Plant Science Research

ISSN 0972-8546

Effect of climatic and physical factors on yield and quality of essential oil of *Pandanus odorifer* (Forssk.) Kuntze

J. K. Behera[1], I. S. Sandeep[2], N. Nasim[2], S. Nayak[2], S. Mohanty[2] and V. V. Rama Rao[1Ψ]

[1] Fragrances and Flavour Development Centre, MSME, Govt. of India, Kannauj 209 726, Uttar Pradesh

[2] Centre of Biotechnology, Siksha O Anusandhan University, Bhubaneswar 751 003, Odisha

ARTICLE INFO

Article history:
Received : 14 December 2017
Accepted : 20 December 2017

Keywords:

Pandanus odorifer
Kewda
essential oil
climate
physical factors

ABSTRACT

Pandanus odorifer (Forssk.) Kuntze., commonly known as Kewda, is one of the major bioresources of Ganjam district, Odisha used in small scale perfumery industry for aromatic compound extracted from the male inflorescences. In order to establish the effect of climatic and physical factors on perfume yield and quality, oil samples of Kewda flowers collected from the coastline of Ganjam, Odisha, India were analyzed. Gas chromatography (GC) analysis revealed the presence of Phenyl ethyl methyl ether or PEME (75-85%) and Terpinen-4-ol (4-10%) as major constituents of Kewda oil. The climatic factors were found to significantly influence the yield and quality of Kewda oil. The yield percentage of major constituents was found to be low in summer, followed by rainy season and winter season. The physical factors like quality of flowers, time of plucking and process of distillation also had major effect on the yield and quality of the oil.

1. Introduction

The genus *Pandanus* (Pandanaceae) is comprised of about 500 species, which are distributed in the tropical parts of the world extending from Western Africa in the west to the Polynesia, Pacific island in the east and in Northern Australia. Some 36 are reported to occur in India (Padhy *et al*., 2016). Of these, *Pandanus odorifer* (Forssk.) Kuntze. (Syn. *Pandanus odoratissimus* Linn) has widespread distribution and has several vernacular names such as Ketakee, Gandhapushpa, Sthiragandha, Indu Kalika, Jambala (Sanskrit); Kewra, Keora, Kewda, Gagandhul (Hindi); Kedgi, Kevda, Keora (Marathi); Kewoda (Gujarati); Kiya, Ketakee (Odia); Keya, Kedki, Keori (Bengali); Thazhai, Thalay, Thazhampoo (Tamil); Mogali, Gajangi (Telugu); and Kaitha, Kaida, Thala (Malayalam). In India, the plant grows widely along the entire coast of Indian peninsula and the Andamans. The species also occurs in the coastal regions of Iran, Malaysia, Mauritius, Myanmar, Java, China, Taiwan and southern islands of Japan (Panda *et al*., 2010). Though the plant is distributed all along the coast of Odisha from Ganjam in south-west to Baleswar in the North-east, it is fairly abundant in the coastal belt of Ganjam district. Especially, a stretch of about 4 km along the coast between the two rivers Bahuda and Rushikulya supports luxuriant growth of several populations of Kewda. Traditionally, the plants are used as vegetative fence around agricultural fields and planted along the coast as a wind breaker and soil binder (Panda *et al*., 2009; 2010). Besides, the leaves and roots are processed and used in local cottage industry for making mats, bags, baskets etc. and flowers in folklore and traditional medicine (Dutta *et al*., 1987). The plant is dioecious and the inflorescences of male plants are the source of essential oil or '*Rooh*' (2 Phenyl ethyl methyl ether, terpinen-4-ol, phenyl ethyl alcohol etc.) and perfume or '*Ittar*' (Panda *et al*., 2009). Generally, the female plants do not bear floral bouquets and develop into fruits (Rout *et al*., 2005). Essential oil of the Kewda has various commercial applications as food additives, aromatherapy, ayurvedic medicines, hair oils, agarbattis (incense sticks), lotions, cosmetics, soaps and perfume and significantly supports the local economy in

Ψ *Corresponding author; Email:* vvrao_ffdc@rediffmail.com

Ganjam district (Panda *et al*., 2012). The populations growing in this belt are considered superior in terms of the quantity and quality of essential oil like higher yield and essence value of the flowers (Fig. 1).

Fig. 1: *Pandanus odorifer* (Forssk.) Kuntze plant with flower

The small-scale perfumery industries located along the coast of Ganjam, Odisha are thriving due to abundance of Kewda plants in the region (Panda *et al*., 2009). The post-process wastes of distilled flowers are also being used in production of low grade cardboards by the paper mills and as manure.

The chemistry of the essential oil of kewda suggests that its major constituent, Phenyl ethyl methyl ether (PEME) contents varies between 75 to 85 percent among different populations and geographical regions (Sahu & Mishra, 2007; Misra *et al*., 2000; Mahalingam *et al*., 2012; Raina *et al*., 2004). Significant variation in major constituents of kewda oil has been attributed to edaphic and climatic factors. The present study was undertaken to establish whether climatic and physical factors play any role with regard to variation in major constituents of Kewda essential oils among the germplasms of different regions of Ganjam district of Odisha. The essential oils from flowers of seven male populations of *Pandanus odorifer* (KW-I to KW-VII) growing naturally along the coastline of Ganjam were analysed to suggest suitable physical conditions of flowers and climatic requirement to obtain quality essential oil with higher oil yield.

2. Materials and methods

2.1. Collection and processing of flowers

A survey was carried out along the coastline of Ganjam district of Odisha covering a distance of 80 km from Biswanathpur to Rushikulya River bank region for sample collection. For determining the effect of weather conditions on the contents of major constituents of the Kewda essential oil, male flowers were collected from seven populations from three zones (Table 1). The flowers were collected in the early morning between 8.0-9.0 AM in three prominent seasons (summer, rainy and winter). The collected flowers were hydro-distilled within an hour of collection in laboratory using Clevenger’s apparatus at FFDC Extension Unit, Berhampur. Traditional method of distillation practiced in the area was also used. In order to assess the effect of flower condition on the yield and quality of the essential oil, the decayed flowers, semi-bloomed flowers and buds were also collected and distilled as per the process described earlier.

Table 1

Geographical locations of collection of populations of *P. odorifer* from Ganjam district, Odisha

Populations	Regions	Lattiude	Longitude
KW-I	Rushikulya River Belt	$19^0$23’11.77"N	$85^0$1’36.53"E
KW-II	Chamakhandi	$19^0$10’15.27"N	$84^0$56’0.33"E
KW-III	Kalipalli	$19^0$17’48.97"N	$84^0$55’37.37"E
KW-IV	Keluapalli	$19^0$13’3.7"N	$84^0$48’29.84"E
KW-V	Indrakhi	$19^0$11’44.59"N	$84^0$49’20.5"E
KW-VI	Markandi	$19^0$11’35.02"N	$84^0$48’32.79"E
KW-VII	Mantridi	$19^0$11’14.94"N	$84^0$46’0.69"E

2.2. Distillation

Freshly plucked flowers collected from aforesaid locations were chopped to smaller pieces of about 1-2 inches length after the green parts of the flowers are removed as they contain no oil. After that the chopped flowers were added to the round bottom flask of 5 L capacity with water in the ratio of 1:3. It was connected to the Clevenger’s apparatus and kept on the heating mantle for hydro-distillation for 3 hours at a stock temperature of 60-70^{0}C. The first distillate came in a vaporized form at a temperature of 100-110^0 C after one and half hour and about 80% of the essential oil could be recovered during this phase. The remaining oil was extracted after completion of 3 hours of distillation.

2.3. Traditional distillation (Deg-Bhapka) method

The fresh Kewda flowers harvested from the fields in the early morning (6 AM) were graded and dressed up by removing the green leaf materials in order to maintain the aroma of the oil. The selected flowers were kept in the traditional copper stills (*Deg*) which could accommodate 400-500 flowers in a batch for distillation (Sahu *et al*., 2007). In each still, depending upon the number of flowers added, 80-110 L of redistilled water was added. The process of heating was done using firewood. At a temperature of 60 - 70^{0}C, the vaporization process started. The same is collected in the container locally known as “*Bhapka”*. In each batch, approximately 12-15 L of hydrosol (mixture of water and essential oil) was collected. The process of DEG-BHAPKA (traditional distillation method) took about 2-3 hrs (Sahu *et al*., 2007). After completion of the distillation process of a day, the collected hydrosols of all the batches were redistilled to recover the essential oil of Kewda and oil was separated from water using separating funnel.

2.4. Collection and storage of essential oil

After completion of hydro-distillation, the extracted essential oil of Kewda was collected and stored in air tight glass vials for further analysis. The percent oil yield was measured using standard methods.

2.5. Gas Chromatography (GC) analysis

Gas Chromatography (GC) analysis was done with nitrogen as carrier gas flowing at a rate of 1.0 ml/min. Flame Ionizing Detector (FID) supplied with air and hydrogen flowing at a rate of 350 ml/min and 35 ml/min respectively was used. The essential oil sample injected volume was 0.02µl. Initial oven temperature was 50^0C and hold time was 5 mins. Constituents of Kewda oil were analyzed through GC/GC-MS and compounds identified by comparing their mass spectral data with those in NIST library (Stein, 1990). Further, the major constituents of Kewda oil were analyzed employing Gas Chromatography system (GC, Agilent Technologies 6890 System, USA).

3. Results and discussion

3.1. Variation in constituents of essential oil in different seasons

The major constituents of essential oil showed significant variation in response to climatic variables and among different individuals and populations occurring in a range of habitats. The PEME (2 phenyl ethyl methyl ether) and terpinen-4-ol constitute about 90-95 % of the essential oil of Kewda. Results of the present study revealed that the contents of PEME and terpinen-4-ol are inversely proportional. While the PEME value decreased in summer, terpinen-4-ol increased during this period. In rainy season, which is the peak flowering period for Kewda, an increase in the PEME value and decrease in terpinen-4-ol content could be observed and this trend continued till flower harvest in winter. The PEME and terpinen-4-ol contents varied between 65.42-77.19% and 9.08-16.50 % during summer respectively. In rainy season, variation in PEME content was in the range of 70.38-81.5% and terpinen-4-ol between 7.95-15.53%, whereas during winter harvest the PEME values ranged between 80.05-83.62% and terpinen-4-ol from 4.68% to 9.97%.

3.2. Annual average variation in major constituents among different populations

The annual average variation in major constituents among different populations is shown in Fig. 2. It revealed

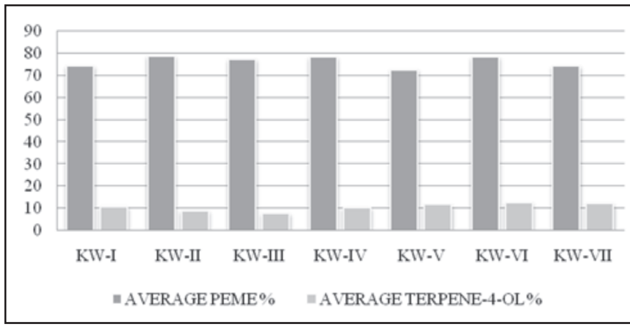

Fig. 2: Annual average variation in major constituents (PEME and Terpinen-4-ol) among different populations of Kewda

that the percentage of PEME varied from 72.57-78.85 % and terpinen-4-ol from 7.71-12.41 % among the populations studied.

3.3 Annual average variation in major constituents in the species

Comparative analysis of cumulative PEME and terpinen-4-ol percentage in the essential oil of Kewda showed significantly higher average PEME (81.71 %) in winter compared to summer (71.64 %) and rainy season (75.52 %), while terpinen-4-ol contents were found to be the lowest (7.54 %) in winter compared to 12.61 % in summer and 11.49 % during rainy season (Fig. 3).

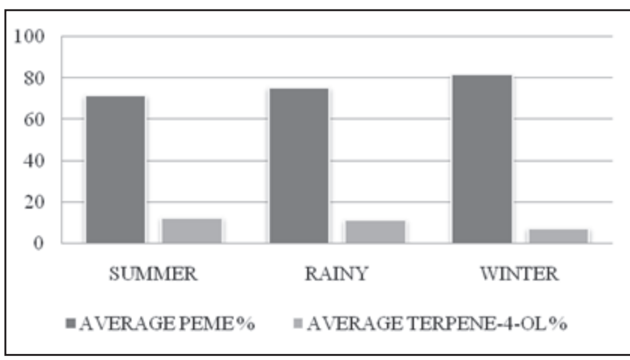

Fig. 3: Annual average variation in major constituents (PEME and Terpinen-4-ol) in the species (*P. odorifer*)

3.4. Effect of seasons on the essential oil yield

The essential oil yield of Kewda was significantly influenced by the seasonal variations in climatic parameters. In most of the populations, the oil yield was significantly higher during the winter season (0.34 %), while it decreased to 0.17% during the summer months (Fig. 4). Climate play a big role in plant secondary metabolite production as reported by several workers (Sandeep *et al*., 2016; Hassiotis *et al*., 2014; Løvdal *et al*., 2010;Mølmann *et al*., 2015;Tayade *et al*., 2013; Payyavula *et al*., 2012). In the present investigation, the essential oil yield was found to be high in winter followed by rainy and summer season and is in agreement with the earlier findings.

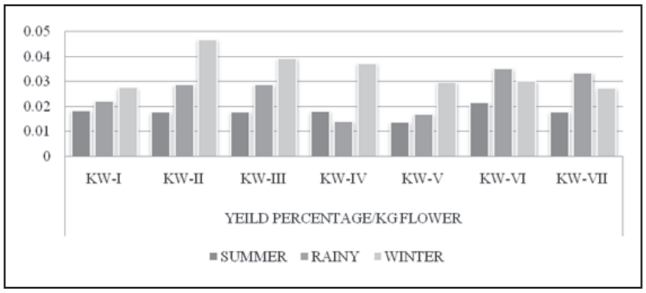

Fig. 4: Yield percentage of essential oil of Kewda per kg fresh weight of flower in different seasons

3.5. Impact of flower conditions on contents of major constituents of essential oil

The PEME and terpinen-4-ol contents in the essential oil varied due to physical conditions of the flowers. Fresh flowers had more PEME (81.70%) and less terpinen-4-ol (12.54 %) contents, where as in preserved and decayed flowers as low as 76.23 % of PEME and high terpinen-4-ol (14.66 %) values were determined. Interestingly, highest PEME (87.55 %) and lowest terpinen-4-ol (7.46%) contents were detected in flower buds of Kewda (Fig. 5).

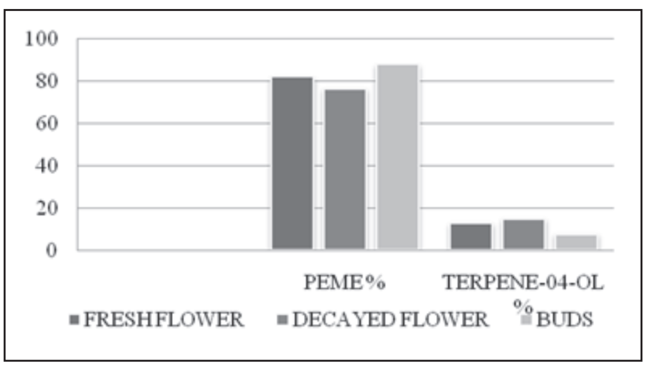

Fig. 5 Variation in major constituents (PEME and Terpinen-4-ol) of essential oil of Kewda at different stages of flower development

3.6. Impact of flower conditions on the yield of essential oil

The data obtained from the present study showed that the fresh flowers give better oil yield compared to decayed and buds. The fresh flower has a percent yield of 0.037% per Kg fresh weight whereas decayed and buds had an average yield of 0.013 and 0.010% per Kg fresh weight of flowers (Fig. 6).

Flower buds were found to contain higher quantity of good quality essential oil. The essential oil of Kewda is a very useful raw material for many value added products in Ayurvedic medicines, perfumery and other pharmaceutical products. Due to high demand and cost of Kewda oil, adulteration does take place and quality control becomes a key issue for authentication of the product. Therefore, the current study on variation in quality and quantity of essential oil in Kewda and how environment and genotypes modify the oil yield will provide baseline data for quality control of the oil. Further, the present work could be helpful for selection of sites for cultivation, provide clues for deciding correct time of harvesting and right stage of flower collection to ascertain better oil yield.

Fig. 6: Variation in essential oil yield percentage at different stages of flower development

Table 2

Variation in major constituents of essential oil of Kewda in different seasons and among populations of Ganjam district, Odisha

Locations/ Populations	Seasons	Average	
		Peme %	Terpinen-04-ol %
KW-I	Summer	67.4306083	13.92807667
	Rainy	75.447848	10.593688
	Winter	80.0502129	7.766814286
KW-II	Summer	71.7048225	12.0545675
	Rainy	81.8398467	8.751156667
	Winter	83.0345233	5.933706667
KW-III	Summer	75.1007125	9.08480125
	Rainy	76.2730275	7.954645
	Winter	80.8480667	6.111936667
KW-IV	Summer	77.193084	11.77467
	Rainy	77.4609925	9.189055
	Winter	80.2827733	9.972891667
KW-V	Summer	65.4275725	16.505385
	Rainy	70.388554	13.947032
	Winter	81.90102	4.682375
KW-VI	Summer	75.609705	12.45288
	Rainy	75.694915	15.5301625
	Winter	83.62995	9.263813333
KW-VII	Summer	69.0460967	12.50985667
	Rainy	71.54673	14.4955525
	Winter	82.281206	9.089582

4. Conclusion

The results demonstrated that significant variation do exist in major constituents of Kewda essential oil due to climatic variables and stages of flower development. Considerable changes in percentage of major constituents such as PEME and terpinen-4-ol were detected in samples collected in different seasons and at different stages of flower development. The analysis of quality parameters revealed that winter crop gives better quality oil compared to summer and rains. Similarly, in terms of yield too, winter crop was the best. The PEME and terpinen-4-ol percentage was recorded high in buds and fresh flowers, whereas decayed flowers had lower percentage of these constituents.

Acknowledgements

The authors are thankful to the Department of Biotechnology, Government of India, New Delhi for financial support and to the Principal Director of FFDC, Kannauj for providing necessary facilities and encouragement.

References

Dutta, P. K, Saxena, H.O. and Brahmam, M. (1987). Kewda perfume industry in India. Economic Botany 41: 403-410.

Hassiotis, C. N., Ntana, F., Lazari, D. M., Poulios, S. and Vlachonasios, K. E. (2014). Environmental and developmental factors affect essential oil production and quality of *Lavandula angustifolia* during flowering period. Industrial Crops and Products 62: 359-366.

Løvdal, T., Olsen, K. M., Slimestad, R., Verheul, M. and Lillo, C. (2010). Synergetic effects of nitrogen depletion, temperature, and light on the content of phenolic compounds and gene expression in leaves of tomato. Phytochemistry 71: 605–613.

Mahalingam, R., Bharathidasan, R., Ambikapathy, V. and Panneerselvam, A. (2012). Phytochemical compound analysis of *Pandanus odoratissimum*. Asian Journal Plant Science & Research 2: 228–231.

Misra, R., Dash, P. K. and Rao, Y. R. (2000). Chemical composition of the essential oils of Kewda and Ketaki. Journal of Essential Oil Research 12: 175–178.

Mølmann, J. A., Steindal, A. L., Bengtsson, G. B., Seljåsen, R., Lea, P. and Skaret, J. (2015). Effect of temperature and photoperiod on sensory quality and contents of glucosinolates, flavonols and vitamin C in broccoli florets. Food Chemistry 172: 47-55.

Naqvi, A. A. and Mandal, S.(1996). Investigation of the essential oil of *Pandanus fascicularis* Lam. by GC/MS. Journal of Essential Oil Research 8: 571–572.

Padhy, S., Dash, S. K., Panda, B. B., Misra, M. K., Padhi, N. P., Sahu, D., Das, M., Nayak, S. P., Dash, B., Rama Rao, V. V., Mohanty, P. C. and Padhy, R. (2016). *Pandanus fasicularis* Lamk. (Kewda): The Prime vegetation in the hinterland biodiversity of coastal Odisha with unique ethnic utility, genetic variation and economics- A review. Journal of Biodiversity 7(1): 33-49.

Panda, K. K., Das, A. B. and Panda. B. B.(2009). Use and variation of *Pandanus tectorius* Parkinson (*P. fascicularis* Lam.) along the coastline of Orissa, India. Genetic Resource Crop Evolution 56: 629- 637.

Panda, K. K., Das, A. B. and Panda, B. B. (2012). Genomics of *Pandanus*: A useful plant resource of coastal Odisha, India. Lambert Academic Publishing, Germany, pp. 88.

Panda, K. K., Sahoo, B., Das, A. B. and Panda, B. B.(2010). Use of RAPD markers to detect sex differences in *Pandanus tectorius* Parkinson, an important bioresource plant in Orissa, India. International Journal of Biodiversity Science, Ecosystem Services & Management 6: 28-34.

Payyavula, R. S., Navarre, D. A., Kuhl, J. C., Pantoja, A. and Pillai, S. S.(2012). Differential effects of environment on potato phenylpropanoid and carotenoid expression. BMC Plant Biology 12: 39.

Raina, V. K., Kumar, A., Srivastava, S. K., Syamsundar, K.V. and Kahol, A. P.(2004). Essential oil composition of "Kewda" (*Pandanus odoratissimus*) from India. Flavour Fragrance Journal. 19: 434–436.

Rout, P. K., Misra, R., Sahoo, S., Sree, A. and Rao, Y. R. (2005). Extraction of Kewda (*Pandanus fascicularis* Lam.) flowers with hexane: composition of concrete, absolute and wax. Flavour Fragrance Journal 20: 442 - 444.

Sahu, D., and Mishra, M. K. (2007). Ecology and traditional technology of screw pine perfume industry in coastal Orissa. Indian Journal of Traditional Knowledge 6(2): 253 - 261.

Sandeep, I. S., Kuanar, A., Akbar, A., Kar, B., Das, S., Mishra, A., Sial, P., Kumar, P., Nayak, S. and Mohanty, S. (2016). Agroclimatic zone based metabolic profiling of turmeric (*Curcuma longa* L.) for phytochemical yield optimization. Industrial Crops and Products 85: 229–240.

Tayade, A. B., Dhar, P., Kumar, J., Sharma, M., Chauhan, R. S., Chaurasia, O. P. and Srivastava, R. B. (2013). Chemometric profile of root extracts of *Rhodiola imbricata* Edgew. with hyphenated Gas Chromatography Mass Spectrometric Technique. PLoS One 8 (1): e52797 (DOI:10.1371/journal.pone.0052797)