

Insecticidal efficacy of lichens and their metabolites—A mini review

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ABSTRACT

Interest in natural products possessing insecticidal activity is increased because of the drawbacks such as high cost, environmental pollution, effects on non-target organisms, and the emergence of resistant pests that are associated with the use of synthetic insecticides. Lichens are composite organisms comprised of a photobiont and a mycobiont. Lichens are used traditionally worldwide and many studies have shown the promising pharmacological properties of lichens, including insecticidal activity. The present review highlights the potential of lichen extracts and their metabolites as insecticidal agents. An extensive literature survey carried out revealed promising insecticidal properties of solvent extracts and metabolites of lichens against plant pests and insect vectors that transmit human diseases. Lichen metabolites such as usnic acid, atranorin, vulpinic acid, fumarprotocetraric acid, barbatic acid, norstictic acid, and diffractaic acid exhibit insecticidal activity. It appears from the literature survey that lichens and their metabolites can be employed as insecticidal agents to prevent and control insect pests that cause damages to plants and transmit diseases such as malaria, dengue, filariasis, and others.

INTRODUCTION

Insects are not only important as the means of pollination in plants (such as bees) but also they are considered potentially dangerous because many insects cause damages to crop (in both pre and postharvest conditions). The problems associated with the pests have originated with the origin of agriculture. Herbivorous insects, especially coleopterans drastically affect plant performance. Many plant pests are polyphagous in nature and feed on various plants including agricultural crops in their larval as well as adult stages. Together with plant pathogens such as bacteria and fungi, insect infestation results in severe damage to crops and in many cases huge economic loss to farmers. Besides, some insects are known to be vectors of transmission of plant diseases (Crawley, 1989; D'Arcy and Nault, 1982; Moyal *et al.*, 1988; Oerke, 2006; Patole, 2017; Popp *et al.*, 2013 Quisenberry and Schotzko, 1994). The crop loss due to insects is quite higher

in developing countries. The available data indicate that insect pests are shown to destroy around 18%–20% of crop production (at a value of >US\$ 470 billion) annually worldwide (Sharma *et al.*, 2017). A study by Oliveira *et al.* (2014) reveals an average annual loss of 7.7% in crop production in Brazil by insects with total annual economic losses of around US\$ 17.7 billion.

Termites are often considered as troublesome as they cause damage to wooden structures. Besides, some insects are also known to be the vectors of transmission of disease-causing agents that cause serious illness to humans. The mosquito genera such as *Aedes*, *Anopheles*, and *Culex* are known to transmit dreadful pathogens which cause severe disease in humans. Diseases such as malaria, dengue, yellow fever, Japanese encephalitis, chikungunya, and filariasis are transmitted through mosquito vectors. Hence, control of insects is a key factor in the management of crop loss and in the prevention of human diseases that are transmitted through insect vectors (Chen and Wilson, 2005; Dar and Wani, 2010; Ghaly and Edwards, 2011; Oberemok *et al.*, 2015; Ricci *et al.*, 2012; Singh *et al.*, 2015; Verma *et al.*, 2009).

The management of these insect pests is commonly achieved using synthetic chemicals. The synthesis and subsequent use of insecticides dramatically decreased the threat of crop

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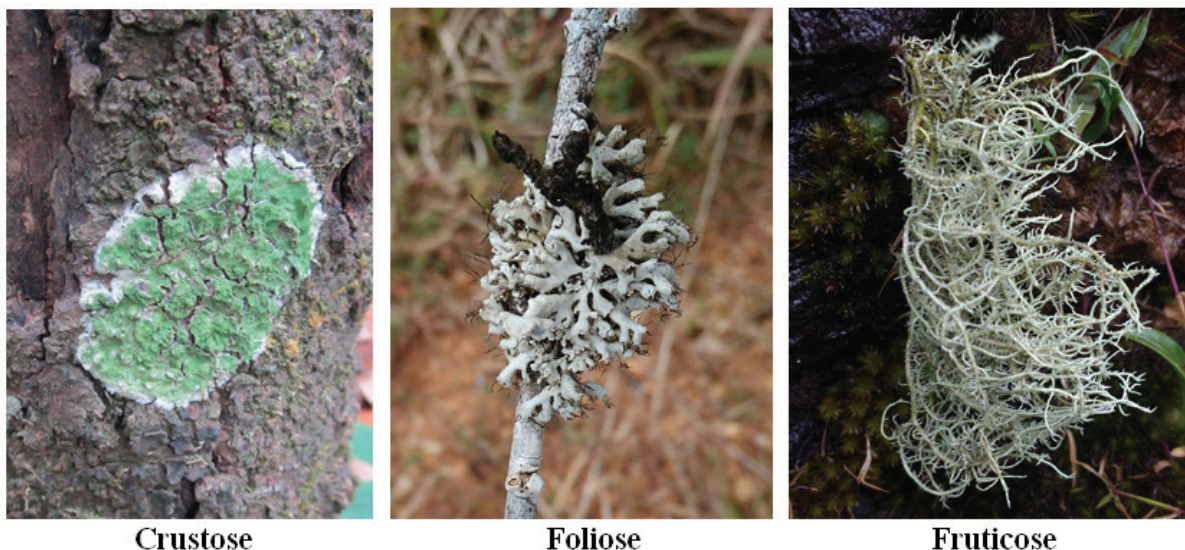


Figure 1. Growth forms of lichens (Photographs by Prashith Kekuda).

damage due to feeding activity of insect pests and also decreased the diseases that are transmitted through insects. These chemical agents may act as ovicidal, larvicidal, and adulticidal agents. Although successful, their usage is often associated with a number of drawbacks such as high cost, residual effect in the environment leading to pollution problems, deleterious effects on non-target organisms, and ill effects in humans and animals through contamination of food and water. Besides, the emergence of resistance in insect pests is one of the most serious concerns with the use of synthetic insecticides. This situation necessitated the search for alternatives which are safe to use and not associated with the development of resistance in insect pests. Biological control is one of the potential alternatives for insect control. Microorganisms (such as bacteria, fungi, and viruses) and natural products including lichens and their metabolites appear to be promising as biopesticides. Biopesticides can replace, at least in part, few of the hazardous chemical agents when used in integrated pest management approaches (Cetin *et al.*, 2008; Ghosh *et al.*, 2012; Gupta and Dikshit, 2010; Kumar, 2012; Nanayakkara *et al.*, 2005; Oliveira *et al.*, 2014; Patel *et al.*, 2016; Ramanujam *et al.*, 2014; Rodriguez-Saona *et al.*, 2016; Verma *et al.*, 2009; Vijayakumar *et al.*, 2010). In this review, a detailed literature survey was carried out to compile information available on the insecticidal activity of lichens and lichen metabolites by referring journals and search engines such as Google Scholar, PubMed, and ScienceDirect.

Lichens

Lichens are composite organisms comprising of a photosynthetic partner (a photobiont, representing an alga or a cyanobacterium) and a fungal partner (a mycobiont, representing an ascomycetes or basidiomycetes member) and represent an ecologically stable symbiosis. Lichens are omnipresent and thrive well even under harsh environmental conditions. Lichens grow on various substrates such as rocks (saxicolous), bark (corticolous), plastic (pasticolous), leaves (foliicolous), and soil (terricolous) and

occur in growth forms viz. crustose, foliose, and fruticose (Fig. 1). It is often known that the lichens growing on rocks accelerate the process of weathering. Lichens are known as indicators of air pollution. Lichens have found tremendous ethnobotanical importance. Many lichens have been used as food and a source of medicine, spice, and dyes since time immemorial. Lichens produce a range of primary and secondary metabolites. Most of the secondary metabolites (lichen substances) produced are unique to lichens and seldom occur in other organisms. Pathways such as acetyl polymalonyl, shikimic acid, and mavalonic acid pathways are involved in the synthesis of lichen metabolites. Extracts and metabolites of lichens exhibit biological activities such as antioxidant, anticancer, insecticidal, herbicidal, enzyme inhibitory, anti-microbial, anti-viral, anti-inflammatory, anti-pyretic, and analgesic activities (Brisdelli *et al.*, 2013; Chen *et al.*, 2000; Devkota *et al.*, 2017; Grube, 2001; Kranter *et al.*, 2008; Kuldeep and Prodyut, 2015; Lucking, 1998; Saklani and Upreti, 1992; Shukla *et al.*, 2014; Valadbeigi *et al.*, 2014; Zavarzina and Zavarzin, 2006).

Insecticidal activity of lichens

A number of studies carried out by various researchers revealed the insecticidal potential of lichens against plant pests (that cause crop damage and transmit plant diseases), pests that damage wood and other items and insect vectors that transmit human diseases. It is shown that crude solvent extracts and purified metabolites of lichens exhibit insecticidal activity against insects that are considered to be plant pests and vectors of many dreadful human diseases. A brief detail on the insecticidal activity of extracts and metabolites of lichens is presented below.

Insecticidal activity of solvent extracts of lichens

Studies have shown that crude solvent extracts of lichens exhibit insecticidal potential against plant pests and mosquito vectors. Methanolic extract obtained from *Ramalina conduplicans*

Table 1. Insecticidal activity of solvent extracts of lichens.

Lichen	Extract	Target insect	Reference
<i>Parmotrema cristiferum</i> and <i>Dirinaria applanata</i>	Methanol extract	II instar larvae of <i>A. aegypti</i>	Kekuda <i>et al.</i> (2015)
<i>Lecanora muralis</i> , <i>Letharia vulpina</i> and <i>Peltigera rufescens</i>	Pooled solvent extract	IV instar larvae and adults of <i>Leptinotarsa decemlineata</i>	Emsen <i>et al.</i> (2013)
<i>Cladonia substellata</i>	Chloroform extract	<i>A. aegypti</i> larvae	Bomfim <i>et al.</i> (2009)
<i>R. montagnei</i>	Hexane, dichloromethane, acetone and ethyl acetate extracts	III instar larvae of <i>Culex quinquefasciatus</i>	Balaji <i>et al.</i> (2012)
<i>Parmotrema tinctorum</i> , <i>Ramalina pacifica</i> , <i>R. nervulosa</i> , <i>Roccella montagnei</i> , <i>Usnea galbinifera</i>	Methanol extract	II instar larvae of <i>A. aegypti</i>	Vinayaka <i>et al.</i> (2009b)
<i>Parmotrema pseudotinctorum</i>	Methanol, chloroform, ethyl acetate, petroleum ether and acetone extract	II instar larvae of <i>A. aegypti</i>	Vinayaka <i>et al.</i> (2010)
<i>Anaptychia ciliaris</i> subsp. <i>ciliaris</i>	n-hexane and methanol extracts	<i>Culiseta longiareolata</i> larvae	Cetin <i>et al.</i> (2013)
<i>Ramalina hossei</i> and <i>R. conduplicans</i>	Methanol extract	II instar larvae of <i>A. aegypti</i>	Kumar <i>et al.</i> (2010)
<i>Ramalina usnea</i>	Methanol extract	III instar larvae of <i>A. aegypti</i>	Moreira <i>et al.</i> (2016)
<i>Heterodermia leucomela</i>	Methanol extract	II and III instar larvae of <i>A. aegypti</i>	Karthik <i>et al.</i> (2011)
<i>Letharia vulpina</i> and <i>Peltigera rufescens</i>	Pooled solvent extract	<i>Sitophilus zeamais</i> adults	Yildirim <i>et al.</i> (2012b)
<i>Parmotrema reticulatum</i> , <i>P. kamatti</i> , <i>P. tinctorum</i> , <i>Parmelia erumpens</i> , <i>Leptogium papilosum</i> , and <i>R. montagnei</i>	Methanol extract	III instar larvae of <i>A. aegypti</i> , <i>Anopheles stephensi</i> , and <i>C. quinquefasciatus</i>	Khader <i>et al.</i> (2018)

is shown to exhibit dose-dependent insecticidal activity against II instar larvae of *Aedes aegypti*. At a concentration of 10 mg/ml and higher, >50% mortality of larvae was observed (Vinayaka *et al.*, 2009a). Solvent extracts of *Roccella montagnei* were tested for insecticidal activity against the larvae of *Helicoverpa armigera* in terms of their effect on insect development. On mixing the extracts at different concentrations to the diet and allowing the larvae to feed, a considerable reduction in larval development, pupation, and moth emergence was observed indicating insecticidal activity (Balaji *et al.*, 2007). Yildirim *et al.* (2012a) observed the insecticidal activity of an extract of *Usnea longissima* against adults of *Sitophilus granarius*. The extract was effective dose dependently and the maximum effect was observed on longer exposure time. At concentration 10 mg/ml and time 96 hours, a mortality rate of 98.98% was observed. The study of Swathi *et al.* (2010) revealed the potential of *Everniastrum cirrhatum* to act against the larvae of *A. aegypti*. It was observed that II instar larvae were susceptible to a higher extent when compared to III instar larvae.

Emsen *et al.* (2012a) screened the extracts of *Cladonia foliacea* and *Flavoparmelia caperata* for insecticidal activity against grain weevil *S. granarius*. The extracts have shown adulticidal activity which was concentration and time-dependent. The LC50 value for *F. caperata* and *C. foliacea* at 96 hours was found to be 0.107 and 0.354 mg/ml, respectively. Emsen *et al.* (2015) evaluated the insecticidal activity of extracts of three lichens viz. *Lecanora muralis*, *Letharia vulpina*, and *Peltigera rufescens* against adults of *Sitophilus granarius* (wheat weevil). The extract treatment resulted in dose dependent mortality of insects with higher activity observed after longer exposure time. The LC50 values for *L. muralis*, *L. vulpina*, and *P. rufescens* extracts were found to be 0.666, 0.505, and 0.328 mg/ml, respectively. Extracts of lichens such as *Cladonia*, *Leptogium*, *Hypogymnea*, *Lecanora*, *Lobaria*, *Pseudocyphellaria*, *Sticta*, *Parmeliella*, *Pannaria*, *Leptroloma*, *Everniastrum*, *Hypotrachyna*,

Parmotrema Pertusaria, *Dirinaria*, *Heterodermia*, *Ramalina*, *Roccella*, *Stereocaulon*, *Ocellularia*, *Caloplaca*, *Myriotrema*, and *Usnea* from Sri Lanka were shown to display insecticidal activity against II instar larvae of *A. aegypti* (Nanayakkara *et al.*, 2005). More information on the insecticidal potential of solvent extracts of lichens is presented in Table 1.

Insecticidal activity of lichen metabolites

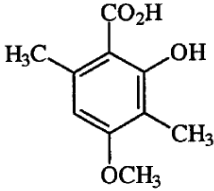
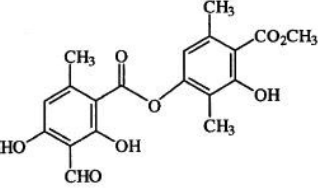
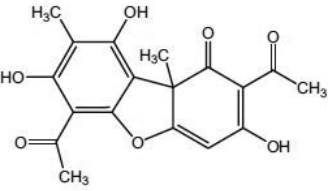
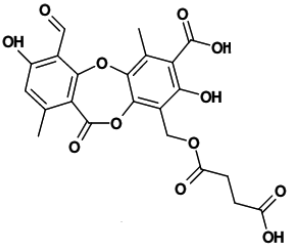
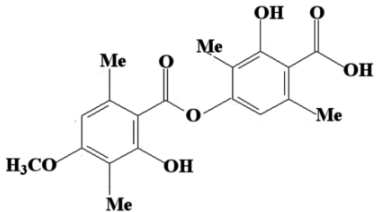
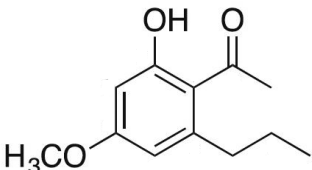
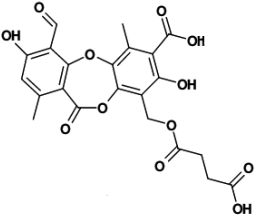
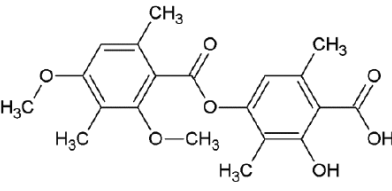
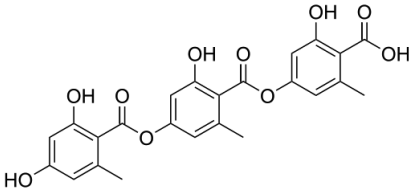
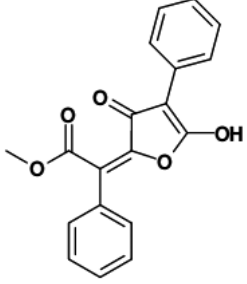
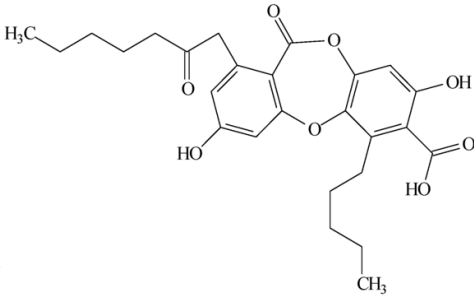
Studies have shown that isolated constituents from lichens exhibit insecticidal activities against a range of insect pests. The antiherbivorous role of lichen compounds such as atranorin, zeorin, lecanoric acid, gyrophoric acid, usnic acid and fumarprotocetraric acid is documented against coleopteran insects (Nimis and Skert, 2006). Lichen compounds viz. (+) and (–)-usnic acid, vulpinic acid, and stictic acid were screened for insecticidal activity against larvae of the polyphagous *Spodoptera littoralis* by toxicity and antifeedant assays. (+) and (–)-usnic acid and vulpinic acid exhibited a strong mortality of larvae while stictic acid did not cause larval mortality (Emmerich *et al.*, 1993). Lichen compounds such as atranorin, parietin, oxyphysodic acid, norstictic acid, fumarprotocetraric acid, calycin and vulpinic acid were shown to exhibit insecticidal activity against polyphagous herbivorous insect *Spodoptera littoralis* (Giez *et al.*, 1994). The lichen compounds viz. usnic acid, vulpinic acid, and physodic acid are shown to exhibit insecticidal activity (Dayan and Romagni, 2001). The lichen metabolites viz. (–)-usnic acid and (+)-usnic acid were shown to be effective against III–IV instar larvae of *Culex pipiens* (Cetin *et al.*, 2008).

Silva *et al.* (2009) purified lectin from *Cladonia verticillaris* and subjected the lectin to insecticidal assay against the termite *Nasutitermes corniger*. The lectin was shown to display termiticidal activity against both soldiers and workers. Cetin *et al.* (2012) revealed the insecticidal potential of lichen substances viz. (+)-Usnic acid, atranorin, 3-hydroxyphysodic acid and gyrophoric acid, against the second and third instar

Table 2. Insecticidal activity of compounds isolated from lichens.

Lichen	Compound	Target insect	Reference
<i>Usnea longissima</i>	Diffractaic acid and usnic acid	<i>S. granarius</i>	Yildirim <i>et al.</i> (2012a)
<i>U. longissima</i>	Diffractaic acid and usnic acid	Larvae and adults of <i>Leptinotarsa decemlineata</i>	Emsen <i>et al.</i> (2012b)
<i>C. substellata</i>	Usnic acid	<i>A. aegypti</i> larvae	Bomfim <i>et al.</i> (2009)
<i>Heterodermia leucomelos</i>	3,6-Dimethyl-2-hydroxy-4-methoxybenzoic acid	II instar larvae of <i>A. aegypti</i>	Kathirgamanathar <i>et al.</i> (2006a)
<i>Pyxine consocians</i>	Cabraleadiol monoacetate, 4-O-methylcryptochlorophaeic acid, lichexanthone	II instar larvae of <i>A. aegypti</i>	Kathirgamanathar <i>et al.</i> (2006a)
<i>Letharia vulpina</i>	Atranorin and vulpinic acid	Larvae of yellow-striped armyworm <i>Spodoptera ornithogalli</i>	Slansky (1979)
<i>U. longissima</i>	Usnic acid and diffractaic acid	<i>S. zeamais</i> adults	Yildirim <i>et al.</i> (2012b)
<i>Leproloma sipmanianum</i>	3,6-dimethyl-2-hydroxy-4-methoxybenzoic acid	II instar larvae of <i>A. aegypti</i>	Kathirgamanathar <i>et al.</i> (2006b)
<i>Lepraria atrotomentosa</i>	(+)-Usnic acid	<i>Glyptotermes dilatatus</i> (termite pest of tea)	Kathirgamanathar <i>et al.</i> (2006b)

Table 3. Lichen metabolites with reported insecticidal activity (Cetin *et al.*, 2008; Kathirgamanathar *et al.*, 2006a; 2006b; Martins *et al.*, 2018; Moreira *et al.*, 2016).

		
3,6-dimethyl-2-hydroxy-4-methoxybenzoic acid	Atranorin	Usnic acid
		
Fumarprotocetraric acid	Barbatic acid	2-hydroxy-4-methoxy-6-propyl-methyl benzoate
		
Norstictic acid	Diffractaic acid	Gyrophoric acid
		
Vulpinic acid	Physodic acid	

larvae of *Culiseta longiareolata* were studied. The toxicity observed was in the order: gyrophoric acid > (+)-usnic acid > atranorin > 3-hydroxyphysodic acid. In a study, compounds viz. 2-hydroxy-4-methoxy-6-propyl-methyl benzoate and the (+)-usnic acid isolated from the methanol extract of *Ramalina usnea* by column chromatography were capable of exhibiting larvicidal activity against III instar larvae of *A. aegypti* with LC50 value of 4.85 and 4.48 µg/ml, respectively (Moreira *et al.*, 2016). Study carried out by Martins *et al.* (2018) showed dose-dependent insecticidal activity of usnic, fumarprotocetraric, and barbatic acids isolated from the lichens *Cladonia substellata*, *C. verticillaris*, and *Cladia aggregata*, respectively, against the termite *Nasutitermes corniger*. It is shown in a recent study that fluorine-containing usnic acid and the fungus *Beauveria bassiana* were shown to exhibit synergistic insecticidal activity against Colorado potato beetle larvae (Kryukov *et al.*, 2018). More information on the insecticidal activity of lichen metabolites is presented in Table 2. Structure of some lichen metabolites (that are reported to possess insecticidal properties) is shown in Table 3.

CONCLUSIONS

The extensive literature review carried out revealed the marked potential of solvent extracts and isolated metabolites of lichens to exhibit insecticidal activity against various pests, especially coleopterans (e.g., *Sitophilus* and *Leptinotarsa*) causing damage to plants and insect vectors such as *Aedes*, *Anopheles*, and *Culex* that transmit dreadful diseases to humans. Lichen compounds such as usnic acid, fumarprotocetraric acid, barbatic acid, vulpinic acid, atranorin, diffractaic acid, gyrophoric acid, and physodic acid are shown to possess insecticidal properties indicating their role against insects. Lichens appear to be promising resources of insecticidal compounds and may be employed to prevent and control insect pests.

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CONFLICTS OF INTEREST

None declared.

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