INTERNATIONAL JOURNAL OF AGRICULTURE & BIOLOGY
1560–8530/2006/08–2–186–190
http://www.fspublishers.org

Comparative Physical Examination of Various Citrus Peel Essential Oils

MUHAMMAD MUSHTAQ AHMAD[1], SALIM-UR-REHMAN, FAQIR MUHAMMAD ANJUM AND EHSAN ELAHI BAJWA
Institute of Food Science and Technology, University of Agriculture, Faisalabad–38040, Pakistan
[1]Corresponding author's e-mail: dr_mushtaq786@hotmail.com

ABSTRACT

Physical parameters of various citrus peel essential oils were determined in this study. Essential oils from the peels of Kinnow (*C. reticulata*, var. mandarin), Fewtrell's early (*C. reticulata*, var. tangerine), Malta (*C. sinensis* var. malta), Mousami (*C. sinensis* var. mousami), grape fruit (*C. paradisi*) and eureka lemon (*C. limon*) were extracted by applying cold expressing method. Eureka lemon had the highest peel portion i.e. 45.0%. Similarly, Malta peel had the highest oil yield i.e. 1.21%. Physical evaluation of oils delineated that Fewtrell's early peel oil had the lowest specific gravity i.e. 0.841, whereas, highest refractive index and optical rotation were found in Grape fruit (1.472) and Malta (93.257) oils. Similarly, the lowest evaporation residue (3.122%) has been observed in Grape fruit peel oil. All the essential oils were found soluble in different ratios, ranging from slight haziness to clearly soluble in 95% alcohol. Flavor of these oils was also evaluated through olfactory method. Eureka lemon and Mousami peel oils had the pleasant sharp and strongest flavor followed by kinnow, Fewtrell's early, Malta and Grape fruit.

Key Words: Essential oils; Citrus fruits

INTRODUCTION

Essential oils are vegetable products whose constituents are basically complex mixture of terpenic hydrocarbons and oxygenated derivatives such as aldehydes, alcohols and esters. These are accumulated mainly in secretory cavities scattered through out the fruit peels and leaf. Among many sources, citrus fruits peels are the most familiar and rich source of essential oils. Peels of citrus fruit comprise of two layers, red outer layer as flavedo and inner white layer as albedo (Nagi *et al*., 1977). The flavedo layer contains essential oils in the range of 0.5 to 3.0 kg/ton of fruit (Sattar *et al*., 1992).

Essential oils of these fruits are composed of complex mixture of 200 natural plant components (Diaz *et al*., 2004) that can be grouped in to two fractions: a volatile fraction constitutes 90 - 95% of the whole oil and contains monoterpenes; aliphatic sesquiterpenes and oxygenated derivatives; bicyclic terpenes and sesquiterpenes; aromatic hydrocarbons; aliphatic aldehydes, alcohols and esters. Non-volatile residue constitutes from 1 - 10% of the whole oil and contains fatty acids, esters carotenoids, couramins, psoralens, paraffine waxes, flavonoids and steroids, which accumulate in balloon like cells (oil glands), glandular trichomes and oil or resin ducts (Clifford, 1999).

Physical behavior of essential oils is designed by the natural plant components present in them. Physical parameters are widely quoted to evaluate purity and quality of essential oils. In trading of these oils, any impurity or adulteration, if practiced, can also be detected easily by matching the physical characteristics of essential oils with the already established required standards (Guenther, 1955).

The information on these physical attributes regarding Pakistani citrus peel oils is scanty. Therefore, the main objective of this study was to determine and compare the physical attributes of various citrus peel oils.

MATERIALS AND METHODS

Research was conducted at the Institute of Food Science and Technology, University of Agriculture and Food Technology Section, Ayub Agricultural Research Institute, Faisalabad. Fully ripened fresh fruits of Kinnow, Fewtrell's early, Malta, Mousami, Grape fruit and Eureka lemon were procured from the commercial orchard in the area of Okara district and local market. Chemicals used in this study were procured from the Sigma Aldrich, USA. The following steps were taken in this study.

Extraction of essential oils. Citrus fruits were peeled manually, % peel portion was calculated and then shredded to a size of 2.0 × 0.3 cm, by using citrus peel shredder (Turnsbull's, Model No. 266069, UK.). Cold expressing was done at ambient temperature using locally made hydraulic press at 15000 psi. The extraction mixture so obtained was centrifuged under controlled conditions of time (45 min.), temperature (28 - 30^{o}C) and rpm (15000) by using refrigerated centrifuge (Model No. ALC - 4227 R, Germany).

After centrifugation, each oil was separated by using separating funnel. The oils so obtained of each fruit were then treated with sodium sulfate in order to remove any traces of moisture % yield was calculated and then stored at

4°C in amber colour bottles.

Physical examination. Physical examination regarding specific gravity and refractive index (AOAC, 2000) and evaporation residue, solubility, optical rotation and flavor test (Guenther, 1955) of these oils was carried out in this study. Each parameter was repeated thrice. The data was analysed statistically by using the completely randomized design (CRD) and DMR Test to determine the level of significance (Steel *et al.,* 1997).

RESULTS AND DISCUSSION

% peels of various citrus fruits. The data regarding % peel portion and yield of essential oils of Kinnow, Fewtrell's early, Malta, Mousami, Grape fruit and Eureka lemon are presented in Table I.

The results indicated that % peel portion of Eureka lemon was the highest and ranked at top with 45.0%, followed by Mousami 41.0%, Grape fruit 38.0%, Malta 37.0%, Kinnow 32.0% and Fewtrell's early 30.0%. These results are in line with findings of various scientists. Weiss (1997) found that peel portions of sweet orange, lemon and mandarin were 25.0, 40.0 and 28.0%, respectively. Similarly, it was found that various citrus fruits such as mandarin, orange, grape fruit and lemon had the peel in the range from 25.0 - 45.0% (Hakim & Harris, 2001)..

In another study (Manthey & Grohmann, 2001), it was observed that the peel portions of various citrus peels such as tangerine, orange, grape fruit and lemon were in the range from 25.6 - 33.0, 21.5 - 38.1, 33.7 - 36.4 and 32.0 - 46.6%, respectively. Although, there are some sort of difference between the % peels of the current citrus fruits and similar previous research studies mentioned above, however, this difference could be due to the variation in climate and soil conditions (Huet, 1991).

% yield of essential oils. After extracting through cold expressing method, the % peel oil of each fruit was calculated and the results are given Table I. The results indicated that oil yield of Malta variety was 1.21%, the highest among all the citrus fruits, under this study. The second highest yield has been observed in Eureka lemon peel, which was 1.12%, whereas, Mousami, Grape fruit, Kinnow and Fewtrell's early followed afterward and had 0.98, 0.73, 0.32 and 0.22%, respectively.

The results of this study are supported by the previous studies of various scientists. Weiss (1997) delineated that the total oil contents of sweet orange, eureka lemon and mandarin were 0.80, 0.90 and 0.80%, respectively. He also mentioned that the oil yield of bergamot orange was 0.45 - 0.65% depending mainly on climacteric conditions, with 0.50% as an average. Ohloff (1990) described that yield of cold pressed peel oils of orange, bergamot and petitgrain was 0.5% each; whereas, mandarin oil had 0.2% yield. Similarly, he also mentioned that 850 kg of carefully picked orange flowers yielded 1 kg of neroli oil after steam distillation. Yield of citrus essential oil differs with individual plant species ranging in most cases from 0.2 to 2.0% (Anonymous, 2004a).

Table I. Percents peel portion and yield of various citrus peel essential oils

Name of fruit	Percent peel portion	Percent oil yield
Kinnow	32	0.32
Fewtrell's early	30	0.22
Malta	37	1.21
Mousami	41	0.98
Grapefruit	38	0.73
Eureka lemon	45	1.12

From the above results and discussion it is inferred that cold expressed method applied in this present study was the most favorable method and the yields of Kinnow, Fewtrell's early, Malta, Mousami, Grape fruit and Eureka lemon peel oils were matching and conforming the results of previously done similar scientific studies. However, the oil yield of Kinnow and Fewtrell's early were low as compared to other fruits, which could be due to the presence of some gums in the peels as has been experienced during oil extraction phase in this study.

Physical Examination of Oils

Specific gravity. The essential oils of Kinnow, Fewtrell's early, Malta, Mousami, Grape fruit and Eureka lemon were subjected to analysis for specific gravity as a first physical parameter. The results obtained were then analysed statistically for F-value and comparison of means (Table II & III). The results of comparison of means revealed that the specific gravities of Kinnow, Fewtrell's early, Malta, Mousami, Grape fruit and Eureka lemon oils were 0.842, 0.841, 0.845, 0.846, 0.855 and 0.846, respectively.

The results of specific gravity in present investigation are corroborated and substantiated by the previous work of Guenther (1964), who thoroughly studied the physical properties of oils of Brazilian mandarin peel, Palestine sweet orange peel and California lemon peel and found that the specific gravities (15°C) were in the range of 0.854 to 0.858. In another study (Keterson *et al*., 1971), the specific gravities of cold expressed lemon oils were observed to be in the range from 0.852-0.858. Similarly, while preparing the material specifications of various essential oils, Statfold Seed Oil Development Ltd. (2004) described the specific gravities of sweet orange, tangerine peel, grape fruit peel, lemon and lime peel, oils as 0.850, 0.848, 0.855, 0.861, 0.861, respectively.

In another study (British Pharmaceutical Codex, 2004), oils obtained by mechanical means from the fresh peels of the sweet orange (*C. aurantium*, var. Linn.), bitter orange (*C. aurantium* var. Bigaradia) and lemon (*C. medica*, var. Linn.) were analysed for specific gravities, which were found to be in the range from 0.847 to 0.854 (0.842 to 0.846 at 25°C). In the Certificate of Analysis (Lockey, 2004a.b), the specific gravities of tangerine and grape fruit essential oils were calculated as the same i.e. 0.847 and 0.851. Greenwood Associates, Inc. (2004) also studied the specific

Table II. Analysis of variance of physical parameters of citrus peel oils

Parameters	Source	Degree of Freedom	Sum of Squares	Mean squares	F-value
Specific gravity	Treatments	5	0.000	0.00007568	136340.881**
	Error	12	0.000	0.00000000055	
Refractive index	Treatments	5	0.000	0.000022	3.848*
	Error	12	0.000	0.0000056	
Optical rotation	Treatments	5	629.424	125.885	2134.915**
	Error	12	0.708	0.059	
Evaporation residue	Treatments	5	89.432	17.886	22900.664**
	Error	12	0.009	0.00078	

Highly significant at P<0.01, Significant at P<0.05

Table III. Mean values of physical parameters of essential oils

Name of oils	Specific gravity	Refractive index	Optical rotation	Evaporation residue
		Means		
Kinnow	0.842d	1.464c	84.447d	5.255b
Fewtrell's early	0.841^{e}	1.469b	92.560b	4.108c
Malta	0.845c	1.469b	93.257a	2.533f
Mousami	0.846b	1.469b	92.390b	3.749d
Grapefruit	0.855a	1.472a	86.705c	9.290a
Eureka Lemon	0.846b	1.469b	76.666e	3.122e
LSD value	0.01779	0.004210	0.4321	0.01779

Mean values sharing same letters are non-significant to each other (P<0.05).

gravity of lemon oil, which was in the range from 0.849 to 0.855.

Refractive index. The results of refractive indices of peel oils are presented in Table II and III. The statistical analysis of the results of this parameter were, revealed that the oils are significantly different to each other ($P < 0.05$). The comparison of means of these oils, are ranged from 1.464 to 1.472.

These results of refractive index are corroborated by the findings of many scientific works. Statfold Seed Oil Development Ltd (2004) in the Material Specifications Sheets, specified that the refractive index of orange sweet, tangerine, grape fruit, lemon and lime (single fold) oils were 1.477, 1.474, 1.476, 1.474 and 1.475, respectively. Similarly, the Certificate of Analysis (Lockey, 2004a, b) specified that the refractive indices of tangerine and grape fruit essential oils were 1.474 and 1.476. In other studies (Greenwood Associate, 2004), it was found that the refractive indices of lemon peel oils were in the range from 1.473 - 1.476 and 1.474 - 1.476, respectively.

Optical rotation. In this study, the optical rotations of the peel oils of Kinnow, Fewtrell's early, Malta, Mousami, Grape fruit and Eureka lemon were 84.447, 92.560, 93.257, 92.390, 86.705 and 76.666, respectively. The analysis of variance (Table II) was calculated on the basis of these results, which showed highly significant difference among all the peel oils ($P < 0.01$). The comparison of means given in Table III indicated that Malta has the highest mean value (93.257) and Eureka lemon the lowest (76.666).

These results are also supported by the previous work of various scientists. Guenther (1964) thoroughly studied the physical properties of oils of Brazilian mandarin peel, Palestine sweet orange peel and California lemon peel and found optical rotation in the range of + 63 to + 74 degree. In another study (British Pharmaceutical Codex, 1973), oils obtained by mechanical means from the fresh peels of the sweet orange (*C. aurantium*, var. Linn) and bitter orange (*C. aurantium* var. Bigaradia) and lemon (*C. medica*, Linn. var.) were found to be having an optical rotation in the range from + 95 to + 98 and + 58 to + 65 degree at 20^{o}C, respectively.

Weiss (1997) observed that the optical rotations of Indian cold pressed mandarin, sweet orange, grape fruit and lemon oils were + 95, + 95 to + 98, + 93 and + 67 to + 80 degrees, respectively. He also found that an optical rotation of cold pressed peel oil of Brazil orange was in the range from +94 to +99. Greenwood Associates (2004) in the process of developing Product Specifications suggested that the optical rotations of lemon oil ranged between + 57 to + 65 degree.

Evaporation residue. The evaporation residue of peel oil of each citrus fruit in this study was determined. F-value was then calculated by analyzing the results of this parameter statistically (Table II), which showed the highly significant difference ($P < 0.01$) among all the essential oils. The comparison of means (Table III) showed that Malta oil had the lowest evaporation residue i.e. 2.533%; whereas, Grape fruit oil had the highest i.e. 9.290%. The other oils such as Kinnow, Fewtrell's early, Mousami and Eureka lemon had 5.255, 4.108, 3.749 and 3.122%, evaporation residues, respectively.

Regarding this quality parameter, the current results are confirmed by the previous findings of Shaw *et al.* (1971) and Shaw and Coleman (1974), who determined that lime and orange oils contained 7 and 1% of non-volatile oil constituents, respectively. Wolfred *et al.* (1971) reported that orange, tangerine, grape fruit and lemon oils have 1.5, 4.0, 7.0 and 2.0% of the evaporation residue as non-

volatiles.

Another measure of citrus oil non-volatiles, reported by Moshonas and Shaw (1971 & 1974) showed that evaporation residue in tangerine oil and grape fruit essence, were 4 and 6%, respectively. Similarly, Weiss (1997) also observed that the evaporation residue of cold pressed oils of Indian mandarin, sweet orange, grape fruit and lemon were 2.93, 2.53, 6.30 and 2.40%, respectively. In order to set the standards of essential oils, British Pharmaceutical (1973) described that the evaporation residue of the bergamot oil was in the range from 5 - 6%. Statfold Seed Oil Development Ltd. (2004) also observed that non-volatile residue of the sweet orange oil was 11%.

Solubility in alcohol. In this study, solubility values of the essential oils were determined by mixing 90 and 95% ethyle alcohols, separately. The results in Table IV showed that all the essential oils were found to be insoluble and created haziness when 90% ethyl alcohol was added in to each oil. However, when the solubility of these alcohols was checked against 95% ethyl alcohol, they showed different behaviour. The Kinnow oil was insoluble with slight haziness in even 10 mL of alcohol, whereas, Fewtrell's early, Malta, Mousami, Grape fruit and Eureka lemon were clearly soluble in 9.2, 8.7, 9.0, 9.5 and 7.0 mL of ethyl alcohol.

These results are substantiated by the research work of various scientists. Weiss (1997) observed that the essential oils of mandarin, sweet orange and lemon have the solubility level of 3:5 in 85%, 1:2 - 3 in 90%, 1:3 in 95% v/ v alcohols, respectively. Cambodian Green Consultancy Services (2002), while developing the export standards of essential oils, observed that citronella oil had 1:2 v/ v solubility level in 80% alcohol. Similarly, Greenwood Associates (2004) studied that the solubility level of cold pressed lemon oil was 1:3 v/v in 90% alcohol. British Pharmaceutical (1973) in its codex also mentioned that the oils of orange and lemon have the solubility level of 1:7 and 1:12 v/ v in absolute alcohol, respectively.

Flavor test. The essential oils of Kinnow, Fewtrell's early, Malta, Mousami, Grape fruit and Eureka lemon were tested for flavor, in this study. Results in (Table V) revealed that Kinnow peel oil possessed a pleasant and strong flavor. Fewtrell's early and Malta peel oils also exhibited pleasant but stronger flavor than Kinnow peel oil. Similarly, the flavor of Mousami peel oil was also pleasant but the strongest in flavor perception. Whereas, Grape fruit peel oil showed some-what varied behavior and had pleasant but weak in flavor as compared to others. However, Eureka lemon oil was highly prominent and exhibited pleasant sharp and strongest flavor, among them.

These results are confirmed by the data sheet of Statfold Seed oil Development Ltd. (2004), which indicated that the odor of tangerine and grape fruit peel oils was fresh, sweet orange like and pleasant citrus like, respectively. Similarly, the oils of orange sweet and bitter orange and lemon obtained (British Pharmaceutical Codex, 1973) by mechanical means from the fresh peels of these fruits had characteristic odor and pleasant and fragrant odor, respectively. Similarly, Brazilian orange oil had orange fresh juicy sweet flavor (Anonymous, 2004b).

CONCLUSION

From the above results and discussions, it is concluded that physical measurements and purity of citrus peel oils of Kinnow, Fewtrell's early, Malta, Mousami, Grape fruit and Eureka lemon, carried out in this study, are corroborated and closely matched with the scientific out puts of similar previous studies. These delineations also confirmed the efficiency, validity and reliability of mechanical mean of cold expressing extraction method. Although, some sort of differences between the results of present and previously conducted scientific contributions have been experienced, which might be due to several significant environmental and physiological factors such as tissues age (maturity), climatic

Table IV. Solubility in alcohol of essential oils of various citrus peels (25°C)

Name of fruits	Solubility in 90% Alcohol		Solubility n 95% Alcohol	
	R1	R2	R1	R2
Kinnow	Insoluble with haziness	Insoluble with haziness	Soluble with slight haziness in 10.0 mL	Soluble with slight haziness in 10.0 mL
Fewtrell's early	Insoluble with haziness	Insoluble with haziness	Clearly soluble in 9.2 mL	Clearly soluble in 9.2 mL
Malta	Insoluble with haziness	Insoluble with haziness	Clearly soluble in 8.7 mL	Clearly soluble in 8.7 mL
Mousami	Insoluble with haziness	Insoluble with haziness	Clearly soluble in 9.0 mL	Clearly soluble in 9.0 mL
Grapefruit	Insoluble with haziness	Insoluble with haziness	Clearly soluble in 9.5 mL	Clearly soluble in 9.5 mL
Eureka lemon	Insoluble with haziness	Insoluble with haziness	Clearly soluble in 7.0 mL	Clearly soluble in 7.0 mL

Table V. Flavor test of essential oils of various citrus peels

Name of fruit	No. of replications	
	R1	R2
Kinnow	Pleasant with strong flavor	Pleasant with strong flavor
Fewtrell's early	Pleasant with stronger flavor	Pleasant with stronger flavor
Malta	Pleasant with stronger flavor	Pleasant with stronger flavor
Mousami	Pleasant with strongest flavor	Pleasant with strongest flavor
Grapefruit	Pleasant with less strong flavor	Pleasant with less strong flavor
Eureka lemon	Pleasant with sharp and strongest flavor	Pleasant with sharp and strongest flavor

season, harvesting time, size of the fruit, soil type, type of extraction method and length of time the fruit is permitted to stay in the cold room and storage conditions (Guenther, 1964; Huet, 1991; Weiss, 1997).

REFERENCES

Anonymous, 2004a. Citrus Fruits. http://alfa.ist.utl.pt/ field/flaves/ sec3/ citrus.html. Accessed on 17/9/2004

Anonymous, 2004b. Lemon Oil. Lemon Oil Analysis Batch kb01028. file://C:\Documents%20and %20and %20settings\ Food%20 Technology\ My%20Documents\Lemon. Accessed on 26/7/2004

A.O.A.C., 2000. *Official Methods of Analysis*, Vol. II, 16th Edition, Virginia 22201, Arlington

British Pharmaceutical Codex, 1973. Pharmaceutical Society of Great Britian, London, United Kingdom

Cambodian Green Consultancy Services, 2002. *Export Development of Essential Oils and Spices by Cambodia*, Pp: 70–2

Clifford, A.A., 1999. Extraction of natural products with superheated water. In: Proceedings of the GVC- Fachausscss 'High Pressure chemical engineering' Mar. 3-5, Karlsruhe, Germany

Diaz, S., S. Espinosa and E.A. Brignole, 2005. Citrus peel oil deterpenation with supercritical fluids: optimal process and solvent cycle design. *J. Supercritical Fluids*, 35: 49–61

Greenwood Associates, Inc., 2004. *Product Specification. Lemon oil from inland fruit*. 310 S. Happ Road, Northfield, Illinois 60093, USA

Guenther, E., 1955. *The Essential Oils. History, Origin in Plants Production Analysis*. Vol. 1, D. Van Nostrand Co. Inc. NY

Guenther, E., 1964. *The Essential Oils. Individual Essential Oils of the Plant Families Rutaceae and Labiatae*, Vol. III, Van Nostrand Co. Inc. NY

Hakim I.A. and R.B. Harris, 2001. Joint effects of citrus peel use and black tea intake on the risk of squamous cell carcinoma of the skin. *BMC Dermatol*. 1: 3

Huet, R., 1991. Les huilles essentielles d' agrumes. *Fruits*, 46: 551–76

Kesterson, J. W., R. Henderickson and R. J. Braddock (1971). Florida Citrus Oils. *Florida Agric. Exp. Sta. Bull*. 749

Lockey, P., 2004a. *Live Life Naturally. Tangerine Essential Oil Certificate of Analysis*. http://www.cedarvale.net/essential oils/ tangerinecofa. htm. Accessed on 22/ 7/ 2004

Lockey, P., 2004b. *Live Life Naturally. Grapefruit Essential Oil Certificate of Analysis*. http://www.cedarvale.net/essential oils/ grape fruit whitecofa. htm. Accessed on 22/ 7/ 2004

Manthey, J. A. and K. Grohmann, 2001. Phenoles in citrus peel by-products concentrations of hydroxycinnamates and polymethoxylated flavones in citrus peel molasses. *J. Agric Food Chem.*, 49: 3268-73

Moshonas, M.G. and P.E. Shaw, 1971. Analysis of volatile flavour constituents from grape fruit essence. *J. Agric. Food Chem*., 19: 119–20

Moshonas, M.G. and P.E. Shaw, 1974. Quantitative and qualitative analysis of tangerine peel oil. *J. Agric. Food Chem*., 22: 282–4

Nagi, S., P.E. Shaw and M.K. Veldhuis, 1977. *Citrus Science and Technology*. Vol. I. Pp: 427–62. Avi Publishing Co., Inc. Westport, Connecticut

Ohloff, G., 1990. What makes an essential oil smell the way it does. The Aromatic plant project. *Scents Fragr*. 17: 124–9

Sattar, A. and S. Mahmud, 1992. Citrus Oil, Composition of Monoterpenes of the Peel Oil of Orange, Kinnow, and Lemon. *Pakistan J. Sci. Ind. Res*., 29: 196–8

Shaw, P.E. and R.L. Coleman, 1974. Quantitative composition of cold pressed orange oils. *J. Agric. Food Chem*., 22: 785–7

Shaw, P.E., R.L. Coleman and M.G. Moshonas, 1971. Quantitative analysis of Persian lime oil. *Proc. Fla. State Hortc. Soc*. 84: 187–9

Statfold Seed Oil Development Ltd., 2004. http://www.statfold-oils. co.uk Accessed on 22/7/2004.

Steel, R.G.D., J.H. Torrie and D. Dickey, 1997. *Principles and Procedures of Statistics, A Biometrical Approach*. 3rd Edi. McGraw Hill Book Co. New York

Weiss, E.A., 1997. *Essential Oil Crops*. Pp: 417–515. CAB Intl. N.Y. USA

Wolferd, R.W., J.W. Kesterson and J.A. Attaway, 1971. Physico-chemical properties of citrus essential oils from Florida. *J. Agric. Food Chem*., 19: 1097–105

(Received 20 October 2005; Accepted 12 December 2005)