

## Essential oil composition from the flowers of *Alstonia scholaris* of Bangladesh

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### Abstract

The essential oil isolated from the flowers of *Alstonia scholaris* and growing all over the country of Bangladesh, was analyzed by gas chromatography-mass spectrometry (GC-MS). Sixty compounds were identified in the flower oil. The essential oil compositions of flower oil 2-Dodecyloxirane (31.83%), Benzene, 1,2-dimethoxy-4-(2-propenyl)- (8.49%), Spinacene (6.09%), 1,54-Dibromotetrapentacotane (5.13%), 2,6,10,15-Tetramethylheptadecane (4.91%), Terpinyl acetate (3.74%), Linalool (2.22%), Tritetracontane (2.17%), 1-Cyclohexanol, 2-(3-methyl-1,3-butadienyl)-1,3,3-trimethyl- (1.78%), 9-Methyl-5-methylene-8-decen-2-one (1.58%) as the main constituents.

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### Introduction

*Alstonia scholaris* (Apocyanaceae) which is commonly known as Chhatim in Bangali and Chatium in Hindi. This Plant is growing to play a vital role in maintaining human health and improving the quality of human life from thousands of years and serves to human the valuable components of medicines, seasonings, beverages, cosmetics and dyes. Herbal medicine contains natural substances that can promote health and reduce illness. The plant research has increased all over the world and a large body of evidence has been collected to show immense potential of medicinal plants used in various traditional systems from long before or Last few decades. Furthermore many western drugs had their origin in plant extract. There are many herbs, which are used to treat cardiovascular problems, liver disorders, central nervous system, digestive and metabolic disorders. They give their potential to produce significant therapeutic effect and can be used as drug or supplement in the treatment, management of various diseases. Herbal drugs or medicinal plants, their extracts and their isolated compounds have exhibits spectrum of biological activities.

The plant grows throughout the humid regions of India, especially in West Bengal, Bangladesh and west-coast forests of south India (Wiar, 2006). The plant grows throughout the humid regions of India, especially in West Bengal and west-coast forests of south India. The plant is used in Ayurvedic, Unani and Sidhha/Tamil types of alternative medicinal systems (Khare, 2007). *Alstonia scholaris* is known

to be a rich source of alkaloids and there is interest among the scientist to use this for therapeutic purposes. Amongst the chemical classes present in medicinal plant species, alkaloids stand as a class of major importance in the development of newer drugs because alkaloids possess a great variety of chemical structures and have been identified as responsible for pharmacological properties of medicinal plants. However, of the large variety of the alkaloids (about 180 alkaloids) isolated, so far only few have been assessed for biological activities (Versha *et al.*, 2003). Almost all the parts of plant (bark, flower, root) are found to contain active principles. The species *Alstonia scholaris* is used in commercial formulation Ayush (Sastri, 1962). The bark of this plant contains alkaloid ditamine and echitamine, echitenine, echicaoutchin, an amorphous yellow mass, echicerin in acicular crystals, echitin in crystallized scales, echitein in rhombic prisms (a crystallisable acid) and echiretin an amorphous substance, resembling an alkaloid, a fatty acid and fatty resinous substances. An uncrystallisable bitter principle called ditain was isolated and ascribed the febrifuge properties of the drug (Nadkarni, 1976).

Traditional bark is bitter, astringent, acrid, thermogenic, digestive, laxative, anthelmintic, febrifuge, antipyretic, depurative, galactogogue, stomachic, cardiogenic and tonic. It is useful in fever, malarial fever, abdominal disorders, diarrhoea, dysentery, dyspepsia, leprosy, skin diseases, pruritus, tumours, chronic and foul ulcers, asthma, bronchitis, cardiopathy, elminthiasis, agalactia and debility. The milky exudate is bitter and is good for ulcers, vitiated

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conditions of vata and otalgia (Kirtikar and Basu, 1980; Nadkarni, 1976). The preparation of infusion i.e. 1 to 2 ounces of tincture, 1 to 2 drachms diluted in water and of ditanin 5 to 10 grams given two or three times a day and an extract is prepared from the fresh bark and given in milk in cases of leprosy. It is also used as an anthelmintic (Nadkarni, 1976). Milky juice is applied to ulcers and to rheumatic pains; mixed with oil and dropped into ear it relieves earache. The bark acts in certain cases as a powerful galactagogue. Juice of the leaves with that of fresh ginger-root or zedoary is administered to women after confinement. The zedoary drug is also used in cases of snake-bite (Nadkarni, 1976). The active constituents of the plant include antimalarials, CNS depressants, anticancers, antituberculosis, antidysentrics and galactopoeitics (Sastri, 1962; Nadkarni, 1976; Kirtikar and Basu, 1980).

The Plant of pacifies vitiated vata, pitta, skin diseases, fever, malaria, diarrhea, dysentery, indigestion, intestinal worms, tumors, malignancy, ulcers, wounds cough, bronchitis, insanity, epilepsy and general debility. The antimicrobial property of the plant constituents of *Alstonia scholaris* are alkanes, alkanols and sterols (Goyal et al., 1995). Khan et al. (2003) evaluated the antibacterial activity of the petrol, dichloromethane, ethyl acetate, butanol fractions of crude methanolic extracts of the leaves, stem and root barks of *Alstonia scholaris* and reported that butanol fraction exhibited broader spectrum of antibacterial activity. Methanol extracts of root barks of *Alstonia macrophylla*, *Alstonia glaucescens*, and *Alstonia scholaris*, collected from Thailand, have been assessed for cytotoxic activity against two human lung cancer cell lines, MOR-P (adenocarcinoma) and COR-L23 (large cell carcinoma), using the SRB assay. Pleiocarpamine, O-methylmacralstonine and macralstonine were all considerably less active than villalstonine (Keawpradub et al., 1997).

The plant is used in Ayurvedic, Unani and Sidhha/Tamil types of alternative medicinal systems. Leaves and bark are rich in Echitamine, Echitamine chloride, Scholarine, Scholaricine, monoterpenoid indole alkaloids, iridoids, coumarins, flavonoids, simple phenolics, steroids, saponins and tannins were also found in the plant (Dey, 2011). The plant is traditionally being used in fever (Rajakumar et al., 2010), cancer, tumour, jaundice, hepatitis, malaria and skin diseases (Mollik et al., 2010).

The chemopreventive effect of various doses of hydroalcoholic extract of *Alstonia scholaris* on the benzo(a)pyrene (BaP) induced fore stomach carcinoma in female mice. The pre or post-treatment of mice with 4 mg/ml ASE also significantly reduced

the frequency of BaP-induced MN in the splenocytes of treated animals (Jagetia et al., 2003). Their study demonstrated that the extract prepared from the summer collection and the fractions containing the alkaloids were highly effective in cell killing.

## Materials and Methods

### Plant material

The flowers of *Alstonia scholaris* were collected from the plants grown in the campus of Bangladesh Council of Scientific and Industrial Research (BCSIR) Laboratory, Chittagong during March 2013. The voucher specimen (Y-112) was deposited in the herbarium of BCSIR Laboratory, Chittagong.

### Extraction of essential oil

Flowers were harvested from healthy, well-grown plants. Freshly harvested flowers (300 g) were grounded in a blender separately. The grounded flowers were subjected to hydro-distillation using Clevenger apparatus for 4 h for isolation of oils separately. The oil samples were stored at 0°C in air-tight containers after drying them over anhydrous sodium sulfate and filtered before going to GC-MS analysis.

### GC-MS analysis

The essential oils from flower of *Alstonia scholaris* were analyzed by GC-MS electron impact ionization (EI) method on GC-17A gas chromatograph (Shimadzu) coupled to a GC-MS QP 5050A Mass Spectrometer (Shimadzu); fused silica capillary column (30 m x 0.25 mm; 0.25 mm film thickness), coated with DB-5 ms (JandW); column temperature 100°C (2 min) to 250°C at the rate of 3°C/min; carrier gas, helium at constant pressure of 90 Kpa. Acquisition parameters full scan; scan range 40 - 350 amu. Samples were injected by splitting and the split ratio 1:20.

### Identification of the compounds

Compound identification was done by comparing the National Institute of Standards and Technology (NIST) library data of the peaks with those reported in literature, mass spectra of the peaks with literature data. Percentage composition was computed from GC peak areas on DB-5 column without applying correction factors.

## Results and Discussion

The chemical compositions of volatile oil of flower of *Alstonia scholaris* are shown in Table 1. The oil yield varied from 0.10% to 0.02% for the

Table 1. Chemical constituents of the flower of *Alstonia scholaris*

S/N	Compound name	Percentage
1	2-Dodecyloxirane	31.83
2	Benzene, 1,2-dimethoxy-4-(2-propenyl)-	8.49
3	Spinacene	6.09
4	1,54-Dibromotetrapentacotane	5.13
5	2,6,10,15-Tetramethylheptadecane	4.91
6	Terpinyl acetate	3.74
7	Linalool	2.22
8	Tritetracontane	2.17
9	1-Cyclohexanol, 2-(3-methyl-1,3-butadienyl)-1,3,3-trimethyl-	1.78
10	9-Methyl-5-methylene-8-decen-2-one	1.58
11	7-Hexylicosane	1.39
12	Methyltridecanoate	1.23
13	Palmitic acid	1.01
14	Oxirane, 2,2-dimethyl-3-(3,7,12,16,20-pentamethyl-3,7,11,15,19-heneicosapentaenyl)-, (all-E)-	0.88
15	[(4-Chloro-[1,2,3]dithiazol-5-ylidene)(nitro)methyl](2,6-dimethylphenyl)diazene	0.86
16	2,6-Dimethyl-2,6-undecadien-10-ol	0.82
17	Rhamnitol, 1-O-octyl-	0.74
18	Eugenol	0.68
19	2,6,10,15-Tetramethylheptadecane	0.68
20	1,3-Dioxolane, 2-(5-bromopentyl)-	0.65
21	4-Terpineol	0.62
22	E-2-Tetra decen-1-ol	0.61
23	10-Heneicosene (c,t)	0.61
24	2-Methylenecholestan-3-ol	0.51
25	2,4-Bis(dimethylbenzyl)phenol	0.50
26	2,3-Bis(acetyloxy)propyl laurate	0.48
27	Cyclopentadecanone, oxime	0.44
28	cis-Myrtanol	0.41
29	4-Terpineol acetate	0.31
30	Alpha, alpha, 4-trimethylbenzyl carbanilate	0.21
31	Nerolidyl acetate	0.25
32	2-(5-Bromopentyl)-1,3-dioxolane	0.36
33	Dihydroartemisinin, 10-O-(t-butyloxy)-	0.26
34	Malonic acid, bis(2-trimethylsilylethyl ester	0.32
35	2,3-Dipentyl-2-cyclopropene-1-carboxylic acid	0.34
36	4-(1,3,2-Dioxaborinan-2-yl)-2-nitrobenzoic acid	0.38
37	Dithioerythritol, O,O',S,S'-tetra kis(trimethylsilyl)-	0.29
38	Benzeneacetic acid, 3-[(trimethylsilyloxy)-, trimethylsilyl ester	0.33
39	Methyl 10-methylundecanoate	0.29
40	6,11-Dimethyl-2,6,10-dodecatrien-1-ol	0.15
41	Dithioerythritol, O,O',S,S'-tetra kis(trimethylsilyl)-	0.19
42	2-Methylenecholestan-3-ol	0.30
43	N,N'-Diaacetylenethylenediamine	0.15
44	1-Heptatria cotanol	0.24
45	2,3-Bis(acetyloxy)propyl laurate	0.24
46	2,3-Bis(acetyloxy)propyl laurate	0.33
47	3,7,11,15-Tetramethylhexadecyl acetate	0.23
48	1-Ethyldecylacrylate	0.15
49	6,11-Dimethyl-2,6,10-dodecatrien-1-ol	0.15
50	2,6-Bis(t-butyl)-4-(dimethylbenzyl)phenol	0.14
51	9,9-Dimethoxybicyclo[3.3.1]nona-2,4-dione	0.34
52	t-Butoxyformamid, N-methyl-N-[4-(1-pyrrolidinyl)-2-butylnyl]-	0.37
53	1,6,10,14-Hexa decatetraen-3-ol, 3,7,11,15-tetramethyl-, (E,E)-	0.26
54	3,7,11,15-Tetramethyl-2-hexadecen-1-ol	0.26
55	Methyl(3-hydroxy-14-methyl-7-oxopodocapan-13-yl)acetate	0.25
56	Ethyl 6-(diethoxyphosphoryl)hexanoate	0.12
57	Capric ether	0.16
58	Imidazole-2-carboxylic acid	0.16
59	3,6-Dimethyl-2,4-heptadione	0.17
60	3-Buten-2-amine, 4-(2,6,6-trimethyl-1-cyclohexen-1-yl)-	0.13
61	Malonic acid, bis(2-trimethylsilylethyl ester	0.14

flower on a fresh weight basis (W/W). There were similarities on the library of such major compounds such as 2-Dodecyloxirane (31.83%), Benzene, 1,2-dimethoxy-4-(2-propenyl)- (8.49%), Spinacene (6.09%), 1,54-Dibromotetrapentacotane (5.13%), 2,6,10,15-Tetramethylheptadecane (4.91%), Terpinyl acetate (3.74%), Linalool (2.22%), Tritetracontane (2.17%), 1-Cyclohexanol, 2-(3-methyl-1,3-butadienyl)-1,3,3-trimethyl- (1.78%), 9-Methyl-5-methylene-8-decen-2-one (1.58%).

The bark is acrid, bitter, appetizer, laxative, anthelmintic, a reputed remedy for malaria and also stated to be efficacious in heart diseases, asthma, leucoderma, tumours and very good for treatment of chronic ulcers, chronic diarrhoea. It is used as an astringent, tonic and febrifuge (Kirtikar and Basu, 1975; Singh and Khan, 1990). Alcoholic extract of stem bark showed anticancer activity in HS1 human sarcoma in the embryonated egg (Anonymous, 1987). It also finds its use as antidyseric and antiseptic (Chopra *et al.*, 1994).

The principal constituents were reported to be linalool (35.7%), cis and trans linalool oxides, alpha-terpineol and terpinen-4-ol (Dung *et al.*, 2001). Atta-ur-Rahman *et al.* reported the isolation of an anilinoacrylate alkaloid, scholaricine, from the leaves of *Alstonia scholaris* to which structure 2-(demethylschoarine) has been suggested (Rahman *et al.*, 1985; Rahman, 1986). They also reported the isolation of 19, 20-dihydrocondylocarpine alkaloid from the leaves of *Alstonia scholaris* (Rahman *et al.*, 1986). Atta-ur-Rahman *et al.* (1987) also isolated 19, 20-Z- Vallesamine and 19, 20-EVallesamine from *Alstonia scholaris*. Lagunamine (19-hydroxytubotaiwine), angustilobine B acid and losbanine (6,7-seco-6-norangustilobine B) were obtained from the leaves of Philippine *Alstonia scholaris*, together with tubotaiwine, its oxide and 6,7-seco-angustilobine B, 17-OAcetylechitamine was isolated from the bark of the plant along with echitamine (Yamauchi *et al.*, 1990). Macabeo *et al.* reported the isolation and structural elucidation (MS and NMR) of first seco-uleine alkaloids, manilamine (18-hydroxy-19,20-dehydro-7,21-seco-uleine) and N4-methyl angustilobine B) from the (pH 5) alkaloid extract of Philippine *Alstonia scholaris* leaves together with the known indole alkaloids 19,20-(E) vallesamine, angustilobine B N4- oxide, 20(S)-tubotaiwine and 6,7-seco-angustilobine B (Macabeo *et al.*, 2005).

## Conclusions

It may be concluded that *Alstonia scholaris* is growing widely in Bangladesh, may be utilized as a source for the isolation of natural 2-Dodecyloxirane (31.83%), Benzene, 1,2-dimethoxy-4-(2-propenyl)- (8.49%) and Spinacene (6.09%) respectively, for medicinal and commercial use.

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