Journal of Applied Research on Medicinal and Aromatic Plants 3 (2016) 136–141

Contents lists available at ScienceDirect

Journal of Applied Research on Medicinal and Aromatic Plants

journal homepage: www.elsevier.com/locate/jarmap

Short communication

Drying methods and distillation time affects essential oil content and chemical compositions of *Acorus calamus* L. in the western Himalayas

Rakesh Kumar[a,*], Saurabh Sharma[b], Shivani Sharma[a], Neeraj Kumar[c]

[a] *Agrotechnology of Medicinal, Aromatic and Commercially Important Plants Division, India*
[b] *Academy of Scientific and Innovative Research, India*
[c] *Natural Product Chemistry and Process Development Division, CSIR- Institute of Himalayan Bioresource Technology, (Council of Scientific and Industrial Research), Post Box No. 6, Palampur 176 061, HP, India*

ARTICLE INFO

Article history:
Received 3 June 2015
Received in revised form 7 April 2016
Accepted 18 June 2016
Available online 5 July 2016

Keywords:
Acorus calamus
Drying methods
Distillation time
Essential oil
Beta asarone

ABSTRACT

Experiments were conducted to study the effect of drying methods and distillation time on essential oil content and composition of *Acorus calamus* L. rhizomes under western Himalaya during 2013–2014. In the first experiment, the effect of different drying methods (sun drying, shade drying, oven drying at 40 °C for 60 h and 70 °C for 24 h) on essential oil content and composition of vacha (*Acorus calamus*) rhizomes was studied. Volatile oil content and composition of dried rhizomes were determined by Clevenger type apparatus and GC–MS method, respectively. Drying the rhizomes under sun recorded significantly higher essential oil content (3.3 ± 0.0%) as compared to other methods. However, β-asarone (53.9 ± 0.1%) which is not desired in higher concentration was low in oven dried rhizomes at 40 °C for 60 h as compared to rhizomes of other drying methods. In second experiment, the effect of distillation time (6 h, 12 h, 18 h and 24 h) was studied. Analysis of data revealed that the essential oil increased significantly with distillation time up to 24 h. Concentration of low boiling point essential oil constituents *viz*., linalool, bornylene decreased with increase in distillation time but trend reversed for high boiling point constituents *viz*., β-asarone and acorone. Thus drying of rhizomes in oven at 40 °C for 60 h and distillation of dried rhizomes for 24 h maintained the quality of essential oil.

1. Introduction

Acorus calamus L. (Family: Araceae) commonly known as "sweet flag" is a semi- aquatic plant found in moist habitats such as the banks of pond, rivers and streams and swamps throughout Asia, Europe and North America. Although it is cultivated throughout India but it grows wild in abundance all over India, ascending to 2200 m in the Himalayas mainly in damp marshy places (Raina et al., 2003). The rhizomes of *A. calamus* and their essential oil are widely used in the flavoring industry and production of alcoholic beverages. Various pharmacological activities of *A. calamus viz*., anticonvulsant, antispasmodic, cardiovascular, anti-inflammatory, antioxidant, antidiarrhoeal, antimicrobial, antibacterial, anticancerous and antidiabetic has been reported earlier (Rajput et al., 2014; Sharma et al., 2014).

The chemical composition of essential oil from leaves and rhizomes of *A. calamus* showed extensive variations and the composition mainly depends on source of the plant part and the ploidy level of plant source; along with variation due to difference in soil constituents, climate and time of collection (Venskutonis and Dagilyte 2003; Singh et al., 2001). Beside this several factors *viz*., agrotechniques, post harvest techniques such as drying and distillation time can also influence the essential oil and chemical composition of aromatic crops.

Literature survey revealed that divergent drying methods have been used previously for drying different medicinal and aromatic plants. To maintain their quality, sun and shade drying have been used successfully for drying of plant material but they have disadvantage of slowness of the process, exposure to environmental contamination, uncertainty of weather and high labour requirements (Omidbaigi, 2005 and Ozcan et al., 2005). Room temperature or shade drying is the traditional technique being used to preserve medicinal plants, as the low temperatures are thought to protect the active components from degradation. However, this drying procedure is slow and metabolic processes may be long lasting which may lead to a lower quality of the plant materials; for example,

* Corresponding author.
E-mail addresses: rakeshkumar@ihbt.res.in, rakeshkrana@yahoo.com (R. Kumar).

http://dx.doi.org/10.1016/j.jarmap.2016.06.001
2214-7861/

color changes and loss in active ingredients (Fennell et al., 2004). In contrast with conventional methods, oven drying with different temperatures have been experienced cost and time effective in recent three decades for drying most of the plant materials. Likewise, effect of drying on essential oil composition of several crops such as *Rosmarinus officinalis* (Blanco et al., 2002b; Szumnya et al., 2010), *Mentha piperita* (Rohloff et al., 2005), *Mentha haplocalyx* (Shu et al., 2013), *Origanum vulgare* (Figiel et al., 2010), Basil (Ghasemi et al., 2013a), *Lavandula officinalis* (Zheljazkov et al., 2012), *Juniperus phoenicea* (Ennajar et al., 2010) and *Agastache foeniculum* (Sourestani et al., 2014) has been studied earlier.

Essential oil should be distilled for proper time to release all their active constituents. Distillation can determine value of the oil or can diminish the value. The length of distillation time was found to alter both essential oil content and composition of other aromatic plants such as *Rosa damascena* (Baydar et al., 2008), *Mentha piperita*, *Cymbopogon flexuosus* and *Cymbopogon martinii* (Cannon et al., 2013), *Lavandula angustifolia* (Zheljazkov et al., 2013a), *Juniperus scopulorum* (Zheljazkov et al., 2013b) and *Salvia officinalis* (Zheljazkov et al., 2014). There is little or no information on postharvest factors *viz.*, drying methods and distillation time effect on essential oil content and composition of *A. calamus* so an attempt has been made to study the effect of these factors in the western Himalayas.

2. Material and methods

2.1. Experimental site

The study was conducted from 2013 to 2014 at the experimental farm of CSIR-Institute of Himalayan Bioresource Technology, Palampur (1325 m amsl, 32°06′05″N, 76°34′10″E), India. The soil of experimental field was clayey in texture, acidic in reaction (pH 6.0), low in available nitrogen (241.5 kg ha^{-1}), high in available phosphorus (22.1 kg ha^{-1}) and potassium (417.9 kg ha^{-1}) and high in organic carbon (1.2%).

2.2. Experimental details

Well decomposed farm yard manure (FYM) @ 30 t ha^{-1} was thoroughly mixed in the soil before transplanting. Rhizomes of accession IHBT/AC/II (PLP16511) were cut into small pieces of 5 cm length and were planted in the plot during March 2013. A row to row spacing of 50 cm and plant to plant spacing of 50 cm was adopted in a plot of 3 m × 2 m. Hand weeding was done three times at monthly intervals. Need based water management was adopted. No pesticide spray was applied.

2.2.1. Drying methods

A. calamus rhizomes were harvested in the month of February 2014. The root and rhizomes samples were washed with tap water, separated and rhizomes were dried as per the treatment. Four treatments *viz.*, sun drying, shade drying, oven drying at 40 °C for 60 h, and oven drying at 70 °C for 24 h were performed. Sun drying of rhizomes under open air sunny conditions was done and it took 240 h until constant weight. Shade drying under conventional open air conditions in room temperature (25 °C) and it took for 720 h until constant weight. Maximum daily temperature during sun drying was between 32 and 35 °C. Furthermore, a cloth was laid between plant material and ground to avoid plant material contamination. Plant material was distributed as a thin layer for enhancing the speed of drying. Plant material was dried in hot air oven (MAC make, Model MSW-211, Macro Scientific Works Pvt. Ltd.) at two continuous temperatures (40 °C and 70 °C). The average moisture content after drying rhizomes at 40 °C and 70 °C for 60 h and 24 h, respectively was 0.67 g/g.

2.2.2. Distillation time

Four treatments of distillation time *viz.,* 6 h, 12 h (six hours for two successive days), 18 h (six hours for three successive days), 24 h (six hours for four successive days), respectively, were used. Sun dried rhizomes were used for hydro distillation. In each treatment 1000 g dry rhizomes were used. Distillation time was considered as the time after first drop of distillate was received. At the end of distillation time, the power was turned off and Florentine vessel was removed from the apparatus. Four replicates were used for evaluating essential oil content and two replicates were used for essential oil composition. Both experiments were conducted in randomized block design.

2.3. Essential oil extraction

The quantities of essential oils obtained at the end of distillation in both experiments were measured as mL and % ratios (v/w) were determined according to standard procedure described in European Pharmacopoeia (1975). The quantities of acorus oils obtained at the end of distillation were measured as mL and percent ratios (v/w) were determined by multiplying oil content with oil density i.e. 0.92 gm mL^{-1}. All the essential oil samples were dried over anhydrous sodium sulphate and were stored at 4 °C until analysed by GC and GC–MS analysis.

2.4. Gas chromatography analysis

Analysis of oil was carried out on Shimadzu GC-2010 gas chromatograph with FID detector and a DB-5 capillary column. 10 μL oil was dissolved with dichloromethane in GC vial. The operating condition was as follows: carrier gas nitrogen, hydrogen and air with a flow rate of 1.24 mL min^{-1}, the oven temperature was programmed as follows: 70 °C (4 min) and then 70 °C–230 °C at 4 °C min^{-1}, injector and detector temperatures were set at 250 °C.

2.5. Gas chromatography–mass spectrometry

Analysis of the oil was carried out on GC–MS (QP2010 Shimadzu, Tokyo, Japan) equipped with AOC 20i Auto sampler and DB-5 capillary column (SGE International, Ringwood, Australia) of 30 m length, 0.25 mm i.d. and 0.25 μm film thickness. 10 μL oil was dissolved with dichloromethane in GC–MS vial. Temperature was programmed from 40 to 220 °C at 4 °C min^{-1}, held isothermally at 70 °C and 220 °C for 4 and 15 min, respectively. Mass spectrometer source temperature, 200 °C interface temperature, 250 °C; injector temperature, 250 °C. Sample injection volume 2 μL (dilution: 5 μL oil in 2 mL dichloromethane, HPLC grade); and mass scan 40–800 amu. Helium was used as a carrier gas with 1.28 mL min^{-1} flow rate. The retention indices were calculated for all volatile constituents using homologous series of *n*-alkanes (C_8 —C_{24}).

2.6. Identification of components

The retention index was calculated for all volatile constituents using homologous series of *n*- alkanes (C_8 —C_{24}). The components of oil were identified by matching their mass spectra with those stored in the computer library namely Wiley, New York mass spectral (MS) library, National Institute of Standards and Technology, NIST (Stein, 2005), their retention indices (RI).

2.7. Statistical analysis

The design of the experiment was randomized block design (RBD). The data was analysed by software SYSTAT-12 (Systat Software Inc., Chicago, Illinois, USA). Least significant difference (LSD)