VOLATILE CHEMICAL CONSTITUENTS OF PATCHOULI (POGOSTEMON CABLIN (BLANCO) BENTH.: LABIATAE) FROM THREE LOCALITIES IN PAPUA NEW GUINEA

Stewart W Wossa¹, Topul Rali² and David N Leach³

ABSTRACT

Fresh aerial parts of patchouli (Pogostemon cablin) were obtained from Rabaul, Port Moresby and Inawabui in the Mekeo area of the Central Province. The volatile oil constituents were extracted by exhaustive hydrodistillation where each of the patchouli samples afforded oil yields of about 0.1 percent. Detailed chemical investigations of the oils indicated patchouli alcohol to be the main constituent in the oils from the three localities. It was noted that the patchouli alcohol composition in the cultivar from Rabaul, Port Moresby and Mekeo were 71.8, 71.7 and 43.7 percent compositions respectively, suggesting that the patchouli oil from PNG can attract a ready market on the basis of its high patchouli alcohol contents in the patchouli oil as opposed to between 30 – 40 percent composition from other patchouli oil producing countries.

Keywords. Pogostemon cablin, Labiatae, Patchouli, Essential oil composition, Patchouli alcohol, Sesquiterpene.

INTRODUCTION

Patchouli oil is obtained from *Pogostemon cablin* (Blanco) Benth, a member of the plant family Labiatae. It is a predominantly tropical herbaceous and perennial plant species with wide distribution in most South East Asian nations where it has also been documented as having many significant uses in traditional medicine and agriculture (Guo 2001). The leaves and stem contain a yellowish and viscous oil that has a unique and intense camphoraceous odour with many useful applications, hence high market value in the perfumery industry.

On the basis of its traditional uses as alternative medicine, agricultural pest control agent and applications in the perfume industry, detailed phytochemical studies were pursued to identify the chemical constituents responsible for the perceived activities. In one such study, the acetone extracts of the leaves were found to contain sesquiterpene hydroperoxide, which showed significant trypanocidal activities (Kiuchi et al. 2004) Another study also identified the cytotoxic Licochalcone A. and 5.7-dihydroxy-3',4'dimethoxyflavanone as the main chemical compounds from the aerial parts (Park et al., 1998). The hexane extracts were also noted to contain patchouli alcohol, pogostol, stigmast-4-en-3-one,

retusin and pachypodol, which showed antiemetic activities (Yang et al. 1999).

The study of the chemical components within the essential oil extracts were found to be made up of patchouli alcohol, delta-guaiene, alpha-guaiene, seychellene, alpha-patchoulene, aciphyllene, transcaryophyllene (Feng et al. 1999; Zhao et al. 2005; Guan et al. 1994). Luo and co-workers (1999). investigated the patchouli oil from the Gaoyao County, China and noted the stem to contain high pogostone content while the leaves contained high patchouli alcohol contents. While the chemical components were noted to be the same from different regions studied, the compositions were found to vary between regions as influenced by various environmental factors (Singh et al. 2002) Yan et al. 2002). Furthermore, the oil composition from different cultivating locations and different harvesting times were obviously different (Luo et al. 2002), suggesting that detailed chemical study was required to establish the reasons for such differences. Such studies revealed that there were two main chemotypes in patchouli; one being the pogostone-type and the other being the patchoulioltype on the basis of the molecular evidences (Liu et al. 2002). The pogostone-type contained rich oxygenated components especially pogostone in the volatile oil while the patchouliol-type contain rich patchouliol, delta-guaiene, alpha-guaiene and other non-oxygenated components (Luo et al. 2003).

¹ Faculty of Science, University of Goroka, PO Box 1078, Goroka, Eastern Highlands Province, PNG.

² Chemistry Department, University of Papua New Guinea.

³ Centre for Phytochemistry and Pharmacology, Southern Cross University, Australia

Morphological studies of the leaves of patchouli also indicated the leaves to contain external trichomes as well as specialized internal accumulatory cells where concentrations of the sesquiterpenes were noted to be exceptionally higher than other parts of the plant (Henderson et al. 1970). Further biosynthetic studies revealed high concentrations of the enzyme sesquiterpene cyclase (patchould) synthase) in the leaves, which was postulated to be responsible for the conversion of the farnesyl pyrophosphate, through biosynthetic mechanisms, to the cyclic sesquiterpenes (alpha and betapatchoulene, alpha-bulnesene and alpha-guaiene) found in patchouli oil (Munck and Croteau, 1990; Croteau et al. 1987). Another study of the leaf extracts revealed the presence of patchoulan-1,12diol, which was also postulated to be the main precursor of nor-patchoulenol (Trifilieff 1980).

Interesting biological activities have been reported for the extracts of patchouli by various authors. The patchouli oil has been reported to show activity against three strains of methicillin-resistant Staphyloccocus aures (Edwards-Jones et al. 2004). cytotoxic (Park et al. 1998), bactericidal activity against Campylobacter jejuni and Listeria monocytogenes (Friedman et al. 2002), antibacterial and antifungal activities (Pattraik et al. 1996; Osawa et al. 1990), unusual tissue destructive effects on the exoskeletons of Formosan subterranean termites, Coptotermes formosanus Shiraki (Zhu et al. 2003), effective mosquito repellency effects (Trongtokit et al. 2005), showed promise as alternative preservative of perishable foods (Holley and Patel 2005), antiemetic activities (Yang et al. 1999) and trypanocidal activities (Kiuchi et al. 2004). Such interesting biological activities led to a number of synthetic studies to attempt their synthesis on a laboratory scale (Magee et al. 1995; Niwa et al. 1984. Niwa et al. 1987: Corv et al. 1990) including microbial assistance in the conversion of the substrates into the desired products (Arantes et al.

Currently, patchouli oil is one of the important natural essential oils because of its base and lasting aromatic character. The yellowish brown coloured viscous oil obtained from the leaves and stems have an intensely camphorous odour, a character that is utilized in the cosmetic industries to scent perfumes, flavour toothpastes and other health and self-care products. The global demand for patchouli oil has been noted to increase annually with Indonesia as the major producer of this oil (Robbins 1983, Tao 1983). The main buyers of the patchouli oil are the USA, Switzerland and France.

Preliminary studies on the chemical constituents in the PNG cultivar of patchouli (Wossa et al. 2004a) has shown the PNG oil to contain superior patchouli alcohol content at 70 percent as compared to the oil from Indonesia, Philippines, Malaysia, China and other South East Asian nations, which have patchouli alcohol compositions between 30 - 40 percent. In view of the economic potential in the cultivation and extraction of patchouli oil as an alternative agribusiness in PNG, we report herein the chemical constituents in the volatile oil extracts from patchouli from three localities and discuss these data in the light of the global market trends. This report is part of an ongoing study to document the chemical constituents in the essential oils obtained from the diversity of aromatic plants of PNG (Rali et al. 2003; Wossa et al. 2004a; 2004b; 2005).

MATERIALS AND METHODS

Samples of Patchouli were obtained from Rabaul, Port Moresby and the Mekeo area of the Central Province. The stem cuttings of the Rabaul cultivar was grown in a Port Moresby backyard garden to compare its oil yield and chemical composition with that of the samples from Rabaul. A native cultivar from the Mekeo area was also used in this study for comparative analysis of the chemical constituents.

The matured leaf samples of the patchouli were collected while fresh and the volatile oils obtained through exhaustive hydrodistillation. The distillates were extracted with diethyl ether and the ether removed under reduced pressure to afford yellowish brown-coloured pure oils. The oils were dried with anhydrous magnesium sulphate and stored at 4 degrees Celsius until further analysis. The analyses of the oils were done using a gas chromatograph coupled to a mass spectrometer (GC and GC/MS) and the individual components identified on the basis of their individual retention indices.

The analyses of the oil constituents were done as previously described (Wossa *et al.* 2005). The individual oil constituents were identified on the basis of their respective retention indices and confirmed by comparison with the mass spectral data of the authentic reference compounds or with the library of the published data (Adams 1995).

RESULTS AND DISCUSSION

The results of the GC and GC/MS analysis of the patchouli oil obtained from Rabaul, Port Moresby and Inawabui in Mekeo (Table 1) indicate that the Rabaul cultivar of patchouli has a higher patchouli

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Table 1. Volatile chemical constituents (% area) of Pogostemon cablin from three localities in PNG

Chemical Constituents	Rabaul	Port Moresby	Mekeo
alpha-pinene	-		1.2
alpha-guaiene	7.5	4.6	7.8
seychellene	3.9	3.2	6.2
gamma-patchoulene	3	i i	3.7
peta-patchoulene	140		1.8
alpha-patchoulene	1.7	1.5	1.4
delta-guaiene	9.9	6.5	9.5
pogostol	5.1	-	-
Patchouli alcohol	71.8	71.7	43.7
beta-caryophyllene	-	1.1	2.4
aciphyllene	:#::	1.2	
beta-patchoulene	<u> 26</u>	2.2	-
viridiflorol	+	1.4	-
Selina-3,7(11)-diene	-	1	5.1
Benzyl benzoate	42	-	1.7
C ₁₅ H ₂₂ O		6.6	0.7
C ₁₅ H ₂₄	-	9	1.9
C ₁₅ H ₂₆ O	-	-	0.9

 ⁼ not detected.

alcohol content than the Mekeo cultivar with compositions at 71.8 and 43.7 percent compositions respectively. Such differences can be expected due to the various environmental and genetic factors that may be involved in the genesis of the different chemical constituents in the two cultivars of patchouli. Similarly, the Port Moresby cultivar was found to be superior in yield and composition of the patchouli alcohol than the Mekeo cultivar with compositions at 71.7 percent and 43.7 percent respectively.

On the other hand, the chemical constituents and composition of the Port Moresby cultivar were noted to be similar to that of the Rabaul cultivar. This result was as expected because the cuttings were from the Rabaul cultivar and were cultivated in the backyard garden in Port Moresby to see the possible effects of environmental factors on oil yield and chemical composition. The similar oil yield and chemical constituents and composition led us to infer that the environmental factors had minimal effect on the yield and composition of the patchouli oil. These results further suggest that the Rabaul cultivar has a higher patchouli alcohol content compared to that reported from other South East

Asian cultivars. The comparison of the patchouli alcohol contents from the Mekeo cultivar with that of the other South East Asian cultivars can be categorized as being similar on the basis of the patchouli alcohol composition at around 40 percent, however they differ in the composition of the other constituents. This study, further shows that the three patchouli cultivars analyzed so far belong to the patchouliol-type with high patchouli alcohol contents and other non-oxygenated constituents.

On the basis of the chemical compositional data on patchouli oil presented, it can be realized that the patchouli alcohol content in the patchouli oil from Rabaul are higher compared to those reported from other patchouli oil producing countries in the region. This implies that patchouli oil from PNG can be readily accepted on the global patchouli oil market on the basis of the high patchouli alcohol contents. These results further suggest that patchouli can be encouraged and cultivated as a rural based agro industry in PNG. With the current global market for patchouli oil fluctuating between US\$ 12 – 27 per kilogram, PNG could do well in introducing patchouli oil production as an alternative income earner in the country.

It is therefore recommended that further studies into the effects of the soil nutrients and chemistry, irrigation systems, age of crop at harvest, other important environmental as well as genetic factors be pursued to identify the optimum conditions for maximum yield of the patchouli alcohols with higher patchouli alcohol content from the patchouli cultivars of PNG. Such studies will pave the way for commercial production of patchouli oil in PNG as an alternative revenue earner.

CONCLUSION

This study has given new insight into the commercial potential for the patchouli oil production in PNG in terms of its oil yield and chemical constituents. The Rabaul cultivar of patchouli was found to contain higher patchouli alcohol content compared to the Mekeo cultivar while the patchouli alcohol content in the Mekeo cultivar were noted to be within the marketable range. It was also noted that the cultivation of the cuttings from the Rabaul cultivar in Port Moresby gave similar yield and composition, suggesting that the environmental factors had minimal effect on the oil yield and chemical composition.

In view of the commercial potential in the cultivation and extraction of the patchouli oil as an alternative revenue earner in the country, further studies into the soil nutrients and chemistry, the irrigations systems age of plant at harvest, other environmental factors and genetic composition needed to be established to assist farmers in cultivating *Pogostemon cablin* for the extraction of its oil for the global patchouli oil markets

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REFERENCES

ADAMS, R.P. (1995) "Identification of Essential Oil Components by Gas Chromatography/Mass Spectrometry", Allured Pub. Corp., Carol Stream, IL.

- ARANTES, S.F., HANSON, J.R. AND HITCHCOCK, P.B. (1999). "The microbiological hydroxylation of the sesquiterpenoid patchoulol by *Mucor plumbens*" Phytochemistry, **52(4)**: 635 638.
- CORY, R.M., BAILEY, M.D. AND TSE, D.W.C. (1990). A divergent approach to patchouli sesquiterpenes: Synthesis of 3-oxopatchouli alcohol, 5-oxo-7-hydroxy-13-norcycloseychellene, 6-methoxy-4,12-dehydro-13-norcycloseychellene and patchouli alcohol". Tetrahedron Letters, 31(47): 6839 6842.
- CROTEAU, R., MUNCK, S.L., AKOH, C.C., FISK, H.J. AND SATTERWHITE, D.M. (1987). "Biosynthesis of the sesquiterpene patchoulol from farnesyl pyrophosphate in leaf extracts of *Pogostemon cablin* (patchouli): mechanistic considerations". Arch. Biochem. Biophys., 256(1): 56 68.
- EDWARDS-JONES, V., BUCK, R., SHAWCROSS, S.G., DAWSON, M.M. AND DUNN, K. (2004). "The effect of essential oils on methicillin-resistant *Staphyloccocus aures* using dressing model". Burns, 30(8): 772 777.
- FENG, Y., GUO, X. AND LUO, J. (1999). "GC-MS analysis of the volatile oil of *Herba Pogostemonis* collected from Leizhou County". Zhong Yao Cai, 22(5). 241 243. (Article in Chinese with English Abstract).
- FRIEDMAN, M., HENIKA, P.R. AND MANDRELL, R.E. (2002) "Bactericidal activities of plant essential oils and some of their isolated constituents against Campylobacter jejuni, Escherichia coli, Listeria monocytogenes, and Salmonella enterica" Journal of Food Protection, 65(10): 1545 1560
- GUAN, L.; QUAN, L.H.; XU, L.Z. AND CONG, P.Z. (1994). "Chemical constituents of *Pogostemon cablin* (Blanco) Benth." Zhongguo Zhong Yao Za Zhi, **19(6):** 355 356, 383. (Article in Chinese with English Abstract).
- GUO, J.-X. (2001). "International Collation of Traditional and Folk Medicine". Volume 4, World Scientific, Singapore. pp 99 100.
- HENDERSON, W., HART, J.W., HOW, P. AND JUDGE, J. (1970). "Chemical and morphological studies on sites of sesquiterpene accumulation in *Pogostemon cablin* (patchouli)". Phytochemistry, 9(6): 1219 1228.

- HOLLEY, R.A. AND PATEL, D. (2005). "Improvement in shelf-life and safety of perishable foods by plant essential oils and smoke antimicrobials". Food Microbiology, 22(4): 273 – 292.
- KIUCHI, F., MATSUO, K., ITO, M., QUI, T.K. AND HONDA, G. (2004). "New Sesquiterpene Hydroperoxides with Trypanocidal Activity from *Pogostemon cablin*". Chem. Pharm. Bull., **52(12)**: 1495 1496.
- LIU, Y.P., LUO, J.P., FENG, Y.F., GUO, X.L. AND CAO, H. (2002). "DNA profiling of *Pogostemon cablin* chemotypes differing in essential oil composition". Yao Xue Xue Bao, **37(4)**: 304 308. (Article in Chinese with English Abstract).
- LUO, J.P., LIU, Y.P., PENG, Y.F., GUO, X.L. AND CAO, H. (2003). "Two chemotypes of *Pogostemon cablin* and influence of region of cultivation and harvesting time on volatile oil composition". Yao Xue Xue Bao, 38(4): 307 310. (Article in Chinese with English Abstract).
- LUO, J., GUO, X. AND FENG, Y. (2002). "Constituents analysis on volatile oil of *Pogostemon cablin* from different collection time cultivated in Hainan". Zhong Yao Cai, 25(1): 21 23. (Article in Chinese with English Abstract).
- LUO, J., FENG, Y., GUO, X. AND LI, X. (1999). "GC-MS analysis of volatile oil of Herba Pogostemonis collected from Gaoyao County". Zhong Yao Cai. 22(1): 25 28 (Article in Chinese with English Abstract).
- MAGEE, T.V., STORK, G. AND FLUDZINSKI, P. (1995). "A total synthesis of rac-patchouli alcohol". Tetrahedron Letters, 36(42): 7607 7610.
- MUNCK, S.L. AND CROTEAU, R. (1990). "Purification and characterization of the sesquiterpene cyclase patchoulol synthase from *Pogostemon cablin*" Arch. Biochem. Biophys., 282(1): 58 64
- NIWA, H., HASEGAWA, T., BAN, N. AND YAMADA, K. (1984). "Stereocontrolled total synthesis of (±)-hydroxypatchouli alcohol and the corresponding (±)-carboxylic acid metabolites of patchouli alcohol and (±)-norpatchoulenol". Tetrahedron Letters, 25(26): 2797 2800.
- NIWA, H., HASEGAWA, T., BAN, N. AND YAMADA, K. (1987). "Stereocontrolled total synthesis of (±)-norpatchoulenol and two metabolites of patchouli

- alcohol. (\pm)-hydroxypatchouli alcohol and the corresponding (\pm)-carboxylic acid". Tetrahedron, **43(5):** 825 834.
- OSAWA, K., MATSUMOTO, T., MURAYAMA, T., TAKIGUCHI, T., OKUDA, K. AND TAKAZOE, I. (1990). "Studies on the antibacterial activity of plant extracts and their constituents against periodontopathic bacteria". Bull. Tokyo Dental College, 31(1): 17 21.
- PARK, E.J., PARK, H.R., LEE, J.S. AND KIM, J. (1998). "Licochalcone A: An inducer of cell differentiation and cytotoxic agent from *Pogostemon cablin*". Planta Medica, **64(5)**: 464 466.
- PATTRAIK, S., SUBRAMANYAM, V.R. AND KOLE, C. (1996). "Antibacterial and antifungal activities of the essential oils in vitro". Microbios., 86(349): 237 246.
- RALI, T., LEACH, D.N. AND WOSSA, S.W. (2003)

 "Preliminary Analysis of the Essential Oil
 Compositions in Some Aromatic Plants Species
 of Papua New Guinea", Abstracts of the 5th New
 Guinea Biological Conference, University of
 Goroka, Eastern Highlands Province, Papua
 New Guinea, September 2003.
- ROBBINS, S.R.J. (1983). "Natural essential oils Current Trends in production, marketing and demand" Perfumer and Flavorist, 8: 75 – 82
- SINGH, M., SHARMA, S. AND RAMESH, S. (2002). "Herbage, oil yield and oil quality of patchouli [Pogostemon cablin (Blanco) Benth.] influenced by irrigation, organic mulch and nitrogen application in semi-arid tropical climate". Industrial Crops and Products, 16(2): 101 107.
- TAO, C. (1983) "China's burgeoning aromatic industry". Perfumer and Flavorist, 7: 1
- TRIFILIEFF, E. (1980) "Isolation and the postulated precursor of nor-patchoulened in patchouli leaves" Phytochemistry, 19(11): 2467
- TRONGTOKIT, Y., RONGSRIVAM, Y., KOMALAMISRA, N. AND APIWATHNASORN, C. (2005). "Comparative repellency of 38 essential oils against mosquito bites". Phytotherapy Research, 19(4): 303 309.
- WOSSA, S.W., RALI, T. AND LEACH, D.N. (2004a) "Analysis of the Essential Oil Compositions of Some Selected Spices of Papua New Guinea". PNG J. Agric. Forest. Fish., 47(1-2): 17-20.

- WOSSA, S.W., RALI, T. AND LEACH, D.N. (2004b)
 "The Chemistry of the Aromatic Plant Diversity
 of Papua New Guinea: The Family Rutaceae".
 Abstracts and Proceedings of the 6" New Guinea
 Biological Conference, State University of Papua,
 Manokwari, Indonesia, August 2004.
- WOSSA, S.W., RALI, T. AND LEACH, D.N. (2005).

 "Analysis of the Volatile chemical constituents of Tumeric (*Curcuma longa* Linn: Zingiberaceae)". PNG J. Agric. Forest. Fish., 48(1-2): 21 24.
- YAN, Z., QIU, J., CAI, Y. AND LIAO, G. (2002).
 "Study on nutritional characteristics of Pogostemon cablin". Zhong Yao Cai, 25(4): 227 230. (Article in Chinese with English Abstract).
- YANG, Y., KINOSHITA, K., KOYAMA, K., TAKAHASHI, K., TAI, T., NUNOURA, Y. AND WATANABE, K. (1999). "Antiemetic principles of *Pogostemon cablin* (Blanco) Benth.". Phytomedicine, 6(2): 89 93.
- ZHAO, Z., LU, J., LEUNG, K., CHAN, C.L. AND JIANG, Z.H. (2005). "Determination of patchoulic alcohol in *Herba Pogostemonis* by GC-MS-MS". Chemical and Pharmaceutical Bulletin (Tokyo), 53(7): 856 860.
- ZHU, B.C., HENDERSON, G., YU, Y. AND LAINE, R.A. (2003). "Toxicity and repellency of patchouli oil and patchouli alcohol against subterranean termites Coptotermes formosanus Shiraki (Isoptera: Rhinotermitidae)". J. Agriculture and Food Chemistry, 51(16): 4585 4588.