EXTRACTION AND PURIFICATION OF ESSENTIAL OILS FROM LIME FRUIT *Citrus aurantifolia* (Christom) Swing), SWEET ORANGE (Citrus sinensis (L.) *Osbeck*) and LEMON GRASS (*Cymbopogon citratus* (DC) Stapf.)

***Muhammed, H.M. and Adebola, M.O.**

Department of Biological Sciences, Ibrahim Badamasi Babangida University, Lapai, Niger State, Nigeria.

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ABSTRACT

The extraction and purification of essential oils were carried out from two citrus species: *Citrus aurantifolia*, (lime fruits), *Citrus sinensis* (orange fruits), as well as *Cymbopogon citratus* (lemon grass), by solvent extraction method using soxhlet apparatus. The oils so extracted from the samples were purified by separating with separating funnels. They were then analyzed by thin layer chromatography and other methods of qualitative analysis. An average of 2.8 g of oil was extracted from the peels of 50 g lime fruits (*Citrus aurantifolia*) while 6.3 g of oil was extracted from the peels of 50 g sweet oranges (*Citrus sinensis*) and 2.7 g of oil was extracted from 28 g *Cymbopogon citratus* (lemon grass), respectively. The solvent extraction method yielded good quality essential oils from the three species.

Key words: Essential oils, solvent extraction, thin layer chromatography, purification.

INTRODUCTION

Compounds produced by living organisms are known as natural products. According to Mosher (1987), essential oils are products of plants. They are volatile, odoriferous oil products of plant origin which are distillable. Since they are products of plants, they can be classified as natural products. These oils are distinguished from fatty oils by their volatility, non-greasiness and non-saponifying properties (Theimer, 1992). In our everyday life, people consume oils in one form or the other through the use of products in which these oils are incorporated, but hardly does anybody seem to care about how these oils are made available, once the edible vegetable oils used in cooking are in continuous supply (Mosher, 1987).

Citrus oils are mixtures of very volatile components such as terpenes and oxygenated compounds (Sato *et al*., 1996). Limonene, a monoterpene, is the major component of lime and other related citrus essential oils (Lanças and Cavicchioli, 1990). These oils are used in the pharmaceutical, perfumery and food industries (Huet, 1991), and the quality of the oils is related to the value of total aldehydes, basically citral content, which is between 4-5% (Shaw, 1979). The common commercial methods to produce the oils from citrus fruits and peels are machine cold pressing and distillation. However, the oils obtained by distillation deteriorate easily and develop off-flavours due to the instability of the terpene hydrocarbons present, particularly *d* limonene (Yamauchi and Sato, 1990).

Oils of citrus species and lemon grass have been used as flavouring ingredients, manufacture of soaps, beverages, medicine, food and also promote secretion of digestive juices into the stomach and thus help in digestion of food. Generally, all the extracted oils give fragrance characteristic of the plant materials from which they were obtained (Mahran, 1993). The aim of this study was to provide information on how essential oils are extracted from *Citrus aurantifolia* (lime fruits), *Citrus sinensis* (orange fruits), and *Cymbopogon citratus* (lemon grass) plants and purified from various qualitative analyses.

* Author for correspondence.

MATERIALS AND METHODS

COLLECTION OF PLANT MATERIALS

Fresh fruits of *C. aurantifolia and C. sinensis* were purchased from a local market in Lapai, Niger State, Nigeria, while *Cymbopogon citratus* leaves were collected from the botanical garden of Ibrahim Badamasi Babangida University, Lapai. The identitification of the plant was confirmed by botanists in the Herbarium unit of the Department of Biological Sciences, Ibrahim Badamasi Babangida University, Lapai. The plants were properly cleaned with sterile distilled water and kept in the refrigerator for further use.

EXTRACTION OF ESSENTIAL OILS

Essential oils of the plant materials were extracted by solvent extraction method of Chopra and Kanwar (1991) and all operations were carried out at room temperature (28°C). Solvents used for the extraction were commercial n-hexane from El Hali Chemical Stores, Sokoto, Nigeria. Samples of 50 g of *C.sinensis*, *C.aurantifolia* peels and 28 g of *C. citratus* were used for the extraction using soxhlet apparatus. The oil was collected in a conical flask and stored at 10°C prior to analysis.

SEPERATION OF THE OIL CONSTITUENTS

Thin layer chromatography was used to separate the compounds present in the oil extract and to determine their purity. The components spots were identified by comparing the distance travelled by the component (DCP) over the distance travelled by the solvent front (DSF). The Retention factor (R_f) was calculated as DCP/DSF (Landgrebe, 2004).

QUALITATIVE ANALYSIS OF THE ESSENTIAL OILS

This was done using the method of Chopra and Kanwar (1991). The parameters examined were saponification value (MgKOH/g), peroxide value (mg/kg), iodine value (g/g), and pH value.

RESULTS

The oils extracted from the peels of *C. sinensis*, *C.aurantifolia* and leaves of *Cymbopogon citratus* were liquid at room temperature (28°C). The colour of the oils varied from yellow in *C.aurantifolia* to light green in *C. sinensis* and green in *Cymbopogon citratus*. The quantities of essential oil obtained were 2.8 g from *C. aurantifolia*, 6.3 g from *C.sinensis* and 2.7 g from *C. citratus* (Table 1).

THIN LAYER CHROMATOGRAHY

The thin layer chromatography (Table 2) revealed that the samples varied in their R_f values: *Cymbopogon citratus* (0.43), *C.sinensis* (0.82) and *C.aurantifolia* (0.36).

QUALITATIVE ANALYSIS

The pH values (Table 3) were 6.73, 7.42 in *C.sinensis*, *C.aurantifolia* and 7.11 in *Cymbopogon citratus*, respectively. Peroxide values (MgO_2/g) were 950 in *C.sinensis*, 200 in *C.aurantifolia* and 1,400 in *Cymbopogon citratus*. Iodine values (MgKOH/g) were 11.43 in *C.sinensis*, 10.16 in and 3.81 in *Cymbopogon citratus* (Table 3). The saponification values (MgKOH/g) were 43.24 in *C.sinensis*, 48.48 in *C.aurantifolia and* 47.05 in *Cymbopogon citratus* (Table 3).

Table1: Essential oils extracted from the *Citrus aurantifolia*, *Citrus sinensis* and *Cymbopogon citratus*

	C. aurantifolia	*C. sinensis*	*Cymbopogon citratus*
Quantity of samples used (g)	50	50	28
Quantity of essential oil obtained (g)	2.8	6.3	2.7
Colour of oil samples obtained	Yellow	Light green	Green

Table 2: Thin layer chromatography of the oil samples extracted.

Samples	DSF(cm)	DCP(cm)	R_f value
C. aurantifolia	14.2	5.2	0.36
C. sinensis	17.0	14.0	0.82
Cymbopogon citrates	15.0	6.5	0.43

Table 3: Qualitative analysis of essential oils of *Citrus sinensis*, *Citrus aurantifolia*, and C. citratus

Parameters analysed	*Citrus sinensis*	*Citrus aurantifolia*	*Cymbopogon citratus*
State at room temperature	Liquid	Liquid	Liquid
Colour	Light green	Yellow	Green
PH value	6.73	7.42	7.11
Peroxide value(MgO$_2$/g)	950	200	1400
Iodine value(MgKOH/g)	11.43	10.16	3.81
Saponification value(MgKOH/g)	43.24	48.48	47.05

DISCUSSION

Essential oils derived from plants have been recognised since antiquity to possess biological activities including antibacterial, antifungal and antioxidant properties. With growing interest in the use of essential oils in food, pharmaceutical and agricultural industries, a systemic examination of these extracts has become increasingly important (Ibrahim *et al.*, 2010). The high quality essential oils obtained in this study using solvent extraction method conformed with what Ferial *et al.* (2004) reported in Jojoba seed oil collected using a similar method.

From the result obtained from the thin layer chromatography, the samples were easily detected because they are naturally coloured (Dawodu *et al.*, 2009). The R_f values of 0.36, 0.43 and 0.82 obtained from this study conformed with Raven and Evert (1999), that pigments with lower R_f values adsorbed at the polar sites in a thin layer chromatography depends on the polarity of the mobile phase, and the more polar is the pigment, the faster and stronger it will adsorb down the plate .

At room temperature (28°C), the oils were liquid. The colours of the oils varied from yellow (*C.aurantifolia*), light green (*C.sinensis*) to dark green in *C. citratus*. This could be improved upon by bleaching (Dawodu *et al.*, 2009). The $_pH$ value is an indicator for edibility of oil and suitability for industrial use with *C.sinensis* (6.73), *C.aurantifolia* (7.42) and *C. citratus* (7.11). These fall within the recommended codex of 0.6 and 10 for virgin and non-virgin edible fats and oils, respectively (Adelaja, 2006). The results suggest that the oils are edible and can also be used in the manufacturing of paints and vanishes (Ibrahim *et al.*, 2010) as comparable to the values of edible oils such as palm oil, pumpkin seed oil, palm kernel oil which have been reported by Dosumu and Ochu (1996). *C.aurantifolia* and *C. citratus* were slightly akaline with pH of 7.42 and 7.11, respectively. *C.sinensis* was slightly acidic. Iodine value is a valid factor to monitor lipid oxidation (Naz *et al.*, 2004), the iodine value which is useful in predicting the drying properties of oils was found to be 11.43 for *C.sinensis*, 10.16 for *C.aurantifolia* and 3.81 for *Cymbopogon citratus,* which does not support the specified 220-270 mg/iodine/100 g for drying oils (Akinhanmi *et al.*, 2008). The iodine value is also an index for assessing the ability of oil to go rancid (Dawodu *et al.*, 2009). Peroxide value is one of the most widely used tests for oxidative rancidity in oils and fats. The high peroxide values of the oils characterized the majority of the conventional oils above 20 mg O_2/g which will result to rancid tastes (Adelaja, 2006). The saponification values that were obtained from the three samples have high values of 43.24, 47.24 and 48.48. Akanni *et al.* (2005) noted that the oils contain high molecular weight fatty acid and low level of impurities.

CONCLUSION

Considering the rich quality of these essential oils from the three samples, creating awareness on the importance of these oils to the urban populace will go a long way in supplementing them with other oils which are very expensive to buy.

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