

Application of FT-IR Spectroscopy for Rapid and Simultaneous Quality Determination of Some Fruit Products

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Abstract: Detection of adulteration is a technical problem. In our work, we have demonstrated the capability of FT-IR spectroscopy as a simple, rapid and accurate method for simultaneously determining sugars, pectin and organic acid contents (citric acid) in some natural and synthetic fruit products. FT-IR technique used to detect the adulterants added such as glucose syrup, synthetic flavor and pigment (allura red & sunset yellow) in jams (strawberry & apricot), apricot sheet and juices (orange, apple and strawberry). The authenticity of strawberry jam was identified by several spectral bands assigned to fructose as deformation OCH, COH and CCH detected at 1425-1414 cm^{-1} . While, spectral peaks due to the presence of glucose, sucrose, citric acid, pectin and allura red pigment were identified at 1029-1045 cm^{-1} , 1058-1061 cm^{-1} , 1351-1378 cm^{-1} , (700 - 705 and 917 - 927 cm^{-1}) and 631 - 633 cm^{-1} , respectively in synthetic strawberry jam. Also, spectral band of fructose in fruity apricot jam was detected at (1414-1416 cm^{-1}); and spectral band of synthetic pigment (sunset yellow) were detected at (770-762 cm^{-1}) in synthetic apricot jam. Apricot sheet could be replaced with carrots as a cheap food material. Adulterated apricot sheet (100% Carrot) was identified with presence of a certain peak at 1089 cm^{-1} , which disappeared in 100% apricot sheet. Also, in carrot-adulterated apricot sheet the intensity of the peaks were higher than those of unadulterated one at the same concentration. FT- IR spectra of synthetic strawberry, orange and apple juices were dominated by specific peaks that attributed to corresponding synthetic pigments at (1637 - 1644 cm^{-1}), (1419 - 1421 cm^{-1}) and (1053 - 1056 cm^{-1}), respectively. The synthetic juices characterized with specific spectral bands of stretching C=O ester of aldehydic and ketonic groups in synthetic flavor at (1726-1731 cm^{-1}) and stretching CO of sucrose at (996-963 cm^{-1}), while these functional groups disappeared in natural juices. Results concerning the prediction of other quality traits using reference analyses were discussed. The obtained results indicated that, FT-IR technique could be easily adapted to detect any adulterants added in jam and juice products.

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Introduction

Authenticity is an important food quality criterion. Regulatory bodies and food processor increasingly demand rapid methods for confirming food authenticity. In recent years, the soft drinks and jams industry have received great attention because of their biological activities and sensory qualities, and have become very important agricultural product in the world (Gurdeniz *et al*, 2007). The products of food industries such as jams and juices have grown in Egypt within the last decade, so that quality control demands using fast and easy technique to evaluate the quality control attributes and discover any adulterants added such as synthetic flavors and colorants become essential.

The adulteration of food products is of primary importance for consumers, food processors and industries. From the legislative point of view, the quality standards were established through the requirement of quality labels that specify the chemical composition of each product (Gallardo *et*

al, 2009). The adulteration frequently involves the replacement of high cost ingredients with cheaper substitutes. Jams and soft drinks are frequently subjected to be adulterated with other synthetic flavors and colorants of lower commercial value (Flores *et al*, 2007). Although the adulteration is done for economic reasons, the action can affect the chemical composition and quality parameters of food (Christy *et al*, 2004).

Several analytical techniques have been developed for detection and quantification of adulteration and authentication of food products, such as mass spectrometry using new type of ion source, direct analysis in real time (Vaclavik *et al*, 2009), nuclear magnetic resonance (NMR) spectroscopy (Jafari *et al*, 2009), infrared spectroscopy (Lerma-Garcia *et al*, 2010), Raman spectroscopy (Heise *et al*, 2005), fluorescence (Poulli *et al*, 2007), gas chromatography (Jafari *et al*, 2009), high performance liquid chromatography (Flores *et al*, 2006) and differential scanning

calorimetry (**Chiavaro et al., 2009**).

Some of these methods are time consuming, expensive, generally destructive of the sample material, and require a high degree of technical knowledge when interpreting the data. Optical spectroscopy techniques have the potential to replace or at least complement some of the classical laboratory methods. In many cases, they allow for a reliable analysis in quality control during production where speed and simplicity of analysis are crucial advantages. FT-IR spectroscopy has shown to be useful for a range of adulteration problems in food sector such as in strawberry purees (**Kemsley et al., 1996**), jam (**Defernez et al., 1995**) and extra virgin olive oil (**Lai et al., 1994**). Apricot and strawberry fruit quality is a multicomponent concept, defined by physical, physiological and biochemical attributes such as firmness, skin and flesh color, ethylene production, respiration rate, sugars, organic acids, pigments, phenolic compounds and volatiles (**Guillot et al., 2006**).

Most instrumental techniques currently required for measuring these parameters are long, expensive and involve a considerable amount of manual work. Therefore, there is a demand for new and rapid analytical methods for assessing quality attributes. Recently, Fourier transform mid-infrared (FT-IR) spectroscopy has become a well-accepted method for the determination of food constituents since it achieves high analysis speed and requires little or no sample preparation. FT-IR spectroscopy often coupled with chemometrics has been used to study different quality attributes in many food samples including fruits, vegetables or beverages e.g. epicuticular wax of apple (**Veraverbeke et al., 2005**), polymethoxylated flavone of orange oil residues (**Manthey, 2006**), vitamin C in powdered mixture and liquid (**Yang & Irudayaraj, 2002**). FT-IR spectroscopy has been widely used for must and wine analysis (**Fernandez & Agosin, 2007**). Moreover, it has become an alternative method for sugar analysis (**Bellon-Maurel et al., 1995**), in food such as mango juices (**Duarte et al., 2002**), soft drinks and fruit juices (**Ramasami et al., 2004**). More recently, this technique has been applied for the analysis of acids in fruits, and in particular apple and tomato (**Beullens et al., 2006**).

At present, FT-IR spectroscopy is often applied in the analysis of plant cell wall polysaccharides. It is a simple, fast and non-destructive method for investigation of fruit juices composition and monitoring the enzymes activity. This technique coupled with chemometrics has been used to study different quality attributes in many food samples including fruits, vegetables or beverages (**Veraverbeke et al., 2005**), olive pulp cell wall

polysaccharides (**Coimbra et al., 1999**), must and wine analysis (**Fernandez & Agosin, 2007**). On the other hand, this technique has become an alternative method for the laborious sugar analysis (**Bellon-Maurel et al., 1995**), in food, e. g. soft drinks and fruit juices (**Ramasami et al., 2004**).

However, there is a few articles available related to the use of FT-IR spectroscopy for analysis of jams and juices adulterated with synthetic flavors and colorants. Therefore, in this research study, we adopted FT-IR spectroscopy to detect and evaluate the adulterants added to jams and juices during their processing.

2. Materials and Methods:

Fresh mature carrot, strawberry, apricot, orange and apple fruits were purchased from local market. Natural juices and jams of strawberry, apricot, apple and orange; and apricot sheet were prepared in Food Technology Lab., National Research Centre.

Strawberry, orange and apple synthetic flavors were obtained from Greatco Company for flavors and fragrances industries, 6th October City, Egypt.

To evaluate strawberry and apricot jam quality, three jam samples of each fruit were prepared using 100% sucrose, 50% sucrose + 50% glucose syrup and 100% glucose syrup. Synthetic jams were prepared by using synthetic flavors, pigment and pectin. Also, the authenticity of apricot sheet, carrot as a cheap food material was used as adulterant in apricot sheet, where sheets of 100% apricot (control) and their adulterated sheets of 50% or 100% carrot were prepared; and identified by FT-IR spectral data. Natural and synthetic strawberry, orange and apple Juices were prepared in Food Technology lab, National Research Center.

Determination of quality traits using reference analyses

Products were analyzed for Total Soluble Solids (TSS) using Digital Hand-held "Pocket" Refractometer, pH was measured by digital pH meter (Hanna, Italy) and viscosity was measured using Brookfield apparatus (spindle No. 2, speed 200 rpm and at 26.8°C). Color (L^* , a^* , b^* and reflectance spectra) was measured using Hunter Lab. Instrument, standardized with white tile (LX No 16379).

The properties of the samples were identified using chemical and physical methods in parallel with FT-IR, where moisture, protein, carbohydrate and ash of fruits products were determined according to **AOAC (1990)**. The spectra or fingerprints of the samples were obtained using FT-IR spectroscopy. The samples of FT-IR (FT-IR-6100 Jasco, Japan) were prepared by using potassium bromide disks.

All results were evaluated statistically using

1425-1414 cm^{-1} in natural jam and adulterated jam with a cheaper sweetener (glucose). The bands around 3390-1640 cm^{-1} due to stretching (OH), (CH) asymmetric, stretching (C=O) and deformation (OH). Another spectral peaks identified due to the presence of glucose, sucrose, citric acid, pectin and allura red pigment at 1029-1045 cm^{-1} , 1058-1061 cm^{-1} , 1351-1378 cm^{-1} , (700 - 705 and 917 - 927 cm^{-1}) and 631 - 633 cm^{-1} , respectively (Guillot *et al.*, 2006).

From the previous results it could be concluded that, the FT-IR bands of fructose in fruity jam and the bands of synthetic color (allura red) in synthetic jam could be used to detect the adulteration of strawberry jam. Also, replacing sucrose with a cheaper sweetener (glucose syrup) was detected. These results are in agreement with those of Defernez *et al.*, (1995) and Kemsley *et al.*, (1996).

Table (3): Representative FT-IR spectra (cm^{-1}) of natural, adulterated and synthetic strawberry Jam.

Assignment	Fresh Fruits	Natural Jam			Synthetic Jam (Flavor + allura red)	
		Sucrose (100%)	Adulterated by replacing sucrose with glucose		Sucrose (100%)	Glucose (100%)
			Glucose (50%)	Glucose (100%)		
Stretching (OH)	3380	3388	3386	3390	3388	3376
CH asymmetric (Stretching)	2931	2931	2931	2930	2931	2929
Stretching (C=O)	1730	---	---	---	---	---
Deformation (OH)	1622	1639	1640	1638	1640	1639
Deformation (OCH, COH, CCH) of fructose	1415	1425	1414	1415	---	---
Citric acid band	1378	---	1365	1365	1351	1365
Citric acid band	1239	1253	1295	1259	1254	1261
Stretching (C-O) of pectin	---	---	1139	1147	---	1147
Stretching (CO, CC and CCO) of sucrose	---	1060	1061	---	1058	---
Vibrational (CO, CCC and vibrational asymmetric of pyranose ring) of glucose	1045	---	1042	1030	---	1029
Rocking (CH ₃) of pectin	927	918	927	925	917	926
Deformation (CH) of fructose	---	818	824	818	---	---

Apricot fruit is limited around the year and adulteration of its products is expected. So we are challenged in the near future to find a fast and easy specific analytical method to confirm the authenticity of apricot products. Hence, natural and synthetic apricot jam were made at laboratory scale as shown in Photo (2), and evaluated by FT-IR spectroscopy.

Photo (2) showed that, there was no observed difference in general appearance of natural apricot jam (100% sucrose) and its adulterated jam with replacing 50% or 100% (GS). Table (4) and Figure (2) show the assignments of spectral bands of the natural apricot, natural apricot jam and synthetic jam. In fruity and synthetic apricot jam the differences were detected as similar in fruity and synthetic strawberry jam. Moreover, there is a specific band at 770-762 cm^{-1} for sunset yellow pigment in synthetic apricot jam. The natural β -carotene pigment and synthetic sunset yellow pigment were identified in fresh apricot fruit, natural apricot jam and synthetic apricot jam at 1630-1650 cm^{-1} that attributed to Stretching

(H-C=CH) of β -carotene and sunset yellow, respectively.

The obtained results suggest that, FT-IR spectroscopy could be used to differentiate between natural and synthetic apricot jam by using spectral band of synthetic pigment (sunset yellow at 770-762 cm^{-1}), and spectral band of fructose in fruity jam (1414-1416 cm^{-1}). Apricot sheet is one of the most important apricot products especially in Ramadan month; so, it may be replaced with carrots as a cheap food material. Photo (3) shows sheets of 100% apricot (control) and their adulterated sheets of 50% or 100% carrot.

The color quality of the studied apricot sheets were evaluated instrumentally by using Hunter color parameter (L^* , a^* , b^* and uv-visible spectra analysis). Hunter color parameter (L^* , a^* , b^*) as shown in Table (5) indicated that, the sheet (apricot: carrot 1:1) had the highest value of lightness (L^*), redness (a^*) and yellowness (b^*), where they reached 33.05, 18.45 and 14.76, respectively, while the sheet of carrot 100% replacement ranked as the second of L^* , a^* and b^* ; and the sheet containing apricot 100% was ranked as a third.

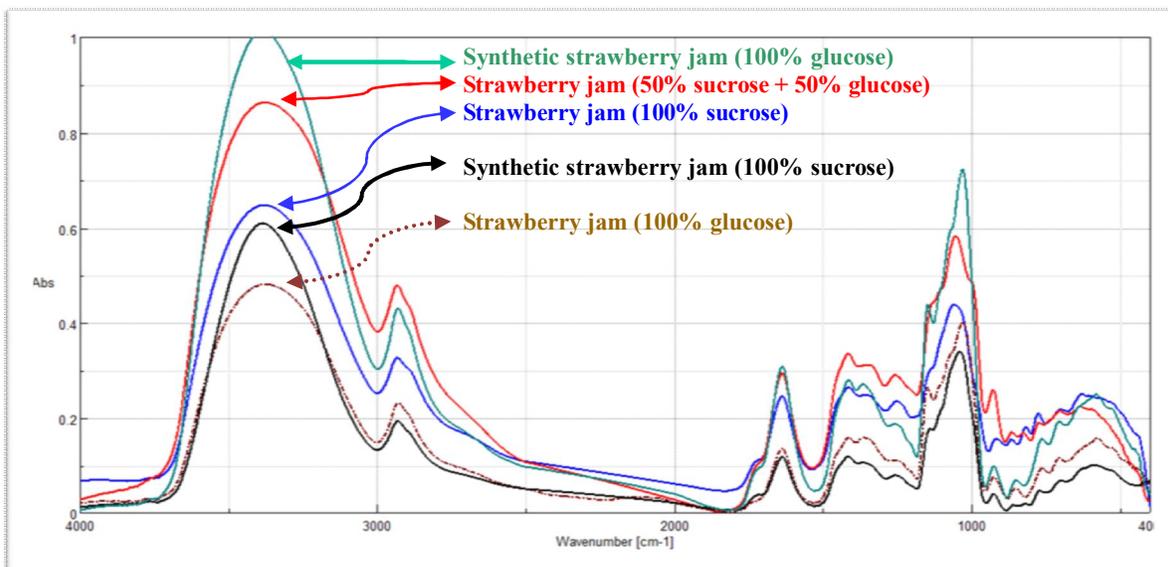


Figure (1): FT-IR spectra (cm^{-1}) of natural, adulterated and synthetic strawberry Jam.



A = Synthetic B = Sucrose + Glucose (1:1) C = Glucose (100%) D = Sucrose (100%)

Photo (2): Apricot jam and its natural, adulterated and synthetic jam.

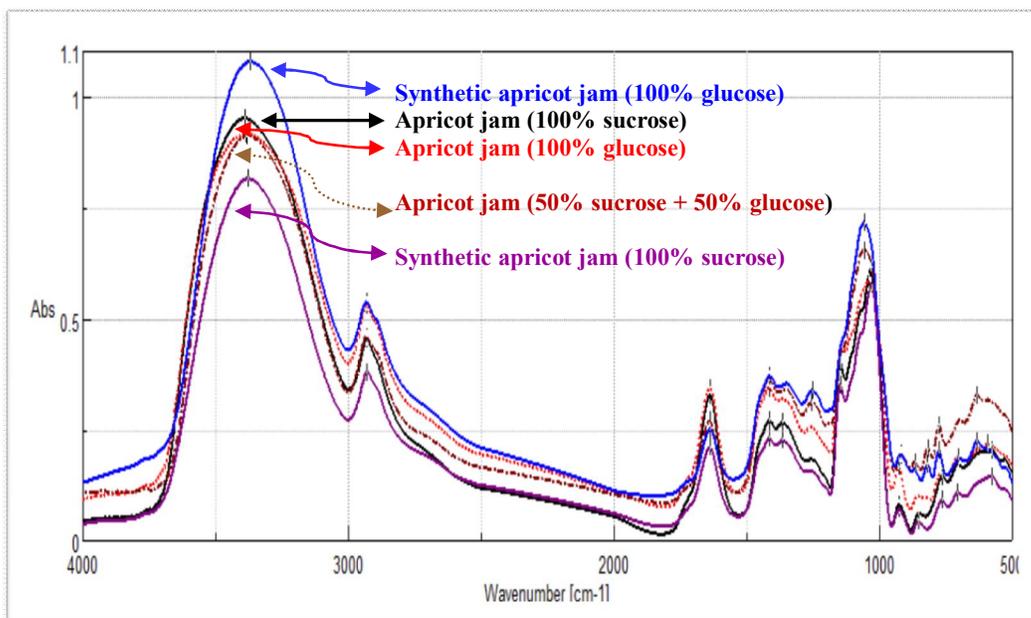


Figure (2): FT-IR spectra (cm^{-1}) of natural, adulterated and synthetic apricot Jam.

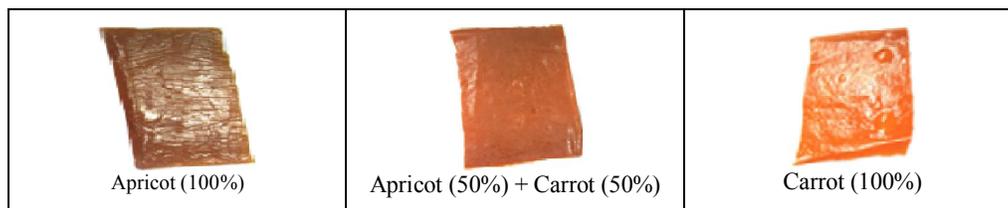


Photo (3): Apricot sheet (100% Apricot) and its adulterated sheets with 50% or 100% carrot.

Table (5): Hunter color parameter of apricot sheet (100% apricot) and its adulterated sheet with 50% or 100% carrot.

Sample	L*	a*	b*
Apricot 100%	23.82 ^c ± 0.4	13.23 ^c ± 0.37	11.18 ^b ± 0.33
Apricot: carrot (1:1)	33.05 ^a ± 0.37	18.45 ^a ± 0.6	14.76 ^a ± 0.32
Carrot 100%	30.5 ^b ± 0.9	14.67 ^b ± 0.58	14.23 ^a ± 0.25
L S D	1.213	0.685	0.598

Table (4): FT-IR spectra (cm⁻¹) of apricot fruits and their natural and synthetic jams.

Assignment	Apricot Fruit	Natural Apricot Jam (citric + pectin)			Synthetic Apricot Jam (citric + pectin + flavor + sunset pigment)	
		Sucrose (100%)	Sucrose : Glucose (1:1)	Glucose (100%)	Sucrose (100%)	Glucose (100%)
Stretching (OH)	3371	3385	3382	3391	3371	3385
Stretching (CH) asymmetric	2930	2930	2933	2930	2928	2931
Stretching (C=O)	1718	-----	-----	-----	-----	-----
Stretching (H-C=CH) of β-carotene and sunset yellow	1650	1639	1639	1630	1639	1638
Deformation (CH)	1455	-----	-----	-----	-----	-----
Deformation (OCH, COH, CCH) of fructose	1381	1415	1416	1414		
Citric acid band	1288	1364	1347	1365	1362	-----
Citric acid band	1232	1257	1257	1258	1258	1261
Stretching (C-O) of pectin	1136	1139	-----	1147	1147	-----
Stretching (CO, CC and CCO) of sucrose	1045	1058	1053	-----	1052	-----
Vibrational (CO, CCC and vibrational asymmetric of pyranose ring) of glucose	-----	1040	1029	1030	-----	1035
Rocking (CH ₃) of pectin	926	925	927	926	926	927
Deformation (CH) of fructose	837	820	825	827	-----	-----
Sunset yellow pigment	-----	-----	-----	-----	762	770

Visible spectroscopy was used to evaluate the spectra in apricot sheet (100% apricot) and its adulterated sheets (apricot + carrot 1:1 and 100% carrot). Spectra of β-carotene pigment showed two neighboring spectral bands at 450 and 475 nm. Natural apricot sheet (100% apricot) showed the same bands as found in standard β-carotene. The

adulterated sheets of apricot plus carrot (1:1) and 100% carrot showed spectral band at (425, 450 and 475 nm); and a single broad band at 475 nm, respectively as illustrated in Figure (3).

The obtained results of Hunter color parameter and spectrophotometer show the need to develop other technique to detect and confirm the

adulteration of apricot sheet. So, FT-IR was used to detect the variation between apricot sheet and its adulterated sheets of carrots.

Data reported in Table (6) and Fig (4) showed that, 100% carrot sheet was identified with specific peak at 1089 cm^{-1} , while 100% apricot sheet was characterized by a specific peak at 576 cm^{-1} , and 1374 cm^{-1} was appeared in the adulterated sheet (50% carrot). The spectra of all samples were characterized with the following assignment, $3424\text{-}3428\text{ cm}^{-1}$ (OH stretching), 2925 cm^{-1} (CH stretching asymmetric and symmetric at

$2858\text{-}2857\text{ cm}^{-1}$), $1743\text{-}1745\text{ cm}^{-1}$ (stretching C=O ester) of pectin, $1640\text{-}1647\text{ cm}^{-1}$ (vibrational CO bond), $1457\text{-}1449\text{ cm}^{-1}$ (stretching CH), $1155\text{-}1162\text{ cm}^{-1}$ (C-C stretching), $1042\text{-}1025\text{ cm}^{-1}$ (vibrational CO of sucrose) and $707\text{-}721\text{ cm}^{-1}$ (β -carotene), respectively. These assignments are in agreement with **Bureau *et al.* (2009)**. Also, Figure (4) showed that, the peaks' intensities of carrot-adulterated sheet of apricot were the highest among 100% carrot and 50% carrot at the same concentrations.

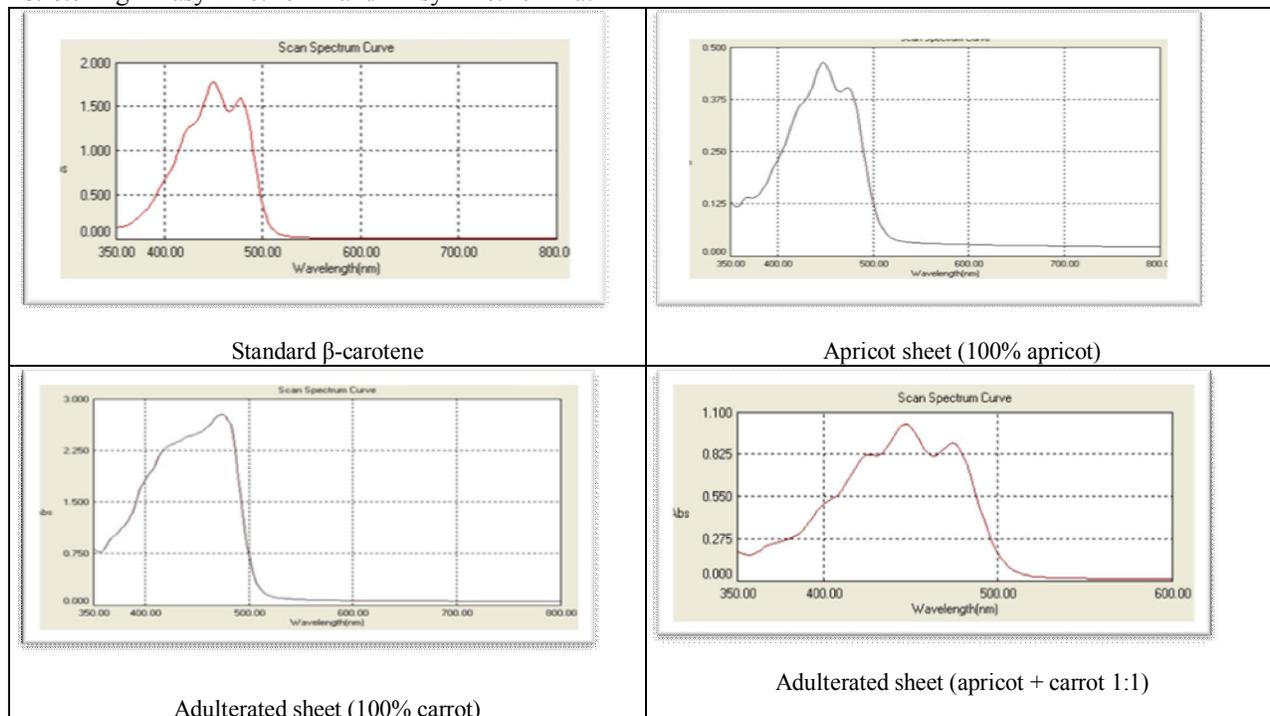


Figure (3): Reflectance of apricot sheet (100% apricot) and their adulterated sheets with 50% or 100% carrot.

Table (6): FT-IR assignment of apricot sheet (100% apricot), and its adulterated sheets (apricot + carrot 1:1 & 100% carrot).

Assignment	Adulterated Apricot Sheet		
	Apricot Sheet Apricot (100 %)	Carrot + Apricot (1:1)	Carrot (100 %)
Stretching O-H	3425	3428	3424
Stretching C-H asymmetrical	2925	2925	2925
Stretching C-H symmetrical of CH_3	2858	2857	2857
Stretching C=O ester of pectin	1739	1745	1743
Stretching (H-C=CH)	1647	1647	1640
Stretching CH	1449	1457	1457
Stretching C-O-C ring	---	1374	---
Stretching C-H asymmetrical	1155	1162	1155
C-C stretching of carbohydrates	---	---	1089
Vibrational CO of sucrose	1025	1030	1042
β -carotene	707	721	711

Natural and synthetic strawberry, orange and apple juices were prepared at laboratory scale as

shown in Photo (4). Table (7) showed that, natural strawberry juice characterized with its higher

viscosity (101 cp) and lower TSS than synthetic juice, so that TSS does not reflected the authenticity of the strawberry juice.

Also, physical quality of natural orange, apple and strawberry juices were determined and compared with synthetic juices. Table (7) showed that, natural orange and apple juices was slightly affected with replacing fruits with synthetic flavor and color, where viscosity of natural and synthetic orange juice were 43.8 and 50.6 cp, respectively; also natural and synthetic apple juice were 18.6 and 18.5 cp, respectively. The same trend was observed in TSS and pH. This slight variation in physical properties indicated that we need another

fast and easy method to identify the authenticity of orange and apple juices.

Color quality of natural and synthetic strawberry, orange and apple juices were evaluated separately. Table (8) showed that, all natural juices were characterized with its higher color parameter of lightness (L^*), redness (a^*) and yellowness (b^*) than synthetic juices. This result could depend on juice dilution, type and concentration of synthetic pigment. So, the possibility of using FT-IR as a fast and easy technique to detect authenticity of some fruit juices was examined.

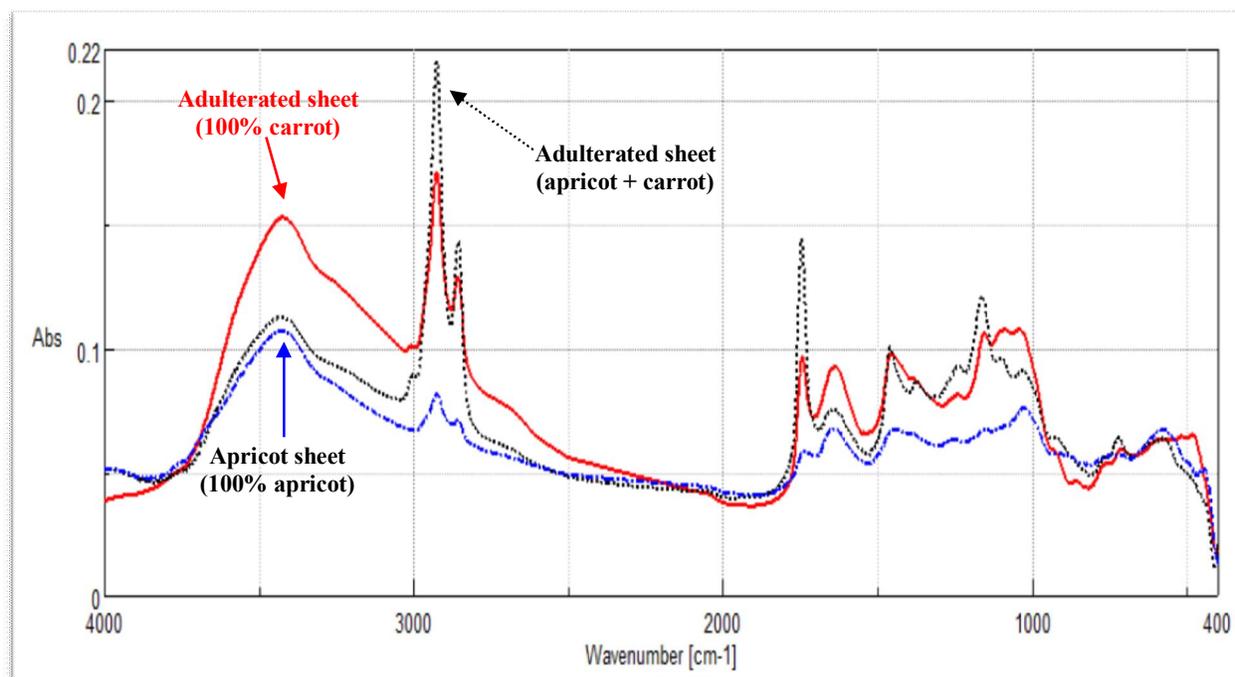


Figure (4): FT-IR spectra (cm^{-1}) of apricot sheet (100% apricot) and their adulterated sheets with 50% or 100% carrot.

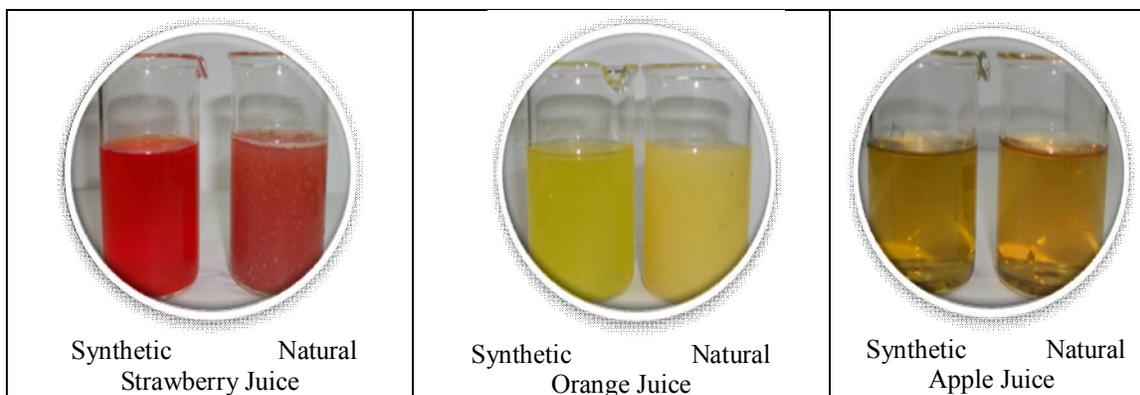


Photo (4): Natural and synthetic strawberry, orange and apple Juices.

Table (7). Physical properties of natural and synthetic strawberry, orange and apple juices.

Physical Parameter	Strawberry juice		Orange Juice		Apple Juice	
	Natural	Synthetic	Natural	Synthetic	Natural	Synthetic
pH	3.67±0.17	2.13±0.01	3.47 ± 0.34	2.14 ± 0.01	3.63 ± 0.02	2.14 ± 0.06
TSS (Brix)	12.1±0.05	15.87±0.29	13.55±1.22	17.87 ± 0.64	9.56 ± 0.19	13.6 ± 0.06
Viscosity (cp)	101.00±1.00	20.07±0.81	43.8 ± 0.5	50.6 ± 0.3	18.6 ± 0.4	18.5 ± 0.1

Table (8): Hunter color parameters of natural and synthetic strawberry, orange and apple juices.

Sample	L*	a*	b*
Natural strawberry juice	29.85 ^a ± 0.72	30.25 ^a ± 1.03	12.7 ^b ± 0.44
Synthetic strawberry juice	9.31 ^c ± 0.35	13.19 ^e ± 0.38	4.8 ^t ± 0.21
Natural Orange Juice	52.91 ^a ± 1.156	4.76 ^c ± 0.0404	45.72 ^b ± 1.91
Synthetic orange juice	6.84 ^e ± 0.0902	0.383 ^d ± 0.005	4.00 ^e ± 0.623
Nature apple juice	20.60 ^a ± 0.49	3.24 ^a ± 0.072	8.26 ^a ± 0.062
Synthetic apple juice	6.03 ^b ± 0.252	0.56 ^b ± 0.0199	2.35 ^b ± 0.0551

Natural strawberry, apple and orange juices were evaluated by FT-IR spectral data as shown in Table (9). Stretching (OH), (CH) asymmetric, deformation (OH), deformation of (CH) functional groups in natural orange, apple and strawberry juices were identified in

the region from 3441-1415 cm⁻¹. While, stretching (CO), (CC) and (CCO) of sugar and stretching C-OH ring were located at 1045-1058 cm⁻¹ and 924-930 cm⁻¹, respectively. These assignments are in agreement with **Irudayaraj and Tewari (2003)**.

Table (9): Assignments of natural orange, apple and strawberry juices.

Assignments	Orange	Apple	Strawberry
Stretching (OH)	3441	3421	3430
Stretching (CH) asymmetric	2930	2938	2931
Deformation (OH)	1632	1638	1622
Deformation (CH) of fructose	1426	1424	1415
Stretching (CO, CC & CCO) of sugar	1053	1058	1045
Rocking CH ₃ of pectin	930	924	927

FT- IR spectra of synthetic strawberry, orange and apple juices were dominated by specific peaks that were attributed to synthetic pigment at 1637 - 1644 cm⁻¹, 1419 - 1421 cm⁻¹ and 1053 - 1056 cm⁻¹ (Table 10). From the FT-IR results of natural and synthetic juices, we can conclude that, the synthetic juices can be characterized with specific bands of stretching C=O

ester of aldehydic and ketonic groups in synthetic flavor which used in the manufacturing process of synthetic juices at 1726-1731 cm⁻¹ and stretching CO of sucrose at 996-963 cm⁻¹, while these functional groups disappeared in natural juices (**Ramasami et al., 2004**).

Table (10): Assignment of synthetic strawberry, orange and apple juice.

Assignment	Orange juice	Apple juice	Strawberry juice
Stretching O-H	3378	3395	3401
Stretching C-H asymmetrical	2932	2932	2932
Deformation OH	1644	1644	1637
Stretching C=O ester	1731	1726	1730
Stretching (COO ⁻¹) of pectin	1419	1421	1419
Vibrational CH ₃ deformation	---	---	1384
Citric acid bands	1248	1257	1262
Stretching C-O-C of glycosidic ring	1133	1134	1139
Stretching CO of sucrose	1055	1056	1053
Stretching CO ester of synthetic flavor	997	996	663
Stretching C-OH	929	928	926
Vibrational CH	863	863	834
CH ₂ group deformation	593	594	605

In conclusion, the results of this work recommend FT-IR spectroscopy as a potential analytical rapid, economic and nondestructive tool to determine the quality attributes and to detect any adulterants, added in food industries, of some food products as jams and juices.

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