Proximate Composition, Antibacterial and Anthelmintic Activity of *Capsicum frutescens* (L.) Var. *Longa* (Solanaceae) Leaves

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**INTRODUCTION**

Chilies are high in vitamin A and C, but low in calories and sodium. Chilies contain potassium, magnesium and folic acid. Capsaicinoids are responsible for pungency and are considered as active compounds. Capsaicin accounts for about 50 to 70% of the total capsaicinoids. Other components are dihydrocapsaicin, nordihydrocapsaicin, homocapsaicin and homodihydrocapsaicin. When eaten, capsaicin stimulates the release of endorphins, which give a pleasurable feeling. Moreover, chilies are believed to increase blood circulation, relieve rheumatic pain, treat mouth sores and infected wounds, reduce blood clots and aid digestion by stimulating saliva flow.

**ABSTRACT**

Introduction: *Capsicum frutescens* (L.) var. *longum* (Solanaceae) is a well used tropical and subtropical medicinal plant. The present study was undertaken to investigate proximate composition, antibacterial and anthelmintic activity of *C. frutescens* leaves.

Methods: The leaf material was tested for proximate parameters namely carbohydrates, proteins, crude fibre, crude fat, ash and moisture. The powdered leaf material was extracted using methanol. Antibacterial activity of the methanolic extract was tested by agar well diffusion method. Anthelmintic activity of the methanolic extract was studied using Indian earthworm.

Results: The protein, ash, crude fat and crude fibre contents were not appreciably high. The carbohydrate content was comparatively higher. Among microelements, iron was found to be in high concentration. Potassium content was found to be high than phosphorus. The methanolic extract showed dose dependent antibacterial and anthelmintic activity. Among the bacteria studied, *S. aureus* was found to be more susceptible to the methanolic extract followed by *K. pneumoniae* and *P. aeruginosa*. The anthelmintic effect of all concentrations was lesser when compared to standard. Phytochemical analysis revealed saponins, tannins, alkaloids, glycosides and steroids in methanol extract.

Conclusion: The leaves may be used as a good source of carbohydrates and other nutrients. The methanolic extract could be used in the treatment of bacterial and helmintic infections. The presence of various phytochemicals might be responsible for the biological potential of the extract. Further studies on isolation of constituents from the extract and their biological activities are under investigation.

**Key words:** *Capsicum frutescens*, *longum*, Nutritive composition, Agar well diffusion, Anthelmintic, *Pheretima posthuma*.
and gastric juice flow\textsuperscript{[1]} Capsaicin has been tested for its effects on experimental carcinogenesis and mutagenesis. There is no solid evidence showing that chili and capsaicin are carcinogenic in humans. In contrast, many studies reveal substantial antioxidant, antigenotoxic and antitumor effects of chili extracts and capsaicin.\textsuperscript{[2–3]} Therefore, capsaicin is suggested as an important dietary phytochemical with antioxidant and chemopreventive activities. \textit{Capsicum frutescens} (L.) \textit{var. longum} (Solanaceae) is a well used tropical and subtropical medicinal plant. It is known as Bird chilli in English, Kutaviraa in Ayurveda and Surkh mirch in Unani. It is cultivated as a condiment crop. It is more woody and taller with small pod like berries and innumerable small flat seeds. The crimson or orange red fruits are elongate conical some what flattened and very pungent. The pungent principles are present in the flesh rind and seeds as well. It acts as stimulant, accelerates oxygenation of cells, encourages adrenal glands to produce corticosteroids, and increases gastrointestinal secretion. It is also carminative, antispasmodic and antiseptic. It is applied externally, in painful muscle spasms in areas of shoulder, arm and spine; for treating arthritis, rheumatism, neuralgia, lumbago and chilbains. The British Pharmacopoeia reported rubefacient and vasostimulant action. The plant contains hydroxybenzoic acid, hydroxycinnamic acid and ascorbic acid. Fruits contain up to 1% capsaicin.\textsuperscript{[4–5]} The literature survey revealed scanty information on biological activities of \textit{C. frutescens} leaves. The present study was undertaken to investigate the proximate composition, antibacterial and anthelmintic activity of \textit{C. frutescens} leaves.

**MATERIALS AND METHODS**

**Collection and Identification**

The plant material was collected during April 2010 from Hosanagara Taluk of Shivamogga district, Karnataka, India. The plant sample was identified by Dr. K.G Bhat, MGM College, Udupi, Karnataka. A voucher specimen (Voucher No: KU/AB/KA-2036) was deposited in the University Herbaria, PG Department of Studies and Research in Applied Botany, Shankaraghatta-577451 for future reference.

**Solvent Extraction and Phytochemical Analysis**

The leaves were separated from the plants, washed 2–3 times with tap water and once with sterile water, shade dried and powdered. For extraction, a known amount of powder (100 gm) was subjected to soxhlet extraction and exhaustively extracted with methanol for about 48 hours. The extract was filtered, concentrated in vacuum under reduced pressure using rotary flash evaporator and dried. The extract was subjected to phytochemical screening to detect the presence of secondary metabolites.\textsuperscript{[6–7]}

**TABLE 1 : Proximate composition of \textit{C. frutescens} leaves.**

<table>
<thead>
<tr>
<th>PARAMETER</th>
<th>CONTENT</th>
</tr>
</thead>
<tbody>
<tr>
<td>Moisture (%)</td>
<td>26.9</td>
</tr>
<tr>
<td>Ash (%)</td>
<td>1.52</td>
</tr>
<tr>
<td>Crude fibre content (%)</td>
<td>2.68</td>
</tr>
<tr>
<td>Total carbohydrates (%)</td>
<td>49.01</td>
</tr>
<tr>
<td>Total protein (%)</td>
<td>2.68</td>
</tr>
<tr>
<td>Crude Fat (%)</td>
<td>1.52</td>
</tr>
<tr>
<td>Nutritive value (cal/100 g)</td>
<td>290.6</td>
</tr>
</tbody>
</table>

**Determination of Nutritive Composition**

The nutritive composition of powdered leaves was carried out to determine proximate and mineral composition using various techniques. The moisture content was determined by drying powder in oven at 100°C until constant weight, ash by incineration in a muffle furnace at 550°C for 48 h, proteins by nitrogen determination using Kjeldahl method and conversion of nitrogen to proteins by the factor 6.25. Fat was determined by Bligh dyer technique and crude fiber by successive digestion of the defatted sample with 0.26 N sulphuric acid and 0.23 N potassium hydroxide solutions. Percentage carbohydrate was calculated using the formula: 100—(percentage of ash + percentage of moisture + percentage of fat + percentage of protein). Nutritive value was determined by: Nutritive value = 4 × percentage of protein + 9 × percentage of fat + 4 × percentage of carbohydrate.\textsuperscript{[8]}

**Screening for Antibacterial Activity**

The antibacterial efficacy of the methanol extract was tested against \textit{Staphylococcus aureus}, \textit{Klebsiella pneumoniae} and \textit{Pseudomonas aeruginosa} by agar well diffusion method.\textsuperscript{[9]} Briefly, wells of 6 mm were bored in Muller-Hinton agar plates inoculated with 24 hours old broth cultures of test bacteria. The extracts (10, 25, 50 and 100 mg/ml of 10% dimethylsulfoxide (DMSO)), standard (Rifampicin, 1mg/ml) and control (10% DMSO) were added into the labeled wells. The plates were incubated at 37°C for 24 hours in upright position and the zone of inhibition was recorded. Experiment was carried in triplicate and the average reading was noted.
Proximate Composition, Antibacterial and Anthelmintic Activity of *Capsicum frutescens* (L.) Var. *Longa* (Solanaceae) Leaves

Nutritive composition of *C. frutescens* leaves was represented in the Table 1. The carbohydrate content of leaf was found to be 49.01%. The protein content (2.68%) was less. The ash, crude fat and crude fibre contents were not appreciably high. The nutritive value was 290.6 cal/100g.

The result of antibacterial activity of methanol extract is shown in Table 2. Results were recorded as presence or absence of zones of inhibition around the well. The inhibitory zone around the well indicated the absence of bacterial growth and it was reported as positive and absence of zone as negative. In this study, the extract has shown inhibition of test bacteria in a concentration dependent manner. Among bacteria, *S. aureus* was found to be more susceptible to extract followed by *K. pneumoniae* and *P. aeruginosa* as revealed by wider inhibition zones. Standard antibiotic caused more inhibitory activity than methanol extract. No inhibition of test bacteria was observed in case of control and 10 mg/ml concentration of methanol extract. It appears that overall the bacteria were found to be sensitive to extract. The reasons for this could be that the components from the plant were active against microorganisms and are most often obtained through solvent extraction.

### TABLE 2: Antibacterial activity of methanol extract of *C. frutescens* leaves.

<table>
<thead>
<tr>
<th>TREATMENT</th>
<th>CONCENTRATION (mg/ml)</th>
<th>INHIBITION ZONE IN cm</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td><em>S. aureus</em></td>
</tr>
<tr>
<td>Control</td>
<td>10%</td>
<td>0.0</td>
</tr>
<tr>
<td>Standard</td>
<td>1</td>
<td>3.8 ± 0.31</td>
</tr>
<tr>
<td>Methanol extract</td>
<td>10</td>
<td>0.0</td>
</tr>
<tr>
<td></td>
<td>25</td>
<td>0.8 ± 0.13</td>
</tr>
<tr>
<td></td>
<td>50</td>
<td>1.0 ± 0.09</td>
</tr>
<tr>
<td></td>
<td>100</td>
<td>1.3 ± 0.14</td>
</tr>
</tbody>
</table>

Screening for Anthelmintic Activity

The anthelmintic assay was performed on adult Indian earthworm *Pheretima pasthuma* due to its anatomical and physiological resemblance with the intestinal roundworm parasite of human beings. Standard drug (Piperazine citrate, 1%) and different concentrations of methanol extract (10, 25, 50 and 100 mg/ml of DMSO) were poured into labeled petriplates containing saline. A saline control was kept. Six worms of nearly equal size were introduced into each of the plates. Observations were made for the time taken for paralysis and death of worms. Paralysis was said to occur when the worms were not able to move even in normal saline. Death was concluded when the worms lost their motility followed with fading away of their body colors. Death was also confirmed by dipping the worms in slightly warm water.[10,11]

RESULTS

Preliminary phytochemical analysis of *C. frutescens* leaves showed the presence of phytoconstituents namely saponins, tannins, alkaloids, glycosides and steroids in methanol extract.

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### TABLE 3: Anthelmintic activity of methanol extract of *C. frutescens* leaves.

<table>
<thead>
<tr>
<th>EXTRACT/ DRUG</th>
<th>CONCENTRATION (mg/ml)</th>
<th>PARALYSIS TIME (IN MIN)</th>
<th>DEATH TIME (IN MIN)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Saline</td>
<td>0.85%</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Methanol extract</td>
<td>10 mg/ml</td>
<td>198.35 ± 13.09</td>
<td>256.23 ± 29.83</td>
</tr>
<tr>
<td></td>
<td>25 mg/ml</td>
<td>173.21 ± 08.73</td>
<td>221.39 ± 24.12</td>
</tr>
<tr>
<td></td>
<td>50 mg/ml</td>
<td>131.10 ± 29.18</td>
<td>167.16 ± 13.64</td>
</tr>
<tr>
<td></td>
<td>100 mg/ml</td>
<td>110.42 ± 16.93</td>
<td>151.09 ± 18.50</td>
</tr>
<tr>
<td>Standard</td>
<td>1%</td>
<td>85.15 ± 10.24</td>
<td>114.28 ± 08.91</td>
</tr>
</tbody>
</table>
The result of anthelmintic activity of methanol extract of *C. frutescens* leaves is shown in Table 3. The extract exhibited marked anthelmintic activity by causing paralysis and death of worms and the effect was found to be dose dependent. The anthelmintic effect of all the concentrations of extract tested was lesser when compared to standard drug (1% piperazine citrate).

**DISCUSSION**

Medicinal plants, as a group, comprise approximately 8000 species and account for about 50% of all the higher flowering plant species of India. The traditional healers have used this resource since time immemorial for the benefit of mankind. Plants produce a diverse range of bioactive molecules, making them rich source of different types of medicines. Over 50% of all modern clinical drugs are of natural plant origin and natural products play an important role in drug development programs in the pharmaceutical industry. The medicinal value of plants lies in some chemical substances that produce a definite physiological action on the human body. The most important of these bioactive constituents of plants are alkaloids, tannins, flavonoids, and phenolic compounds. Phytomedicines derived from plants have shown great promise in the treatment of various diseases including viral infections. Single and poly herbal preparations have been used throughout history for the treatment of various types of illness. In this study, the preliminary phytochemical analysis of methanol extract of *C. frutescens* leaves showed the presence of phytoconstituents namely saponins, tannins, alkaloids, glycosides and steroids.

Relevant to human existence and living is good nutrition. All human beings require a number of complex organic compounds as added caloric requirements to meet the need for their muscular activities. Carbohydrates, fats and proteins form the major portion of the diet, while minerals and vitamins form comparatively a smaller part. The increasing populations of the world food demands have overwhelmed the available land resources. It has been reported that protein-calories malnutrition deficiencies is a major factor responsible in nutritional pathology. The dietary fibre plays an important role in decreasing the risks of many disorders such as constipation, diabetes, cardiovascular diseases, obesity etc. The carbohydrates are main source and store of energy. They are the starting substances for biological synthesis of many compounds.

Antibiotics provide the main basis for the therapy of infections. Over use of antibiotics has become the major factor for the emergence and dissemination of multi-drug resistant strains. Bacteria have the ability to transmit and acquire resistance. Multi-drug resistant strains of pathogens are widely distributed in hospitals and are increasingly being isolated. Infectious diseases caused by bacteria, fungi, viruses, and parasites remain a major threat to public health, despite tremendous progress in human medicine. Their impact is particularly great in developing countries because of the relative unavailability of medicines and the emergence of widespread drug resistance. Interest in plants with antimicrobial properties has revived as a result of current problems associated with the use of antibiotics. Antimicrobial activities of tannins, flavonoids, saponins, terpenoids and alkaloids have been documented. The methanol extract of *C. frutescens* leaves was found to possess most of these phytoconstituents. The antibacterial activity of extract in this study could be chiefly due to the presence of these phytoconstituents and is suggestive of the possible use of the plant in treatment of bacterial infections as most strains have already developed resistance to most of the currently used antibiotics.

Helminth infections are among the most common infections in man, affecting a large proportion of the world’s population. Parasitic helminthes affect human beings and animals by causing considerable hardship and stunted growth. Most diseases caused by helminthes are of a chronic and debilitating in nature. During the past few decades, despite numerous advances made in understanding the mode of transmission and the treatment of these parasites, there are still no efficient products to control certain helminthes and the indiscriminate use of some drugs has generated several cases of resistance. Furthermore, it has been recognized recently that anthelmintic substances having considerable toxicity to human beings are present in foods derived from livestock, posing a serious threat to human health. The origin of many effective drugs is found in the traditional medicine practices and in view of this several workers have undertaken studies pertaining to testing of natural compounds for their proclaimed anthelmintic activity. The traditional medicines hold a great promise as a source of easily available effective anthelmintic agents to the people, particularly in developing countries, including India. Indigenous system of medicine reports a number of natural sources for their anthelmintic efficacy. However, their scientific evaluation as compared to commercial anthelmintics is limited. Many plants have proven to possess anthelmintic activity in vitro and in vivo. Tannins were found to possess anthelmintic activities. Reported anthelmintic effect of tannins is that they can bind to free proteins in...
the gastrointestinal tract of host animal or glycoprotein on the cuticle of the parasite and may cause death.[30-31] Preliminary phytochemical analysis revealed the presence of tannins in the methanol extract which might be responsible for the anthelmintic effect of extract. The result of the present study is suggestive that the extract could be used in the control of round worm infections such as Ascarisis, hookworm infections etc as the worms used in the study are in anatomical and physiological resemblance with the intestinal round worms.

CONCLUSION

The results of the present study revealed that the leaves contain considerable quantity of carbohydrates. The plant may be used as a source of carbohydrates and other nutrients. The extract could be used in the treatment of bacterial and helmintic infections. The presence of various phytochemicals might be responsible for the biological potential of the extract. Further studies on the isolation of constituents from extract and their biological activities are under investigation.

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REFERENCES