

CHEMICAL COMPOSITION OF THE ESSENTIAL OIL OF *AMOMUM ULIGINOSUM*J. Mailina^{1,*,**}, M. A. Nor Azah¹, Y. Y. Sam¹, S. L. L. Chua¹ & J. Ibrahim²¹Forest Research Institute Malaysia, 52109 Kepong, Selangor Darul Ehsan, Malaysia²Faculty of Allied Health Sciences, Universiti Kebangsaan Malaysia, Jalan Raja Muda Abdul Aziz, 50300 Kuala Lumpur, Malaysia

The genus *Amomum* (Zingiberaceae) contains about 150 to 180 species. It is widespread in South-East Asia with 18 species reported in Peninsular Malaysia (Holttum 1950, Xia *et al.* 2004). In Malaysia, the fruits and seeds of *A. cardamomum* (pelaga) are widely used by many communities in traditional herbal medicines and as spices and flavourings (Burkill 1966). Antioxidant, anti-malarial, anti-diabetes, anti-microbial and anti-bacterial activities have been reported in *Amomum* spp. (Holdsworth & Mahana 1983, Jitoe *et al.* 1992, Kamchonwongpaisan *et al.* 1995, Heilmann *et al.* 2001, Kwon *et al.* 2003, Mathew *et al.* 2003). The chemical composition of the essential oils and extracts of *A. subulatum*, *A. cannicarpum*, *A. testaceum*, *A. linguiforme* and *A. koenigii* have been examined (Ajit & Subhan 1995, Dong *et al.* 1999, Sirat *et al.* 2001, Mathew *et al.* 2003, Pura Naik *et al.* 2004, George *et al.* 2006). Major components of *A. cannicarpum* were found to be β -terpineol (13.4%), β -pinene (9.4%) and α -pinene (6.9%) (George *et al.* 2006). Meanwhile, the major component reported from the pericarp (husk) oil of *A. subulatum* (large cardamom) was 1,8-cineole (38.7%) (Pura Naik *et al.* 2004). *Amomum linguiforme* oil was a source of methyl chavicol (93.2%) (Ajit & Subhan 1995). β -Pinene (15.8%), 1,8-cineole (12.7%), fenchone (14.9%) and *trans*-pinocarveol (10.9%) were some of the major constituents reported from the fruits of *A. testaceum* (Sirat *et al.* 2001). Besides essential oils, eicosenones and methylated flavanols have been reported from other *Amomum* species (Dong *et al.* 1999).

Amomum uliginosum belongs to the tall ginger group. The stems are tall and slender like the common langkas. It can be found in Thailand, Peninsular Malaysia and Borneo. It is such a common ginger that with a little searching, one can easily find the plants growing at forest edging the road, stream or riverbanks. The first thing one will notice is the distant leafy stems that emerge from long running rhizomes found just below the ground surface. These stems can attain to the height of 2 to 3 m, with about 16–20 pairs of leaves on the upper half of the stem. Leaves are large but narrow, about 50 cm long and 5–11 cm wide. The characteristics of the leaves are the short petioles and long narrow tips in a drooping position (Holttum 1950).

Like the leafy stems, the inflorescences of *A. uliginosum* also emerge from the running rhizome, usually a short distance from the stem. However, the inflorescence stalk is short and buried in the soil and only the inflorescence head can be seen above the ground. Flowers are arranged close together, opening about 2–4 each time. The most distinct part of the flower is the concave white lip, which has a bright yellow band at the centre and two red lines beside it. The fruits are easy to distinguish; they are rounded, hairy and red in colour, like little rambutans. The rhizome has essential oil with ginger aroma. This impels us to study the chemical constituents that contribute to the odour. The chemical constituents of this species have not been reported.

The rhizomes of *A. uliginosum* were collected from the Tapah–Cameron Highlands Road, Pahang. A voucher specimen (FRI47241) was deposited at the herbarium of the Forest Research Institute Malaysia. The fresh rhizomes (870 g) were cut into small pieces and hydrodistilled in a Clavenger apparatus for six hours. The oily layer obtained was dried over anhydrous sodium sulphate. The yield was calculated based on dry weight basis of the plant materials. The rhizome oil yielded 0.03% of slightly pale yellow oil with a characteristic ginger odour. Analysis of the rhizome oil by gas chromatography (GC) was conducted using model Shimadzu GC-2010 capillary chromatograph which was equipped with a flame ionization detector (FID) using the split mode injection technique. The following conditions were employed: helium as carrier gas; similar temperature for injector and detector at 250 °C. The chromatograph used a non-polar capillary column EQUITY-1 (30 m \times 0.25 mm, film thickness 0.25 μ m). Operating conditions for the columns: initial oven temperature, 60 °C for 10 min, 60 °C to 230 °C at 3 °C min⁻¹ for 10 min. Gas chromatography/mass spectrometry (GC/MS) analysis was conducted on Hewlett-Packard GC/MSD 5890 Series II 5971A (70eV direct inlet) with similar conditions as described in GC programs using DB-1 (30 m \times 0.25 mm, film thickness 0.25 μ m).

The constituents were identified by matching their mass spectra with database in the Wiley Library software, co-chromatography of some chemical constituents with authentic samples and comparison of

their retention indices with literature values (Jennings & Shibamoto 1980, Menut *et al.* 1995, Ibrahim *et al.* 1996, Bastide *et al.* 1997, Dung *et al.* 1997, Bordoloi *et al.* 1999, Foray *et al.* 1999, Menon *et al.* 2001, Ibrahim *et al.* 2002). From the chromatographic analysis, about 47 compounds (excluding unidentified compounds) were identified and they accounted for 81.9% of the oil. The chemical constituents of the rhizome oil are listed in Table 1.

β -Pinene and α -pinene were the major components of the rhizome oil contributing to 29.9% and 10.4% of the total oil respectively. The other major monoterpenoids (C_{10}) were α -terpineol (7.6%), isopinocampone (5.1%), 1,8-cineole (2.7%) and p-cymene (2.2%). Other compounds that were present in appreciable amount were limonene (1.3%) and camphene (1.0%). The sesquiterpenoids (C_{15}) identified were spathulenol (1.8%), β -caryophyllene (1.6%) and (*E*)- β -farnesene (1.4%). Meanwhile, the remaining constituents (monoterpenoids and sesquiterpenoids) identified were present at less than 1%. Methyl thymyl ether contributed to 0.9% of the total oil. α -Fenchyl alcohol, α -fenchyl acetate, β -chamigrene and globulol gave the same concentration (0.7%). The other appreciable compounds were γ -terpinene and terpinolene (0.6%), myrcene, pinocarvylacetate, alloaromadendrene and epiglobulol (0.5%), camphor, α -humulene and viridiflorol (0.4%). From the mass fragmentation analysis the unidentified components were assigned the formulae $C_{12}H_{20}O_2$ and $C_{15}H_{24}$ which indicate that they are monoterpenoids and sesquiterpenoids. Based on this and previous studies, the result showed that most of the *Amomum* species are promising source of monoterpenoid components. These findings may be useful in providing information on identification of plant species in taxonomy studies.

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Table 1 Percentage composition of *Amomum uliginosum* oil

No.	Compound	RI	Concentration (%)	Methods of identification
1	Tricyclene	897	0.1	B
2	α -Thujene	929	0.1	A,B
3	α -Pinene	938	10.4	A,B,C
4	Camphene	949	1.0	A,B,C
5	Sabinene	953	0.1	A,B
6	β -Pinene	979	29.9	A,B,C
7	Myrcene	988	0.5	A,B,C
8	α -Phellandrene	1012	0.2	A,B,C
9	p-Cymene	1016	2.2	A,B,C
10	1,8-Cineole	1027	2.7	A,B,C
11	Limonene	1030	1.3	A,B,C
12	(<i>E</i>)- β -ocimene	1040	0.3	A,B,C
13	γ -Terpinene	1055	0.6	A,B,C
14	Fenchone	1076	0.3	A,B
15	Terpinolene	1082	0.6	A,B
16	Linalool	1092	0.3	A,B,C
17	α -Fenchyl alcohol	1102	0.7	A,B,C
18	Camphor	1127	0.4	A,B,C
19	Isopinocampone	1159	5.1	B
20	Terpinen-4-ol	1172	4.8	A,B,C
21	α -Terpineol	1185	7.6	A,B,C
22	Myrtenol	1188	0.3	A,B,C
23	α -Fenchyl acetate	1195	0.7	A,B
24	Methyl thymyl ether	1213	0.9	B
25	Endobornyl acetate	1276	0.3	A,B,C
26	Pinocarvylacetate	1286	0.5	B
27	Myrtenyl acetate	1311	0.2	A,B
28	α -Copaene	1375	0.1	A,B
29	Cyperene	1388	0.1	B
30	α -Gurjunene	1396	0.2	A,B
31	β -Caryophyllene	1416	1.6	A,B
32	Zingiberene	1434	0.1	A,B
33	β -Santalene	1436	0.1	A,B
34	α -Humulene	1450	0.4	A,B,C
35	Alloaromadendrene	1457	0.5	A,B
36	γ -Selinene	1471	0.2	A,B
37	β -Maaliene	1477	0.1	A,B
38	β -Chamigrene	1480	0.7	B
39	Valencene	1486	0.2	B
40	Eremophilene	1490	0.3	B
41	β -Bisabolene	1503	0.3	A,B
42	δ -Cadinene	1526	0.1	A,B,C
43	(<i>E</i>)- β -farnesene	1570	1.4	A,B
44	Spathulenol	1576	1.8	A,B
45	Globulol	1579	0.7	A,B
46	Epiglobulol	1586	0.5	B
47	Viridiflorol	1588	0.4	A,B
48	Unidentified compounds	-	3.8	B
			Total:	85.7

Percentages were obtained by peak area normalization on non-polar column EQUITY-1, A = retention index, B = mass fragmentation, C = co-injection with authentic sample.

REFERENCES

- AJIT, K. H. & SUBHAN, C. N. 1995. Methyl chavicol—the major component of the rhizome oil of *Amomum linguiforme* Benth. *Journal of Essential Oil Research* 7: 325–326.
- BURKILL, I. H. 1966. *A Dictionary of the Economic Products of the Malay Peninsula*. Vol. 1 (A–H). Ministry of Agriculture and Cooperatives, Kuala Lumpur.
- BASTIDE, J. M., MILHAU, G., VALENTINE, A., BENOIT, F. & MALLIE, M. 1997. *In vitro* antimalarial activity of eight essential oils. *Journal of Essential Oil Research* 9: 329–333.
- BORDOLOI, A. K., SPERKOVA, J. & LECLERCQ, P. A. 1999. Essential oils of *Curcuma aromatica* Salibs. from Northeast India. *Journal of Essential Oil Research* 11: 537–540.
- DONG, H., GOU, Y. L., CAO, S. G., CHEN, S. X., SIM, K. Y., GOH, S. H. & KINI, R. M. 1999. Eicosenones and methylated flavonols from *Amomum koenigi*. *Phytochemistry* 50: 899–902.
- DUNG, N. X., TRUONG, P. X., KY, P. T. & LECLERCQ, P. A. 1997. Volatile constituents of the leaf, stem, rhizome, root and flower oils of *Curcuma harmandii* Gagnep. from Vietnam. *Journal of Essential Oil Research* 9: 677–681.
- FORAY, L., BERTRAND, C., PINGUET, F., SOULIER, M. & ASTRE, C. 1999. *In vitro* cytotoxic activity of three essential oils from *Salvia* species. *Journal of Essential Oil Research* 11: 522–526.
- GEORGE, V., MATHEW, J., SABULAL, B., DAN, M. & SHIBURAJ, S. 2006. Chemical composition and antimicrobial activity of essential oil from the rhizomes of *Amomum cannicarpum*. *Fitoterapia* 77: 392–394.
- HEILMANN, J., BRUN, R., MAYR, S., RALI, T. & STICHER, O. 2001. Minor cytotoxic and antibacterial compounds from the rhizomes of *Amomum aculeatum*. *Phytochemistry* 57: 1281–1285.
- HOLDSWORTH, D. K. & MAHANA, P. 1983. Traditional medicinal plants of Huon Peninsula, Morobe province, Papua New Guinea. *International Journal of Crude Drug Research* 21: 121–133.
- HOLTUM, R. E. 1950. The Zingiberaceae of the Malay Peninsula. *Garden's Bulletin Singapore* 13(1): 1–249.
- IBRAHIM, J., ABU SAID, A., ABDUL RASHIH, A., NOR AZAH, M. A. & NORSIHA, A. 1996. Chemical composition of some *Citrus* oils from Malaysia. *Journal of Essential Oil Research* 8: 627–632.
- IBRAHIM, J., NORSIHA, A., HIONG, A. B. & ABU SAID, A. 2002. Chemical composition of the essential oils of *Cinnamomum cordatum* Kosterm. *Flavour and Fragrance Journal* 17(3): 212–214.
- JENNINGS, W. & SHIBAMOTO, T. 1980. *Qualitative Analysis of Flavor and Fragrance Volatiles by Glass Capillary Gas Chromatography*. Academic Press, New York.
- JITOE, A., MASUDA, T., TENGAH, I. G. P., SUPRAPTA, D. N., GARA, I. W. & NAKATANI, N. 1992. Antioxidant activity of tropical ginger extracts and analysis of the contained curcuminoids. *Journal of Agricultural and Food Chemistry* 40: 1337–1340.
- KAMCHONWONGPAISAN, S., NILANONTA, C., TARNCHOMPOO, B., THEB-TARANONTH, C., THEBTARANONTH, Y., YUTHAVONG, Y., KONGSAEREE, P. & CLARDY, J. 1995. An antimalarial peroxide from *Amomum krervanh* Pierre. *Tetrahedron Letter* 36(11): 1821–1824.
- KWON, K. B., KIM, J. H., LEE, Y. R., LEE, H. Y., JEONG, Y. J., RHO, H. W., RYU, D. G., PARK, J. W. & PARK, B. H. 2003. *Amomum xanthoides* extract prevents cytokine-induced cell death of RINm5F cells through the inhibition of nitric oxide formation. *Life Sciences* 73: 181–191.
- MATHEW, J., SHIBURAJ, S. & GORGE, V. 2003. Antimicrobial activity of *Amomum cannicarpum*. *Fitoterapia* 74: 476–478.
- MENUT, C., LAMATY, G., BESSIERE, J. M., SEULEIMAN, A. M., FENDERO, P., MAIDOU, E. & DENAMGANAI, J. 1995. Aromatic plants of tropical central Africa. XXII. Volatile constituents of *Croton aubrevillei* J. Leonard and *C. zambesicus* Muell. Arg. *Journal of Essential Oil Research* 7: 419–422.
- MENON, N. A., CHACKO, S. & NARAYANAN, C. S. 2001. Free and glycosidically bound volatiles of Pepper (*Piper nigrum* L.). *Journal of Essential Oil Research* 13: 166–169.
- PURA NAIK, J., JAGAN MOHAN RAO, L., MOHAN KUMAR, T. M. & SAMPATHU, S. R. 2004. Chemical composition of the volatile oil from the pericarp (husk) of large cardamom (*Amomum subulatum* Roxb.). *Flavour and Fragrance Journal* 19(5): 441–444.
- SIRAT, H. M., HONG, L. F. & KHAW, S. H. 2001. Chemical compositions of the essential oil of the fruits of *Amomum testaceum* Ridl. *Journal of Essential Oil Research* 13: 86–87.
- XIA, Y. M., JOHN KRESS, W. & PRINCE, L. M. 2004. Phylogenetic analyses of *Amomum* (Alpinioideae: Zingiberaceae) using ITS and matK DNA sequence data. *Systematic Botany* 29(2): 334–344.