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Effects of Phytochemicals in the Post Embryonic Development of the Lesser Mealworm - *Alphitobius Diaperinus* (Panzer) (Insecta: Coleoptera: Tenebrionidae)

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Abstract: **Coleopteran insects constitute the major insects causing havoc damage to the stored food grains both for human consumption and sowing. Lesser mealworm (*Alphitobius diaperinus*) is considered as a minor pest causing damage to different stored grains especially in poorly maintained grain processing warehouses. Additionally it is an important pest of poultry houses where it lives in poultry droppings and litter. Its larvae and adults are also suspected to be health risk to humans as they are the vector of pathogens of many viruses, bacteria and protozoa causing serious diseases in birds. Synthetic insecticides are widely used for the control of stored grain coleopteran pests but development of pesticide resistance, accumulation of toxic residues in food and consequent health hazards creating more and more problems than resolving them due to indiscriminate use of broad-spectrum insecticides has been creating serious health hazards. These problems have led the scientists to introduce new insecticides that are equally lethal to the insects but very less harmful to the consumers. With a view to this objective the insecticidal efficacy of neem, eucalyptus oil, sesame oil and mustard oil have been tested to report their various physiological effects on *A. diaperinus* during post embryonic developmental stages. Adults are broadly-oval, moderately convex, black - 5.8 to 6.3 mm long in length. Eggs are about 1.5 mm in length, creamy white with slightly rounded ends. Their incubation period is 6-7 days. Larvae show 6 instars attaining 7 - 11 mm long at last instar. Pupae are creamy white to tan coloured, 6-8 mm long, with 6 days duration; the pharate adults appear brown that gradually become black. The larvae show decline in survivality; the size and weight gradually increase in successive instars. Treatment with different neem, eucalyptus oil, sesame oil and mustard oil depicted substantial decline in survival rate and sizes of larval instars starting from 4th instars. Increased duration of respective instars actually starts from third instars and increased duration of respective instars is maximum following nimbecidine treatment and minimum in mustard oil treatment.**

Keywords: **Lesser mealworm, *Alphitobius*, Phytoproducts, Neem, Eucalyptus oil, Sesame oil, Mustard oil.**

1. INTRODUCTION

A variety of stored grain insect pests render the stored food completely unfit for consumption as well as for sowing that affect the quality and quantity of the concerned seeds. According to FAO report 10% of the stored foods are lost in this way [1]. Such high levels of losses are attributed to the unscientific post-harvest management practices and poorly designed storage programme. With the advent of chemical insecticides, the most commonly used method of controlling stored grain pests is the application of synthetic contact insecticides and fumigants [2]. The indiscrimi- nate use of broad-spectrum insecticides has created more problems than resolving them. The development of pesti- cide resistance, accumulation of toxic residues in food, serious health hazards for human applicators and undesirable environmental pollution are acute critical problems that have drawn attention of the scientists to adopt alternative, natural pesticides that are safer and environmentally acceptable [3], [4]. Constant scientific survey eventually have recommended use of ecofriendly alternatives, with a special focus on the plant-derived insecticidal compounds [5], [6]. Botanical insecticides have long been treated as alternative to synthetic chemical insecticides for pest management, because these are less harmful to the environment due to their biodegradation [3]. The use of these traditional materials has recently stimulated research to establish the scientific basis for their continued use regarding their efficacy, active constituents and effective application technology [7]. But in spite of the huge potentialities of phytoproducts as insecticides, only a handful of such compounds are currently used in post harvest management of seeds [7].

Over the past three decades, neem, *Azadirachta indica* has come under close scientific scrutiny as a source of natural pesticide [8]. Neem products have been shown to be effective protectants of grains against infestation by grain weevils, grain borers armyworm and several species of storage moths [9]. The characteristic odour of neem mate rials permitting the closed storage environment presumably repelled insects and bitter compounds in neem materials mixed with the stored grain discouraged insect feeding. Probably, the oil present in neem seed or kernel also inhibits egg deposition on grains. Singh (1993) [10] has reviewed the role of neem products on the behaviour, growth, development, survival and reproduction of stored grain insects. The advantage of neem treatment is that it does not impair the germination of stored seed [11].

In addition to neem, attentions have also been drawn for the pesticidal properties of eucalyptus oil [12]. [13], [14], [15]; oil of *Sesamum indicum* [16], [17]; Cucurmin [18], Mustard [19]; Garlic is considered as the heavenly spice having innumerable medical use. It has proved to be a potent antimicrobial and insecticidal agent [20], [21], [22], [23]. Essential oil from chenopodium, sesame and garlic were also tested against different insect pests [24].

Of all the stored grain insects, Coleopteran insects constitute a major percentage of the stored grain major and minor pests. The lesser mealworm, *Alphitobius diaperinus* (Panzer), is a cosmopolitan stored product minor pest of wheat, barley, millet, cottonseed, oatmeal, soybeans, cowpeas, oilseed products, and tobacco especially in poorly maintai- ned grain processing warehouses. It is well adapted for warm, humid conditions. Additionally it is an important inhabitant of poultry or brooder houses where it lives in poultry droppings and litter and is considered a significant pest in the poultry industry [25]. The beetle also consumes birds' feed and irritates them by biting them [26].

Alphitobius is also known to be vector of many pathogens. It spreads many bird diseases [27]. It also transmits different animal viruses [28]. It also transmits bacteria like *Campylobacter jejuni*, *Salmonella typhi*, *E. coli*, and *Staphylococcus* [29]. This beetle can become a household pest if old litter is used as compost.

In spite of different attempts to explore different possible measures to control *Alphitobius*, [30], [31], [32], [33], no attempt has been made to show deleterious effects of phytoproducts on this pest. With a view to this unexplored field, present investigation was undertaken to report the effects of nimbecidine, sesame oil, mustard oil and eucalyptus oil on the post embryonic developmental stages and fecundity of *A. diaperinus.*

2. MATERIALS AND METHODS

Experimental insect: The insect (both larvae and adults) were collected from a neighbouring warehouse of the Food Corporation of India. They were specifically collected from damp, almost dark sites from the underneath of the polythene sheets of the wheat stacks. In the laboratory, they were maintained in dark containers at 28 +/- 2^{o} C and 70 – 75% RH. on wheat. To check fungal infection, 2% Nepazin was mixed with the wheat. Temperature and humidity were routinely examined by thermometer and hygrometer. By routine examination from the lower stratum of the culture pots pupae were separated and hatched adults were kept in separate pots to assess the normal developmental profiles. Their incubation period is 4 - 7 days and it takes 40 – 50 days to complete the life cycle. After mating, a female beetle can lay over 2,000 eggs (with an average 200 - 400). Females produce eggs for most of their life. Adults can live up to 3-12 months;

Preparation of phytoproducts:

Preparation of neem extracts: Leaves, fruits, and bark were collected from neem trees (*Azadirachta indica*) and were used to prepare water extract of neem. Respective parts of neem were weighed and emulsified in 1000 ml of distilled water with a blender and the suspension was sieved through a muslin cloth. A powder was prepared by drying respective parts at 35–40°C in an oven and these were grinded to form powder.

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Preparation of sesame oil extracts: Sesame (*Sesamum indicum* L.) is an oleaginous seed of the family Pedaliaceae. Sesame seeds are allowed to be grinded in a traditional Ghani and the extracted oil was refined to be used for treatment.

Preparation of eucalyptus oil: Fresh eucalyptus leaves were taken in a pot and were allowed to boil to extract oil which were collected and used to treat the insect.

Preparation of mustard oil: Mustard seeds **(***Brassica compestris*) were allowed to be grinded in a traditional Ghani and the extracted oil was refined to be used for treatment.

All the developmental stages were divided into control and four treated groups and their morphological and durational changes were recorded separately. Mortalities were recorded at every 24 hours until all larvae or pupae had either died or developed to the next stage.

3. RESULTS

Alphitobius is a holometabolous insect showing adult, eggs, larvae and pupae in its life cycle. Characteristic features of these stages are as follows:

Adults: Lesser mealworm adults are broadly-oval, moderately convex, black or brownish-black and usually shiny in appearance. Colour can be variable depending on age or 'strain' of lesser mealworm. Length is approximately 5.8 to 6.3 mm. Antennae are densely clothed with short yellowish hairs, with the terminal segment lighter in colour. The head is deeply emarginate in front, has a distinct clypeal groove, and the surface is coarsely punctured. Eyes are also emarginated.

The prothorax is twice as broad as long, slightly narrowed from base to apex, with sides feebly curved and narrowly margined. There is a shallow impression on each side of the pronotum in the middle near the base. The pronotal disk is finely and sparsely punctured, with punctures much coarser laterally.

The elytra have moderately impressed striae with finely punctured, feebly convex intervals. Elytral punctures are sparse and nearly as large as those of the striae. The ventral surface of the lesser mealworm is dark reddish-brown, with the prosternal process horizontal between coxae and having a prominent apex.

Eggs: Eggs are about 1.5 mm in length, creamy white to tan coloured and slender with slightly rounded ends.

Larvae: Larvae hatch in 6-7 days and complete development to the adult stage in 40 to 100 days, depending on temperature and food quality. Several studies have been conducted on *A. diaperinus* development on varied diets, at varying ambient temperatures and microhabitats, and among other poultry house inhabiting species. Larvae consume spilled feed, manure, and in some cases dead or sick birds and cracked eggs.

Larvae have a segmented body with three pairs of legs and tapering abdominal tip posteriorly. They are approximately 7 - 11 mm in length at last instar. Freshly-emerged larvae are a milky colour but gradually a shade of brown colour are observed; before moulting they again assume dark colour. There are 6 larval instars. The duration of each instar is 3, 5, 5, 6, 8 and 12 days respectively.

Pupae: Pupae are approximately 6 to 8 mm in length, creamy white to tan coloured, with legs tucked alongside the body. Pupal period is 6 days. At the end of pupal period, the whole body appears brownish and newly hatched pharate adults appear brown that gradually turns into black.

Declining survival rate: Following the rule of natural selection gradually the number of surviving individuals in successive instars decline by 34%. The size and weight however gradually increase in successive instars; it shows 200 times increase in size and substantial increase in weight (Table 1).

Effects of different phytoproducts: Treatment with different phytoproducts depicted substantial decline in survival rate showing 44%, 44%, 44% and 42% decline in number in respect of treatment with nimbecidine, eucalyptus oil, sesame oil and mustard oil. Increased duration of respective instars actually starts from third instars and duration increase is maximum following nimbecidine treatment and minimum in mustard oil treatment. Declining size of respective instars in different treatments appeared significant from 4th instar onwards (Table 2).

Effect of different doses of Nimbecidine against different developmental stages: 50 gm semi crushed wheat grains were treated with 0.5ml, 0.4 ml, 0.2 ml, 0.1 ml of nimbecidine and 20 adults were subjected to toxic effects of

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nimbecidine for several weeks to report the detrimental effects of this phytoproduct on the post embryonic developmental stages of this beetle (Table – 3, 4).

4. DISCUSSION

In spite of the tremendous advent of the chemical insecticides, due to their hazardous side effects, different phytoproducts have long been treated as alternative harmless insecticides that has minimum side effects and from the view point of imparting resistance to insects and toxic residual effects in grains and for their biodegradable property, use of phytoproducts has been gaining importance day by day. The insecticidal activity of different essential oils and plant extracts against different stored-product pests has been evaluated [34], [35], [36], [37]. [38]. In spite of the wide-spread recognition that many phytoproducts possess insecticidal properties, only a handful of phytopesticides are directly in use because the commercialization of new botanical insecticides can be hindered by a number of issues [39]. These products presently constitute 1% of the world insecticide market [40]. Essential oils from different plant species possess ovicidal, larvicidal, and repellent properties against various insects and are regarded as environmentally compatible pesticides [41].

Neem (*Azadirachta indica*) has long treated as the miracle plant for its spectacular role in traditional medicine for the cure of multiple diseases including cancer for centuries [42], [43], [44]. Apart from its medicinal roles, it has long been used in controlling the infestation of stored grains. Different vegetative parts of neem, like flower [45], wood chipping [46], neem oil from seed and kernel [47] etc. have been tested against different insects for their control.

Nimbecidine is a totally natural neem-oil based product with *Azadirachtin* as the most effective insect growth regulator molecule in addition to Meliantriol, Salanin, Nimbin. It acts as an insect repellant, antifeedant, growth regulator and mating disruptor and has been proved to be really effective against a wide range of pests [48], [49], [07], [50], [51], [52], [53]. It also corroborates the findings of Jilani *et al* (1988) {54] showing significant reduction in the number of progeny emergence, delayed development, failure of pupation and abnormal pupae and adults of *T. castaneum* treated with Calneem oil.

Data of present investigation showed a dose dependent effect (Table -1) which might not due to the *Azadirachtin* content since other active ingredients of nimbecidine may be responsible for such dose-dependent effect [48]. Present data showing reduced progeny production in *A. diaperinus* corroborates the observation of Lale and Mustapha (2000) [55] where they reported significant reduction in oviposition and adult emergence in *Callosobruchus maculatus* treated with neem seed oil. Mortality of *A. diaperinus* in nimbecidine treated wheat corroborates the findings of Dunkel *et al* (1990) [56] who noticed significant mortality of *S. oryzae* in neem-based insecticide Margosan-O treated wheat.

As the pupae are non feeding stage, their mortality could not be explained by any of the known modes of action of neem i.e. antifeedant effects and growth regulator effects. Treating stored grain with nimbecidine can disrupt insect feeding by making the treated materials unattractive or unpalatable and as a consequence, insect growth, survival and reproduction are adversely affected [57]. Azadirachtin, inhibits the release of prothoracicotropic hormones and allatotropins, thereby affecting metamorphosis in insects [58].

Neem has been treated as an anti-feedant, an oviposition deterrent (anti-egg laying), a growth inhibitor and a chemosterilizer. [59]. Azadirachtin, a tetranortriterpenoid compound, closely mimics the hormonal action of Edison, which is necessary for reproduction in insects. Substantial decline in the level of ovarian protein corroborates this data. Disruption of feeding process may be attributed to the decline in the concentration total body protein in other organs.

Since essential oils of different plants are a complex mixture of components (unlike chemical pesticides which are based on a single product), they work together within a plant and it is unlikely that pests will become resistant to them. Since eucalyptus oil is particularly strong when in vapourized form, they could also be used commercially as a fumigant (gaseous pesticide) for stored products and impregnated into packaging to prevent insect infestation. The study explains that eucalyptus essential oil could have a large role in the control of pests and provide an alternative to chemical pesticides.

The vapour of mustard oil can penetrate the grain and kill, the internal larvae and the pupa of *S. zeamais* within 24 h, like other grain fumigants such as phosphine and methyl bromide. Of all the developmental stages, pupae proved to be most resistant to the fumes of oil because they have the least respiratory rate [60] because the developmental stage of low respiration rate absorbs less quantity of the fumigant in a given time period. In this study also, the bioactive compound of mustard oil seem to be penetrated the larvae or pupa through spiracles.

During the pupal period oxygen consumption initially declines and then rises again, due to changes in the enzymes systems regulating energy release. It is well accepted that the developmental phases of insects with higher metabolic rate are more susceptible to gas exchange incited by an insecticide resulting in the higher energy expense for homeostasis maintenance. (Emekci *et al.*, (2002) [61]. reported that in cockroaches (*Blattella germanica)* CO_2 production rate increased when exposed to sub lethal concentrations of AITC (the major component of the mustard oil), which led to conclude that AITC might also act on the energy metabolism and may be acting as inhibitor of oxidative phosphorylation, and thus interrupting ATP formation. All the phytoproducts used in this investigation act as anti-feedant, oviposition deterrent (anti-egg laying), a growth inhibitor and chemosterilizer and these phytoproducts indirectly act as phytoinsecticide.

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REFERENCES

[1] Wolpert, V. 1967. Needless losses. *Far Eastern Econ. Rev*. 55.

[2] Chaube, M.K. 2008. Fumigant toxicity of essential oils from some common spices against pulse beetle, Callosobruchus chinensis (Coleoptera: Bruchidae). *J. Oleo. Sci*.57:171–179.

[3] Isman, M.B. 2006. Botanical insecticides, deterrents and repellents in modern agriculture and an increasingly regulated world. *Annu. Rev Entomol* 51:45–66.

[4] Wang, C.F. Yang, K, Zhang, H.M. Cao, J. Fang, R. Liu, Z.L., Du, S.S. Wang, Y.Y. Deng, Z.W, and Zhou, L. 2011. Components and insecticidal activity against the maize weevils of Zanthoxylum schinifolium fruits and leaves. *Molecules;* 16(4):3077-88.

[5] Cosimi, S., Rossi, E., Cioni, P.L., Canale, A . 2009. Bioactivity and qualitative analysis of some essential oils from Mediterranean plants against stored-product pests: Evaluation of repellency against *Sitophilus zeamais* Motschulsky,*Cryptolestes ferrugineus* (Stephens) and *Tenebrio molitor* (L.). *J. Stored Prod. Res.* 45: 125–132

[6] Nesci, A., Barra, P., Etcheverry, M. 2011. Insecticidal activity of synthetic antioxidants, natural phytochemicals, and essential oils against an Aspergillus section Flavi vector (*Oryzaephilus surinamensis* L.) in microcosm. *J. Pest Sci.* 84: 107–115.

[7] Obeng-Ofori, D. 2007. The use of botanicals by resource poor farmers in Africa and Asia for the protection of stored agricultural products, *Stewart Postharvest Rev* 3:6.

[8] [8] Schmutterer, H. 2002.The Neem Tree (*Azadirachta indica* A. Juss.) and Other Meliaceous Plants: Sources of Unique Natural Products for Integrated Pest Management, Medicine, Industry and Other Purposes. *Neem Foundation, Mumbai, India*, 893 pp.

[9] Tanzubil, P. B., and McCaffery, A. R. 1990. Effects of azadirachtin and aqueous neem seed extracts on survival, growth and development of the African armyworm, *Spodoptera exempta*. *Crop Prot*. 9, 383–386.

[10] Singh, R.P. 1993. Neem for the management of stored grain insects in developing countries. *World Neem Conf.* Bangalore, India, 1993. *Souvenir*, pp. 69.

[11] Kareem, A., Saxena, R.C. Boncodin, M.E.M. Krishnasamy, V. and Seshu. D.V. 1989. Neem as seed treatment for rice before sowing: effects on two homopterous insects and seedling vigor. *J. Econ. Entomol*. 82: 1219-1223.

[12] Batish, D.R., Singh, H.P, Kohli, R.K. 2008. Eucalyptus essential oil as a natural pesticide. *Forest Ecology and Management.* 256: 2166-2174.

[13] Alzogaray, R.A., Lucia, A., Zerba, E.N., Masuh, H.M. 2011. Insecticidal activity of essential oils from eleven Eucalyptus spp. and two hybrids: lethal and sublethal effects of their major components on Blattella germanica. *J. Econ. Entomol.*;104(2):595-600.

[14] Mossi, A.J., Astolfi, V., Kubiak, G., Lerin, L., Zanella, C., Toniazzo, G., Oliveira, D., Treichel, H., Devilla, I.A., Cansian, R., Restello, R. 2011. Insecticidal and repellency activity of essential oil of Eucalyptus sp. against Sitophilus zeamais Motschulsky (Coleoptera, Curculionidae). J. Sci. Food Agric. ;91(2):273-277.

[15] Sugumar, S., Clarke, S.K., Nirmala, M.J., Tyagi, B.K., Mukherjee, A., Chandrasekaran, N. 2014. Nanoemulsion of eucalyptus oil and its larvicidal activity against Culex quinquefasciatus. *Bull. Entomol Res.*;104(3):393 - 402.

[16] Mesbah, H.A,, Mourad, A.K., Rokaia, A.Z. 2006. Efficacy of some plant oils alone and/or combined with different insecticides on the cotton leaf-worm Spodoptera littoralis (Boisd.) (Lepidoptera: Noctuidae) in Egypt. *Commun. Agric. Appl. Biol. Sci.*;71(2 Pt B):305-28.

[17] Regmi, H., Kafle, L., Gc, Y.D., Shih, C.J. 2012. Efficacy of natural products against Callosobruchus chinensis (Coleoptera: Bruchidae) in Nepal. *J. Econ. Entomol*.;105(3):1095-9.

[18] Roy, G.C. Chakraborty,K. Nandy,P and Moitra M.N. 2014. Pros and Cons of Curcumin as Bioactive Phyto-compound for Effective Management of Insect Pests *American Scientific Research Journal for Engineering, Technology, and Sciences (ASRJETS):7(1): 31-43*

[19] Paes , J.L.; Faroni, L.R.D.'A.; Dhingra,O. D.; Cecon , P.R.; Silva,T.A.2012. Insecticidal fumigant action of mustard essential oil against *Sitophilus zeamais* in maize grains. Crop Protection, 34:56-58.

[20] Park, I.K., Choi, K.S., Kim, D.H., Choi, I.H., Kim, L.S., Bak, W.C., Choi, J.W., Shin, S.C. 2006. Fumigant activity of plant essential oils and components from horseradish (Armoracia rusticana), anise (Pimpinella anisum) and garlic (Allium sativum) oils against Lycoriella ingenua (Diptera: Sciaridae). Pest Manag Sci. 62(8):723-8.

[21] Park, I.K., and Shin, S.C. 2005. Fumigant activity of plant essential oils and components from garlic (Allium sativum) and clove bud (Eugenia caryophyllata) oils against the Japanese termite (Reticulitermes speratus Kolbe). *J. Agric. Food Chem.* ;53(11):4388-92.

[22] Rattanachaikunsopon, P.and Phumkhachorn, P. 2009. Antimicrobial activity of elephant garlic oil against Vibrio cholerae in vitro and in a food model. Biosci. Biotechnol Biochem.; 73(7): 1623-7.

[23] Yang, F.L., Li, X.G., Zhu, F., Lei, C.L. 2009. Structural characterization of nanoparticles loaded with garlic essential oil and their insecticidal activity against Tribolium castaneum (Herbst) (Coleoptera: Tenebrionidae). J Agric. Food Chem. ;57(21):10156-62.

[24] Cloyd, R.A. and Chiasson, H. 2007. Activity of an essential oil derived from Chenopodium ambrosioides on greenhouse insect pests. J. Econ. Entomol. ;100(2):459-66.

[25] Dinev, I. 2013. The darkling beetle (Alphitobius diaperinus) - A health hazard for broiler chicken production. *Trakia J. Sci.* 11(1), 1-4.

[26] Kozaki, T., *et al.* 2008. Comparison of two acetylcholinesterase gene cDNAs of the lesser meal worm, Alphitobius diaperinus, in insecticide susceptible and resistant strains. *Archives of Insect Biochem. and Physiol.* 67(3), 130-38].

[27] Arunraj, C., *et al*. 2013. Lesser mealworm, Alphitobius diaperinus (Panzer, 1797) (Coleoptera: Tenebrionidae) menace in poultry farms in south India. *J. Biopesticides* 6(1), 84-86

[28] Watson, D.W.,*et al*. 2000. Limited transmission of turkey coronavirus in young turkeys by adult Alphitobius diaperinus (Coleoptera: Tenebrionidae). *Journal of Medical Entomology*37(3): 480-83.

[29] Steelman, C. D. 2008. Comparative susceptibility of adult and larval lesser mealworms, Alphitobius diaperinus (Panzer) (Coleoptera: Tenebrionidae), collected from broiler houses in Arkansas to selected insecticides. *J. Agri. Urban Entomol.* 25(2), 111-25.

[30] Geden, C.J. and Steinkraus, D. C. 2003. Evaluation of three formulations of *Beauveria bassiana* for control of lesser mealworm and hide beetle in Georgia poultry houses. *J. Econ. Entomol.* 96: 1602-1607.

[31] Hinton, J.L. and Moon, R.D. 2003. Arthropod populations in high-rise, caged layer houses after three manure cleanout treatments. *J. Econ. Entomol.* 96: 1352-1361.

[32] Salin, C., Vernon, P., Vannier, G. 2003. Cold resistance in the lesser mealworm Alphitobius diaperinus (Panzer) (Coleoptera: Tenebrionidae). Cryo. Letters.;24(2):111-8.

[33] Watson, D.W., Denning, S.S., Zurek, L., Stringham, S.M., Elliott, J. 2003. Effects of lime hydrate on the growth and development of darkling beetle, *Alphitobius diaperinus*. *Int. J. Poultry Sci.* 2: 91-96.

[34] Tunc, I., Berger, B.M., Erler, F., Dagli, F. 2000. Ovicidal activity of essential oils from five plants against two stored-products insects. *J. Stored Prod. Res.* 36*:* 161–161

[35] Kim, S.I., Park, C., Ohh, M.H., Cho, H.C, Ahn, Y.J. 2003. Contact and fumigant activities of aromatic plants extracts and essential oils against *Lasioderma serricorne* (Coleoptera: Anobiidae). *J. Stored Prod. Res.* 39*:*11–11.

[36] Lee, S.E., Peterson, C.J., Coats, J.R. 2003. Fumigation toxicity of monoterpenoids to several stored-product insects .*J. Stored Prod. Res.* 39*:* 77–77.

[37] [37] Cetin, H. and Yanikoglu, A. 2006. A study of the larvicidal activity of *Origanum* (Labiatae) species from Southwest Turkey. *J. Vector Ecol.* 31*:* 118–118.

[38] Ayvaz, A., Karaborklu, S., Sagdic, O. 2009. Fumigant Toxicity of Five Essential Oils Against the Eggs of *Ephestia kuehniella* Zeller and *Plodia interpunctella* (Hübner) (Lepidoptera: Pyralidae). *Asian J. Chem.* 21*:*596–596.

[39] Isman, M.B. 1997. Neem and other botanical insecticides commercialization. *Phytoparasiti-Ca*. 25*:* 339–339.

[40] Rozman, V., Kalinovic, I., Korunic, Z. 2007. Toxicity of naturally occurring compounds of Lamiaceae and Lauraceae to three stored-product insects. *J. Stored Prod. Res.* 43*:*349–355.

[41] Isman, M.B . 2000. Plant essential oils for pest and disease management. *Crop Protection.* 19*:* 603–603.

[42] Gunadharini, D.N., Elumalai, P., Arunkumar, R., Senthilkumar, K., Arunakaran, J. 2011. Induction of apop- tosis and inhibition of PI3K/Akt pathway in PC-3 and LNCaP prostate cancer cells by ethanolic neem leaf extract. *J Ethnopharmacol*.;134(3):644-50.

[43] Paul, R., Prasad, M, Sah, N.K. 2011. Anticancer biology of Azadirachta indica L (neem): a mini review. *Cancer Biol Ther.*;12(6):467-76.

[44] Hao, F., Kumar, S., Yadav, N. and Chandra, D. 2014. Neem components as potential agents for cancer prevention and treatment. *Biochim. Biophys. Acta.*;1846(1):247-57.

[45] Siddiqui, B.S., Ali, S.T., Rajput, M.T., Gulzar, T., Rasheed, M., Mehmood, R. 2009. GC-based analysis of insecticidal constituents of the flowers of Azadirachta indica A. Juss. *Nat Prod Res.*;23(3):271-83.

[46] Howard, A.F., Adongo, E.A., Hassanali, A., Omlin, F.X., Wanjoya, A., Zhou, G. Vulule, J. 2009. Laboratory evaluation of the aqueous extract of Azadirachta indica (neem) wood chippings on Anopheles gambiense. (Diptera: Culicidae) mosquitoes. *J. Med. Entomol.;* 46 (1):107-14.

[47] Adnan SM, Uddin MM, Alam. MJ., et al (2014). Management of mango hopper, Idioscopus clypealis, using chemical insecticides and Neem oil. *The Scientific World Journal* Volume 2014: Article ID 709614,

[48] Schmutterer, H. 1990. Properties and potentials of natural pesticides from neem tree. *Annu. Rev. Entomol* .35:271–298.

[49] Isman, M.B. 2006. Botanical insecticides, deterrents and repellents in modern agriculture and an increasingly regulated world. *Annu. Rev Entomol* 51:45–66.

[50] Anjali, C.H., Sharma, Y., Mukherjee, A., Chandrasekaran, N. 2012, Neem oil (Azadirachta indica) nanoemulsion--a potent larvicidal agent against Culex quinquefasciatus. *Pest Manage Sci.*;68(2):158-63

[51] Scudeler, E.L and dos Santos, D.C. 2013. Effects of neem oil (Azadirachta indica A. Juss) on midgut cells of predatory larvae Ceraeochrysa claveri (Navás, 1911) (Neuroptera: Chrysopidae). *Micron*.;44:125-32

[52] Benelli, G., Murugan, K., Panneerselvam, C., Madhiyazhagan, P., Conti, B., Nicoletti, M. 2015. Old ingredients for a new recipe? Neem cake, a low-cost botanical by-product in the fight against mosquito-borne diseases. *Parasitol. Res*. 114(2):391-7.

[53] Martínez ,L.C., Plata-Rueda, A., Zanuncio, J.C., Serrão, J.E. 2015. Bioactivity of six plant extracts on adults of Demotispa neivai (Coleoptera: Chrysomelidae). *J. Insect Sci.;*15.pii:34.

[54] Jilani, G., Saxena, R.C., Rueda, B.P. 1988. Repellent and growth inhibiting effects of turmeric oil, sweet flag oil, neem oil and Margosan-O on red flour beetle, Tribolium castaneum (Tenebrionidae: Coleoptera). *J .Econ .Entomol* 81:1226–1230.

[55] Lale, N.E.S. and Mustapha, A. 2000. Potential of combining neem (Azadirachta indica A.juss) seed oil with varietal resistance for the management of the cowpea bruchid, Callosobruchus maculatus (F.). *J. Stored Prod. Res* 36:215–222

[56] Dunkel, F.V., Sriharan, S., Niziyimana, E., Serugendo, A 1990. Evaluation of neem kernel extract (Margosan-O) against major stored insect pests of beans and sorghum in Rwanda. In: Fleurat-Lessard F, Ducom P (eds) Proceedings, 5th international working conference of stored-product protection, Bordeaux, France. Escuela Agricola Panamericana, Tegucigalpa, Honduras, pp 527–535.

[57] Saxena, R.C., Milani, G., Abdul-Kareetn, A. 1988. Effects of neem on stored grain insects. In: Jacobson M (ed) Focus on phytochemical pesticides, vol. I. The neem tree. CRC, Boca Raton, pp 97–111

[58] Banken, J. A.O. and Stark, J. 1997. Stage and age influence on the susceptibility of Coccinella septempunctata after direct exposure to Neemex, a neem insecticide. *J. Econ. Entomol*. 90:102–1105

[59] Khater, H. F. and Khater, D.F. 2009. The insecticidal activity of four medicinal plants against the blowfly Lucilia sericata (Diptera: Calliphoridae). Int. J. Dermatol. 48(5):492-7

[60] Mahroof R. Subramanyam B. Throne JE. Menon A. (2003). Time-mortality relationships for Tribolium castaneum (Herbst) life stages exposed to elevated temperatures. J. Econ. Entomol. 96: 1345-1351.

[61] Emekci M. Navarro S. Donahaye EJ. Rinder M. Azrieli, A. (2002). Respiration of Tribolium castaneum (Herbst) at reduced oxygen concentrations. J. Stored Prod. Res. 38: 413-425.

APPENDIX - A

Table – 1. Gradual decline in the survival rate (number), duration of different post embryonic stages (days), size (mm), colour and weight (mg) of different stages:

Parameters	**Egg**	**1st Instar**	**2nd Instar**	**3rd Instar**	**4th Instar**	**5th Instar**	**6th Instar**	**Pupa**	**Adult**
Survival rate	50+/- 9	48+/- 8	47+/- 9	46+/- 7	44+/- 9	42+/-8	40+/-8	35 +/-10	33+/-12
Duration	6-7	3-4	5-6	5-6	6-7	8-9	12-14	6-7	
Size (mm)	1.5	2.2+/-0.1	3.2+/-0.2	4.3+/-0.4	5.4+/-0.5	7.7+/-0.2	8.8 +/-0.3	5.8+/-0.6	6.1+/-0.5
Body Color	Milky white	Creamy white	Light brown	Brown	Deep brown	Dark Brown	Dark Brown	Off white	Black
Weight (mg)	0.0004	0.0005	0.0010	0.0018	0.0036	0.007	0.009	0.010	0.015

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Table – 2. Gradual decline in the survival rate (number), duration (days), size (mm) and weight (mg) of different post embryonic stages in response to nimbecidine, eucalyptus oil, sesame oil and mustard oil treatment against untreated insects:

Parameters	Egg	1st Instar	2nd Instar	3rd Instar	4th Instar	5th Instar	6th Instar	Pupa	Adult
Survival rate	50+/- 9	48+/- 8	47+/- 9	46+/- 7	44+/- 9	42+/- 8	40+/- 8	35 +/-10	33+/-12
Nimbecidine	50 +/- 6	42+/- 5	45+/- 4	41+/- 6	40+/- 5	36+/- 4	35+/- 4	29+/- 4	28+/- 4
Eucalyptus oil	50 +/- 4	45+/- 6	44+/- 8	44+/- 6	43+/- 6	35+/- 3	35+/- 3	31+/- 3	28+/- 3
Sesame oil	50 +/- 6	44+/- 4	44+/- 6	45+/- 3	42+/- 5	35+/- 4	35+/- 4	30+/- 5	28+/- 4
Mustard oil	50 +/- 4	45+/- 4	43+/- 4	44+/- 4	43+/- 4	38+/- 4	36+/- 2	31+/- 4	29+/- 3
Duration	6-7	3-4	5-6	5-06	6-07	8-09	12-14	6-07	
Nimbecidine	6-7	3-4	5-6	5-11	6-13	8-15	12-19	6-11	
Eucalyptus oil	6-7	3-4	5-6	5-09	6-11	8-14	12-18	6-10	
Sesame oil	6-7	3-4	5-6	5-10	6-12	8-13	12-17	6-10	
Mustard oil	6-7	3-4	5-6	5-08	6-11	8-12	12-17	6-09	
Size (mm)	1.5	2.2+/-0.1	3.2+/-0.2	4.3+/-0.4	5.4+/-0.5	7.7+/-0.2	8.4+/-0.3	5.8+/-0.6	6.1+/-0.5
Nimbecidine	1.5	2.2+/-0.2	3.2+/-0.2	4.0+/-0.2	4.8+/-0.2	6,9+/-0.3	7.6+/-0.1	5.1+/-0.3	5.8+/-0.2
Eucalyptus oil	1.5	2.2+/-0.1	3.2+/-0.2	4.1+/-0.2	4,9+/-0.1	7.1+/-0.1	7.9+/-0.3	5.2+/-0.4	6.0+/-0.1
Sesame oil	1.5	2.2+/-0.1	3.2+/-0.2	4.1+/-0.3	5.1+/-0.2	7.0+/-0.2	7.8+/-0.2	5.3+/-0.4	5.9+/-0.3
Mustard oil	1.5	2.2+/-0.2	3.2+/-0.2	4.2+/-0.4	5.0+/-0.3	7.2+/-0.3	8.1+/-0.3	5.4+/-0.2	6.0+/-0.2
Body Color	Milky white	Creamy white	Light brown	Brown	Deep brown	Dark Brown	Dark Brown	Off white	Black
Weight (mg)	0.0004	0.0005	0.0010	0.0018	0.0036	0.0071	0.0092	0.014	0.015
Nimbecidine	0.0004	0.0005	0.0010	0.0015	0.0032	0.0066	0.0081	0.012	0.011
Eucalyptus oil	0.0004	0.0005	0.0010	0.0016	0.0035	0.0068	0.0086	0.013	0.013
Sesame oil	0.0004	0.0005	0.0010	0.0018	0.0033	0.0069	0.0086	0.014	0.014
Mustard oil	0.0004	0.0005	0.0010	0.0017	0.0033	0.0070	0.0088	0.013	0.012

Table 3. Mean number of *Alphitobius diaperinus* larvae, pupae and adults that emerged from wheat treated with different dosages of Nimbecidine at different times after oviposition period. Data are mean ± SE (n=5 taking 20 insects).

Doses % (v/v)	1st instar larva	2nd instar larva	3rd instar larva	4th instar larva	5th instar larva	6th instar larva	Pupa	Adult
Control	53±5.2	48.6±5.81	45±4.04	44±3.51	42.6±3.8	39±4.72	23±2.64	32±4.16
0.2ml	31±3.2	26±3.05	24.6±0.3	23±0.57	22±1.15	20±1.15	15±2.88	11±2.08
0.1ml	34.6±2.4	31±2.08	30.3±1.8	27.6±1.2	24.3±2.3	23±2.64	17.6±1.4	16±1.15
0.05ml	38.3±1.2	37±1.9	34.3±1.4	33±1.52	29±2.08	26±3.05	18.6±1.8	16.3±2.1
0.025ml	39±0.57	36.6±0.88	35.3±0.3	32.3±1.2	30±.6.7	28.3±2.2	26.3±2.0	24.6±2.6

Number of progeny emergence (mean ± S.E.). Means in the same column followed by the same lower case letter do not differ significantly, means in the same rows followed by the same uppercase letter do not differ significantly at $P < 0.05$ (Holm–Sidak test).

Table 4. Toxicity of different dosages of Nimbecidine in wheat grains to *Alphitobius diaperinus* adults at a temperature of 25°C and 65–70% RH. Data are means ± SE (n=5 of 20 insects each.Exposure period (d)% mortality (mean ± SE)

Dosage % (v/v) ml	2 days	6 days	10 days
Control	0±0.00	0±0.00	0±0.00
0.5	6.66±3.52	11.66±0.88	18±1.154
0.4	6±1.15	8.66±0.66	12±1.15
0.2	3.33±0.88	6±0.57	9±0.57
0.1	1±0.57	1.66±0.88	3.33±0.88

* Means in the same column followed by the same lowercase letter do not differ significantly, means in the same rows followed by the same uppercase letter do not differ significantly at $P < 0.05$ (Holm–Sidak test).