A Comparative Study on Antioxidative Properties of Carameled Reducing Sugars; Inhibitory Effect on Lipid Oxidative and Sensory Improvement of Glucose Carameled Products in Shrimp Flesh

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ABSTRACT

The aim of the present study was to compare three reducing sugars (D-glucose, D-arabinose, D-mannose) with respect to the antioxidative activity characteristics of their Caramelisation Products (CPs) prepared by monitoring UV-absorbance, browning intensity, ferric ion reducing capacity, Fe^{2+} ion chelating activity as well as DPPH radical scavenging activity. Moreover, the effect of CPs from D-glucose on prevention of lipid oxidation and improvement of sensory characteristics were examined in Indian shrimp flesh (Penaeus indicus) during its frozen storage for a duration of 6 months. The reducing sugars were heated at 100°C at pH 10 for 100 minutes to generate CPs. Results revealed greater antioxidant activity of the hexoses, D-glucose and D-mannose, than that of the pentose sugar, D-arabinose, as evidenced by the higher UV-absorbance, browning development, reducing power, chelating as well as DPPH radical scavenging activity. Results of the chemical analysis including TBARs and FFA proved that the addition of CPs from glucose (G-CPs) could retard the formation of compounds produced over lipid oxidation in G-CPs-added shrimps (P< 0.05). More desirability in sensory attributes of the shrimps treated with G-CPs was expressed than in the untreated samples during the 6 months of storage. These results reflect that G-CPs can be utilized as additives in seafood with appropriate antioxidative and techno-functional properties.

Keywords: Antioxidant activity, Caramelisation products, Lipid oxidation, Shrimp, Sugar.

INTRODUCTION

Shrimp is considered as one of the seafoods of high nutritional value and acceptability around the world. Shrimp as well as other seafood products are susceptible to lipid oxidation because of containing high levels of polyunsaturated fatty acids (PUFA) (Harris and Tall, 1994). Shrimp meat is biochemically alive after the shrimp’s death with organic decomposition of the meat probable to occur for various reasons (Pedraja, 1970). Immediately after the catch, freezing can improve the shrimp flesh quality (Karel et al., 1975). Frozen shrimp is a product of high commercial value and there is an increasing trend towards its competitive price and extended shelf life. However, some such important changes as color fading and lipid oxidation can affect shrimp quality during long periods of storage (Tsironi et al., 2009). Various synthetic and natural antioxidants are utilized to prevent lipid oxidation in foods subjected to long term storage. In recent years, there has been increasing concerns regarding the appropriateness of synthetic additives (Varelzis et al., 1997). In fact, synthetic antioxidants including BHT and BHA may cause some such serious

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problems as liver damage and carcinogenicity in laboratory animals (Wang et al., 2000). Therefore, there has been an interest coming forth in the use of natural antioxidants.

Caramelisation, one of the most important heat-favored reactions leading to non-enzymatic browning during food processing and storage (Manzocco et al., 2001), represents an interesting research area for the implications in food stability and technology, as well as in nutrition and health. This reaction occurs when sugars, normally monosaccharides, are being heated to high temperatures without amino groups (Davies and Labuza, 2005) and also simultaneously during the development of brown color caused by the Maillard reaction (Buera et al., 1987). Caramelisation process produces the desirable flavors and colors which are characteristic of many such food products as dark beer, coffee, confectionery and peanuts. Moreover, caramelized sugar delivers the nutritional benefits of the original with a considerable proportion of less sugar. As demonstrated by the recent researches, caramelisation occurs to a larger extent under alkaline condition (Benjakul et al., 2005; Phongkanpal et al., 2006). Preventive effect of acetone extracts from glucose caramelisation against oxidative damage in soybean oil has been proved (Rhee and Kim, 1975). More recently, investigators observed that caramelisation products play a key role in extending the shelf-life of fatty foods by deterring undesirable lipid oxidation reactions (Benjakul et al., 2005). Moreover, according to the results of some recent epidemiological studies, they could exert a role in the prevention and/or protection against such degenerative and chronic diseases as cancer and cardiovascular disease (Aeschbacher, 1990).

Parameters controlling the progression of caramelisation reactions include sugar type, heating time, temperature, pH, buffer type as well as concentration. Studying the effect of sugar type in caramelisation products will bring new insights into industrial applications and focus, for example, on the utilization of sugars rendering more functionality in food industry. The choice of sugar to be incorporated into a food formulation as an additive or preservative can be affected by the sugar type and the criteria as relates economical or technological applications. In the case of economical application, there is more attempt to use cheaper sugars with appropriate biological (antioxidant activity) or techno-functional (sweetening, color development, flavor promotion) properties in food processing and storage (Okazaki and Makino, 2004; Kwak and Lim, 2004). This would prompt industrialists to benefit from scientific studies on sugars’ effects, by producing particular food products.

In recent researches, several such reducing sugars as hexoses and pentoses were compared in terms of their antioxidant potentials as Carameled Products (CPs) and/or Maillard Reaction Products (MRPs) such as Benjakul et al. (2005) (D-fructose, D-glucose, D-ribose D-xylose); Laroque et al. (2008) (ribose, xylose, arabinose, glucose and fructose), Phongkanpal et al. (2006) (D-glucose and D-fructose) while little information is available on a comparison of CPs antioxidant activity prepared from aldohexose sugars like D-mannose and aldopentose sugars such as D-arabinose.

With the above scenario, different CPs derived from D-glucose, D-arabinose and D-mannose prepared under specific conditions were examined for their antioxidative activity. Beyond the presentation of experimental results on the mode of action of CPs, this paper also provides a survey on lipid oxidation of Indian shrimp (Penaeus indicus) flesh treated by D-glucose CPs during its frozen storage.

**MATERIALS AND METHODS**

Thiobarbituric acid reagent, D-glucose, D-arabinose, D-mannose, potassium ferricyanide, ferric chloride, trichloroacetic acid, silicon oil, phenolphthalein, as well as...
sodium carbonate buffer were purchased from Merk (Dermastadt, Germany). 1, 1 diphenyl-2picrylhydrazyl (DPPH), sodium phosphate buffer, sodium sulphate, ethanol, acetic acid, and chloroform were procured from Sigma (St. Louis, USA). All chemicals were of analytical grade.

Sample Preparation

Throughout the study, aldohexos sugars, D-glucose and D-mannose, an aldopentose sugar, D-arabinose were selected as sugar reactants due to differences in the reaction rates associated with their chemical structure. Carameled products (CPs) were prepared by dissolving the sugars in 0.05M sodium carbonate buffer, pH 10, to give a final concentration of 0.05M. Then each sugar solution was heated in a silicon oil bath at 100°C for 100 minutes in screw-capped test tubes. As many previous researchers have proved that carameled products (CPs) and Maillard reaction products (MRPs) from reducing sugars rendered the highest rate of antioxidative activity at alkaline pH during heating and at 100°C for 100 minutes. The absorbance ratio (A_{270}/A_{420} and A_{285}/A_{420}) was also calculated to monitor the transformation of UV-absorbance compounds into brown polymers.

Physical and Chemical Analysis

Measurement of UV-absorbance

The appearance of intermediate products was monitored by the absorbance of diluted CP solutions at 270 and 285 nm (Benjakul et al., 2005). Appropriate dilution (50-fold dilution) was made using distilled water and the absorbance measured at 270 and 285 nm through spectrophotometry (Shimadzu UV 1601, Tokyo, Japan) for determining UV-absorbance.

Formation of Browning Polymers

The browning intensities of CP samples of 10-fold dilution were assessed, utilizing a spectrophotometer (Shimadzu UV 1601, Tokoyo, Japan) by reading the absorbance at 420 nm, against distilled water, as an index of the brown polymers formed at alkaline pH during heating and at 100°C for 100 minutes. The absorbance ratio (A_{270}/A_{420} and A_{285}/A_{420}) was also calculated to monitor the transformation of UV-absorbance compounds into brown polymers.

Test for Ferric Ion Reducing Capacity (Fe^{3+} to Fe^{2+})

The ferric ion reducing capacity was determined according to the method of Oyaizu (1986) with some modifications. A volume of 0.5 ml of potassium ferricyanide was mixed with 0.5 ml of 0.2 M sodium phosphate buffer, pH 6.6 and 0.5 ml of each sample diluted 20-fold. The contents were incubated at 50°C for 20 minutes and 0.5 ml of 10% (w/v) trichloroacetic acid then added to the reactive mixture. Thereafter the mixture was mixed with 2 ml of distilled water and 400 µl of 0.1% (w/v) ferric chloride solution. The blank was prepared in the same manner except that 1% potassium ferricyanide being replaced by distilled water. The absorbance was recorded at 700 nm using a Shimadzu UV-1601 spectrophotometer (Tokyo, Japan). Absorbance increases with an increase in ferric ion reducing capacity.

Fe^{2+} Ion Chelating Activity

The potential of CPs to chelate the prooxidative transitional metal ion Fe^{2+} was investigated as described by Tang et al. (2002) with some modifications. Eight hundred µl of distilled water plus 0.1 ml volumes of 2 mM FeCl_{3} were added to 200 µl of CP samples. The reaction mixture was later added with 0.2 ml of 5 mM ferrozine
and allowed to stand at room temperature for 20 minutes. Changes in color were monitored at 562 nm with a spectrophotometer (Shimadzu UV 1601, Tokyo, Japan). The Fe\(^{2+}\) chelating activity was calculated as follows:

\[
\text{Chelating activity\%} = \left[1 - \left( \frac{A_{\text{sample}}(562\text{nm})}{A_{\text{blank}}(562\text{nm})} \right) \right] \times 1
\]

1,1-Diphenyl-2-picrylhydrazyl (DPPH) Radical Scavenging Activity

DPPH radical scavenging activity was assessed according to the method of Yen and Hsieh (1995) with some modifications. 400 µl of CPs from different sugars was mixed with 2 ml of freshly prepared 0.12 mM DPPH methanolic (95%) solution. Thereafter a 45-fold dilution was made for the samples using distilled water as a diluent. The resulting solution was well agitated and then incubated at 25°C for 30 minutes, then measured colorimetrically at 517 nm in a Shimadzu UV-1601 spectrophotometer (Tokyo, Japan) against a blank of distilled water. Lower absorbance at 517 nm represents higher DPPH scavenging activity according to the following equation:

\[
\text{Scavenging activity\%} = \left[1 - \left( \frac{A_{\text{sample}}(517\text{nm})}{A_{\text{blank}}(517\text{nm})} \right) \right] \times 1
\]

Preparation of CPs-added Shrimp

Fresh Indian shrimp (Penaeus indicus) samples were supplied from a local market and transported to the chemical laboratory of Fishery Department, Gorgan University, in iced-boxes. The edible portions were removed, rinsed with tap water and finely chopped into pieces of 8 g (on the average). CPs derived from D-glucose by heating at 100°C and at pH 10 for 100 minutes, were added to shrimp slices in a way as to cover the whole surface of the samples. This was used as the antioxidant treatment. Distilled water was added to a portion of shrimp samples to represent the control. The ratio of shrimp flesh to either CPs or distilled water was 1:9 (w/w) to obtain 10% as a whole. Prior to an addition of CPs to the shrimp slices, 6M HCl was applied to reduce the pH of CPs to a neutral one. The mixtures were then packaged in polyvinylidene film and styrofoam trays. The packages were initially blast-frozen at -40°C for 12 hours, then stored at -18°C for 6 months. At each sampling occasion (0, 1, 3, and 6 months past storage), samples of each treatment were taken for subsequent lipid oxidation and as well for sensory analysis.

Inhibition of Lipid Peroxidation

Assessments of oxidation in shrimp samples added with CPs (D-glucose) were achieved through measurements of thiobarbituric acid reactive substances (TBARs) as well as free fatty acids (FFA), according to the procedures of Tarladgis et al. (1969) and Egan et al. (1997), respectively. To determine TBARs, 10 g of shrimp sample was homogenized for each group, prepared for TBARs analysis, with the color development measured at 538 nm using a lightwave spectrophotometer (S2000 UV/VIS diode array spectrophotometer). The TBARs was expressed as mg malonaldehyde kg\(^{-1}\) of shrimp sample and measured according the following equation:

\[
\text{TBARs(mgmalonaldehydekg}^{-1}) = 7.8 \times D
\]

Where, \(D\) is the absorbance of the solution against the blank sample prepared by adding 5ml of distilled water plus 5 ml of TBA solution.

Free fatty acid (FFA) content was reported as described by Egan et al. (1997). Results were expressed as % oleic acid (i.e. the cm\(^3\) 0.1N NaOH used in the titration corresponding to % oleic acid).
Sensory Analysis

Sensory assessments included the evaluation of three parameters (flavor, color and odor) conducted by five trained panelists. According to the method of Karmer and Twigg (1996), 5 categories of acceptability were ranked: excellent quality (5), good (4), acceptable (3), fair (2) and finally reject (1). Samples of the treatments were taken at regular intervals, steam-cooked and served warmly to the panelists. The scores were then analyzed.

Statistical Analysis

Experiments of lipid prooxidation in shrimp samples as assessed by TBA and FFA were conducted as a 2×4×3 factorial design (two treatment levels, four storage time level, and three replicates). ANOVA was employed to search for significant differences among levels of each factor and interactions between treated factors. Data from reducing sugar experiments, TBA and FFA were subjected to ANOVA followed by least significant difference test (LSD) using statistical analysis system SAS (SAS institute) where $\alpha = 0.05$. Analysis of sensory attribute scores was performed through Kruskal-Wallis $H$ test as a non-parametric test using statistical package of SPSS, Ver. 11.5 (SPSS Inc., Chicago, IL, USA). Diagrams were made employing the Microsoft Excel 2003 (Microsoft Corporation, USA). All the data, except for sensory measurements (5 replicates), were expressed as means±standard errors of triplicate determinations.

RESULTS AND DISCUSSION

Changes in UV-absorbance

UV-absorbance was employed to monitor the intermediate degradation products of non-enzymatic browning reactions (Ajandouz et al., 2001). The changes in UV-absorbance of different CPs derived from aqueous solutions of D-glucose, D-arabinose and D-mannose are indicated in Figure 1 (a, b). In the present study, glucose and mannose were found to have higher UV-absorbance at 270, 285 nm than aldopentose, arabinose. The UV-absorbance value of glucose CPs was in agreement with the results obtained by Benjakul et al. (2005) who reported that glucose exhibited higher accumulation of intermediate products than pentose sugars, ribose and xylose. The results of UV-absorbance measurement at
270 and 285 nm were in accordance with those reported by Phongkanpal et al. (2006) who determined values of 35 and 28 for UV-absorbance of glucose-CPs at 270 and 285 nm, respectively.

**Changes in Browning Intensity**

The final stage of the browning reaction was monitored by the increase in absorbance at 420 nm (Ajandouz et al., 2001). The UV absorbing and colorless compounds formed at the intermediate stages contributed to such brown pigment formations as melanoidins in both Maillard and caramelization reactions (Benjakul et al., 2005). Studies on melanoidin formation have been summarized in different review articles (Friedman, 1996; Rizzi, 1997). Figure 1-c depicts the changes in browning intensity of CPs derived from aqueous solutions of D-glucose, D-arabinose and D-mannose. In this research, results indicated that the browning development of D-glucose and D-mannose CPs occurred in the same manner following heating at 100°C for 100 minutes (P> 0.05). These sugars were more likely to undergo browning via caramelisation than D-arabinose which showed a significant (P< 0.05) lower browning development. The differences in browning, found among sugars might be associated with their different relative structural stabilities including mutarotation, opening of the hemiacetal ring and enolization of the sugar (Buera et al., 1987). Furthermore, the development of browning might be influenced by the type of sugar, pH and duration of heating. Buera et al. (1987) reported that rates of browning development of reducing sugars via caramelisation processes were in the following descending order: fructose> xylose> lactose> maltose> glucose.

However, as discovered by Laroque et al. (2008), the browning development was favored with MRPs from pentoses (ribose, xylose, and arabinose), compared to that of hexoses MRPs (glucose and fructose). In the browning development, the order of reactivity according to which aldoses are more reactive than ketoses and pentoses than hexoses was well stated and demonstrated in many studies (Jing and Kitts, 2002, 2004; Kwak and Lim, 2004). Under the present experimental conditions, distinction could not be significantly made between the relative browning degree of mannose and glucose (P> 0.05).

The absorbance ratio of sugar solutions heated for 100 min at 100°C is shown in Table 1. The highest ratios of \( A_{270} / A_{420} \) and \( A_{285} / A_{420} \) were observed in CPs derived from D-glucose and D-mannose. The increased absorbance ratios of these hexose sugars suggested that their intermediate products were generated to a larger extent with lower transformation to brown polymers than D-arabinose when heated at 100°C for 100 minutes under alkaline conditions. From the results here and previous ones by Benjakul et al. (2005), it could be concluded that the formation of brown polymers from different intermediates varied with the type of sugar. Lu et al. (2005) declared that diglycine–glucose reaction mixtures had a higher degree of browning, followed by glycene–glucose, and triglycine–glucose reaction mixtures. Thermolysis causes dehydration of sugar molecules with the introduction of either double bonds or of anhydro rings. Introduction of double bonds leads to unsaturated rings and conjugated double bonds, absorbing light and producing color. Unsaturated rings will condense to polymers, leading to the development of color (BeWiller and Whistler, 1996).

**Table 1. Changes in the absorbance ratio of CPs from different sugars during heating at 100°C for 120 (Min).**

<table>
<thead>
<tr>
<th>Sugar</th>
<th>( A_{270}/A_{420} )</th>
<th>( A_{285}/A_{420} )</th>
</tr>
</thead>
<tbody>
<tr>
<td>D-glucose</td>
<td>10.25</td>
<td>8.4</td>
</tr>
<tr>
<td>D-mannose</td>
<td>10.5</td>
<td>8.81</td>
</tr>
<tr>
<td>D-arabinose</td>
<td>6.09</td>
<td>5.63</td>
</tr>
</tbody>
</table>

**Changes in Reducing Capacity**

Previous studies have reported that the reducing capacity of a compound may serve as a significant indicator of its potential
Antioxidant Properties of Caramelized Sugars

Thus, it appears necessary to determine the reducing power of CPs derived from reducing sugars to evaluate their antioxidant potentials. The changes in the reducing capacity of CPs derived from aqueous solutions of D-glucose, D-arabinose and D-mannose are shown in Figure 2-a. Regarding the presented results, CPs from D-glucose and D-mannose indicate a similar reducing capacity (P > 0.05) whereas D-arabinose CPs represent the lowest capacity under alkaline conditions. Benjakul et al. (2005) reported that CPs from glucose showed higher reducing power than those from ribose and xylose under alkaline conditions. Thus, it can be concluded that CPs from hexose sugars rendered the greater reducing power, as compared with CPs from pentose. This is in agreement with Yoshimura et al. (1997) who reported that MRPs from a glucose–glycine mixture had a higher reducing power, especially when the heating time increased. The result of ferric ion reducing capacity of CP glucose, in this research, was in accordance with the result of Benjakul et al. (2005) whilst it showed a double increase in comparison with reducing power rate proposed by Phongkanpal et al. (2006). During heating of sugar solutions, especially under alkaline conditions, reducing compounds and Maillard reaction products might be formed and these could exhibit antioxidative activity (Benjakul et al., 2005; Yen and Hsieh, 1995). The reducing power of CPs might be due to hydrogen-donating potential (Shimada et al., 1992).

**Changes in Fe\textsuperscript{2+} Ion Chelating Activity**

The Fe\textsuperscript{2+} ion is the most powerful prooxidant among various species of metal ions (Yomauchi et al., 1988). Ferrous iron can interact with hydrogen peroxide in a Fenton reaction to produce the hydroxyl free radical, which can initiate lipid oxidation (Hultin, 1994). Figure 2-b represents the changes in the Fe\textsuperscript{2+} ion chelating activity of CPs derived from aqueous solutions of D-glucose, D-arabinose and D-mannose. CPs from hexoses, glucose and mannose, significantly (P < 0.05) revealed the highest chelating activity of Fe\textsuperscript{2+} than CPs from arabinose. In the present research, chelating activity of the hexoses was in accordance with the results of CPs from fructose suggested by Benjakul et al. (2005) when concentrations of the sugar solution increased (> 15 µl). A lot of information exists on the metal ion binding affinity of MRPs and the effect of melanoidins, as a possible mechanism for their antioxidant activity (Jing and Kitts, 2004; Morales et al., 2006).
Changes in the DPPH Radical Scavenging Activity

DPPH is a chromogen-radical-containing compound that can directly react with antioxidants. Since the stable radical DPPH can accommodate many samples in a short period and is sensitive enough to detect active ingredients, DPPH assay has been widely used to evaluate the antiradical activity of pure antioxidant compounds, plant and fruit extracts, and food materials (Piao et al., 2004; Yu et al., 2002). The changes in the DPPH radical scavenging activity of CPs derived from aqueous solutions of D-glucose, D-arabinose and D-mannose are illustrated in Figure 2-c. The DPPH radical scavenging activity of CPs from D-glucose and D-mannose were 0.7 times higher than that of D-arabinose. Benjakul et al. (2005) declared that CPs from glucose exerted greater DPPH radical scavenging activity as compared with CPs from ribose and xylose. In this research, the rate of DPPH radical scavenging activity for glucose CPs was two times that of Phongkanpal et al. (2006) who determined a rate of approximately 15% for CPs derived from glucose under alkaline pH and heating at 100°C for 100 minutes.

Values of DPPH radical scavenging activity of sugar solutions have been reported by several workers but they are difficult to be compared because of the differences in sample sources, sample preparations, methodology details, standards used, among others. The rate of the reaction depends on the rate at which the sugar ring opens to the reducible, open-chained form (Davies et al., 1998). For example, Sumaya-Martinez et al. (2005) reported that the ribose caramelization contributed to the antiradical activity and browning reactions at 95 and 115°C. Kim and Lee (2009) reported that the antiradical activity of the MRP s derived from ribose was 11-fold that of MRPs derived from glucose due to the acyclic form of ribose.

There is a lot of information about Maillard reaction whilst available data on caramelisation reaction is scarce (Yen and Hsieh, 1995; Morales et al., 2005; Benjakul et al., 2005). In the latest citation, antioxidative activity of CPs in accordance with MRPs in such high-temperature cooked products, as grilled or roasted products has been proposed (Benjakul et al., 2005). It was suggested that CPs able to reduce the DPPH radical to the yellow-colored diphenylpicrylhydrazine. The reduction of alcoholic DPPH solution in the presence of a hydrogen-donating antioxidant is due to the formation of the non-radical form, DPPH-H. When the DPPH radical is scavenged by antioxidants through the donation of hydrogen to form a stable DPPH-H molecule, the color is changed from purple to yellow (Shon et al., 2003). Therefore it
was proposed that either intermediates or the final brown polymer could function as hydrogen donors.

**Changes in Lipid Oxidation**

Lipid hydrolysis occurred along the frozen storage of Indian shrimp (Figure 3a). In comparison with the initial material, the release of free fatty acid (FFA) in control and in samples with CPs involving glucose (G-CP) increased with the storage time and reached the highest mean value after 6 months each sample. This may be due to the effect of lipid hydrolyzing enzymes (mainly lipase and phospholipase) in decomposing the fats (phospholipids and triglycerides) (Serdaroglu and Felekoglu, 2005). In the present study, the release of FFA in G-CP treated samples was significantly ($P \leq 0.05$) lower than that in the control on months 3 and 6 (1.98 for G-CP treated sample versus 2.86 for the control on month 6) (Figure 3-a). Accordingly, inhibitory effects of caramelisation product on lipid hydrolysis could be concluded. The authors could not find any study evaluating the effect of caramelisation products on the lipid hydrolysis of shrimp or fish meat. Since the release of FFA content increased with time, as found in this study, it is reported that there exists a relationship between FFA release and lipid oxidation (Serdaroglu and Felekoglu, 2005) as well as loss of freshness (Özogul et al., 2005). In this study, G-CP conspicuously reduced the rate of lipid damage in frozen shrimp meat.

TBARS, an index of malonaldehyde concentration, is a widely used indicator for the assessment of degree of secondary lipid oxidation (Weber et al., 2008). Malonaldehyde is one of the main end-products of lipid oxidation. The changes in the formation of TBARS in both samples are shown in Figure 3-b. A significant increase ($P \leq 0.05$) in the TBARS value was observed in both treatments during the storage time, in particular, the highest value obtained on month 6 for the control (1.16). In the present study, the use of G-CP reduced the rate of TBARS formation in shrimp meat as compared with the control ($P \leq 0.05$). The results obtained from TBARS assessment are in agreement with the previous work by Benjakul et al. (2005) who reported that adding caramelisation product of fructose reduced the formation of TBARS in comminuted saury (*Cololabis saira*) during iced storage. However, it is concluded that G-CP can retard lipid oxidation in shrimp meat during frozen storage due to its good chelating activity (chelating the ferrous ions) (Figure 2-b), DPPH radical scavenging (scavenging free radicals) (Figure 2-c) as well as high reducing power (being a good hydrogen or electron donor) (Figure 2-a).

**Changes in Sensory Attributes**

Seafoos are particularly sensitive to oxidative rancidity (Khayat and Schwal, 1983) the development of which may produce some undesirable changes in flavor (Karähadian and Lindsay, 1989), color (Haard, 1992) and external qualities (Undeland and Lingnert, 1999). Figure 4 shows the results of the sensory analysis of shrimps during frozen storage. The effect of storage time on sensory attributes of the shrimps varied between the control and G-CP treatment. The sensory quality of both samples significantly ($P< 0.05$) decreased with increased storage time, except for flavor score of G-CP treated shrimp which was not significant ($P> 0.05$). Sensory analysis indicated that the use of G-CP reduced the rate of meat deterioration in comparison with control. Although some information is available on antioxidative properties of caramelisation products, there is no study evaluating the effect of caramelisation products on sensory attributes of shrimp or fish meat. In the present study, the panelists gave the higher scores to the shrimp flesh treated by G-CP at all sampling occasions and also characterized a desirable light brownish color with a new appealing flavor and smell.
Figure 4. A comparison of flavor (a), odor (b) and color (c) scores in shrimp treated by CPs of D-glucose and the control during frozen storage. Bars indicate the standard errors of the means (n= 5).

CONCLUSIONS

Antioxidant properties, desirable color formation and appealing flavor of the CPs-treated frozen stored shrimp reveals that glucose caramelisation products offer a strong radical scavenging role and an oxidative inhibitory activity as well as more acceptability for the consumers. These findings provide an interesting and important scenario to a better understanding of the potential exploitation of caramelisation products for technological and functional purposes by seafood product manufacturers. For further evaluation, comparisons of G-CP at various concentrations and different preparation methods of carameled products are proposed with the employment of microbiological analysis and assessment of toxicological safety, not investigated in the present trail.

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