

## PHYSIOLOGICAL CHANGES OF IRRADIATED AND DISEASED MULBERRY SILKWORM, *BOMBYX MORIL*.

ABULYAZID I<sup>1</sup>, SOUAD M. MAHMOUD<sup>2</sup>, EL SHAFEI A. M.<sup>3</sup> AND TAHA, R. H<sup>2</sup>.

1- Atomic Energy.

2- Sericulture Res. Dept. , Agric. Res. Cent. Cairo- Egypt.

3- Faculty of Science, Ain Shams Univ.

(Manuscript received 25 November 2005)

### Abstract

The effect of radiation during egg stage with three different doses 10,20 and 30 Gy on healthy and diseased larvae with Flacherie (bacterial disease) was investigated.

Different biochemical parameters of haemolymph viz Total protein concentration, Transaminases enzymes; Glutamic- Oxaloacetic Transaminase (GOT) and Glutamic- Pyruvic Transaminase (GPT) which are responsible for silk synthesis and Alkaline and Acid Phosphatase for their important functions in metabolism, growth and synthesis of protein were estimated.

### INTRODUCTION

The effect of Gamma, X and UV rays on insects was studied by different scientists, Datta and Ray, 1977, Sengupta *et al.* 1977, Ovesenska and Jova, 1984, Kanarev and Dothi, 1985 and Yusifov *et al.* 1990 for two reasons, the production of these rays naturally in cosmic specially Gamma rays which lead to changes in different living organisms.

Also, to control insects through the usage of Gamma rays which extremely affect the genetic material DNA (Muller, 1962). These rays are necessary for normal being and growth of living organism (Kuzin, 1986) and lead to changes in different living organisms and cause mutation then evaluation.

Several authors selected different neutron fluence to irradiation silkworm eggs and found that , there was an appropriate fluence range could be applied to promote metabolism and physiological function in the silkworm body (Ruiying *et al.* 1985).

Because of Gamma radiation may be a useful tool in exploring a genetic variation for quantitative characters in silkworm (Singh, 1990).

The production of these rays in cosmic rays to control insect through the usage of Gamma rays which extremely affect the genetic material DNA (Muller, 1962).

Therefore, the present study was carried out to investigate the effect of Gamma rays on some biochemical parameters related to protein synthesis and production in the haemolymph and silk gland of healthy and diseased larvae from irradiated and non irradiated eggs at late stage of development.

## MATERIALS AND METHODS

Silkworm egg were obtained from The Sericulture Research Department of The Plant Protection Research Institute in Giza.

Irradiation was conducted at the Gamma Cell unit installed at the Middle Eastern Regional Radio Isotope Center for The Arab Countries in Dokki during the embryo stage with 10,20 and 30 Gy. Larvae hatched from irradiation eggs were reared under the laboratory condition according to Krishanswami, 1978.

Infected larvae with Grasserie and Flascherie were collected and be a part from control

The haemolymph was collected from healthy and diseased larvae according to (Takai and Tamashiro ,1975 ). Total protein concentration was estimated accroding to (Henry 1964 ).Glutamic –Oxaloacetic Trasaminase(GOT) and Glutamic – Pyruvic Transaminase (GPT) activities were estimated colourmetrically according to (Ritman and Frankel 1957) .Alkaline phosphatase was estimated colourmetrically according to (Kind and King 1954). Acid phophatase activity was estimated colourmetrically according to (Moss, 1984). The obtained data were manipulated statistically by using T-TEST analysis.

## RESULTS

### A-Haemolymph Biochemical Studies:

Different biochemical parameters of flascherie disease in irradiated and non-irradiated haemolymphal samples of *Bombyx mori* L. were reported in Table (1)

#### Total protein:

- a- Estimation of Total protein concentration in healthy and diseased non irradiated larvae revealed high significant difference between diseased and healthy (control) ones. It was reduced by 65.670 compared to the control in diseased larvae Table (1)

- b- The same trend was observed in 10 and 20Gy irradiated groups. The effect of irradiation on protein concentration in healthy larvae haemolymph revealed that 10Gy and 20 Gy doses increase the protein concentration ( 12.337 and 10.3, respectively). Low dose of 10Gy was higher than 20 Gy dose as revealed in Table 1.

#### **Transaminases enzymes:**

**a-** The estimation of Glutamic-Oxaloacetic Transaminase (GOT) activity showed a non-significant difference between Flacherrrie and healthy non-irradiated haemolymphal samples. Also, the activity of GOT in 10Gy and 20 Gy irradiated diseased larvae was not significantly differ with healthy ones.

**b-** On the other hand, Glutamic-Pyruvic Transaminase (GPT) activity of Flacherrrie diseased larvae showed a highly significant difference with healthy non-irradiated larvae as shown in Table (1).

Ones in 10 Gy dose of diseased larval haemolymph The GPT activity showed a significant difference with healthy , and a highly significant difference with healthy ones in the 20Gy irradiated larvae.

#### **Phosphatases enzymes:**

**a-** Estimation of Alkaline Phosphatase (AkP) activity for non-irradiated larvae, showed a significant difference between Flacherrrie diseased larvae and healthy ones while 10Gy irradiated larval haemolymph diseased with Flacherrrie was highly significantly differ with healthy ones and recorded a non-significant difference in AkP activity comparing to healthy ones in 20Gy irradiated haemolymphal samples.

Estimation of Acid Phosphatase (AcP) activity for non-irradiated group in diseased samples showed a highly significant difference comparing to healthy ones. But no significant differences was recorded in diseased larval haemolymph irradiated with 10Gy and 20Gy comparing with healthy ones .

#### **Silk gland Biochemical Studies:**

The same biochemical parameters were estimated for silk gland samples and reported in Table (2).

#### **Total protein:**

Total protein concentration of Flacherrrie diseased silk gland samples was not significantly differ with healthy (control) ones. The same trend was observed in 10Gy and 20 Gy irradiated samples.

**Transaminases enzymes:**

Glutamic-Oxaloacetic Transaminase (GOT) activity in Flacherrie diseased samples was highly significantly differ with healthy (control) ones. Also, the same trend was observed in 10 and 20Gy irradiated samples. On the contrary, The activity of Glutamic-Pyruvic Transaminase (GPT) in diseases larvae was not significantly differ with healthy ones in non-irradiated samples.

Also, the same in irradiated group with 10 Gy dose, Table 2.

On the contrary, in 20Gy irradiated silk gland samples of Flacherrie diseased larvae, GPT activity was highly significantly differ with healthy ones.

**Phosphatases enzymes:**

The estimated Alkaline Phosphatase (AkP) activity of Flacherrie diseased samples showed a non-significant difference with healthy (control) non-irradiated samples. In 10Gy irradiated silk gland samples of Flacherrie diseased larvae AkP was significantly differ with healthy ones,. While 20Gy irradiated samples showed a highly significant difference of compared to healthy ones.

Estimation of Acp of diseased samples of non-irradiated larvae showed a significant difference with healthy (control) ones asignificant difference between Flascherie diseased and healthy larvae was recorded in 10 Gy and highly significant difference in 20 Gy Table (2).

**DISCUSSION AND CONCLUSION****Haemolymph biochemical studies:**

Changes occurring in the haemolymph following pathological infections and irradiation could be expected to reflect physiological changes in tissues (Young and Lovell 1971; Abulyazid 1998).

**Total protein concentration:**

In the present study, total protein concentration in Flacherrie diseased larvae reduced by about (65.67%) compared with that of control. Mckinstry and Steinhauer (1970) in *Galleria mellonella*, Salem, et al (1980c) in *Philosamia ricini* and Barakat (1997) in *Galleria mellonella*, observed that, by increasing the dose of *Bacillus cereus* the protein levels decreased than control. This is may be due to their sharing in the formation of factors and enzymes that activate the phagocytic response of the haemolymph. Beard (1945) concluded that, the pathogenic infection produces drastic changes in the haemolymph proteins of the infected host. The larval death from

*Bacillus cereus* infection may result from the production of proteolytic enzymes (Bucher 1960) or from the reduction in haemolymph proteins (Lysenko 1972). The effect of irradiation on protein synthesis and inhibition in lepidopterous larvae was studied by several investigators, (Desrosier 1970) concluded that, ionizing radiation induced denaturation in proteins. Protein synthesis in lepidopterous cells is regulated in response to irradiation by means of synthesis of several protein types and inhibition of others (Rand and Koval 1994). This may explain the increment of protein concentration in 10Gy dose and the reduction of its level in 20Gy dose in the present study.

The present results concerning the effect of diseases on the protein concentration in fifth larval instar from irradiated eggs. It was found that, Flacherrie disease of 10Gy irradiated group reduces the protein concentration by about 58.76% deviated from control and by about 45.5 % in 20 Gy dose.

Biochemical parameters of Silkworm are known to be altered due to invasion by infectious pathogens (Ambika, *et al*/1996). Therefore, it was important to observe the change in enzymes activities, which have an important role in the formation of silk proteins as well as protein concentration during bacterial and viral infections.

#### **Transaminases enzymes:**

Silk proteins are synthesized in the haemolymph and silkgland from precursor amino acids by enzymatic transamination reactions (Mahmoud 1988). Glutamic acid is formed by amino transfer from aspartic acid by Glutamic-Oxaloacetic Transaminase (GOT) or from alanine by Glutamic-Pyruvic Transaminase (GPT), it is probably a very significant enzymatic activity in the final stage of development (Gowda and Ramaiah 1976).

The present investigation revealed that, GPT records lower activity than GOT enzyme in the haemolymph of healthy larvae. This result was confirmed by (Gowda and Ramaiah 1976) they recorded that, formation of alanine through enzymatic transfer of the amino group from either glutamic acid or aspartic acid was very slight after the second moult in *B. mori* L. larvae then decrease to undetectable levels at the final stage of development.

The obtained results revealed that, Flacherrie disease decreases the activities of both GOT and GPT enzymes by about (44.121 and 31.01%, respectively). Also, in 10Gy irradiated haemolymph decreases the activities of GOT and GPT enzymes by about (32.204 and 3.823%, respectively) and in 20Gy irradiated larvae, Flacherrie

disease decreases the activities of GOT and GPT enzymes by about (28.09 and 38.26%, respectively).

Mordue and Goldworthy (1973) and Tsou, *et al* (1979) found that, the changes in transaminases activities might be correlated with the protein metabolism. The trend of results in the present study support this suggestion, the protein concentration was related to the transaminases activities in the haemolymph of healthy and diseased samples of irradiated and non-irradiated groups. The interrelationships between protein synthesis and transaminases levels was confused by the hormonal control of protein synthesis and the neurosecretory hormones which involved in the regulation of transaminase levels (Ramadan, *et al* 2002).

#### **Phosphatases enzymes:**

Phosphatases enzymes were determined for their important functions in metabolism including the phosphate cycle, tissue transformation, growth, nerve action and synthesis of fibrous proteins (Mahmoud 1988). Alkaline Phosphatase plays an important role in the degradation and synthesis of tissue proteins (Denuce' 1952). Acid Phosphatase is important in biological processes that need high level of energy, such as development, growth, maturation and histolysis (Ray, *et al* 1984).

The presented results showed that, Acid Phosphatase predominates over Alkaline Phosphatase in the haemolymph of fifth larval instar of *B. mori* L. Pant and Lacy (1969) suggested that, Acid Phosphatase predominates over Alkaline Phosphatase when processes of tissue histolysis, histogenesis, differentiation and transformation are intensive in *Philosamia ricini*. Sridhara & Bhat (1963) noticed a steady increase in the activities of both enzymes in *B. mori* L. from hatching till the spinning stage followed by a conspicuous decrease at each moult. (Mahmoud 1988) recorded the absence of Alkaline Phosphatase in the haemolymph of *B. mori* L. and *Philosamia ricini* larvae. The absence of phosphatases enzymes in insect haemolymph may be an adaptation process to the vast energy needs during the formation of silk (Sridhara and Bhat 1963).

In the present study it was found that, Flacherrie disease increases the activity of Alkaline Phosphatase by about (70.454%), while it decreases the activity of Acid Phosphatase by about (57.915%) in the haemolymph of non-irradiated larvae. Wolfersberger (1984) found that midgut cells of lepidopterous larvae were rich in Alkaline Phosphatase activity. Due to destruction of midgut cells by bacteria or endotoxins, changes in basement membrane permeability took place, which permit an

ever increasing flow of midgut contents to the haemolymph (Loulouides and Heimpel 1969). The highly alkaline midgut contents increase the PH of the haemolymph to 8.1 (Heimpel and Angus 1959). This may explain the high Alkaline Phosphatase activity in the haemolymph of Flacherrie diseased larvae in this study. Due to bacterial infection, the acidity of haemolymph decreases (Aruga 1994). This may explain the decrease in Acid Phosphatase activity in the haemolymph of Flacherrie diseased larvae in the present study.

Results revealed that the inhibition in AcP activity by bacterial infection in the haemolymph of non-irradiated larvae reflect most probably a strong inhibition of ecdyson as postulated by (Bassal and Ismail 1985). Irradiation with 10 and 20Gy doses disturb this trend.

The results obtained revealed that, irradiation increases the activity of Alkaline Phosphatase, while it decreases the activity of Acid Phosphatase in the haemolymph of healthy larvae. Flacherrie disease decreases the activities of both Alkaline and Acid Phosphatases in 10Gy irradiated larvae by about (80.238 and 16.918%) deviation from control.

On the other hand, in 20Gy irradiated larvae, Flacherrie disease increases the Alkaline Phosphatase activity by about (10.503%) deviation over control and increases the Acid Phosphatase activity by about (29.019%) deviation over control.

### **Silkgland biochemical studies:**

#### **Total protein concentration:**

Silkgland protein concentration was estimated during the fifth larval instar to reveal the effect of diseases on the normal and irradiated larvae. It was found that, the protein levels in the silkgland are lower than in the haemolymph of healthy non-irradiated larvae. Mahmoud (1988) disagreed with these results, there was a higher levels of proteins in the silkglands of *B. mori* L. and *Philosamia ricini* during the fifth larval instar. Passent and Szafranski (1970); Singh, *et al* (1985) mentioned that, the silkglands in *B. mori* L. and *Philosamia ricini* formed proteins gradually during the fifth larval instar and reached a maximum level before spinning process.

Flacherrie disease increases protein concentration in the silkgland by about (62.799%) deviated over control.

Irradiation with 10 and 20Gy doses of healthy larvae increases the protein concentration in the silk glands. This may be due to the increase in silk gland cells resulting in an increase in the formation of liquid silk proteins (Aruga 1994).

Petkove, *et al* (1996b); Carneiro (1997) concluded the same results as irradiation stimulates the synthesis and secretion of silk proteins and also stimulates the increment of protein and nitrogen contents in the silk gland.

In the present study, Flacherrie disease of 10Gy irradiated samples decreases the protein concentration of 20Gy irradiated samples by about (33.202%) deviated from control. While, it increases protein concentration of 20Gy irradiated samples in the silk gland by about (83.536%) deviated over control

#### **Transaminases enzymes:**

Transaminases enzymes were involved in the uptake of nitrogen from mulberry leaves by silk gland and body tissues, which resulted in the subsequent promotion of silk protein synthesis (Li and Zhu 1985). Also transamination reactions are important in the formation of alanine from both aspartic and glutamic acids (koide, *et al* 1955; Gregoire, *et al* 1960).

The changes in GOT and GPT activities due to diseases in normal and irradiated silk gland were studied. GOT records a higher activity than GPT in the healthy irradiated and non-irradiated silk glands. This result was supported by (Singh, *et al* 1985) in *Philosamia ricini* and (Mahmoud 1988) in *B. mori* L. and *Philosamia ricini*. This might be due to the prevalence of aspartate and glutamate, which are the substrates of GOT enzyme, in the posterior silk gland (Fournier 1979).

In the present study, Flacherrie disease increases the GOT activity by about (11.321%) deviated over control, while it decreases the GPT activity by about (4.204%) deviated from control.

Irradiation with gamma rays promotes transaminases activation in healthy silk glands and by increasing the dose the activity increase and consequently protein levels. It is well known that in insects and other animals, transaminases are the key enzymes in the formation of non-essential amino acids and in the metabolism of nitrogen (Mordue and Goldworthy 1973). They concluded that changes in transaminases levels have been correlated with protein metabolism, as concluded in the present study.



Flacherrie disease in 10Gy irradiated larvae increases the GOT activity by about (7.284%) deviated over control, while it decreases the activity of GPT in the silk gland by about (9.479%) from control.

In 20Gy irradiated samples disease decreases the activities of both GOT and GPT by about (13.300 and 30.830%, respectively) deviated from control.

#### **Phosphatases enzymes:**

Acid and Alkaline Phosphatases are the key enzymes involved in the release of inorganic phosphorous and may be involved in meeting the increased energy demands (Barker and Lloyds 1973) and in DNA synthesis (Moore and Frazier 1976).

The obtained results revealed that, Acid Phosphatase predominates over Alkaline Phosphatase in the silk glands of healthy larvae. Sridhara and Bhat (1963), Mahmoud (1988) confirmed this result in the silk gland of *B. mori* L., while in the silk gland of *Philosamia ricini*, Alkaline Phosphatase was more active than Acid Phosphatase (Sirdhara and Bhat 1963; Zohdy 1965; Mahmoud 1988). Drihlon and Busnel (1954) concluded that, the Alkaline Phosphatase is located in cells that are most active in synthesis of fibrous proteins.

Flacherrie disease decreases the activities of both Alkaline and Acid Phosphatases in the silk gland by about (59.292 and 84.179%, respectively) deviation from controls.

Irradiation with gamma rays increases the activities of both phosphatases enzymes in the silk gland of healthy larvae. According to Wallis and Fox (1968), Alkaline Phosphatase enzyme may be under genetic control. Since the irradiation affect the genetic material in the irradiated samples (Abulyazid 1998), the Alkaline Phosphatase showed an increase in the present study due to irradiation in the haemolymph and silk gland samples.

Flacherrie disease in 10Gy irradiated silk glands, decreases the activities of both Alkaline and Acid Phosphatases by about (74.379 and 37.615%, respectively) deviated from controls.

While, in 20Gy irradiated silk glands it decreases both enzymes by about (97.448 and 67.963%, respectively) deviated from their controls. Also, Grasserie disease decreases their activities by about (94.896 and 87.527%, respectively) deviated from controls.

In the present study, infection with bacteria in haemolymph and silk gland samples decreases protein concentration and AcP activity and this trend did not

change with irradiation. These results disagree with the results obtained by (Mushtaq and Shakoori 1996) who mentioned that, *Bacillus thuringiensis* did not disturb biochemical components analyzed in the adult beetles of *Tribolium castaneum* except AcP and soluble protein.

### REFERENCES

1. Abdel Hafez, M. M.; Abd El-Naby, A. A; El-Malla, M. A Abdel Fattah, M. S. and Abbass, M. G. (1984): Some biochemical studies on the Egyptian cotton leafworm, *Spodoptera littoralis* (Boi III- Changes in GOT and GPT enzymes activities during different developmental stages. *Minia J. Agri. Res. & Dev.*, Vol. 6(4): 541-554.
2. Ambika, T.; Narayanaswamy, T. K.; Gobindan, R. and Krishnamoorthy, R. V. (1996) : Changes in some organic components in tolerant and susceptible breeds of *Bombyx mori* L. due to infection by *Beauveria bassiana* (Bals.). *Bull. Sericu. Res.*, (7): 55-59.
3. Barakat, E. M. (1997): A comparative study on the immune response of the Wax moth, *Galleria mellonella* (Linn.) to some biotic and abiotic materials. Ph. D. thesis. Faculty of science, Ain Shams Univ.
4. Beard, R. L. (1945): Studies on the Milky disease of Japanese beetle larvae. *Cann. Agri. Exp. Sta. Bull.*, 491:505-509.
5. Bucher, G. E. (1960): Potential bacterial pathogens of insects and their characteristics. *J. Insect Pathol.*, 2:172-195.
6. Datta, R. K. and Roy, G. C. (1977): X-ray induced mutagenesis in silkworm. Annual Research and Administrative Report (1976-1977), Berhampur (West Bengal). Central Silk Board, Bombay- 2, India 1011-1017.
7. Denuce, G. M. (1952): Recherchs sur le systeme phosphatasique de glandes sericigenes chez le ver a soie, *Bombyx mori* L .
8. Desrosier, N. W. (1970): The technology of food preservation. AVI Publishing Company, INC. London, PP.: 313.
9. Drihlon, A. and Busnel, R. G. (1954): Recherthes sur les phosphates d'insects et enparticulier des tubes des malpighi et du tube digesif. *Bull. Soc. Zool.*, France, 70: 40-47.

10. Gowda, V. T. and Ramaiah, T. R. (1976): Effect of induced polyhedrosis on haemolymph transaminases in the larvae of the Silkworm, *Bombyx mori* L. J. Inver. Pathol., 28 (2): 271-272.
11. Gregoire, L. (1955): Effects of ionizing radiation on coagulation. "The Physiology of Insecta ."VOL III, Academic Press.,New York and London, PP.: 153-185.
12. Heimpel, A. M. and Angus, LA. (1959): The site of action of crystalliferous bacteria in lepidopterous larvae. J. Insec. Pathol., (I): 152-170.
13. Henry, R. J. (1964): Clinical chemistry. Harper and Row Publishers, New York, PP.: 181.
14. Kanarev, G. and Dothi Chan, (1985): Effect of laser irradiation of silkworm eggs of *Bombyx mori* development and productivity . Zhivotnov'd Nauki 22:47-53.
15. Kind, P. R. N. and King, E. J. (1954): Estimation of plasma phosphatase by determination of hydrolysed phenol with amino antipyrine. J. Clin. Path., 7: 322-326.
16. Krishnaswami, S. (1978): New Technology of silkworm rearing. Bull. No. 2. Central Silk Boare India, 4: 111-116.
17. Kuzin, A. M. (1986): Structure metabolic theory. Hauka; 284.
18. Louloudes, S. J. and Heimpel, A.M. (1969): Mode of action of *Bacillus thuringiensis* toxic crystals, in larvae of the Silkworm, *Bombyx mori* ]. Invest. Pathol., 14: 375-380.
19. Lysenko, O. (1972): Some characteristics of *Galleria mellonella* haemolymph proteins. J. Insect. Pathol., 19: 335-341.
20. Mckinsty, D. M. and Steinhauer, A. L. (1970): Disc electrophorasis of septicemic and melanized plasma from Greater waxmoth larvae, *G. mellonella*. J. Invert. Pathol., 16: 123-126.
21. Moss, D. W. (1984): Methods of enzymatic analysis. Ed. H.U.Bergmeyer, Varlag. Chemie, 3rd edition, 4: 92-106.
22. Muller, A. (1962): Efficiency of radical production by x-rays in dry proteins and nucleic acids.Intern J. Radia. Biology and Related Studies in Physics, Chemistry and Medicine, 5:192-200.

23. Ovesenka, L. and Jova, U. (1984): Effect of helium-neon laser irradiation of *Bombyx mori* breed in egg stage on the productive characters. Genet. Sel. 17: 394-403.
24. Pant, R. and Lacy, P. S. (1969): Phosphatase activity in *Philosamia ricini* during development. Indian J. Biochemi., 6: 154-156.
25. Ramadan, K.; El-Bermawy, S. M. and Abulyazid, I. I. (2002): Biochemical studies on the pupal stage of Mediterranean fruit fly, *Ceratitidis Capitata* (Wied) after irradiation of egg stage with chronic gamma dosages. J. Molecular Biology. (Unpublished).
26. Rand, A. and Koval, I. M. (1994): Coordinate regulation of proteins associated with radiation resistance in cultured insect. (eus. Radiation Research, 138 (1): 513-516.
27. Ray, A.; Rao, C. G. P.; Srivevi, R. and Ramamurty, P. S. (1984): Changes in acid phosphatase activity in *Spodoptera litura* (Lepi: Noctuidae) during the post embryonic and adult development. Entomon., 9(3y 161-167
28. Ritman, S. and Frankel, S. (1957) :Am. J. Clin. Pathol., 28: 56-66
29. Ruiying, Z.; Infen, Z.; Dingzh, C. and Jinxian, R. (1985): Study on increasing production of natural silk by using low irradiation. Trans. Am. Nucl. Soc., 46: 407-410.
30. Salem, M. S.; Eid, M. A. and ElO Maasarawy, S. A. S. (1980c): Biochemical studies on the haemolymph of the Eri- Silkworm, *Philosamia ricini* (Boisd.) under varied conditions. Proc. 1<sup>st</sup> Conf. Pi, Prot. Res. Ins., 11: 223-236.
31. Sengupta, A.; Datta, R.; Das, S. K. (1977): Gamma ray induced mucagenesis in silkworm for improvement of deteriorated evolved race D14b. Ann. Res. And Admin. Dep. (1976-1977) Berhampour (West Bengal). Central Silk Board. Bombay 2, India. 1.17-1.19.
32. Shigematsu, H. (1958): Synthesis of blood protein by the fat body in the Silkworm, *Bombyx mori* L. Nature, 102; 880-881.
33. Shigematsu, H. and Takeshita, H. (1948): Changes in quantity of nucleic acid and protein in the fat body of the Silkworm in a course of contracting Jaundice. J. Sericul. Sci., Japan, 30:66-70.

34. Shylaja, M. and Ramaiah, T. (1984): Isoenzymes of glutamate oxaloacetate transaminase in the larvae of Silkworm, *Bombyx mori* L. Infected with Nuclear Polyhedrosis Virus. *Experientia*, 40(7): 717-718.
35. Singh, R.; Nagaraju, J.; Vijayaraghavan, K. and Prema, L.(1990): Radiation sensitivity of the Silkworm, *Bombyx mori* L. *Indian J.Seric.*, 29(1): 1-7.
36. Takai, G. H. and Tamashiro, M. (1975): Changes observed in haemolymph proteins of the Lawn armyworm, *Spodoptera maturitia acronyctoides* during growth, development and exposure to a Nuclear Polyhedrosis Virus (NPV). *J. Inver. Pathol.*, 26:147-158.
37. Tu-Zhen, L. A.; Shirai, K. A.; Kanekatsu, R. A.; Kiguchi, K.A.; Wallis, B. B. and Fox, A. S. (1968): *Phosphatase isoenzyme: Isoenzymes.* Chapman and Hall LTD, 11 New Fetter Lane., London. PP:267.
38. Young, S. Y. and Lovell, J. S. (1971) :Haemolymph proteins of *Trichoplusia ni* during the course of a Nuclear Polyhedrosis Virus (NPV) infection. *J. Invert. Pathol.* ,17:410-418.
39. Yusifov, N. I. Kuzin, A. M.; Agaev, F. A.; Atieva, S. G. (1990): The effect of low level ionizing radiation on embryogenesis of silkworm, *Bombyx mori*. *Radiat. Environ. Biophys.* 29: 323-327.

Table (1): Diseased and healthy haemolymphal biochemical assay in the irradiated and non-irradiated larval stage of *Bombyx mori* L.

	Total Protein (g/dL)			Glutamic-oxaloacetic transaminase GOT (Unit/ml)			Glutamic-Pyruvic Transaminase GPT (Unit/ml)			Alkaline-phosphatase AkP (U/100 ml)			Acid-phosphatase AcP (U/100 ml)		
	0.0 Gy	10 Gy	20 Gy	0.0 Gy	10 Gy	20 Gy	0.0 Gy	10 Gy	20 Gy	0.0 Gy	10 Gy	20 Gy	0.0 Gy	10 Gy	20 Gy
Healthy Control	8.41 ± 0.21	12.33 7± 0.24	10.3 ± 0.21	137.5 ± 34.3	108.16 7± 21.1	163.16 7± 30.8	22.8 ± 0.41	34.0 ± 0.29	43.90 ± 0.58	2.64 ± 0.20	3.279 ± 0.32	3.47 5 ± 0.28	369.9 97 ±10.9	97.4 55± 14.0 4	332.9 63 ± 12.47
Flacherrie	2.887 ±0.05 ***	5.087 ± 0.19** *	4.679 ±0.22 ***	76.83 3 ±14.4 *	73.333 ± 4.8*	117.33 3 ± 31.9*	15.73 ±0.37 ***	32.7 ± 0.25**	27.10 ±0.26 ***	4.50 ± 0.25**	0.648 ±0.00 3***	3.84 ±0.2 4*	155.7 12±1 2.07** *	80.9 68±3 5.5*	429.5 87 ±133. 01*
% Change	- 65.67 2	- 58.76 6	- 54.57 3	- 44.12 1	- 32.204	- 28.090	- 31.01	- 3.823	- 38.26 9	70.45 4	- 80.23 8	10.5 03	- 57.91 5	- 16.9 18	29.01 96
Grasserie	49.76 7 ±0.26 ***	17.76 8 ±0.05 8***	24.37 2 ±0.05 8***	192.5 ± 25.4*	187.37 ± 0.27**	225.0 ± 0.29*	34.0 ± 0.29** *	57.33 ± 0.44** *	55.20 ± 0.11** *	0.573 ± 0.04** *	2.837 ± 0.05*	3.64 5 ±0.1 7*	38.38 ±12.1 0***	110. 45 ± 0.28*	74.76 ± 0.32** *
% Change	491.7 60	44.02 2	136.6 21	40.00	73.223	38.202	49.12 3	68.61 7	25.74 0	78.29 5	13.48 0	4.89 2	89.62 7	13.3 34	77.54 7

P &lt; 0.001 highly significant (\*\*\*)

P &lt; 0.01 Significant (\*\*)

P &lt; 0.05 non-significant (\*)

Table (2): Diseased and healthy silk gland biochemical assay in the irradiated and non-irradiated larval stage of *Bombyx mori* L.

	Total Protein (g/dL)			Glutamic-oxaloacetic transaminase GOT (Unit/ml)			Glutamic-Pyruvic Transaminase GPT (Unit/ml)			Alkaline-phosphatase AkP (U/100 ml)			Acid-phosphatase AcP (U/100 ml)		
	0.0 Gy	10 Gy	20 Gy	0.0 Gy	10 Gy	20 Gy	0.0 Gy	10 Gy	20 Gy	0.0 Gy	10 Gy	20 Gy	0.0 Gy	10 Gy	20 Gy
Healthy Control	0.376 ± 0.10	0.506 ± 0.06	0.741 ± 0.07	97.9 ± 0.06	106.63 3± 0.35	130.75 ± 0.20	46.46 ± 1.19	48.53 ± 3.71	50.25 7 ± 1.26	1.017 ± 0.25	2.295 ± 0.31	1.68 5 ± 0.25	9.898 ± 1.16	21.6 14± 0.17	76.60 9 ± 1.07
Flacherrie	0.612 ±0.06*	0.338 ± 0.02*	1.360 ±0.42*	108.9 33 ±0.27***	114.4 ± 0.29***	113.36 ± 0.25***	44.50 7 ±0.19*	43.93 ± 0.53*	34.76 3 ±0.27***	0.414 ± 0.03*	0.588 ±0.11**	0.04 3± 0.00 2***	1.566 ±0.84**	13.4 84±2 .24**	24.54 3 ±0.46***
% Change	62.79 9	- 33.20 2	83.53 6	11.32 1	7.284	- 13.300	- 4.204	- 9.479	- 30.83 0	- 59.29 2	- 74.37 9	- 97.4 48	- 84.17 9	- 37.6 15	- 67.96 3
Grasserie	0.256 ±0.09*	0.573 ±0.15*	0.083 ±0.00 7***	102.8 66 ±0.31 8***	117.53 3 ± 0.33***	98.0 ± 0.312**	42.66 7 ± 0.46*	43.93 ± 0.39*	40.32 ± 0.21**	0.595 ± 0.11*	0.430 ± 0.02**	0.08 6 ±0.0 04***	6.161 ±3.01*	30.4 52 ± 1.07*	9.555 ±0.18 9***
% Change	- 31.91 5	13.24 1	- 88.79 9	5.073	10.222	- 25.048	- 8.164	- 9.479	- 19.77 2	- 41.49 5	- 81.26 3	- 94.8 96	- 37.75 5	40.8 90	- 87.52 7

P < 0.001 highly significant (\*\*\*)

P < 0.01 Significant (\*\*)

P < 0.05 non-significant (\*)

## مشاكل النخيل و إنتاج التمور بإدارة أوقاف صالح الراجحي

رمزى أبو عياته

الإدارة الزراعية بإدارة أوقاف صالح الراجحي- المملكة العربية السعودية

التعريف بالإدارة الزراعية بإدارة أوقاف صالح الراجحي.

التحول من الزراعات التقليدية إلي الزراعات النظيفة ثم الزراعة العضوية لجزء من مساحة مشروع الباطن.

تسجيل مشروع نخيل الباطن في موسوعة غينيس للمعلومات العامة والأرقام القياسية مايو ٢٠٠٥. أهم آفات وأمراض النخيل بمشاريع الإدارة

أولاً: أهم الآفات الحشرية:

الحفارات ( حفار العزوق- حفار الساق ذو العزوق الطويلة - حفار السعف).

سوسة طلع النخيل.

دبور البلح.

ديدان البلح ( فراشة التمر الكبرى- فراشة التمر الصغرى).

النمل الأبيض.

الحشرات القشرية.

حشرات المخازن ( خنفساء سورنيام- فراشة المخازن-ذبابة الدروسفيلا).

عدم تسجيل سوسة النخيل الحمراء بمنطقة القصيم.

ثانياً: أهم الآفات المرضية:

مرض موت القسائل الدبلودي.

مرض الخامج أو تعفن الطلع.

٣- مرض الذبول الوعائي.

مرض جرب أو لفحة العرجون.

مرض السعفة المحروقة.

مرض تبقع الأوراق.

العظم الجاف ( اليباس).

مرض تعفن الجذور.



ثالثا: الآفات الأكاروسية:

أكاروسات حلم الغبار ( الغبيرة)

رابعا: الآفات النيماطودية

(نيماطودا تعقد الجذور)

خامسا: القوارض

سادسا: الطيور البرية

سابعا: الحشائش

(عريضة ورقيقة الأوراق) معمرة وحولية.

الطرق الوقائية

أولا: الطرق الوقائية لغرس الفسائل:

الطرق الوقائية لأختبار الفسيلة.

الطرق الوقائية قبل غرس الفسيلة.

الطرق الوقائية أثناء غرس الفسيلة.

الطرق الوقائية بعد غرس الفسيلة.

ثانيا : الطرق الوقائية في بساتين النخيل :

١- التغذية المتوازية ( الأسمدة ( معدنية - عضوية ) مياه).

الإكتشاف المبكر للإصابة ( الفحص الدوري الحقلّي المستمر).

استخدام المصائد الضوئية ( سواء مصدر كهربي - خلايا شمسية).

استخدام المصائد الفرمونية - الكرمونية (سوسة النخيل).

للتنبؤ بالإصابة.

قطع وحرق الأجزاء المصابة.

ثالثا: الطرق الوقائية للحد من الإصابة في المستودعات:

الحصاد المبكر والفرز أول بأول.

استبعاد التمور المتساقطة بأحواض النخيل وتحويلها لتمور أعلاف.

تطهير المستودعات بخليط من مبيد حشري (قطري عديم الرائحة).

استخدام المصائد الكهربائية الصاعقة بالمستودع.

فلسفة الإدارة في إختيار المبيدات الكيميائية

تأمين مادة ناشرة.

تغيير المبيدات المستخدمة سنويا.

تكون قابلة للخلط ( فطرية - حشرية ) ٢٥٠ . ٠٠٠ .

مبيدات سائلة أفضل من البودرة خاصة التي ترش علي الثمار للأكاروسات.

### الطرق العلاجية (الزراعات النظيفة)

١-إجراء رشه وقائية علاجية بمبيد فطرى بعد الحصاد(نوفمبر)

٢-إجراء رشه وقائية علاجية بمبيد(حشري- فطرى) خلال نهاية مارس وأوائل إبريل. الرش بناء على البيانات السابقة في ظهور الآفات.

٣-إجراء رشة وقائية علاجية للأكاروسات حلم الغبار بمبيد اكاروس مناسب خلال الفترة من نهاية مايو إلى نهاية يونيو بناءً على البيانات الموجودة بمواعيد ظهور الآفة.

٤-إجراء رشة علاجية للحشائش خلال الفترة من منتصف فبراير - حتى منتصف مارس بمبيد جلاتيوسيت.

### مع ملاحظة:

أ-التعشيب أولاً بعد الحصاد ثم الرش بعد أن يصل ارتفاع الحشيشه إلى ١٠سم.

ب-إضافة ٢ كجم يوريا لكل ١٠٠٠ لتر ماء مع المبيد.

ج-استخدام المادة الناشرة.

د-زيادة المعدل بالنسبة لحشيشه الحلقا والحقنه.

هـ-عدم التعشيب بعد إجراء عمليات الرش.

كيفية علاج بؤر الإصابة:

١-حفار العزوق(العنقرة).

٢-النمل الأبيض.

٣-الحشرات القشرية.

٤-الذبول الوعائي.

٥-جرب العزوق.

٦-النيماتودا.

طرق مكافحة القوارض باستخدام:

محطات الطعوم السامة وبعض المبيدات.

غاز الفوستوكسين.

**ملحوظة:**

لا يقتصر خطورة القوارض علي الخسائر في الفسائل والتمور بل تدمير حواف الأحواض مما يسبب تهريب وتسريب مياه الري.

**أهمية تحليل متبقيات المبيدات في:**

-التمور .

-النوي .

-التربة .

كأساس لتدعيم برامج مكافحة.

**برامج الوقاية والمكافحة في حقول النخيل العضوية:**

١-مكافحة الحشرة القشرية ← زيت معدني.

٢-مكافحة الحفارات ← مصادد ضوئية.

٣-مكافحة الفراشات ← مصادد ضوئية + مبيد النيم + التريكوجراما.

٤-مكافحة الاكاروسات ← مبيد بيكو + مبيد النيم.

٥-مكافحة اللفحات ← كبريت ميكروني.

**المشاكل التي تواجهنا في الإدارة بخصوص****الوقاية والمكافحة لآفات النخيل**

١-تشقق الثمار في مرحلتي الخلال والبشر (المسبب غير معروف حتى الآن).

٢-عدم وجود طريقه فاعله لمكافحة الحشائش في حقل الزراعة العضوية.

٣-في ظل تحريم استخدام مبيدات بروميد الميثايل والفوسفوكسين فما البديل لتبخير التمور ١٠٠٠٠ طن سنوياً.

٤-هل نطمع في وجود مصادد فرمونية للحشرات القشرية والحفارات.

**الطموحات:**

١-وضع حلول مناسبة للمشاكل التي أثرت بالشريحة السابقة.

٢-وضع توصية مناسبة تتعلق بـ:

١-إعداد مواصفة للتمور محليه.

٢-إصدار مجلة للنخيل والتمور علي المستوي المحلي أو العربي.

٣-التوسع في المبيدات الحيوية لخدمة الزراعة العضوية وذلك إن أمكن.

## توصيات المؤتمر الدولي الثالث لمعهد بحوث وقاية النباتات

٢٦-٢٩ نوفمبر ٢٠٠٥

في ختام أعمال المؤتمر الدولي الثالث لمعهد بحوث وقاية النباتات أعلن السيد الأستاذ الدكتور احمد عبده حامد مقرر عام المؤتمر التوصيات العامة في الجلسة الختامية للمؤتمر وهذا نص البيان الرسمي للتوصيات:

قام معهد بحوث وقاية النباتات - مركز البحوث الزراعية بتنظيم وعقد المؤتمر الدولي الثالث للمعهد تحت عنوان " دور مكافحة الآفات في الإدارة المتكاملة للمحاصيل وأثرها على سلامة البيئة والمنتجات الزراعية " خلال الفترة من ٢٦ - ٢٩ نوفمبر ٢٠٠٥ م بمقر المركز الدولي للزراعة بمصر .

وقد عقد المؤتمر لتحقيق عدة محاور رئيسية وهي استخدام التقنيات الحديثة لإدارة المحاصيل الزراعية بمفهوم أعم وأشمل لمكافحة الآفات من بداية الزراعة وحتى جنى المحصول وكذلك تنمية مهارات مكافحة الآفات بعد حصاد وجمع المحاصيل المختلفة والتخلص من بقايا المحاصيل وذلك لكسر دورة حياة الآفات التي تسكن بداخلها وتهاجم المحاصيل التالية لها كما يؤدي ذلك إلى وجود بيئة نظيفة خالية من مخلفات المحاصيل

كذلك التوسع في برامج تربية وتقييم السلالات النباتية المقاومة التي تقاوم وتتحمل الإصابة بالآفات والأمراض ويتضمن ذلك التعاون الوثيق في مجالات الهندسة الوراثية وتربية المحاصيل ومكافحة الآفات والأمراض

كما تتضمن محاور المؤتمر دور المكافحة في وقاية المنتجات الزراعية بعد الحصاد و أثناء التخزين وذلك لرفع كفاءة التصدير من حيث الكم والجودة .

وقد أكد السادة العلماء المشاركين في المؤتمر من مصر والدول العربية والأجنبية على ضرورة استخدام مركبات آمنة للإنسان والحيوان والمحاصيل الزراعية وذلك للحفاظ على البيئة والمنتجات الزراعية والتركيز على دور الأعداء الحيوية في المكافحة والحد من الاستخدام المسرف والخطأ للمبيدات الحشرية .

• حضر المؤتمر أكثر من ٤٠٠ من العلماء والباحثين من الجامعات ومراكز البحوث والشركات العاملة في المجال وقد حضر المؤتمر ما يقرب من ٤٥ باحثا وعالما من ست عشرة دولة عربية و أمريكية .

- تضمنت أعمال المؤتمر ١٦ جلسة علمية شملت جميع محاور المؤتمر هذا بالإضافة إلى ورشة عمل على هامش أعمال المؤتمر عن مشاكل النخيل و إنتاج التمور في الوطن العربي.
  - نوقشت خلال هذه الجلسات عدد ١١٧ ورقة علمية تركز محتواها على البحوث التطبيقية التي تسهم في حل مشاكل الإنتاج الزراعي.
- وقد أقر السادة الباحثين والعلماء المشاركين بالمؤتمر عدة توصيات محددة دون إسهاب لتكون في متناول التطبيق الفعلي لأهداف المؤتمر أمام صانعي القرار:
- ١- إرسال برقية تهنئة وشكر وتقدير الى معالي السيد المهندس احمد الليثي وزير الزراعة واستصلاح الأراضي بمناسبة فوز سيادته في الانتخابات البرلمانية ولدعمه وتشجيعه للمؤتمر.
  - ٢- إرسال برقية شكر وتقدير إلى السيد الأستاذ الدكتور عبد العظيم الطنطاوي بدوي رئيس مركز البحوث الزراعية لدعمه وتشجيعه للمؤتمر مع الاقتراح بان يقوم سيادته بإرسال برقية شكر إلى السيد رئيس الجمهورية لرعايته للبحث العلمي والعلماء.
  - ٣- العمل على تأصيل فكرة إدارة المحصول من بداية تجهيز الأرض للزراعة وحتى الحصاد للسيطرة على عدد كبير من الآفات وخاصة التي تقضى جزء من حياتها في التربة .
  - ٤- التوسع في إنشاء قواعد البيانات الخاصة بالآفات والحد الحرج والحد الحرج الاقتصادي .
  - ٥- الاهتمام بتعريف المشاكل الزراعية بدقة وبصفة خاصة مشاكل الضرر الناشئ عن الآفات والأمراض حتى يمكن اتباع التوصية المناسبة في التوقيت المناسب حيث يؤدي ذلك إلى إنتاج زراعي آمن وتقليل تكاليف الإنتاج وبالتالي زيادة العائد الاقتصادي للمحصول .
  - ٦- التوسع في نشر برامج التنبؤ لتوقع الإصابة والاستعداد لها قبل حدوثها.
  - ٧- التركيز على استخدام المركبات الحيوية و المستخلصات النباتية كبديل نظيف وأمن على المحاصيل والحيوانات والإنسان والبيئة .
  - ٨- تشجيع وإكثار الأعداء الحيوية من طفيليات ومفترسات وممرضات حشرية متخصصة معزولة من البيئة المحلية واستخدامها في برامج مكافحة ضمن التتابعات الموضوعة في خطة المكافحة كبديل للمبيدات .
  - ٩- عدم التوسع في استخدام الممرضات الحشرية المنتجة في الخارج إلا بعد التأكد من وجود هذه السلالات في البيئة المحلية مع الاهتمام بعناصر الأمن الحيوي لهذه الممرضات.
  - ١٠- تشجيع وتنمية برامج تربية النباتات المقاومة للإصابة .

- ١١- الاهتمام بتدوير المخلفات النباتية للاستفادة من بقايا المحاصيل الزراعية بهدف التخلص من الآفات وعدم حرق هذه المخلفات مما يؤثر على سلامة البيئة .
- ١٢- توفير الوسائل البديلة والأمنة لمكافحة آفات المخازن للحفاظ على المخزون الغذائي وعدم تلوثه بمركبات سامة لرفع الكفاءة التصديرية للمحاصيل المختلفة .
- ١٣- زيادة الدعم للبحث العلمي والباحثين حتى يمكن للباحثين القيام بعملهم على اكمل وجه والخروج بنتائج ملموسة في كافة مجالات الزراعة .
- ١٤- توثيق التعاون بين الهيئات البحثية والأجهزة التنفيذية لتحقيق أقصى استفادة من نتائج البحوث العلمية.
- ١٥- الاهتمام بدور المدارس الحقلية وتدعيمها .
- ١٦- الاستفادة من وسائل الإعلام السمعية والمرئية والمقروءة في توصيل نتائج البحوث الحديثة إلى المزارع.
- ١٧- الاهتمام بدور الإرشاد الزراعي في توصيل نتائج البحوث إلى المزارعين.
- ١٨- عرض وتسويق لنتائج البحوث التي تسهم في حل المشاكل التطبيقية المؤثرة في الإنتاج القومي .
- ١٩- دعم وتشجيع استمرارية المشاريع البحثية و التطبيقية و التي أثبتت حالياً نجاحها في مجال مكافحة البيولوجية والحفاظ على البيئة والتي تضمن إعادة التوازن الطبيعية للبيئة المصرية بعد الاستخدام العشوائي للمبيدات .
- ٢٠- التركيز على الحفاظ على حقوق الملكية الفكرية عند النقل من المصادر المختلفة وبصفة خاصة شبكة المعلومات الدولية خاصة انه الآن اصبح النقل غير الشرعي مجرم دولياً ومحلياً .
- ٢١- التوصية بعقد ورشة عمل متخصصة لمشاكل النخيل و إنتاج التمر في مصر في أقرب فرصة ممكنة .
- ٢٢- إنشاء جمعه عربية مختصة بآفات النخيل.
- ٢٣- تدعيم أجهزة الحجر الزراعي في المنافذ المختلفة بأحدث الأساليب للكشف عن الحشرات وسهولة تعريفها حتى لا تتسرب الى البلاد آفات جديدة.
- ٢٤- التوصية بعقد المؤتمر الدولي الرابع لمعهد بحوث وقاية النباتات في نوفمبر ٢٠٠٨ .