The Intake of Red Cabbage Anthocyanines in Ice-Cream

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Abstract: The present study aimed to extract anthocyanines from red cabbage for coloring the ice-cream instead of the synthetic colors to avoid the harmful effect on the health. Also, the study aimed to study its stability under the effect of some factors (pH-values – temperatures – both of them). Anthocyanine pigments were stable at low pH-values. The degradation percentage was increased by increasing in the pH-values. Red cabbage anthocyanin was stable at 50 and 70°C, but it being degradation with high ratio after 75°C. Acidified extract was more stable than citric acid (2%) extract. The results showed that the use of acidified extract was the best coloring agent to color ice-cream.


1. Introduction
Food colorants play a very important role in enhancing the aesthetic appeal of Food. The practice of adding colors to food dates back to ancient times. One of the earliest records of the use of food colorants is that of the coloring of wine as early as 400 BC. Until the discovery of the first synthetic dye is 1856 by Sir/ William Henry Perkins, mankind had been relying on pigments extracted from plants, animals and minerals as food colorants. Since then, due to the superior of synthetic dyes with respect to tin cortical strength, line stability and low cost, synthetic dyes are used more extensively than the natural extracts in the coloring of food (Yuan – Kun and Hwee-Peng, 2002).

Anthocyanins are well known as water – soluble-colorants. Anthocyanins are almost ubiquitous in the plant kingdom and are responsible for most of the red, blue and purple colors of flowers and fruits. These water-soluble pigments are also widely used in the food industry to color many dairy products (Ginat, 2008).

Red cabbage has been shown to be a very interesting anthocyanin source for coloration of soft drinks due to: (1) extended pH – region of coloration compared to other anthocyanin sources, providing natural blue color in neutral solution; (2) low sensitivity to photo – degradation from pH 3.7 and (3) excellent heat stability allowing heat treatment of colored products with only limited loss of color (Dyrby, et.al, 2001).

The objective of this study is as follows:
(1) Extraction of anthocyanins from red cabbage by two methods and report the best method for extraction.
(2) Determination the effects of pH value, temperature and time on the stability of the extracted pigments.
(3) Evaluation the possibility of using the isolated anthocyanins as natural colorants for ice-cream.
(4) Production of the food product with high nutrition value. Children love it and richer by antioxidants (anthocyanins) as well as without artificial colors.

2. Material and Methods
Materials:
1- Red cabbage, sugar, skim milk powder (low fat) used in this study was made in Finland (Vallo) and pure coconut oil were purchased from local market.
2- All chemicals were obtained from Future Time Est. Jaddah, KSA.

Methods:
The natural red pigment (anthocyanins) was extracted by two methods as follows:-
(1) Ethanol acidified with HCl 1% (Bilykm, 1979).
(2) Citric acid solution 2% (Du and Francis, 1975).

In each extraction trail 100g of red cabbage was mixed with 500 ml of solvent extract at ambit temperature (25°C ± 5°C) overnight. Then, it was filtered and concentrated by rotary evaporator at 40°C (Franci, 2000).

Determination of Total Anthocyanins:
A small aliquot of the concentrated extract was diluted with the extracting solvent to yield the optical density. This diluted extract was put in dark bottle and left for 2 hrs after this period; the extract absorbance was measured by spectrophotometer at 520nm, (Giustic and Wrolstad, 2005).

Total anthocyanins pigment (mg/L) = (A x MW x DF x 1000) / (E x 1)
Where: A is the absorption value obtained at 520nm; MW is the molecular weight of anthocyanin (494 g/mol); DF is the dilution factor and χ is the molar absorptive (26,300 L/mol).

**Determination of factors affecting on anthocyanin stability:**
1. Effect of different pH values ranged from 3 at 9 at room temperature (25°C ± 5°C).
2. Effect of different temperatures (50, 75 and 90°C).
3. Effect of different pH values and different temperatures on the stability of anthocyanins (Castellar *et al.*, 2003).

**Preparation of Ice-cream:**
Ice-cream was prepared to contain 8% fat (pure coconut oil), 15% sucrose (sugar), 0.25% CMC as stabilizer, 5 ml and 10 ml concentrated anthocyanins and 5 ml of strawberry flavoring agent. All mixes were pasteurized at 80°C for 30 second as given by Arbuckile (1986). The mixes were cooled to 5°C and kept at this temperature for 4 hours prior to freezing. After aging each batch was frozen in an experimental ice-cream freezing machine. The resultant ice-cream was packaged in cups 100 ml and placed in the deep freezer at (-18°C) for hardening for 24 hours (Rothwell, 1976).

**Color parameters:**
Color values were measured with Minolta Chroma Meter (CM-3600d, Minolta, and Ramsey, NJ) (Ruiz *et al.*, 2005). The measurements were displayed in “L”, “a” and “b” values.

**Sensory Evaluation:**
A sensory evaluation of color acceptability was made in the products. This test was carried out according to Reitmees and Nonnecke (1991).

**Statistical Analysis:**
Statistical analysis was carried out on data of sensory evaluation applying analysis of variance (ANOVA) followed by multiple compa.

**3. Results and Discussion**

**Quantification of Anthocyanins of Red Cabbage:**
Table (1) was showed the photometric quantification on anthocyanins of red cabbage. Extract of acidified ethanol showed the highest quantification of anthocyanins (135.98 mg/100 g as cyanidin-3,5-diglucoside) while citric acid (2%) gave the lowest (120.18 mg/100 g on fresh weight). The obtained results agree with Malien-Aubert *et al.* (2001) reported that, red cabbage contained (141.9 mg/g) as acylated anthocyanins. On the other side, Piccaglia *et al.* (2002) stated that, the anthocyanins content in red cabbage corresponds to 125 mg/100 g fresh plant tissue.

### Table (1) Quantification of Anthocyanins of Red Cabbage

<table>
<thead>
<tr>
<th>Extraction Solvent</th>
<th>Concentration (mg/100 g as cyanidin-3,5-diglucoside)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ethanol+HCl (1%) (acidified ethanol)</td>
<td>135.98</td>
</tr>
<tr>
<td>Citric acid (2%)</td>
<td>120.18</td>
</tr>
</tbody>
</table>

**Factors Effecting on Anthocyanins Stability:-**

1. **Effect of different pH values on the stability of anthocyanins in red cabbage:-**
   Data illustrated in figure (1) showed the results of the effect of different pH values on the natural red pigment in red cabbage. The pH values ranged from 3 to 9. This was confirmed using the spectrophotometer, which showed different absorbance values at different pH value, data in fig. (1) Indicated that, acidified ethanol extract was more stable at pH (4 – 6). While, citric acid (2%) extract less stable. Citric acid (2%) extract was stable at pH 4-5. McDoogall *et al.* (2007) decided that, red cabbage anthocyanins are stable over a broader pH range while, Sapers *et al.* (1981) reported that, pH had little effect on color stability of red cabbage.

![Fig.1](image-url) - Effect of different pH values on the stability of anthocyanins in red cabbage

2. **Effect of different temperature and time of the stability of anthocyanins in red cabbage Extracts:**
   Several physical and chemical factors have a negative effect on the stability of anthocyanins, one of the most important being heat. Measurements of changing in absorbance at wavelength 520 nm of anthocyanins extracts from red cabbage heated within different temperature (50, 75 and 90°C) for different times (10, 20 and 30 min.) are shown in figs (2). Data indicated that, red cabbage anthocyanins were stable at 50 and 75°C but it recorded high degradation after 75°C. Acidified ethanol extract was more stable than citric acid (2%) extract. Henry (1996) decided that, anthocyanins from red cabbage contain a significant quantity of mono and di-acylated anthocyanins and are particularly stable.
3- Effect of different pH values (3-9) and different temperatures (50, 75 and 90˚C) on anthocyanins from red cabbage:

The stability of red cabbage color at three different pH values (3-9) and different temperatures (50, 75 and 90˚C) were illustrated in fig. (3). The results of the stability were reported as percentage degree of the degradation over a period of 30 min. In the two red cabbage extracts at different temperature, the lowest percentage degree of degradation was recorded at pH 3, but it was least stable at pH 9. The acidified ethanol extract recorded the best thermal stability of red cabbage extract. From fig. (3), it could be reported that, the percentage degree of anthocyanin degradation increased by increasing of temperatures and pH – values. These results were agreement with Walkowiak – Tomezak & Czapstei (2007) and Ginat & El-Samahy (2008).

Application in Ice-cream:

Table (2) shows that, the color parameters by Hunter lab for the ice-cream colored by anthocyanins which was extracted from red cabbage. Results represented the values of color parameters as “L”, “a” and “b” values. The “L” values of the ice-cream with acidified ethanol extract by two levels showed high “L” values than ice-cream with citric acid (2%) extract by two levels. However, the other parameters gave the best data with acidified ethanol extract than which by citric acid (2%).

Sensory Evaluation of Ice-cream:

Organoleptic evaluation of food products is of primary importance, since it reflects the consumer preference for respective food products. Accordingly, it is necessary to run a test for the sensory aspects of the food before marketing to avoid any possible hazards that could be happened in the market (Ginat and El-Samahy, 2008).

Results in Table (3) represent the scores of panel test given for ice-cream colored by acidified ethanol extract (5 and 10 ml) and citric acid (2%) extract (5 and 10 ml) from red cabbage. These results indicate...
that, the use of acidified ethanol extract is the best coloring agent. These results were agreed with these results obtained from the color parameters measured by Hunter lab.

Table (2): Color parameters of Ice-cream by using anthocyanins extracts from red cabbage:

<table>
<thead>
<tr>
<th>Color Addition</th>
<th>L</th>
<th>a</th>
<th>b</th>
</tr>
</thead>
<tbody>
<tr>
<td>5 ml acidified ethanol extract</td>
<td>58.63</td>
<td>21.72</td>
<td>-1.09</td>
</tr>
<tr>
<td>10 ml acidified ethanol extract</td>
<td>61.13</td>
<td>26.05</td>
<td>-2.98</td>
</tr>
<tr>
<td>5 ml citric acid (2%) extract</td>
<td>52.74</td>
<td>18.57</td>
<td>-0.98</td>
</tr>
<tr>
<td>10 ml citric acid (2%) extract</td>
<td>55.69</td>
<td>22.43</td>
<td>-1.01</td>
</tr>
<tr>
<td>Artificial Color (as counted)</td>
<td>22.87</td>
<td>63.56</td>
<td>17.93</td>
</tr>
</tbody>
</table>

Table (3): Sensory Evaluation of Ice-cream Prepared by using red cabbage Extracts:

<table>
<thead>
<tr>
<th>Source of color</th>
<th>Color of Ice-Cream</th>
</tr>
</thead>
<tbody>
<tr>
<td>Artificial color (control)</td>
<td>5.00 **</td>
</tr>
<tr>
<td>5 ml acidified ethanol extract</td>
<td>7.70 **</td>
</tr>
<tr>
<td>10 ml acidified ethanol extract</td>
<td>8.50 a</td>
</tr>
<tr>
<td>5 ml citric acid (2%) extract</td>
<td>5.80 b</td>
</tr>
<tr>
<td>10 ml citric acid (2%) extract</td>
<td>6.95 b</td>
</tr>
<tr>
<td>LSD</td>
<td>0.9383</td>
</tr>
</tbody>
</table>

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Reference: