PROVENANCE EFFECT ON MORPHO-CHEMICAL, MORPHO-GENETIC CHARACTERIZATION OF OIL OF *Hydnocarpus pentandra*

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By
DHATHRI N. R.

DEPARTMENT OF BIOCHEMISTRY
COLLEGE OF AGRICULTURE, DHARWAD
UNIVERSITY OF AGRICULTURAL SCIENCES,
DHARWAD – 580 005

JUNE, 2014

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INTRODUCTION

Every country in the world is improving their infrastructure rapidly. With the improvement of roads for transport facility world over, the automobile sector is witnessing a gigantic growth. Particularly diesel engines are used widely for in-sea and on-land transportation.

Worldwide petroleum consumption has steadily increased to about 4100 million tonnes in 2009, with an average annual growth rate of about 1.5 per cent in the past 20 years(Balat and Balat, 2009). In recent years, ever increasing trend of energy consumption due to industrialization and development has caused serious threat to the energy security and environment. Global fossil fuel consumption grew 0.6 million barrels per day and cost $ 111.26 per barrel in 2011 which means a 40 per cent increase than 2010 level. Current reserve of liquid fuel has the capacity to meet only half of the usual energy demand until 2023 (Owen, 2010). Such growth would affect stability of ecosystems and global climate and exert considerable pressure on global oil reserves. The crude oil as a result is becoming scarcer, more expensive and a highly volatile commodity.

Diesel engine has become the major source of air pollution as its emissions contain harmful waste like black smoke, hydrocarbon and oxides of nitrogen apart from causing noise pollution. The diesel engine emissions contribute heavily for the depletion of ozone layer, green house effect and acid rain productions which become causes for many human diseases and the degradation of the environment. The predicted shortage of petroleum resources, the increasing demand of energy and consequent pollution problems have forced us to search the substitute of petroleum derivatives such as biomass, alcohol, vegetable oils, wind, solar and hydrogen *etc.* This scenario has led to a renewed interest in the use of vegetable oils for making biodiesel because of their bio degradable, non-toxic, less polluting and renewable nature (Pintoa *et al.,* 2005; Murugesan *et al.,* 2009; Demirbas, 2009).

Biodiesel fuel can be defined as medium length (C_{16} ± C_{18}) chains of fatty acids and is comprised mainly of mono-alkyl fatty acid esters. Transesterification is the key and the foremost important step to produce the clear and environmentally safe fuel from vegetable oils. Chemically the oils/fats consist of triglyceride molecules of three long chain fatty acids. They differ by the length of carbon chains, orientation and position of double bond in these chains. Thus biodiesel refers to lower alkyl esters of long chain fatty acids which are synthesized either by transesterification with lower alcohols or by esterification of fatty acids.

Active research programs to reduce reliance on fossil fuels by use of bio-based alternatives and sustainable fuel sources are being pursued worldwide. Bio-diesel, a clean renewable fuel, is considered as one of the best candidates for substitution of diesel fuel. Bio-diesel can be produced from both edible and non edible oils and animal fats. In India edible oil production is not sufficient rather India is an importer of edible oils. Worldwide biodiesel is produced mainly from edible oils, but in India, non-edible oil seeds are available in abundance such as Pongamia (*Pongamia pinnata*), Jatropha (*Jatropha curcas*), Mahua (*Madhuca indica*), Neem (*Azadirachta indica*) and Rubber seed (*Hevea brasiliensis*) oil *etc.*, which can be tapped for biodiesel production (Nagaraj and Mukta, 2004; Sarviya*et al.,* 2012).

Biodiesel is expanding at a very rapid rate because of increasing demand, necessary policy support and technological availability. India consumes approximately 40 million tonnes of diesel and ranked fifth in the world after U S, China, Russia and Japan in terms of fossil fuel consumption. Recently, Government of India launched “National Mission on Bio-diesel” with a review to find a cheap and renewable liquid fuel based on vegetables oils. Realizing the mission to reduce India’s dependency on petroleum imports would be successful if a holistic approach is adopted for biofuels feedstock cultivation. Such a strategy needs to address alternate multipurpose crop plantation and method of producing biofuels without affecting the local ecology and farmer’s needs (Yamane *et al.*, 2006; Meher *et al.*, 2006).

Two species namely, *Jatropha curcas* and *Pongamia pinnata* are favoured in India because of their contrasting plant characteristics and the species selected should match the site characteristics. However, *Jatropha curcas* and *Pongamia pinnata* are not suitable in heavy rainfall areas like Western Ghat region. There is a need to search for suitable species providing high oil content suitable high rainfall areas. The percentage and composition of oils vary from species to species as well as from region to region due to different agro-climatic environments (Zafar *et al.,* 2003). A methodical research should therefore be carried out on variety of other oil yielding crops and other alternatives as biodiesel feed stock that are suited to the diverse socioeconomic and environmental conditions in rural India. Substitution of even a small fraction of total consumption by biodiesel will have a significant impact on the economy, agriculture and the environment.

Hydnocarpus pentandra (Buch.-Ham.) Oken, belongs to family Flacourtiaceae. Synonyms are *H. laurifolia* Sleumer and *H. wightiana* Blume. This family consists of about 85-89 genera and 800-1250 species distributed throughout tropical and subtropical region of the world. In Kannada it is known as Chalmogra yenne mara, Mirolhakai, Surti, Suranti, Toratti. The genus *Hydnocarpus* is the major group of Angiosperms. is available in moist deciduous and semi-evergreen forests of Western Ghats, India up to 850 m. *H. pentandra*is found in tropical forests along the Western Ghats, the Konkan southwards and below the Ghats in Kanara and Malabar in damp conditions especially near water. It is also very common inTravancore up to 609 m. (Sambamurty, 2005).

In India, it grows in tropical forests along Western Ghats, along the coast in Maharashtra to Kerala, Assam, Tripura, often planted on road sides in hilly areas. The tree is found in South East Asia, chiefly in Indo Malayan region, cultivated in Sri Lanka, Nigeria and Uganda. Trees of the species grow to a height of 12–15 m and in India trees bear fruits in May and June. The fruits are ovoid, 10 cm in diameter with a thick woody rind. Internally they contain 10-16 black seeds embedded in the fruit pulp. The seeds account for 20 per cent of the fruit weight. A typical tree produces 20 kg of seed per annum. The kernels make up 60–70 per cent of the seed weight and contain pale yellow oil. The seed oil from *Hydnocarpus* species is known as chaulmoogra oil. The oil is unusual in not being made up of straight chain fatty acids but acids with a cyclic group at the end of the chain. It is valued for its seed oil. Seed oil is anti-inflammatory, alterative, tonic, used as local application in rheumatism, sprains, braises sciatica and chest affection. Seed and seed oil are also used in leucoderma, worm infection, polyuria, eye diseases and sinus wounds (Kshirsagar and Upadhyay, 2009; Narayanan *et al*., 2011). It is used for treatment of lepromatous leprosy and is effective in decreasing size of nodules, anaesthetic patches, skin lesion (Yadav and Sardesai, 2002) and antihelmintic action against human tapeworm (Raj, 1975).

Tree borne oils species are becoming popular and *Hydnocarpus pentandra* is one of those species which is least exploited for its oil content and biodiesel potential. The present investigation was therefore, carried out with the following objectives:

1. To study the effect of different climatic conditions on growth and yield of *Hydnocarpus pentandra* in Agro climatic Zone 9 of Karnataka.
2. To study the effect of different climatic conditions on oil yield and oil quality parameters of *Hydnocarpus pentandra.*
3. To study the biodiesel yield and biodiesel quality parameters of *Hydnocarpus pentandra*
4. To study theseed cake quality parameters of *Hydnocarpus pentandra.*

REVIEW OF LITERATURE

The review of literature pertaining to the influence of provenance on growth, productivity, seed quality, fatty acid profile and seed cake quality and biodiesel are presented below. The information on these aspects in *Hydnocarpus pentandra* is very meager, with intense scrutiny of the literature; it was found that little work has been done on these aspects. Hence the reviews on other biofuel species or tree species have also been included.

2.1 Effect of different climatic conditions on growth and yield of tree species

The rate of growth and productivity of tree species varies greatly with site quality and other climatic, edaphic and topographic factors. Studies on growth and productivity of *H. pentandra* in relation to site conditions are meager and hence, an effort has been made to review the existing literature relating to species-site interactions in general among various tree species.

2.1.1 Variation in physical and chemical properties of Forest soils

Singh and Totey (1985) reported maximum increase in cation exchange capacity, exchangeable cations and organic matter in the soil under miscellaneous plantation flowers by Teak and Eucalyptus in Bhata soil of Raipur (MP).

Vitousek and Denslow (1987) found high availability of soil N in the mature forest under all soil types at La Selva.

Stewart and Vander (1988) compared chemical properties of Kalhari sand to a depth of 150 cm under Miombo woodland and the adjacent Eucalyptus plantation in Central Zimbabwe. Soils at both sites were strongly acidic and infertile nitrogen, mineral nitrogen and available phosphorus in the surface 10 cm under Miombo were more than double the values measured under Eucalyptus and the total nitrogen was significantly higher in the 20-30 cm depth, difference in these nutrients below 30 cm exchangeable potassium and magnesium were higher under Eucalyptus between 10-15 cm and it was estimated that there was double the quantity of exchangeable potassium in soil under the plantation.

Narain *et al.* (1990) reported soil nutrient status, their dynamics and chemical characteristics of soil, under Sal, Eucalyptus and Brush wood forests in Doon valley. Organic carbon, total nitrogen, available potassium and exchangeable calcium and magnesium in soil were higher in brush wood forest followed by Eucalyptus and Sal but available phosphorus was more under Sal. Accumulation of nutrients was conspicuous in surface soil, while lower depths did not show much change in nutrient status and also reported the soil physical properties under brush wood. Eucalyptus and Sal forests increased bulk density and decreased pore space under brush wood forest followed by Eucalyptus and then Sal forests in Doon valley.

Vadiraj and Rudrappa (1990) reported nutrient status of soil under five year plantation of *Casurina equisetifolia, Eucalyptus camaldulensis, Tamarindus indica* and *Mangifera indica* at Hoskote near Bengaluru. They observed decrease in pH, available P_2O_5 and K_2O and increase in organic carbon in all the plantations. However, electric conductivity remained unaffected.

Balagopan *et al.* (1992) reported the nature and properties of soils in monocultures of Teak, Eucalyptus and mixed stands of Teak and Bombax plantations. Among the soil properties studied, the chemical properties differed in these plantations in comparison to physical properties. The relatively low values for pH, organic carbon, exchangeable bases and exchange acidity in Teak and Eucalyptus in relation to Teak and Bombax plantations necessitate positive measures in the former plantations to preserve and enhance the fertility of the land.

Soil physical properties such as bulk density, porosity and water holding capacity were distinctly improved under plantations of *Casurina equisetifolia*, *Eucalyptus tereticornis, Cieba pentandra* and *Leucania leucacephala* as compared to rest of the species (Prathiban and Rai, 1994).

Forest tree species such as *Tectona grandis, Terminalia tomentosa, Pongamia pinnata, Gmelina arboria, Eucalyptus tereticornis, Acacia auriculiformis* and *Casurina equisetifolia* grown on lateritic soil showed that organic carbon, available nitrogen, potassium and phosphorus increase significantly in the surface layer under plantations (Chavan *et al.*, 1995).

Animon *et al.* (1999) reported soil pH under *Acacia auriculiformis* and *Eucalyptus tereticornis* plantation at three soil depths *viz.*, 0-10, 10-30 and 30-60 cm revealed that *A.auriculiformis* did not show any trend with increase in depth (5.4, 5.3 and 5.5), whereas *E. tereticornis* showed increase in pH with depth (5.0, 5.6 and 5.7). Potassium content was highest under *A. auriculiformis* (244.3, 209.1 and 134.4 kg/ha) as compared to *E. tereticornis* (128.8, 76.5 and 61.6 kg/ha) at three depths, respectively. Phosphorus content was lower under *A. auriculiformis* (14.9, 10.7 and 6.9 kg/ha) as compared to *E. tereticornis* (15.9, 14.3 and 12.6 kg/ha) at three depths, respectively. Organic carbon content of soils was found to decrease under all the plantations with soil depth. Higher organic carbon content was noticed under Acacia soils, while it was lower under Eucalyptus. Comparatively more organic carbon content under Acacia was attributed to high accumulation of litter and its slow decomposition.

Guo *et al.* (2002) reported that accumulation of biomass and nutrients (N, P, K, Ca, Mg and Mn) during the first 3-year rotation of three Eucalyptus short rotation forest species (*E. botryoides, E. globulus* and *E. ovata*) irrigated with meat works effluent compared with no irrigation. *E. globulus* had the highest biomass and nutrient accumulation either irrigated with effluent or without irrigation. After 3 year growth, *E. globulus* stands irrigated with effluent accumulated 72 oven dry tonnes per ha of above ground total biomass with a total of 651 kg nitrogen, 55 kg phosphorus, 393 kg potassium, 251 kg calcium, 35 kg magnesium and 67 kg manganese. Effluent irrigation increased the accumulation of biomass, nitrogen, potassium, phosphorus and manganese, but tended to reduce the leaf area index and leaf biomass, and decreased the accumulation of calcium and magnesium.

Forest soil properties, including the quantity and quality of soil organic carbon stocks, are influenced by the complex interactions of climate, soil type, management, and tree species (Lal, 2005).

Syed *et al.* (2006) reported that soil samples collected at a distance of 5, 10, 15, and 20 m from the Eucalyptus trees with two different depths indicated that soil pH of both the depths was alkaline. Electrical conductivity (EC) of the samples varied from 0.08 to 0.35 dS per m. The organic matter content was low in both the depths. The effect of distance on various soil properties, pH, EC, organic matter, phosphorus and potassium decreased with distance from the trees in the surface soil, while in the subsoil, organic matter and phosphorus decreased, whereas pH, EC and potassium increased with distance.

The soil surface properties underneath *Acacia salicina*, *Pinus halepensis* and *Eucalyptus occidentalis* in a 10 year-old common garden experiment established on a degraded *Stipa tenacissima steppe* in southern Tunisia have improved soil properties as compared to those of open areas (Jeddi *et al.,* 2009).

2.1.2 Variations in growth and productivity of tree species

Lopez (1987) observed geological differences in provenance of *Brosinm alicastrum* tree of moist tropical forest. Significant difference in plant height and stem girth due to provenances was observed in Eucalyptus.

De Jesus (1988) reported that at the age of 7 years, the Eucalyptus trees were all single-stemmed with an average height of about 20 m and DBH of about 19 cm. The *Eucalyptus grandis* and *E. tereticornis* showed standing volume of 88.5 and 205 m^3 per ha at the age of 4 and 10 years, respectively in Asarori (Dehradun) (Dwivedi, 1993).

The *Eucalyptus tereticornis* when grown at Jodigenhalli (Bengaluru) with the spacing of 2 x 2 m produced 4.44 m height and DBH growth of 4.227 cm, respectively at the age of 5 years, 6.35 m and 5.211 cm, respectively at 6 years, 7.31 m and 6.513 cm, respectively at the age of 7 years and 9.76 m and 8.683 cm, respectively at the age of 11 years (Kushalappa, 1993).

Gera*et al.* (1995) studied 20 provenances or seed sources of *Dalbergia sissoo.* Roxb. Scattered over wide range in India were studied for the pattern of genetic variation and character association. The results revealed the presence of highly significant variations among the provenances for height, number of branches and survival per cent. There are fair differences between phenotypic and genotypic co-efficient of variability indicating that these parameters are sensitive to environmental changes, the estimates of heritability and expected genetic gain were low to moderate for all characters, except number of branches which registered comparatively higher values of heritability (61.95%) and genetic gain (28.45%).

Hossain *et al.* (1997) reported the growth and biomass production of some Acacia and Eucalyptus species in degenerated Sal forest areas of Bangladesh. *A. auriculiformis* and *E. urophylla* produced higher growth and biomass in comparison to other species or seed lots.

Performance trial of 25 provenances of six species of Eucalyptus revealed significant difference with respect to height, diameter breast height, basal area and volume (Chandra *et al.*, 1998).

Growth performance of some tree species at the age of 9 and 12 years with the 2 x 2 and 4 x 4 m spacing, respectively revealed that at the age of 9 years, *Leucaena leucacephala, Eucalyptus camaldulensis, Acacia albida* and *A. auriculiformis* produced 29.3, 26.2, 21.3 and 18.4 m height and the DBH of 24.1, 24.1, 27.4 and 20.9 cm, respectively. *Albizia lebbeck* (4 x 4 m), *Azadiractha indica* (4 x 4 m), *Acacia nilotica* (3 x 3 m) and *Tamarindus indica* (4 x 4 m) showed highest growth of 12.2, 11.4, 10.5 and 11.2 m and the DBH of 18.2, 13.7, 15.2 and 12.7 cm, respectively at the age of 12 years (Rao *et al.*, 2000).

Among Eucalyptus, branch and foliage weight formed a relatively small fraction of the total above-ground biomass in more mature stands (Madgwick *et al.,* 2003).

Dogra and Sharma (2009) reported that *Eucalyptus hybrid* trees with diameter at breast height ranging between 7.1 and 56.3 cm and height from 10.0 to 42 m across Punjab state in. *E. hybrid* grown under Punjab's climatic conditions attained an average air dry timber density of 550 ± 28 kg per m^3.

2.1.3 Seed morphological characters

Agro-ecological conditions largely comprising edaphic and environmental factors can have more than one effect on the performance of the seeds. Success in the establishment and productivity is a function not only of the species used but also the source of seed within the species The place of production has greater influence on seed quality characteristics such as, seed weight, shelling percentage, oil content *etc.* The quality of the seed influenced by the weather conditions such as temperature, relative humidity, photoperiod, wind velocity, soil type, nutrition and soil chemical reactions vary from location to location (Lopez, 1987).

Bagchi and Singh (1992) reported that among provenances of *Acacia nilotica* from each of which seed length, seed width and seed thickness were measured and the data revealed significant difference with respect to all these parameters and they concluded that the higher inter-cluster distance in different groups indicates higher variability within the groups and it is quite likely that extreme individuals from different groups may help in selection.

Narayanaswamy (1994) collected groundnut seeds of JL-24 from 8 locations and TMV-2 from 12 locations. He observed significant differences among different locations for test weight (range varied from 38.2g to 46.5g for JL.24 and 25.7g to 34.90g for (TMV-2).

Deshpande and Mahadevappa (1996) evaluated the seeds of rice genotypes (Pushpa B, Mangala B, Pragthi B and Madhu B) produced from Bangalore, Mandya, Shimoga and Muidgere for various seed quality parameters. Significant differences were noticed due to provenances for seed characters and seed quality parameters. Seeds produced at Mandya and Bangalore recorded higher 1000 seed weight and density, as compared to the seeds produced at Shimoga and Mudigere.

Suresh *et al.* (1997) studied the effect of seed source on seed and seedling quality and revealed that 100 seed weight recorded significant trend among the different seed sources.

Thapliyal and Dhiman(1997) reported that there were significant difference among seed sources of *Pinus roxburghi* in seed weight, seed weight significantly correlated with cotyledon numbers, germination per cent whereas, germination value significantly correlated with seedling height, seed weight and cotyledon numbers with longitude of the seed source.

Devagiri *et al.* (1998) studied sixty seed sources of *Dalbergia sissoo* and observed that there was significant variation for several pod and seed traits between seed sources. Seed weight and number of seeds per pod showed large amount of variation and could be taken as an important criteria in selecting and delineating seed source.

Sindhuveerendra *et al.* (1999) reported that Sandal (*Santalum album*) seeds from various seed sources showed significant variation for morphologically and physiological traits. Data on seed length, seed width and seed weight showed significant amount of variation among different seed provenances.

Tyagi *et al.* (1999) reported that for seed characters the different provenances showed significant difference among seven provenances and the provenances revealed the existence of genetic variability in *Grewia optiva*.

Nautiyal *et al.* (2000) reported that seeds of *Quercus leucotrichophora* collected from seven places in Ghahwal and Kumaun regions varying considerably in their longitude, latitude and altitude and tested for their seed attributes. Provenance differed from each other in mean seed weight.

Similarly significant difference with respect to 100-seed weight was reported due to provenances in Acacia (Devarnavadagi *et al*., 2003). They also reported significant difference in oil content, Azardiractin content, collar girth, germination and vigour due to provenances in Neem.

Dhanai *et al.* (2003) conducted a study to identify suitable seed source for production of quality seedlings for mass afforestation in agroforestry systems at central Himalayas in *Albizia chinensis*. Significant variations were recorded among 13 populations for pod and seed morphology. Altitude of seed sources was found non- effective. Among all the pod- seed characters, number of seeds/ pod and seed weight were found most variable characters and number of seed / pod had higher genotypic, phenotypic and environmental co-efficient of variability.

Neelannavar and Manjunath (2003) carried out a study to estimate the genetic divergence among 30 seed sources of *Albezia lebbek* (L.) collected from all over India, to identify the promising seed source to be utilized in the breeding programme. Observations on seven characters namely seed length (mm), seed breadth (mm), seed thickness (mm), seed shape index, seed volume and 100 seed weight (g) were recorded. The study revealed that among the characters studied 100 seed weight contributed (44.37%) largely to genetic diversity followed by the seed shape index (15.63%).

Nayak *et al.* (2005) seeds of *Azardiracta indica* collected from different agro climatic zones of Karnataka showed significant difference with respect to germination and other seed quality parameters.

Kumar *et al.* (2007) collected 48 accessions of Pongamia and the data on various seed parameters, *viz*., pod length, pod width, seed length, seed width and 100 seed weight were recorded. The seed and seed oil content differed significantly among different accessions of Pongamia, the results of the study indicated that the pods and seeds of different accessions were polymorphic for seed length, seed width and 100 seed weight, oil content varied from 14.30 to 33.51 percent among the seeds of different accessions of Pongamia, indicating the considerable scope for selection of genotype for high oil content.

Dabgar *et al.* (2007) collected seed samples of Jatropha from Uttar Kannada, Haveri and Shimoga of Karnataka to identify superior seed source, a total of 25 promising seed sources were collected. Data on various seed parameters *Viz.,* seed length, seed width and 100 seed weight were recorded. The seed oil content differed significantly among different seed sources of *Jatropha curcas*. The minimum values for seed length (16.42 mm) in CPT-23 (Bisalakoppa), seed width (9.9 mm) in CPT-22 (Ahrekoppa), 100 seed weight (53.75 g) in CPT- 4 (Byathnalaz), where as maximum values were recorded in CPT 15 with respect to seed length (19.45 mm) seed width (12.54 mm) 100 seed weight (77.69 g).

Kaushik *et al.* (2007) variability in seed traits and oil content of 24 accessions of *Jatropha curcas* collected from different agroclimatic zones of Haryana state, India were assessed. There were significant differences in seed size, 100-seed weight and oil content between accessions. Maximum seed weight was recorded in seeds collected from IC-520602 and the least weight was recorded in IC-520587. Seed weight had positive correlation with seed length, breadth and thickness.

Twenty seed sources of *Pongamia pinnata* collected from Konkan region of Maharashtra from different agro climatic zones showed pod length varied from 14.50 to 69.69 mm and pod thickness also varied significantly among all seed sources. Average pod weight varied from 2.80 to 7.64 g. Maximum value for seed length (27.75 mm) was observed and maximum average seed weight (3.18 g) was recorded. The seed thickness varied from 5.00 to 10.00 mm (Raut *et al*., 2011).

Genetic variation patterns of Khilek (*Senna siamea*) were studied by Chakrit *et al.* (2012). Analysis of variance revealed significant differences among provenances in all the study traits except the relative growth rate in some stages of development. The provenance effect as determined by broad sense heritability was 50–83 per cent for seed morphometric traits, 53–85% for height, 33–62per cent for diameter at ground level, 14–67per cent for the height relative growth rate and 11–50per cent for the diameter at ground level relative growth rate. Significant correlations were found within seed mophometric traits and growth characters while the inter-character correlations between seed and growth characters were very weak.

2.2 Effect of different climatic conditions on oil yield and oil quality

2.2.1 Oil yield

Dhawan *et al.* (1981) reported that there was not much effect of location on oil content of Groundnut kernels (bunch types). The range reported was from 46.75 to 49.33 per cent at Ambala and at Sirsa it was 46.87 to 49.00 per cent of oil content.

Hydnocarpus kurzii contains 30.10 per cent oil and *Hydnocarpus wightiana* 63.0 per cent oil in their seeds (Bringi, 1987).

Kumaran *et al.* (2003) reported that there is a considerable variation in the oil content of different genotypes of *Pongamia pinnata* (karinja). The elite germplasm identified accession IC-546669 followed by IC546671 were found to be promising with respect to pod, seed, seedling traits and seed oil content genotypes.

Puri and Swamy (2003) have reported that seed of Jatropha collected from moist localities had higher oil content than dry localities. Among the dry localities also oil content in seed was also affected by different soil types.

Ginwal *et al.* (2004) have reported that, seed of Jatropha collected from different sources varied significantly for oil content.

Kaushik *et al.* (2007) reported variability in seed traits and oil content of 24 accessions of *Jatropha curcas* collected from different agroclimatic zones of Haryana state, India. There were significant differences in oil content between accessions. Oil variability ranged from 28.00 per cent in IC-520589 to 38.80 per cent in IC-520601. High heritability and genetic gain was recorded for oil content (99.00 and 18.90per cent). Seed weight had positive correlation with seed length, breadth, thickness and oil content.

Rao *et al.* (2011) studied variations in *Pongamia pinnata* collected from 11 different locations. There were significant differences observed in seed morphology and oil content. Plant height and number of branches exhibited much higher values of both phenotypic and genotypic variance than observed in the seed characters.

Seeds of *Jatropha curcas* collected from the outskirts region of the Bardoli (Gujarat) on extraction using light petroleum ether (60-80℃) by Soxhlet apparatus showed that the seeds consist of 46.31 per cent (dry w/w) oil (Nayak and Patel, 2010).

Saroj and Singh (2011) evaluated tree borne oilseeds in India, Mahua seed contain 30-40 percent fatty oil called mahua oil. Jatropha seeds with oil content of around 37 per cent. The air dried kernels of neem contains 27.5 per cent of oil.

Adebayo *et al.* (2011) conducted experiment on *Jatropha curcas* and reported that the percentage of oil yield got from the seed is 39.7 per cent.

Babagana *et al.* (2012) reported that, *Balanite aegyptiaca* seeds extracted with hexane (40-60ºC) yielded 34.52 per cent oil.

Bobade and Khyade (2012) conducted experiment on *Pongamia pinnata* and reported that the oil yield varies along with the change in extraction method, 24 per cent oil yield in mechanical expeller, 31 per cent in soxhlet extraction, and 27 per cent in cold percolation method.

Mishra *et al.* (2012) studied *Simarouba glauca* and reported that seeds contain 40 per cent oil and kernel contains 55-65 per cent.

Sanjay (2013) reported the presence of 75 per cent oil content in *Calophyllum inophyllum* seeds.

2.2.2 Fatty acid profile of oils

Cole and Cardoso (1939) studied the fatty acid composition of *Hydnocarpus anthelmintica* oil and reported the presence of palmitic acid (16:0, 7.5%), oleic acid (18:1, 12.30%), hydnocarpic acid (67.8%), gorlic acid (1.40%), chaulmogric acid (8.70%) and lower homologus hydnocarpic acid (0.10%).

The major fatty acids in *Pongamia pinnata* crude oil were palmic acid, stearic acid, lenoleic acid and ecosenoic acid. Oleic acid occurred in highest amount (44.24%), stearic (29.64%) and palmitic (18.58%) acids were the next in quantity. Hiragonic and octadecatrienoic acids were present in trace amounts (0.88%) (Shameel *et al.*, 1996).

Azam *et al.* (2005) reported fatty acid profile of *Hydnocarpus kurzii*, oil is rich with palmitic acid (16:0, 4.0%), oleic acid (18:1, 14.60%), hydnocarpic acid (39.40%), gorlic acid (19.50%) and chaulmogric acid (22.50%). He also reported the presence of palmitic acid (16:0, 1.80%), oleic acid (18:1, 6.90%), hydnocarpic acid (48.70%), gorlic acid (12.20%), chaulmoogric acid (27.00%) and chaulmogric homolog (3.40%) in *Hydnocarpus athelmintica.*

Kumar and Satyawati (2008) reported that, the oil fraction of Jatropha contains saturated fatty acids mainly palmitic acid (16:0, 14.10%) and stearic acid (18:0, 6.7%). Unsaturated fatty acids consisted of oleic acid (18:1, 47.0%) and linoleic acid (18:2, 31.6%).

Chhetri *et al.* (2008) studied two plant species, soapnut (*Sapindus mukorossi*) and jatropha (*Jatropha curcas*.) as newer sources of oil for biodiesel production. Soapnut oil was found to have average of 9.1 per cent free fatty acid (FFA), 84.43 per cent triglycerides, 4.88 per cent sterol and 1.59 per cent others. Jatropha oil contains approximately 14 per cent FFA, approximately 5 per cent higher than soapnut oil.

Akbar (2009) reported that, the lipid fraction of Jatropha oil seed contains oleic acid (44.7%) and linoleic acid (32.8%) as the dominant fatty acids while palmitic acid and stearic acid were the saturated fatty acids.

Nzikou *et al.* (2009) reported that, oil of *Jatropha curcas* seeds contain high levels of unsaturated fatty acids, especially oleic (up to 40.10%) and linoleic (up to 37.60%). The dominant saturated acids were palmitic (up to 15.63%) and stearic (up to 5.78%).

Seed oil of *Jatropha curcas* collected from the outskirts region of the Bardoli (Gujarat) on GC analysis showed presence of palmitic acid (16.69%), stearic acid (7.67%), oleic acid (40. 39%) and linoleic acid (33.09%). Oil contains high percentage of unsaturated fatty acid (75.64%). The study shows that fatty acids composition of the *J. curcas* oil is rich in oleic and linoleic acids and the oil can be classified as unsaturated oil.(Nayak and Patel, 2010)

Saroj and Singh (2011) reported the composition of typical air dried kernel of *J. curcas*. Major fatty acid composition of oil is palmitic acid (19.4%), stearic acid (21.2%), oleic acid (42.1%), linoleic acid (14.9%) and arachidic acid (1.4%).

The fatty acid composition of the *Pongamia pinnata* oil was determined by Gas Chromatography. The oil was rich with palmitic acid (9.77%), stearic acid (7.33%), oleic acid (35.72%), linoleic acid (19.17%), linolenic acid (4.17%), arachidonic acid (1.29%), behenic acid (5.84%) and lignoceric acid (24:0, 2.4%) (Natesan and Chandra, 2011).

Babagana *et al.* (2012) reported that *Balanite aegyptiaca* seeds extracted with hexane (40-60ºC) yielded 34.52 per cent oil. Gas Chromatography (GC) of oil indicated the presence of three fatty acids in the extracted oil and were mainly identified as; Palmitic acid, linoleic acid and stearic acid with 14.73, 75.86 and 9.40 per cent respectively.

Oscar (2012) examined seeds from different sources and reported that, oils are in reasonable levels in terms of yield (20.92 to 48 per cent) except breadfruit, groundnut, African bean seed and soybean, which contain 9.71, 19.2, 12.23 and 16 per cent, respectively. Most of them (breadfruit, melon seed, coconut seed, soybean and Dacryodes), contain mainly unsaturated fatty acids judging by their high iodine value which exceeded 100, and therefore are suitable for use in biofuel production.

Thiruvengadaravi *et al.* (2012) studied *Pongamia pinnata* oil for fatty acid profile and reported the presence of palmitic acid (16:0, 10.6%), stearic acid (18:0, 6.8%), oleic acid (18:1, 49.4%), linoleic acid (18:2, 19.0%), arachidic acid (20:0, 4.1%), behenic acid (22:0, 5.3%) and lignoceric acid (24:0, 2.4%).

Sixty seed powders and seed oils from various sources of *Pongamia pinnata* in southern Thailand were scanned by NIRS. All samples were analysed for oil content by hexane extraction and fatty acid composition by gas chromatography. Calibration equations were developed for oil content and individual fatty acids *viz.* palmitic acid (16:0), stearic acid (18:0), oleic acid (18:1), linoleic acid (18:2), and behenic acid (22:0), are identified (Chutarat *et al.*, 2013).

2.3 Biodiesel yield and biodiesel quality parameters

Tomasevic and Marinkovic (2003) reported conversion of Sunflower frying oil into biodiesel through alkaline trans-esterification at 25ºC, 6:1 methanol to oil molar ratio. They have reported 90 per cent biodiesel yield.

Dmytryshyn *et al.* (2004) studied the conversion of Canola oil into biodiesel by two-stage process, with potassium hydroxide as a catalyst. The conversion efficiency was 87 per cent at 25ºC, 6:1 methanol to oil molar ratio.

Zullaikah *et al.* (2005) reported the conversion of rice bran oil into biodiesel by two-step acid-catalyzed Sulfuric acid at 60ºC, 10:1 methanol to oil molar ratio. They have reported 96 per cent biodiesel yield.

Adebayo *et al.* (2011) conducted study on *Jatropha curcas*, and the oil yield of seed and biodiesel output of the oil were studied. There was a significant difference between physico-chemical properties of biodiesel from the fossil diesel. They also reported that the specific gravity, flash point, kinematic viscosity, iodine value of biodiesel was, 0.876 g per/cm^3, 170ºc, 4.8 cSt, and 7.64 respectively.

Calophyllum inophyllum (Punnakka) seed oil was transesterified using alkaline catalyst to produce biodiesel. The optimum conditions of this stage are methanol to oil ratio of 6:1, catalyst concentration of 1.0 per cent, reaction temperature of 60^{o}C, reaction time of 30 min and settling time of 60 min. biodiesel yield 92.5 per cent. The biodiesel properties are comparable with the ASTM 6751-02 standards set for biodiesel and also with those for petroleum diesel (Ramaraju and Kumar, 2011).

Anil *et al.,* (2011) conducted study on *Simarouba glauca*, reported that the content of free fatty acid, iodine value, saponification value were 0.65 mg KOH per gram of oil, 53.90 g per 100 g of oil, 174.55 mg KOH per gram of oil respectively. Analysis have been also done in fatty acid composition and was reported as palmitic acid, steric acid, oleic acid, linoleic acid, linolenic acid as 10.90, 25.66, 58.18, 3.30 and 0.35 per cent, respectively.

Bobade and Khyade (2012) conducted study on *Pongamia pinnata* oil for biodiesel production and reported a yield of 90 per cent. The biodiesel parameters are density (0.860g/cc), calorific value (3700 Kcal/kg), cetane number (41.70), acid value (0.46 mg/KOH) and iodine value (91) respectively.

Transesterification of Jatropha and Neem oils in methanol was studied with the use of potassium hydroxide as a catalyst. The biodiesel produced from Jatropha oil have properties within the acceptable range for automotive gas oil while the Neem biodiesel properties are not within the range. The Neem biodiesel properties obtained are flash point (160ºC), density (0.64 kg/m^3), pour point (10ºC), cloud point (15ºC) while that for Jatropha biodiesel are flash point (146ºC), density (0.42 kg/m^3), pour point (2ºC) and cloud point (3ºC) respectively. The fuel properties obtained for Neem biodiesel are higher in value than those obtained for Jatropha biodiesel (Mohammed *et al.*, 2012).

Biodiesel was produced from a non edible oil source, *Silybum marianum* L by alkaline trans-esterification, a maximum conversion of (95.0%) was obtained by using potassium hydroxide as a catalyst with a concentration of 0.90 per cent, 6:1 methanol to oil molar ratio, 60ºC reaction temperature and a reaction duration of 100 min. The properties of the produced biodiesel are found to conform to the ASTM standard specifications (Abdelrahman *et al.*, 2012).

Babagana *et al.*(2012) reported the yield of the biodiesel from *Balanite aegyptiaca* seed oil after 12-24 hours reaction was 90 per cent. The oil exhibited good physical and chemical properties which can be used in biodiesel production as the fuel properties were within ASTM 6751 standard specifications.

Pongamia pinnata seed was used to synthesize biodiesel (fatty acid methyl esters) by tranesterification with methanol in the presence of two different base catalysts *viz.* NaOH and KOH at a predetermined optimum temperature of 60˚C, stirring speed of 300 rpm for 45 minutes. A percentage conversion of 68 and 73 per cent respectively was achieved by NaOH and KOH catalyzed transesterification reaction. Biodiesel properties obtained are flash point (222 and 228ºC), density (0.790 and 0.774 kg/m^3) and fire point (252 and 262 ºC) respectively for biodiesel obtained from KOH and NaOH catalyzed transesterification reaction (Arun *et al.*, 2013).

2.4 Deoiled cake and its quality parameters

Sumitra *et al.* (2007) reported that edible oil cakes have high nutritional value; especially have protein content ranging from 15 to 50 per cent.

Ramchandra *et al.* (2006) reported the manurial values of *Jatropha curcas* and *Pongamia pinnata* oil cakes with nitrogen (%) phosphorus (%) and potassium (%) are 3.85, 2.09, 1.68, and 5.50, 1.00, 1.00 per cent respectively.

Kumar and Satyawati (2008) reported that, Jatropha seed cake is as a straight fertilizer its properties were compared with those of other organic fertilizers with regard to nitrogen, phosphorus and potassium content: Nitrogen-3.2-4.44, 5.0, 0.97; Phosphorus 1.4–2.09, 1.0, 0.69; Potassium-1.2–1.68, 1.5, 1.66 per cent of *J. curcas* oil cake, Neem oil cake and Cow manure respectively.

Proximate analyses of un-decorticated and decorticated seeds and cake revealed that both the oil seed and cake contain valuable nutrients namely soluble carbohydrate: seed (7.96%), cake (24.69%), protein: seed (20.78%), cake (31.06%), fat: seed (51.20%), cake (19.40%), mineral matter: seed (7.75%), cake (11.10%) for un-decorticated castor seeds and cake respectively. The decorticated seed cake gave higher values of these nutrients than the un-decorticated, for instance carbohydrate: seed (8.86%), cake (24.88%), nitrogen: seed (21.87%), cake (35.43%), ether extract: seed (55.50%), cake (25.10%), total ash: seed (9.40%) and cake (7.14%) (Annongu and Joseph, 2008).

Osman *et al.* (2009) conducted experiment on *Pongamia pinnata* seed cake and reported that pongamia cake contains nitrogen (4.28 %), phosphorous (0.40 %), potassium (0.74 %), magnesium (0.17%), zinc (59 ppm), boron (19 ppm) and sulphur (1894 ppm).

Raphael *et al.* (2010) reported that after oil extraction from the seeds of *Jatropha curcas* a residue matter or cake remains as a by-product. Generally, about 50 or up to 75 per cent of the weight of seeds remains as press-cake. This press-cake contains mainly proteins and carbohydrates. Nitrogen, phosphorus and potassium levels of the cake are 6, 2.8 and 0.9 per cent, respectively.

Mostafa *et al.* (2011) reported proximate nutritional compositions of *Swietenia mahagoni* seed cake. The proximate nutritional composition of the seed cake was found to contain moisture (14.37%), minerals (16.36%), fats (19.42%), crude fiber (19.60%), protein (8.76%) and carbohydrate (21.49%).

Defatted kernels (expeller cake) after the extraction of Karanja seed kernels with hexane contained 31.9 per cent protein, 7.8 per cent oil and 4.2 per cent alkaloids (Rahman *et al*., 2011).

Proximate composition, mineral elements, anti-nutrients (toxicants) and lipid characteristics were determined for almond (*Terminalia catappa*) seeds. The results obtained showed the proximate composition was moisture (25.23 %); ash (5.00%); lipid (32.73 %); crude fibre (33.66 %); crude protein (3.11 %); carbohydrate (25.47%) and caloric value 534.2 Kcal. Mineral elements determined were P (10.0 mg/100g), Na, K, Fe, Mg (26.4 mg/100g) and Ca (36.1mg/100g) (Akpabio, 2012).

Inekwe *et al.* (2012) studied proximate composition of seed and seed cake of *Jatropha curcas* from India, Kaduna and Edo, using standard methods. The proximate analysis for the seed revealed that the (%) presence of crude protein was (24.72±0.00) for Kaduna, (21.11±1.09) for India and (28.87±1.24) for Edo; and these increases were significant ($p < 0.05$). Crude lipid for Edo (29.95±0.50) was significantly ($p < 0.05$) lower than that of Kaduna (37.85±4.11). Crude carbohydrate for Kaduna (15.51±0.97) was significantly lowers ($p < 0.05$) than that of India (18.58±1.64). Crude fibre contents were not significant ($p > 0.05$). Also, the Ash and Moisture contents were not significant ($p > 0.05$). Seed is not edible (due to its toxic potentials); it has great nutritional potentials, especially as a source of animal feed. It is therefore a good potential source of animal nutrition if subjected to appropriate detoxification processes.

Aisha *et al.* (2013) conducted experiment on proximate analysis of wild castor seed and reported that potassium (K) was the highest mineral contained in the castor seed (1850 mg/ml) followed by sodium (Na) with 45 mg/ ml, then Phosphorus (P) (2.56 mg/ml) and Magnesium (Mg) with 1.2 mg/ml. The least mineral contained was Calcium with a value of 0.35 mg per ml. The ash content was 3.5 per cent, crude fiber (1.0%), crude protein (11.11%) and carbohydrate content was 28.39 per cent.

MATERIAL AND METHODS

Present study was conducted in Agro-climatic Zone-9 of Karnataka. Experiments are undertaken in the already existing trees in natural forests. Four experimental sites are chosen based on the different levels of altitude. In each site three trees are chosen for the study. Following studies were conducted

3.1 Study area

Uttara Kannada district, Karnataka is located between 13^0 to 16^0 N latitude and 74^0 to 76^0 E longitude and extended over an area of 2000 square kms. The district comes under the Agro-climatic Zone-9 of Karnataka *i.e.,* Hilly Zone. The main geographic feature of the Uttara Kannada district is the Western Ghats or Sahyadri range, which runs from north to south through the district. East of the Sahyadris is the Balaghat upland, part of the vast Deccan plateau. Moisture-bearing winds come from the West and yearly rainfall averages 1,000-4500 mm, and as high as 5,000 mm on the west-facing slope of the Sahyadris. East of the crest is the rain shadow of the Sahyadris, which receive as little as 1,000 mm annually. Much of the rain falls in the June–September Monsoon. The district's high rainfall supports lush forests, which cover approximately 70 per cent of the district. The North Western Ghats moist deciduous forests cover the Sahyadris below 1000 meters elevation. Many trees shed leaves in the drier months. In pockets above 1000 m elevation lie the evergreenNorth Western Ghats montane rain forests.(Plate 1a and 1b).

3.2. Site conditions

The sampling sites lie between 14^0 52’-14º 53’N to latitude and 74^0 58'-74^0 67’ E longitude. Four different locations based on different levels of altitudes were chosen. The rainfall in these sites ranges between 2000-3200 mm per year and the altitude ranges between 400-550 m (Table 1 and 2).

3.2.1 Treatment details

Treatments: Four provenances (3 candidate plus trees from each provenance) and three fruit sizes.

Replication: Three

Design: Split plot

3.2.2 Seed source

Seeds were drawn randomly form the pods harvested from the identified candidate plus trees, three each from each location.

3.2.3 Collection of samples:

Samples were collected from four different location of Hilly zone in the month of May, 2013. The samples were immediately preserved in polyethylene bags and transferred to experimental laboratory. The geographic information of these sites was recorded by using GPS model. In each location, 3 trees were selected for collection of fruits, soil sampling and recording of tree growth parameters. (Plate 2a, 2b and 2c)

3.3 Stand enumeration

Stand enumeration was done as per the method of Chaturvedi and Khanna (1982).

3.3.1 Diameter at breast height (m)

The diameter at breast height (DBH) was recorded in “m” with the help of tree caliper at 1.37 m above the ground level.

3.3.2 Total height (m)

The total height (m) of the tree from base to tip was recorded using Ravi altimeter.

3.3.3 Bole height (m)

The height (m) of the main stem from the base to first branch of the tree was recorded by using Ravi Altimeter.

Plate 1a: Agro-climatic zones of Karnataka

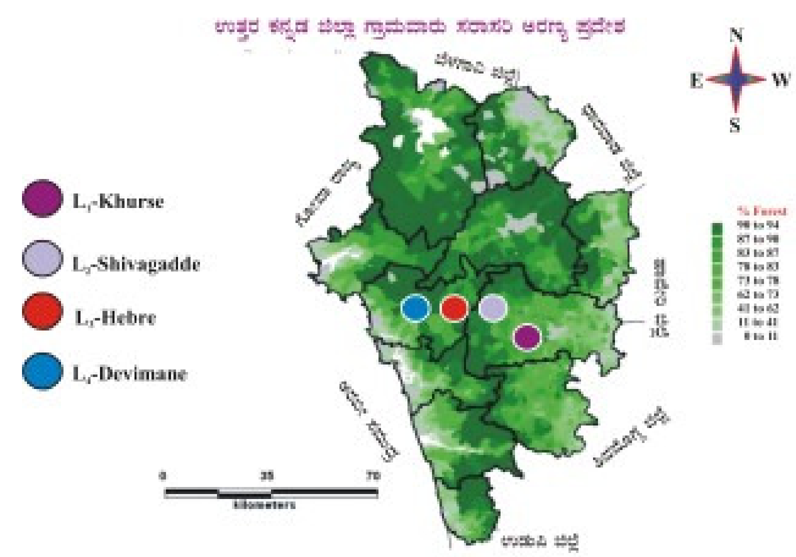

Plate 1b: Study area in Uttara Kannada district

Table 1. Details of area selected for the current study

Locations	Place	Taluk	District
Location-1 (L_1)	Khurse	Sirsi	Uttara Kannada
Location-2 (L_2)	Shivagadde	Sirsi	Uttara Kannada
Location-3 (L_3)	Hebre	Kumta	Uttara Kannada
Location-4 (L_4)	Devimane	Kumta	Uttara Kannada

Table 2. Site factors of the study area

Locations	Place	Latitude	Longitude	Altitude (m)	Mean Annual rainfall (mm)	Annual rainy days	Mean Annual temperature (°C)	Relative humidity (%)
Location-1 (L_1)	Khurse	14º 53’500-14º53’859 N	74º 67’ 236-74º 67’ 703E	541-549	2000	84	31	79
Location-2 (L_2)	Shivagadde	14º 52’484-14º 52’653 N	74º 62’220-74º 62’363 E	442-447	2450	92	30	84
Location-3 (L_3)	Hebre	14º 52’230-14º 52’599 N	74º 58’734-74º 61’958 E	428- 440	2900	110	28	86
Location-4 (L_4)	Devimane	14º 52’347-14º 52’405 N	74º 60’579-74º 60’671 E	400-413	3400	116	28	91

Plate 2b: Fruits of *Hydnocarpus*

Plate 2a: *Hydnocarpus pentandra* tree

Plate 2c: Seeds of *Hydnocarpus pentandra*

The spread of crown of the tree was measured in two directions perpendicular to each other. The average of two values was recorded as the crown diameter

3.3.5 Tree Basal area (m^2)

The basal area of the tree was determined by using following formula

$$\text{Tree Basal Area (TBA)} (m^2) = (DBH/200)^2 \times 3.142$$

Where, DBH = Diameter at breast height

3.14 is constant

3.4 Soil analysis

3.4.1 Collection and preparation of soil sample

The soil samples were collected from the field and brought to the laboratory in polythene bags. The soil samples were dried under shade, pounded with wooden pestle and mortar, passed through 2 mm sieve for the analysis of available N, P and K. For organic carbon, soil samples were again pounded in an agate mortar and passed through 0.5 mm sieve and preserved in polythene bag for further analysis.

3.4.2 Soil properties

3.4.2.1 Available nitrogen(kg/ha)

The available soil nitrogen was estimated by micro-Kjeldahl method (Black, 1965). Soil sample was digested in Conc.H_2SO_4 with a pinch of digestion mixture comprising salicylic acid, K_2SO_4 and Na_2SO_3. The digested sample was distilled with excess of 40 per cent sodium hydroxide. Ammonia thus released was trapped in 2 per cent boric acid and the ammonium borate thus formed was titrated against previously standardised 0.05 N Hcl.

3.4.2.2 Available phosphorus(kg/ha)

Available phosphorus in soil was extracted by Bray and Kurtz No. 1 extracting solution (0.03 N NH_4F in 0.025 N HCl) (Bray and Kurtz, 1945). The extract phosphorus was determined by chlorostannous reduced molybdophosphoric method using spectrophotometer at 660 nm (Jackson, 1973).

3.4.2.3 Available potassium(kg/ha)

Available soil potassium was determined by extracting the soil sample with neutral ammonium acetate solution and estimated by using flame photometer as outlined by Jackson (1973).

3.4.3 Soil reaction

3.4.3.1 pH

The pH of the soil-water suspension was determined by using a digital pH meter.

3.4.3.2 Electrical conductivity

Electrical conductivity was determined by using Conductivity Bridge.

3.4.3.3 Organic carbon

The organic carbon content of the soil sample was determined by wet oxidation method of Walkey-Black as described by Jackson (1967).

3.5 Fruit/seed and Kernel parameters

Physical characteristics

The fruit material was divided into 3 lots and 50 fruits/ seeds/ kernel were selected at random from each lot based on size. The visual observations of seeds were made to collect information on fruit/seed/kernel colour and shape. The fruit, in terms of the three principal axial dimensions, that is (in mm): length (L), width (W) and thickness (T) was measured using a Vernier caliper (Aerospace, China) with an accuracy of 0.01 mm. The seed/ kernel in terms of the three principal axial dimensions, that is (in mm): length (L), width (W) was measured using image analyser. These measurements were replicated three times to get mean values (Plate 3a, Plate 3b).

3.6 Extraction and quantification oil from seed kernel

3.6.1 Oil isolation

The seeds are sun dried. The dried kernel are blended and mixed thoroughly in a Kenstar food mixer. The powdered sample was stored in an air-tight jar and kept in the refrigerator prior to the analysis. Oil was extracted by soxhlet extraction method using solvent hexane. Moisture in the oil samples were removed by adding anhydrous sodium sulphate.

3.6.2 Percentage yield of oil

The amount of extracted oil was determined and percentage yield of the oil fromeach sample was calculated on the basis of kernel by using following formula:

$$\text{Percentage of oil yield} = \frac{\text{Weight of oil}}{\text{Weight of kernel}} \times 100$$

3.7 Organoleptic and Physico-chemical characterization of oil

3.7.1 Colour and aroma of oil samples are recorded with visual observations and smelling.

3.7.2 Kinematic Viscosity: By using viscometer (AOAC, 1990).

3.7.3 Specific gravity / density

By using densitometer method (AOAC, 1990).

3.7.4 Determination of acid number and free fatty acid

The acid number and free fatty acid of oil was determined by the procedures of AOAC. (1975)

$$\text{Acid value} = (V \times N \times 56.1) / W$$

where, V = volume of potassium hydroxide used,

N = normality of KOH

W = weight in g of the sample

Free fatty acid as oleic acid, per cent by weight = $(28.2 \times V \times N) / W$

$$\% \text{ FFA} \times 1.99 = \text{Acid value}$$

Where, V = volume of potassium hydroxide used,

N = normality of KOH

W = weight in g of the sample

3.7.5 Determination of iodine number

The iodine number was determined by Hanus iodine method (AOAC, 1975).

$$\text{Iodine number} = \{(B - S) \times N \times 12.69\} / \text{Weight of the sample (g)}$$

Where, B = ml of 0.1 N sodium thiosulfate required by blank,

S = ml of 0.1 N sodium thiosulfate required by sample,

N = Normality of sodium thiosulfate solution.

3.7.6 Estimation of saponification number

The saponification number was estimated as per the method of AOAC (1975).

$$\text{Saponification number} = \frac{28.05 \times (\text{titre value of blank-titre value of sample})}{\text{Weight of the sample(g)}}$$

Plate 3a: Fruits size variations in *Hydnocarpus pentandra* collected from different locations

Plate 3b: Seeds obtained from different size fruits of *Hydnocarpus pentandra*

3.8 Chemical characterization of oil

The Fatty acid composition of oil was determined by Gas chromatography mass spectrometry (GC-MS).

3.8.1 Gas Chromatographic- Mass spectrometry analysis of oil

The chemical composition of oil was determined by chromatography technique. GC-MS analyses were carried out on a Shimadzu Chromatopac GC-MS QP2010S GC-MS system equipped with a DB-5 fused silica column (30 m x 0.25 mm i.d.). Oven temperature was 40℃ increasing to 250℃ at a rate of 4℃, transfer line temperature 260℃. The carrier gas was helium with a linear velocity of 31.5 cm/s, split ratio 1/60, Ionization energy 70 eV, scan time 1 sec and mass range of 40-300 atomic mass unit (1 Atomic mass unit = 1.66×10^{-27}). The compounds were quantified by the area normalization method without considering response factors. The components of the oils were identified by comparison of their mass spectra with those of a computer library, and confirmed by comparison of their retention indices with those of data published in the literature.

3.8.2 Biodiesel Production

Transesterification reaction was performed as per the procedure reported by Venkatesan *et al.*, 2012.

Experimental Setup and Steps

Transesterification was carried out in a batch type reactor. This reactor consists of following components.

Experimental Setup

Three necked round bottom flask is used to collect sample of mixture (oil + Methanol + catalyst). Magnetic stirrer and heater provide the stirring and heating effect simultaneously. Temperature controller is used to control the desired heating effect. Stirrer controller is used to control the stirring effect.

Transesterification reaction

A round bottom flask of 500 ml is used for the present analysis. The Hydnocarpus oil (100 ml) in the flask was heated on a hot plate having magnetic stirrer arrangement. The mixture was stirred at the same speed for all test runs. Initially oil was heated to 65℃. Then the methanol 15 ml and the sulphuric acid 0.3 ml are mixed. The mixture is transferred to a reactor. The temperature maintained for the whole transesterification process is about 60 ℃ and heated up to 3 h and reaction vessel was allowed to cool for 3 h. The resultant mixture is taken into the separating funnel containing acidic oil and impurities with glycerol. After 3 to 4 h impurities settles down at the bottom of acidic oil. Impurities with small amount of glycerol are separated by opening the value of the separating funnel.

Initially obtained acidic oil from first step transesterification reaction is heated to 65℃. Then 15 ml methanol and 1 g of NaOH are mixed. The mixture is transferred to a reactor. The temperature was maintained for the whole second step transesterification process at 60℃. The mixture was stirred at the same speed (390 rpm) and heated for to 3 h. and after that allowed to cool.The resultant mixture is taken into the separating funnel containing biodiesel and glycerol. After 3 to 4 h glycerol settles down at the bottom of biodiesel. The glycerol is separated by opening the value of the separating funnel. The washing was carried out in a separating funnel. The separated biodiesel was used for characterization.(Plate 4a, 4b, 4c, 4d).

3.8.3 Physico-chemical characterization of Biodiesel

Colour and aroma, kinematic viscosity, specific gravity / density, acid number, iodine number, saponification number were estimated as per the above listed procedures (3.7)

3.8.3.1 Determination of cetane number

Cetane number was calculated empirically as per Krisnangkura (1986).

Cetane number = 46.3 + (54.58/SN-0.225 × IV)

Where, SN is the saponification number, IV is the iodine value

3.8.3.2 Flash Point: Flash point of the biodiesel was determined by using Pesky-Martens flash point determination unit (Venkatesan *et al.*, 2012).

3.9 Macro, Micronutrients and Proximate Composition of deoiled cake

Hydnocarpus deoiled cake was used for determination of macro and micronutrients content. Proximate composition of deoiled cake samples were analyzed by standard methods.

3.9.1 Estimation of Macro and micronutrients

3.9.1.1 Digestion of sample

1 g of powdered sample was pre-digested with Conc. HNO_3 overnight. Further digestion was carried out with 5 ml of di-acid mixture (HNO_3 + $HClO_4$) until clear solution was obtained. The residue was dissolved in 6 N HCl and the volume was made up to 50 ml using distilled water. Five ml of di-acid mixture served as blank (AOAC, 1990).

3.9.1.2 Nitrogen content

Nitrogen content was determined by micro Kjeldhal method (AOAC, 1990).

3.9.1.3 Phosphorus content

The phosphorus content in the sample was determined by Vanadomolybdophosphoric acid method using spectrophotometer at 660 nm (AOAC, 1990).

3.9.1.4 Potassium content

The potassium content in the sample was determined using flame photometer after making proper dilution.

3.9.1.5 Calcium and Magnesium

Calcium and magnesium content was determined by Versenate titration method (Cheng and Bray, 1951)

3.9.1.6 Sulphur content

The sulphur content (%) in the sample was determined by Turbidimetry method (Chesnin and Yien, 1951) using Cassia gum as stabilizing agent.

3.9.2 Proximate Composition

3.9.2.1 Moisture content

The wet sample obtained immediately after collection was oven dried at 60^{o}C for 24 h with the weight of the wet sample and the weight after drying noted. The drying was repeated until a constant weight was obtained. The moisture content was expressed in terms of loss in weight of the wet sample (AOAC, 1990).

3.9.2.2 Ash Content

1 g of the oven-dried sample in powder form was placed in crucible of known weight. This was ignited in a muffle furnace for 5 hours at 550ºC. The crucible was cooled and weighed and the ash content was expressed in terms of the oven-dried weight of the sample (AOAC, 1990).

3.9.2.3 Crude fibre

2 g of the ground sample was digested in 50 ml of 1.25 per cent H_2SO_4. The solution was boiled for 30 min. after which it was filtered and washed with hot water. The filtrate was also digested in 50 ml of 1.25 per cent NaOH. The solution was heated for 30 min., filtered and washed with hot water and oven dried. Finally the oven-dried residue was ignited in a furnace at 550^{o}C. The fibre content was measured by the weight of the left after ignition and was expressed in term of the weight of the sample before ignition (AOAC, 1990).

3.9.2.4 Crude lipid content

The lipid content was determined by extracting the fat from 5 g of the sample using petroleum ether in a soxhlet apparatus. The weight of the lipid obtained after evaporating off the petroleum ether from the extract gave the weight of the crude fat in the sample (AOAC, 1990).

Plate 4a: Trans-esterification reaction

Plate 4b: Separation of biodiesel and glycerol

Plate 4c: Crude biodiesel and glycerol

Plate 4d: Purified biodiesel

3.10 Statistical analysis

The data of the experiment were analyzed statistically following the procedure described by Gomez and Gomez (1984). The critical difference was calculated wherever the ‘F’ value was found to be significant at 5% or 1% probability level.

EXPERIMENTAL RESULTS

The experimental results obtained in the present investigation are presented in this chapter.

4.1 Growth and productivity performance of *Hydnocarpus pentandra*

The data on growth and productivity performance of *H. pentandra viz.* diameter at breast height (m), total height (m), bole height (m), crown diameter (m) and basal area (m^2/ha) are presented in Table 3 and Fig. 1.

4.1.1 Diameter at breast height (DBH)

Data on diameter on breast height expressed in 'm' are presented in Table 3.

The diameter at breast height (DBH) of *H. pentandra* varied from 0.58 m to 0.81 m. Among these experimental sites, Khurse (L_1) showed the highest DBH (0.81m); followed by Devimane (L_4, 0.76m), Shivagadde ($L_{2,}$ 0.75 m) and the lowest DBH was recorded in Hebre ($L_{3,}$ 0.58m). All locations are significantly differed from each other.

4.1.2 Total height

Data on total height expressed in 'm' are presented in Table 3.

The total height of *H. pentandra* varied from 20.59 to 22.83 m. Among the locations, the highest tree height was recorded in Khurse (L_1, 22.83 m) followed by Shivagadde ($L_{2,}$ 21.03 m), Hebre ($L_{3,}$ 20.73 m) and lowest tree height was recorded in Devimane ($L_{4,}$ 20.59 m). However, all the locations are non significant from each other.

4.1.3 Bole height

Data on bole height expressed in 'm' are presented in Table 3.

The bole height of *H. pentandra* varied from 2.31 to 2.59 m. Among the locations, L_1 showed the highest bole height of 2.59 m followed by L_2 with 2.39 m, L_4 with 2.31 m and lowest bole height of 2.25 m was recorded in L_3. There was no significant variation among the provenance with respect to bole height.

4.1.4 Crown diameter

Data on crown diameter expressed in 'm' are presented in Table 3.

The crown diameter of *H. pentandra* varied from 9.65 to 10.40 m. Among the locations, L_4 showed highest crown diameter (10.40 m) followed by L_2 (10.31 m), L_3 (10.16 m) and lowest was observed in L_1 (9.65 m). There was no significant variation among the sites with respect to crown diameter.

4.1.5 Basal area

Data on basal area expressed in 'm^2' are presented in Table 3.

The basal area of *H. pentandra* varied from 1.99 to 2.87 m^2. Among the experimental locations, L_1 showed the highest basal area of 2.87 m^2 followed by L_2 with 2.15 m^2, L_3 (2.03 m^2) and lowest basal area showed in L_4plantations (1.99 m^2). There was no significant variation among the sites with respect to basal area.

4.2 Soil properties of *Hydnocarpus pentandra* growing areas

4.2.1 Soil pH

Data on soil pH presented in Table 4.

The pH of soil under *H. pentandra* varied from 4.71 to 5.40. Among these locations, Hebre L_4 recorded the highest soil pH of 5.41 followed by L_3 (5.11), L_1 (5.02) and the lowest pH was recorded in L_2 with soil pH of 4.71. There was no significant difference in pH of experimental soils.

Table 3. Growth and productivity performance of *H. pentandra* in different provenance

Locations	Place	DBH (m)	Total height (m)	Bole height (m)	Crown diameter (m)	Basal area (m^2)
Location-1 (L_1)	Khurse	0.81	22.83	2.59	9.65	2.87
Location-2 (L_2)	Shivagadde	0.75	21.03	2.39	10.31	2.15
Location-3 (L_3)	Hebre	0.58	20.73	2.25	10.16	2.03
Location-4 (L_4)	Devimane	0.76	20.59	2.31	10.40	1.99
SE.m±		0.005	1.25	0.17	0.54	0.14
CD @5%		0.14	NS	NS	NS	NS

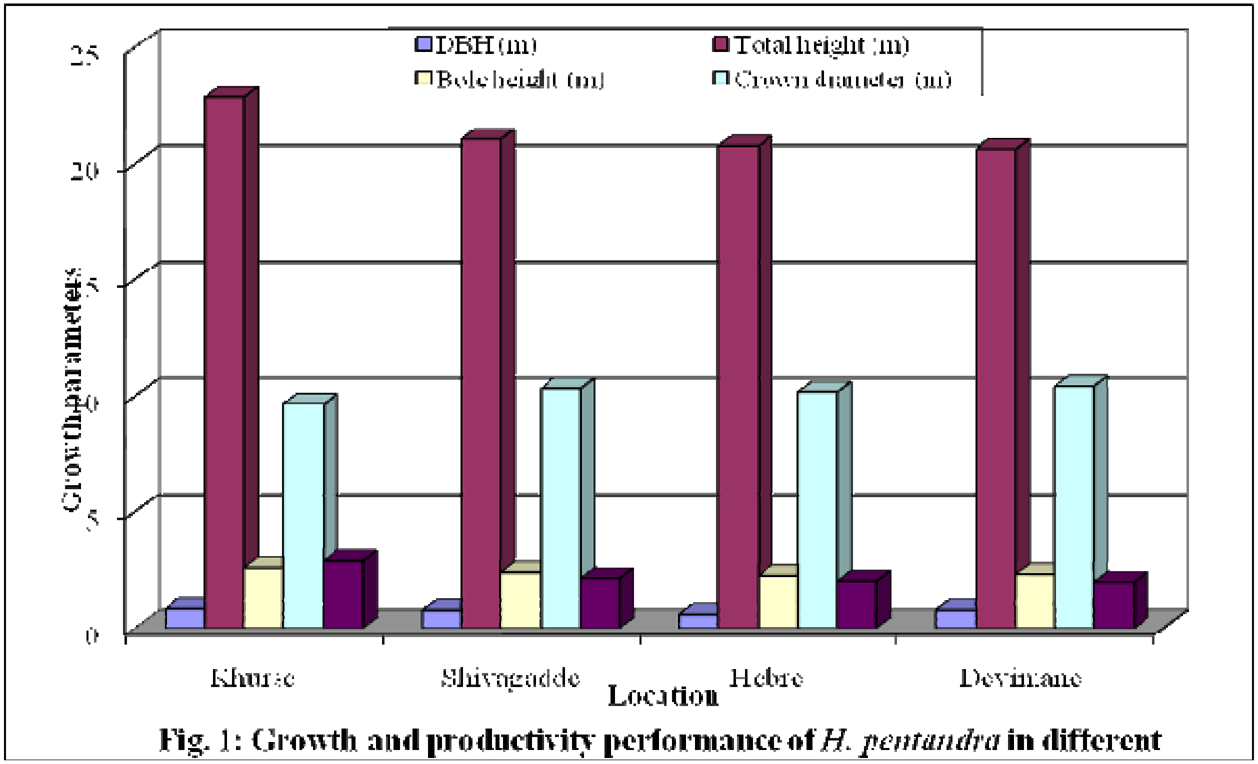

Fig. 1: Growth and productivity performance of *H. pentandra* in different site conditions

Table 4. Soil properties of experimental sites

Locations	Place	Physical properties		Chemical properties			
		pH	EC (dSm^{-1})	N	P	K	Organic Carbon (%)
				(kg/ha)			
Location-1 (L_1)	Khurse	5.02	0.05	114.27	14.03	255.33	0.54
	Control	6.50	0.04	103.23	10.25	150.56	0.38
Location-2 (L_2)	Shivagadde	4.71	0.07	122.00	11.43	265.05	0.73
	Control	6.20	0.05	116.32	9.11	158.45	0.51
Location-3 (L_3)	Hebre	5.11	0.04	121.47	13.49	255.68	0.52
	Control	6.70	0.04	114.54	10.42	165.78	0.42
Location-4 (L_4)	Devimane	5.40	0.04	113.73	9.61	245.19	0.37
	Control	6.10	0.03	104.81	7.22	147.53	0.28
SE.m±		0.24	0.011	2.13	0.85	2.11	0.06
CD @5%		NS	NS	7.37	2.96	7.32	0.22

4.2.2 Electrical conductivity

Data on electric conductivity expressed in 'dS/m' are presented in Table 4.

The electrical conductivity (EC) of soil under *H. pentandra* varied from 0.04 to 0.07 dS per m. Among the locations L_2 showed highest soil EC (0.07 dS/m) followed by L_1 (0.05 dS/m) and lowest was recorded in L_3 and L_4 (0.04 dS/m). There is no significant difference in electric conductivity among the locations.

4.2.3 Available soil nitrogen

Data on available nitrogen expressed in 'kg/ha' are presented in Table 4

The available soil nitrogen under *H. pentandra* varied from 113.73 to 122 kg per ha. Highest soil nitrogen was observed in L_2 (122 kg/ha) followed by L_3 (121.47 kg/ha), L_1 (114.27 kg/ha) and the lowest available nitrogen was found in L_4 (113.73 kg/ha). There was a significant difference between the four locations with respect to available nitrogen.

4.2.4 Available soil phosphorus

Data on available soil phosphorus expressed in 'kg/ha' are presented in Table 4.

The available soil phosphorus under *H. pentandra* varied from 9.61 to 14.03 kg per ha. Among the locations, L_1 showed the highest soil phosphorus with 14.03 kg per ha followed by L_3 (13.49 kg/ha), L_2 (11.43 kg/ha) and the lowest phosphorous was recorded in L_4 (9.61 kg/ha).There was a significant difference in the soil phosphorus content of sites.

4.2.5 Available soil potassium

Data on available potassium expressed in 'kg/ha' are presented in Table 4.

The available soil potassium under *H. pentandra* varied from 245.19 to 265.05 kg per ha. Among the locations, L_2 showed highest soil potassium (265.05 kg/ha), followed by L_3 (255.68 kg/ha), L_1 (255.33 kg/ha) and the lowest available potassium is present in L_4 (245.19 kg/ha). There was a significant difference between the sites with respect to available potassium.

4.2.6 Organic carbon

Data on organic carbon expressed in 'percent' are presented in Table 4

The organic carbon of soil under *H. pentandra* varied from 0.37 to 0.73 per cent. Among the sites, L_2 showed highest soil organic carbon with 0.73% per cent, followed by L_1 (0.54%), L_3 (0.52%) and the lowest organic carbon was observed in L_4 (0.37%). All the locations differed significantly.

4.3 Fruit characteristics of *Hydnocarpus pentandra* in different site conditions

Fruit characteristics of *Hydnocarpus pentandra* are presented in Table 5 and Fig. 2.

4.3.1 Fruit length

Data on fruit length expressed in 'cm' are presented in Table 5.

The fruit length of *H. pentandra* varied from 5.76 to 6.97 cm. Fruit length was significantly influenced by different locations. L_4 recorded the highest fruit length of 6.97 cm followed by L_3 (6.59 cm) and L_2 (6.13 cm) whereas, the lowest fruit length was recorded in L_1 (5.76 cm).

Similarly, fruit length was significantly influenced by fruit sizes, F_1 (large sized fruits) recorded the highest fruit length (7.64 cm) when compared to F_2 (medium sized fruits, 7.19 cm) and the lowest fruit length was recorded in F_3 (small sized fruits, 4.26 cm).

Interaction effect of location and fruit sizes also differed significantly with respect to fruit length. A treatment combination of L_4F_1 recorded the highest fruit length of 8.48 cm and the lowest fruit length of 4.00 cm was recorded with L_1F_3.

Table 5. Fruit parameters of *Hydnocarpus pentandra* in different provenance

Location	Fruit length (cm)				Fruit width (cm)				Fruit weight (g)			
	F_1	F_2	F_3	Mean	F_1	F_2	F_3	Mean	F_1	F_2	F_3	Mean
L_1	6.90	6.39	4.00	**5.76**	6.90	4.18	4.92	**5.33**	22.50	23.54	22.00	**22.68**
L_2	7.29	6.90	4.18	**6.13**	7.53	4.35	5.63	**5.84**	24.67	22.68	22.13	**23.16**
L_3	7.89	7.53	4.35	**6.59**	7.93	4.49	5.24	**5.89**	23.17	20.00	22.71	**21.96**
L_4	8.48	7.93	4.49	**6.97**	4.00	5.42	4.24	**4.55**	26.00	25.28	21.00	**24.09**
Mean	**7.64**	**7.19**	**4.26**		**6.59**	**4.61**	**5.00**		**24.08**	**22.88**	**21.96**	
	S.Em±		CD @5%		S.Em±		CD @5%		S.Em±		CD @5%	
Location (L)	0.14		0.33		0.30		0.74		0.73		NS	
Fruit size (F)	0.10		0.20		0.22		0.46		0.60		1.26	
L x F	0.19		0.41		0.44		0.93		1.19		2.53	

Note: L_1- Khurse, L_2-Shivagadde, L_3- Hebre, L_4- Devimane, F_1-Large fruit, F_2-Medium fruit and F_3-Small fruit

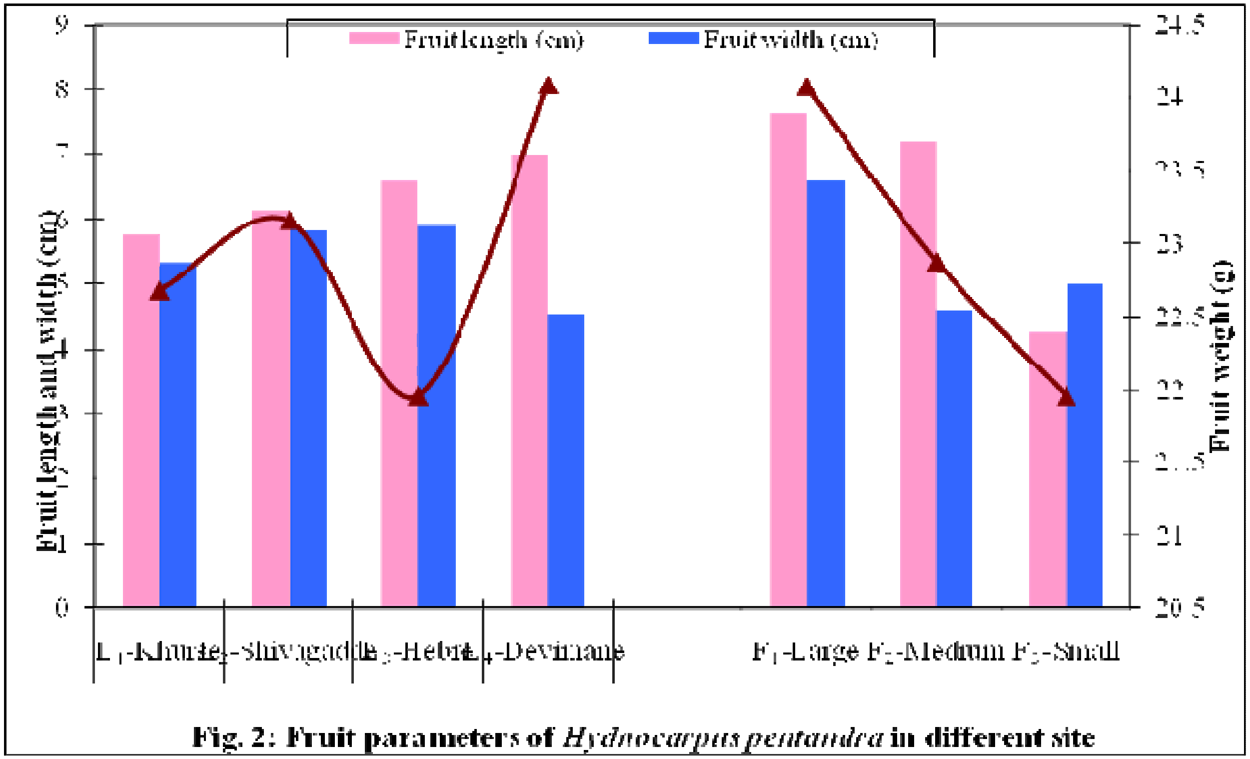

Fig. 2: Fruit parameters of *Hydnocarpus pentandra* in different site conditions

4.3.2 Fruit width

Data on fruit width expressed in 'cm' are presented in Table 5.

The fruit width was significantly influenced by different locations. L_3 recorded the highest fruit width (5.89 cm) followed by L_2 (5.84 cm) and L_1 (5.33 cm) whereas, the lowest fruit width was recorded in L_4 (4.55 cm).

Similarly, fruit width was significantly influenced by fruit sizes, F_1 recorded highest fruit width of 6.59 cm when compared to F_3 (5.00 cm) and lowest fruit width was recorded in F_2 (4.61 cm).

Interaction effect of location and fruit sizes also differed significantly with respect to fruit width. A treatment combination of L_3F_1 recorded the highest fruit width (7.93 cm) and the lowest fruit width of 4.18 cm was recorded with L_1F_2. L_4 F_3 (4.24 cm) combination was on par with L_1 F_2 (4.18 cm).

4.3.2 Fruit weight

Data on fruit weight expressed in 'g' are presented in Table 5

Fruit weight was significantly influenced by different locations. L_4 recorded the highest fruit weight of 24.09 g followed by L_2 (23.16 g) and L_1 (22.68 g) whereas, the lowest fruit weight was recorded in L_3 (21.96 g).

Fruit weight was also significantly influenced by fruit sizes, F_1 recorded the highest fruit weight of 24.08g when compared to F_2 (22.88 g) and the lowest fruit weight was recorded in F_3 (21.96 g).

Interaction effect of location and fruit sizes also differed significantly with respect to fruit weight. A treatment combination of L_4F_1 recorded the highest fruit weight of 26.0 g and the lowest fruit weight of 20.00 g was recorded with combination of L_3F_2. L_3F_2 (20.0 g) and L_4F_3 (21.0 g) combination was on par with each other.

4.4 Seed characteristics of *Hydnocarpus pentandra*

Seed characteristics of *Hydnocarpus pentandra* presented in Table 6 and Fig. 3

4.4.1 Seed length

Data on seed length expressed in 'cm'.

The seed length was not significantly influenced by different locations. However, seed length was significantly influenced by fruit sizes. F_1 recorded the highest seed length (2.94 cm) when compared to F_3 (2.89 cm) and F_2 (2.88 cm). However, there is no significant variation between F_2 and $F_{3.}$

Interaction effects between location and fruit sizes were also differed significantly with respect to seed length. A treatment combination of L_3F_1 recorded the highest seed length of 2.96 cm and the lowest seed length of 2.85 cm was recorded with $L_1F_2$and L_2F_3. L_3F_1 (2.96 cm)combination was on par with L_4F_1 and L_1F_3 (2.95 cm).

4.4.2 Seed width

Data on seed width expressed in cm are presented in Table 6

The seed width was significantly influenced by different locations. L_4 recorded the highest seed width (1.80 cm), followed by L_2 (1.54 cm), L_1 (1.52 cm) and the lowest seed width was recorded in L_3 (1.30 cm).

Similarly, seed width was significantly influenced by fruit sizes. F_3 (large sized fruits) recorded the highest seed width (1.79 cm) when compared to F_2 (1.63 cm) and the lowest seed width was recorded in F_1 (1.19 cm).

Interaction effect of location and fruit sizes also differed significantly with respect to seed width. A treatment combination of L_4F_2 recorded the highest seed width (1.92 cm) and the lowest seed width (0.89 cm) was recorded with combination of L_3F_2. L_1F_1 (0.90 cm) and L_2F_1 (0.93 cm)combination was on par with L_3F_2 (0.89 cm).

Table 6. Seed parameters of *Hydnocarpus pentandra* in different provenance

Locations	Seed length (cm)				Seed width (cm)				Total perimeter (cm^2)				Total area (cm^2)				100 Seed weight (g)			
	F_1	**F_2**	**F_3**	**Mean**	**F_1**	**F_2**	**F_3**	**Mean**	**F_1**	**F_2**	**F_3**	**Mean**	**F_1**	**F_2**	**F_3**	**Mean**	**F_1**	**F_2**	**F_3**	**Mean**
L_1	2.91	2.85	2.95	**2.90**	0.90	1.85	1.80	**1.52**	5.56	4.18	5.84	**5.19**	2.91	1.29	1.75	**1.98**	109.33	105.07	101.40	**105.27**
L_2	2.93	2.87	2.85	**2.88**	0.93	1.87	1.82	**1.54**	5.84	4.35	5.84	**5.34**	2.93	2.87	1.77	**2.52**	115.17	106.57	103.60	**108.44**
L_3	2.96	2.89	2.87	**2.91**	1.29	0.89	1.70	**1.30**	5.70	4.49	5.22	**5.14**	2.96	2.89	1.79	**2.55**	119.00	110.43	106.50	**111.98**
L_4	2.95	2.92	2.89	**2.92**	1.65	1.92	1.84	**1.80**	4.00	5.18	4.20	**4.46**	2.95	2.92	1.79	**2.55**	123.33	112.83	108.67	**114.94**
Mean	**2.94**	**2.88**	**2.89**		**1.19**	**1.63**	**1.79**		**5.28**	**4.55**	**5.28**		**2.94**	**2.49**	**1.77**		**116.71**	**108.73**	**105.04**	
	S.Em±		CD @5%		S.Em±		CD @5%		S.Em±		CD @5%		S.Em±		CD @5%		S.Em±		CD @5%	
Location (L)	NS		0.05		0.10		0.25		0.21		0.51		0.01		0.03		1.37		3.34	
Fruit size (F)	0.01		0.03		0.10		0.20		0.08		0.18		0.01		0.03		1.00		2.12	
L x F	0.02		0.05		0.19		0.41		0.17		0.35		0.03		0.06		2.00		4.24	

Note: L_1- Khurse, L_2-Shivagadde, L_3- Hebre, L_4- Devimane, F_1-Large fruit, F_2-Medium fruit and F_3-Small fruit

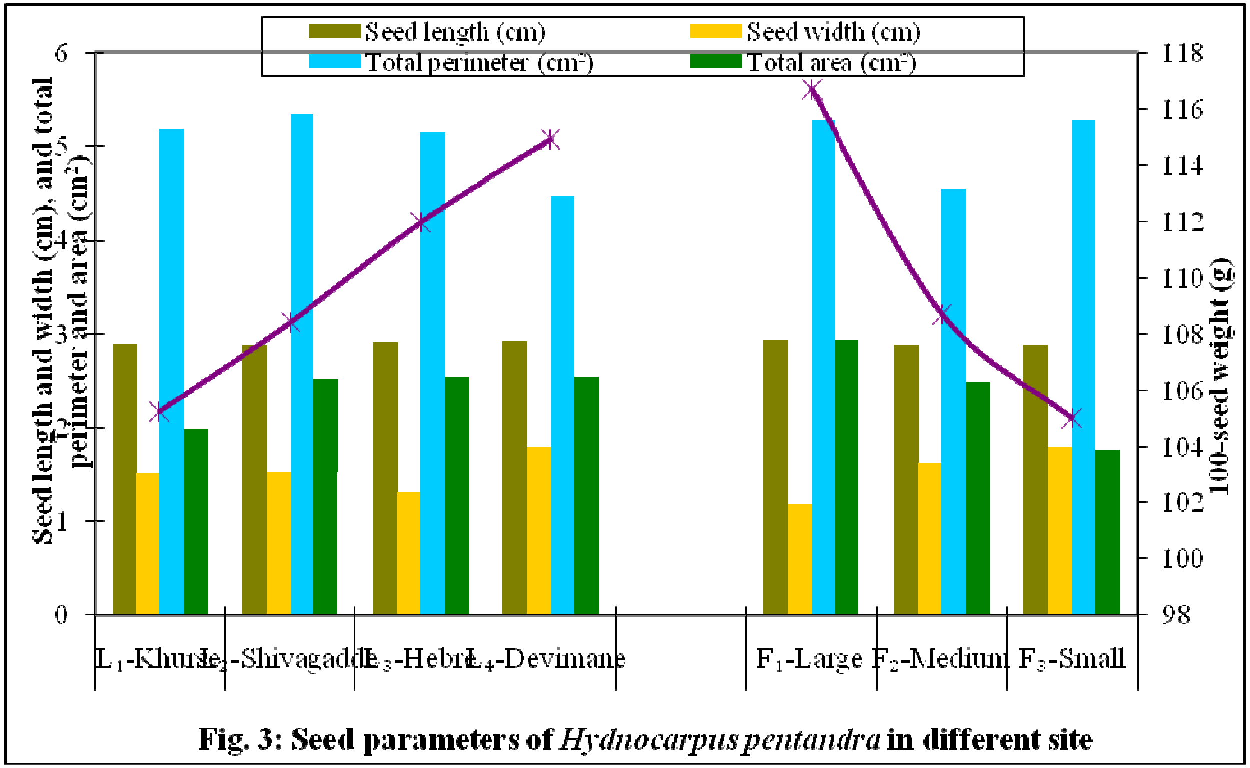

Fig. 3: Seed parameters of *Hydnocarpus pentandra* in different site conditions

4.4.3 Total perimeter of the seed

Data on seed total perimeter expressed in ‘cm^{2}’ are presented in Table 6

The seed total perimeter was significantly influenced by different locations. L_2 recorded thehighest seed perimeter (5.34 cm^2) followed by L_1 (5.19 cm^2) and L_3 (5.14 cm^2) whereas, the lowest perimeter was recorded in L_4 (4.46 cm^2).

Similarly, seed total perimeter was significantly influenced by fruit sizes. F_1 and F_3 recorded the highest perimeter (5.28 cm^2) and lowest was in F_2 (4.55 cm^2).

Interaction effect of location and fruit sizes also differed significantly with respect to total perimeter of seed. A treatment combination of L_1F_1, L_2F_3 and L_2F_1 recorded the highest seed total perimeter (5.84 cm^2) and the lowest perimeter (4.00 cm^2) was recorded with L_4F_1.

4.4.4 Total Area of Seed

Data on total area of seed expressed in ‘cm^{2}’ are presented in Table 6.

Total area of seed was significantly influenced by different locations L_3 and L_4 recorded the highest area (2.55 cm^2) followed by L_2 (2.52 cm^2) whereas, the lowest total area of seed was recorded in L_1 (1.98 cm^2).

Total area of seed was significantly influenced by fruit sizes. F_1 recorded the highest area (2.94 cm^2) followed by F_2 (2.49 cm^2) and the lowest total area was recorded in F_3 (1.77cm^2).

Interaction effect of location and fruit sizes also differed significantly with respect to total area of seed. A treatment combination of L_3F_1 recorded the highest total area (2.96 cm^2) and the lowest total area (1.29 cm^2) was recorded with L_1F_2. L_2F_1 (2.93 cm^2) and L_3F_1 (2.95 cm^2) combination are on par with L_3F_1.

4.4.5 100 seed weight

Data on 100 seed weight expressed in ‘g’ are presented in Table 6.

The 100 seed weight was significantly influenced by different locations. L_4 recorded the highest seed weight (114.94 g) followed by L_3 (111.98 g), L_2 (108.44 g) whereas, the lowest seed weight was recorded in L_1 (105.27g).

100 seed weight was significantly influenced by fruit sizes. F_1 recorded the highest seed weight (116.71g), followed by F_2 (108.73 g) and the lowest seed weight was recorded in F_3 (105.04 g).

Interaction effect of location and fruit sizes also differed significantly with respect to seed weight. A treatment combination of L_4F_1 recorded the highest seed weight (123.33 g) and the lowest seed weight (101.40 g) was recorded with L_1F_3. L_1F_2 (105.07g) combination is in on par with L_1F_3.

4.5 Seed kernel and shell ratio of *Hydnocarpus pentandra*

Seed kernel and shell ratio of *Hydnocarpus pentandra*are presented in Table 7 and Fig. 4

4.5.1 Seed Weight

Data on seed weight expressed in ‘g’ are presented in Table 7.

The single seed weight was significantly influenced by different locations. L_4 recorded the highest seed weight (2.22 g) followed by L_3 (2.02 g) and L_2 (1.94 g) whereas, the lowest seed weight was recorded in L_1 (1.88 g).

Seed weight was significantly influenced by fruit sizes. F_1 recorded the highest weight (2.11 g) when compared to F_3 (1.99 g) and the lowest was recorded in F_2 (1.95 g).

Interaction effect of location and fruit sizes also differed significantly with respect to seed weight. A treatment combination of L_4F_3 recorded the highest weight (2.38 g) and the lowest was recorded with L_1F_3 (1.81g). L_1F_1 (1.97 g), L_1F_2 (1.88 g) and L_2F_2 (1.91 g) combinations are on per with L_1F_3.

Table 7. Seed kernel and shell ratio of *Hydnocarpus pentandra* in different site conditions

Locations	Seed weight (g)				Kernel Weight (g)				Shell weight (g)				Kernel fraction (%)				Shell fraction (%)			
	F_1	**F_2**	**F_3**	**Mean**	**F_1**	**F_2**	**F_3**	**Mean**	**F_1**	**F_2**	**F_3**	**Mean**	**F_1**	**F_2**	**F_3**	**Mean**	**F_1**	**F_2**	**F_3**	**Mean**
L_1	1.97	1.88	1.81	**1.88**	1.09	1.05	1.01	**1.05**	0.87	0.83	0.79	**0.83**	55.73	55.98	56.29	**56.00**	44.27	44.02	43.71	**44.00**
L_2	2.08	1.91	1.85	**1.94**	1.15	1.06	1.03	**1.08**	0.93	0.84	0.81	**0.86**	55.36	55.90	56.01	**55.75**	44.64	44.10	43.99	**44.25**
L_3	2.16	1.99	1.91	**2.02**	1.19	1.10	1.06	**1.12**	0.97	0.88	0.84	**0.90**	55.21	55.68	55.82	**55.57**	44.79	44.32	44.18	**44.43**
L_4	2.25	2.03	2.38	**2.22**	1.23	1.13	1.30	**1.22**	1.01	0.91	1.08	**1.00**	54.96	55.49	54.64	**55.03**	45.04	44.51	45.36	**44.97**
Mean	**2.11**	**1.95**	**1.99**		**1.17**	**1.09**	**1.10**		**0.95**	**0.87**	**0.88**		**55.31**	**55.76**	**55.69**		**44.69**	**44.24**	**44.31**	
	S.Em±		CD @5%		S.Em±		CD @5%		S.Em±		CD @5%		S.Em±		CD @5%		S.Em±		CD @5%	
Location (L)	0.04		0.14		0.02		0.07		0.02		0.07		0.26		NS		0.26		NS	
Fruit size (F)	0.03		0.09		0.02		0.05		0.02		0.05		0.12		0.34		0.12		0.34	
L x F	0.06		0.19		0.03		0.09		0.03		0.09		0.23		NS		0.23		NS	

Note: L_1- Khurse, L_2-Shivagadde, L_3- Hebre, L_4- Devimane, F_1-Large fruit, F_2-Medium fruit and F_3-Small fruit

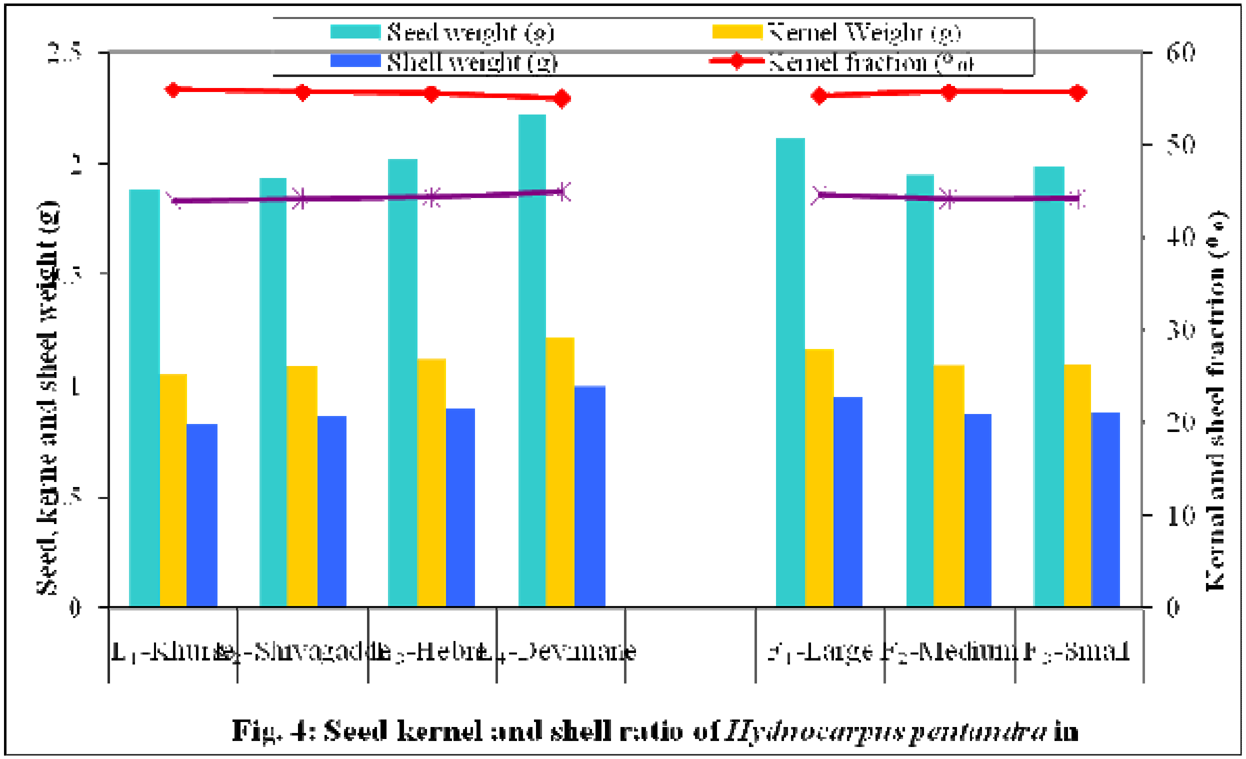

Fig. 4: Seed kernel and shell ratio of *Hydnocarpus pentandra* in different site conditions

4.5.2 Kernel Weight

Data on kernel weight expressed in 'g' are presented in Table 7.

The single kernel weight was significantly influenced by different locations. L_4 recorded highest kernel weight (1.22 g) followed by L_3 (1.12 g) and L_2 (1.08 g) whereas, lowest kernel weight was recorded in L_1 (1.05 g).

Kernel weight was significantly influenced by fruit sizes. F_1 recorded the highest kernel weight (1.17g), followed by F_3(1.10 g) and the lowest weight per kernel was recorded in F_2 (1.09 g).

Interaction effect of location and fruit sizes also differed significantly with respect to kernel weight. A treatment combination of L_4F_3 recorded the highest weight of 1.30 g and the lowest was recorded with L_1F_3 (1.01 g). L_4F_1 (1.23 g) combination was on par with L_4F_3 and L_1F_1 (1.09 g) combination was on par with L_1F_3.

4.5.3 Shell weight

Data on weight of the shell expressed in 'g' presented in Table 7.

The shell weight was significantly influenced by different locations. L_4 recorded the highest shell weight (1.00 g) followed by L_3 (0.90 g) and L_2 (0.86 g) whereas, the lowest weight of the shell was recorded in L_1 (0.83g).

Shell weight was significantly influenced by fruit sizes. F_1 recorded the highest shell weight (0.95 g) when compared to F_3 (0.88 g) and lowest shell weight was recorded in F_2 (0.87 g).

Interaction effect of location and fruit sizes also differed significantly with respect to shell weight. A treatment combination of L_4F_3 recorded the highest weight of the shell (1.08 g) and the lowest weight was recorded with L_1F_3 (0.79 g). L_4F_1 (1.01g) combination was on par with L_4F_3 (1.08 g). L_1F_2 (0.83 g), L_2F_2 (0.84 g) and L_3F_2 (0.88 g) are on par with L_1F_3.

4.5.4 Kernel fraction

Data on kernel fraction expressed in 'percentage' are presented in Table 7.

The effect of different locations and interaction effect of location and fruit size was not significant with respect to kernel fraction. Kernel fraction was significantly influenced by fruit sizes. F_2 recorded the highest kernel fraction (55.76%) when compared to F_3 (55.69%) and the lowest kernel fraction was recorded in F_1 (55.31%).

4.5.5 Shell fraction

Data on shell fraction expressed in 'percentage' are presented in Table 7.

The effect of different locations and interaction effect of location and fruit size was not significant with respect to shell fraction. The shell fraction was significantly influenced by fruit sizes. F_1 recorded the highest shell fraction (44.69%) when compared to F_3 (44.31%) and the lowest shell fraction was recorded in F_2 (44.24%).

4.6 Seed kernel parameters *Hydnocarpus pentandra*

Seed kernel parameters of *Hydnocarpus pentandra*presented in Table 8 and Fig. 5.

4.6.1 Kernel length

Data on length of the kernel expressed in 'cm' are presented in Table 8.

The kernel length was significantly influenced by different locations. L_3 and L_4 recorded the highest length (1.87 cm) each followed by L_2 (1.73 cm) and the lowest kernel length was recorded in L_1 (1.30 cm).

The Kernel length was significantly influenced by fruit sizes. F_1 recorded the highest kernel length (2.15 cm) when compared to F_2 (1.70 cm) and the lowest kernel length was recorded in F_3 (1.23 cm).

Interaction effect of location and fruit sizes also differed significantly with respect to kernel length. A treatment combination of L_3F_1 recorded the highest kernel length (2.17 cm) and the lowest kernel length was recorded with L_1F_2 (0.83 cm). L_2F_2 (2.08 cm) andL_3F_2 (2.10 cm) combinations are on par with L_3F_1. Similarly L_1F_3 (0.96 cm) and L_2F_3 (0.98 cm) combinations are on par with L_1F_2.

Table 8. Seed kernel parameters of *Hydnocarpus pentandra* in different site conditions

Locations	Length (cm)				Width (cm)				Perimeter (cm^2)				Area (cm^2)			
	F_1	F_2	F_3	Mean	F_1	F_2	F_3	Mean	F_1	F_2	F_3	Mean	F_1	F_2	F_3	Mean
L_1	2.12	0.83	0.96	**1.30**	1.70	0.41	0.54	**0.88**	4.56	3.18	4.84	**4.19**	1.91	0.62	0.75	**1.09**
L_2	2.14	2.08	0.98	**1.73**	1.72	1.66	0.56	**1.31**	4.84	3.35	4.84	**4.34**	1.93	1.87	0.77	**1.52**
L_3	2.17	2.10	1.33	**1.87**	1.75	1.68	0.91	**1.45**	4.70	3.49	3.22	**3.81**	1.96	1.89	1.12	**1.66**
L_4	2.16	1.79	1.67	**1.87**	1.74	1.37	1.25	**1.45**	3.00	4.18	3.20	**3.46**	1.95	1.58	1.46	**1.66**
Mean	**2.15**	**1.70**	**1.23**		**1.73**	**1.28**	**0.81**		**4.28**	**3.55**	**4.03**		**1.94**	**1.49**	**1.02**	
	S.Em±		CD @ 5%		S.Em±		CD @ 5%		S.Em±		CD @ 5%		S.Em±		CD @ 5%	
Location (L)	0.09		0.22		0.09		0.22		0.21		0.51		0.09		0.22	
Fruit size (F)	0.14		0.35		0.14		0.31		0.08		0.18		0.14		0.31	
L x F	0.29		0.61		0.29		0.61		0.17		0.35		0.29		0.61	

Note: L_1- Khurse, L_2-Shivagadde, L_3- Hebre, L_4- Devimane, F_1-Large fruit, F_2-Medium fruit and F_3-Small fruit

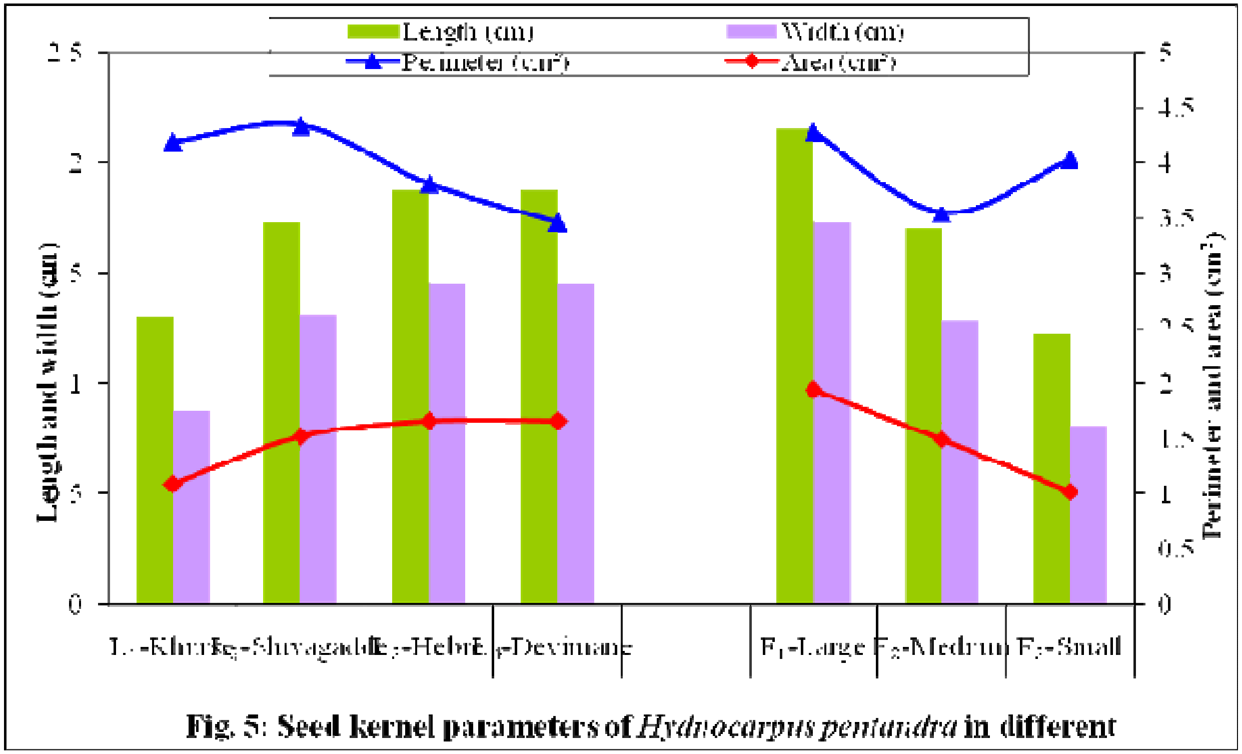

Fig. 5: Seed kernel parameters of *Hydnocarpus pentandra* in different site conditions

4.6.2 Kernel width

Data on width of the kernel expressed in cm are presented in Table 8.

The kernel width was significantly influenced by different locations. L_3 and L_4 recorded highest width (1.45 cm) each followed by L_2 (1.31 cm) whereas, lowest kernel width was recorded in L_1 (0.88 cm).

The Kernel width was significantly influenced by fruit sizes. F_1 recorded highest kernel width (1.73 cm) followed by F_2 (1.28 cm) and lowest kernel width was recorded in F_3 (0.81 cm).

Interaction effect of location and fruit sizes also differed significantly with respect to kernel width. A treatment combination of L_3F_1 recorded the highest kernel width (1.75 cm) and the lowest kernel width was recorded with L_1F_2 (0.41 cm). L_2F_2 (1.66 cm), L_3F_2 (1.68 cm) combinations are on par with L_3F_1. Similarly L_1F_3 (0.54 cm) and L_2F_3 (0.56 cm) combinations are on par with L_1F_2.

4.6.3 Kernel perimeter

Data on perimeter of the kernel expressed in 'cm^2' are presented in Table 8.

The kernel perimeter was significantly influenced by different locations. L_2 recorded the highest kernel perimeter (4.34 cm^2) followed by L_1 (4.19 cm^2) and L_3 (3.81 cm^2) whereas, the lowest kernel perimeter was recorded in L_4 (3.46 cm^2).

The Kernel perimeter was significantly influenced by fruit sizes. F_1 recorded the highest kernel perimeter (4.28 cm^2) followed by F_3 (4.03 cm^2) and the lowest kernel perimeter was recorded in F_2 (3.55 cm^2).

Interaction effect of location and fruit sizes also differed significantly with respect to kernel perimeter. A treatment combination of L_2F_1 and L_2F_3 recorded the highest kernel perimeter of 4.84 cm^2 each. Lowest kernel perimeter was recorded with L_4F_1 (3.00 cm^2). L_2F_2 (3.35 cm^2) combination is on par with L_4F_1.

4.6.4 Kernel area

Data on kernel area expressed in 'cm^2' are presented in Table 8.

The kernel area was significantly influenced by different locations. L_3 and L_4 recorded the highest area of 1.66 cm^2 each followed by L_2 with 1.52 cm^2. The lowest kernel total area was recorded in L_4 (1.09 cm^2).

The kernel area was significantly influenced by fruit sizes. F_1 recorded the highest kernel total area (1.94 cm^2) followed by F_2 (1.49 cm^2) and the lowest kernel total area was recorded in F_3 (1.02 cm^2).

Interaction effect of location and fruit sizes also differed significantly with respect to kernel area. A treatment combination of L_3F_1 recorded the highest kernel area (1.96 cm^2) and the lowest kernel area was recorded with L_1F_2 (0.62 cm^2). All the treatment combinations in all the location with respect to F_1 are on par with each other. Similarly L_1F_3 (0.75 cm^2), L_2F_3 (0.77 cm^2) and L_3F_3 (1.12 cm^2) combinations are on par with L_1F_2.

4.7 Oil yield from seed kernel of *Hydnocarpus pentandra*

Data on oil yield expressed in 'percentage' are presented in Table 9 and Fig. 6

The kernel oil yield was significantly influenced by different locations. L_3 recorded the highest oil yield (49.49%) followed by L_4 (43.76 %) and L_2 (41.41 %) whereas, the lowest oil yield was recorded in L_1 (32.35%).

The kernel oil yield was significantly influenced by fruit size. F_2 recorded the highest oil yield (44.02%) when compared to F_1 (41.25%) and the lowest oil yield was recorded in F_3 (39.99%).

Interaction effect of location and fruit sizes also differed significantly with respect to oil yield. A treatment combination of L_3F_2 recorded the highest oil yield (60.13%) and the lowest oil yield was recorded with L_1F_1 (30.60 %). L_1F_2 (32.65%) and L_1F_3 (33.79%) combination are on par with L_1F_1.

Table 9. Oil yield from seed kernel of *Hydnocarpus pentandra* in different site conditions

Locations	**Oil yield (%)**			
	F_1	**F_2**	**F_3**	**Mean**
L_1	30.60	32.65	33.79	**32.35**
L_2	46.29	41.03	36.89	**41.41**
L_3	42.73	60.13	45.60	**49.49**
L_4	45.35	42.25	43.68	**43.76**
Mean	**41.25**	**44.02**	**39.99**	
	S.Em±		CD @ 1%	
Location (L)	1.26		4.68	
Fruit size (F)	0.80		2.35	
L x F	1.61		4.70	

Note: L_1- Khurse, L_2-Shivagadde, L_3- Hebre, L_4- Devimane, F_1-Large fruit, F_2-Medium fruit and F_3-Small fruit

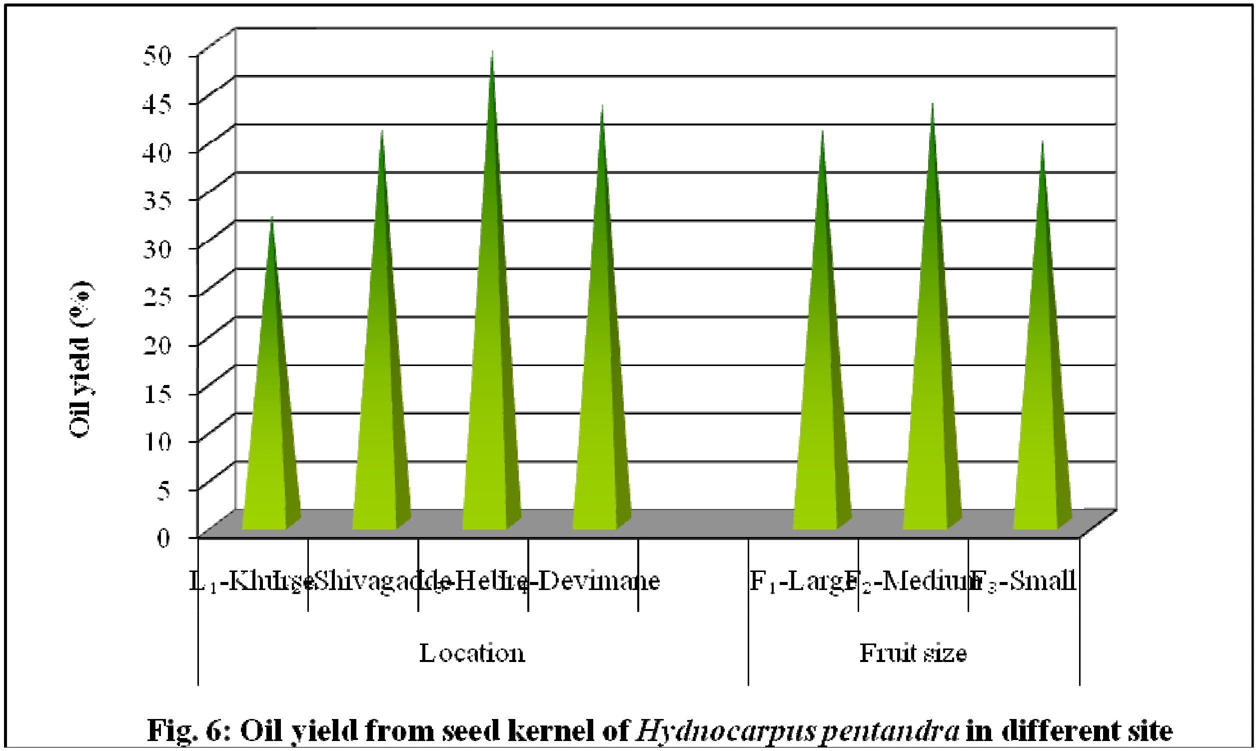

Fig. 6: Oil yield from seed kernel of *Hydnocarpus pentandra* in different site conditions

Table 10. Organoleptic and Physicao-chemical properties of *Hydnocarpus pentandra* seed kernel oil

Parameter	Value
Organoleptic properties	
Colour	Golden yellow
Odour	Agreeable
Physical properties	
Kinematic viscosity @ 40°C (cSt)	7.146
Density @ 30°C (kg/m^3)	950
Acidity (mg KOH/g)	1.040
FFA (as oleic acid)	0.525
Specific gravity at 25ºC	0.950
Saponification value	215.06
Iodine value	92.30

Table 11. GC-MS report of seed oil of *Hydnocarpus pentandra* from Khurse site

Fatty acid	Molecular formula	Carbon nomenclature	Molecular weight	% Area	Retention Time
Palmitic acid	$C_{16}H_{32}O_2$	C16:0	256	2.26	9.880
Stearic acid	$C_{18}H_{36}O_2$	C18:0	280	2.56	10.722
Hydnocarpic acid	$C_{16}H_{28}O_2$	C16:1	280.2	7.94	11.091
Chaulmoogric acid	$C_{18}H_{32}O_2$	C18:1	252.2	57.07	10.257
Gorlic acid	$C_{18}H_{30}O_2$	C18:2	262	30.17	11.174

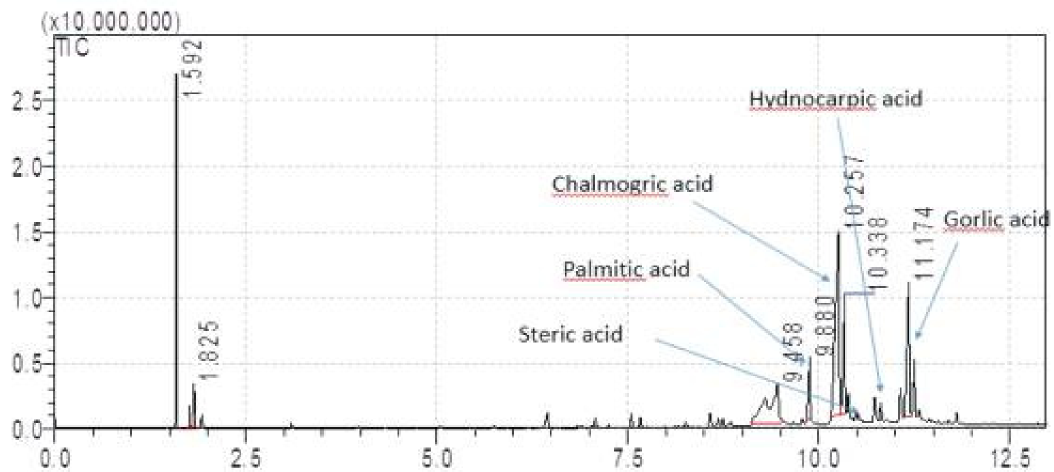

Fig 7: GC-MS chromatogram of seed oil of *Hydnocarpus pentandra* oil from Khurse (L_1)

Table 12. GC-MS report of seed oil of *Hydnocarpus pentandra* from Shivagadde site

Fatty acid Name	Molecular formula	Carbon nomenclature	Molecular weight	% Area	Retention Time
Palmitic acid	$C_{16}H_{32}O_2$	C16:0	256	3.69	9.859
Stearic acid	$C_{18}H_{36}O_2$	C18:0	280	4.06	10.724
Hydnocarpic acid	$C_{16}H_{28}O_2$	C16:1	280.2	3.22	11.082
Chaulmoogric acid	$C_{18}H_{32}O_2$	C18:1	252.2	36.90	10.233
Gorlic acid	$C_{18}H_{30}O_2$	C18:2	262	16.17	11.152

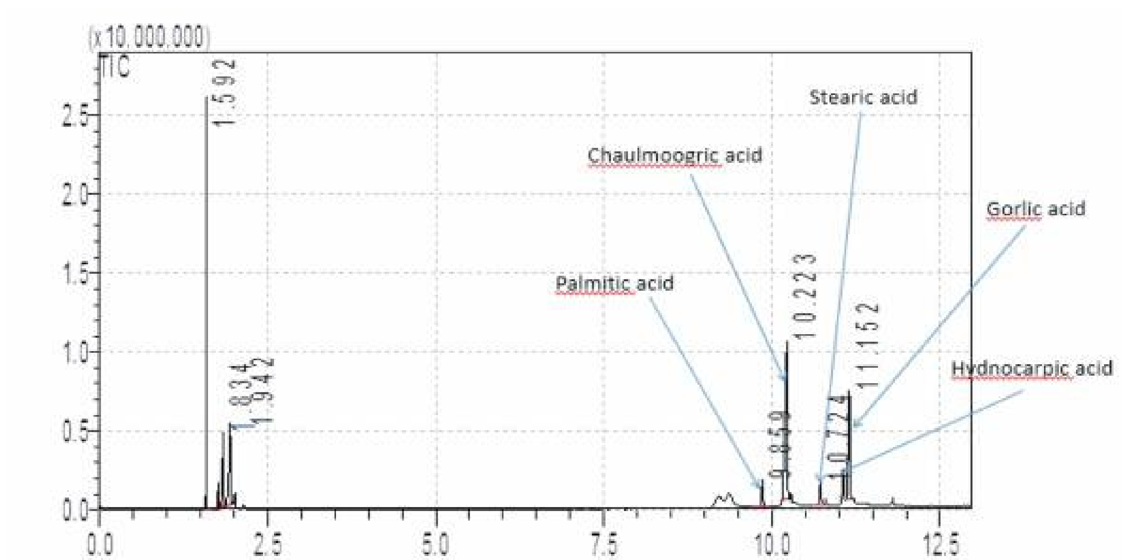

Fig 8: GC-MS chromatogram of seed oil of *Hydnocarpus pentandra* oil from shivagadde (L_2)

Table 13. GC-MS report of seed oil of *Hydnocarpus pentandra* from Hebre site

Fatty acid	Molecular formula	Carbon nomenclature	Molecular weight	% Area	Retention Time
Palmitic acid	$C_{16}H_{32}O_2$	C16:0	256	4.20	9.873
Stearic acid	$C_{18}H_{36}O_2$	C18:0	280	4.51	10.748
Hydnocarpic acid	$C_{16}H_{28}O_2$	C16:1	280.2	5.85	11.079
Chaulmoogric acid	$C_{18}H_{32}O_2$	C18:1	252.2	56.03	10.282
Gorlic acid	$C_{18}H_{30}O_2$	C18:2	262	20.82	11.189

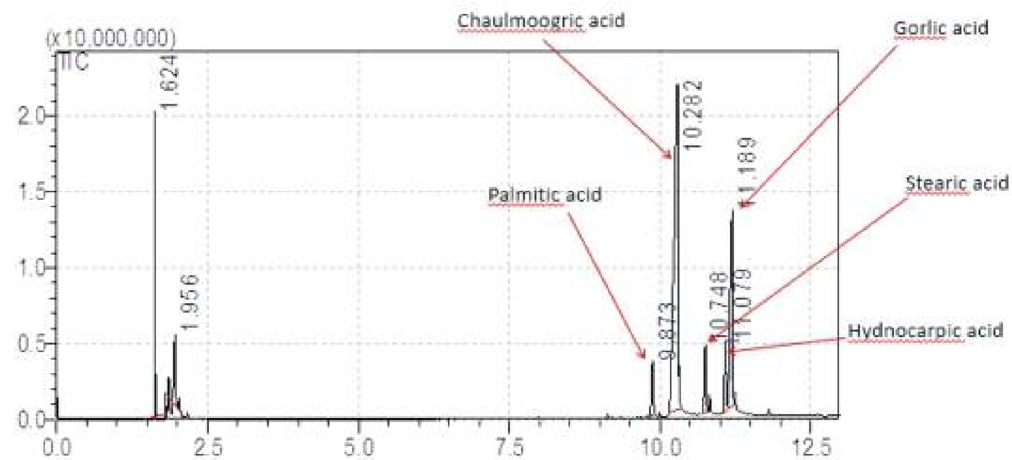

Fig 9: GC-MS chromatogram of seed oil of *Hydnocarpus pentandra* oil from Hebre (L_3)

Table 14. GC-MS report of seed oil of *Hydnocarpus pentandra* from Devimane site

Fatty acid	Molecular formula	Carbon nomenclature	Molecular weight	% Area	Retention Time
Palmitic acid	$C_{16}H_{32}O_2$	C16:0	256	3.48	9.848
Stearic acid	$C_{18}H_{36}O_2$	C18:0	280	5.13	10.716
Hydnocarpic acid	$C_{16}H_{28}O_2$	C16:1	280.2	3.45	11.070
Chaulmoogric acid	$C_{18}H_{32}O_2$	C18:1	252.2	24.57	10.197
Gorlic acid	$C_{18}H_{30}O_2$	C18:2	262	12.02	11.135

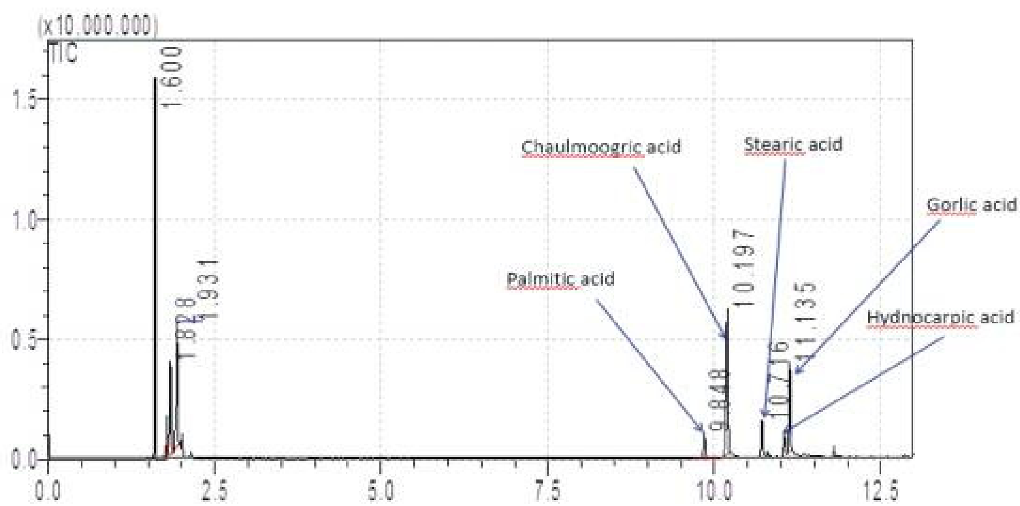

Fig 10: GC-MS chromatogram of seed oil of *Hydnocarpus pentandra* oil from Devimane (L_4)

Table 15. Percentage content of fatty acids in seed oil of *Hydnocarpus pentandra* from Agro-climatic Zone 9 of Karnataka

Fatty acids	Khurse (L_1)	Shivagadde (L_2)	Hebre (L_3)	Devimane ($L4_1$)
Palmitic acid	2.26	3.69	4.20	3.48
Stearic acid	2.56	4.06	4.51	5.13
Hydnocarpic acid	7.94	3.22	5.85	3.45
Chaulmoogric acid	57.07	36.90	56.03	24.57
Gorlic acid	30.17	16.17	20.82	12.02

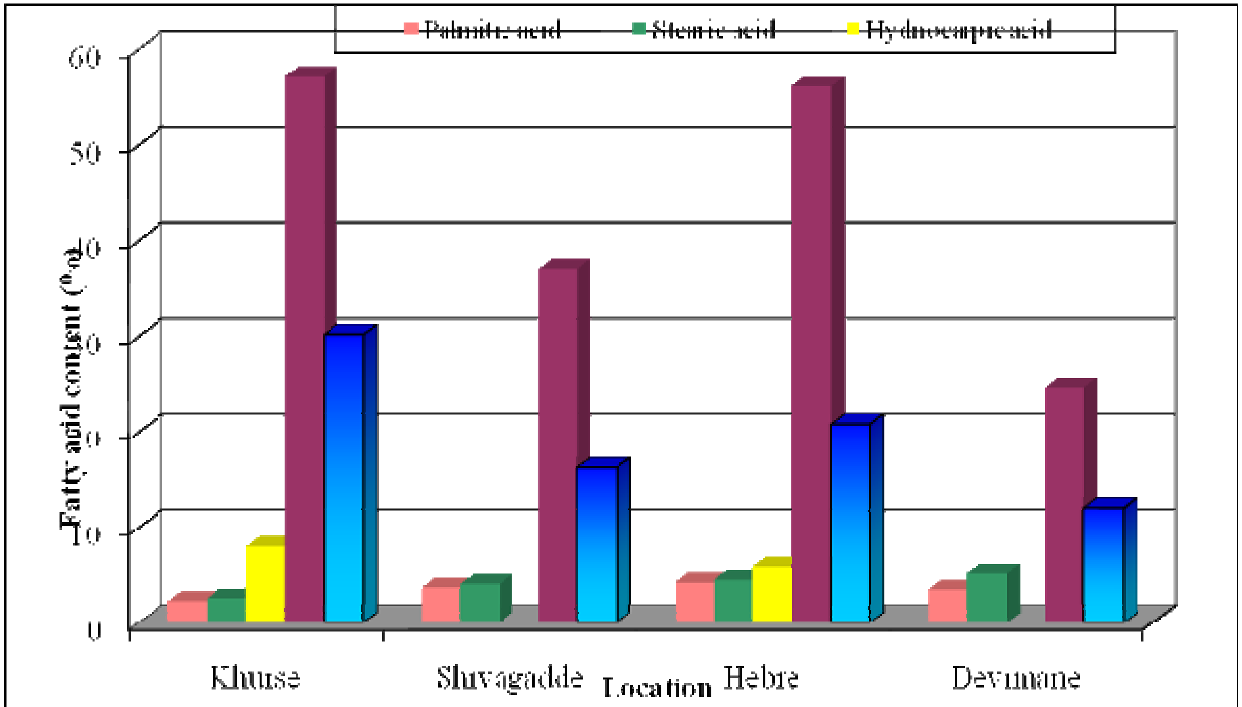

Fig. 11: Percentage content of fatty acids in seed oil of *Hydnocarpus pentandra*

Fig. 11: Percentage content of fatty acids in seed oil of *Hydnocarpus pentandra* from Agro-climatic Zone 9 of Karnataka

Table 16. Phisico-chemical properties of *Hydnocarpus pentandra* methyl ester (Biodiesel)

Parameter	Observed/ Standard value		
	Biodiesel	**Fossil diesel**	**ASTM standard for Biodiesel**
Kinematic viscosity @ 40°C (cSt)	4.104	2.60	1.90-6.0
Density @ 30°C (kg/m^3)	827	820	-
Acidity (mg KOH/g)	0.36	-	0.50 Max.
Specific gravity	0.827	0.820	-
Flash point (°C) (closed cup)	165	65	93 Min.
Fire point (°C)	174	70	-
Saponification value	284.34	-	-
Iodine value	92.30	-	-
Cetane Number	63.93	46	47 Min.

4.8 Organoleptic and Physico-chemical properties of *Hydnocarpus pentandra* oil

Data on organoleptic and physic-chemical properties of *H. pentandra* oil are presented in Table 10.

H. pentandra oil was golden yellow in colour with agreeable odour.

It is found that oil is having kinematic viscosity of 7.146 centi stokes (cSt) (at 40℃), density of 950 (kg/ m^3 at 30℃), acidity was 1.04 (mg KOH/g), specific gravity of 0.950 (at 30ºc), free fatty acid content of 0.525 (as oleic acid equivalent), saponification value of 215.06 and iodine value of 92.30.

4.9 Chemical composition of *Hydnocarpus pentandra* seed kernel oil

Results of qualitative and quantitative analysis of oil from the seeds of *Hydnocarpus pentandra* are shown in Tables 11 to 15. The GC-MS chromatogram of the oil is displayed in Fig. 7-11. The chemical components of the volatile oil were determined by the peak area normalization method. The presence of several overlapping peaks reveals the complexity of the mixture.

Eight to ten compounds were detected in the oils, of which 4 to 5 were well identified as the major compounds in the oil of *H. pentandra*. The major chemical components were palmitic acid, stearic acid, chaulmogric acid, hydnocarpic acid and gorlic acid.

In Khurse area, 5 chemical components were obtained from the seed oil of *H. pentandra*, of which the chaulmogric acid (57.07%) had highest percentage, followed by gorlic acid (30.17%), hydnocarpic acid (7.94%), stearic acid (2.56%) and the lowest was palmitic acid (2.26%) (Table 11 and Fig. 7).

In Shivagadde area, 5 main chemical components were obtained from the seed oil of *H. pentandra*, of which the chaulmogric acid (36.09%) had highest percentage, followed by gorlic acid (16.17%), stearic acid (4.06%), palmitic acid (3.69%) and lowest was hydnocarpic acid (3.22 %) (Table 12 and Fig. 8).

In Hebre area, 5 chemical components were obtained from the seed oil of *H. pentandra*, of which the chaulmogric acid (56.03%) had the highest percentage, followed by gorlic acid (20.82%), hydnocarpic acid (5.85%), stearic acid (4.51%) and the lowest was palmitic acid (4.20%) (Table 13 and Fig. 9).

In Devimane area, 5 main chemical components were obtained from the seed oil of *H. pentandra*, of which the chaulmogric acid (24.57%) had the highest percentage, followed by gorlic acid (12.02%), stearic acid (5.13%), palmitic acid (3.48%) and lowest was hydnocarpic acid (3.45 %)(Table 14 and Fig. 10).

The results from all the four places with respect to the composition of *H. pentandra* seed oil varied except in some major components *viz.* palmitic acid, stearic acid, chaulmogric acid, hydnocarpic acid and gorlic acid. Chaulmogric acid showed high percentage in all the site conditions followed by gorlic acid (Table 15 and Fig. 11).

4.10 Biodiesel yield and Physico-chemical properties of *Hydnocarpus pentandra* methyl ester (Biodiesel)

Data on organoleptic and physic-chemical propertiesof biodiesel (methyl ester of seed oil) from *H. pentandra* are presented in Table 16.

It was found that the biodiesel yield was 82-85 %. Biodiesel of *H. pentandra* was having pale yellow colour with agreeable odour.

It is found that biodiesel is having kinematic viscosity of 4.104 cSt (at 40℃), density as 827 (kg/m^3 at 30℃), acidity was 0.36 (mg KOH/g), specific gravity of 0.827 (at 30ºc), saponification value as 284.34, flash point of 165ºC, fire point 174ºC and cetane number of 63.93.

4.11 Nutrient content of *Hydnocarpus pentandra* de-oiled cake

Macro and micro nutrients of *Hydnocarpus pentandra* cake is presented in Table 17-18 and Fig. 12-13.

Table 17. Nitrogen, Phosphorus and Potassium content of *Hydnocarpus pentandra* de-oiled cake

Locations	Nitrogen (%)				Phosphorus (%)				Potassium (%)			
	F_1	**F_2**	**F_3**	**Mean**	**F_1**	**F_2**	**F_3**	**Mean**	**F_1**	**F_2**	**F_3**	**Mean**
L_1	4.49	3.72	2.73	**3.65**	0.22	0.21	0.20	**0.21**	1.23	1.40	1.09	**1.24**
L_2	2.95	2.25	1.58	**2.26**	0.26	0.27	0.25	**0.26**	0.64	1.08	1.66	**1.13**
L_3	3.20	2.47	1.77	**2.48**	0.34	0.33	0.31	**0.33**	1.03	1.36	1.27	**1.22**
L_4	3.51	2.68	2.27	**2.82**	0.49	0.44	0.39	**0.44**	1.48	1.29	1.03	**1.27**
Mean	**3.54**	**2.78**	**2.09**		**0.33**	**0.31**	**0.29**		**1.10**	**1.28**	**1.26**	
	S.Em±		CD @1%		S.Em±		CD @1%		S.Em±		CD @1%	
Location (L)	0.13		0.48		0.001		0.003		0.07		NS	
Fruit size (F)	0.14		0.40		0.01		0.02		0.06		0.17	
L x F	0.27		NS		0.01		0.03		0.11		0.33	

Note: L_1- Khurse, L_2-Shivagadde, L_3- Hebre, L_4- Devimane, F_1-Large fruit, F_2-Medium fruit and F_3-Small fruit

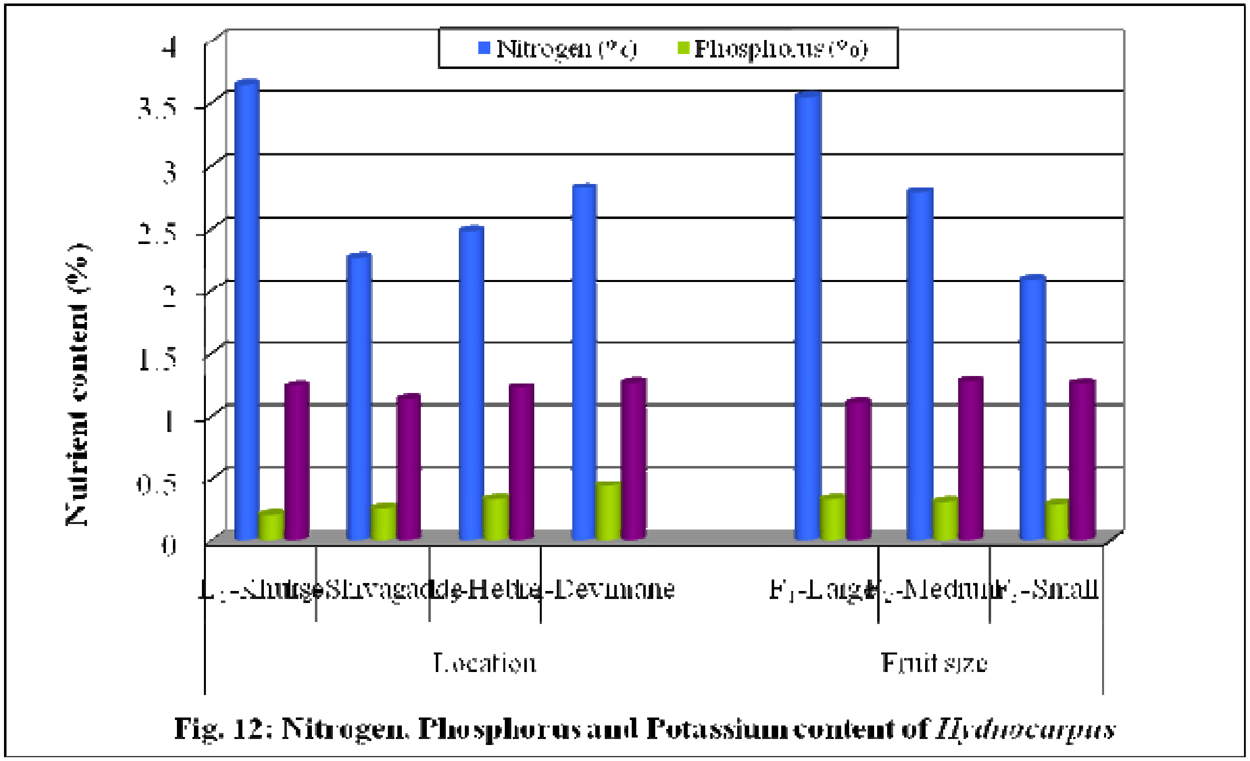

Fig. 12: Nitrogen, Phosphorus and Potassium content of *Hydnocarpus pentandra* de-oiled cake

Table 18. Calcium, Magnesium and Sulphur content of *Hydnocarpus pentandra* de-oiled cake

Locations	Calcium (%)				Magnesium (%)				Sulphur (%)			
	F_1	F_2	F_3	Mean	F_1	F_2	F_3	Mean	F_1	F_2	F_3	Mean
L_1	0.47	0.46	0.45	**0.46**	0.32	0.31	0.30	**0.31**	0.12	0.11	0.10	**0.11**
L_2	0.51	0.52	0.50	**0.51**	0.36	0.37	0.35	**0.36**	0.16	0.17	0.15	**0.16**
L_3	0.59	0.58	0.56	**0.58**	0.44	0.43	0.41	**0.43**	0.24	0.23	0.21	**0.23**
L_4	0.74	0.69	0.64	**0.69**	0.59	0.54	0.49	**0.54**	0.39	0.34	0.29	**0.34**
Mean	**0.58**	**0.56**	**0.54**		**0.43**	**0.41**	**0.39**		**0.23**	**0.21**	**0.19**	
	S.Em±		CD @1%		S.Em±		CD @1%		S.Em±		CD @1%	
Location (L)	0.001		0.003		0.001		0.003		0.001		0.003	
Fruit size (F)	0.01		0.02		0.01		0.02		0.01		0.02	
L x F	0.01		0.03		0.01		0.03		0.01		0.03	

Note: L_1- Khurse, L_2-Shivagadde, L_3- Hebre, L_4- Devimane, F_1-Large fruit, F_2-Medium fruit and F_3-Small fruit

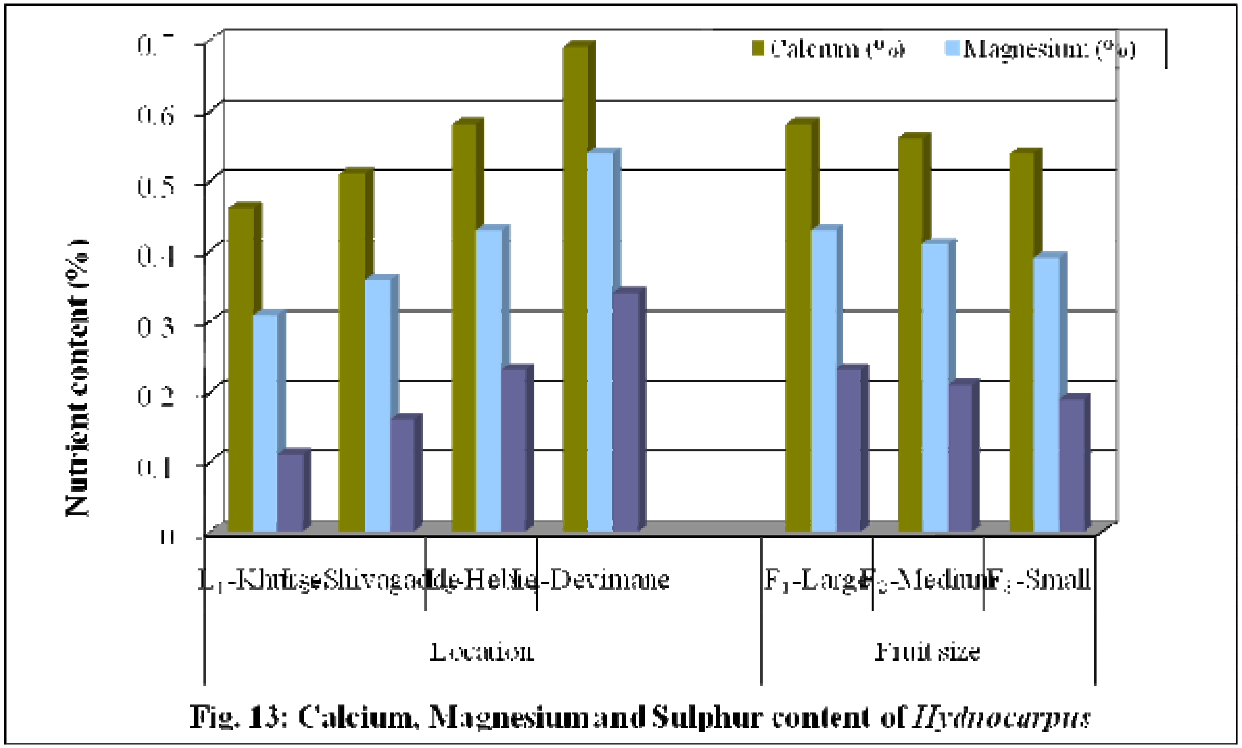

Fig. 13: Calcium, Magnesium and Sulphur content of *Hydnocarpus pentandra* de-oiled cake

4.11.1 Nitrogen content

Data on nitrogen content in de-oiled cake expressed in 'percentage' was presented in Table 17.

Nitrogen content of de-oiled cake was significantly influenced by different locations. L_1 recorded the highest nitrogen content (3.65%) followed by L_4 (2.82%) and L_3 (2.48%) whereas, the lowest nitrogen content was recorded in L_2 (2.26%).

Nitrogen content of de-oiled cake was significantly influenced by fruit sizes. F_1 recorded the highest nitrogen content (3.54%), followed by F_2 (2.78%) and the lowest nitrogen content was recorded in F_3 (2.09%). Interaction effect of location and fruit sizes was non-significant with respect to nitrogen content.

4.11.2 Phosphorous content

Data on phosphorous content of de-oiled cake expressed in percentage are presented in Table 17.

The phosphorous content was significantly influenced by different locations L_4 recorded the highest phosphorous content (0.44%) followed by L_3 (0.33%) and L_2 (0.26%) whereas, the lowest phosphorous content was recorded in L_1 (0.21%).

Phosphorous content of cake was significantly influenced by fruit sizes. F_1 recorded highest phosphorous content (0.33%) when compared to F_2 (0.31%). and lowest phosphorous content was recorded in F_3 (0.29%).

Interaction effect of location and fruit sizes also differed significantly with respect to phosphorous content. A treatment combination of L_4F_1 recorded the highest phosphorous content (0.49%) and the lowest phosphorous content was recorded with L_1F_3 (0.20%). L_1F_1 (0.22%) and L_1F_2 (0.21%) combination are on par with L_1F_3.

4.11.3 Potassium content

Data on Potassium content of de-oiled cake expressed in percentage presented in Table 17.

Effect of location on potassium content of cake was found to be not significant.

The potassium content was significantly influenced by different locations. L_4 recorded the highest potassium content (1.27%) followed by L_1 (1.24%) and L_3 (1.22%) whereas, the lowest potassium content was recorded in L_2 (1.13%).

Interaction effect of location and fruit sizes also differed significantly with respect to potassium content. A treatment combination of L_4F_1 recorded the highest potassium content (1.48%) and the lowest potassium content was recorded with L_2F_1 (0.64%). L_1F_1 (1.23%), L_1F_2 (1.40%), L_3F_2 (1.36%), L_4F_2 (1.29%), L_2F_3 (1.66%) and L_3F_3 (1.27%) combinations are on par with L_4F_1.

4.11.4 Calcium content

Data on calcium content of de-oiled cake expressed in percentage are presented in Table 18.

Calcium content was significantly influenced by different locations. L_4 recorded the highest calcium content (0.69%) followed by L_3 (0.58%) and L_2 (0.51%) whereas, lowest calcium content was recorded in L_1 (0.46%).

Calcium content was significantly influenced by fruit sizes, F_1 recorded the highest calcium content (0.58%) when compared to F_2 (0.56%) and the lowest calcium content was recorded in F_3 (0.54%).

Interaction effect of location and fruit sizes also differed significantly with respect to calcium content. A treatment combination of L_4F_1 recorded the highest calcium content (0.74%) and the lowest calcium content was recorded with L_1F_3 (0.45%). L_1F_1 (0.47%) and L_1F_2 (0.46%) combinations are on par with L_1F_3.

4.11.5 Magnesium content

Data on magnesium content of de-oiled cake expressed in 'percentage' are presented in Table 18.

The magnesium content of cake was significantly influenced by different locations L_4 recorded highest magnesium content (0.54%) followed by L_3 (0.43%) and L_2 (0.36%) whereas, lowest magnesium content was recorded in L_1 (0.31%).

Magnesium content was significantly influenced by fruit sizes, F_1 recorded the highest magnesium content (0.43%) followed by F_2 (0.41%) and the lowest magnesium content was recorded in F_3 (0.39%).

Interaction effect of location and fruit sizes also differed significantly with respect to magnesium content. A treatment combination of L_4F_1 recorded the highest magnesium content (0.59%) and the lowest magnesium content was recorded with L_1F_3 (0.31%). L_1F_1 (0.32%) and L_1F_2 (0.31%) combinations are on par with L_1F_3.

4.11.6 Sulphur content

Data on sulphur content de-oiled cake expressed in percentage are presented in Table 18.

The sulphur content of de-oiled was significantly influenced by different locations L_4 recorded the highest width (0.34%) followed by L_3 (0.23%) and L_2 (0.16%) whereas, the lowest sulphur content was recorded in L_1 (0.11%).

Sulphur content was significantly influenced by fruit sizes, F_1 recorded highest sulphur content (0.23%) when compared to F_2 (0.21%) and lowest sulphur content was recorded in F_3 (0.19%).

Interaction effect of location and fruit sizes also differed significantly with respect to sulphur content. A treatment combination of L_4F_1 recorded the highest sulphur content (0.39%) and the lowest total sulphur content was recorded with L_1F_3 (0.10%). L_1F_1 (0.12%) and L_1F_2 (0.11%) combinations are on par with L_1F_3.

4.12 Proximate composition of *Hydnocarpus pentandra* de-oiled cake

Proximate composition of *Hydnocarpus pentandra* de-oiled cake presented in Table 19 and Fig. 14.

4.12.1 Moisture content

Data on moisture content of de-oiled cake expressed in 'percentage' are presented in Table 19.

The moisture content in de-oiled cake was significantly influenced by different locations L_4 recorded highest moisture content (12.67%) followed by L_1 (12.50%) and L_3 (11.06%) whereas, lowest moisture content of cake was recorded in L_2 (7.92%).

Similarly, moisture content of de-oiled cake was significantly influenced by fruit sizes, F_2 recorded highest moisture content of (12.82%) when compared to F_1 (11.02%) and lowest moisture content was recorded in F_3 (9.27%).

Interaction effect of location and fruit sizes also differed significantly with respect to moisture content. A treatment combination of L_4F_1 recorded the highest moisture content (14.84%) and the lowest moisture content was recorded with L_2F_1 (6.37%). L_1F_2 (13.99%) and L_3F_2 (13.60%) combinations are on par with L_4F_1. Similarly L_2F_3 (6.58%) combination is on par with L_2F_1.

4.12.2 Crude fiber content

Data on crude fiber content in de-oiled cake expressed in percentage are presented in Table 19.

In case of crude fiber content in de-oiled cake was significantly influenced by different locations L_4 recorded the highest crude fiber (6.92%) followed by L_2 (6.06%) and L_1 (5.86%) whereas, the lowest crude fiber content was recorded in L_3 (5.31%).

Table 19. Proximate composition of *Hydnocarpus pentandra* de-oiled cake

Locations	Moisture (%)				Crude fibre (%)				Crude fat (%)				Ash content (%)			
	F_1	F_2	F_3	Mean	F_1	F_2	F_3	Mean	F_1	F_2	F_3	Mean	F_1	F_2	F_3	Mean
L_1	12.60	13.99	10.92	**12.50**	6.18	6.24	5.17	**5.86**	0.03	0.09	0.03	**0.05**	9.10	10.49	7.42	**9.00**
L_2	6.37	10.80	6.58	**7.92**	6.62	5.05	6.50	**6.06**	0.02	0.02	0.02	**0.02**	2.87	7.30	3.08	**4.42**
L_3	10.26	13.60	9.32	**11.06**	4.51	7.85	3.57	**5.31**	0.02	0.02	0.09	**0.04**	6.76	10.10	5.82	**7.56**
L_4	14.84	12.90	10.26	**12.67**	9.09	7.15	4.51	**6.92**	0.01	0.02	0.02	**0.02**	11.34	9.40	6.76	**9.17**
Mean	**11.02**	**12.82**	**9.27**		**6.60**	**6.57**	**4.94**		**0.02**	**0.04**	**0.04**		**7.52**	**9.32**	**5.77**	
	S.Em±		CD @ 1%		S.Em±		CD @ 1%		S.Em±		CD @ 1%		S.Em±		CD @ 1%	
Location (L)	0.23		0.84		0.20		0. 73		0.01		0.03		0.23		0.84	
Fruit size (F)	0.24		0.69		0.23		0.67		0.01		NS		0.24		0.69	
L x F	0.47		1.38		0.46		1.35		0.02		NS		0.47		1.38	

Note: L_1- Khurse, L_2-Shivagadde, L_3- Hebre, L_4- Devimane, F_1-Large fruit, F_2-Medium fruit and F_3-Small fruit

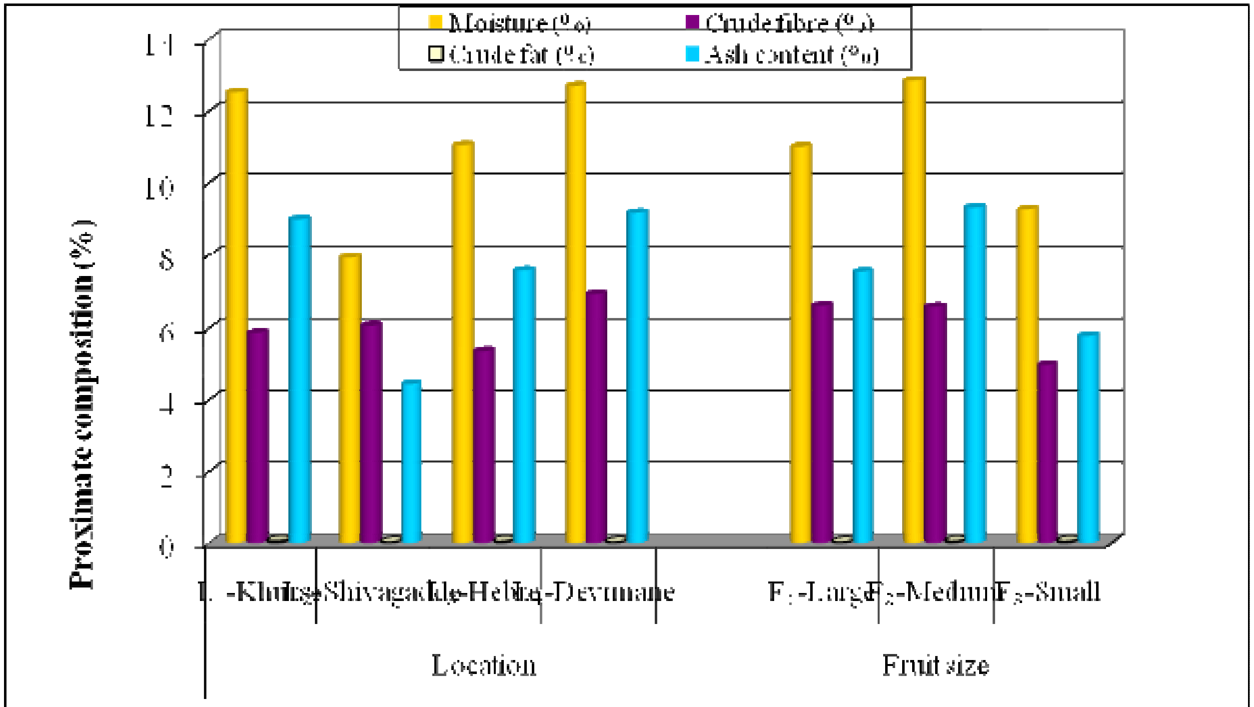

Fig. 14: Proximate composition of *Hydnocarpus pentandra* de-oiled cake

Fig. 14: Proximate composition of *Hydnocarpus pentandra* de-oiled cake

Crude fiber content in cake was significantly influenced by fruit sizes, F_1 recorded the highest crude fiber content (6.60%) when compared to F_2 (6.57%) and the lowest crude fiber content was recorded in F_3 (4.94%).

Interaction effect of location and fruit sizes also differed significantly with respect to crude fiber content in de-oiled cake. A treatment combination of L_4F_1 recorded the highest crude fiber content (9.09%) and the lowest crude fiber content was recorded with L_3F_3 (3.57%). Combinations L_3F_1 and L_4F_3 with 4.51 per cent each are on par with L_3F_3.

4.12.3 Crude fat content

Data on crude fat content in de-oiled cake expressed in 'percentage' are presented in Table 19.

Crude fat in de-oiled cake significantly was influenced by different locations. L_1 recorded the highest crude fat (0.05%) followed by L_3 (0.04%) whereas, the lowest crude fat in was recorded in L_2 and L_4 (0.02%).

Effect of fruit size and interaction effect of location and fruit sizes on crude fat content of cake was found to be not significant.

4.10.4 Ash content

Data on ash content in de-oiled cake expressed in percentage are presented in Table 19.

The ash content in de-oiled cake was significantly influenced by different locations. L_4 recorded highest ash content (9.17 %) followed by L_1 (9.00%) and L_3 (7.56%) whereas, lowest ash content was recorded in L_2 (4.42%).

Similarly, ash content in cake was significantly influenced by fruit sizes, F_2 recorded the highest ash content (9.32%) followed by F_1 (7.52%) and the lowest ash content was recorded in F_3 (5.77%).

Interaction effect of location and fruit sizes also differed significantly with respect to ash content in de-oiled cake. A treatment combination of L_4F_1 recorded the highest ash content (11.34%) and the lowest ash content was recorded with L_1F_2 (2.87%). L_1F_2 (10.49%) and L_3F_2 (10.10%) combination are on par with L_4F_1. Similarly L_2F_3 (3.08%) combination is on par with L_2F_1.

DISCUSSION

Fossil fuels such as petroleum, coal and natural gas are the main sources of energy. However, the increasing reliance of mankind on fossil fuels has increased the depletion rate of resources and global warming threats. As a result, alternative fuels must be found out to reduce the reliance on fossil fuel sources and to reduce greenhouse gas emissions as well. Biomass derived fuels could be one of the solutions, because they are renewable and act as a carbon sink. Biodiesel is one of the liquid biofuel that is gaining increasing attention as an alternative fuel to petro diesel.

However, the use of vegetable oils as a source for biodiesel production competes with its use as a source of food. Therefore, cheap resources must be used as raw materials for producing biodiesel. Numerous new plant sources are therefore being researched to yield 'high value added' products from it. India has great potential to produce alternative energy instead of fossil fuels derived from the renewable natural resources in the form of plants. Little work has been done so far on germplasm collection and its evaluation for chemical composition (oil content) of seeds of India (Ginwal *et al.,* 2005).

The *Hydnocarpus pentandra* has been found in natural forest in Agro-climatic Zone-9 (Hilly Zone) of Karnataka state. The zone is highly variable with respect to climatic, topographic and edaphic conditions (Gaonkar, 1991). Despite the importance *H. pentandra*as a major medicinal tree species, studies focusing on the effect of different provenance on its growth, productivity, oil quantity, oil quality and its biodiesel production potential are surprisingly very scanty. The knowledge of species-site interaction will be highly useful in selecting sites for future plantations in order to increase the production per unit area. The existing natural population in forest in the Agro-climatic Zone-9 offered great opportunity to study the response of *H. pentandra* to provenance conditions. Climate and topography of 9^{th} zone varied greatly in rainfall, number of rainy days and altitude. It is imperative to understand the growth and productivity in relation to varied site conditions under which it is being grown. The results obtained in the present investigation are discussed hereunder.

5.1 Growth and productivity performance of *Hydnocarpus pentandra*

Several climatic factors like moisture, rainfall, number of rainy days, temperature, wind, *etc.* effect the vegetation. Among the climatic factors, altitudes rainfall and number of rainy days are most important. In the present study, the analysis of variance showed significant effect of provenance on various growth and productivity parameters such as DBH, total height, bole height, crown diameter and basal area. The growth and productivity parameters examined such as DBH (0.81 m), height (22.83 m), bole height (2.59 m), basal area (2.87 m^2) and except crown diameter (9.65 m) (Table 3) at different provenance were found superior under low rainfall area (Khurse) than rest of the sites. The Khurse location was characterized by mean annual rainfall (2000 mm) and less number of rainy days (84) (Table 2) and adequate moisture availability. This might have enhanced the growth of trees in the initial years and accelerated it at the subsequent litter fall leading to improved soil properties (Table 4). Most of the climatic and edaphic factors exhibited positive trend with almost all the growth and productivity parameters, except pH which showed negative association with them. This may be due to increase in organic carbon content of the soil which decreases the pH of the soil (Table 4). It is evident from the present investigation that the abundance of moisture coupled with rich nutrient status of their soils might have enhanced the growth (except DBH) and productivity of the species under the low rainfall area (2000 mm). The poor growth attributes were observed in the *H.pentandra* grown under heavy rainfall areas (3400 mm) and with more number of rainy days (116) (Table 2). It may be noted here that soil properties were found to be poor under high rainfall area because of leaching of nutrients and organic carbon along with runoff water as compared to rest of the sites.

The *H.pentandra* grown under moderate rainfall (Shivagadde) exhibited next superiority with respect to various growth and productivity parameters *viz.,* bole height (2.39 m), DBH (0.75 m), total height (21.03 m), basal area (2.15 m^2) and crown diameter (10.31 m) (Table 3). The region because of its moderate mean annual rainfall 2450 mm and moderate number of rainy days(92) have probably supported the rich growth of the *H.pentandra* in the initial years and thus accelerated its growth, productivity and moderate litter fall, thereby enriching the soil properties (Table 4). The growth and productivity attributes exhibited positive and significant association with the most of the climatic and edaphic factors, except electrical conductivity and pH which showed negative association. This may be due to increase in the nitrogen and phosphorus content of soil and pH. The availability of abundant moisture coupled with rich soil nutrients might have enhanced the growth and productivity in

Shivagadde (moderate rainfall area) area. However, the DBH and basal area were comparable with other three locations. This may be attributed to the low temperature and high humidity of the location.

The *H.pentandra* grown under high (Hebre) and very high (Devimane) rainfall areas were next in the rank of productivity parameters.

The trees under high rainfall areas (Hebre) occupied next place with respect to various growth and productivity parameters *viz.*, total height (20.73), bole height (2.25 m), basal area (2.03 m^2), crown diameter (10.16 m) and low in DBH (0.58 m) and bole height (2.25 m) (Table 3). The region because of the growth and productivity attributes exhibited positive and significant association with most of the climatic and edaphic factors. This may be due to increase in the nitrogen and phosphorus content of the soil (Table 4). The availability of abundant moisture coupled with rich soil nutrients might have enhanced the growth (except DBH and bole height) and productivity inHebre region 2900 mm. However, the total height and basal area of the tree was comparable with the other three sites which may be attributed to the temperature and humidity of the location.

The trees under heavy rainfall areas (Devimane) exhibited low superiority over other locations with respect to various growth and productivity parameters *viz.,* bole height (2.31 m), but low in total height (20.59 m), high in DBH (0.76 m), high in crown diameter (10.40 m) and low in basal area (1.99 m^2) (Table 3). The region comes under heavy annual rainfall 3500 mm and number of rainy days (116) (Table 2) might have supported the rich growth of the plantation in the initial years leading to acceleration of its growth. The low productivity or poor performance observed in this region may be attributed to its less depth soil properties like nitrogen, potassium and phosphorus, which might have leached out due to runoff water led to may be due to poor fertility build up and competition for nutrients by the seedlings because of its profuse natural regeneration in these locations (Table 4).

The study showed a few strong associations of soil properties with tree growth characters. Banerjee *et al.* (1986) reported the good growth of teak even under the soil pH of 4. Akinsanmi (1985) showed a significant association between the volume increment of teak with rainfall and soil pH. Ezenwa (1988) reported that soil pH is having influence on tree heights and basal area. Major nutrients such as available NPK showed a positive association with stand growth and productivity.

Similar results reported by Harwood *et al.* (1997) revealed that in New Guinea provenances of *Eucalyptus pellita* exhibited good performance relative to other Eucalyptus species in certain tropical environments, in both seasonally dry and year-round-humid climates and on soils of relatively low to high fertility. Although they can tolerate a dry season of up to 5–6 months and annual rainfall of only 1200 mm (Pegg and Wang, 1994; Pinyopusarerk *et al*., 1996), under these conditions their advantage over Queensland provenances of *E. pellita* appears to be the minor. The superiority of the New Guinea over the Queensland provenances appears clearest on lowland tropical sites with high annual rainfall (over 1600 mm) and a short (4 months or less) dry season.

Dickinson and Sun (1995) reported that *Eucalyptus urophylla* and *E. pellita* grown on a lowland site with higher rainfall and grown on more fertile *i.e.*, kraznozem soil, survival and health of both the species was excellent. *E. urophylla* slightly outgrew *E. pellita* in this trial, although its stem form had inferior growth and was much faster than that on the infertile, poorly textured Cardwell soil.

5.2 Influence of *Hydnocarpus pentandra* on soil properties

Hydnocarpus pentandra exerts influence on both the soil physical and chemical properties. The species shed good amount of litter material on to the soil resulting in enrichment of the soil properties. Soil fertility is determined by the presence or absence of plant nutrients *i.e.* macro and micronutrients. Nitrogen plays an important role in utilization of carbohydrates, phosphorus in energy transformation and potash in the activation of enzymes, osmotic regulation and protein synthesis (Samuel *et al*., 1985). Nutrients taken up by the plants are used for their growth and development activities and their concentration at root surface plays a key role in meeting these requirements (Wild and Jones, 1988). It may be noted here that the improvement in the physical and chemical properties of soil under all the soils under *H. pentandra* was better (Table 4).

5.2.1 Soil physical properties

Soil pH was low under all the locations. In the present study, a significant variation among locations for soil pH was recorded and it varied from 4.71 (L_2) to 5.40 (L_4) with an overall mean of 5.06 (Table 4). It revealed that the soils of all studied locations are acidic in nature. The soil pH was found to be reduced under all the locations. It may be observed from Table 4 that the soil pH was 5.02 in Khurse site, 4.71 at Shivagadde site, 5.11 at Hebre site and 5.40 in Devimane location.

The reduction in pH noticed under *H. pentandra* may be attributed to higher organic matter content and high microbial activities resulting in the production of high amount of organic acids, which in turn might have lowered the pH of the soil as compared to their respective controls. These results are in conformity with the findings of Animon *et al.* (1999) who have reported that soil pH under *E. tereticornis* ranged between 5.0 and 5.7 with depth. Syed *et al.* (2006) in Eucalyptus and Singh and Totey (1985) reported maximum increase in cation exchange capacity, exchangeable cations and organic matter in the soil under miscellaneous plantation flowers by Teak and Eucalyptus in Bhata soil of Raipur (MP).

The soils under the *H.pentandra* with the oblong conical canopy were more subjected to the action of the environmental factors leading to enhanced leaching of bases which might have led to relatively higher acidity of soils in plantations observed in this investigation (Mary and Sankaran, 1991). A slight difference in the soil reaction at different sites is attributed to the differences in the species composition of the vegetation (Vadiraj and Rudrappa, 1990).

The soil under the *H.pentandra* showed higher electrical conductivity than their corresponding control sites. The increase in electrical conductivity was higher in Shivagadde site (0.07 dS/m) followed by Khurse (0.05 dS/m) and Hebre and Devimane sites both exhibited EC value of 0.04 dS per m each (Table 4). Among these four sites there was no significant difference. Syed *et al.* (2006) reported that soil electrical conductivity of the soil samples in plantations varied from 0.08 to 0.35 dS per m. This indicates that there was no contribution of the litter in increasing the cations in the soil under plantation significantly as compared to control location.

5.2.2 Soil chemical properties

The higher nitrogen content was observed under *H. pentandra* tree was in the range of 113.73 to 122 kg per ha than their respective control locations (103.23-116.32 kg/ha). The increase in nitrogen content was high under Shivagadde location (122 kg/ha) followed by Hebre location (121.47 kg/ha) and low under Devimane (113.73 kg/ha) (Table 4). Stewart and Vander (1988) reported that soils at Kalhari and Miombo sites were strongly acidic and infertile nitrogen, mineral nitrogen and available phosphorus in the surface 10 cm under Miombo were more than double the values measured under Eucalyptus and the total nitrogen was significantly higher at the depth of 20-30 cm.

The higher phosphorus content was observed under *H. pentandra* in the range of 9.61 to 14.03 kg per ha than their respective control sites (9.11-10.42 kg/ha). The soil at Khurse had higher phosphorus content (14.03 kg/ha), followed by Hebre site (13.49 kg/ha) whereas phosphorus content at Devimane was low (9.61 kg/ha) (Table 4). Similar results have been observed by Animon *et al.* (1999) in *Eucalyptus tereticornis* and Syed *et al.* (2006) in Eucalyptus species.

In the present study, the soil under *H. pentandra* exhibited higher potassium content (245.19-265.05 kg/ha) than their respective control sites (147.53-165.78 kg/ha). The potassium content was highest in Shivagadde site (265.05 kg/ha) and lowest in Devimane site (245.19 kg/ha) (Table 4). The observations on higher nitrogen, phosphorus and potash content of the soils under plantations may be attributed to the addition of organic matter in the form of litter fall under plantations as compared to their respective control locations. The significant differences between the plantation sites may be due to the climatic factors and edaphic factors *etc.* It may also be due to the leaching effect in high rainfall areas for low potassium. Stewart and Vander (1988) reported that exchangeable potassium and magnesium were higher under Eucalyptus plantation. The observations of Animon *et al.* (1999) in *Eucalyptus tereticornis*, Syed *et al.* (2006) in Eucalyptus species and Guo *et al.* (2002) in *E. tereticornis* lend support to the findings of the present investigation.

Organic carbon was found to be on the higher side under *H. pentandra* than their respective control sites. The soil at Shivagadde site recorded organic carbon content of 0.73 per cent and Devimane site recorded the lowest value of 0.37 per cent (Table 4). In general, the organic carbon content of the soil reflects the differences in quantity of leaf litter fall and the rate of its decomposition. This may be attributed to addition and incorporation of organic matter into the soil. Vadiraj and Rudrappa (1990) reported decrease in pH, increase in organic carbon status of soil under five year plantation of *Casurina equesetifolia, Eucalyptus cameldulensis, Tamarindus indica* and *Mangifera indica* at Hoskote near Bengaluru. Similar results have been observed by Swami and Proctor (1997) in Eucalyptus, Teak and Acacia. Chavan *et al.* (1995) reported that forest tree species such as *Tectona grandis, Terminalia tomentosa, Pongamia pinnata, Gmelina arboria, Eucalyptus*sps, *Acacia* *ariculiformis* and *Casurina equesetifolia* grown on lateritic soil showed significant increase in the organic carbon, available nitrogen, potassium and phosphorus in the surface layer under plantations than their respective control sites.

5.3 Fruit, Seed and Kernel morphometric traits of *Hydnocarpus pentandra*

Seed characters are important when evaluating trees of unknown genetic potential especially in a tree like *H. pentandra* where little work has been done in terms of scientific evaluation. Considerable variation in growth, chemical composition of seed and seed traits at the level of provenance, variety or progeny has been reported in several multipurpose tree species such as Albizzia, Acacia and Prosopis (Wanyancha *et al.,* 1994; Goel and Behl, 2001), which are widely used in agroforestry systems. The knowledge on this type of variation would be useful for selection especially in a tree where the desired growth pattern, phenology and yield component distribution is yet to be defined due to its wide adaptability and end use pattern. Variation in seed characteristics, growth and branching pattern, and yield attributes and components of *H. pentandra* can be of great potential in tree improvement programmes, particularly selection of genotypes having more oil content and yield in addition to suitability in agroforestry systems.

5.3.1 Fruit morphometric traits of *H. pentandra*

The fruit length, width and fruit weight are found to be 5.76 to 6.97 cm, 4.55 to 5.89 cm, and 21.96 to 24.09 g, respectively (Table 5). Corresponding values for the Jatropha seed (Garnayak *et al.*, 2008; Sirisomboon *et al.*, 2007) are 18.65 to 21.02, 11.34 to 11.97 and 8.91 to 9.58 mm. The length and stem-end diameter of neem nut are 14.56 and 7.72 mm, respectively (Visvanathan *et al.,* 1996). The importance of these dimensions in determining aperture sizes and other parameters in machine design have been discussed by Mohsenin (1980) and highlighted lately by Omobuwajo *et al.* (1999).

The *Jatropha curcus* fruits obtained from Thailand were characterized by larger dimensions (1.3 g/seed) than genotypes from Mexico (0.53 to 0.74 g/seed) as reported by Makkar *et al.* (1998).

According to Mathur *et al.* (1984), variability occurring in fruit traits might be attributed to genetic in nature as a result of adaptation to various environmental conditions prevailing throughout their distributional range. However, environmental conditions cannot explain alone all the variability observed in traits. Accessions from the same agro ecological zone often presented various fruit traits. It is clear that one part of the variability is under genetic control. Some genotypes seem to produce large fruit traits than others (Kaushik *et al.,* 2007).

The size and shape of fruits is variable depending on the structure and form of the ovary and environmental conditions under which plant is growing. It is evident from the result that fruits from heavy rainfall area (L_4, Devimane) were found to be superior with respect to fruit length (6.97 cm), fruit weight (24.09 g) except fruit width (4.24 cm) (Table 5). There is a positive correlation excising between amounts of rainfall with the fruit length. Highest fruit length of 6.97 cm was observed in heavy rainfall area (L_4, Devimane) and smaller fruit size of 5.76 cm was observed in low rainfall area (L_1, Khurse). This is in line with study made by Dwivedi (1993) in *Azadirachta indica* and Devagiri *et al.* (1998) in *Dalbergia sissoo*. They found that the variation observed in the fruit characters may be attributed to adverse environment and differences in their distribution range.

Pavithra *et al.* (2013) also reported provenance variation and genetic variability in pod and seed traits with 232 candidate plus trees of *Pongamia pinnata* collected from selected agro-ecological zones of southern peninsular India. Significant variation (P <0.01) for pod and seed traits across zones and provenances within zones was recorded. The southern dry and transition zone of Karnataka showed the highest mean value for all the pod and seed traits.

5.3.2 Seed and kernel morphometric traits of *H. pentandra*

Results of the study demonstrated significant provenance variations in seed and kernel related traits of *H. pentandra* (Table 6 and 7). Several authors have reported similar findings for other species such as; *Cordia africana* (Loha *et al.*, 2006), *Dalbergia sissoo* (Gera *et al.*, 2000), *Faidherbia albida* (Ibrahim *et al.*, 1997), *Strychnos cocculoides* (Mkonda *et al.*, 2003) and *Tectona grandis* (Jayasankar *et al.,* 1999; Sivakumar *et al.*, 2002).

Multiple factors may induce and maintain variation in seed size some of which could be that large seeds may be favoured because they produce larger and more vigorous seedlings with better chances of survival than small seeds and conversely the small seeds may have a selection advantage due to wider and more effective dispersal. Therefore, contradictory evolutionary pressures mediated by biotic as well as abiotic agents potentially act upon plants to produce the seed size distributions observed in natural plant populations (Eriksson, 1999).

Seed character variations in pongamia have been reported from different geographical regions (Kaushik *et al.*2007). Similarly, it has been show that other trees have wide variations in relation to habitat have also been reported in *Azadirachta indica* by Kundu and Tigerstredt (1997).

Seeds from heavy rainfall area (L_4, Devimane) were found to be superior with respect to seed length (2.92 cm), seed width (1.80 cm), total area (2.55 cm^2) and 100-seed weight (114.94 g) except total perimeter (4.46 cm^2) (Table 6). There is a positive correlation excising between amounts of rainfall with the 100-seed weight. The size and shape of seeds is variable depending on the structure and form of the ovary and environmental conditions under which plant is growing. Variation in morphological characters of the seeds among the wild accession of *H. pentandra* may due to fact that the species is widely adapted to a broad range of edaphic conditions. Seeds collected from heavy to moderate rainfall areas are having higher seed parameters than lower rainfall areas. Similar studies showed a significant association between the seed parameters with rainfall, indicating wet zones are the most favourable to some tree species (Maes *et al.*, 2009). As described by several authors (Kaushik *et al.*, 2007; Ginwal *et al.*, 2005), variation in seed sources with respect to their morphological characters could be due to fact that the species grows over a wide range of rainfall, temperature and soil type.

High variability was observed in *H. pentandra* seed traits among the accessions. 100-seed weight ranged from 101.40 to 123.33 g. Variability in environmental conditions of mother trees may be explained the variation observed in seed traits. Indeed, *H. pentandra* were collected in different locations that present different climatic conditions and soil characteristics. Soil and climate of the place of seed origin are considered as important factors influencing the seed traits (Salazar and Quesada, 1987). Such variations in habitat have been reported in *Jatropha curcas* (Ginwal *et al.*, 2005; Kaushik *et al.,* 2007) and others tree species (Gera *et al.*,1978; Jindal *et al.*, 1999; Kumar *et al.*, 2004).

Variation in kernel was found to be less pronounced as compared to other seed- related characters.

Seeds from heavy and very heavy rainfall area (L_4, Devimane and L_3, Hebre) were found to be superior with respect to kernel length of 2.11 and 2.17 cm respectively, kernel width of 1.75 and 1.75 cm, and total area (1.95 and 1.91 cm^2 respectively. However, there is a variation with respect to perimeter (L_3, 3.81 cm^2; L_4, 3.46 cm^2) (Table 7).

Current study revealed a significant influence of rainfall and altitude on all seed-related characters except total perimeter (Table 6 and 7). All provenances that were compared were found to be significantly different.

5.4 Yield and composition of oil from the seeds of *Hydnocarpus pentandra*

5.4.1 Seed oil yield

The oil content of the seed samples of *H. pentandra* from Agro-climatic Zone-9 (Hilly Zone) of Karnataka state showed significant variations in the oil potential as well as in the chemical composition of oils. The amount of oil was obtained by soxhlet method using hexane as solvent system. All the extracted oils from *H. pentandra* seeds under study were of golden yellow to yellow coloured mobile liquids having agreeable smell predominantly (Table 9).Cole and Cardoso (1939) also reported similar colour and smell characteristics of *Hydnocarpus anthelmintica* seed oil. It may be recalled here that the highest oil yield was obtained with seeds of *H. pentandra* from Hebre site (L_3, 49.49%), while the lowest was from Khurse site (L_1, 32.35%). The medium sized fruits have yielded more oil when compare to other fruit sizes. The average yield from medium sized fruits is 44.02 per cent. Highest oil yield was observed medium sized fruit at L_3 (60.13%), which also recorded maximum oil yield among the test samples. The present findings are in agreement with the studies of Bringi (1987), who had also reported that *Hydnocarpus kurzii* contains 30.10 per cent oil and *Hydnocarpus wightiana* 63.0 per cent oil in their seeds.

Data (Table 7 and 9) showed seed length to be strongly correlated with seed width, seed area and exocarp thickness. Strong positive correlation was also seen between seed diameter and exocarp thickness. However, morphological parameters were not consistently correlated to oil content. These results are consistent with the finding of Kaura *et al.* (1998) who have observed similar correlations in *Azadirachta indica* (Neem).

The oil yield in any given situation is determined by the rate at which biomass accumulates between successive harvests and the concentration of oil in the seeds. On the other hand, biomass production in oil plantations is strongly influenced by the climatic and edaphic features of a given planting location and the quality of establishment of plantings (Whiffin, 1982; Wildy *et al.*, 2000). The bioactivity of the essential oil depends upon the type and nature of the constituents and their individual concentrations (Brooker and Kleinig, 2006) and which are highly determined by the genotype (Barton *et al.*, 1991). However, the environment may also influence the oil yields. Oil yield further varied markedly between seasons and in relation to site-specific edaphic factors including season, location, climate, soil type, leaf age, fertility regime, the method used for drying the plant material and the method of oil extraction (Doran and Bell, 1994).

The four regions of Hilly zone showed different oil yields (32.35 to 49.49%). Among these regions, Hebre (L_3), showed the highest oil percentage in *H. pentandra* seeds followed by Devimane (L_4), Shivagadde (L_2) and the lowest in Khurse region (Table 9 and fig.11). Kaushik *et al.* (2007) also reported variability in oil content of 24 accessions of *Jatropha curcas* seeds collected from different agroclimatic zones of Haryana state, India. There were significant differences in oil content between accessions. Oil variability ranged from 28.00 per cent in IC-520589 to 38.80 per cent in IC-520601.

Seed oil content was observed to be the highest in moist localities with high (Hebre, L_3, 2900 mm, 49.49%) to very high rainfall regions (Devimane, L_4, 3400 mm, 43.76%). The lowest seed oil content was observed in dry localities with low rainfall (Khurse, L_1, 2000 mm, 32.35%) to moderate rainfall and Puri and Swamy (2003) also reported that seed of Jatropha collected from moist localities had higher oil content than dry localities. Among the dry localities also oil content in seed was also affected by different soil types. They attributed these observations to different agro-climates and soil composition.

In spite of their larger size, seeds of *Calophyllum inophyllum* from Australian provenances contained lower amount of oil as compared to Sri Lankan provenances. However, seeds from Cardwell, Australia and Colombo, Sri Lanka recorded similar oil contents; 30.8 and 31.5% respectively. Interestingly, both provenances share similar mean annual rainfalls (2,000 mm). In contrast, the seeds from Anuradhapura, Sri Lanka and Yeppoon, Australia recorded the highest oil contents, despite sharing the lowest mean annual rainfall in their respective countries (Hathurusingha *et al.*, 2010).

Sangwan *et al.* (2001) reported that oil yield increased in the wet period following a dry season and then fell with approaching the next dry season. The greatest accumulation of recently mature biomass with high oil yield values was expected to occur in the canopy when light, temperature and water were not limiting the growth (List *et al.*, 1995) which coincided with the summer time. It may seen from the literature (Elaissi *et al.*, 2007) that yields of oil from *Eucalyptus oleosa* from the center of Tunisia were richer in the essential oil than that of south with a difference of 2.25 per cent. The yield variability in the total essential oil of *E. oleosa* species growing in Tunisia was attributed to the soil conditions and ecological, climatic conditions, age of plant and the season of the harvest.

Seeds store a large amount of storage materials which are mostly carbohydrates (Ibrahim *et al.,* 1997) and in harsher conditions they produce slightly smaller oil-rich seeds (Rose 1988). Seeds with very high oil content are sometimes dormant (Dweck and Meadows 2002). It is an adaptation to an unfavourable environment which could reduces the competition for available resources and improves the survival rate of germinated seedlings.

A significant negative correlation and positive correlation was recorded for latitude with 100-seed weight and seed germination, respectively. Altitude positively correlated with 100-pod weight and negatively correlated with oil content. Similar trend was observed in the study, the oil content was observed to be lower at higher altitude. Phenotypic coefficient of variation was higher than the genotypic co-efficient of variation indicating the influence of environment (Pavithra *et al.*, 2013).

5.4.2 Fatty acid composition of *Hydnocarpus pentandra* seed oil

The GC-MS chromatogram of the oils is displayed in Figs. 7-10. The fatty acid components of the oils were determined by comparing their retention time with the retention time of standard compounds. Concentration of the detected or identified compounds was determined by the GC-MS QP2010S software considering the peak area or peak height. The presence of several overlapping peaks shows the complexity of the mixture. Eight to ten compounds were mainly detected in oils, of which 5 were well identified as the major compounds in *H. pentandra* oil (Tables 11 to 15). The major components were palmitic acid (2.26-4.20%), stearic acid (2.56-5.13%), oleic acid (1.46%), chaulmogric acid (24.57-57.07%), hydnocarpic acid (5.85-7.94%) and gorlic acid (12.02-37.35%) (Table 15). In addition, there were some other differences and similarity between oil compositions of *H. pentandra* seed oil.

Similar studies were carried out by Cole and Cardoso (1939), who studied the fatty acid composition of *Hydnocarpus anthelmintica* oil and reported the presence of palmitic acid (16:0, 7.5%), oleic acid (18:1, 12.30%), hydnocarpic acid (67.8%), gorlic acid (1.40%), chaulmogric acid (8.70%) and lower homologus hydnocarpic acid (0.10%).

Azam *et al.* (2005) also reported fatty acid profile of *Hydnocarpus kurzii*, oil is rich with palmitic acid (16:0, 4.0%), oleic acid (18:1, 14.60%), hydnocarpic acid (39.40%), gorlic acid (19.50%) and chaulmogric acid (22.50%). He also reported the presence of palmitic acid (16:0, 1.80%), oleic acid (18:1, 6.90%), hydnocarpic acid (48.70 %), gorlic acid (12.20 %), chaulmogric acid (27.00%) and chaulmogric homolog (3.40%) in *H. pentandra*.

It may be noted here that there are no reports on the chemical composition of *H. pentandra* seed oil. It is, therefore, for the first time that the analysis of *H. pentandra* seed oil grown in Agro-climatic zone-9 of Karnataka is being reported through this investigation.

The highest oil was extracted in the lowest altitude from *Thymus praecox* (2.73%), while it was the other way round at the highest altitude (1.98%). The highest thymol rate (69.08%) was ensured in the highest altitude. There were significant differences among the populations for quantity of oil with active contents. However, amount of oil and its components depends upon many factors. Edaphic, climatic and genetic factors were reported to bring in chemical and compositional variations in oil of different plant species (Tkachev *et al.,* 2006; Mirjalili *et al*., 2007; Lyra *et al*., 2008; Echeverrigaray *et al*., 2009; Hazzit and Baaliouamer, 2009; Toncer *et al*., 2010). Farah *et al.* (2006) reported that there could be differences in quantity of oils among populations in *Myrtus communis.*

In the present study, altitude and rainfall was found to significantly influence chaulmoogric acid, hydnocarpic acid and gorlic acid content and large individual differences were found in fatty acid content of *H. pentandra*. At lower altitude, highest chaulmogric acid, hydnocarpic acid and gorlic acid content was found in *H. pentandra*. Similar findings were reported by Doran and Bell (1994) and Da Silva *et al.* (2006).

Similar variations in chemical composition in oil have been reported by previous workers in India, Pakistan and also across the world (Siminshameel *et al*., 1996; Kumar and Satyawati, 2008; Akbar *et al*., 2009; Nzikou *et al*., 2009; Nayak and Patel, 2010 and Thiruvengadaravi *et al*., 2012).

The results obtained from the qualitative and quantitative analysis of the seed oils of *H. pentandra* have been shown in Table 15. The chaulmooogric acid was high in Khurse seed sample (L_1, 57.07%) fig7. Similarly hydnocarpic acid and gorlic acid content are highest in Khurse seed sample with 7.94 and 37.35 per cent, respectively. On the basis of the results obtained, it may be inferred that the seed oil of *H. pentandra* from the Khurse was rich in chaulmogric acid, hydnocarpic acid and gorlic acid content. Among the four regions, Khurse region with low rainfall and dry conditions was the best place to grow the *H. pentandra*. This oil may be used for medicinal purpose and as a substitute for the imported material. *H. pentandra* with chaulmogric acid and hydnocarpic acid content is suggested as its good source for medicinal use.

It is evident from results (Tables 15 and figs. 11) that the chemical composition of oils varied in the seeds collected from different areas. The results revealed differences in the composition of their oils with respect to data in the literature. These differences may be attributed to the geographical and climatic factors. It may be concluded from the present study that *H. pentandra* is a potential source of oil and it can grow well in climatic conditions. It should be further characterized for various commodities of cosmetics, medicinal and pharmacological attributes. The study on seed oil of the *H. pentandra* has shown that there is potential for commercial exploitation both for biofuel and as medicinal chaulmoogra oil in India for both the domestic and export markets. Much will depend on how this species perform in different regions of the country.

This study indicated that climatic and edaphic conditions exercise appreciable effect on proportional yields of the desirable oil yield and compounds chaulmogric acid, gorlic acid and hydnocarpic acid content. However, the variations between species and locations in terms of total oil yield tended to dominate over those relating to variations in balance of specific oil constituents. It may be noted that the production of total oil by sapling or coppice leaf biomass varied more than 10-fold between species and locations. Nevertheless, a small, but highly reproducible, negative correlation was evident between proportion of oil as chaulmogric acid versus other relatively major compounds, hydnocarpic acid and gorlic acid. It may be inferred that there may be other, not yet identified, effects which influence balance between constituents when species are grown at different locations. All the above-mentioned oil constituents are closely related structurally and stem from the same precursor based branch in the biosynthetic pathway for oil synthesis in Hydnocarpus seeds (Dewick, 1997) providing evidence that minor climatic or edaphic differences between locations might influence efficiency or inhibition of enzymes responsible for specific interconversions.

5.4.3 Physical properties of *Hydnocarpus pentandra* seed kernel oil

In order to address the shortages of feedstock for the production of biodiesel, new non-edible oil sources need to be exploited (Mustafa, 2011). High fatty acid non-edible oil can be employed to produce biodiesel. The industrial value of vegetable oils depends on their fatty acids composition and the ease with which it can be modified (Berchmans and Hirata, 2008; Ramos *et al.*, 2009). While feedstock with a low level of free fatty acids (FFAs) can be alkali catalyzed during the biodiesel production process, feedstock's with significant FFAs amounts perform better in the presence of acid catalysts (Demirbas, 2009). The quality of oil is of significant importance for production of biodiesel. Hence, the physical parameters are to be assessed before any non-edible oil can be used for biodiesel production.

The observed physical properties of *H. pentandra* oil such as kinematic viscosity (7.146 cSt at 40ºC), density (950 kg/m^3 at 30ºC), acid value (1.04 mg KOH/g), percentage free fatty acids (0.525 as oleic acid equivalent), specific gravity (0.950), saponification value (215.06) and iodine value (92.30) (Table 10).

Akbar *et al.,* (2009) studied the lipid fraction of Jatropha oil seed oil and analyzed for their chemical and physical properties such as acid value, percentage free fatty acids (% FFA), iodine value, peroxide value and saponification value as well as viscosity, and density.

Nayak and Patel (2010) also studied the physical properties of *Jatropha curcas* oil. The physicochemical properties shows acid value (36.46), iodine value (106.00 mg/g) and saponification value (194.70 mg/g). The unsaponifiable matter was 1.02 per cent.

Similar observations of physical properties of non-edible seed oils been reported by previous workers in India, Pakistan and also across the world (SiminShameel *et al.*, 1996; Kumar and Satyawati, 2008; Akbar *et al.*, 2009; Nzikou *et al.*, 2009; Nayak and Patel, 2010 and Thiruvengadaravi *et al.*, 2012).

5.5 Biodiesel Yield and Quality parameters of *Hydnocarpus pentandra*

The problem with substituting triglycerides for diesel fuel is mostly associated with high viscosity, low volatility and polyunsaturated characters. However the direct use of vegetable oils as fuel can cause numerous engine problems like poor fuel atomization, incomplete combustion and carbon deposition on fuel injector and engine fouling (Sridharan and Mathai, 1974; Williamson and Badr, 1998; Karaosmanoglu*et al.,* 2000;Encinar *et al.,* 2002). This is due to the presence of higher viscosity (about 11-17 times higher than petroleum diesel) of vegetable oils. Hence the viscosity of vegetable oils can be reduced by several methods which include blending of oils, micro emulsification, pyrolysis and transesterification (Ma and Hanna, 1999). Among this transesterification is widely used for industrial biodiesel production. Because it gives high yield with low temperature, pressure and short reaction time.

Transesterification is a chemical reaction involving triglycerides with alcohol in the presence of catalyst. For transesterification process methanol was commonly used. Because methanol is cheaper than ethanol and the recovery of un-reacted methanol is easier (Zhou and Boocock, 2003). In the case of base catalyst potassium hydroxide (KOH) or sodium hydroxide (NaOH) are used, because it is less expensive and easy to handle in storage and transport. However most of the studies showed best properties of biodiesel was obtained by using potassium hydroxide as catalyst (Encinar, *et al*., 2005; Karmee and Chadha 2005; Dorado, *et al*., 2004; Ugheoke *et al*., 2007). Besides, some studies showed best result with the use of sodium hydroxide as fuel (Felizardo*et al*., 2006; Vicente*et al*., 2004). Nevertheless base catalyzed transesterification method is commonly employed for biodiesel production. Since, it is more economical method than other methods (Singh*et al*., 2006). Bases catalyze the reaction by removing a proton from the alcohol, which acids donate protons to the carbonyl group making the reactions more reactive. Besides acids and bases, transesterification can also be catalyzed in the presence of enzymes (Hama *et al*., 2004; Shimada *et al*., 2002).

5.5.1 Biodiesel yield from *H. pentandra* seed oil

Naika *et al.* (2008) has discussed the mechanism of a dual process adopted for the production of biodiesel from Karanja oil containing free fatty acid (FFA) up to 20 per cent. The first step is acid-catalysed esterification by using 0.5 per cent H_2SO_4, alcohol 6:1 molar ratio with respect to the high FFA Pongamia oil to produce methyl ester by lowering the acid value, and the next step is alkali-catalysed transesterification The yield of biodiesel from high FFA Karanja oil by dual step process has been observed to be 96.6-97 per cent.

The Indian non-edible oils are often contaminated with FFAs, depending on the process of oilseed collection, expelling and storage condition, and contamination of oil/oilseed with moisture. The conventional alkali-catalysed route of biodiesel production does not work out effectively with high FFA feedstock such as Karanja oil. However, the dual-step process of transesterification using acid-catalysed and followed by base-catalysed reaction proves effective in producing the appropriate quality of biodiesel as per the ASTM specification (Prafulla and Deng, 2009).

The optimum reaction conditions for transesterification of *H. pentandra* seed oil was by a two step acid-base process using methanol as reagent and H_2SO_4 and NaOH as catalysts for acid and base reactions respectively. Use of 0.3 per cent of H_2SO_4 acid followed by 1 per cent NaOH, base-catalysed reaction proves effective in producing the appropriate quality of biodiesel. The yield of methyl esters from *H. pentadra* oil under the optimal condition was 82-85 per cent.

The experimental study revealed that, the optimum reaction conditions for methanolsis of oil was 0.3 per cent of H_2SO_4 and 1 per cent NaOH as catalyst, alcohol/oil molar ratio 6:1, reaction temperature 65℃, and rate of mixing 360 rpm for a period of 3 h.

Karthikeyan *et al.* (2013) also reported dual-step process of transesterification of *H. pentandra* seed oil (Marotti) using acid-catalysed and followed by base-catalysed reaction as optimum reaction conditions for transesterification for producing the appropriate quality of biodiesel. In our study also it is observed that, single step base-catalysed tranesterification has not yielded high quantity of biodiesel. The yield was highest in a two stage process.

The observed poor yield in single stage reaction of *H. pentandra* may be due to non availability of base catalyst (NaOH) for transesterification as reported by Sahoo and Das (2009). They have observed that, biodiesel production from feed stocks *viz.* Jatropha, *Pongamia pinnata* and Polanga oil, with high FFA is extremely difficult using alkaline catalysed transesterification process. This is because the alkaline catalysts react with FFAs to form soap that prevents the separation of the glycerin and ester. A two step transesterification are developed to convert the high FFA oils to its ester. The first step reduces the FFA content of the oil to less than 2 per cent. The viscosity of biodiesel is nearer to that of diesel.

Meher *et al.* (2006) has carried out transesterification of Karanja oil with methanol for the production of biodiesel. The yield of methyl esters from Karanja oil under the optimal condition was 97-98 per cent. The experimental study revealed that, the optimum reaction conditions for methanolsis of Karanja oil was 1 per cent KOH as catalyst, alcohol/oil molar ratio 6:1, reaction temperature 65℃, and rate of mixing 360 rpm for a period of 3 h. The yield of methyl esters was >85% in 15 min and reaction was almost complete in two hours with a yield of 97-98%. The reaction was incomplete with a low rate of stirring at 180 rpm, whereas stirring at high rpm was a time efficient process.

Prafulla and Deng (2009) also reported a two-step and single-step transesterification process was used to produce biodiesel from high free fatty acid non-edible oil and edible vegetable oils, respectively. This process gives yields of about 80-85 per cent for Pongamia oil using potassium hydroxide as a catalyst. It was found that the high FFA oils could not be transesterified with the alkali catalyst transesterification process

The separation and / or extraction of these components and their subsequent treatments through thermal, chemical, catalytic, bio-chemical (enzymatic) and other methods to yield highly desirable value added products is now a subject of considerable research efforts worldwide. The concept of conversion and manufacturing of such components is now termed a 'bio-refinery' where biodiesel is but one of the useful commercial products.

5.5.2 Physico-chemical properties of Biodiesel from *H. pentandra* seed oil

Certain important fuel property such as density, kinematic viscosity, acidity, specific gravity, saponification vale, iodine value, cloud point, pour point and flash point of synthesized biodiesel, crude *H. pentandra* oil were studied and compared with biodiesel standards. Viscosity and flash point are the important parameter that has to be considered (Table 16).

In case of *H. pentandra* biodiesel, it is observed that the kinematic viscosity 4.104 cSt (at 40°C), density of 827 kg per m^3 (at 30°C), acidity was 0.36 (mg KOH/g), specific gravity was 0.827 (at 30ºc), saponification value was 284.34, flash point 165ºC, fire point 174ºC and cetane number 63.93 (Table 16). The biodiesel produced from *H. pentandra* oil have properties within the acceptable range for automotive gas oil.

Similar findings were reported by Karthikeyan *et al.*, (2013). They reported that biodiesel obtained from *Hydnocarpus pentandra* seed oil (Marotti) through two-stage transesterification process is having kinematic viscosity (6.12 cSt), flash point (165ºC), fire point (176 ºC), specific gravity (0.86 kg/m^3), cloud point (12.5ºC) and cetane value of 52.2 respectively.

Viscosity is the measure of resistance to flow of a liquid due to internal friction of one part of a fluid moving over another affects the atomization of a fuel upon injection into the combustion chamber and thereby, ultimately leads to the formation of engine deposits. The higher the viscosity, the greater is the tendency of the fuel to cause such problems (Ma, 1999). The value of kinematic viscosity is reported in the Table 16. It was found that the viscosity of the synthesized biodiesel (4.104 cSt) was within the prescribed specifications of biodiesel standards.

Viscosity is also known to increase with increasing chain length (Rodrigues *et al.*,2006; Ramirez *et al.*, 2012), whereas branching seems to play a secondary role (Knothe and Steidler, 2005). Although, the majority of the examined MEs have a chain length between 17 and 18 carbon atoms

Density is also defined as its weight per unit volume, although, this quantity is more properly called specific weight. The density of *H. pentandra* biodiesel obtained was 827 (kg/m^3 at 30 ºC). This also in agreement with the findings of Adebayo *et al.* (2011) who reported the density of biodiesel from *Pongamia pinnata* seed oil as 876 (kg/m^3).

Specific Gravity is a relative measure of the density of a substance. The specific gravity of *H. pentandra* biodiesel is 0.827. The reported value is in agreement with the studies carried out by Karthikeyan *et al.*, (2013) who also reported specific gravity of 0.860. It is interesting to note that, the observed specific gravity of *H. pentandra* biodiesel is far better than the reported value by Karthikeyan *et al.*, (2013). The specific gravity of *H. pentandra* biodiesel is very close to the fossil diesel (0.820)

The flash point of biodiesel is greater than that of diesel. The flash point of a volatile liquid is the lowest temperature at which it can vapourize to form an ignitable mixture in air. The fire point is defined as the temperature at which the vapour continues to burn after being ignited. The flash point is often used as a descriptive characteristic of liquid fuel, and it is also used to help characterize the fire hazards of liquids. Therefore it has been considered as an important fuel property. The value of flash point of *H. pentandra* biodiesel is 165 ºC. The higher value of flash point as observed in this study has a higher safety than the petroleum diesel for transport purposes (Mehdi and Hamid, 2011).

Specifically, the (average) values of flash point for 26 examined feedstock's range from 127.7^{o}C (for the most saturated FAME, coconut) to 174.5^{o}C (peanut ME), with the mean value from all feed stocks being 163.3ºC. European specifications require biodiesel fuels to have at least 101 ºC flash points, whereas in the US the minimum required level is lower (93 ºC) ; both are meant to determine a lower limit of purity in the final FAME, and are easily met by *H. pentandra* biodiesel.

Closed cup testers normally give lower values for the flash point than open cup and are a better approximation to the temperature at which the vapour pressure reaches the lower flammable limit (Ritesh *et al.*, 2011). In the present study closed cup test method was used, indicating biodiesel of *H. pentadra* is safer and meets the national and international standards.

One of the most influential properties of the diesel fuel is the dimensionless cetane number (CN), which represents the ignitability of the fuel, particularly critical during cold starting conditions. Low cetane numbers lead to long ignition delay, *i.e.* long time between fuel injection and start of combustion. Cetane Number is the ability of fuel to ignite quickly after being injected. Higher the cetane value, the better the ignition quality of fuel. This is one of the important parameters, which is considered during the selection of FAMEs for use as biodiesel. For this, different countries/organization has specified different minimum values. Biodiesel standards of USA (ASTMD 6751), Germany (DIN 51606) and European Organization (EN 14214) have set this value as 47, 49 and 51, respectively (Azam *et al.*, 2005).

In the present study, *H. pentandra* biodiesel has cetane value of 63.93 which is higher than the prescribed values.

Another important criterion for biodiesel is its degree of unsaturation, which is measured as iodine value. To an extent, the presence of unsaturated fatty acid component in biodiesel is required as it restricts the biodiesel from solidification. However, with higher degree of unsaturation, biodiesel are not suitable for biodiesel as the unsaturated molecules react with atmospheric oxygen and are converted to peroxide, cross-linking at the unsaturation site can occur and the material may get polymerized into a plastic-like body. At high temperature, commonly found in an internal combustion engine, the process can get accelerated and the engine can quickly become gummed up with the polymerized biodiesel. To avoid this situation, biodiesel standards have set a minimum limit of iodine value in their specifications. The biodiesel of *H. pentandra* is having iodine value less than the maximum permitted limit (92.30).

Generally, biodiesel with higher cetane number are favored for use as biodiesel. However, with increase of cetane number, iodine number decreases which means degree of unsaturation decreases. This situation will lead to the solidification of biodiesel at higher temperature. To avoid this situation, the upper limit of cetane number (65) has been specified in US biodiesel standard (ASTMPS 121–99). In this study it was observed that cetane value is less than the upper limit of cetane number.

Besides, the concentration of linolenic acid and acid containing four double bonds in FAMEs should not exceed the limit of 12 and 1 per cent, respectively (Klopfenstein, 1985). In the present study it is observed that, *H. pentandra* biodiesel is devoid of highly unsaturated fatty acids like linolenic acid. Therefore, the biodiesel of this species is suitable for the production of biodiesel.

Biodiesel is gaining increasing acceptance in the market as an environmental friendly alternative diesel fuel (Jidon and Narko, *et al.* 2010). It is non-toxic, biodegradable, and free of sulphur or any carcinogenic compounds (Demirbas, 2002). The demand and cost of edible oils prevents its use in the production of biodiesel. So, a large variety of plants that produce non-edible oils are considered for biodiesel production. In India, there are several non-edible oil seed species such as *Jatropha curcas* (Jatropha), *Pongamia pinnata* (Karanja), *Azadirachta indica* (Neem), *Madhuca indica* (Mahua) *etc.*, which could be utilized as a source for production of oil (Meher *et al.*, 2006) and can be grown in large scale on non-cropped marginal lands and waste lands (Gerhard, 2005).

Biodiesel of *H. pentandra* gives slightly lower brake thermal efficiency, higher smoke density, lower in hydrocarbon and oxides of nitrogen as compared to neat diesel for all load conditions (Karthikeyan *et al.,* 2013).

H. pentandra having higher oil content in their seed/kernel was found to be most suitable for use as biodiesel and it meet the major specification of biodiesel standards of USA, Germany and European Standard Organization. Hence, *H. pentandra* biodiesel could be suitable for DI diesel engine without any modification on the existing condition (Karthikeyan *et al.,* 2013).

5.6 Nutrient content of *Hydnocarpus pentandra* de-oiled cake

In the present study, nutrient content of *Hydnocarpus pentandra* de-oiled cake was studied for nitrogen, phosphorous, potassium, calcium, magnesium and sulphur content. The macro and micro nutrients were significantly influenced by both the location and fruit sizes (Table 17).

Oil seed cake as a manure has been shown to influence the chemical properties of soil such as soil reaction, electrical conductivity, nutrient transformation and availability, buffering capacity, *etc.*

Addition of oil cakes serves as a source of organic matter and both major and micro nutrients and hence it enhances the organic carbon as well as nutrient (Tiyagi and Alam, 1995) in soil significantly.

Neem seed cake contains more nitrogen (2-5%), phosphorus (0.5-1.0%), calcium (0.5-3%), magnesium (0.3–1%) and potassium (1-2 %) (Radwanksi and Wickens, 1981). The Jatropha oil cake is acidic in reaction and medium in electrical conductivity and relatively rich in NPK besides sufficient organic matter content. It contains 4.89 per cent nitrogen, 1.52 per cent P_2O_5, 1.49 per cent of K_2O. Because of rich nutrient composition, the addition of the composted organic wastes improve availability of NPK calcium as well as key micronutrients such as iron, copper, and manganese in soil (Chaturvedi, 2011).

In our study of *H. pentandra* cake, it is observed that nitrogen content ranged from 1.58 to 4.49 per cent, phosphorous content was ranged from 0.20 to 0.49 per cent, and potassium content was ranged from 0.64 to 1.66 per cent. Calcium content was ranged from 0.46 0.74 per cent, magnesium content was ranged from 0.30 to 0.59 per cent; sulphur content was ranged from 0.10 to 0.39 per cent.

Similar results are reported by Elnasikh *et al.* (2011) from *Pongamia pinnata* cake. Pongamia seed cake contains 3.9 percent of nitrogen, 0.9 per cent of phosphorous, 1.2 percent of potassium.

Castor cake contains 58 per cent organic matter, 5 per cent of N, 2 per cent of P and 1.5 per cent of K. It also has traces of nutrients like manganese, zinc and copper, thus making it a balanced fertilizer. It can be applied for any type of soil, with its high content of organic matter. This cake is rich in N and hence it can be used as organic fertilizer (Ramachandran *et al.* 2007).

Pongamia seed cake contains high NPK values and it is an excellent organic fertilizer. In addition, cake contains relatively high content of micronutrients. Application of cake with nitrogenous fertilizers, gives a synergistic result by slow down the process of conversion of nitrogenous compounds into ammonia, nitrites and nitrates and improving N use efficiency. It is suited for crops like rice, wheat, cotton, sugarcane, fruits, vegetables, spices, coconut, tea, coffee, flower plants and aromatic plants. (Osman *et al.*, 2009).

Increase in electrical conductivity and exchangeable cation in association with oilcake application is also reported besides enhancing micronutrient content in soil. Application of neem cake along with FYM enhanced the organic carbon content in soil. In laboratory tests, addition of 3per cent neem cake to soil increased soil pH from 5.2 to 8.7 (Abbasi *et al.*, 2005). Neem seed cake significantly increased electrical conductivity, exchangeable calcium, iron, manganese, copper and zinc content (Elnasikh *et al.*, 2011) in soil.

Resource-poor farmers will be constrained to invest more in organic N source and to encourage them for increased use of organic manures suitable policy mechanisms are needed. There is a clear impact of use of Pongamiacake on profitability of crops reflecting higher additional B: C Ratio *viz.*, 5.03 and 8.57 in case of use of cake and cake + fertiliser, respectively in production of soybean while 1.81 and 1.75 in case of maize. Similarly, the impact of sub-treatments on growth parameters and treatments receiving Pongamia cake was significant and had higher values over two other treatments. Substituting hundred percent N with Pongamia cake also gave higher yield over farmer's practice (Osman *et al.*, 2009)

H. pentandra de-oiled cake with high to moderate nutrient can be effectively used for supplementing macro and micro nutrient needs of agriculture.

5.7 Proximate composition of *Hydnocarpus pentandra* de-oiled cake

Inadequate availability of pastures due to shrinking grazing lands in light of intensive cultivation and chronic shortage of protein and energy rich animal feeds due to enhanced needs of ever increasing human population are some of the major constraints for obtaining optimum productivity of livestock and poultry in India and other South East Asian countries. Hence, the animal nutritionists are compelled to explore the possibilities of feeding non-edible and non-competitive agro-industrial by-products to meet the nutritional requirements of animals. These unconventional feeds were though found to be promising, long term feeding that too at higher levels adversely affected the animal performance due to the presence of toxic factors.

Such non-edible agro-industrial by-products, which are hitherto wasted otherwise despite rich nutritional value, can be converted into wholesome animal feeds after evolving suitable processing technology which could be easily adoptable by farmers as well as by the industry. One among them is *H. pentandra* de-oiled cake and in this study its potential is evaluated for its proximate composition.

The high fibre content can act better on the digestive system without giving much problem of constipation. The fibre content of almond seed cake is low when compared to the value of 8.2per cent obtained for African mango (Oke and Umoh 1978).

The moisture content of almond seed cake was 25.23 per cent. This result therefore shows that almond seed has high moisture content, hence cannot be preserved for a long time. This value is high when compared to 5.5 and 5.1 per cent for cashew nut (Fetuga *et al.,* 1974).

The ash content of almond seed was 5 per cent. Ash content signifies the level of mineral present in the sample. The ash content of almond seed cake is high when compared to 3.3 per cent recorded for cashew nut (Fetuga *et al.,* 1974). The fibre content of almond seed cake was 3.11 per cent. This is significantly high when compared to 0.8 per cent obtained from cashew nut and 2.5 per cent for African oil bean and melon seed cake. Karanj cake, containing~28-34per cent crude protein, is not commonly used as a feed for livestock and poultry due to the harmful effects of anti-nutritional/toxic factors present in the seed cake (Akpabio, 2012)

Aderibigbe *et al.* (1997) reported the fibre content of *Jatropha curcas* meals between 3.9 and 4.5 per cent NDF on dry matter basis was lower than that in soybean meal (17.2% NDF). Similarly, seed meals from provenances showed varied fibre content with Cape Verde (4.7%), Senegal (5.6%), Burkina Faso (5.3%), India (4.5%) and Nicaragua (3.8%).

In the present study, proximate analysis of *H. pentandra* de-oiled cake was studied. The proximate compositions were significantly influenced by both the location and fruit sizes. Moisture content in the de-oiled cake ranged from 6.37 to 14.84 per cent, crude fiber 3.57 to 9.09 per cent, crude fat 0.01 to 0.09 per cent and ash content was ranged from 2.87 to 11.34 per cent.

The present findings are in agreement with the work of Saetae and Suntornsuk (2010); they reported that the sterilized *J. curcas* seed cake contained approximately fat (3%), ash (6%) and fiber (9%) on dry basis.

Ramakrishna and Mishra (2011) reported that Pongammia oil cake contains 10.29 per cent moisture, 10.17 per cent ash. They also reported 10.09 per cent moisture and 14.63 per cent ash in case of Madhuca and 9.22 per cent moisture and 8.07 per cent ash in case of Jatropha.

Jatropha curcas seeds are usually not given much attention in nutritional research due to the presence of the toxicants and anti-nutrients. However, the result of the proximate analysis of the seeds showed presence of crude lipid also crude fibre and ash, showing that it is a good source of minerals for animal nutrition. It is therefore a good potential source of animal nutrition if subjected to appropriate detoxification processes (Inekwe, 2012).

The proximate analysis of un-decorticated and decorticated full-fat castor seeds and cake showed that both contain high quantities of valuable nutrients, protein, nitrogen free extract, ether extract, crude fibre and high total ash content signifying the presence of high levels of macro- and micro-minerals. The presence of high nutrients suggests that castor seeds and cake may serve useful alternative feedstuffs for livestock if properly processed since previous works showed that though castor seed contain high nutrients, it harbors toxic substances namely ricin, ricinine, hydrocyanide, allergens and other alkaloids. These toxicants have limited the use of castor seeds for feeding purposes for man or animals (Aisha *et al*., 2013).

The cake left after extraction contained only 0.01 per cent of residual karanjin. By far, solvent extraction was found to be the best method of detoxification of karanj seed cake. Incorporation of deoiled karanj cake between 24 and 30per cent in the concentrate mixtures of growing calves (Gupta *et al*., 1981; Konwar and Banerjee, 1987) and kids (Srivastava *et al*., 1990) apparently produced no adverse affects in the performance, confirming the reduction of karanjin levels upon deoiling.

Similarly the alkali treatment of unconventional protein feed supplements (Neem Seed kernel cake, Mahua seed cake, sal seed cake *etc.*), was successfully tried for their detoxification (Joshi *et al*., 1990; Katiyar *et al*., 1991, 1993; Bhar and Katiyar, 1996; Gowda *et al*., 1998)

The use of Hydnocarpus oil and other plant parts as traditional medicine has been a long custom especially in the rural parts of India. Similar applications of Hydnocarpus oil in flavour and fragrances, as condiment or spice, in medicines, as antimicrobial or insecticidal agents and to protect stored products is found in many parts of the world (Dhanasekaran *et al.*, 2013; Joshi and Harijan, 2014).

Two species namely, *Jatropha curcas* and *Pongamia pinnata* are favoured in India because of their contrasting plant characteristics and the species selected match the site characteristics. However, *Jatropha curcas* and *Pongamia pinnata* are not suitable in heavy rainfall areas like Western Ghat region(Zafar *et al.,* 2003). There was a need to search for suitable species which can be cultivated in high rainfall areas and also with high oil content. In the present study helps in identifying *Hydnocarpus pentandra*, a tree species native to western ghat with high oil content and its potentiality for biodiesel production. Among the number of species available in the country, *H. pentandra* with potential oil yield can serve as a raw material for both pharmaceutical and biofuel purpose.

One of the main advantages of using Hydnocarpus species for oil production is its suitability for cultivation in heavy rainfall areas, which is central to the economic production of oil. Despite the available potential of Hydnocarpus species to contribute towards the country's economy in oil sector, the present information on oil production in general and on yield influencing factors in particular is highly deficient. To that end, this study helps in understanding the influence of abiotic factors like rainfall, altitudes temperature and soil properties on oil yield and oil quality of *H. pentandra* in Agro-climatic Zone-9 of Karnataka.

Future line of work

1. *Hydnocarpus pentandra* is a potential source of oil and it can grow in diverse type of climatic conditions. Work on characterization of its oil in various commodities like medicinal, pharmacological and other attributes is scanty which may be taken up in near future.
2. The study on oil of *Hydnocarpus* species has shown that there is potentiality for commercial exploitation of *Hydnocarpus* oil in India for both the domestic and export markets. Much will depend on how this species perform in different regions of the country. It may be standardized in future.
3. If intended to use *H. pentandra* in the production of oils, further investigation may be undertaken for fabricating oil expellers' to extract oil at small scale industrial level. There is also a need to study the most appropriate age for harvesting and harvesting time for better oil yield.
4. The study of fatty acid profile of oil indicated variation in Hydnocarpic acid and chaulmogric acid contents, which are of medicinal importance. Hence, there is a need to undertake detailed studies in future to identify promising source.
5. The knowledge of genetic variability and association between pod and seed traits is considered to provide considerable help in genetic improvement of the species. Further studies are needed on these lines in *H. pentandra*.
6. Studies are needed to evaluate the de-oiled cake for both as organic manure and cattle feed.
7. Looking at the potential of *H. pentandra* in the present study, establishment of plantations of high yielding species/provenance in different Agro-climatic Zones of Karnataka and India may be explored.

SUMMARY AND CONCLUSIONS

An attempt was made through the present study to analyze the influence of different location on the growth, productivity and chemical composition, seed oil and biodiesel composition of *Hydnocarpus pentandra* (Buch.-Ham.) Oken during 2012-14 at four sites in Uttar Kannada district, Karnataka, which form parts of central Western Ghats is known for natural forests. Being Hilly Zone it provides a wide variation in climatic, topographic and edaphic factors. The present study was aimed at (1) assessing the growth and productivity of *H. pentandra* in different site conditions, (2) determining the influence of *H. pentandra* on soil properties as compared to the control sites, (3) studying the seed traits of *H. pentandra* in different provenance, (4) studying yield and quality of *H. pentandra* oil under different provenance, (5) studying the biodiesel quality of *H. pentandra* and(6) assessing the de-oiled cake of *H. pentandra* for nutrients and proximate composition. The salient futures of study are as follows:

1. Provenances were classified by taking in to account of the rainfall and altitude. In the present study, *Hydnocarpus pentandra* each under four conditions were evaluated for various growth, productivity, oil yield, oil quality, biodiesel yield and biodiesel quality, nutrient and proximate composition.

2. The analysis of variance showed significant effect of provenance on various growths and productivity parameters such as DBH, total height, bole height, crown diameter and basal area of the *H. pentandra*.

3. Khurse location (L_1) low rainfall 2000 mm showed superiority in growth (DBH, 0.81 m) and also good performance in productivity *viz.*, height (22.83m), bole height (2.59 m) and basal area (2.87 m^2) as compared to other experimental locations.

4. The soil under *H. pentandra* exhibited better physical and chemical properties as compared to their respective control sites. The soil properties such pH, electric conductivity (dS/m), available nitrogen (kg/ha), available phosphorus (kg/ha), available potassium (kg/ha) and organic carbon (%) were studied. There was a significant difference between the four locations with respect to soil properties. Shivagadde location (L_2) moderate rainfall of 2450 mm showed superiority with nitrogen (122 kg/ha), potassium (265.05 kg/ha) and organic carbon (0.73%) as compared to other experimental locations.

5. The fruit size parameters of *H. pentandra* were significantly influenced by different locations. Interaction effect of location and fruit sizes also differed significantly. A treatment combination of L_4F_1 (Devimane- Large fruit) recorded the highest fruit length (8.48 cm), L_3F_1(Hebre- Large fruit) recorded the highest fruit width (7.93 cm) and L_4F_1 (Devimane- Large fruit) recorded the highest fruit weight of 26.0 g.

6. The seed size parameters of *H. pentandra* were significantly influenced by different locations. Interaction effect of location and fruit sizes also differed significantly. A treatment combination of L_3F_1(Hebre- Large fruit) recorded highest seed length of 2.96 cm, L_4F_2 (Devimane- Medium fruit) recorded the highest seed width (1.92 cm), L_1F_1 (Khurse- Large fruit), L_2F_3 (Shivagadde- Small fruit)and L_2F_1(Shivagadde- Large fruit) recorded the highest seed total perimeter of 5.84 cm^2 each, L_3F_1(Hebre- Large fruit) recorded the highest area (2.96 cm^2) and L_4F_1(Devimane- Largefruit) recorded the highest seed weight of 123.33 g.

7. The seed kernel parameters of *H. pentandra* were significantly influenced by different locations. Interaction effect of location and fruit sizes also differed significantly. A treatment combination of L_3F_1 (Hebre- Large fruit) recorded the highest kernel length (2.17 cm), L_3F_1 recorded the highest kernel width (1.75 cm), L_2F_1(Shivagadde- Large fruit) and L_2F_3(Shivagadde- Small fruit) recorded the highest kernel perimeter of 4.84cm^2 each and L_3F_1(Hebre- Large fruit) recorded the highest kernel area (1.96 cm^2).

8. The effect of different locations and interaction effect was not significant with respect to kernel and shell fraction. Kernel fraction was significantly influenced by fruit sizes. F_2 (medium fruit)

recorded highest kernel fraction (55.76%). F_1 (large fruit) recorded highest shell fraction (44.69%).

9. Interaction effect of location and fruit sizes also differed significantly with respect to oil yield. A treatment combination of L_3F_2 (Hebre- Medium fruit) recorded the highest oil yield (60.13%) and the lowest oil yield was recorded with L_1F_1 (Khurse- Large fruit) (30.60 %). L_1F_2 (32.65%) and L_1F_3 (33.79%) combination are on par with L_1F_1(Khurse- Large fruit).

10. *H. pentandra* seed oil color was found to be golden yellow having agreeable smell.

11. *H. pentandra* seed oil was having kinematic viscosity 7.146 (cSt, at 40℃), density of 950 (kg/ m^3 at 30℃), acidity was 1.04 (mg KOH/g), specific gravity of 0.950 (at 30ºc), free fatty acid content of 0.525 (as oleic acid equivalent), saponification value of 215.06 and iodine value of 92.30.

12. Eight to ten compounds were mainly detected in oils, of which 5 were well identified as the major compounds in *H. pentandra* oil. The major components were palmitic acid (2.26-4.20%), stearic acid (2.56-5.13%), oleic acid (1.46%), chaulmogric acid (24.57-57.07%), hydnocarpic acid (5.85-7.94%) and gorlic acid (12.02-37.35%). In addition, there were some other differences and similarity between oil compositions of *H. pentandra* seed oil.

13. Biodiesel of *H. pentandra* was found to be pale yellow in colour with agreeable odour having kinematic viscosity of 4.104 cSt (at 40℃), density as 827 (kg/m^3 (at 30℃), acidity of 0.36 (mg KOH/g), specific gravity of 0.827 (at 30ºc), saponification value as 284.34, flash point of 165ºC, fire point 174ºC and cetane number of 63.93.

14. Interaction effect of location and fruit sizes also differed significantly with respect to macro and micro nutrients content of *H. pentandra* de-oiled cake except no nitrogen content. A treatment combination of L_4F_1 recorded the highest phosphorous content (0.49%), L_4F_1(Devimane- Large fruit) recorded the highest potassium content (1.48%), L_4F_1(Devimane-Large fruit) recorded the highest calcium content (0.74%), L_4F_1 (Devimane- Large fruit) recorded the highest magnesium content (0.59%) and L_4F_1 recorded the highest sulphur content (0.39%).

15. Interaction effect of location and fruit sizes also differed significantly with respect to proximate composition of *H. pentandra* de-oiled cake except crude fat content. A treatment combination of L_4F_1 (Devimane- Large fruit) recorded the highest moisture content (14.84%), L_4F_1(Devimane- Large fruit) recorded the highest crude fiber content (9.09%) and L_4F_1 recorded the highest ash content (11.34%).

16. It was concluded from the present study that *H. pentandra* has the potential to capture Medicinal (Chaulmogric and Hydnocarpic acid) and biofuel sector, due to its high oil content biodiesel production potential and for large seed afforestation programmes (non-edible oil sources) in heavy rainfall areas.

17. Future line of work has been suggested.

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PROVENANCE EFFECT ON MORPHO-CHEMICAL, MORPHO-GENETIC CHARACTERIZATION OF OIL OF *Hydnocarpus pentandra*

DHATHRI N. R. **2014** **Dr. P. RAMANA**

Major Advisor

ABSTRACT

Provenance influence on growth, productivity, seed quality, fatty acid profile, seed cake quality and biodiesel production of *Hydnocarpus pentandra* were evaluated in Uttara Kannada district (Agro-climatic Zone-9) of Karnataka during 2013-2014. Experiments are undertaken in the already existing trees in natural forests at four different locations with varying altitudes (400-550 m) and rainfall (2000-3200 mm). There was a significant effect of site conditions on various growth and productivity parameters. The soil under *H. pentandra* exhibited better physical and chemical properties as compared to their respective control sites. The fruit, seed and kernel parameters were significantly influenced by different locations and fruit sizes. Kernel fraction was significantly influenced by fruit sizes. Seed oil yield varied from 30.61 to 60.13 per cent and was golden yellow in colour having agreeable smell. The GC-MS chromatogram of the oil indicated the presence of 8 to 10 compounds, of which 4 to 5 were identified as the major compounds viz. palmitic acid (2.26-4.20%), stearic acid (2.56-5.13%), chaulmoogric acid (24.57-57.07%), hydnocarpic acid (5.85-7.94%) and gorlic acid (12.02-30.17%). The methyl esters (biodiesel) having kinematic viscosity (4.104 cSt), density (827 kg/m^3), acidity (0.36 mg KOH/g), specific gravity (0.827), saponification value (284), flash point (165ºC), fire point (174ºC) and cetane number (63.93) with 82-85 per cent yield was obtained under the optimal condition. The biodiesel parameters fit into the ASTM standards. Macro, micro nutrients and proximate composition of de-oiled cake differed significantly with respect to location and fruit sizes. *H. pentandra* has the potential to capture medicinal and biofuel sector, due to its high oil content, oil quality and biodiesel production potential.