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Altitudinal Variation in Essential Oil Content in Leaves of *Zanthoxylum alatum* Roxb. A High Value Aromatic Tree from Uttarakhand

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ABSTRACT

In the present investigation two essential oil components linalool and limonene quantity were estimated in the leaves of male and female plants of different altitudes (775, 1100 and 1650 m). The plant material collected from different sites was named as Z-1, Z-2 and Z-3, respectively. The essential oil obtained was dark greenish black in bulk, yellowish green in thin layers, very fluid and had an odor of ether. In Z-1, Z-2 and Z-3 female leaves linalool was found to be 34.1, 27.2 and 11.7% and limonene to be 1.6, 2.4 and 6.4%, respectively. In male leaves also significant variation was found in the quantity of linalool and limonene in plants of different altitudes. In Z-1, Z-2 and Z-3 male leaves linalool was found to be 35.6, 19.8 and 10.0% and limonene to be 2.8, 4.0 and 4.5%, respectively.

Key words: *Zanthoxylum alatum* Roxb., leaf, essential oil, linalool, limonene, sex, altitude

INTRODUCTION

The genus *Zanthoxylum alatum* Roxb. is known as winged prickly ash. It is a member of family Rutaceae. It is found as forest undergrowth and in hot valleys up to 1800 m in the Himalayas. In Garhwal region the plant has dioecious nature and is found in altitudes ranging from 700 to 1800 m above sea level (asl). The genus *Zanthoxylum alatum* Roxb. is a plant used highly for medicinal as well as religious purposes by the Hindus in Northern India. Some other synonyms of the plant are *Z. armatum* DC, *Z. planispinum, Z. alatum subtrifoliolatum* (Franch.) etc. The English name of the plant is winged prickly ash and the vernacular name is timru or nepali dhaniya. The plant is a member of family Rutaceae. It is found as forest undergrowth and in hot valleys up to 1800 m in the Himalayas. It extends to temperate latitudes in E. Asia and E. North America. The seeds are grounded with black salt along with a little quantity of chilli and common salts and are given during stomach disorders. The seeds and bark are used as aromatic tonic during fever and cholera (Debl *et al*., 2009).

In Uttarakhand the Bhotiya tribal community uses this plant more than any other ethnic group due to its availability near their winter settlements. It is used in curing various common ailments such as toothache, common cold, cough and fever. Common stomach complaints are also treated with its soup. Most people consider the tree to have religious significance and magical properties (Kala *et al*., 2005). The fruit contains 1.5% essential oil called as *Zanthoxylum* oil.

The root is used in toothache, stomachache, fever, rheumatism, paresis, boils and as an insecticide and pesticide. The fruit is used in the treatment of stomachache, cough, colic vomiting, diarrhea and paresis and as an aromatic, stimulant and pesticide. The small branches, seeds and stem bark are prescribed in Fever, diarrhea and cholera (Kanjilal, 1997; Kirtikar and Basu, 1993; Anonymous, 1976).

A large amount of biochemical work has been done on the essential oil of this plant. Most of the work available deals with the composition of essential oil obtained from seeds, bark, wood, branches and leaves (Hussain *et al*., 1998). Yoshihito *et al*. (2000) from Nepal found that by GC and GC/MS analysis linalool and limonene were the main constituents of essential oil of *Z. armatum* DC. Jain *et al*. (2001) found that the essential oil of seeds of this plant had two major components linalool and limonene. It is suggested that the seeds of *Z. alatum* can be used as a commercial source for the isolation of linalool. Tiwary *et al*. (2007) also reported linalool and limonene as major components of essential oil from *Z. armatum* seeds. The sexual behaviour of this plant in Garhwal is mostly dioecious. In the present investigation an attempt was made to carry out sex based variation in the quantity of the oil present in leaves of plants situated at different altitudes. This kind of sex and altitude dependent variation work has not been reported earlier.

MATERIALS AND METHODS

The leaves sample of both male and female plant parts were collected from the forests of different sites (Table 1). The plant material was collected in the month of March during the morning hours in 2009. The oil was extracted by hydro distillation method for 6 h in a Clevenger-type apparatus at the Botany department of Government Degree College Karanprayag (Chamoli) Uttarakhand. The essential oil obtained in batches was dried over anhydrous sodium sulfate and after filtration, stored under refrigeration until tested and analyzed.

Gas chromatography: Analysis by GC were performed by using HP 6890 gas chromatograph equipped with a FID detector and a HP-S fused silica column (30X032 mmX0.2SlJm film thickness). Nitrogen was used as a carrier gas during analysis. The injector and detector temperature were maintained at 210 and 230°C, respectively. The column oven temperature was programmed from 60 to 220°C with an increase in rate of 3°C mm^{-1}.

RESULTS AND DISCUSSIONS

The essential oil obtained was dark greenish black in bulk, yellowish green in thin layers, very fluid and an odor of ether. The oil was found to be soluble in alcohol, ether and alkaline solutions. The leaf Analysis of female plant of different sites showed a considerable variation in linalool and limonene content. The data is shown in Table 2. In Z-1, Z-2 and Z-3 female plant's bark linalool was found to be 34.1, 27.2 and 11.7% while limonene was found to be and 1.6, 2.4 and 6.5%, respectively. In male leaves also significant variation was found in the quantity of linalool and

Table 1: List of various accessions of *Zanthoxylum armatum*

Accession No.	Site name	Altitude (meter asl)
Z-1	Gwar	775
Z-2	Semi	1100
Z-3	Kumud	1650

Table 2: The quantity of linalool and limonene in the leaves of male and female plants of various altitudes

	Female leaf		Male leaf	
Accession No.	Limonene	Linalool	Limonene	Linalool
Z-1	1.59	34.06	2.76	35.57
Z-2	2.43	27.19	4.52	19.80
Z-3	6.46	11.67	4.00	10.00

limonene in plants of different altitudes. In Z-1, Z-2 and Z-3 male plant leaves linalool was found to be 35.6, 19.8 and 10.0% and limonene to be 2.8, 4.0 and 4.5%, respectively. The mean quantity of linalool was found to be much higher than limonene in both male and female plant's leaves.

Previously work on the medicinal properties of the chemical composition of species (Kalia *et al*., 1999; Mehta *et al*., 1981; Kokate *et al*., 2001).

In the present study the quantitative Analysis of two essential oil components linalool and limonene was made in order to see that how these two vary in the leaves of male and female plants present at various altitudes. The analysis was GC based and it was found that the mean amount of linalool was much more in both male and female leaves as compared to limonene. The amount of linalool in the present study varied from 11.7 to 34.1% and limonene from 1.6 to 6.4% in female leaves. In male leaves the linalool varied from 10.0 to 35.6% and limonene from 2.8 to 4.5%.

A significant variability was found in the quantity of linalool and limonene in leaves of female and male plants. In female leaves the limonene content was found to be highest at high altitude while it was found to be decreasing in low altitude. The linalool in female plant leaves was found to be highest (34.1%) at low altitude while it decreased in higher altitude. In male plants the leaves had high content of linalool (35.6%) at low altitude while it decreased at higher altitude. The limonene content in male leaves was found to be highest in high altitude (4.5%) while it decreased in low altitudes.

The oils from *Z. armatum* are potential sources of linalool (Kirtikar and Basu, 1993). The essential oils collected from plants growing in different parts of the world show a great variation in the composition as well as content of particular constituent(s) due to geographical or environmental factors. Earlier reported linalool content from fruits of *Z. alatum* from J and K was 64.1 of 2.3% yield with unreported limonene content out of 6 constituents of essential oil (Sharma *et al*., 1966). As per Yoshihito *et al*. (2000) the quantity of two main compounds in the essential oil obtained from Nepal *Z. armatum* plants were linalool (62.2%) and limonene (12.6%). As per Jain *et al*. (2001) the linalool content and limonene content in *Z. armatum* seeds was found to be 71 and 8.2%. Indian workers reported linalool and limonene as major components of essential oil from *Z. armatum* seeds in India (Tiwary *et al*., 2007). Bhattacharya *et al*. (2009) reported linalool to be 23.3% and limonene to be 12.9% from the fruits *Z. nitidum* while limonene to be 33.1% from the leaf material of Assam region. According to Upadhyaya and Ashok (2010) the linalool content from the fruits of *Z. armatum* was found to be 28.9%. To the best of our knowledge no information is available about the sex and altitude dependent variation in the linalool and limonene content of this plant. In the present study from lower altitudes the leaves of both male and female plants produced high content of linalool, whereas the limonene content in both sexes was found to be highest at high altitudes. Both the male and female plants possess essential oil the quantity of which varied with altitude. Study concludes that the *Zanthoxylum alatum* requires more detailed explorations conservation strategy of the plant.

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