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The Properties of Selected Medicinal Plants Against *Bovicola ovis* and *Amblyomma varigatum:* A Review

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Abstract: One of the major health concerns affecting livestock production are ectoparasites, particularly lice and ticks. Health problems caused by ectoparasites such as mange mites, lice, keds and ticks result in serious economic losses to smallholder farmers. Ectoparasites are described as the predominant causes of pre-slaughter skin defects and losses in the tanning industry. Lice is a blood sucker causing great irritation to the animal and forcing them to scratch and bite at themselves. Ticks affect livestock directly by causing skin damage opening up wounds which make the animal susceptible to secondary infection, cause toxicosis and tick-borne diseases. Several reports have shown that the use of some of acaricides towards the control of ectoparasites is constrained by growing development of resistance, high toxicity and environmental concerns though their application is the most widely employed approach in many countries. Ethno-veterinary medicine specially the use of medicinal plants could be a promising area of alternative tick and *Bovicola ovis* control strategies. According to WHO estimate, between 35, 000 and 70, 000 plant species are used in folk medicine worldwide. *Calpurnia aurea, Otostegia integrifolia, Nicotiana tabaccum* and *Jatropha curcas* are traditionally used against *B. ovis* and *Amblyoma varigatum* in ethno-veterinary practices. Use of such an alternative means of treatment can promise to substitute the use of synthetic drugs which have a widespread drug resistance especially in developing countries.

Key words: Ectoparasites · *Calpurnia aurea* · *Otostegia integrifolia* · *Nicotiana tabaccum* · *Jatropha curcas*

INTRODUCTION

Parasitic diseases are a global problem and considered as a major obstacle in the health and product performance of livestock [1]. One of the major health concerns affecting livestock are ectoparasites, particularly ticks and tick-borne diseases (TTBD). Ticks are very significant and harmful blood sucking external parasites of mammals, birds and reptiles throughout the world [2]. Ticks affect livestock directly by causing skin damage opening up wounds which make the animal susceptible to secondary infection and cause toxicosis and paralysis in some instances. Indirectly and more importantly, ticks act as vectors of fatal diseases, for example babesiosis and theileriosis [3].

B. ovis is a blood sucker causing great irritation to the sheep and forcing them to scratch and bite at themselves [4]. The parasites are reported to cause discomfort and annoyance that leads to reduction of weight gain, wool growth and milk production due to nervousness and improper nutrition of the animals in which they spend less time feeding. They reduce and down grade the value of the skin. In addition, it reduces the growth rates and also causes paralysis and injuries which lead to secondary infection and expose the animal to cutaneous myiasis. Further economic losses result from the effects of feeding and scratching on the skin with hard nodules (cockle) reducing the value of the hide and causes loss of wool [4].

Ticks and *B. ovis* also creates financial burdens of diagnostic, therapeutic or preventive programs at flock, community and national levels. Ectoparasite infections have been responsible for major economic loss in leather industry in which approximately more than half of the skin reject occurs [5]. All these imply ectoparasites pose serious economic losses to the resource poor farmers, the tanning factories and the national economy [6].

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The widely accepted and most used method of ectoparasite control has been the use of commercial chemical acaricides. In many developing countries, however, the commercial acaricides may be inconsistently available or not available at all [7]. Other challenges emanating from the use of these modern products include the development of widespread host resistance, ever-increasing cost of acaricides, environmental toxicity of chemicals, residuals in animal products and harm to non-target organisms [8]. Issues including pest resistance, product residues, withdrawal of active ingredients, undesirable environmental persistence and unacceptable risks to nontarget organisms are among those driving research to identify alternative approaches. This has compelled researchers to explore other methods that can be used as alternatives. One approach is to explore and integrate the existing conventional methods of health management and ethno-veterinary practices [7].

Ethno-veterinary medicine specially the use of medicinal plants could be a promising area of alternative tick and *B. ovis* control strategies [9]. The World Health Organization estimates that about 80% of the population in developing countries depends on traditional medicine for their primary health care needs [10]. Most of the rural small-holder farmers cannot afford the cost of modern drugs and therefore resort to Indigenous Technical Knowledge [11].

The medicinal properties of plants are due to the presence of certain specific substances, referred to as bioactive principles which may be stored in organs like roots, leaves, stem bark, fruits and seeds [9, 12]. Among the medicinal plants *Calpurnia aurea* (Aition) Benth. *Jatropha curcas* L. (Euphorbiaceae), *Nicotiana tabacum* L. (Solanaceae), were mainly used in ethno-veterinary medicine [13]. Botanical surveys have already shown that *C. aurea, Otostegia integrifolia, N. tabaccum* and *J. curcas* are traditionally used against ectoparasites of livestock in ethno-veterinary practices [14, 15]. Therefore, this paper was commissioned to review acaricidal and insecticide efficacy evaluation of traditionally selected medicinal plants namely *C. aurea, O. integrifolia, N. tabaccum* and *J. curcas* against *A. varigatu and B. ovis.*

An Overall Review of Ectoparasites and Medicinal Plants

Ectoprasitism: The term ectoparasites or external parasites refer to "parasites, with few exceptions, that live or burrow into the surface of their host's epidermis" [16]. Ectoparasites acquire blood meal from their host without penetrating the entire body of their host. It is only the mouth parts that are inserted into the integument of the host. However, there are reports on ticks occurring in the subcuticular regions or deeper regions of the host skin [17]. The association between arthropod ectoparasites and vertebrate hosts may take on a variety of forms. In some cases, the parasite may be totally dependent on the host, alternatively, the parasite may feed, or live only occasionally on the host, without being dependent on it [16].

The effect of skin parasitism usually depends on the size of invading population, on the manner on which the parasite ekes out its existence and the state of nutrition of the host animal when infected. The damage ectoparasites inflict may be mechanical, but the situation is complicated also by host reactions to the presence of the particular parasite, their secretion and excretion. Young animals are generally more susceptible to ectoparasites because of higher region of accessible surface to the body volume and poor grooming behavior [18].

Lice Infestation: The lice belong to the order Phthiraptera which is divided into four suborders; Anoplura, Amblycera, Ischnocera and Rhynchophthirina. Rhynchophthirina is a very small suborder that includes just two species, one of which is a parasite of elephants and the other for warthogs. Amblycera and Ischnocera are known as chewing lice while Anoplura are described as sucking lice [16].

Both biting and sucking lice affect small ruminants. The important species in sheep and goats are found in the genus *Bovicola* and *Linognathus.* Lice usually are unable to survive for more than 1-2 days off their host and tend to remain with a single host animal throughout their lives. Most species of louse are highly host specific and many species specialize in infesting only one part of their host body and transfer to new hosts is by body contact, particularly under condition of close confinement [18]. The most common lice affecting sheep are the sheep body louse (*B. ovis*, formerly called *Damalinia ovis*). The adults are about 1.6 mm long and have a pale brown body with dark bands. The young lice (nymphs) are smaller with a cream colored body and a brown head, but no bands on the body [19].

Obviously, a critical factor in the likelihood of infestations beginning from non-sheep sources is the period for which lice can live away from sheep. Although the standard statement is that lice will not survive for more than 4 to 5 days after removal from sheep, it now seems that the potential period of survival is much longer than this. Laboratory studies were conducted to investigate the relative periods of survival of the different life stage of *B. ovis* and the effects of temperature and the presence of wool on survival period. Large nymphs survived significantly longer than both small nymphs and adults and both age groups of nymphs survived longer than adults. This pattern was consistent across all temperatures. In a comparison of survival, at 4°C, 20°C, 25°C and 36.5°C lice consistently lived longest at 25°C and lice lived significantly longer at both 20°C and 36.5°C than at 4°C. Significant mortalities may also be caused by rapid reversal of temperature gradients in the fleece as sheep walk from shade into sunlight. Eggs fail to hatch at humidities above 90% and if the fleece remains saturated for more than 6 h many nymphs and adults can drown [20].

Bovicola ovis are susceptible to extremes in temperature and humidity and move up and down the wool fiber to accommodate these changes. They prefer to live at 37°C and 70-90% humidity. Above 39°C the number of eggs laid is reduced and at 45°C no eggs are laid. On a hot day the fleece temperature on exposed parts of a sheep, with less than 25 mm of wool, may range from 45°C near the skin to 65°C at the wool tip. These temperatures are too hot for eggs and young lice to survive. Also lice and eggs do not survive extended periods of very low temperatures. Lice and their eggs do not survive for very long off the sheep. Survival of lice in wool on fences and in yards is very short. This is due to lack of food, exposure to sunlight and desiccation as well as temperature fluctuations between night and day [21].

When inspecting sheep for lice, at most times of the year greatest attention should be paid to the sides of the sheep. However, soon after shearing, inspections should also include the neck and lower body regions and areas where longer wool has been left. It should be noted that the chance of detecting lice in the early stages of an infestation is very low. For example, for a sheep with 10 lice, the probability of detecting the infestation by inspecting 10 fleece partings is less than 5%. Even with 40 partings the probability is less than 20% [22].

Biting lice feed on the skin and scurf. Being highly active, *B. ovis* is usually considered to be most pathogenic in sheep and it can cause great irritation so that the sheep are restless and have their grazing interrupted. Rubbing and/or bitting leads to wool loss, excoriations and serum exudation. The exuded serum from the wounds cause wool matting and the wound itself may attract blowflies. *B ovis* infestation in sheep is reported to cause an allergic dermatitis referred to as 'scatter cockle' [23].

Lice infestations in small ruminants in Ethiopia were reported with an overall prevalence of 1.52% in goats and 2% in sheep [24] from central Ethiopia; 0.53% in goats and 0% in sheep [25] from southern range land; 14.25% in goats from Komblocha [26]; and 2.4% in sheep and 28.8% in goats [27]. The louse species identified were 0.8% *B. ovis* and 1.2% *Linognathus* species in sheep and 1.52 % *Linognathus* species in goats in central Ethiopia and 11.54% *Linognathus stenopsis* and 2.71% *B. capre* in goats from Kombolcha. However, results obtained from the examination of fresh sheep pelts showed a much higher infestation rate of 89.55% [28].

The highest prevalence of lice were those recently reported in sheep from Assela by Hailu [29] who identified *Linognathus* spp. (75.5%) and *B. ovis* (67.1%) as well as Asnake *et al*. [30] who recorded *Linognathus ovillus* (14.6%) and *B. ovis* (36.1%). Other reports were *B. ovis* infestation in 15.3% sheep and 27.9% goats and *L. ovillus* in 27.9% sheep in Tigray [31] and *B. ovis* 26.64% in sheep in Wolayta Sodo [32]. In the main, the lice species of sheep and goats identified in several studies conducted so far in Ethiopia were *B. ovis* and *L. stenopsis* [29].

Ticks Infestation: Ticks are obligate, blood feeding ectoparasites of vertebrates, particularly mammals and birds. They belong to three families Ixodiadae, Argasidae and Nutelielidae. Ixodidae, known as hard ticks, contain almost all the species of ticks of veterinary importance. The second family Argasidae, known as soft ticks contains relatively small number of species of veterinary importance [33]. According to Wall and Shearer [16], ticks are primarily parasites of wild animals and only about 10% of the species feed on domestic animals, primarily sheep and cattle.

The body of tick comprises of two main regions: gnathosoma and idiosoma. Gnathosoma includes the basis capituli and mouthparts. The mouthparts consist of a pair of four-segmented palps, a pair of two-segmented chelicerae and a hypostome. Ticks use the chelicerate to penetrate the epidermis of their host and insert the hypostome with retrograde teeth into the wound. The retrograde teeth on the hypostome, together with the cement material secreted by tick's salivary glands, enhances attachment of tick to its host [17].

The life cycle of ticks vary widely. Some species pass their entire life on the host, others pass different stages of the life cycle on successive host and others are parasitic only at the certain stages. Most ticks spend more time off the host, but are totally dependent on the host for sustenance. They are subjected to microenvironment condition when on the ground and thus tend to be more endemic in specific types of area. Ticks can exist for a long period of time without feeding [18].

Attachment to the host causes irritation of the skin, with subsequent ulceration and sometimes secondary bacterial infections. In addition, tick wounds may become infested by screw-worms or other agents of myiasis and are also associated with the spread of bovine dermatophilosis (streptotrichosis) caused by bacteria known as *Dermatophilus congolensis.* Heavy infestations of ticks can result in anaemia, particularly in small animals and the restlessness caused by the presence of large numbers of ticks can lead to a significant loss of weight and condition [34]. Ticks are important vectors for diseases like babesiosis, anaplasmosis and erlichiosis in domestic ruminants. They are known to exacerbate non specific disease symptoms like anemia, toxicosis and paralysis [3].

The tropical bont tick, *A. variegatum* Fabricius, is a three-host tick that originated in Africa. It has since spread to several countries, including the Caribbean islands, where it is known as the 'Senegalese tick' [35].

Amblyomma variegatum, the 'tropical bont tick', are relatively large and have a bright colouration that makes them easily identifiable. They sometimes have bright, yellow-gold colouration that is seen in the males that led to the common name, 'Antigua gold tick'. Females are usually brown and when fully engorged can be the size of a "nutmeg" (approximately 2 to 3 centimeters long) [36].

In females the scutum is smaller with a wide posterior angle and straight sides. Due to the scutum being smaller, it only provides partial coverage of the dorsal surface, which, as feeding or engorgement commences, covers a progressively smaller percentage of her body. The posterior lips of the female genital aperture forms a wide shaped "U" [37].

The 'tropical bont tick' has had a huge effect on the livestock industry, primarily through its transmission of heartwater disease, *Ehrlichia ruminantium* (formerly *Cowdria ruminantium*) and their association with dermatophilosis, *D. congolensis*. The tropical bont tick has also been implicated as a vector or potential vector for several diseases that include Crimean Congo haemorrhagic fever virus, Dugbe virus, yellow fever virus, *Rickettsia africae* (African tick bite fever) and Jos virus. The subsequent damage from the mouthparts predisposes the host to infection from various diseases such as dermatophilosis [35].

In the Caribbean, only heartwater disease and dermatophilosis have yet been detected in the hosts and have demonstrated clinical symptoms. The testing of ticks and seropositive blood tests of cattle have led to the conclusion that African tick bite fever is widespread in the islands, but there have been few positive human case reports [38].

In Ethiopia, ticks occupy the first place amongst the external parasites by the economic loss they incurred when they infest livestock particularly cattle [39]. Extensive surveys have been carried out, in Ethiopia, on the distribution of tick on livestock in different region of the country like Gamogofa [40], the highland areas of Harer and Diredawa [41]. *A. variegatum* is the most widely distributed tick species in Ethiopia. Other tick species such as *Rhipicephalus evertsi*, *Hyalomma marginatum rufipes*, *Hyalomma truncatum*, *Amblyomma cohaerens*, *Amblyomma gemma*, *Amblyomma lepidum* and *Rhipicephalus pulchellus* were also frequently reported in many tick surveys carried out in the country [42].

Ectoparasites Control Methods: Farmers mostly rely on the use of chemical acaricides and repellents to control ticks and limit the production losses. In order to reduce contact between ticks and vertebrate hosts, chemical repellents such as N, N-diethyl-m-toluamide (DEET) and permethrin are extensively used [43]. Organophosphates (diazinon, fampur, phosmet, dichlorvos), synthetic pyrethroids (resmethrin, deltamethrin, permethrin), carbamates (carbofuran, propoxur), growth regulator (fenoxycarb, methroprene), amitraz, fipronil and methandiol that are currently being used for tick control. Although clearly effective at reducing transmission of tick-borne pathogens to livestock, repeated heavy applications of pesticides to hosts can cause considerable mortality in non-target arthropods through environmental contamination [44]. Moreover, evolved resistance to acaricides, which is a well-known problem with mosquitoes, is a persistent issue for tick species such as *Rhipicephallus microplus* that are chronically exposed by virtue of their close association with cattle to which the acaricides are applied [45, 46].

Challenges of Ectoparasites Control: Problems posed by synthetic acaricides, resistant tick are on the rise due especially to increased frequency in the application of acaricides. For instance, *R. microplus* has developed resistance to synthetic pyrethroids and amitraz; amitraz, chlorfenvinphos and cypermethrin against *Boophilus decolouratus* [47]. The resistance mechanism of ticks such as *R. microplus* to acaricides (coumaphos and diazinon) has been linked to an enhanced cytochrome P 450 monooxygenase-mediated detoxification [48]. Environmental pollution is a serious problem posed by the use of synthetic acaricides in tick control. Chemical compounds such as dichloro-diphenyl-trichloro-ethane (DDT), endosulfan and endosulfan sulphate are toxic and bioaccumulate in nature [49].

Alternative Approaches to Chemical Control: Ethno-veterinary plants use for tick control is very important in Africa and other developing countries since a greater proportion of livestock farmers are small-scale and most of these are in rural areas where cultural practices are still preserved. Plant extract preparations are developed by farmers rather than scientists due to lack of finance to purchase synthetic acaricides which force them to depend on traditional methods of tick control [50].

Traditional knowledge on the use of ethno-veterinary plants for tick control is fast disappearing due to the lack of documentation since this type of knowledge has been transferred orally. Furthermore, the efficacy of most plants that have been traditionally used hasn't been scientifically tested. Due to the economic and medical importance of ticks, it is necessary to screen some ethno-veterinary plants that have acaricidal properties and could be used widely. Some of the advantages of promoting research on ethno-veterinary include the development of plant-derived semiochemicals which may be easily accessible by the rural communities and their low toxicity and biodegradability; thus, the need for their conservation. Plants are increasingly being recognized as possible sources of anti-tick agents. The use of plants or plant-based products for the control of arthropod ectoparasites on livestock is widespread among small scale livestock keepers in Africa [51, 52]. This practice is typically community-based and as a result, the plant species used for such purposes may vary from one community to another. Furthermore, knowledge on such practices is orally transferred from one generation to another and often lacks scientific validation. A number of studies have so far been conducted to validate the use of plants for tick control. For instance, most recently Zorloni *et al*. [53] demonstrated that extracts of *C. aurea* leaves used by the Borana people of northern Kenya and Southern Ethiopia to treat lice infestations in humans and calves. Besides, Magano *et al*. [54] and Thembo *et al*. [55] have described that *C. aurea* had anti-tick properties.

Herbal Treatment

Study Plants: Plants have provided the basis for the traditional treatment of different types of diseases and still offer an enormous potential source of new chemotherapeutic agents. Different parts of plants have been used to treat ectoparasites both in animals and man. These are roots, barks, leaves and seeds [15, 14, 56]. The following are plants selected for this study based on their traditional use by farming communities.

***Calpurnia Aurea*:** *Calpurnia aurea* belongs to Fabaceae family and is commonly known as *Natal laburnum*. It is a small, multi-stemmed tree (3-4 m) tall. The leaves are about 15-25 cm long, each bearing 5-20 pairs of ovate to oblong leaflets, light green and 2-5 cm long, ending with a terminal one (Fig. 1). The flowers are bright yellow, in racemes and the fruits are flat brownish pods [57].

This plant is widely distributed in Ethiopia. In Western Ethiopia, the juice of crushed leaves and bark is used for tick control [58]. In South-Western Ethiopia, the leaves of *C. aurea*, mixed with other plant species, are crushed and squeezed to obtain a juice, which is applied through the auricular route for 2 days to treat earache in humans. In the same area, the plant is traditionally used to treat rheumatism [59]. Antibacterial and antioxidant activity of *C. aurea* have been reported [60] and the plant has been used to treat bacterial dermatitis [61].

Calpurnia leaves and powdered roots are used to destroy lice and to relieve itches. Unspecified parts are used to destroy maggots and the leaves are used to treat allergic rashes, particularly those caused by caterpillars. In East Africa, leaf sap is used to destroy maggots in wounds. In Nigeria, the seeds are used to treat abscesses. In Ethiopia, it is used to treat stomach complaints, headache, eye diseases, scabies and skin infection caused by ticks and as an insecticide as well [62].

Calpurnia aurea extracts are used in southern Ethiopia to protect stock against ticks. Acetone, hexane and water leaf extracts of *C. aurea* collected in southern Ethiopia were tested for repellent/attractant and acaricidal properties on unfed adult *R. pulchellus* ticks. In contrast to many other plant species evaluated, *C. aurea* extracts did not have repellent properties, but rather had a slight attractant capacity. With 20% and 10% acetone extracts, all ticks were either killed or their mobility severely compromised after 1 ml of extract was topically applied on

Fig. 1: A representative plant species of *Calpurnia aurea* (Sisay Alemu and Jelalu Kemal, 02/01/2015)

the abdomen. At 5% concentration, 85% of ticks were still affected. However, 10% aqueous solution had a marked effect. The results proved the efficacy of the traditional use of this extract and may lead to a product that can be used commercially to protect animals against tick infestation, under subsistence as well as industrialized conditions [57].

Calpurnia aurea is used for the treatment of amoebic dysentery and diarrhoea in animals, killing head lice in humans and ticks in animals, syphilis, diarrhoea, leishmaniasis, tapeworm, trachoma, *Tinea capitis*, wound, scabies, elephantiasis and different swellings [62]. The antioxidant activities of the stem extract of *C. aurea* as determined by the total phenol, flavonoids and FRAP methods were higher than that of the leaves. On the other hand, the leaf extract of the plant has higher level of total flavonols and proanthocyanidins.

The leaf extract also has higher radical scavenging activity as shown in 1, 1-Diphenyl-2-picrylhydrazyl (DPPH) and 2, 2¿-azinobis-3- ethylbenzothiazoline-6-sulfonic acid (ABTS) assay. The leaf extract showed activity against seven of the bacterial organisms [60]. *C. aurea* are important to most terrestrial arthropods and this may apply even to those whose major habitat is not vegetation. Ticks demonstrate those newly hatched larvae move up vegetation to assist contact with a passing mammalian host. This tick behaviour can be exploited with success for off-host semiochemical-assisted tick control strategy [17].

Otostegia Integrifolia: There are more than 20 species of *Otostegia*. Many of these species have circumboreal distribution in Mediterranean region and Asia. Only a small number of the most common species with the most obvious fruiting bodies have been evaluated for biological activity. There are more than 65 new and novel compounds have been isolated. Naturally, the species of this genus traditionally been used as an ophthalmia, mosquito repellency, antimicrobial activity, antihyperglyceamic activity and antioxidant activity which prevent different kind of ailment [57].

Otostegia integrifolia is widely distributed in Ethiopia and a very common shrub in overgrazed hillsides or old and abandoned farms. The plant is easily recognized and stands out due to the grayish color of the leaves (Fig. 2). The yellow flowers are also showy when in full bloom. Medicinally, the genus *Otostegia* is very important. Also this plant have genuine antiplasmodial activity along with its safety profile observed in the study could make the leaf extract of *O. integrifolia* a potential addition to the ant malarial armamentarium and also provide scientific support for the ethno-medicinal use of the plant [63].

Otostegia integrifolia Benth, commonly known by its vernacular name ‘Tinjut’, is claimed to have insecticidal properties and often used as fumigant for pots and houses [64]. Other reports also indicated the application of the leaves of the plant for the treatment of tonsillitis, uvulitis, lung diseases, stomachache, malaria and hypertension [65, 66]. Moreover, species of genus *Otostegi*a have shown to possess antiulcer, antispasmodic, antidepressant, anxiolytic and sedative activities [65].

In Northern Ethiopia, *Otostegia* is commonly used to smoke utensils for sterilization. It is also a ritual custom for a mother to cleanse herself with the smoke on the tenth day after giving birth to a child before leaving her confinement to resume normal daily activities. Various ethno-botanical surveys have shown the traditional use of the plant as antimalarial [65].

Fig. 2: A representative plant species of *Otostegia integrifolia* (Sisay Alemu and Jelalu Kemal, 08/06/2015)

A B

Fig. 3: Picture demonstrating plant species of *Jatropha curcas.* A; The plant; B: The seed. (Sisay Alemu and Jelalu Kemal 08/03/2015)

Jatropha curcas: *Jetropha curcas* Linn belongs to the family Euphorbiaceae and are used in traditional folklore medicine to cure various ailments in Africa, Asia and Latin America. The plant is native to North America but now thrives well in Africa and Asia. It is easy to establish as it grows relatively quickly with high yields and it is one of the promising biodiesel plants [67] (Fig. 3).

Traditionally, it is used to cure diseases like cancer, piles, snake bites, paralysis and dropsy [68]. The antimicrobial and larvicidal activities of the leaves of the plant [69], stem and bark [70] and the insecticidal property [71] of *J. curcas* seed oil were already reported. There were many previous reports on the acaricidal activity of essential oils from these plants against *R. annulatus* [72].

The antiovipositional and ovicidal effects of *J. curcas* against *Callosobruchus maculatus* Fab were also reported. The larvicidal effect of methanolic leaf extract of *J. curcas* against the first and fourth instar larvae of *Culex quinquefasciatus* was reported [69]. In another study, the acetone extract of *J. curcas* leaves extended the duration of the various larval instars and of pupation of *Aedes aegypti* even at very low concentration and showed toxicity at higher concentrations [73].

The insecticidal and acaricidal activities of *J. curcas* seed oil were previously reported. The oil of *J. curcas* seed was identified as efficacious in controlling sarcoptic mange in sheep when combined with ascorbic acid. *J. curcas* seeds showed high content of unsaponifiable matter and the insecticidal activity of seed oil was attributed to the presence of sterols and triterpene alcohols [74].

The extract of *J. curcas* was highly effective in controlling hatching of eggs laid by the treated ticks. The eggs laid by the treated ticks were apparently glossy in their appearance. Thus, it is evident from the results of study that the oviposition was not at all inhibited but the eclosion was prevented. The ethanolic extracts of *J. curcas* induced a significant concentration dependent decrease in egg mass production and complete blocking of the hatching of the laid ova [75].

Fig. 4: Picture showing plant species of *Nicotiana tabaccum* (Sisay Alemu and Jelalu Kemal, 16/03/2015)

The moulting hormones, ecdysteroids play an important role in the regulation of salivary gland function, production of pheromones, oogenesis and oviposition [76] in ticks. Ecdysone is metabolized by specific cytochrome P450 isozymes to the active form, 20 hydroxy ecdysone (20 HE). This is then transported through haemolymph to the target cells, where it binds to the ecdysone receptor to cause gene transcription [77] several phytochemicals, in particular, the flavones have the potential to interact with the vertebrate estrogen receptor as agonists or antagonists. Apigenin inhibit both mammalian and insect cytochrome P450 isozyme expression and activity. The flavones, apigenins (apigenin 7-O-β-D-neohesperidoside, apigenin 7-O-β- D-galactoside), orientin, vitexin, vicenin II and the biflavone di-C-β-Dglucopyranoside-methylene-(8, 8')-biapigenin were isolated from the leaves of *J. curcas* [78]. The ethanolic extract of the leaves of *J. curcas* at low concentrations can significantly inhibit the hatching of laid eggs and can be considered as a possible alternative for the control of ticks [79].

Nicotiana Tabaccum: Tobacco belongs to the large family Solanaceae, genus *Nicotina*. They are mostly stout herbs of ten feet or more, with a number of them being annual or perennial in growth (Fig. 4). One outstanding feature of the tobacco plant is the extensive leaf area it produces. A number of tobacco genus have found their use in cigar manufacture and as nicotine for use as an insecticide. Local farmers in South-Western Nigeria have found an extract of tobacco plant to be useful in the treatment and prevention of lice [80].

In traditional medicine, *N. tabaccum* is used for the treatment of diseases like toxic-shock syndrome, pneumonia, boils, carbuncles, etc. However, its extracts might also be used in healing of wounds, be effective against methicillin-resistant *Staphylococcus aureus* (MRSA) and controlling certain plant diseases. The ethyl acetate extract of *N. tabaccum* effective against *Ervinia carotovora* [81]. The plant originated from America, however, now, it has been frequently cultivated in the Indian sub-continent where it is called as 'tambaku'. The plant is known throughout the world due its narcotic chattels. Whole herb, dried leaves and stalks of *N. tabaccum* are traditionally used in the subcontinent for its emetic, purgative, analgesic, antispasmodic, sedative and insecticidal properties. *N. tabaccum* is also used in the ethno-veterinary practice as an anthelmintic [82]. Dipeolu and Ndungu [83] have demonstrated acaricidal efficacy of a natural product based on ground mixture of *N. tabaccum* leaves and a mineral salt mined from around Lake Magadi of Kenya. Larvae and nymphs of *R. appendiculatus* were killed on the ears of calves within 24 h and large numbers of adult ticks were found dead *in vitro* within 2-3 days of application of the product.

Hot water, acetone, chloroform and methanol extracts of the leaf of *N. tabaccum* were tested against the larvae of *C. quinquefasciatus* [84]. The crude aqueous and methanol extracts of *N. tabaccum* were investigated *in vitro* and *in vivo* for anthelmintic activity against *Haemonchus contortus* [82]. *N. tabaccum* extracts were tested for pesticidal activity against *Tribolium castaneum* and shown to be very active against *B. microplus* [85].

Plant Extraction and Phytochemical Screening Tests

Plant Extraction: Plant-derived substances have recently become of great interest owing to their versatile applications. Medicinal plants are the richest bio-resource of drugs of traditional systems of medicine, modern medicines, nutraceuticals, food supplements, folk medicines, pharmaceutical intermediates and chemical entities for synthetic drugs [86]. Extraction (as the term is used pharmaceutically) is the separation of medicinally active portions of plant and animal tissues using selective solvents through standard procedures. The products so obtained from plants are relatively complex mixtures of metabolites, in liquid or semisolid state or (after removing the solvent) in dry powder form and are intended for oral or external use. These include classes of preparations known as decoctions, infusions, fluid extracts, tinctures, pilular (semisolid) extracts or powdered extracts. Such preparations have been popularly called galenicals, named after Galen, the second century Greek physician [87].

The purpose of standardized extraction procedures for crude drugs (medicinal plant parts) is to attain the therapeutically desired portions and to eliminate unwanted material by treatment with a selective solvent known as menstruum. The extract thus obtained, after standardization, may be used as medicinal agent as such in the form of tinctures or fluid extracts or further processed to be incorporated in any dosage form such as tablets and capsules. These products contain complex mixture of many medicinal plant metabolites such as alkaloids, glycosides, terpenoids, flavonoids and lignans [88]. The general techniques of medicinal plant extraction include maceration, infusion, percolation, digestion, decoction, hot continuous extraction (Soxhlet), aqueous-alcoholic extraction by fermentation, counter-current extraction, microwave-assisted extraction, ultrasound extraction (sonication), supercritical fluid extraction and phytonic extraction (with hydrofluorocarbon solvents). For aromatic plants, hydrodistillation techniques (water distillation, steam distillation, water and steam distillation), hydrolytic maceration followed by distillation, expression and enfleurage (cold fat extraction) may be employed. Some of the latest extraction methods for aromatic plants include headspace trapping, solid phase micro-extraction, protoplast extraction, microdistillation, thermomicrodistillation and molecular distillation [88].

The basic parameters influencing the quality of an extract are plant part used as starting material, solvent used for extraction and extraction procedure. While effect of extracted plant phytochemicals depends on the nature of the plant material, its origin, degree of processing, moisture content and particle size whereas the variations in different extraction methods that will affect quantity and secondary metabolite composition of an extract depends upon type of extraction, time of extraction temperature, nature of solvent, solvent concentration and polarity [86].

Fresh or dried plant materials can be used as a source for the extraction of secondary plant components. Many authors had reported about plant extract preparation from the fresh plant tissues. The logic behind this came from the ethno medicinal use of fresh plant materials among the traditional and tribal people. But as many plants are used in the dry form (or as an aqueous extract) by traditional healers and due to differences in water content within different plant tissues, plants are usually air dried to a constant weight before extraction. Other researchers dry the plants in the oven at about 40°C for 72 h. In most of the reported works, underground parts (roots, tuber, rhizome, bulb etc.) of a plant were used extensively compared with other above ground parts in search for bioactive compounds possessing antimicrobial properties [56].

Successful determination of biologically active compounds from plant material is largely dependent on the type of solvent used in the extraction procedure. Properties of a good solvent in plant extractions includes, low toxicity, ease of evaporation at low heat, promotion of rapid physiologic absorption of the extract, preservative action, inability to cause the extract to complex or dissociate. The factors affecting the choice of solvent are quantity of phytochemicals to be extracted, rate of extraction, diversity of different compounds extracted, diversity of inhibitory compounds extracted, ease of subsequent handling of the extracts, toxicity of the solvent in the bioassay process, potential health hazard of the extractants [89]. The choice of solvent is influenced by what is intended with the extract. Since the end product will contain traces of residual solvent, the solvent should be non-toxic and should not interfere with the bioassay. The choice will also depend on the targeted compounds to be extracted [56].

Phytochemical Screening: Phytochemicals are bioactive chemicals of plant origin. They are regarded as secondary metabolites because the plants that manufacture them may have little need for them. They are naturally synthesized in all parts of the plant body; bark, leaves, stem, root, flower, fruits, seeds, etc. i.e. any part of the plant body may contain active components [90]. The quantity and quality of phytochemicals present in plant parts may differ from one part to another. In fact, there is lack of information on the distribution of the biological activity in different plant parts essentially related to the difference in distribution of active compounds (or active principles) which are more frequent in some plant parts than in others [91].

Phytochemicals have been recognized as the basis for traditional herbal medicine practiced in the past and currently in vogue in parts of the world [92]. In the search for phytochemicals that may be of benefit to the pharmaceutical industry, researchers sometimes follow leads provided by local healers in a region [56]. Following such leads, plant parts are usually screened for phytochemicals that may be present. The presence of a phytochemical of interest may lead to its further isolation, purification and characterization. Then it can be used as the basis for a new pharmaceutical product. Successful determination of biologically active compounds from plant material is largely dependent on the type of solvent used in the extraction procedure [90].

CONCLUSION

The review demonstrated that the four medicinal plants, *C. aurea, N. tabaccum* and *J. curcas* have remarkable insecticidal and acaricidal activities against *B. ovis* and *A. varigatum* which deserves further studies on isolation and characterization of the responsible active components of the selected medicinal plant materials.

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