CONCLUSION

*Dysdercus koenigii* (Fabr.) (Heteroptera: Pyrrhocoridae) is widely distributed in India and is a minor/sporadic pest of cotton crops in Punjab and Uttar Pradesh. It also feeds on okra, maize, and pearl millet, etc. The insects were maintained in a B.O.D. chamber at a temperature of 28±2°C and relative humidity of 70-80 percent and under long day (14L:10D) photoperiod. A maximum of 35-40 adults were kept in a jar with sterilized sand on its bottom and provided water soaked cottonseeds as food. The eggs were laid by the females on the cottonseed or on moist sand and were collected in a petriplate. The food was changed daily while the jars were changed twice a week in order to maintain hygienic conditions.

The present study is an attempt to understand the impact of synthetic insecticides and neem formulations on the biology of *D. koenigii*. Therefore, desired concentrations i.e. 0.01, 0.02 and 0.04 percent of multineem (8 EC) and neemjeevan (0.3 EC) and 0.001, 0.002 and 0.004 percent of imadacloprid (Confidor, 200 SL), monocrotophos (Hilcron, 36 SL) quinolphos (Byrusil, 25 EC), and oxydemeton-o-methyl (Metasystox, 25 EC) were applied topically @ 1μl/IV instar on the thoracic terga by means of a microapplicator.

Multineem caused mortality in IV instar and delayed mortality in V instars also. 0.04 percent multineem was more effective than other concentrations tested. The longevity of treated IV instar is prolonged as well as increase in moulting V instar. Surviving females derived from treated IV instar laid 61.66, 51.55 and 37.66 eggs at 0.01, 0.02 and 0.04 percent concentrations respectively. Egg hatching was significantly less than control. Emerged adults derived from treated IV instar were significantly malformed except those treated with lower concentration. Multineem prolonged premating, pre-oviposition and oviposition periods and post oviposition period was significantly reduced. The longevity of surviving mated and unmated females was reduced and survival of males was also affected.

The highest mortality of IV instar was at 0.04 percent of neemjeevan. The duration of IV and V instar was insignificantly enhanced. Adult emergence was not significantly decreased. The fecundity of surviving
individuals derived from treated IV instar was greatly affected. Fertility was significantly reduced at 0.04 and 0.01 percent concentrations. The highest number of deformed individuals was produced at 0.04 percent concentration. Premating period was insignificantly increased while preoviposition period was significantly reduced at 0.004 percent. Oviposition period was not affected significantly. The post-oviposition period was considerably reduced. The longevity of mated and unmated males and females reduced.

0.004 percent of imidacloprid gave highest mortality of IV and V instars. The longevity of surviving individuals of IV and V instar was prolonged. Emergence of adults was found to be insignificant. Fecundity was considerably affected and the adult females laid significantly less number of eggs. Fertility was also significantly inhibited at higher concentrations than at lower. Malformations were high at 0.004 percent of imidacloprid. The premating, pre-oviposition and oviposition period is enhanced in individuals obtained from treated IV instar than the untreated. The post-oviposition period was significantly decreased. The longevity of mated and unmated females and males was considerably decreased.

Total mortality of IV and V instars caused by 0.004 percent of monocrotophos was higher than 0.001 and 0.002 percent concentrations. The longevity of surviving IV and V instars was prolonged. Emerged adults were less in number as compared to untreated ones. The fecundity was statistically analyzed and found to be significant. Hatching was markedly reduced while malformed individuals were more in number at 0.004 percent concentration than other treatments. Premating and preoviposition period was reduced and oviposition period was prolonged, while, postoviposition period decreased. Longevity of mated males and females was significantly reduced.

0.001, 0.002 and 0.004 percent concentrations of quinolophos gave mortality of IV and delayed mortality in V instars. Longevity of the survivors of IV and V instars was slightly increased. Adult emergence was substantially reduced. The fecundity and fertility was also significantly reduced in females. Malformed individuals were also produced at different concentrations but 0.004 percent produced higher number. Premating, preoviposition and
oviposition periods were substantially increased. While postoviposition period was significantly shortened. The longevity of mated and unmated females and males derived from treated IV instar was significantly decreased.

Oxydemeton-o-methyl caused mortality in IV instar and delayed mortality obtained in V instar. The longevity of surviving nymph significantly increased in comparison to untreated control. The eggs per female was significantly decreased and hatching also. Highest malformed individuals were produced at 0.004 percent and the lowest at 0.001 percent concentration. The pre-mating, preoviposition and oviposition periods were delayed and post-oviposition period was significantly decreased. Longevity of mated males was significantly decreased whereas insignificant in unmated males. Longevity of mated and unmated females was significantly decreased.

Efforts have also been made to study the quantitative estimation of protein of gonads obtained from treated IV instar to ascertain the effect of these insecticides at different age interval i.e 1-day, 4-day and 7-day old. The quantity of protein in multineem treatment was decreased in 1-day old female but insignificant. In 4-day old untreated female, total ovarian protein was increased to 42.855mg/100mg, while the inhibition of protein in treated one was insignificant. In 7-day old females the inhibition was also insignificant as compared to control. Almost same trend of inhibition was estimated in neemjeevan treatment.

Ovary protein of survivors of 1-day old treated imidacloprid was partially affected. Almost insignificant inhibition was observed in 4-day old female. In 7-day old female 0.001 percent of insecticide caused significant inhibition of ovary protein but 0.002 and 0.004 percent did not. Ovary protein of 1, 4 and 7-day old females derived from IV instar treated with 0.001, 0.002 and 0.004 percent concentrations of monocrotophos was significantly and insignificantly reduced. Two fold increase in the amount of ovary protein in 7-day old untreated females was observed.

0.001 and 0.002 percent of quinolphos did not cause significant inhibition of ovary protein in 1- and 4-day old females. In 7-day old female, the amount was significantly decreased at all concentrations. 0.004 percent of
oxydemeton-o-methyl was proved to inhibit the protein of ovary more significantly than other concentrations tested. Inhibition of ovary protein was significant in 4-day and 7-day old females.

Testes protein of males derived from IV instars was more inhibited by synthetic insecticides than neem products at different age intervals. Multineem did not significantly inhibit the testis protein. The amount of testis protein decreased from 21.940 mg/100mg in 1-day old untreated control to 14.443, 13.806 and 14.061 mg/100mg at 0.01, 0.02 and 0.04 percent concentrations respectively of neemjeevan. While 40.235 mg/100mg in 4-day untreated males and attained a peak i.e 46.305 mg/100mg in 7-day old males. 0.004 percent of monocrotophos caused a significant inhibition of total protein in 1-day old males. Whereas, 4 and 7-day old males derived from IV instar treated with 0.001, 0.002 and 0.004 percent concentrations affected the level of protein adversely. Quinolphos seems to be most promising which significantly inhibited the amount of testis protein. In 1-day old females derived from quinolphos treated IV instar caused a significant reduction of testis protein. In 4-day old male derived from treated IV instar the testis protein was significantly reduced while it did not offer in 7-day old males. Males derived from oxydemeton-o-methyl treated with IV instar nymphs showed significant decrease in testis protein in comparison to untreated control.

Efforts have been made in the present study to quantify the cholesterol at different age intervals in the ovary of surviving adult of *D. koenigii* derived from treated IV instar nymph for 0-day, 1-day, 2-day, 3-day, 4-day, 5-day and 6-day old. In 0-day old female derived from 0.01, 0.02 and 0.04 percent concentration of multineem did not show marked inhibitory effect while, significant inhibition was recorded in the ovary of 1-day old and 2-day female. Whereas cholesterol was insignificantly decreased in 3-day old female. 0.04 percent of multineem caused a marked reduction in the quantity of cholesterol than at 0.01 and 0.02 percent recorded in 4-day old female. In 5-day old females 0.04 percent of multineem gave significantly lesser amount of cholesterol whereas 6-day old female did not. In 0-day old females derived
from neemjeevan treated IV instar the ovarian cholesterol was statistically insignificant. A significant inhibitory effect at different concentrations of neemjeevan, was however obtained in 1-day, 2-day old females whereas in 3-day old inhibition caused by the neemjeevan was insignificant. A significant inhibition was obtained in 4-day old females derived from 0.04 percent concentration. Inhibitory result caused by 0.04 percent concentration on 5-day and 6-day old was also found to be significant.

0-day old females derived from IV instar treated with imidacloprid showed a significant decrease in ovarian cholesterol whereas 0.004 percent gave insignificant result. In 1-day and 2-day old the inhibitory effect caused by aforesaid concentrations are highly significant in relation to untreated control. In 3-day old untreated female the level of cholesterol was greatly increased in comparison to 0-day, 1-day and 2-day old females. While the total effect in cholesterol in 4-day old was statistically insignificant. A significant reduction in ovarian cholesterol was expressed in 5-day and a substantial in 6-day. Almost similar reduction obtained in monocrotophos treatment. Quinolphos was found to be highly effective causing a significant reduction in the level of ovary cholesterol in the female of *D. koenigii* derived from treated IV instar. A significant decrease in the quantity of ovary cholesterol was found in the 0-day, 1-day, 2-day, 4-day old female. Insignificant inhibition was obtained on 3-day and 5-day old females. Inhibitory effect caused by oxydemeton-o-methyl was significant in 0-day, 1-day, 2-day and 5-day old whereas insignificant result was observed in 3-day, 4-day and 6-day old females.

Testes cholesterol of surviving males of different age intervals obtained from insecticide treated IV instar was estimated. Quantity of cholesterol in untreated adult was high at beginning of age and then decreased to 1.808 mg/100mg and again increase to 3.488 mg/100mg in 4-day old male and then falls to 1.435 mg/100mg on 6-day. In 0-day, 1-day, 3-day and 4-day old males, the inhibitory effect caused by different concentration of multineem was not significant in comparison to control and considerably increase in 2-day, 5-day and 6-day old males In 0-day, 1-day, 3-day and 4-day old males, the decrease was more with 0.04 percent concentration than that of 0.01 and
0.02 percent of neemjeevan. A highly significant decrease in the quantity of testes cholesterol was obtained in the 2-day, 5-day and 6-day old males. In 0-day, 1-day, 3-day and 4-day old the inhibition of cholesterol was found to be insignificant when it was treated with imidacloprid. In 2-day, 5-day and 6-day old males, a highly significant reduction in cholesterol was obtained by monocrotophos treatment while in 0-day, 1-day and 4-day old males the level of cholesterol was not significant. In 2-day old males derived from 0.004 percent concentration treated IV instar manifested a significant inhibitory effect in testis cholesterol while other concentrations didn’t affect considerably. Inhibitory effect caused by quinolphos on the quantity of testis cholesterol was significant in 2-day, 5-day and 6-day old males derived from IV instar treated with different concentrations. Although reduction in the level of cholesterol also was obtained in the 0-day, 1-day, 3-day and 4-day old adults by quinolphos. Oxydemeton-o-methyl also significantly inhibited testes cholesterol in the males of 2-day, 5-day and 6-day old derived from IV instar treated with different concentrations. 0-day, 1-day, 3-day and 4-day old derived from IV instar treated with 0.001, 0.002 and 0.004 percent of oxydemeton-o-methyl the level of inhibition was not significant as compared to that of untreated males.

Histopathology of reproductive organs of males and females of D. koenigii is also included in the present study in order to understand the degenerative changes occurred due to insecticides as well as neem formulations. Ovary of 4-day old female of D. koenigii is milky white and consists of seven telotrophic meroistic ovarioles. Each ovariole is composed of a terminal filament, germarium, vitellarium and pedicel. The ovariole is enclosed by an outer epithelial sheath and inner the tunica propria. The distal part of ovariole is germarium, which contains germ cells, trophocytes and prefollicular tissue. The central part is occupied by central core, trophic core to which trophocytes send finger like projections and lay down their nutritive components in to the trophic core. The posterior end of trophic core bears a number of cytoplasmic projections, the nutritive cords which extends to the developing oocytes. The prefollicular tissue consists of spindle- shaped cells
oriented with their long axis at right angles to the long axis of the ovariole. In vitellarium seven to eight oocytes are seen at various stages of development arranged according to maturity. The young oocytes gradually grow larger by incorporation of yolk and enclosed by 2-3 layers of follicular epithelium connected with nutritive cord to receive nutrition. It is a previtellogenic phase of oocyte when it leaves the distal part the nutritive cord is withdrawn. The nucleus of young oocyte, germinal vesicle is large in the previtellogenic phase whereas smaller in vitellogenic phase. The follicles are separated from each other by interfollicular tissue.

Histological degenerations have occurred in the ovary of 4-day old adult females derived from 0.04 percent multineem treated IV instar. The size and shape of germarium is invariably affected. Cells of prefollicular tissue are smaller but spindle in shape. Pycnosis was not observed in these cells. Tunica propria was almost intact and no change occurred in this layer. Trophic core looses its smoothness and becomes irregular in shape. Nucleus of the primary oocytes lying in the centre and chromatin material remained clumped. Cytoplasmic nutritive cord in multineem treatment loses it compactness. In vitellarium, follicular epithelial cells are 2 or 3 layers thick, which enclose the young oocytes but the cells are no more columnar in shape as has been observed in the untreated control. The nutritive cords are detached from young oocytes. Ooplasm is thick and condensed. Tunica propria is irregular in the proximal part of vitellarium. Chromatin material is highly dispersed which form a network in the nucleoplasm. Cytoplasm of these cells is vacuolated and containing certain endobodies. 4-day adult females derived from 0.004 percent of monocrotophos treated IV instar showed that growth of ovariole is significantly affected and size of ovariole is reduced. Tunica propria is slightly detached from germarium and vitellarium. Trophocytes are also not well developed becoming loosely packed and a few of them show pycnosis. The size of vitellarium is considerably reduced as compared to untreated control. The distal part of vitellarium consists of 3 developing oocytes and they do not attain their respective size as well as cytoplasmic cords are probably detached from them. Nucleus of oocyte,
germinal vesicle is significantly smaller than the untreated control. Chromatin granules are condensed. Yolk which is synthesized outside of the ovary is greatly impaired, therefore size of oocytes are not increased by incorporation of yolk. Though follicular epithelium is stretched and their cells are atrophied neither columnar nor squamous. Interfollicular cells are also highly disorganized and are compressed between the two oocytes. The female obtained from 0.004 percent oxydemeton-o-methyl treated IV instar showing a number of degenerations of different magnitude in the germarium and vitellarium. The germarium did not develop as much as compared to untreated control. The size of trophocytes is significantly reduced and a few of them are showing pycnosis. Only a few primary oocytes are found at the proximal part of germarium. The chromatin material is highly dispersed forming a network in vitellarium. The follicular cells which surround the young oocytes are highly irregular. The interfollicular layer between the two young oocytes is obliterated and compressed as well as disorganized in such a way that gives an impression of mass of overlapping cells. The ooplasm becomes thick and condensed. Nuclei of follicular cells didn’t divide amitotically because of oxydemeton-o-methyl.

4-day old male of *D. koenigii* consists of paired testis red in colour. Each testis is made up of seven testicular follicles. The epithelium of follicle is fibrillated in appearance. The distal most part of follicle is known as germarium contains germ cells, which divide mitotically to produce primary spermatogonia. Later on enclosed in a sheath as a cyst, which divide mitotically and then increased in size to become spermatocytes which undergo two mitotic divisions to produce four spermatids. These are transformed into spermatozoa in the zone of transformation where bundles are also formed. Testis of 4-day old adult male derived from 0.04 percent of multineem treated IV instar showed some histological degenerations but not significant. The spermatogonia insignificantly affected but a few of them showing clumping of chromatin materials as well as cytoplasm vacuolated. The size of the spermatocytes get decreased as compared to control. Testis obtained from survivor of 4-day old adult male derived from 0.004 percent
monocrotophos treated IV instar showed considerable histological degenerations. The spongy epithelium became irregular and more cavities are developed. Some of the spermatogonia showing their chromatin material are stained dark. A number of changes occurred in the spermatocytes i.e. shape is changed from hexagonal to elliptical and oval. The number of spermatids formed declined with monocrotophos treatment. The sperms are also formed but their number is quite less as compared to control. Histological changes occurred in the testis of 4-day old male obtained from IV instar treated with 0.004 percent oxydemeton-o-methyl which showed that the tunica externa is intact while spongy epithelial layer is showing more cavities than the control. Fibrillated testicular epithelium becomes weak and loses its compactness. In germarium, a few nuclei of spermatogonia are having clumped chromatin material. In certain spermatocytes mitotic division did not occur and their nuclei contain clumped chromatin materials and vacuolated. Similarly a number of degenerative changes occurred in the zones of maturations and transformation; number of spermatids decreased and scattered, degenerated and shrunken. Number of sperm bundles decreased and sperms are freely floating in proximal part of testis.