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RESEARCH ARTICLE

Studies on Ascorbic acid content of some wild edible fruits from Ahmednagar District, Maharashtra (India).

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Abstract

Numerous wild edible fruits from different families are distributed in Western Ghat (Maharashtra), India. Tribal people consume these fruits as natural source of food supplement. Ten wild edible fruits of *Diospyros melanoxylon, Pithecellobium dulce, Carrisa congesta, Lantana camera var. aculeata, Opuntia **stricta**, Aegle marmelos, Terminalia catappa, Ziziphus mauritiana* and *Elaegnus conferta, .Limonia accidisma* possess high nutritional potential and medicinal properties. The present communication deals with the investigation of vitamin-C contents of these ten wild edible fruits using standard protocols. The ascorbic acid content was compared with some commonly consumable fruits. Present study focused on the food value and exploration of underutilized edible fruits in Maharashtra, India.

Introduction:-

Nutritional potential of fruits make noteworthy contribution to standard of living and good health in human beings. There is need of identification and exploration of the wild underutilized edible fruits for executing the increasing demand of nutritionally potential diet for growing populations (Andersen et al., 2003; Sena et al., 1998). Since, wild fruits are found to be the most important source of micronutrients; nutritional rich supplements can be fulfilled by fruits. Hence, efforts must be devoted on sustainable use of underutilized wild plants resources as nutrient supplements against enhancing food and nutrition problems in developing countries. As a part of current scenario of safeguarding of good health, worldwide researchers were studying the nutritional potential of various types of wild edible underutilized plants (Lockeett et al., 2000; Ogle et al., 2001).

Enormous literature has evidenced that several wild plant fruits are edible since ages. However, the use of wild fruits as a food has decreased due to improvement and hybridization in commercially cultivated fruit plants. On the other hand increase in urbanization and gradual exploitation of forests is resulting in disappearance of several valuable wild species. Several people in remote areas are still using them as a supplement to complete their basic food requirement. Some are preserved for use during famine period or some time sold in the market. Now a day, the popularity of these wild fruits has declined. Hence, special attention should be focused on maintenance and popularisation of these natural resources as a of future food supply. Therefore, there is need to achieve scientific and systematic knowledge of wild edible fruits for cultivation. Hence, special attention is being focused on the study of vitamin-C content of underutilized wild fruits.

Especially, ascorbic acid (vitamin-C) is considered as a key factor in the quality determination of fruits (Vazhacharickal et al.,2015). It has excellent solubility in water with both acidic and reducing properties. It is being an essential nutrient in humans and it has functions as a cofactor in several enzymatic reactions (Kalt et al., 1999;

Padayatty et al., 2003; Gardner et al., 2000). The required vitamin-C intake in humans is approximately 60-95 mgs per day and the maximum intake level is 2000 mgs per day (Carr and Frei, 1999; Levine et al., 1995; Levine et al., 1999; Naidu, 2003). However, overdose of vit-C may cause diarrhoea, iron overload disorders, kidney stone formation and deficiency causes scurvy, muscle pain, skin lesions, fatigue and bleeding (Naidu, 2003; Carr and Frei, 1999; Holick and Chen, 2008; Mayland et al., 2005; Weber et al., 1995; Jacob, 1990; Jacob et al., 1987). It plays an important role in the construction and defence of human connective tissue, immunity, Alzheimer's disease, stroke and coronary heart disease (Padayatty et al., 2003; Morris et al., 1998; Gale et al., 1995).

There are two bioactive forms of vitamin-C are known such as L-ascorbic acid and L-dehydroascorbic acid (DHAA). Mostly, L-ascorbic acid form is often encountered in conjunction with small amounts of dehydroascorbic acid may also present as its primary oxidation product. There are different methods available for determination of ascorbic acid as reported in literature (Christie and Wiggins, 1978, Sauberlich et al.,1982; Pachla et al.,1985). The Association of Official Analytical Chemists (AOAC) has designated two official methods for the determination of vitamin C as (a) The dye-titration method (b) The micro-fluorometric method (AOAC, 1984). The dye titration protocol was found to be most suitable in terms of ease of conduction, rapid, use of common desk reagents and large number of samples can be analyzed in a short time. The micro-fluorometric method is more sensitive than redox titrimetic method but requires more sophisticated or expensive equipments and expertise for getting accurate results.

Plant Species considered for the determination of vitamin-C:-

(I) *Diospyros melanoxylon Roxb*

Family- Ebenaceae

Vernacular names- '*Temburini'.*

Locality- Distributed in Ratanwadi, Bhandardara, Tal- Akole, Dist- Ahmednagar, Maharashtra State, India.

Common in dry tropical and sub-tropical forests with black or dark brown bark and woody. Leaves are thick pale green and hairy. Fruits are yellowish and fleshy with enlarged calyx like brinjal. Leaf fall has been observed during March to April, flowering and fruiting observed during April to June. Fruits o are useful against diarrhoea, dry cough and hypertension. Leaves contain flavones and triterpenes (Mallavadhani et al., 2004)

(II) *Pithecellobium dulce(Roxb.) Bth*

Family- leguminosae(fabaceae)

Vernacular name- *'Vilayati chinch'.*

Occurrence- India

Locality- Distributed in Ratanwadi, Bhandardara, Tal- Akole, Dist- Ahmednagar, Maharashtra State, India.

The genus contains approximately 100 to 200 species of shrubs and small trees. The fruits are in the form of pod, which turns pink when ripe and opens to expose an edible pulp. It has medicinal applications against convulsions. It contains carotene, phenylalanine, tryptophan, saponine a sterol glucoside and ascorbic acid (Crane et al 1984).

(III) *Lantena camara L. var. aculeata (L.) Moldenke*

Family- Verbenaceae

Vernacular name- *'Tantani' and 'Ghaneri'.*

Locality- Distributed in Ratanwadi, Bhandardara, Tal- Akole, Dist- Ahmednagar, Maharashtra State, India.

It is the most extensive species of this genus growing in tropical sub-tropical & temperate regions. It is being a large scrambling evergreen strong smelling shrub. The fruit are fleashy berries in cluster, shiny globular in shape. Ripened fruits are black in colour consisting caryophyllene, 1-alpha phellandrene, lantadene A, lantadene, quinine and Lantanine. It has various medicinal properties viz. vulneary, diaphoretic, carminative, antispasmodic and tonic wounds, ulcers, swelling, tumors and rheumatism. Fruit are known to have antibacterial, antifungal, anti-inflammatory and anti-fertility activity (Ganesh et al 2010, Kumar et al 2010, Srivastava et al 2011).

(IV) *Carissa congesta* Wight

Family- Apocynaceae

Vernacular names- *'Karvand'.*

Locality- Distributed in Ratanwadi, Bhandardara, Tal- Akole, Dist- Ahmednagar, Maharashtra State, India.

This species belongs to rank growing strategy, woody, climbing shrub with evergreen leaves. Leaves are opposite oval or elliptic, 1 to 3 inch long and dark green in colour. The flowers are fragrant, tabular with hairy lobes, twisted to the left in the bud. Fruits are developed at terminal clusters, broad ovoid or round, dark purple or nearly black when ripe and bitter (Kirtikar et al., 2003).

(V)*Opuntia stricta* (Haw.) Haw.
Family- Cactaceae
Vernacular names- 'Nivdung'.
Locality- Distributed in Ratanwadi, Bhandardara, Tal- Akole, Dist- Ahmednagar, Maharashtra State, India.
It is spread in the form of large colonies can be used as ornamental plant in gardens and later on it has been used as a natural agricultural fencing. Fruits are oval elongated berry with a thick receptacle and juicy pericap Fruit and stem have been traditionally used as medicine in several countries (Griffith, M. P. 2004, Hunt et al 2006).

(VI) *Aegle marmelo* (L.) Corr.
Family- Rutaceae
Vernacular names- *'Bel'.*
Locality- Distributed in Ratanwadi, Bhandardara, Tal- Akole, Dist- Ahmednagar, Maharashtra State, India.
It has great mythological significance for Hindus. Fruit is being used for breakfast as a mixture pulp and palm sugar or in the form of syrup. The syrup has therapeutic agent and relives diarrhoea and dysentery. It is rich in minerals and vitamin content, coumarins, alkaloids, sterols and essential oil (Ansari P. Y. 2005, Singh S. 2000, Prince et al 2005).

(VII) *Terminalia catappa* L.
Family- Combretaceae
Vernacular names- 'Deshi badam'.
Locality- Distributed in Shendi, Bhandardara, Tal- Akole, Dist- Ahmednagar, Maharashtra State, India.
Terminalia catappa is a large tropical tree in the Leadwood tree family Combretaceae. The leaves contain several flavonoids, tannins and saponine. Fruit are used in different herbal medicines for various purposes (Hnawia et al 2011).

(VIII) *Ziziphus mauritiana* Lam.
Family- Rhamnanceae
Vernacular names- *'Bor'.*
Locality- Distributed in Shendi, Bhandardara, Tal- Akole, Dist- Ahmednagar, Maharashtra State, India.
Ziziphus mauritiana is native to temperate and subtropical region. It is a spiny evergreen shrub or small tree upto15 m high, with trunk 40 cm or more in diameter. The fruit are applied on cuts and ulcers are employed in pulmonary ailments and fever. Fruit are mixed with salt and chilli pepper is given in indigestion and biliousness (Grice AC, 1996; Taraneh, E and Asna, U., 2012).

(IX)*Limmonia acidissma* L.
Family- Rutaceae
Vernacular names- *'Kavath'.*
Locality- Distributed in Shendi, Bhandardara, Tal- Akole, Dist- Ahmednagar, Maharashtra State, India.
It is a native and common to dry plain, apparently drought tolerant and best adapted to light soil. Wood apple is useful for curing scurvy and in relieving flatulence. Seedless pulp of fruit is beneficial for the treatment of dysentery, diarrhoea and piles (Rathayake et al, 2009; Bandara et al, 1988)

(X) *Elaegnus conferta* Roxb.
Family- Elaeagnaceae
Vernacular names- *'Ambal'.*
Locality- Distributed in Shendi, Bhandardara, Tal- Akole, Dist- Ahmednagar, Maharashtra State, India.
Elaeagnus conferta is native to temperate and subtropical regions of Asia. It is deciduous or evergreen shrubs or small trees. The fruits used as medicine for the treatment of indigestion. The fruit are rich in Ca, Fe, Mn and P (He F. J. and MacGregor G.A., 2008; Feng et al,1986)

Material and Methods:-

Collection of Fruits and Sampling:
Wild fruits of *Diospyros melanoxylon, Pithecellobium dulce, Carrisa congesta, Lantena camera, Opuntia stricta, Aegle marmelos, Limmonia acidissma, Terminalia catappa, Ziziphus mauritiana* and *Elaegnus conferta*. collected from Shendi and Ratanwadi area (Bhandardara) Tal- Akole, Dist- Ahmednagar(MS), India. The raw and ripened fruit samples were collected and dried under shade.

(A) Preparation of the plant materials for chemical analysis:

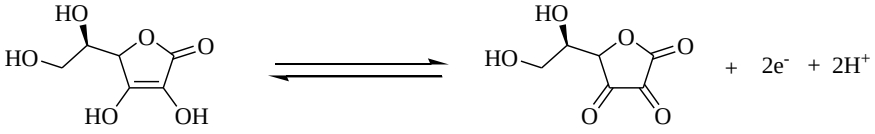
All ten fruit samples were dried in shade in order to remove residual moisture. The shade dried fruits were ground into powder using grinder, filtered and stored in polythene bags. The stored food powders were used as such for further lipid analysis.

(B) Chemical Analysis:

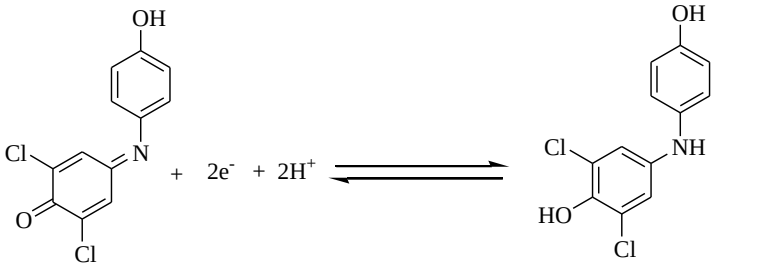
Determination of Vitamin-C all fruit samples were conducted in Research Laboratory, Department of Botany, Shri Muktanand College, Ganagapur, Dist- Aurangabad by titrimetic method.

(C) Principle:

The ascorbic acid in wild underutilized fruits can be determined by using redox titration method. In general, acid-base titration is not suitable because there are many acids and bases in food material. Hence, the 2,6-dichloroindophenol (DCP) dye has been used as colored titrant because it has ability to oxidize only ascorbic acid. It has dark blue in neutral and basic solutions and red in acidic solutions. The oxidation of ascorbic acid and reduction of DPC can be represented in Figure 1.



Oxidation of L-Ascorbic acid



In base-Dark blue colour
In Acid-Red pink colour

Reduced DCP-Colourless

Reduction of 2,6-dichlorophenol endophenol

Figure 1. Redox reactions involved in estimation of ascorbic acid

(D) Preparation of ascorbic acid solution (standard):

Weigh accurately ascorbic acid (100mg) and dissolve it distilled water (5 ml). Dilute this solution to 100ml with 4% oxalic acid solution in distilled water. Concentration of Ascorbic acid is 1mg/ml.

(E) Preparation of the dye solution:

Weigh accurately 42 mg of sodium bicarbonate ($NaHCO_3$) in 5ml distilled water. To this solution 52 mg of DCPIP (Mol wt. = 268.1g/mol) was added and solution was diluted to 200 ml with distilled water.

(F) Preparation of Sample Solution:

500 ml of fruit powder was mixed with 10 ml distilled water and mixture was stirred for 30 minutes and filtered using Whatman No.1 filter paper (Dilution Factor was 10 ml).

(G) General procedure for the determination of ascorbic acid in the fruit sample:

(G-1) Determination of V_1 ml (Volume of dye required for 10 ml of standard ascorbic acid solution):

Fill the burette with dye solution. 10 ml of ascorbic acid solution was taken in 100 ml conical flask. The ascorbic acid solution was titrated against dye solution from burette. Burette reading (ml) was recorded when solution in the conical flask turns pink and colour was persists for 30 sec. In this way three burette readings were recorded as X_1(7.4 ml), X_2 (7.5 ml) and X_3 (7.5 ml). From these readings, constant burette reading was recorded as V_1 ml= 7.5 ml).

(G-2) Determination of V_2 ml (Volume of dye required for 5 ml of sample solution):-

5 ml of the sample solution was pipette out in a 100 ml conical flask. To this solution 10 ml of 4% oxalic acid and titrated against the dye (V_1ml). Burette reading (V_2ml) was recorded when solution in the conical flask turns pink and colour was persists for 30 sec. The amount of the dye consumed is equivalent to the amount of ascorbic acid.

Amount of ascorbic acid in mg/100 gm of fruit sample was determined by following formula

$$\textbf{Weight of Vit-C} = \frac{\text{Wt.of Dye (mg)}}{\text{V1 ml}} \times \frac{\text{V2 ml}}{\text{Volume of Sample (ml)}} \times \frac{\text{10 (Working Std)}}{\text{Wt.of Sample (mg)}} \times 100$$

Result and Discussion:

In continuation to our ongoing research on applicability of wild fruits in India [37-39] the determination of ascorbic acid (Vit-C) content of ten wild edible plant viz. *Diospyros melanoxylon, Pithecellobium dulce, Carrisa congesta, Lantena camera, Opuntia stricta, Aegle marmelis, Limmonia acidissma, Terminalia catappa, Ziziphus mauritiana* and *Elaegnus conferta*. using redox titrimetric method. The results obtained were represented in Table 1. Wild edible fruits have been serving as good source of nutritional components such as protein, carbohydrate and vitamin for the local residents. Earlier reports indicated that vitamin-C content in *Pithecellobium dulce* (White) 82.6 mg 100 g^{-1} and Red-79.7 mg 100 g^{-1}, *Carissa congesta* (1.32 mg/g), *Aegle marmelos* (0.77 mg/g), *Lantana camara* (6.585 mg 100 g^{-1}) and *Opuntia stricta* (30.2 mg 100 g^{-1}) (Fernando et al, 2013; Kubola et al, 2011; Asadu et al, 2015; Albano et al, 2015) found in various regions. The variation in vitamin-C content may be due to adaptation of plants in different environmental soil conditions or the harvesting of fruits at different time or the climatic factors and the species variation (Sabir and Raiz, 2005). The ascorbic acid (vitamin C) content of ten wild fruit species are generally found in Maharashtra (India) indicated a valuable nutrition like other regularly consumable fruits such as orange, grape, lemon etc, as shown in Table 2. (Ighodalo *et al.*, 1991). As a part of our ongoing research on nutritional analysis of some wild fruits, herein we have determined the ascorbic acid content of ten wild fruits found in Maharashtra (India) by redox titration method. The results obtained indicate that vitamin-C content of *Pithecellobium dulce (*Roxb.*) Bth.* was 68.60 for white fruits and for red fruits it was found 61.34 mg/100gm. *Carissa congesta* was 36.68 mg/100gm, *Opuntia stricta* 31.68 mg/100gm. For *Lantana camara var. aculeata* was found 29.04 mg/100gm. For *Prunus amygdalus* ????????? 29.04 mg/100gm. Whereas, for *Diospyros melanoxylon* Roxb, *Aegle marmelos, Ziziphus mauritiana, Elaegnus conferta*. Vitamin-C content was found 10.56, 8.66, 2.64,13.2 mg/100gm respectively (Table 1).

Table 1 Determination of Ascorbic acid (Vitamin-C) contents of some wild fruit samples.

Sr.No	Plant Name	Family	Common Name	Amount of Ascorbic acid (Vitamin-C) (mg/100gm)
1.	*Diospyros melanoxylon* Roxb.	Ebenaceae	Temburini	10.56
2.	*Pithecellobium dulce (*Roxb.*)Bth.*	Fabaceae	Vilayati Chinch	68.60 61.34
3.	*Carissa congesta Wight.*	Apocynaceae	Karvand	36.68
4.	*Lantana camara L. var. aculeate* (L.)*Moldenke*	Verbenaceae	Ghaneri	29.04
5.	*Opuntia stricta (Haw.)Haw.*	Cactaceae	Nivdung	31.68
6.	*Aegle marmelos* (L.) *Corr.*	Rutaceae	Bel	8.66
7.	*Limmonia acidissma* L.	Rutaceae	Kavath	15.84
8.	*Terminalia catappa L*	Combretaceae	Deshi badam	29.04
9.	*Ziziphus mauritiana Lam.*	Rhamnaceae	Bor	2.64
10.	*Elaegnus conferta* Roxb.	Elaeagnaceae	Ambal	13.2

Table 2. Relative Vitamin C and protein content of *Spondias pinnata* and *Elaeagnus pyriformis* with selected well known fruits.

Common name of the fruit	Vitamin C (mg/100 g)	Common name of the fruit	Vitamin C (mg/100 g)
Grape	10	**Tembhurni**	**10.56**
Apricot	10	**Vilayati Chinch**	**68.60** **61.34**
Plum	10	**Karvand**	**36.68**
Watermelon	10	**Ghaneri**	**29.04**
Banana	9	**Nivdung**	**31.68**
papaya	60	**Beal**	**8.66**
Strawberry	60	**Kavath**	**15.84**
Orange	53	**Almond**	**29.04**
apple	48	**Bor**	**2.64**
Lemon	53	**Ambal**	**13.2**

Conclusion:

Determination of ascorbic acid content of wild edible fruit was conducted using redox titration method. The ascorbic acid content was further compared with some of the commonly consumable fruits (Ighodalo *et al*., 1991). The comparative results were represented in Table 2. This indicates the vitamin C potential of wild fruits under investigation were good to excellent for consumption. Hence it will be more beneficial than commonly consumable fruits when vitamin C rich supplement was required.

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