bd}	2.84 ± 0.70^{Cc}
	4	4.46 ± 0.25^{Ae}	3.40 ± 0.20^{Be}	1.52 ± 0.25^{Cd}
	5	3.59 ± 0.23^{Af}	2.31 ± 0.32^{Bf}	1.90 ± 0.38^{Bd}
	Degradation (%)	**91.23**	**93.36**	**94.09**
Apple Juice (pH 3.04)	**0**	40.53 ± 3.78^{Aba}	38.75 ± 0.42^{Ba}	41.79 ± 0.90^{Aa}
	1	32.41 ± 0.38^{Ab}	27.93 ± 0.59^{Bb}	20.72 ± 0.82^{Cb}
	2	29.39 ± 0.51^{Ac}	24.51 ± 0.17^{Bc}	13.92 ± 0.70^{Cc}
	3	27.05 ± 0.26^{Ad}	20.88 ± 0.53^{Bd}	9.72 ± 0.28^{Cd}
	4	25.45 ± 0.50^{Ae}	18.58 ± 0.30^{Be}	6.60 ± 0.24^{Ce}
	5	23.32 ± 0.42^{Af}	15.61 ± 0.67^{Bf}	4.77 ± 0.73^{Cf}
	Degradation (%)	**42.45**	**59.72**	**88.59**

Capital letters on the same line are a comparison of the process temperature. There is no statistically significant difference between the samples symbolized with the same letters ($p<0.05$). Small letters in the same column are the comparison of heating time. There is no statistically significant difference between the samples symbolized with the same letters ($p<0.05$). Mean±Standard deviation, n=6.

Table S2. The changing of *Berberis crataegina* DC. TPC and AA

pH	Time (h)	TPC (mg GAE/L)			DPPH (% inhibition)		
		70°C	80°C	90°C	70°C	80°C	90°C
2	0	1.54±0.04^{Aa}	1.32±0.04^{Bc}	1.60±0.03^{Ca}	88.28±0.60^{Ac}	79.80±0.38^{Bd}	67.22±2.96^{Ca}
	1	1.55±0.03^{Aa}	1.32±0.02^{Bc}	1.58±0.02^{Aa}	89.20±0.46^{Abc}	80.95±0.61^{Bc}	68.48±0.94^{Ca}
	2	1.46±0.04^{ABb}	1.41±0.03^{Bab}	1.50±0.04^{Ab}	89.30±0.59^{Ab}	82.04±0.53^{Bb}	68.77±2.21^{Ca}
	3	1.53±0.04^{Aa}	1.39±0.03^{Bb}	1.25±0.01^{Cc}	89.70±0.46^{Aab}	83.83±0.73^{Ba}	62.38±1.20^{Cb}
	4	1.38±0.02^{Ac}	1.39±0.04^{Aab}	1.26±0.02^{Bc}	89.92±0.71^{Aab}	83.42±0.69^{Ba}	67.14±2.14^{Cab}
	5	1.42±0.02^{Abc}	1.41±0.01^{Aa}	1.22±0.02^{Bd}	90.62±0.57^{Aa}	84.12±0.74^{Ba}	65.68±3.36^{Cab}
3	0	1.51±0.03^{Aab}	1.43±0.02^{Ba}	1.35±0.02^{Ca}	90.62±0.22^{Aa}	90.29±1.26^{Aab}	88.68±0.68^{Ba}
	1	1.49±0.01^{Abc}	1.42±0.02^{Ba}	1.29±0.01^{Cb}	90.66±0.46^{Aa}	90.48±0.20^{Aa}	87.62±0.86^{Ba}
	2	1.51±0.02^{Aab}	1.39±0.03^{Bb}	1.25±0.02^{Cc}	90.81±0.32^{Aa}	89.12±0.43^{Bbc}	85.73±0.69^{Cb}
	3	1.52±0.03^{Aa}	1.38±0.01^{Bbc}	1.21±0.02^{Cd}	91.05±0.23^{Aa}	88.44±0.54^{Bc}	81.92±1.35^{Cc}
	4	1.47±0.01^{Ac}	1.38±0.01^{Bbc}	1.19±0.02^{Cd}	91.09±0.57^{Aa}	90.28±0.87^{Aab}	79.98±0.54^{Bd}
	5	1.43±0.02^{Ad}	1.36±0.01^{Bc}	1.30±0.02^{Cb}	90.61±0.20^{Aa}	90.50±0.52^{Aa}	76.24±0.43^{Be}
4	0	1.66±0.03^{Aac}	1.31±0.01^{Ba}	1.35±0.03^{Ba}	91.70±0.32^{Bab}	92.29±0.22^{Ab}	90.43±0.40^{Ca}
	1	1.59±0.03^{Ab}	1.29±0.01^{Bb}	1.31±0.01^{Bab}	91.75±0.37^{Aa}	91.44±0.58^{Aa}	90.43±0.60^{Ba}
	2	1.57±0.03^{Abc}	1.29±0.01^{Bb}	1.30±0.02^{Bbc}	91.81±0.10^{Aa}	91.67±0.16^{Aa}	89.19±0.43^{Bb}
	3	1.59±0.02^{Ab}	1.28±0.01^{Bc}	1.29±0.02^{Bbc}	91.65±0.20^{Aa}	91.21±0.28^{Ba}	88.39±0.22^{Cc}
	4	1.57±0.03^{Abc}	1.25±0.02^{Bd}	1.27±0.01^{Bc}	91.67±0.12^{Aa}	91.20±0.28^{Ba}	87.05±0.30^{Cd}
	5	1.54±0.01^{Ac}	1.25±0.02^{Bd}	1.27±0.02^{Bc}	91.53±0.06^{Ab}	91.46±0.26^{Aa}	86.96±0.63^{Bd}
5	0	1.27±0.02^{Ca}	1.31±0.02^{Bb}	1.35±0.01^{Aa}	76.85±0.59^{Bd}	79.47±0.52^{Abc}	79.60±0.17^{Ac}
	1	1.32±0.03^{Aab}	1.30±0.01^{Ab}	1.33±0.03^{Aab}	77.78±0.70^{Bc}	80.46±0.69^{Aa}	80.90±0.47^{Aa}
	2	1.33±0.02^{Aab}	1.30±0.01^{Ab}	1.31±0.05^{Aab}	81.96±0.92^{Aa}	80.04±0.22^{Bab}	79.82±0.38^{Bc}
	3	1.34±0.01^{Bb}	1.45±0.05^{Aa}	1.28±0.01^{Cb}	82.28±1.01^{Aa}	80.12±0.43^{Cab}	80.67±0.25^{Bab}
	4	1.32±0.01^{Bb}	1.47±0.05^{Aa}	1.29±0.03^{Bb}	81.64±0.30^{Aa}	79.42±0.38^{Cbc}	80.35±0.38^{Bab}
	5	1.29±0.02^{Bab}	1.47±0.02^{Aa}	1.32±0.02^{Bab}	79.48±0.26^{Bb}	78.69±0.45^{Cc}	80.45±0.33^{Ab}
6	0	1.30±0.01^{Ba}	1.52±0.05^{Aa}	1.31±0.02^{Ba}	57.89±0.74^{Cb}	60.86±0.67^{Bbc}	64.56±0.62^{Aa}
	1	1.27±0.02^{Bb}	1.43±0.02^{Ab}	1.22±0.01^{Cb}	58.06±0.46^{Bb}	62.40±0.50^{Aa}	54.82±0.88^{Ccd}
	2	1.28±0.01^{Bb}	1.41±0.04^{Ab}	1.22±0.01^{Cb}	57.48±1.12^{Bb}	61.53±0.39^{Ab}	53.66±0.79^{Cd}
	3	1.25±0.01^{Bcd}	1.33±0.03^{Ac}	1.20±0.01^{Cc}	62.55±1.74^{Aa}	58.37±0.71^{Cc}	59.87±0.90^{Bb}
	4	1.25±0.01^{Ac}	1.24±0.01^{Ad}	1.21±0.01^{Ad}	62.10±1.19^{Aa}	60.56±0.68^{Bd}	55.77±0.75^{Cc}
	5	1.23±0.01^{Ad}	1.23±0.02^{Ad}	1.23±0.03^{Ad}	62.43±1.35^{Aa}	58.51±0.99^{Bd}	53.94±1.00^{Cd}
7	0	1.26±0.02^{Ca}	1.33±0.03^{Bb}	1.38±0.01^{Aa}	44.68±1.64^{Aa}	43.00±1.37^{Ab}	35.51±0.40^{Bab}
	1	1.18±0.03^{Bb}	1.23±0.02^{Ac}	1.20±0.02^{ABd}	42.46±3.00^{Aa}	44.30±0.80^{Aab}	33.06±0.71^{Bcd}
	2	1.16±0.01^{Cb}	1.19±0.02^{Bd}	1.22±0.01^{Ac}	26.92±1.59^{Cbc}	44.03±0.96^{Aab}	31.85±0.86^{Bd}
	3	1.16±0.01^{Cb}	1.38±0.03^{Aa}	1.31±0.01^{Bb}	27.54±2.71^{Cbc}	44.82±1.25^{Aab}	36.05±1.24^{Ba}
	4	1.16±0.02^{Cb}	1.32±0.03^{Ab}	1.23±0.00^{Bc}	25.09±1.83^{Cc}	44.88±0.90^{Aa}	34.21±1.14^{Bbc}
	5	1.12±0.02^{Cc}	1.38±0.03^{Aa}	1.30±0.02^{Bb}	27.60±1.96^{Ab}	43.44±0.95^{Bab}	34.91±1.01^{Cab}
Apple juice	0	3.92±0.09^{Aa}	3.79±0.23^{Ab}	3.98±0.04^{Ac}	88.38±0.31^{Bb}	89.24±0.52^{Aa}	87.84±0.62^{Ba}
(pH 3.04)	1	3.89±0.08^{Ba}	3.86±0.12^{Bb}	4.37±0.10^{Ab}	86.27±2.00^{Ac}	86.75±0.34^{Abc}	86.02±0.51^{Ab}
	2	3.68±0.13^{Cb}	3.97±0.22^{Bb}	4.35±0.15^{Ab}	89.30±0.83^{Aa}	86.47±0.80^{Bcd}	86.55±1.36^{Bab}
	3	3.72±0.07^{Bb}	4.32±0.07^{Aa}	4.40±0.07^{Ab}	89.21±1.61^{Aab}	87.99±0.95^{Aab}	84.50±3.58^{Bb}
	4	3.73±0.06^{Cb}	4.38±0.16^{Ba}	4.86±0.10^{Aa}	89.44±1.12^{Aab}	85.33±1.00^{Bd}	85.63±2.22^{Bb}
	5	3.67±0.07^{Cb}	4.39±0.13^{Ba}	4.79±0.13^{Aa}	89.11±0.50^{Aa}	86.03±0.76^{Ccd}	87.53±1.13^{Ba}

Capital letters on the same line are a comparison of the process temperature. There is no statistically significant difference between the samples symbolized with the same letters ($p<0.05$). Small letters in the same column are the comparison of heating time. There is no statistically significant difference between the samples symbolized with the same letters ($p<0.05$). Mean±Standard deviation, n=6.

Table S3. The changing of *Berberis crataegina* DC. color values (L^*, $h°$, and C^*)

pH	Time (h)	L^*			$h°$			C^*		
		70°C	80°C	90°C	70°C	80°C	90°C	70°C	80°C	90°C
2	0	47.48±0.07^{Cf}	48.04±0.02^{Bf}	49.43±0.04^{Af}	23.17±0.08^{Cc}	23.34±0.04^{Ba}	24.07±0.03^{Af}	68.68±0.05^{Ca}	68.77±0.06^{Ba}	69.16±0.03^{Aa}
	1	49.09±0.07^{Ce}	51.03±0.05^{Be}	56.89±0.07^{Ae}	23.48±0.08^{Aab}	22.78±0.01^{Bb}	21.32±0.02^{Ce}	68.19±0.01^{Ab}	66.76±0.01^{Bb}	59.22±0.06^{Cb}
	2	50.44±0.11^{Cd}	53.17±0.05^{Bd}	60.63±0.06^{Ad}	23.60±0.17^{Aa}	22.29±0.07^{Bc}	21.02±0.01^{Cd}	67.97±0.12^{Ac}	64.78±0.06^{Bc}	51.52±0.05^{Cc}
	3	51.51±0.01^{Cc}	54.78±0.07^{Bc}	63.20±0.49^{Ac}	23.41±0.02^{Ab}	20.92±0.08^{Bd}	22.40±0.12^{Cc}	67.22±0.05^{Ad}	61.55±0.05^{Bd}	44.27±0.31^{Cd}
	4	52,.07±0.02^{Cb}	55.33±0.02^{Bb}	64.67±0.05^{Ab}	22.91±0.03^{Cd}	18.92±0.09^{Be}	24.52±0.07^{Ab}	66.19±0.15^{Ae}	57.78±0.08^{Be}	38.74±0.07^{Ce}
	5	52.81±0.02^{Ca}	56.97±0.10^{Ba}	66.12±0.29^{Aa}	22.55±0.03^{Ce}	19.41±0.08^{Bf}	25.84±0.54^{Aa}	65.43±0.03^{Af}	55.49±0.14^{Bf}	35.16±0.50^{Cf}
3	0	54.54±0.02^{Be}	53.00±0.03^{Cf}	55.91±0.05^{Ae}	14.38±0.02^{Ad}	13.68±0.02^{Bd}	13.56±0.01^{Cf}	56.11±0.01^{Ba}	56.91±0.02^{Aa}	53.87±0.04^{Ca}
	1	56.43±0.03^{Cd}	56.79±0.08^{Bd}	60.64±0.02^{Aa}	15.73±0.09^{Bc}	15.66±0.01^{Bb}	18.50±0.08^{Ae}	54.24±0.06^{Ab}	52.27±0.11^{Bb}	41.86±0.07^{Cb}
	2	57.24±0.10^{Cc}	58.50±0.06^{Ba}	59.53±0.10^{Ab}	16.10±0.07^{Cb}	16.64±0.01^{Ba}	19.15±0.01^{Ad}	52.47±0.07^{Ac}	47.92±0.30^{Bc}	35.63±0.10^{Cc}
	3	58.28±0.14^{Ab}	57.77±0.08^{Bb}	58.24±0.05^{Ad}	16.27±0.04^{Bb}	15.27±0.21^{Cc}	19.81±0.03^{Ac}	50.58±0.16^{Ad}	44.18±0.15^{Bd}	31.67±0.04^{Cd}
	4	58.51±0.14^{Aab}	56.01±0.32^{Ce}	58.11±0.21^{Bd}	16.20±0.05^{Ba}	13.43±0.05^{Ce}	21.18±0.03^{Ab}	49.19±0.03^{Ae}	40.86±0.50^{Be}	28.55±0.10^{Ce}
	5	58.59±0.06^{Ba}	56.94±0.03^{Cc}	58.95±0.12^{Ac}	16.14±0.01^{Ba}	14.96±0.25^{Cc}	23.29±0.12^{Aa}	46.74±0.02^{Af}	37.12±0.07^{Bf}	26.60±0.15^{Cf}
4	0	63.89±0.06^{Cf}	64.73±0.32^{Bf}	65.88±0.02^{Af}	8.61±0.05^{Bf}	7.98±0.04^{Cf}	11.13±0.05^{Af}	34.61±0.08^{Aa}	33.17±0.07^{Ba}	30.96±0.01^{Ca}
	1	67.80±0.04^{Cd}	69.51±0.02^{Ae}	72.30±0.12^{Be}	17.79±0.11^{Ce}	21.59±0.05^{Be}	38.98±0.21^{Ae}	30.89±0.03^{Ac}	29.16±0.03^{Bb}	26.60±0.11^{Cb}
	2	67.23±0.07^{Ce}	71.28±0.02^{Bd}	74.21±0.00^{Ad}	18.18±0.10^{Cd}	28.93±0.07^{Bd}	49.17±0.16^{Ad}	31.67±0.05^{Ab}	27.41±0.01^{Bc}	25.98±0.01^{Cd}
	3	68.52±0.01^{Cc}	72.53±0.05^{Bc}	75.84±0.09^{Ac}	22.00±0.12^{Cc}	34.65±0.12^{Bc}	56.69±0.47^{Ac}	30.15±0.02^{Ad}	26.32±0.04^{Bd}	25.84±0.04^{Ce}
	4	69.23±0.02^{Cb}	72.62±0.02^{Bb}	75.99±0.01^{Ab}	24.56±0.08^{Cb}	36.54±0.10^{Bb}	59.67±0.25^{Ab}	29.44±0.05^{Ae}	26.02±0.01^{Be}	26.46±0.05^{Cc}
	5	69.88±0.04^{Ca}	74.01±0.10^{Ba}	77.71±0.38^{Aa}	27.24±0.13^{Ca}	42.42±0.15^{Ba}	65.62±0.75^{Aa}	28.66±0.03^{Af}	25.18±0.03^{Bf}	26.31±0.19^{Cc}
5	0	69.99±0.06^{Bf}	70.94±0.01^{Ae}	69.48±0.00^{Ce}	12.61±0.09^{Cf}	17.28±0.03^{Af}	17.20±0.05^{Bf}	19.99±0.03^{Ae}	18.64±0.01^{Bf}	19.23±0.00^{Cf}
	1	71.82±0.03^{Ce}	73.40±0.03^{Ae}	73.05±0.14^{Bd}	27.82±0.04^{Ce}	45.58±0.07^{Be}	56.38±0.18^{Ae}	20.01±0.01^{Ce}	20.81±0.01^{Be}	23.98±0.20^{Ae}
	2	72.52±0.01^{Bd}	74.02±0.05^{Ad}	73.86±0.14^{Ac}	34.82±0.12^{Cd}	52.99±0.07^{Bd}	62.93±1.30^{Ad}	20.17±0.02^{Cd}	22.16±0.05^{Bd}	26.64±0.44^{Ad}
	3	72.86±0.10^{Bc}	74.51±0.02^{Ac}	74.69±0.19^{Ab}	39.98±0.39^{Cc}	57.31±0.23^{Bc}	68.27±0.08^{Ac}	20.47±0.05^{Cc}	23.28±0.06^{Bc}	29.42±0.13^{Ac}
	4	73.24±0.24^{Cb}	74.80±0.14^{Bb}	75.47±0.15^{Aa}	45.04±0.59^{Cb}	61.11±0.44^{Bb}	70.31±0.01^{Ab}	20.88±0.04^{Cb}	24.29±0.15^{Bb}	30.78±0.01^{Ab}
	5	73.76±0.06^{Ca}	74.89±0.13^{Ba}	75.50±0.03^{Aa}	48.32±0.35^{Ca}	62.56±0.14^{Ba}	71.15±0.01^{Aa}	21.37±0.00^{Ca}	25.31±0.03^{Ba}	31.70±0.12^{Aa}
6	0	72.92±0.02^{Bd}	73.34±0.04^{Ad}	72.65±0.07^{Cd}	29.22±0.12^{Cf}	33.39±0.11^{Bf}	34.20±0.15^{Af}	13.09±0.00^{Cf}	13.18±0.02^{Bf}	13.28±0.01^{Af}
	1	73.85±0.00^{Bb}	74.22±0.01^{Ac}	73.40±0.01^{Cc}	54.37±0.07^{Ce}	63.30±0.05^{Be}	67.69±0.01^{Ae}	17.49±0.02^{Ce}	21.97±0.01^{Be}	26.88±0.02^{Ae}
	2	73.87±0.00^{Ba}	74.34±0.00^{Ab}	73.93±0.08^{Bb}	59.58±0.05^{Cd}	66.56±0.02^{Bd}	70.11±0.04^{Ad}	19.73±0.01^{Cd}	24.42±0.02^{Bd}	30.41±0.25^{Ad}
	3	73.81±0.04^{Bc}	74.64±0.23^{Aa}	74.48±0.03^{Aa}	62.38±0.01^{Cc}	68.26±0.00^{Bc}	71.01±0.03^{Ac}	21.44±0.00^{Cc}	26.24±0.28^{Bc}	31.76±0.20^{Ac}
	4	73.85±0.08^{Bbc}	74.85±0.19^{Aa}	73.84±0.03^{Bb}	64.26±0.03^{Cb}	69.43±0.04^{Bb}	70.85±0.04^{Ab}	22.85±0.07^{Cb}	27.02±0.44^{Bb}	33.00±0.02^{Ab}
	5	73.84±0.02^{Bbc}	74.87±0.07^{Aa}	75.12±0.47^{Aa}	65.22±0.01^{Ca}	70.21±0.03^{Ba}	71.32±0.00^{Aa}	23.77±0.04^{Ca}	29.09±0.04^{Ba}	33.89±0.16^{Aa}
7	0	70.79±0.01^{Ad}	67.36±0.02^{Bd}	64.38±0.04^{Cc}	62.56±0.17^{Af}	53.56±0.06^{Cf}	56.87±0.09^{Be}	12.31±0.04^{Af}	11.30±0.00^{Cf}	11.90±0.01^{Be}
	1	70.48±0.02^{Af}	69.70±0.03^{Bb}	67.86±0.29^{Ca}	68.58±0.01^{Be}	68.66±0.05^{Bb}	68.99±0.14^{Ab}	23.51±0.04^{Ce}	26.89±0.09^{Be}	31.47±0.87^{Ad}
	2	70.68±0.01^{Ae}	70.20±0.11^{Ba}	67.68±0.53^{Ca}	69.04±0.01^{Ad}	69.06±0.01^{Aa}	68.99±0.11^{Ab}	26.12±0.01^{Cd}	29.11±0.06^{Bd}	33.35±1.34^{Acd}
	3	70.88±0.05^{Ac}	69.73±0.05^{Bb}	68.21±0.83^{Ca}	69.31±0.02^{Bc}	68.39±0.01^{Cc}	70.27±0.05^{Aa}	28.12±0.06^{Cc}	29.61±0.11^{Bc}	33.01±0.36^{Ab}
	4	71.17±0.04^{Ab}	69.54±0.01^{Bcb}	67.50±0.43^{Ca}	69.71±0.09^{Aa}	68.12±0.01^{Bd}	68.37±0.43^{Bc}	30.09±0.12^{Cb}	31.04±0.33^{Bb}	36.06±0.32^{Ab}
	5	71.29±0.04^{Aa}	69.53±0.70^{Bb}	66.93±0.06^{Cb}	69.40±0.05^{Ab}	68.02±0.02^{Be}	67.92±0.00^{Cd}	30.57±0.12^{Ca}	32.53±0.13^{Ba}	36.55±0.20^{Aa}
Apple Juice (pH 3.04)	0	53.91±0.06^{Cf}	54.43±0.07^{Bf}	55.79±0.02^{Af}	26.97±0.07^{Cf}	28.06±0.02^{Bf}	28.70±0.02^{Af}	50.97±0.07^{Ba}	51.23±0.06^{Aa}	50.09±0.04^{Ca}
	1	56.41±0.04^{Ce}	59.38±0.01^{Be}	63.24±0.15^{Ae}	30.66±0.04^{Ce}	36.14±0.08^{Be}	43.80±0.10^{Ae}	49.47±0.03^{Ab}	47.81±0.06^{Bb}	44.38±0.07^{Cb}
	2	57.30±0.04^{Cd}	60.24±0.03^{Bd}	65.08±0.02^{Ad}	32.50±0.03^{Cd}	38.33±0.12^{Bd}	49.63±0.09^{Ad}	48.29±0.08^{Cc}	46.32±0.01^{Bc}	42.69±0.02^{Ad}
	3	58.17±0.02^{Cc}	61.47±0.03^{Bc}	66.35±0.28^{Ac}	34.59±0.03^{Cc}	41.33±0.13^{Bc}	54.78±0.38^{Ac}	47.43±0.02^{Ad}	44.79±0.06^{Bd}	42.31±0.15^{Ce}
	4	58.99±0.09^{Cb}	62.56±0.01^{Bb}	67.18±0.13^{Ab}	36.23±0.02^{Cb}	44.68±0.04^{Bb}	58.89±0.21^{Ab}	46.55±0.08^{Ae}	43.92±0.07^{Be}	42.35±0.05^{Ce}
	5	59.68±0.06^{Ca}	63.18±0.06^{Ba}	67.73±0.18^{Aa}	37.99±0.02^{Ca}	47.17±0.03^{Ba}	62.15±0.01^{Aa}	45.77±0.07^{Af}	43.29±0.09^{Bf}	42.96±0.21^{Cc}

Capital letters on the same line are a comparison of the process temperature. There is no statistically significant difference between the samples symbolized with the same letters ($p<0.05$). Small letters in the same column are the comparison of heating time. There is no statistically significant difference between the samples symbolized with the same letters ($p<0.05$). Mean±Standard deviation, n=6