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PROPERTIES OF CAROTENOID PIGMENTS EXTRACTED FROM YELLOW CARROT AND PUMPKIN AS A FOOD COLORANTS

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ABSTRACT

Carotenoids from yellow carrot (*Daucus carota* L.) and pumpkin (*Cucurbita* spp.) were extracted, identified by TLC and HPLC. The adsorption of concentrated yellow pigment to solid matrixes (lactose, dextrin, flour, starch and skim milk) was studied. The effects of pH and temperature on the pigments were evaluated and the thermal stability of pigments was determined. The extracted yellow pigments were used for colouring hard candy, noodle and glazing jelly as healthy food products.

The concentration of carotenoids in yellow carrot and pumpkin was 148.58 and 126.49 mg/100g, respectively. The identification results by TLC which confirmed by HPLC showed five fractions of carrot carotenoids. They were lutein (5.49%), cryptoxanthin (8.24%), α-carotene (39.56%), β-carotene (44.51%) and canthanthin (2.20%). On the other hand four peaks were appeared in case of pumpkin carotenoids namely isozeaxanthin (4.42%), zeaxanthin (5.76%), β-carotene (57.58) and bixin (32.24%).

The obtained results proved that, the best carrier for adsorption of carrot carotenoids was starch followed by lactose, while lactose was the most effective adsorpant for carotenoids extracted from pumpkin followed by flour. The tested carotenoids exhibit good stability to heat and the highest retention rate of carotenoids for carrot and pumpkin was observed at pH 7.0 while the lowest retention rates were at pH 2.0 and 9.0.

Addition of 0.39% carotenoid pigments extracted from carrot as a natural colorants to noodle prepared from wheat 72% extraction and 0.13% carotenoids pigment to glazing jelly and hard candy were the best treatments and improved the sensory qualities as indicated by panelists. Results suggest that carotenoids could be successfully utilized as natural coloring agents for the purpose of making some healthy food products.

keywords: Carotenoid, carrot, pumpkin, extraction, identification, adsorption, food products, sensory evaluation.

INTRODUCTION

Carotenoids represent the most widespread group of naturally occurring pigments in nature (Gareth *et al.*, 1998). Increasing demand for natural β-carotene has resulted in grouping interest in extracting β-carotene from vegetable products (Vega *et al.*, 1996). Besides their pro-vitamin A activity, α- and β-carotene are considered to act as antioxidants (Graham, 1988) in the diet and they may have important effects in cancer prevention and reduction of the risk of heart disease. Carotenes are used commercially as natural food colorants and they have been added to many products (Britton, 1992; Rogers, 1993; Baumy, 1997 and Rizk, 1997).

The effect of processing and storage on β-carotene retention is of much concern to the technologist as is the raw product s nutritional value (Rogers, 1993 and Helena *et al.*, 1998).

The objective of this research was performed to: (a) evaluate the carotenoid levels in yellow carrot and pumpkin grown in Egypt ; (b) determine the effect of pH, temperature and thermal stability on the investigated pigment levels ; (c) study the pigment effectiveness as influenced by absorptive supports (d) investigate the feasibility of utilizing the carotenoids as natural coloring agent in some food products.

MATERIALS AND METHODS

Materials:

Samples of mature yellow carrot (*Daucus carota* L.) and pumpkin (*Cucurbita* spp.) were purchased from local market in Cairo, Egypt, during the winter months (December – March). Chemicals and solvents were reagent grade.

Methods:

Extraction and determination of yellow carotenoids:

Yellow carotenoids were extracted from carrot and pumpkin samples using acetone and petroleum ether solvents. To measure total carotenoids, 5ml of petroleum ether extract of the sample was pipetted to a 100ml volumetric flask containing 3ml acetone and diluted to mark with petroleum ether. The color was measured using spectrophotometer Jenway 6105 (UV.VIS.) at 452nm. The total carotenoids content was calculated using the following equation (Ranganna 1979).

Conc. of carotene as read

$$\text{Carotenoid (mg /100g)} = \frac{\text{from standard curve (µg/ml) . Final volume . Dilution}}{\text{ml of the extract used . weight of sample}} \cdot 100$$

Concentration and adsorption of carotenoids in solid supports:

The collected extracts of carotenoids were concentrated by the removal of solvent in a rotary vacuum evaporator at 40°C. The concentrated pigments were adsorbed to solid matrixes (lactose, dextrin, flour, starch, and skim milk) in different ratios (1:1, 1:2, 1:3, and 1:4 carrier/pigment) and the mixtures were dried in oven at 40°C for 24hrs.

Identification of carotenoids:

Carotenoid pigments extracted from yellow carrot and pumpkin were identified by thin-layer chromatography (TLC) according to the method described by Eder (1996). The purified carotenoids of carrot and pumpkin were then analyzed by High Performance Liquid Chromatography (HPLC) according to the method reported by Heinonen (1990).

Stabilization of carotenoids:

Effect of pH:

Changes in carotenoids (carrot and pumpkin) as a function of pH were measured according to the method described by Elbe *et al.* (1974) as follows: 1ml of carotenoids solution was mixed with 4ml of 0.1M McIlvaines buffer of various pH ranging from 2.0-9.0. The buffer solutions were introduced into 10ml vials which were flushed repeatedly with nitrogen gas to limit oxygen. Vials were maintained at 4°C in the dark, and absorbance readings were made initially and after 7 days using spectrophotometer at 452 nm.

Effect of temperature:

The method described by Saguy (1979) with some modifications was used to study the effect of temperature on carotenoids. 1ml of carotenoids solution and 9ml of optimum buffer into unsealed vials were placed in a thermostatically controlled water bath at different temperatures ranging from 50-100°C for 30 min. The samples were further cooled down immediately in an ice water bath and absorbance measurements were read by spectrophotometer at 452 nm.

Thermal stability of pigments:

Holding carotenoids (carrot and pumpkin) solution at 70 to 100°C was extended for 180 min in water bath and removed each 30 min then cooled immediately in an ice bath followed by measuring absorption of the solution at 452 nm.

Technological methods:

1-Hard candy processing:

Hard candy was manufactured in the laboratory using the traditional procedure as described by Staniec (1994). The formula of the control sample is shown in Table (1). Water, sucrose, corn syrup, and citric acid were mixed together and heated to reach 157.5°C with continues stirring and then the mixture was cooled to reach 110°C. Color and flavor were added and then formulation and cooling of the product were done to reach room temperature and then packaging was carried out. For the control treatment, synthetic yellow color (Tartrazin) was added by a ratio 6.3 mg /100g of mixture. Different ratios of the investigated yellow pigments were added by 0.13, 0.26, and 0.39%.

Table (1): Formula used for the control hard candy.

Ingredients	weight (%)
Sucrose	48.48
Corn syrup	25.90
Water	25.12
Flavoring oil	0.21
Citric acid	0.15

2- Manufacturing of noodle:

Noodle samples were prepared from flour (72% extraction) in the laboratory using the method described by Collins and Pangloli (1997) using the past Matic 1000 Simac Machine corporation; Millano, Italy. Each sample was manufactured by using 150g wheat flour and blended with different ratios of investigated yellow natural pigments 0.13, 0.26, and 0.39%. Enough water was added for each mixture and the mixing times was ranged from 4-6 min to get a plastic homogenous mass. The produced dough was kneaded and extruded in a continuous extrusion press equipped with special die shape. Noodle was cut to appropriate lengths, hardened for 15 min in air and drying at 40°C for 24hrs. The product was packed in polyethylene bags for further analysis.

3-Glazing jelly processing:

Glazing jelly was prepared in the laboratory using ingredients given in Table (2) as follows: sucrose and caragenan mixture was boiled first in the water, then calcium chloride (dissolved in a small volume of water) was added to the mixture. Sorbic acid (dissolved in 2ml isopropyl alcohol) and potassium sorbate (dissolved in water) were added to the mixture. Glucose was lately added with continuous stirring. After the complete dissolving of ingredients heating was stoped and color was added. Formulation and cooling of jelly samples were done in the refrigerator for 5hr. For the control treatment, synthetic yellow color (Tartrazin) was used by ratio 6.3 mg/100g of mixture. Different ratios of the investigated yellow pigments were added by 0.13, 0.26, and 0.39%.

Table (2): Formula used for glazing jelly.

Ingredients	weight (%)
Sucrose	35.85
Water	54.35
Glucose	9.06
Caragenan	0.38
Sorbic acid	0.08
Potassium sorbate	0.13
Calcium chloride	0.15

Organoleptic evaluation:

The organoleptic properties of the processed yellow cooked noodle, hard candy and glazying jelly were assessed by taste panelists of the staff-members of the Food Science Department, Faculty of Agriculture, Ain Shams University.

Statistical analysis:

The obtained data were analyzed according to the Statistical Analysis System (SAS, 1996) published by SAS Institute, Inc, U. S. A.

RESULTS AND DISCUSSION

Determination and identification of carotenoids:

The concentration of carotenoids in carrot and pumpkin were 148.58 and 126.49 mg/100g, respectively. A typical High Performance Liquid Chromatography (HPLC) chromatograms given in Figs. (1 and 2) and data in Table (3) showed the carotenoids composition of carrot and pumpkin pigments, which were separated based on their functional groups into five fractions for the first source and four fractions for the latter source by thin layer chromatography (TLC). In carotenoid analysis, TLC is mainly used for preliminary examinations to give an indication of the number and variety of carotenoids present and to help in the selection of a suitable separation and purification procedure for the given mixture (Eder 1996).

Table (3): Identified carotenoid pigments of carrot and pumpkin.

Source of pigment	HPLC			T LC (R_f)	Identified carotenoids
	Peak	Retention time (min)	Area %		
Carrot	1	3.25	5.49	0.34	*Lutein
	2	5.70	8.24	0.73	*Cryptoxanthin
	3	7.02	39.56	0.88	**α- carotene
	4	11.00	44.51	0.83	**β- carotene
	5	11.82	2.20	0.88	*Canthanthin
Pumpkin	1	7.00	4.42	0.64	Isozeaxanthin
	2	7.60	5.76	0.19	Zeaxanthin
	3	11.00	57.58	0.94	β- carotene
	4	12.10	32.24	0.50	Bixin

Carrot pigments:

*TLC first system: adsorbent (silica gel G), solvents (methylene chloride : ethyl acetate 80:20).

**TLC second system: adsorbent (silica gel G : calcium hydroxide 1:6 w/w), solvents (petroleum ether : methylene chloride 95 : 5).

Pumpkin pigments:

TLC adsorbant (silica gel G), solvents (hexan : ether, 30 : 70).

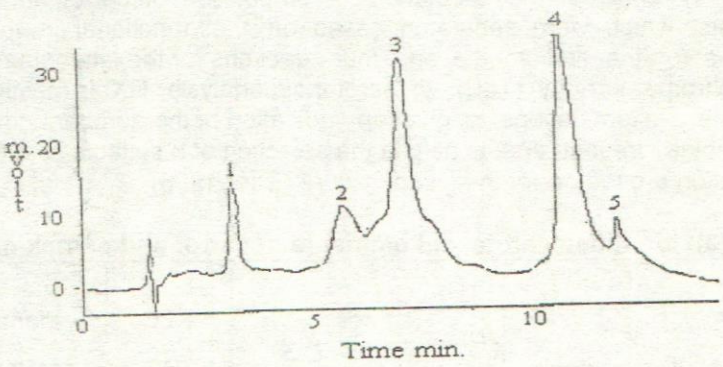

Fig. (1): HPLC chromatogram of carotenoid pigments extracted from yellow carrot.

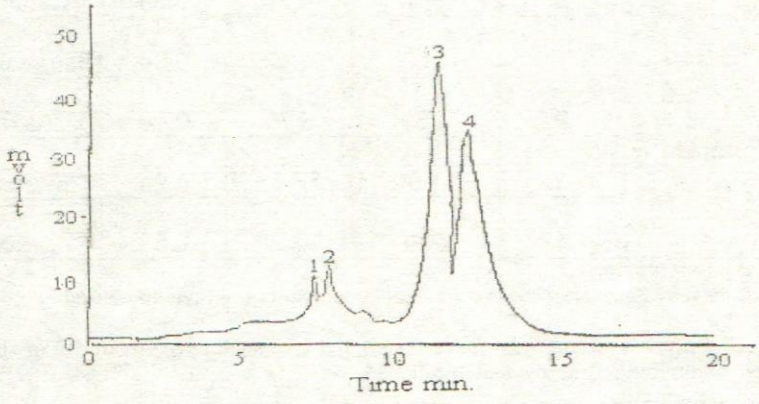

Fig. (2): HPLC chromatogram of carotenoid pigments extracted from pumpkin.

The calculated R_f values for carotenoid fractions of carrot were 0.34, 0.73 and 0.88 in the first system which were identified as lutein, cryptoxanthin and canthanthin, respectively, 0.88 and 0.83 in the second system and were identified as α-carotene and β- carotene. In case of pumpkin carotenoids, four fractions were separated by R_f values 0.64, 0.19, 0.94 and 0.50 were identified as isozeaxanthin, zeaxanthin, β- carotene and bixin, respectively.

These results are confirmed by HPLC into five fractions of carrot carotenoids, they are lutein (5.49%), cryptoxanthin (8.24%), α- carotene (39.56%), β- carotene (44.51%) and canthanthin (2.20%) with a retention time 3.25, 5.70, 7.02, 11.00 and 11.82 min, respectively. On the other hand, four peaks were appeared by using HPLC in case of pumpkin carotenoids namely isozeaxanthin (4.42%), zeaxanthin (5.76%), β- carotene (57.58%) and bixin (32.24%) with a retention time 7.00, 7.60, 11.00 and 12.10 min, respectively. These results are in agreement with those obtained by Watanabe *et al.*, (1988), Heinonen (1990), Wu et al., (1996) and Ana *et al.*, (1998). The aforementioned results proved that the major compounds was β-carotene followed by α- carotene in carrot pigment and β- carotene followed by bixin in pumpkin pigment

Adsorption of carotenoids:

Means of the data obtained for the adsorption of carotenoids extracted from carrot as a natural food colorants at different ratios (1:1, 2:1, 3:1 and 4:1 pigment : carrier w/w) on different carriers (dextrin, flour, lactose, skim milk and starch) are presented in Table (4). Results were statistically analyzed using the Duncan s multiple range tests. In the first ratio (1 : 1), no significant difference was found between dextrin and starch (40.29 mg/100g) which had the highest concentration followed by lactose, flour and skim milk with concentrations 35.10, 27.31 and 21.53 mg/100g, respectively. The best carrier in ratio (2 :1) was lactose 49.50 followed by skim milk, starch, dextrin and flour with concentrations 48.32, 46.06, 26.64 and 19.60 mg/100g, respectively. At ratio (3 :1), skim milk was the best carrier 73.95 followed by starch, dextrin, lactose and flour with concentrations 69.34, 59.88 and 46.65, respectively. At ratio (4 :1), starch has the highest concentration for carrot carotenoids 91.69 followed by lactose and skim milk with concentration 77.21, 67.49 mg/100g but no significant difference between dextrin and flour with concentration 45.23 and 45.22 mg/100g, respectively. Subsequently, the obtained results proved that, the best carrier for carrying pigment was starch followed by lactose. Results may suggest the potency of starch as an efficient dispersing or emulsifying agent that acting as a good carrier for carotenoids. These results are in agreement with those found by Hamed (1985) and Abou El-Maati and Abd-Ellatif (1992).

Table (4): Color concentration (mg/100g carrier) of carotenoids extracted from carrot and suspended on different carriers at different ratios (w /w).

Pigment :Carrier (w/w)	Applied carriers					L.S.D.
	Dextrin	Flour	Lactose	Skim milk	Starch	
1:1	40.29 Ca	27.31 Bc	35.10 Db	21.53 Dd	40.29 Da	1.2709
2:1	26.64 Dd	19.60 Ce	49.50 Ca	48.32 Cb	46.06 Cc	0.903
3:1	65.57 Ac	46.65 Ae	59.88 Bd	73.95 Aa	69.34 Bb	1.8546
4:1	45.23 Bd	45.22 Ad	77.21 Ab	67.49 Bc	91.69 Aa	1.0874
L.S.D.	2.3826	1.7612	1.3305	1.8457	2.0627	

A,B (capital letters) means have the same letter indicated that there is no significant difference between any two different pigment : carriers ratios (P>0.05) within the same column.

a,b (small letters) carriers means have the same letter proved that There is no significant difference between any two different carriers (P>0.05) within the same row.

Table (5) shows the adsorption of pumpkin carotenoids at different ratios (1 :1, 2 :1, 3 :1 and 4 :1 pigment : carrier w/w) on different carriers (dextrin, flour, lactose, skim milk and starch). Results were statistically analyzed using the Duncan's multiple range tests. At first ratio, no significant difference was observed between dextrin, flour and lactose with concentrations 71.92, 71.64 and 72.48, respectively followed by starch 68.71 and skim milk 60.76 mg/100g. It could be also observed that, lactose had the highest concentrations 110.58, 125.31 and 132.73 mg/100g at ratios 2 :1, 3:1 and 4 :1, respectively. This means that lactose was the most effective adsorpant coated carrier material for yellow colorant extracted from pumpkin followed by flour. These results are in agreement with those found by Rizk, (1997).

Thermal stability:

Thermal stability of carotenoids extracted from carrot and pumpkin in solution of pH 7.0 and temperature ranging from 70°C to 100°C were extended for 180 min through they were removed each 30 min and cooled followed by measuring absorption spectra of the solution. Fig. (3) proved that the retention of carotenoids extracted from carrot and pumpkin was decreased significantly with increasing temperatures (70–100°C) and holding time (30–180 min.). At 30 min., the retention rates of caroenoids extracted from carrot for temperature 70, 80, 90, and 100°C were 98.97, 98.62, 96.91 and 95.45 ,respectively, and at 180 min. The retention rates were 95.39, 91.33, 88.93 and 86.36, respectively. The retention rates at 70°C for holding time were greater than those produced by 100°C for holding time (30–180 min.). The retention rates for the carotenoids extracted from pumpkin showed the same trend for the data of retention of carotenoids for carrot. Lower stability of the carotenoids is apparent especially in the region of higher

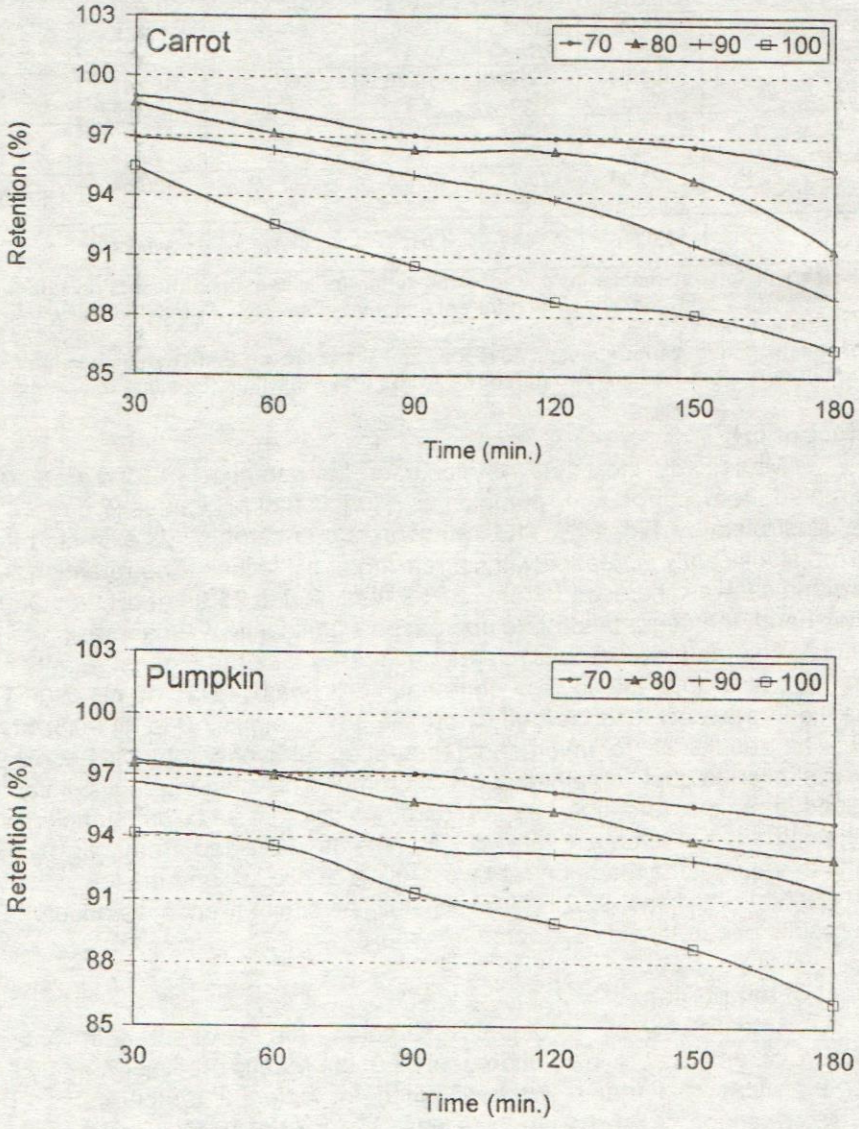

Fig.(3): Thermal stability of carotenoids extracted from carrot and pumpkin (at pH 7.0).

Table (5): Color concentration (mg /100g carrier) of carotenoids extracted from pumpkin and suspended on different carriers at different ratios (w /w).

Pigment :Carrier (w/w)	Applied carriers					L.S.D.
	Dextrin	Flour	Lactose	Skim milk	Starch	
1:1	71.92 Da	71.64 Da	72.48 Da	60.76 Dc	68.71 Db	2.3037
2:1	103.88 Cbc	102.20 Ccd	110.58 Ca	100.04 Cd	105.13 Cb	2.1859
3:1	117.87 Bbc	118.42 Bb	125.31 Ba	113.83 Bd	115.90 Bcd	2.2332
4:1	123.08 Ac	125.14 Ab	132.73 Aa	122.09 Acd	120.54 Ad	1.6506
L.S.D.	2.0215	3.2086	4.0196	2.4824	3.0416	

A,B (capital letters) means have the same letter indicated that there is no significant difference between any two different pigment : carriers ratios (P>0.05) within the same column.

a,b (small letters) carriers means have the same letter proved that There is no significant difference between any two different carriers (P>0.05) within the same row.

Effect of pH:

Means of the data obtained for the retention % of carotenoids extracted from carrot and pumpkin as a function of pH values (7 days/ 4°C) are illustrated by Fig. (4). The retention rate of carotenoids extracted from carrot significantly increased with increasing of pH values. The retention % of carotenoids was increased from 85.59 at pH 2.0 to 95.98 at pH 7.0. On the other hand, the degradation rate decreased significantly with increasing of pH values. The obtained results clearly appeared that, the highest retention % was (95.98) and the lowest degradation % was (4.02) at pH 7.0. The retention rate decreased slightly from 95.98 % at pH 7.0 to 94.90 % at pH 8.0. The results of retention and degradation rates of carotenoids at pH 9.0 were similar to that obtained at pH 3. From the mentioned results, it can be noticed that, the retention rate of carotenoids extracted from pumpkin was higher than the retention rate of carotenoids extracted from carrot. The highest value of the retention rate was 100 % at pH 7.0, and the lowest value of retention rate was 92.21 % at pH 9.0. The similar trend was observed in the results of carotenoids extracted from carrot.

Effect of temperature:

Retention % of carotenoids extracted from carrot and pumpkin as a function of temperature in solution of pH 7.0, the results are illustrated by Fig. (5). Significant differences were obtained for different temperatures. The highest retention % of carotenoids was (97.41 %) at 50°C/ 30 min., and the lowest retention % was (91.87 %) at 100°C/ 30 min. The results of retention % of carotenoids extracted from pumpkin indicated that, the lowest

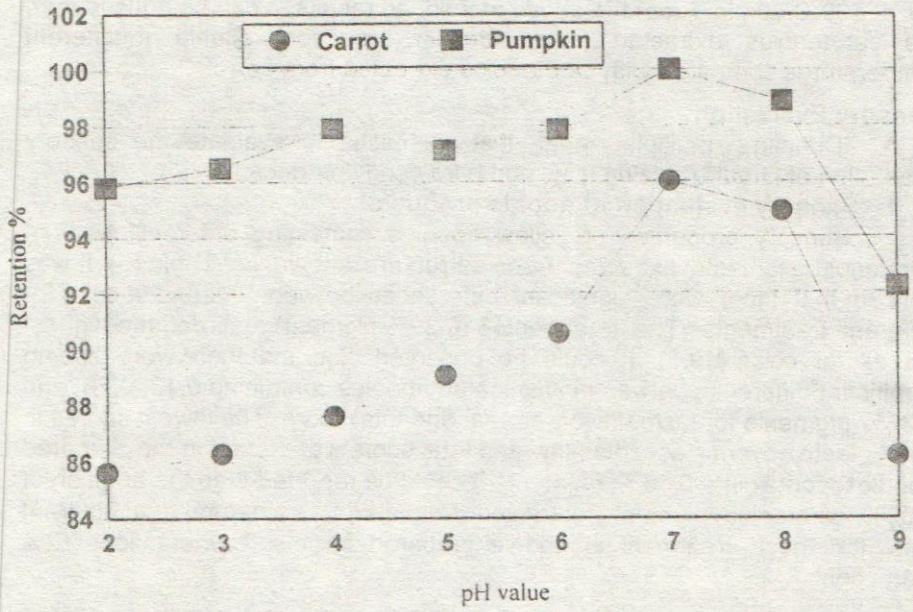

Fig. (4): Retention % of carotenoids extracted from carrot and pumpkin as a function of pH values (7 days / 4°C).

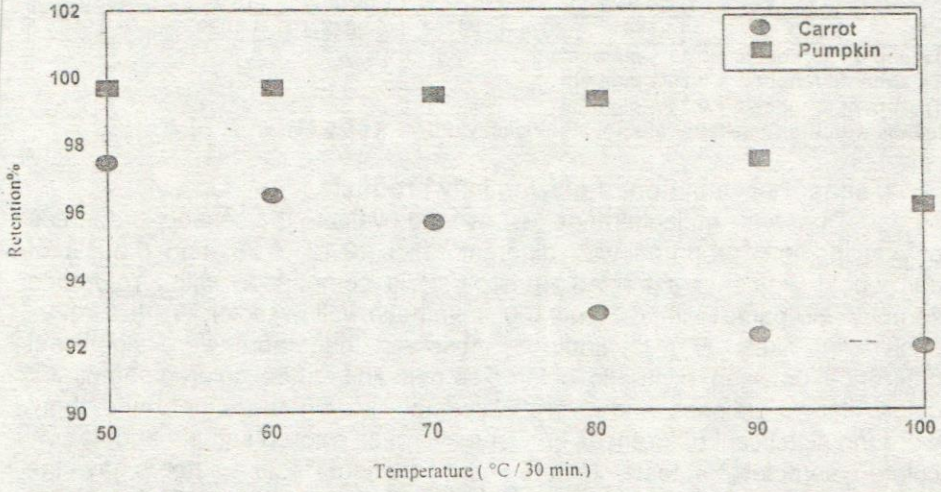

Fig (5): Retention % of carotenoids extracted from carrot and pumpkin as a function of temperature in solutions of pH 7.0.

degradation rates of carotenoids were (0.34 to 0.70%) at (50 - 80°C)/ 30 min. On the other hand, the lowest retention rate of carotenoids was (96.10 %) at 100°C/30min. From the above mentioned results, it can be noticed that, the carotenoids extracted from pumpkin are more stable at different temperatures compared with carotenoids extracted from carrot.

Sensory Evaluation:

Duncan's multiple range test was used to evaluate the sensory evaluation of noodle, glazing jelly, and hard candy products.

1- Sensory evaluation of noodle products:

Sensory properties of yellow noodles containing different ratios of carotenoid pigments extracted from carrot are shown in Table (6). It was noticed that there were significant differences between means of color for different treatments. The treatment 3 (0.39% pigment) recorded the highest scores for color (19.0). It could be observed also that there were also no significant difference between investigated noodles containing 0.13, 0.26, and 0.39% pigments for taste, flavor, aroma, and total score. The lowest scores in color, taste, overall acceptability and total score were found in the prepared noodles containing 0.13% pigment. It could be reported that, the addition of 0.39% carotenoid pigments extracted from carrot as a natural food colorant was the best treatment in noodle prepared from soft wheat flour 72% extraction.

Table (6): Effect of adding carotenoid pigments extracted from yellow carrot as a natural food colorant on selected sensory parameters of tested noodle products.

Treatments	Tested parameters				Overall acceptability	Total score
	Color	Taste	Flavor	Aroma		
1	16.0c	18.6a	18.7a	19.0a	17.1c	89.1a
2	17.8b	18.9a	19.0a	19.0a	18.3b	91.5a
3	19.0a	18.8a	18.5a	18.7a	19.5a	93.2a
L.S.D.	1.1661	1.2094	1.1793	0.9022	1.1099	4.6955

Treatment 1:- noodle + 0.13% pigment.
Treatment 2:- noodle + 0.26% pigment.
Treatment 3:- noodle + 0.39% pigment.
Means with the same letter are not significantly different (P>0.05).

2. sensory evaluation of glazing jelly products:

Duncan's multiple range test used to evaluate the sensory properties of glazing jelly prepared with different ratios (0.13, 0.26, and 0.39%) of carotenoid pigments extracted from carrot in comparison with a reference sample prepared with 6.3 mg/100g synthetic yellow color (Tartrazin) as shown in Table (7). It could be observed that, there were significant difference between synthetic colored sample and natural colored sample for color, clarity, grainess and overall acceptability. On the other hand, there were no significant differences between synthetic colored sample and natural colored samples for taste, texture, and bleeding. Panel members gave the synthetic colored sample superior scores. The treatment containing 0.13% carotenoid pigments extracted from carrot recorded highest scores compared with other treatments.

Table (7): Effect of adding carotenoid pigments extracted from carrot as a natural food colorant on selected sensory parameters of tested glazing jelly products.

Treatments	Tested parameters						Overall accep-tability	Total score
	Color	Taste	Clarity	Texture	Grainess	Bleeding		
1	19.8a	9.9a	19.9a	9.8a	9.8a	10.0a	19.8a	99.0a
2	18.1b	9.8a	16.0c	9.7a	8.5c	10.0a	17.4b	89.5c
3	18.6b	9.8a	16.4c	9.7a	9.0bc	10.0a	18.0b	91.6c
4	18.8b	9.6a	18.8b	9.8a	9.3ab	10.0a	19.0a	95.3b
L.S.D.	0.807	0.3854	0.7234	0.4112	0.7122	0.0	0.8814	2.6051

Treatment 1:- jelly + synthetic yellow color as a control (6.3mg /100g of mixture).
Treatment 2:- jelly + 0.39 % pigment.
Treatment 3:- jelly + 0.26 % pigment.
Treatment 4:- jelly + 0.13 % pigment.
Means with the same letter are not significantly different (P>0.05).

3. Sensory evaluation of hard candy:

Sensory evaluation data of the hard candy with synthetic yellow color (6.3mg/100g) and with different levels of carotenoid pigments extracted from carrot as a natural food colorant (0.13, 0.26, and 0.39%) were statistically analyzed. Data in Table (8) showed that, the mean color, clarity, and overall acceptability scores of hard candy containing 0.13% natural pigment were the highest scores for color, clarity, and overall acceptability. It could be observed that, there were no significant difference between hard candy containing synthetic color and hard candy containing 0.13, 0.26, and 0.39% antural pigment for color, flavor, texture, mouthfeel, and overall acceptability.

Table (8):Effect of adding carotenoid pigments extracted from carrot as a natural food colorant on various sensory parameters of tested hard candy products.

Treatments	Tested parameters					Overall accept-ability	Total score
	Color	Flavor	Texture	Clarity	Mouthfeel		
1	19.0a	14.6a	14.4a	14.7a	14.6a	19.1a	95.5a
2	19.0a	14.0a	14.4a	13.9b	13.9a	18.7a	93.3a
3	18.7a	14.3a	14.2a	14.5ab	14.4a	19.1a	95.1a
4	19.1a	14.3a	14.4a	14.8a	14.3a	19.3a	95.4a
L.S.D.	0.8943	0.6519	0.5815	0.7265	0.7058	0.828	3.2977

Treatment 1:- hard candy + synthetic yellow color as a control (6.3mg /100g).
Treatment 2:- hard candy + 0.39 % pigment.
Treatment 3:- hard candy + 0.26 % pigment.
Treatment 4:- hard candy + 0.13 % pigment.
Means with the same letter are not significantly different (P>0.05).

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خصائص الصبغات الكاروتينية المستخلصة من الجزر الأصفر و القرع العسلى و استخدامها كمواد ملونة طبيعية فى الأغذية

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أجرى البحث لدراسة تركيز الكاروتينات المستخلصة من الجزر الأصفر و القرع العسلى و التعرف على مكوناتها باستخدام طرق الفصل الكروماتوجرافي HPLC, TLC كما تم دراسة أفضل مادة لتحميل تلك الصبغات (اللاكتوز – الدكسترين – الدقيق – النشا – اللبن الفرز) و أفضل نسبة اضافة من صبغة الجزر الأصفر لتحسين اللون فى المكرونة و الحلوى الصلبة و جيلى التجليس، كذلك معرفة تأثير كلا من الـ pH و درجة الحرارة و الثبات الحرارى على الصبغات المختبرة.

و أظهرت النتائج المتحصل عليها أن تركيز الصبغات كانت ١٤٨,٥٨ ، ١٢٦,٤٩ ملليجم/١٠٠ جرام فى الجزر و القرع العسلى على التوالى، و باستخدام طرق التحليل الكروماتوجرافى أمكن التعرف على خمسة مركبات من كاروتينات الجزر و هى ليوتين و كريبتوزانثين و ألفا كاروتين و بيتا كاروتين و كانزانثين بينما وجد أربعة مركبات لكاروتينات القرع العسلى و هى ايزوزيانثين و زيانثين و بيتا كاروتين و بكسين.

هذا و قد ثبت من التجارب أن النشا هو أفضل مادة لتحميل كاروتينات الجزر يليه اللاكتوز بينما فى القرع العسلى كان اللاكتوز هو أفضل مادة يليه الدقيق و لوحظ أن أكبر فقد فى كاروتينات الجزر و القرع العسلى كان على أرقام pH ٢ ، ٩ كذلك لوحظ ثبات الصبغات تحت الدراسة حراريا.

و أوضحت النتائج المتحصل عليها أنه عند اضافه ٠,٣٩% من كاروتينات الجزر الى المكرونه و ٠,١٣% منها الى كلا من جيلى التجليس و الحلوى الصلبه أدى ذلك تحسين الخواص الحسية للأغذية المصنعة، و بذلك يتضح من النتائج أنه يمكن الاقتراح بامكانية استخدام الكاروتينات كمواد ملونة طبيعية بغرض صناعة بعض المنتجات الغذائية ذات خواص صحية مرتفعة و آمنة غذائيا.