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Antimicrobial activity, nutritional profile and phytochemical screening of wild edible fruit of *Catunaregam spinosa* (Thunb.) Tirveng

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Abstract
The aim of the present study is to evaluate the nutritional profile, phytochemical screening and antimicrobial activity of wild edible fruits of *Catunaregam spinosa*. The fruits have been found to rich in nutrients such as crude fiber (12.6%), ash content (11.3%), moisture content (25%), protein (12.5%), lipids (0.018%) and carbohydrates (60.28%) with the energy value of 292±0.7 kcal/100g respectively. The phytochemical screening for the presence of flavonoids, saponin, phenol, tannins, alkaloids, anthraquinones, cardiac glycosides, phlobatannins, terpenoids and steroid in ethanol extract. Ethanol and chloroform extracts of *C. spinosa* showed maximum zone of inhibition against *Bacillus subtilis* followed by *Klebseila pneumonia, Pseudomonas aeruginosa, Escherichia coli* and *Salmonella typhi*. Aqueous extracts of the *C.spinosa* showed maximum zone of inhibition against *Klebseila pneumonia and Bacillus subtilis.* The results showed that the fruits contained higher value of energy level required per day by a person. Consumption of fruits may promote good health and well-being as well as reduce the risk of chronic diseases. These findings confirm that the *C. spinosa* fruit has a potential source for the formulation of new therapeutic drugs.

Keywords: Edible fruits, Nutritional analysis, Energy value, phytochemical analysis and Antibacterial activity

1. Introduction
Wild edible fruits played a vital role in food supplementing to maintain the diet of human beings. The dependence on the wild edible fruits has gradually decline due to exotic fruits have been introduced. But many people in tribal areas still use them as a supplement of their basic need of food. Some of them are preserved for use in dry period or sold in rural market. But the popularity of these wild forms has recently decreased. Apart from their traditional use of food, potentially they have many advantages. They are edible and having nutritional food value, which provides the minerals like sodium, potassium, magnesium, iron, calcium, phosphorus etc. They are immune to many diseases and often used in different formulation of 'Ayurveda' in Indian Folk- medicine. They provide fibres which prevent constipation [1]. It is consider that special attention should be paid in order to maintain and improve this important source of food supply.

Catunaregam spinosa belongs to the family rubiaceae commonly known as maniphal or Emetic nut [2]. In recent years plants and plant secondary metabolites (phytochemicals), previously with unknown pharmacological activities, have been extensively investigated. The *Catunaregam spinosa* fruits are edible and considered to be tonic, alternative, demulcent, diuretic and restorative. The drug is claimed as medicines to cure for piles, anti-dysenteric agent, asthma, jaundice, diarrhea, emetic and gonorrhea [3]. The fruit, seeds and barks posses insecticidal, anti-dysenteric, nauseant, expectorant, anthelmintic, abortifacient properties [4]. The fresh fruits contain high amount of carbohydrates and saponins [5]. The nutritional value describes mainly the percentage of major nutritional bio-molecules such as proteins, carbohydrates, lipids and fibere along with the presence of major minerals and their food value [6]. The fruits are a good source of various bioactive compounds and its importance being much unaware among the society. Hence, the present study has been framed to estimate the nutritional profile, phytochemical screening and antimicrobial activity of wild edible fruits of *Catunaregam spinosa.*

2. Materials and Methods

2.1 Sources of Plant Materials

The fresh fruits of *Catunaregam spinosa* were collected from the Kolli hills, Namakkal district, Tamil Nadu, India. The plants was identified and authenticated in the Botanical survey of India, Southern Regional Centre, Coimbatore, Tamil Nadu. The voucher registration No. BSI/SRC/5/23/2016/Tech/1751/specimen-3.

2.2 Fruit extract preparation

The samples were thoroughly washed with tap water. The fresh and dried edible portion of each sample was cut into tiny piece and crushed by using clean Mortar and Pestle, the grounded samples were stored in a labelled air tight container and placed in the refrigerator at 4°C and used immediately for subsequent analysis.

2.3 Nutritional Analysis

Determination of protein content by Folin-lowry's Method [7], Determination of Total Lipid [8], Determination of Carbohydrate, crude fiber, ash content, Moisture content by A.O.A.C Methods [9].

2.4 Determination of energy value

The total energy value in kcal/100g was estimated by using the method described by Jain and Sharma as shown below [10]. Energy value = 4x percentage of protein + 9x percentage of fat + 4x percentage of carbohydrate.

2.5 Qualitative phytochemical screening

Ethanol, chloroform and aqueous Solvent are used for the extraction. Flask extraction procedure was adapted for extraction. 25 grams of the powdered fruits sample was soaked in the conical flask containing solvent, wrapped with aluminum foil and placed in shaker for 48 hours at 120-130 rpm. After 48 hours, the extracts were filtered using Whatman filter paper No.1 Concentrated all the solvent extracts by vacuum evaporator. Dried extracts were stored at 4°C for further analysis. Phytochemical analysis of each extract has been carried out according to standard protocols [11, 12].

2.6 Antibacterial activity by Disc diffusion method

The antimicrobial assays of Ethanol, Chloroform and Aqueous extracts were performed by each fruits extract was tested for various concentration such as 25, 50, 75 and 100 µl, if different concentrations to see their inhibitory effects against microbial pathogens. Sterile paper discs (6 mm in diameter) prepared from Whatman No. 1 filter paper was impregnated with drug, containing solution placed on the inoculated agar. The inoculated plates were incubated at 37°C for 24 h [13]. The antibacterial activity was evaluated by measuring the diameter of the inhibition zone for the test microorganisms. The pure cultures of *Klebseila pneumonia, Pseudomonas aeruginosa, Escherichia coli, Streptococcus pyogenes, Salmonella typhi* and *Bacillus subtilis* were sub-cultured in nutrient agar.

3. Results and Discussion

The edible portion of the fruits is taken for the finding. The wild Edible fruits of plants can provide healthy alternatives to highly processed foods and pharmaceuticals, bringing greater health into our lives. Apart from their traditional use of food, potentially they have many advantages like antimicrobial properties [14]. They are edible and having fortified nutritional food value, which provides the proteins, lipid and carbohydrates. These are richest source of secondary metabolites with highest antimicrobial properties. They are immune to many diseases and often used in different formulation of 'Ayurveda' in Indian Folk medicine and traditional medicines. Documentation of wild edible fruits plays significant role to enhance the natural food resources which had been used with the help of religious knowledge [15]. Wild edible fruits today needs to be recommended for cultivation due to the reason which, they can serve as food material for ever increasing population [16]. The wild edible fruit are not only food but also contributes the beneficial natural nutrition source as food, diet, nutrition and nutrients to ever increasing population and in food scarcity. The wild edible fruits play an important role in sustainable livelihood of tribal communities residing in forest areas. Increased use of these wild edible fruits may raise as promising solutions on problems of malnutrition [17]. In order to medicate, a wider and sustained acceptance of wild fruits as important dietary components must be encouraged.

3.1 Nutritional analysis

Table 1: Nutritional analysis of *Catunaregam spinosa* fruits

S. No	Proximate analysis	Percentage
1.	Crude Fiber	12.6±0.2
2.	Ash content	11.3±0.2
3.	Moisture content	25±1.15
4.	Lipid	0.18±0.01
5.	Carbohydrate	60.28±4.6
6.	Protein	12.5±0.9
7.	Energy value	292±0.7

The proximate analysis was done in order to find out the nutritional value of *Catunaregam spinosa* fruits. The analyses were performed in triplicate and the values are expressed as mean percentage ± SD. The percentage proximate content of the fruit Catunaregam *spinosa* is shown in Tables-1**.** Proximate analysis is used to estimate the relative amounts of crude fiber, ash content, moisture content, carbohydrates protein, energy value in the fruits of *C. spinosa*. The proximate compositions are: crude fiber (12.6±0.2), ash content (11.3±0.2), moisture content (25±1.15), carbohydrates (60.28±4.6), Protein (12.5±0.9), Lipid (0.18±0.01) Energy value (292±0.7 kcal/100g). There is evidence that dietary fiber has a number of beneficial effects related to its indigestibility in the small intestine [18]. Carbohydrates are one of the most important components in many fruits. The results obtained showed that *C. spinosa* has high Energy value and carbohydrates contents while crude fiber, ash content, moisture content, lipid and protein content were low.

3.2 Phytochemical Screening

The preliminary screening showed in Table-2. The increasing interest in the phytochemical compound which could be relevant to their nutritional incidence and their role in health and diseases. Phytochemical analyzed on the *Catunaregam spinosa* fruits extracts shows the presence of bioactive compound which are known to reveal medicinal properties as well as physiological activities [12]. Screening of the fruits extracts is to find out the presence of phytochemicals such as phenols, tannins, flavonoids, saponins, cardiac glycosides, steroids, terpenoids, phlobatannins and alkaloids. Ten

phytochemical tested to show the presence and absence of these active compounds in the fruits extracts**.** Ethanol extracts have the presence of secondary metabolites such as phenol, tannins, flavonoids, alkaloids, saponins, phlobatannins, steroids, terpenoids and cardiac glycosides.

Table 2: Phytochemical Screening of Fruit extract of *Catunaregam spinosa*

S. No	Secondary metabolites	Aqueous	Ethanol	Chloroform
1.	Alkaloid	-	+	-
2.	Flavonoids	+	+	+
3.	Saponin	+	+	+
4.	Phenol	+	+	+
5.	Tannins	+	+	+
6.	Anthraquinones	-	-	-
7.	Cardiac Glycosides	-	+	-
8.	Phlobatannins	+	+	+
9.	Terpenoids	+	+	+
10.	Steroid	-	+	-

3.3 Antibacterial activity

The antibacterial screening of the *Catunaregam spinosa* fruit extracts were performed against *Klebseila pneumonia, Pseudomonas aeruginosa, Escherichia coli, Streptococcus pyogenes, Salmonella typhi* and *Bacillus subtilis* by the disc diffusion method. The activities of the compounds were compared with standard Chloramphenicol for antibacterial activity. The results of the antibacterial activity of the aqueous, chloroform and ethanolic extracts of *Catunaregam spinosa* against the test organisms are shown in Tables 3. In the case of the ethanol extracts at 100 µg concentration showed more potent antibacterial activity than other extracts tested. Maximum zone of inhibition was observed against the *Bacillus subtilis* of 28.0 mm, followed by *Klebseila pneumonia* of 24.0 mm, and the least was recorded in *Streptococcus pyogenes*. From the chloroform extracts, *Bacillus subtilis* showed maximum antibacterial activity of 26.0 mm zone of inhibition followed by *Pseudomonas aeruginosa* of 16.0 mm and the least was recorded in *Escherichia coli, Klebseila pneumonia* and *Streptococcus pyogenes*. In aqueous extracts, *Klebseila pneumonia* showed highest antibacterial activity of 18.0 mm zone of inhibition, followed by *Bacillus subtilis* of 14.0 mm zone of inhibition, and the least was recorded in *Escherichia coli*, *Pseudomonas aeruginosa* and *Streptococcus pyogenes*. However reported in the present study compaces with the findings of the reported in medicinal plants like [19, 20]. Hence, this fruits of *Catunaregam spinosa* show the high nutritional value, phyto screening analysis of the better remedies against the pathogenic bacteria.

Table 3: Antibacterial activity of the *Catunaregam spinosa* fruit extracts

S. No.	Microorganisms	Std*	Zone of Inhibition (mm)											
			Aqueous				Chloroform				Ethanol			
			25	50	75	100	25	50	75	100	25	50	75	100
	Bacillus subtilis	30	8	10	12	14	18	20	22	26	14	18	24	28
	Escherichia coli	30	2	0	2	6	0	2	2	4	8	10	16	18
	Klebseila pneumonia	30	0	12	12	18	0	0	0	2	14	18	22	24
	Pseudomonas aeruginosa	28	4	0	2	4	6	10	12	16	14	16	20	21
	Salmonella typhi	28	0	0	2	0	2	2	2	0	10	14	16	18
	Streptococcus pyogenes	30	2	0	0	2	2	4	6	2	2	4	6	6

Std* Chloramphenicol

4. Conclusion

The nutritional, Phytochemical and antibacterial activity of *Catunaregam spinosa* fruits extract was studied. From these findings, the scientific evidence is to support traditional medicinal plant for its nutritive value and indicate a promising potential for the development of an antimicrobial drugs. The phytochemical analysis of these plants also helpful for elucidation of bioactive molecules which inhibit the pathogen activity. Hence these *in-vitro* studies concluded that the *Catunaregam spinosa* fruits shows the promising effective to develop the novel antimicrobial drugs.

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