

Qualitative Analysis and Antimicrobial Activities on Leaves of *Senna tora* (L.) Roxb

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ABSTRACT

The plant *Senna tora* (L.) Roxb locally is known as Dan-gywe belongs to family Caesalpiniaceae. The leaves were collected from Mawlamyine University Campus, Mawlamyine Township, Mon State during from November to February 2019. The collected plant was identified and confirmed by using literatures. This research dealt with the study of phytochemical analysis and antimicrobial activities on leaves of *Senna tora* (L.) Roxb. In this paper, the powdered leaves were tested to determine phytochemical properties and antimicrobial activities. According to phytochemical test, alkaloids, amino acid, reducing sugar, phenolic compounds, carbohydrates, starch, glycosides, flavonoids, terpenoids, steroids, tennins and saponins were present. Antimicrobial activities of leaves extracts were determined by using paper disc diffusion method. In these tests, the various solvent extracts (acetone, ethyl acetate, ethanol, methanol, pet-ether and water) of powdered leaves of *Senna tora* (L.) Roxb were investigated on six different microorganisms. Among them, acetone, ethanol and methanol extracts showed the most significant antimicrobial activities against *Aspergillus flavus*. Ethanol extract showed significant activities on six different microorganisms. Methanol extract showed effective activity on *Pseudomonas fluorescens*. Aqueous and pet-ether extracts did not show any antimicrobial activity against on all microorganisms.

Keywords: phytochemical, antimicrobial, *Senna tora* (L.) Roxb

INTRODUCTION

Senna tora (L.) Roxb is annual fetid herb, belongs to family Caesalpiniaceae. The plant is locally known as Dan-gywe in Myanmar (Hundley, 1987 and Kress, *et al*, 2003) and Sickle Senna, Sickle Wild sensitive plant in English name (Kurz,1877). It grows abundantly wild in parts of India, China, Pakistan and Myanmar (Burkill, 1935). It is mostly distributed throughout India, Sri Lanka, West China and tropics, particularly in forest and tribal areas. It's native to Tropical America and Asia only (Cooke, 1903). The whole plant of *Senna tora* (L.) Roxb has been widely used in traditional Indian and South Asian medicine. Young and tender leaves can be cooked as vegetable in soups and salad as Moe-kya-lak-hpak. A decoction of the leaves may be used by the Malays as a mild purgative and as a cure for coughs (Burkill, 1935). The leaves are used in an application for ringworm, skin diseases, hepatoprotective, antioxidant and generally given to children having intestinal order (Kapoor, 2001). In Sri Lanka, the flowers are added to food, used for a natural pesticide in organic farms and as a powder commonly used in the pet food industry (Murni, 2019). The seeds are medicinal throughout the East and used to treat skin disease and laxative. In India, these are pounded and applied for itch and after roasting, used as a substitute for coffee (Burkill, 1935). The roots have a bitter taste and also used as purgative, ringworm and anthelmintic properties (Rastogi, 1993). Herbal plants may save lives if they are used correctly. More than 75% of pure compounds derived from higher plants find their place in modern medicine (Ali, 1998 and Garni, 1998). Herbal medicines are popularized due to their effectiveness, easy availability, low cost and comparatively being devoid of serious toxic effects (Bannerman, 1982).The qualitative analysis of *Senna tora* (L.) Roxb leaves extracts contained bioactive and secondary metabolites components such as tannins, steroids, saponin, terpenoids,

glycosides, flavonoids, phenols, oxalic acids, oxyanthraquinones and anthraquinones derivatives (Murni, 2019). The activity of drug was divided according to the presence of chemical constituents (Ali, 1998). There are more than thousand known phytochemicals. These occur naturally in plants and currently many phytochemicals in clinical trait for a variety of disease (Lewis, 1977). The plants which manifest relatively high levels of antimicrobial action may be sources of compounds that can be used to inhibit the growth of pathogens. Bacterial cells could be killed by the rupture of cell walls and membranes and by the irregular disruption of the intracellular matrix when treated with plant extracts (Murugan *et al.*, 2013). Antimicrobial may be antibacterial, antifungal or antiviral which have different modes of action by which they act to suppress the infection. Plants are the main source of antimicrobials and contain many chemical constituents that effect against different microorganisms (Murni, 2019). In antimicrobial activities, the leaves extract was showed the most effective antimicrobial activity on *Aspergillus flavus*. This is sporophytic and pathogenic fungus which can cause liver cancer through consumption of contaminated feed or aspergillosis through invasive growth in mammals (Sharma, *et al.*, 2010). The fungus is mostly found in soil as sporophytes. It causes significant losses in corn, peanuts, cottonseeds and treenuts (Klich, 2007). Aims and objectives are to investigate the phytochemical properties on the leaves of *Senna tora* (L.) Roxb and to determine the antimicrobial activities of the leaves from the different solvent extracts by using six types of microorganisms.

MATERIALS AND METHODS

The specimens of *Senna tora* (L.) Roxb were collected from Mawlamyine University Campus, Mawlamyine Township, Mon State, during the period from November to February 2019. The collected plant was identified by the help of literatures (Hooker, 1885; Hundley, 1987; Cooke, 1903; Brandis, 1907; Burkill, 1935; Kirtikar and Basu, 1935). In phytochemical and antimicrobial activities tests, the leaves were ground into coarse powder with the help of a suitable mechanical grinder and the powder was stored in a suitable container for extraction. Phytochemical tests of the leaves of *Senna tora* (L.) Roxb has been examined according to the methods mentioned in British Pharmacopoeia (1968), Robinson (1983), Harbone, (1984), Central Council for Research in Unani Formulation (1989), Trease and Evans (2002) and Bandiola (2018). The tests were carried out at the Department of Botany, University of Yangon. It was tested to find out the presence or absence of alkaloids, amino acids, carbohydrates, starches, reducing sugars, phenolic compounds, glycosides, flavonoids, steroids, terpenoids, saponins and tannins. In antimicrobial activities, the air dried powdered leaves of *senna tora* (L.) Roxb were extracted by using, acetone, ethyl acetate, ethanol, methanol, pet-ether and water. The extract was tested on six microorganisms such as, *Aspergillus flavus*, *Bacillus subtilis*, *Candida albicans*, *Escherichia coli*, *Pseudomonas fluorescens* and *Xanthomonas oryzae* by using paper disc diffusion method at the Department of Botany, University of Yangon. In this experiment, assay medium was prepared according to the method described by Cruickshank (1975). Assay medium was boiled, and then 20-25 ml of the medium was poured into each conical flask and plugged with cotton wool and autoclaved at 121 for 15 minutes. Then the flasks were cooled down to 40-45°C and each of 0.3 ml of tested organisms was also added into flask and then, poured into sterilized petridishes. The medium was allowed to set for 2-3 hours. After solidification, paper disc impregnated with sample were applied on the agar plates and incubated at 30 for 24-36 hours. The diameter of inhibition zone appeared around paper disc was measured with the help of a transparent ruler.

Types of microorganisms and diseases

Types of microorganism

1. *Aspergillus flavus*
2. *Bacillus subtilis*
3. *Cndiada albicans*
4. *Escherichia coli*
5. *Pseudomonas fluorescens*
6. *Xanthomonas oryzae*

Diseases

Bronchitis, Aspergillus ear and kernel rot
 Usually pathogenic group, anthrax in animals
 Skin and cardidiasis alimentary tract infection,
 bowel disorders, sores and ring worm.
 Cholera, diarrhoea, dysentery, septic wounds and urinary tract infections
 urinary and respiratory tract infection, soft tissue, burns, bones, joints and ear infections, gastrointestinal infection, bacterial for leaf blight.
 wound infections, boils, food poinson, burn and pneumonia, bacterial for leaf blight

RESULTS

Morphological characters of *Senna tora* (L.) Roxb

An annual fetid herb, slightly woody at the base; stems cylindrical, glabrous. Leaves alternate, unipinnately compound, paripinnate, petiolate; stipules linear, caducous; leaflets 3-4 pairs, opposite; petiolules short; blades obovate-oblong, bases subulate oblique, margins entire, tips mucronate, glabrous. Inflorescence axillary racemes, short, hairy, 2-3 flowered. Flowers bright yellow, bisexual, zygomorphic, pentamerous, hypogynous; bracts linear, acute, caducous. Sepals 5, aposepalous, yellowish green, unequal, oblong to rounded, coriaceous. Petals 5, apopetalous, short claw, unequal, lower two slightly longer, bright yellow, glabrous. Staments 7-10, 7-fertile usually three large and four slightly smaller and 3-sterile or absent, unequal, rarely perfect; filaments filiform to stout; anthers ditheous, basifixed, oblongoid, unequal, dehiscent by apical pores. Pistill 1, ovary superior, oblongoid, unilocular with many ovules, marginal placentation; style short; stigma capitate. Fruits a pod, slender, curved, long, sickle shaped, glabrous. Seeds 20-30, brown or yellowish-brown, hard, rhomboidal, glossy, endospermic. Flowering and fruiting period from November to February.



Fig. (1.1) Habit of *Senna tora* (L.)Roxb



Fig. (1.2) Inflorescences of *Senna tora* (L.)Roxb

Fig.(1.3) Leaves of *Senna tora* (L.)RoxbFig.(1.4) Flowers of *Senna tora* (L.)RoxbFig.(1.5) Stamens and style of *Senna tora* (L.)RoxbFig (1.6) Pods of *Senna tora* (L.)RoxbFig (1.7) Seeds of *Senna tora* (L.)Roxb

Phytochemical tests of the powdered leaves from *Senna tora* (L.) Roxb

Phytochemical tests were concerned with determination of presence or absence of phytochemical in qualitative value. The investigation of these tests confirmed the presence of alkaloids, amino acids, reducing sugar, carbohydrates, phenolic compounds, starches, glycosides, flavonoids, steroids, terpenoids, saponins and tannins.

Table (1). Phytochemical tests of the powdered leaves from *Senna tora* (L.) Roxb

No.	Constituents	Extract	Test Reagents	Observation	Result
1	Amino acids	H ₂ O	Ninhydrin reagent	Pink sport	+
2	Alkaloids	EtOH	1. Dragendroff's reagent 2. Mayer's reagent 3. Wagner's reagent 4. Hager's reagent	Orange ppt White ppt Brown ppt Yellow ppt.	+ + + +
3	Carbohydrates	H ₂ O	10% - naphthol + conc:H ₂ SO ₄	Red ring	+
4	Flavonoids	EtOH	HCl / Mg	Pink color	+
5	Glycosides	EtOH	H ₂ O + NaOH	Yellow Color	+
6	Phenolic compounds	EtOH	H ₂ O + 10% FeCl ₃	Green Color	+
7	Reducing Sugar	H ₂ O	Fehling's solution A and B	Reddish brown ppt.	+
8	Saponins	H ₂ O	Distilled water	Frothing	+
9	Starch	H ₂ O	I ₂ solution	Blue black	+
10	Steroids	EtOH	CHCl ₃ + conc:H ₂ SO ₄	Blue to green	+
11	Tannins	H ₂ O	5% FeCl ₃ + H ₂ SO ₄	Yellowish brown ppt.	+
12	Terpenoids	EtOH	CHCl ₃ + conc:H ₂ SO ₄	Pink colour	+

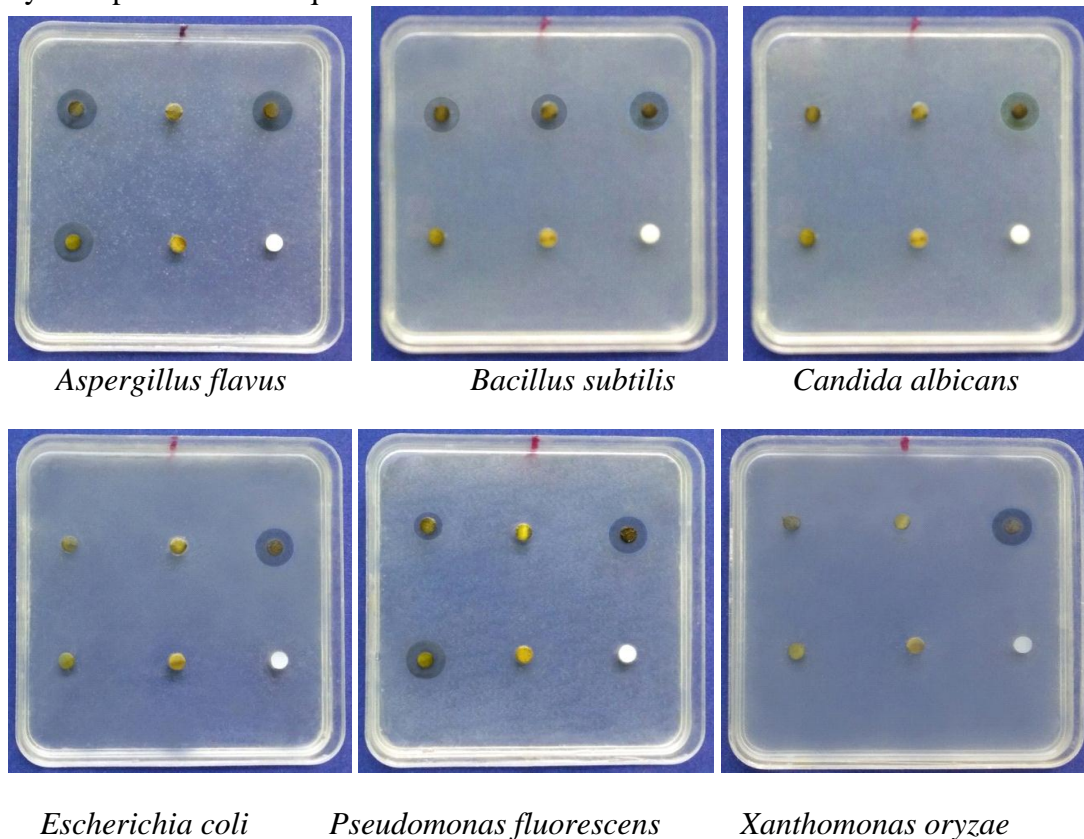
(+) = Presence (-) = Absence

Table (2). Antimicrobial activities of different solvent extracts from *Senna tora* (L.) Roxb

Solvent	<i>Aspergillus flavus</i>	<i>Bacillus subtilis</i>	<i>Candida albicans</i>	<i>E.coil</i>	<i>Pseudomonas fluorescens</i>	<i>Xanthomonas oryzae</i>
Acetone	14mm	10mm	—	—	10mm	—
Ethyl acetate	—	10mm	—	—	—	—
Ethanol	14mm	12mm	12mm	12mm	12mm	12mm
Methanol	14mm	—	—	—	14mm	—
Pet-ether	—	—	—	—	—	—
Water	—	—	—	—	—	—

Paper disc size = 6 mm

According to this result, acetone, ethanol and methanol extracts of the leaves of *Senna tora* (L.) Roxb showed the most significant antimicrobial activities against *Aspergillus flavus*. Methanol extract found to possess most effective activities on *Pseudomonas fluorescens*. Ethanol extract showed significant activities on six different microorganisms. Methanol extract showed effective activity on *Pseudomonas fluorescens* and no inhibition zone was observed for all microorganisms by both pet-ether and aqueous leaves extracts.



DISCUSSION

This paper was carried out to screen for phytochemical analysis and antimicrobial activities on the leaves of *Senna tora* (L.) Roxb extracts. The collected plant *Senna tora* (L.) Roxb (Dan-gywe) was identified and confirmed by the help of literatures (Hooker, 1885; Hundley, 1987; Cooke, 1903; Brandis, 1907; Burkill, 1935; Kirtikar and Basu, 1935). Phytochemical screening of chemical constituents of *Senna tora* (L.) Roxb leaves extracts revealed the presence of phytochemicals which are known to exhibit medical and physiological activities. Murni (2019) stated that the qualitative analysis of *Senna tora* (L.) Roxb leaves extracts contained bioactive and secondary metabolites components such as tannins, steroids, saponin, terpenoids, phenols and mainly the presence of flavonoids and anthraquinone glycoside. Lohar (1975) reported that the leaves of *Senna tora* (L.) Roxb showed the presence of anthraquinone glycosides and flavonoids. Gaykhe, *et al.*, (2018) mentioned that the leaves consist of protein, lipid, amino acids and alkaloids. In this research, amino acid, alkaloids, flavonoids, glycoside, carbohydrates, starches, phenolic compound, reducing sugars, saponins, steroids, tannins and terpenoids were contained in leaves extracts of *Senna tora* (L.) Roxb. Among them, tannins are polyphenolic compounds that interfere with protein synthesis and have shown to have antimicrobial activity. Flavonoids are hydroxylated polyphenolic compounds which found to have antimicrobial activity against an array of microorganisms. Terpenoids mainly used for their aromatic qualities have also been found to be potential agents against inhibiting bacteria. Saponins which are glycosides have been found to have inhibitory effects on gram-positive organism (Murni, 2019). And then the leaves of Dan-gywe are used in the treatment of antioxidant, antifungus, antiarthritic, antidiabetes and anticancer (Gaykhe, *et al.*, 2018). Therefore, the phytochemical analysis revealed that the leaves extracts of *Senna tora* (L.) Roxb have chemical compounds that have been found to possess medicinal properties and antimicrobial activities. In antimicrobial activities, the air dried powdered leaves of *senna tora* (L.) Roxb were extracted by using acetone, ethyl acetate, ethanol, methanol, pet-ether, and water. The extracts were tested on six microorganisms such as, *Aspergillus flavus*, *Bacillus subtilis*, *Candida albicans*, *Escherichia coli*, *Pseudomonas fluorescens* and *Xanthomonas oryzae* by using paper disc diffusion method. In this experiment, acetone, ethanol and methanol extracts of the leaves of *Senna tora* (L.) Roxb showed the most significant antimicrobial activities with zone of inhibition of (14mm) against *Aspergillus flavus*. Methanol extract found to possess most effective activities on zone of inhibition of (14mm) *Pseudomonas fluorescens*. Ethanol extracts were found in more activity on *Bacillus subtilis*, *Candida albicans*, *Escherichia coli*, *Pseudomonas fluorescens* and *Xanthomonas oryzae* (inhibition zone of 12mm). Pet-ether and aqueous extracts did not show any activity against on all microorganisms.

CONCLUSION

The present study was made to detect the presence of reported compounds by using various standard qualitative chemical tests and to look for possible presence of other chemical constituents on the Dan-gywe leaves extracts. In phytochemical analysis, the leaves of *Senna tora* (L.) Roxb contain amino acid, alkaloids, flavonoids, glycoside, carbohydrates, starches, phenolic compound, reducing sugars, saponins, steroids, tannins and terpenoids. Amino acids used for the treatment of liver diseases, fatigue, skeletal muscle damage, cancer prevention, burn and diabetes. Alkaloid has antimalarial, antiasthma, anticancer and hyperglycemic activities. Terpenoid has antimicrobial, antiviral, antifungal, antiparasitic, antioxidant and anti-inflammatory

activities. Therefore, this result indicated due to the presence of these compounds, Dan-gywe leaves possess effective biological properties. In antimicrobial activity tests, acetone, ethanol and methanol extracts were the most significant effects for the extraction of the antimicrobial properties on *Aspergillus flavus*. This fungus produces several mycotoxins, primarily aflatoxin B1 and B2 which cause acute liver damage, cirrhosis and carcinogenic properties. And then, methanol extract found to possess most effective activities on *Pseudomonas fluorescens*. Diseases caused by *Pseudomonas fluorescens* include fever, chills, confusion, rapid heart rate, nausea and vomiting. So, Dan-gywe leaves can prevent diseases caused by *Aspergillus flavus* and *Pseudomonas fluorescens*. The obtained results from this paper *Senna tora* (L.) Roxb leaves contain important chemical constituents that confer upon it as a medicinal agent which has antimicrobial activities. This study provides a support for the uses of Dan-gywe leaves in traditional medicines and its further investigation. The phytochemical analysis and antimicrobial activities as well as inhibitory potential on six microorganisms possessed by *Senna tora* (L.) Roxb leaves highlighted their potential utilization in the development of natural drugs or to treat related diseases or infections.

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