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Review Article

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RED FLOUR BEETLE, TRIBOLIUM CASTANEUM (HERBST): BIOLOGY AND MANAGEMENT

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ABSTRACT

According to an estimate, the world's population will reach up to 9.1 billion till 2050 and food production is projected to increase up to 70%. Developing countries already facing food scarcity contribute most in population growth. In these countries, one in every six children suffers from hunger and malnutrition. In these critical situation, protection of the grains and food products from insect infestation during pre-harvest and post-harvest is urgently necessary it causes mass losses of grains. In several countries, about 15% of grains are lost harvesting or post-harvesting stages. The Food and Agricultural Organization (FAO) has estimated this loss up to 40% in India. Among the culprits of these losses, coleopteran insect species play major roles. Among coleopterans, red flour beetle, Tribolium castaneum causes loss of grains and food products to larger extent in post-harvest stage. T. castaneum is a secondary pest with a wide host range infesting almong, barley, beans, ground nuts, lentils, maize, oat, peas, rice, rye, sorghum, wheat and other processed grain products. Adults and larvae attack grains already damaged by primary pest and feed mainly on the germ. For the management of T. *castaneum* population, several approaches including synthetic insecticides have been applied with lots of success but leaving several environmental and human health issues too. To minimize these adverse effects, plant derived volatile chemicals i.e. essential oils and its constituents alone or in combination have been used and got promising results. Since these are volatile, non-persistent and biodegradable, essential oil based formulations can be used for the management of T. castaneum at large as green eco-friendly insecticide.

Keywords: Biopesticides, Essential oil, Insecticide resistance, Tribolium castaneum.

INTRODUCTION

About 20,000 species of pests including 40 rodents, 150 fungi, 355 mites, 70 moths and more than 600 species of beetles have been reported to damage of agricultural products both qualitative and quantitative at pre-harvest as well as post-harvest stages (Rajendran, 2005). This is a serious problem especially in developing countries where stored-product insect pests cause severe pre- and postharvest losses estimated to be more than 20% in developing countries and up to 9% in developed countries (Pimentel, 2009). Insects deteriorate grain quality in storage making more susceptible to fungal attack, unsuitable for consumption, planting and trade. Among the stored-product insect pests, members of order Coleoptera and Lepidoptera are common (Boyer et al., 2012). Of these, coleopterans are widely distributed and the most serious pests (Stejskal et al., 2015). Both larvae and adults feed on the grain/grain

products causing serious economic losses. The red flour (Herbst) (Coleoptera: beetle, Tribolium castaneum Tenebrionidae) is one of the most damaging insect species that attack stored grain and food products throughout the world (Andrić et al., 2010; Edde et al., 2012). It is a secondary pest of all grains and primary pests of flour and other processed grain products. It infests usually those grains that had already been damaged by other pests. The grains and products become contaminated affected promting fungal growth. Economic losses include unpleasant smell, reduced weight, reduced trade and nutritional value. Presence of this insect pest may also cause allergic responses.

TAXONOMIC POSITION AND DISTRIBUTION

The red flour beetle, T. castaneum is a secondary pest of stored grain and products throughout the world. It is

regarded as one of the most damaging polyphagous insect pest of stored grains and products (Goodman et al., 2012; Neenah, 2014; Devi and Devi, 2015). The adults are longlived and sometimes living more than three years. Several names of red flour beetle viz. Tribolium navale (Fabricius, 1775), Colydium castaneum (Herbst, 1787), Tenebrio castaneus (Schönherr, 1806). Phaleria castanca (Gyllenhal, 1810), Uloma ferruginea (Dejean, 1821). Margus castaneus (Dejean, 1833). Stene ferruginea (Westwood, 1839) Tribolium and ferrugineum (Wollaston, 1854) are being used as synonyms. T. castaneum is considered to have Indian or Indo-Australian origin with a high dispersion rate (Hill, 2002: Ridlev et al., 2011). It is cosmopolitan, but, more common in warmer regions than to temperate regions of the world (Hill, 2002).

MORPHOLOGY

Adults are small, flat, 2.3-4 mm long and red brown coloured (Figure 1)(Mahroof and Hagstrum, 2012). The antennae of *T. castaneum* are composed of eleven segments, with last three abruptly enlarged segments, in contrast with *T. confusum* in which antennal segment enlarge gradually (Figure 2) (Baldwin and Fasulo, 2014). Thorax is slightly darker than the elytra. Elytra are punctuated containing parallel-sided striations. The males have a setaceous lesion on the ventral surface of front femur (Haines, 1991). Adult bears a functional hind wings often used in flight (Ridley *et al.*, 2011). Larvae are initially whitish, but, later become brown and the size reaches up to 5 mm. Its terminal abdominal segment bears a pair of dark and upturned projections.

HOST

T. castaneum, one of the most cosmopolitan pests of stored products, is polyphagous damaging grains and products both quantity and quality (Bachrouch et al., 2010; Neenah, 2014). It attacks a widespread variety of stored grains and its products including beans, biscuits, cornflakes, dried fruits, lentils, maize, rice, sorghum, wheat and wheat flour (Ranga Rao et al., 2010, Devi and Devi, 2015). The damaged grains and products contain frass, carcasses and exuviae. It also turns grey in colour with a pungent unacceptable odour due to benzoquinones, a defensive chemical secreted from their prothoracic and abdominal glands. All these make the grains and products unsuitable for human consumption decreasing trade value as well (Devi and Devi, 2015). Both larvae and adults feed externally on grains, flours and other processed food products. Its larvae consume mostly the germ of the wheat grains (White and Lambkin, 1988).

LIFE CYCLE AND BIOLOGY

T. castaneum is a holometabolous insect showing eggs, larval, pupal and adult phases in the life cycle. The females mate several times and lay about 300-600 eggs. Eggs are white, microscopic and covered with sticky material to which flour particles adhere. About 5-12 days of

incubation, eggs hatch into larvae. The hatched larvae feed on any available food for about 3-4 weeks and undergo pupation. Pupal phase lasts for 5-9 days. It takes 7 to 12 weeks to complete its life cycle, however, life span depends on temperature and relative humidity. A temperature of 27^{0} C has been found to be the most suitable for development. There are four to five generations per year.

Eggs

Eggs are whitish, microscopic (0.61mm long and 0.3mm width) and cylindrical with remnants of flour particles adhering to the surface with the help of sticky substance present over the eggs shell (Li and Arbogast, 1991; Devi and Devi, 2015). This makes their detection in flour very different. The egg period lasts in about 5-9 days (Devi and Devi, 2015).

Larvae

The larvae are minute, 4-5 mm in length, cylindrical, slender, brownish-white, active feeders and well sclerotised when fully grown. Larval phase includes six to seven instars (Devi and Devi, 2015). Duration of larval phase varies depending upon on availability, type and quality of food, temperature and relative humidity (Haines, 1991). It requires 12-13 days with an average of 12.9 days to complete (Haines, 1991; Devi and Devi, 2015).

Pupae

Pupa is brown coloured, quiescent, non-feeding and without cocoon. Pupal length varies between the sexes. The average body length of male pupa is about 3.18 ± 0.3 mm with a body width of 1.07 ± 0.03 mm. The average body length of female pupa is 4.12 ± 0.01 mm with a body width of 1.15 ± 0.01 mm (Devi and Devi, 2015). Pupal duration also varies according to sex, with male pupal period of 6-7days, while female period lasts in 7-9 days (Devi and Devi, 2015).

Mating behavior

Females of T. castaneum show polyandry (a single female can copulate with several males in the same copulation period). Polyandry increases progeny production as during copulation with several males, female receives large amount of sperms and choose cryptically which sperm is used for fertilization (Fedina and Lewis, 2004). It is also important as many sexually active males are non-virgins and may be sperm-depleted (Pai et al., 2005). Polyandry also increases genetic variability in the population increases genetic variability in the population. Females of different geographic regions often show variations in mating behaviour suggesting that polyandry can be advantageous in some populations but not in others (Pai et al., 2005). Males also select females to mate. Males with large number of odour receptors prefer and select mature virgin females by recognizing non-virigin females through scent produced by the reproductive glands. Virgin females lack such scent glands (Arnaud, 1999).

ECONOMIC IMPORTANCE

T. castaneum damage stored grains and food products causing huge economic loss especially in warmer areas of the world. It infests grains already damaged by other pests or during harvest and storage. The affected product becomes contaminated with faeces, has unpleasant smell and loses nutritional and trade value. Quinones secreted from thoracic and abdominal glands deteriorate grain and product quality to worst. Temperature and moisture content affect feeding behaviour. Increase in temperature by 4^oC from 28 to 32^oC increases weight loss of whole grain two times (Majeed et al., 2016). Increase in moisture content of whole grain from 12.2 to 14.2% increases damage to 1.6 times during the first 60 days, 1.3 times during next 60 days and 1.2 times during the last 60 days of infestation (Daniels, 1956). During development, a single T_{\cdot} castaneum larva consumes 13 mg of wheat flour and adult during their lifetime consumes 315 mg of wheat flour (Hagstrum and Subramanyam, 2000). After consuming the kernel of wheat, a larva feeds on the endosperm (Daniels, 1956). During life, adult consumes the entire germ of 7-12 wheat (Karunakaran et al., 2004). T. castaneum can spread infection of Aspergillus flavus, Aspergillus fumigatus, Cladosporium herbarum, Enterococcus faecalis, Penicillium citinum and P. purpurogenum (Channaiah et al., 2010; Yun et al., 2018).

MANAGEMENT

Detection methods

T. castaneum can be difficult to detect at low densities. Simple traps baited with food such as carob beans, ground nuts and cereals can help in detecting individuals when their density is low. At higher densities, adults and larvae can be seen in holes and tunnels in grains and dust produced during feeding. The eggs stuck to storage containers can also be detected. Presence of a pungent odour indicates high density infestation.

Physical control

Proper sanitation of storage area, removal, destruction of all the sources of infestation and sieving of grains can reduce chances of flour beetle infestation. Addition of inert dusts to the grain causes death of insect by desiccation. Suspected grain can be stored in containers with tight fitting lids or in a freezer for 4-5 days or at temperatures of 50° C. This reduces oxygen levels and increases carbon dioxide level causing death of insect. The application of diatomaceous earth powder to adult *T. castaneum* infesting wheat causes cent percent mortality (Aldryhim, 1990).

Molecular control

The RNAi technique is a new eco-friendly method of insect pest management. In this technique, synthetic RNA are used to target and inhibit specific genes within the insect species. This approach has been applied and found effective against *T. castaneum* (Tang *et al.*, 2016; Gao *et al.*, 2017). However, this method has not yet been marketed due high cost of production of the RNA.

Chemical Control

At present, insecticides like dichlorvos, pyrethrins, pyrethroids and insect growth regulators like methoprene and pyriproxyfen are being used frequently (Sutton et al., 2011). Pyrethrin has found wide applications because of its low toxicity against mammals and high efficacy against a broad range of stored-product insect pests. Commercial formulations used often contain synergist like piperonyl butoxide to increase efficacy of pyrethrins (Brooke, 1958). Insect growth regulators cause malformation or reduces emergence. Applications adult of pyethrins/pyrethroids alone or with insect growth regulators give better results insect management (Mullen, 1999). However, the disadvantage associated with these synthetic chemicals is the residual effects and persistence nature.

Biological Control

Different strains of entomopathogenic fungus have shown some hope in the management of stored grain insect pest. Two entomopathogenic fungi, Metarhizium anisopliae and Beauveria bassiana have been reported to show positive results against many stored grain insect pests including T. castaneum (Golshan et al., 2013; Zamani et al., 2013; Sewify et al., 2014; Abdu-Allah et al., 2015; Rumbos and Athanassiou, 2017; Batta, 2018). Various studies have also conducted to explore the possibility of entomopathogenic fungi either alone or in combination with other strategies and received hopeful outcomes (Wakil et al., 2012; Batta and Kavallieratos, 2018). T. castaneum beetles produce and secrete a mixture of quinone containing methy1-1,4-parabenzoquinone, ethyl-1,4-parabenzoquinones and 1pentadecene (Villaverde et al., 2007). These quinones act as an external immune defence to protect the insect against pathogens.

INSECTICIDE RESISTANCE

A wide variety of insecticides has been applied as the main approach for the control and management of *T. castaneum*. These insecticides target the insect's nervous system including voltage-gated ion channels and acetylcholine esterase causing irreversible disruption of neurological function resulting in death of insects. But the continuous application of these insecticides has developed resistance in most of the target insects. In Indian, the first case of insecticide resistance reported was in T. castaneum against malathion (Bhatia and Pradha, 1971). Since then, high cases of insecticide resistance have been reported in T. castaneum (Dhaliwal and Chawla, 1995; Padhee et al., 2002; Arshad et al., 2019). The development of insecticide resistance is a slow and gradual evolutionary process. After the initial exposure to the insecticide, there is a latent period during which resistance genes are segregated and recombine with other genes that make the conditions favourable for resistance development. Now, the target insect species show an increased level of tolerance to the insecticide. Resistance develops gradually followed by a period of rapid development of resistance resulting in explosive growth of the insect's population. There are a variety of mechanisms that cause insecticide resistance in *T. castaneum*. These are:

i. Target site insensitivity: In this, changes in sensitivity of target site inhibit insecticide binding.

ii. Metabolic resistance: In this, production of increased quantity of enzymes leads to increased metabolic detoxification of insecticide.

iii. Lack of penetration: In this, cuticle thickens or undergoes remodelling which prevents penetration of insecticides.

iv. Behavioural resistance: In this, insects adapt some new behaviour to avoid exposure to insecticides.

Target site insensitive resistence

DDT and pyrethroids target voltage-gated sodium ion channels in insects (Narahasi et al., 1992). Cyclodienes and fipronil bind to the gamma aminobutyric acid receptor (Casida, 1993). Organophosphates, carbamates and pyrethroids produce neurotoxicity by inhibiting activity of acetylcholine esterase (Russell et al., 2004; Pang, 2014). These insecticides also affect other target sites such as voltage-gated sodium ion channels and gamma aminobutyric acid receptors. Resistant insect shows normal neurological functions irrespective of the presence of insecticide as they have evolved insensitive acetylcholine receptors which provide resistance to organophosphate and carbamate insecticides. There are mainly four types of target site insensitivity mechanisms viz. altered Acetylcholine esterase resistance mechanism, knockdown resistance mechanism, reduced gamma amino butyric acid receptor sensitivity mechanism and altered acetylcholine receptors.

Altered acetylcholine esterase resistance mechanism: Acetvlcholine esterase is required to breakdown acetylcholine into acetate and choline to terminate synaptic transmission (Pitman, 1991). This enzyme is a target site of organophosphates and carbamates insecticides, which are bound to a serine residue on the active site of acetylcholine esterase making the enzyme inactive. The inhibition of acetylcholine esterase results in accumulation of acetylcholine at the synaptic cleft leading to prolonged binding of acetylcholine to its postsynaptic receptor. This causes neuroexcitation and produces symptoms like tremors, convulsions and death (Siegfried, 2001). Several organophosphates have been used to protect stored grains from insect infestation. But most of them have developed resistance due to insensitive acetylcholine esterase. Insensitivity of acetylcholine esterase occurs due to point mutations in the active site of Ace genes encoding acetylcholine esterase (Kono and Tomita, 2006).

Knockdown resistance mechanism: Pyrethrin, pyrethroids and DDT target voltage gated sodium ion

channels in insects. These bind to the voltage-gated ion channels in axonal membranes reslting in opening of ion channels for a long time and prolonged sodium influx, thereby, depolarizing the axonal membrane and stimulating the neurons to produce repetitive discharges causing paralysis and death in insects (Soderlund and Bloomquist, 1990). In insects, sodium ion channel is encoded by a single gene known as para (Dong, 2010). Changes in the structure of voltage-gated sodium ion channel occur by point mutation or substitution mutation. It reduces the binding affinity of the insecticides to ion channel protein causing insensitivity.

Metabolic resistance

This resistance cause over production of enzymes like cytochrome *P450* monooxygenases, carboxylesterases and glutathione transferases (Gilbert *et al.*, 2005; Feyereisen, 2006). These enzymes detoxify insecticides before exerting their toxic effect. *T. castaneum* has evolved metabolic resistance for organophosphates, carbamates and pyrethroids (Liang *et al.*, 2015; Huang *et al.*, 2019; Wei *et al.*, 2019).

Cuticle resistance mechanism

Generally, contact insecticide penetrates insect's cuticle and reaches the target site to exert its toxic action (Liu *et al.*, 2006). Cuticle resistance is the result of thickening or remodelling of cuticle which reduces the penetration of insecticide into the insect's body (Koganemaru *et al.*, 2013; Lilly *et al.*, 2016). The over expression of laccases and ABC transporters has been reported to be involved in this mechanism.

Behavioural resistance

In this, behaviour of insect is so modified that it reduces exposure period to insecticides. Guedes et al. (2009) reported higher rates of flight take-off in some insects which increases the insect's survival chancesl by reducing exposure time.

ESSENTIAL OILS

The continuous and indiscriminate applications of synthetic insecticides at large cause bioaccumulation and biomagnification in different environmental components. This adversely and severely affects non-target organisms including human and ecosystems. Therefore, plant based insecticides have been attracted as alternative of these synthetic insecticides. In this continuation, various essential oils and its constituents have been explored as repellent, insecticidal, antifeedant and insect growth regulator activities against a variety of stored grain insects. The essential oils are complex mixture of different chemical compounds with multiple modes of action that enhances their activity due to the synergistic action among constituents (Regnault-Roger, 2012). Since essential oils are volatile, biodegradable and non-persistent in nature, these can be a better candidate for insect pest management.

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About 17500 aromatic plant species and more than 3000 essential oil constituents have been identified (Maddocks-Jennings *et al.*, 2005). These are produced as secondary metabolites in the members of Apiaceae, Asteraceae, Combretaceae, Geraniaceae, Gramineae, Lamiaceae, Myrtaceae, Meliaceae, Piperaceae, Rutaceae, Verbenaceae and Zingiberaceae (Bhattarai and Karki, 2004). These are extracted from plant parts like leaves, flowers, roots, buds, rhizomes etc through steam or water distillation. Generally,

essential oils have densities lower than those of water (Perricone *et al.*, 2015). These are lipophilic and, thus, can enter into insect and cause biochemical dysfunction and mortality (Lee *et al.*, 2004). The toxicity of essential oils does not depend only on the chemical compounds but also on many other factors like route of entry, molecular weights and the mechanisms of action. Essential oils with insecticidal activities can be inhaled, ingested or skin absorbed by insects.

X

Figure 1. Tribolium castaneum

T. castaneum: Last three segments enlarge abruptly

T. confusum: Segments enlarge gradually

Figure 2. Difference between antenna of *T. castaneum* and *T. confusum*

Repellent activity

Class: Insecta

Order: Coleoptera

Genus: *Tribolium* Species: *castaneum*

Family: Tenebrionidae Subfamily: Tenebrioninae

Superfamily: Tenebrionoidea

The repellent activity of several essential oils has been reported against T. castaneum. Repellent behaviour of Baccharis salicifolia essential oil have been observed against T. castaneum adults (Garcia et al., 2005). Tagetes terniflora and Artemisia sieberi oil have been reported effective as repellent against adults and fifth instar of T. (Stefanazzi et al., 2006; Negahban and castaneum Moharamipour, 2007). Laurelia sempervirens and Drimys winteri essential oils repelled T. castaneum adults (Zapata and Smagghe, 2010). Schinus molle and Salvia mirzayanii essential oils have been reported for strong repellent activity against T. confuse adults (Abdel-Sattar et al., 2010; Nikooei and Moharramipour, 2011). Repellent activity of Cuminum cyminum, Piper nigrum, Illicium verum, Myristica fragrans, Foeniculum vulgare, Trachyspermum ammi, Anethum graveolens, Nigella sativa. Zingiber officinale, Piper cubeba, Allium sativum essential oils has been determined against T. castaneum adults (Chaubey, 2007a,b; Shukla et al., 2008; Chaubey, 2011; 2013; 2016). Besides complete essential oils, its pure constituents have also been tested for repellent activities (Kim et al., 2010; Chaubey, 2011).

Insecticidal activity

Since essential oils easily vaporise at room without any biochemical decomposition, these can be used as fumigants. The insecticidal activity of several essential oils has been reported against adults and larvae of *T. castaneum*. Essential oils of *Anethum sowa*, *Artemisia annua*, *Lippia alba* and *Elletaria cardomum* have been reported for fumigant toxicity (Tripathi *et al.*, 2000;

Aggrawal et al., 2000; Verma et al., 2000; Huang et al., 2000). Fumigant toxicity of C. cyminum, P. nigrum, I. verum, M. fragrans, F. vulgare, T. ammi, A. graveolens, N. sativa. Z. officinale, P. cubeba, A. sativum essential oils has been determined against T. castaneum adults (Chaubey, 2007a, b; Shukla et al., 2008; Chaubey, 2011; 2013; 2016). Fumigation of essential oils of Carum copticum and C. carvi causes mortality in T. castaneum (Sahaf et al., 2008; Fang et al., 2010). Besides complete essential oils, pure compounds have also been established for fumigant toxicity. Trans-anethole has been reported to show toxicity against T. castaneum (Koul et al., 2007). The toxic effects of Citrus reticulata and C. sinensis oils compounds have been studied against larvae and adult of T. castaneum (Saleem *et al.*, 2013). Fumigation of β -caryophyllene and α -pinene kills the larvae and adult of T. castaneum (Chaubey, 2012).

Antifeedant activity

Antifeedants are compounds or substances that reduce insect feeding. These substances deter feeding by modification the insect behaviour affecting peripheral sensilla of insects (Isman, 2000). Tripathi *et al.* (2002) have reported feeding deterrence activity of *C. longa* leaf essential oil against adult and larvae of *T. castaneum*. Antifeedant activity of *Eucalyptus globulus* and *Lavandula stoechas* essential oils have been observed against *T. castaneum* (Ebdadollahi, 2011). Huamg and Ho (1998) established antifeedant activity of cinnamaldehyde against *T. castaneum*. Antifeedant activity of 1,8-cineole has been demonstrated in *T. castaneum* (Tripathi *et al.*, 2001). Tripathi *et al.* (2002) have reported antifeedant activity of *Curcuma* leaf oil and d-limonene in *T. castaneum*. Chaubey (2012) has evaluated antifeedant activities of *Zingiber officinale* and *Piper cubeba* essential oils as well as pure compounds, α -pinene and β -caryophyllene against *T. castaneum*.

Insect growth regulators activity

Several essential oils and its constituents have been established for their oviposition, growth inhibitory and progeny production inhibitory activities against different insect pests including T. castaneum. These disrupt mating behaviour, growth and metabolism of insects. Z. officinale and *P. cubeba* oils have been evaluated for developmental inhibitory activities against T. castaneum. These two oils inhibit development of larvae to pupae and the pupae to adults (Chaubey, 2011). Essential oil of A. sativum reduces oviposition potential of adults (Chaubey, 2013). C. cyminum, P. nigrum, F. vulgare, T. ammi, A. graveolens, I. verum, M. fragrans and N. sativa essential oils reduce oviposition potential and increase developmental period of T. castaneum. Fumigation inhibits development of larvae to pupae and the pupae to adults and also result in the deformities in different developmental stages of insect (Chaubey, 2007a, b; Shukla et al., 2008). Ajayi and Olonisakin (2011) have been evaluated ovicidal activity of Syzgium aromaticum, Piper guineense and Xylopia aethiopica essential oils against T. castaneum. The three essential oils are able to reduce progeny emergence of T. castaneum. Besides complete oils, constituents have also shown insect growth regulatory activities. Developmental inhibitory activities of α -pinene and β -caryophyllene alone or in binary combination have been determined against 4th instars larvae of T. castaneum. These compounds reduce transformation of larvae into pupae and pupae into adult (Chaubey, 2012). Carvone suppresses egg hatching in T. castaneum (Tripathi et al., 2003).

Mechanism of action

Essential oils and its constituents disrupt insect physiology in different ways. Some of these inhibit acetylcholine esterase enzyme activity or block octopamine receptors (Enan, 2001; Chaubey, 2013). Ryan and Byrne (1975) have reported that essential oils and monoterpenoids cause insect mortality by reversiblely competitive inhibition of acetylcholine esterase by binding to active site of the enzyme (Ryan and Byrne (1975). Some essential oils block gamma aminobutyric acid and induce toxicity in insects (Priestley et al., 2003). Chaubey (2013) has reported that A. sativum essential oil inhibits acetylcholine esterase activity in T. castaneum adults (Chaubey, 2013). Some monoterpenes such as thujone can induce the neurotoxic effects by acting gamma aminobutyric acid receptors in insects (Hold 2000; Ratra and Casida, 2001). Essential oils can also disrupt communication in mating behaviour of insect by blocking the function of antennal sensilla. This lowers fecundity and ultimately the population of insect pest (Ahmad et al., 2001). Caryophyllene oxide inhibits electron transport system by disturbing respiration rate of insects (Emeki *et al.*, 2004). Thus, in conclusion essential oils and its constituents exert its toxicity in insects by interfering with the nervous co-ordination and respiratory system.

Structure activity relationship

The bioactivities of essential oils and their constituents depend on the functional group and molecular weights (Kumbhar and Dewang, 2001). L-Menthol and its acyl derivatives have been evaluated against stored grain insect pests. It has been reported that menthyl propionate and Lmenthol have high insecticidal activity. High activity of menthyl propionate as compared to L-menthol can be due to increasing number of methyl groups in side chain (Aggarwal et al., 2001). Due to nucleophilic properties of methyl group, increase in number of methyl groups on side chain cause increase in negative charge on carbon atom. Therefore, high activity of menthyl propionate may be due to increasing negative charge on carbon atom because of the presence of two methyl groups. Low activity of menthyl benzoate has been observed because they do not have methyl group and the benzene ring is attached directly to menthyl carbon. Activity of menthyl cinnnamate has been found moderate as it has a double bond and benzene ring is not directly attached to carbon atom (Aggarwal et al., 2001; Tripathi et al., 2009). Benzene derivatives (eugenol, isoeugenol, methyl eugenol, safrole and isosafrole) and terpenes (cineole, limonene, p-cymine and α -pinene) have been evaluated for insecticidal activity (Ngoh et al., 1998). It has been observed that derivatives of benzene have higher insecticidal activity than that of terpenes. This toxicity may be due to the active groups in benzene derivatives. The presence of double bond in the side chain of aromatic ring and the substitution of methoxy group play an important role in the toxicity of these analogues. The knockdown and contact activity has been found to increase in methyl eugenol due to further methoxy group. In contrast, when double bond in side chain is nearer to aromatic ring, fumigant toxicity is decreased. However, benzene derivatives have more insecticidal activity than monoterpenes (Ngoh et al., 1998). Further researches involving structure activity are required to enhance insecticidal potency of essential oils and its constituents. Further studies should be carried out to investigate whether the alteration in structure of essential oil constituents can modify mode of action.

Synergistic action of essential oils

Regnault-Roger (1997) reported that the high toxic effect of essential oil may be due to the synergistic effect of essential oil active compounds. Various studies have been carried out to investigate the synergistic effect of essential oils, active components and their combinations against insects. Khalfi *et al.* (2006) studied the insecticidal activity of spearmint essential oils and two active compounds 1,8 cineole and carvone. They found that the activity is due to the synergistic action of 1,8-cineole and carvone. Abbassy *et al.* (2009) reported that terpinen-4-ol and cterpinene increase the insecticidal activity of profenofos and methomyl. The essential oils are a complex of chemical compounds with multiple modes of action that enhances their activity due to the synergistic action.

CONCLUSION

Essential oils are complex mixtures of volatile compounds produced as secondary metabolite in aromatic plants. These oils and its constituents have repellent, insecticidal, antifeedant and growth regulatory activities in insects. These interfere with the nervous and respiratory system of the insect. Due to their volatile and non-persistent nature, essential oils can be used as an alternative against insect pests of stored grain and food products. Most of these oils are selective in their role with little or no harmful effect on the environment and the non-target organisms including human. Thus, essential oils can be used as alternatives in the management of red flour beetle, *T. castaneum*.

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