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Ethnobotanical Exploration and Phytochemical analysis of Selected Medicinal Plants Used in Poisonous Snake Bite Treatment.

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ABSTRACT:
The present study was aimed to explore the traditional knowledge and establish the scientific basis of usage of selected plants claimed by the traditional healers for the management of snake bite. Several field surveys were undertaken to document the traditional knowledge of ethnic people of the study area. Total eight plants were selected for preliminary phytochemical analysis. The study holds importance in the midst of antivenom crisis in the snakebite management, since it unravels the scientific basis of use of these plant species for the management of poisonous snake bite which will be useful for searching the lead compounds from natural products as potential antivenom agents.

I. INTRODUCTION:

Snake bite is an important and serious medico legal problem in many parts of the world, especially in South Asian countries. Estimates of global mortality from snake bite have been reported to range from 50,000 per year (Swaroop and Grab, 1954) to 100,000 per year (Chippaux, 1998). Snake bite is a common acute medical emergency faced by rural population in tropical and subtropical countries with heavy rainfall and humid climate (Banerjee, 1978; Bhat, 1974).There are no accurate records available to determine the exact epidemiological or even mortality in snakebite cases in Maharashtra. In a study of snakebite envenoming in rural Maharashtra, Bawaskar et al., (2008) reported that 182 cases of snakebite were admitted during 12 months at 5 hospitals situated in five different districts of rural Maharashtra.

Still today anti snake venom (ASV) is a specific antidote to snake venom actions. Surprisingly enough still it is used in empirical way to manage the poisonous case. In India, polyvalent ASV is available which contain antibody against cobra, Russell's viper, common krait and saw-scaled viper. Antivenoms are highly effective in the neutralization of systemic alterations in snakebite envenomations. However, neutralization of venom-induced local effects is difficult (Warrell, 1992) owing to the rapid development of tissue damage after envenomation and to the delay to reach health centers where antivenoms are available (Gutierrez et al., 1998). Although it is well known that antiserum is invariably unavailable in remote rural Marathwada, the role of medicinal plants remains largely unnoticed and neglected. The use of plants as alternative for treatment of poisonous bites is important in remote areas where there is no accessibility to hospitals and storage facility for antivenoms. Therefore, it is important to explore their materia medica for alternative venom antidotes that could accompany or substitute conventional antivenoms. Plants are popular alternatives for treatment of poisonous snakebites (Nuno et al., 1994; Selvanayagam et al., 1995). Moreover, some of these traditional methods have a scientific basis (Abubakar et al., 2000; Alam and Gomes, 2003).

Many Indian medicinal plants are recommended for the treatment of snake bite (Chopra et al., 1956). In almost any part of the world, where venomous snakes occur, numerous plant species are used as folk medicine to treat snake bite (Martz, 1992). India has long ancient tradition for treating various ailments with the use of medicinal plants. Taking into consideration the above facts and need to explore the plant wealth of Nanded region, immediate need to find alternatives for the treatment of poisonous snakebite and also the investigators personal observation on the therapeutic usage of many plants of this region by the local community, all these circumstances prompted us to design this study. The study was aimed to explore the potential traditional knowledge of medicinal plants being used for treating poisonous snakebites in different indigenous groups in Nanded district of Marathwada region.

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II. MATERIAL AND METHODS:

1) Field Survey and documentation of traditional knowledge:

Field surveys were undertaken to gather data on the traditional use of medicinal plant species across various traditional healers in Nanded district. Traditional healers, called 'Vaidya' or 'Janata' from different indigenous groups were targeted for documentation of the uses of medicinal plants. Regular visit to local traditional healers is arranged for observation of their treatment method. Local traditional healers having practical knowledge of plants in treating the snakebite were interviewed in four villages of the district. The information was gathered from five traditional vaidyas. Since only these vaidyas are well-known in this region for treating snakebite patients over several years and on the basis of the knowledge they have and beliefs of local people, the traditional healers were selected for interviews.

The ethnomedicinal data were collected through general conversations with the vaidyas. The questionnaires were used to obtain information on medicinal plants with their local names, parts used, mode of preparation and administration. No monetary compensation has been given to the traditional healers for providing the medicinal information.

2) Plant collection and identification:

The standard method was followed with regard to collection of plant material, drying, mounting, preparation and preservation of plant specimens (Jain and Rao, 1976). The identification and nomenclature of the plants were based on the Flora of Marathwada (Naik, 1998). The selected medicinal plants are Argemone Mexicana L., Amorphophallus sylvaticus (Roxb.) Kunth, Calotropis gigantea L., Caesalpinia bonduc (L.) Roxb., Dioscorea pentaphylla L., Eclipta alba L., Solanum nigrum L. subsp. Nigrum and Momordica dioica Roxb., The voucher specimens with catalog number were deposited in the herbarium of Department of Botany, Yeshwant Mahavidyalaya, Nanded.431602.

3) Phytochemical screening of selected medicinal plants:

The qualitative phytochemical tests were carried out to identify the active constituent as described by Sofowara (1993), Trease and Evans (1989) and Harborne (1973). The quantitative phytochemical tests were performed for tannin (Van-Burden and Robinson, 1981), saponin (Obadoni and Ochuko, 2001), flavonoid (Bohm and Kocipai-Abyazan, 1974), alkaloids (Harborne, 1973) and phenols (Lowry et al., 1951).

III. RESULTS AND DISCUSSION:

The Phytochemical characters of the eight medicinal plants clearly revealed the presence of Tannin in five plants viz., Argemone mexicana, Caesalpinia bonduc, Dioscorea pentaphylla, Eclipta alba, Momordica dioica, Solanum nigrum. The quantitative analysis of the selected plants shows that, the plant Argemone mexicana contains more amount of tannins (9.40 gm/100gm) as compare to other plants. The plant Dioscorea pentaphylla has second largest content of tannins (8.92gm/100gm) while the plants viz., Solanum nigrum (8.88gm/100gm), and Eclipta alba (8.80gm/100gm) have at par concentration of tannins. Most of the biological properties of tannins are linked to their ability to form complexes with macromolecules particularly with proteins perhaps these astringent characteristic may explain the antivenome activity of the aqueous extracts of the above plants part. Japanese workers reported about the detoxifying action of persimmon tannin from the unripe fruits of Diospyros kaki as an acknowledged medicine against snake venom envenomation in Japan (Okonogi et al., 1979. Therefore results are agreed with the previously conducted experiment for the screening of plants having potential antivenom agent.

The flavonoids an active chemical constituent is present in all plants. Of all the secondary metabolites of complex formation, the flavonoids are probably the most versatile. These compounds embrace a wealth of possibilities of hydrogen bonding arranged around a relatively small carbon skeleton, capable of interacting with molecular targets. Flavonoids have been held responsible for anti-inflammatory, anti-hepatotoxic, anti-allergic, anti-hypertensive, anti-tumour and many other activities and most important in the present context enzyme inhibiting activity. Flavonoids have been shown to inhibit phospholipase A_2, an important component of snake venom (Alcaraz & Hoult, 1985). In the present study, tested medicinal plant Argemone mexicana, a flavonoid isorhamnetin is cited as an anti-snake venom activity (Mors et al., 2000). Presence of flavonoids revealed its higher content in Argemone mexicana (17.60gm/100gm), Amorphophallus sylvaticus (12.61gm/100gm) and Solanum nigrum (12.06gm/100gm). Lowest concentration was observed in Eclipta alba (2.43 gm/100gm).

The alkaloids are considered as active chemical constituent found in all the tested plants

except Argemone Mexicana and caesalpinia bonduc. The highest concentration of alkaloids is present in Momodica dioica (23.20gm/100gm) followed by Solanum nigrum (20.80 gm/100gm). The result clearly indicated and agreed with the literature that these plants can be exploit for detoxifying and antihypertensive properties since alkaloids is known to be effective for these purposes (Trease and Evans, 1978). There are several reports on the use of roots of Calotropis gigantea as a remedy for snake bite or scorpion bite and anti-pyretic, analgesic, anti-inflammatory, purgative, and muscle relaxant activities (Ahmed et al., 2005). The alkaloids stimulate the vasomotor and respiratory centers in brain. The alkaloids produces immediate predominant but short lived cardio depressant effects and a weak but prolonged cardiotonic effect in heart (Mohanty et al, 2004). The results clearly indicated that the plants selected are worth for the present study.

The phenol content is more in Eclipta alba (320 µg/ml) followed by Argemone mexicana (310 µg/ml). The plants viz., Caesalpinia bonduc (280 µg/ml) and Momordica dioica (280 µg/ml) have at par concentration of phenols. Phenolic compounds are important constituents of anti-snake venom plants. The phenolic compound 4-O-methyl ether was identified as a snake venom neutralizing factor in the Indian anti-snake venom plant Hemidesmus indicus and Primula denticulate (Alam et al., 1994). Another phenolic compound caffeic acid present in Echinacea sp. Known since ancient times for their anti-snake venom activity by North Americans (Mors et al., 2000).

Certainly naturally occurring substances are known to modify the actions of proteins and enzymes, especially the polyphenols (Haslam, 1996). Several plant constituents like flavonoids, quinonoid, xanthene, polyphenols and terpenoids possessed protein binding and enzyme inhibiting properties (Havsteen, 1983; Selvanayagam et al., 1994), which also inhibit snake venom phospholipase A_2 activities (Alcaraz and Hoult, 1985). Therefore, phytochemical profile of the selected medicinal plants clearly revealed that Argemone mexicana, Caesalpinia bonduc, Solanum nigru and, Calotropis gigantea have more content of active chemical constituents and therefore these plants may be more useful for the purposes or objective of the proposed the study.

IV. CONCLUSION:

Conclusively it can be summarized that, the phytochemical screening (Qualitative and Quantitative) of selected medicinal plants and the compilation of traditional literature may be helpful for the development and designing of a novel, safe and effective antivenom agents. Moreover, the investigations made here in can be utilized as supplementary information for formulation of antivenom herbal therapy. Such attempts may provide immediate rescue especially to the snakebite victims such as farmers and labours who are working in the field.

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Table 1: Details of medicinal plants used by traditional healers.

Name of the Plant	Local Name	Family	Part used	Type of snake
Argemone mexicana	Bilayati, Patri, Pivala Dhotra	Papaveraceae	Leaves	Krait and Viper
Amorphophallus sylvaticus	Jungli Suran	Araceae	corm	Krait and Viper
Calotropis gigantea	Pandhari Ruchki, Mandar	Asclepiadaceae	Tender Leaves	Cobra
Caesalpinia bonduc	Gajuka	Caesalpiniaceae	Leaves	Krait and Viper
Dioscorea pentaphylla	Paratwel	Dioscoreaceae	Root tuber	Krait, cobra and Viper
Eclipta alba	Maka	Asteraceae	Leaves	Krait and Viper
Momordica dioica	Ran Karli	Cucurbitaceae	Root tuber	Krait and Viper
Solanum nigrum	Kamoni	Solanaceae	Leaves	Krait and Viper

Table 2: Qualitative analysis of phytochemicals of the medicinal plants used by traditional healers for treating snake bite.

Sr. No.	Plants	Tannins	Flavonoids	Terpenoids	Cardiac glycocides	Alkaloids	Phenols
1.	Argemone mexicana	++	++	++	++	– –	++
2.	Amorphophallus sylvaticus	– –	++	++	++	++	++
3.	Calotropis gigantea	– –	++	++	++	++	++
4.	Caesalpinia bonduc	++	++	++	++	– –	++
5.	Dioscorea pentaphylla	++	++	++	++	++	++
6.	Eclipta alba	++	++	++	++	++	++
7.	Momordica dioica	++	++	++	– –	++	++
8.	Solanum nigrum	++	++	++	++	++	++

Presence of constituents = ++
Absence of constituents = – –

Table No. 3: Quantitative analysis of the Phytochemicals of the selected medicinal plants

Sr. No.	Plants	Tannins (g/100gm)	Alkaloids (g/100gm)	Flavonoids (g/100gm)	Phenols (µg/ml)
1.	Argemone mexicana	9.40 ± 0.12	--	17.6 ± 0.55	310 ± 0.47
2.	Amorphophallus sylvaticus	--	4.00 ± 1.32	12.61± 0.64	235 ± 0.56
3.	Calotropis gigantea	--	12.80 ± 1.71	3.08 ± 0.98	225 ± 0.63
4.	Caesalpinia bonduc	7.60 ± 0.25	--	4.85 ± 0.33	280 ± 0.97
5.	Dioscorea pentaphylla	8.92 ± 0.98	11.70 ± 0.41	5.67 ± 1.11	265 ± 0.27
6.	Eclipta alba	8.80 ± 1.12	12.00 ± 0.88	2.43 ± 0.66	320 ± 0.53
7.	Momordica dioica	8.40 ± 0.82	23.20 ± 0.25	5.09 ± 0.54	280 ± 0.31
8.	Solanum nigrum	8.88 ± 0.99	20.80 ± 0.22	12.06 ± 0.57	275 ± 0.64

Results presented here are the mean values from three independent experiments ± S.D.,