Bioenergetic Deficiency and Oxidative Stress in Rat Liver Mitochondria Exposed to Lambda Cyhalothrin and the Protective Role of Propolis

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Abstract: Mitochondria a primary site of cellular energy generation and oxygen consumption represents a likely target for lambda cyhalothrin (LC) poisoning, a type II synthetic parathyroid insecticide used for agricultural and public health applications. Therefore, the current study was planned to investigate the effect of LC exposure on rat liver mitochondrial electron transport chain as well as the protective effect of 100 mg / Kg bw propolis against LC adverse effects. Male albino rats were subjected to repeated oral sub lethal dose (0.5 mg / Kg bw, 1/50 LD_{50}) of LC for four weeks (3 doses / week) and two weeks for observation. Two of mitochondrial bioenergetic biomarkers; (nicotinamide adenine dinucleotide dehydrogenise) or NADH dehydrogenase (Complex I), adenosine triphosphatase (ATPase) activities and mitochondrial oxidative stress biomarkers; superoxide dismutase (SOD) activity, reduced glutathion (GSH) content and the lipid peroxidation (LPO) levels were *in vivo* assessed in rat liver. The alterations in the previous tested parameters were confirmed by the changes in hepatic mitochondrial proteins pattern induced by LC using sodium dodecyl sulphate polyacrylamide gel electrophoresis (SDS - PAGE) technique. Results showed significant reduction in hepatic mitochondrial; NADH dehydrogenase, ATPase, SOD activities and GSH content in LC treated rats, while the LPO level was significantly increased compare to control. SDS- PAGE depicted some alteration in the mitochondrial proteins pattern. The previous effects were attenuated by propolis supplementation. This study revealed that LC toxication impaired mitochondrial bioenergetics and induces an oxidative stress which may lead to liver dysfunction. Moreover propolis attenuated the LC adverse effect due to its high contents of flavonoids and phenolics compounds supporting it's highly use as antioxidant food supplement.

Keywords: Lambda cyhalothrin, Propolis, Mitochondria, Complex I, ATPase, Oxidative Stress, SDS - PAGE.

1-Introduction

Lambda cyhalothrin is widely used in Egypt and valued to control a wide range of pests in cotton, cereals and vegetables as well as in public health application against insect, ticks and flies **(Abdel Aziz and Abdel Rahem, 2010).** Consistent with LC lipophilic nature **(Michelangeli *et al,* 1990),** LC has been found to accumulate in biological membranes leading to oxidative damage by altering antioxidant systems and increasing lipid peroxidation in mammals **(El-Demerdash, 2007 and Fetoui *et al,* 2009)**.

Animal liver plays a major role in regulating various physiological and chemical functions of animal bodies such as catabolic and anabolic processes as well as synthesis and secretion systems of xenobiotics. Any physiological damage can be repaired but may also accumulate over a period of time and cause many degenerative diseases **(Ames *et al,* 1993)**. LC is rapidly metabolized in the liver via hydrolytic ester cleavage and oxidative pathways by the cytochrome P450 enzymes yield reactive oxygen species (ROS) **(Sankar *et al,* 2012).** Mitochondria are the main energy source in hepatocytes and play a major role in extensive oxidative metabolism and normal function of the liver. This key role also assigns mitochondria a gateway function in the centre of signalling pathways that mediate hepatocytes injury, because impaired mitochondrial functions affect cell survival and contribute to the onset and perpetuation of liver diseases. In physiological conditions, mitochondria are the major source of ROS. While producing energy necessary to convert (Adenosine diphosphate) ADP into (Adenosine triphosphate) ATP, mitochondria also produces superoxide radical ($O_2^{\bullet-}$), hydrogen peroxide (H_2 O_2) and hydroxyl radical ($OH^{\bullet}$) as by-products of the consumption of molecular oxygen in the electron transport chain (**Raha and Robinson 2000**, **Sastre 2000**) and the accumulated $O_2^{\bullet}$ is eliminated by manganese superoxide dismutase (MnSOD), which generates H_2O_2 (**Raha and Robinson 2000**, **Raha *et al,* 2000**). Because mitochondria do not contain catalase (CAT), the mitochondrial redox system, which consists of reduced glutathione (GSH), glutathione peroxidase (GPx) and glutathione reductase (GR), plays a crucial role in reduction of H_2 O_2 and protects mitochondria against peroxidative stress (**Kowaltowski and Vercesi 1999**, **Han *et al,* 2003, Maher 2005)**. Ddisorders in the oxidative reductive status of mitochondria leading to changes of a structure and function of this organelle.

During the last decades, phenolic compounds have received considerable attention due to their antioxidant properties **(Gil -Izquierdo *et al,* 2001)**. Natural products are a promising source for the discovery of new pharmaceuticals. Several works dealing with propolis, which is a resinous hive product collected from plants by honey bees, showing a very complex chemical composition **(Bankova *et al,* 2000)**. Propolis has been used in folk medicine since

ancient times due to its many biological properties, such as antibacterial **(Sforcin *et al,* 2000),** antitumor **(Bazo *et al,* 2002)** and immunomodulatory effects **(Murad *et al,* 2002).** It contains more than 300 biochemical constituents **(Khalil 2006)**. Flavonoids and various phenolics are the most important pharmacologically active constituents in propolis capable of scavenging free radicals and thereby protect lipids from being oxidized or destroyed during oxidative damage **(Nieva Moreno *et al,* 2000).** Propolis has gained popularity and used extensively in healthy drinks and foods to improve health and prevent diseases such as inflammation, heart disease, diabetes and even cancer **(Matsushige *et al,* 1995)**.

Therefore the present work aims to estimate the impairments in mitochondrial bioenergetic function and oxidative stress in the development of liver damage during LC toxication in rats and possible protective role of propolis.

2. Materials and Methods

2.1. Chemicals

Lambda cyhalothrin (Katron 5% EC) [(*R*)-α-cyano-3-phenoxybenzyl (1*S*) - *c i s*-3- [(*Z*)-2-chloro-3,3,3-trifluoropropenyl]-2,2-dimethylcyclopropanecarboxylate and (*S*)-α-cyano-3-phenoxybenzyl (1*R*)-*cis*-3-[(*Z*)-2-chloro-3,3,3-trifluoropropenyl]-2,2 dimethyl cyclopropane carboxylate] was obtained from National Agricultural Chemicals and Investment (El-Watania Co). Propolis was obtained from Natural Factors Coquitlam, Canada. All other chemicals used were of analytical grade purchased from Sigma – Aldrich Co.

2.2. Animals

Male albino rats weighing 150 -160 g were obtained from the Animal House, Faculty of Agricultur, Alexandria University. Animals were housed in groups each of five rats in stainless steel cages and kept under laboratory conditions (25 ± °C, RH 65 –75% and light dark cycle, 12 h light / 12 h dark. Animals were given feed and water *ad libitum.* The animals were acclimatized to laboratory conditions for two weeks before starting the experiment. All procedures involving animals were performed in accordance with the guidelines of the standard procedures laid down by **OECD guideline (2008).**

2.3. Experimental design

Animals were divided into four groups and orally treated 3 doses /week for 4 weeks, then 2 weeks without treatment as a recovery period as the following:

Group I: Rats were administered with water and served as control.

Group II: Rats were administered with 100 mg / Kg bw propolis.

Group III: Rats were administered with 0.5 mg / kg bw LC (1/50 LD_{50}; 23.4 mg / Kg bw which estimated by Department of Mammalian Toxicology, Pesticide Central Laboratory, Agriculture Research Center).

Group IV: Rats were administered with 100 mg / Kg bw of propolis plus 0.5 mg / kg bw of LC. The body weights of control and treated animals were recorded weekly. At the end of the experiment the animals were sacrificed and then livers were removed and immediately washed with physiological saline (0.9 % NaCl), weighed individually and stored at - 20 ^{o}C till used.

2.4. Isolation of liver mitochondria

Rat liver mitochondria were isolated using a slightly modified protocol of **Krause *et al,* (2005).** Liver tissue was homogenized by a 2 ml tight fit glass Teflon homogenizer in buffer solution (250 mM sucrose, 50 mM Tris–HCL, 5 mM EDTA, and 0.5 mM Phenylmethanesulphonyl fluoride (PMSF), pH 7.4), 1:10 (w/v) and then centrifuged at 800g using Sigma 3K30 bench centrifuge for 10 min at 4 ^{o}C. The pellet was discarded and the supernatant was sucking and layered on top of 350 mM sucrose, 50 mM Tris–HCL, 5 mM EDTA, and 0.5 mM (PMSF), pH 7.4, 1:3 (w/v) and then centrifuged at 8,000g for 10 min at 4 ^{o}C. The top layer mitochondrial pellet was washed twice and resuspended in 250 mM sucrose containing 0.5 mM PMSF. Isolated mitochondria were stored at -20 ^{o}C until used.

2.5. Determination of protein

The protein content of the mitochondrial preparations was estimated by the method of **Lowry *et al,* (1951)** using bovine serum albumin as a standard.

2.6. Bioenergetic biomarkers

2.6.1. NADH dehydrogenase (Complex I) activity

NADH dehydrogenase activity was measured by the oxidation of NADH **(Galante and Hatefi, 1978).** Liver mitochondria (40 µg of protein / ml) were mixed with a mixture containing 40 mM phosphate buffer, pH 7.4, 0.1% Sod-Cholate, 1.5 mM potassium cyanide and 1.3 mM potassium ferricyanide and incubated for 1 min at 30 °C then 0.14 mM NADH was added. Decrease of absorption was followed spectrophotometercally at 340 nm for 1-3 minutes. Results were expressed as µmol NADH oxidized / min / mg protein. Sensitivity to rotenone (1µM) was tested in all mitochondrial suspensions.

2.6.2. Mitochondrial ATPase activity

The basic idea of this method is to measure the amount of inorganic phosphate produced from the hydrolytic reaction of ATP by the ATPase. Mitochondria suspension (1 mg protein / ml) were added to a medium containing, 20 mM Tris-HCl, pH 7.6, 5 mM $MgCl_2$ and 5 mM ATP, then the mixture was incubated for 5 min at 37°C in a shaking water bath. The reaction was stopped by the addition of 5% trichloroacetic acid (TCA) and then the inorganic phosphate (Pi) was measured spectrophotometrically at 740 nm according to **Taussky and Shorr (1953).** The concentration of Pi was calculated from a standard curve and the specific activity was expressed as µmole Pi / mg protein /

min. Sensitivity to oligomycin (1µg / ml) was tested in all mitochondrial suspensions.

2.7. Oxidative stress biomarkers

2.7.1. Superoxide Dismutase (SOD) activity

SOD as enzymatic antioxidant was measured spectrophotometrically at 25 °C by the method of **Marklund and Marklund (1974)**, with some modifications. The assay medium was 1.0 ml containing 50 mM Tris – HCl buffer (pH 8.0) and 0.24 mM pyrogallol. Autoxidation of pyrogallol was monitored at 420 nm for 3 min in the absence and presence of enzyme at three concentrations which produced between 30 to 60 % inhibition of pyrogallol. One unit of the enzyme activity is defined as the amount of enzyme which produced 50 % inhibition of pyrogallol autoxidation under the standard assay conditions. Mitochondrial SOD activities were expressed as Units / mg protein.

2.7.2. Reduced GSH content

GSH content as a nonenzymatic antioxidant was determined according to the method of **Beutler *et al* (1963).** The method based on the reduction of 5, 5` dithiobis (2-nitrobenzoic acid) (DTNB) with glutathione to produce a yellow colore measured at 405 nm, the reduced chromogen directly proportional to GSH concentration. The amount of GSH was expressed
as µmol / mg protein.

2.7.3. Lipid peroxidation (LPO) level

LPO was carried out following the procedure of **Ohkawa *et al,* (1979)** using thiobarbituric acid (TBA). Malondialdehyde (MDA) formed as end product of lipids peroxidation. MDA reacts with TBA to generate a colored product that absorbs at 532 nm. The level of lipid peroxidation was expressed as nmoles of MDA released / mg protein.

2.8. Mitochondrial proteins pattern, SDS - PAGE

Equal quantities of protein (42 µg) were loaded per lane and subjected to sodium dodecyl sulphate polyacrylamide gel electrophoresis (SDS - PAGE) (4 % stacking gel and 12 % separating gel) as described by **Laemmli (1970)**. To prepare the samples for electrophoresis, the liver mitochondria were dissolved in the sample buffer (1 ml of 0.5 M Tris-HCl, pH 6.8, 5 ml glycerol, 0.5 gm SDS, 0.5 ml b-mercaptoethanal and 5 mg of Bromophenol blue) and completed to 10 ml with distilled water then incubated at 95 ^{o}C for 4 min. After gel polymerization samples were loaded and the electrophoresis was performed using mini protein cell (Bio-Rad; 0.7 mm thick and 7 cm length at 80 volt with stacking gel and 150 volt with the separating gel for 2 hours. The protein bands were fixed in a solution containing 40 % methanol and 10 % acetic acid for 30 min. The gel was stained for 30 min with 0.1 % Coomassie blue R-250 in the fixative solution. It was destained with 10 % methanol and 7.5 % acetic acid to remove the background. Molecular weights of the different polypeptides on gels were determined as described by **Weber and Osborn (1969)** according to the following equation.

Mobility (RF) = Distance of protein migration / Distance of tracking dye migration

The mobility values of standard proteins were plotted versus the molecular weights on semi-log scaled paper.

2.9. Statistical analysis

Data obtained from the experiments were expressed as mean ±standard deviation (SD). Significant differences of measurement traits were analyzed using one-way analysis of variance (ANOVA) followed by the Student-Newman-Keuls Test. The criterion for statistical significance was set at $p \leq 0.05$. These tests were performed using a computer software CoStat program.

3. Results

3.1. Bioenergetic biomarkers

The *in vivo* adverse effects of repeated oral sublethal dose (0.5 mg / Kg bw) of LC on NADH dehydrogenase, ATPase, SOD activities and GSH and MDA contents in rat liver mitochondria were summarized in Tables (1and 2).

Table (1): NADH dehydrogenase and ATPase activities in rat liver mitochondria exposed to 0.5 mg/kg bw lambda cyhalothrin, 100 mg / kg bw propolis and their combination after six weeks

Groups	Specific activity of mitochondrial	
	NADH µmol NADH oxidized / min / mg protein	**ATPase** µmol Pi / min / mg protein
(I) Control	0.898±0.03^{c}	44.0±0.5^{a}
(II) Propolis	0.900±0.04^{c}	42.5±1.4^{a}
(III) Lambda cyhalothrin	0.434±0.04^{a}	18.5±0.7^{c}
(IV) Their combination	0.660±0.05^{b}	27.4±0.5^{b}

Values are expressed as means (5 rats) ± standard deviation (SD).
Values in column with different letters are significantly different at ($p \leq 0.05$).

3.1.1. NADH dehydrogenase activity:
The present study showed significant reduction in mitochondrial NADH dehydrogenase activities by 51.6 % whereas supplementation with propolis modulates the LC adverse effect to 26.5 % compare to control (Table 1).

3.1.2. ATPase activity:
ATPase activity of LC treated rat liver mitochondria significantly ($p < 0.05$) decreased (57.9 %), while co-administration of propolis attenuate the LC toxic effect to be 37.7 % of control (Table 1).

3.2. Oxidative stress biomarkers

3.2.1. SOD activity:
LC toxication condition (group III) was significantly decreased the activity of SOD to 60 %, whereas propolis supplementation attenuates the LC adverse effect to 24.4 % compared to control (Table 2).

3.2.2. Reduced GSH content:
The present data showed that LC toxication caused significantly ($P \leq 0.05$) decrease in the hepatic mitochondrial GSH content by 67.3 %, while the co-administration of propolis with LC relatively re-stored the GSH levels but still significant compared to the control (Table 2).

3.2.3. Lipid peroxidation (LPO) level:
Results pointed out significant elevation level of LPO in either mitochondria of rats treated with LC (93 %) or co-administration with propolis (20.7 %) compared to control (Table 2).

3.3.Mitochondrial protein pattern, SDS-PAGE

Fig. (1) illustrate the effects of lambda cyhalothrin on the hepato mitochondrial proteins pattern. It was found that the mobility of mitochondrial protein subunit in all lanes was similar to the mobility of control. Liver tissue from LC treated group showed increase in protein intensity relative to control. Also, it was found that protein band around 60 kDa corresponding to the LC mitochondrial subunit which is broader and considered indistinct compare to control patterns was noticed. Two minor bands around 30 and 14 kDa (head arrow), missed in other mitochondrial preparations were found in lane 4.

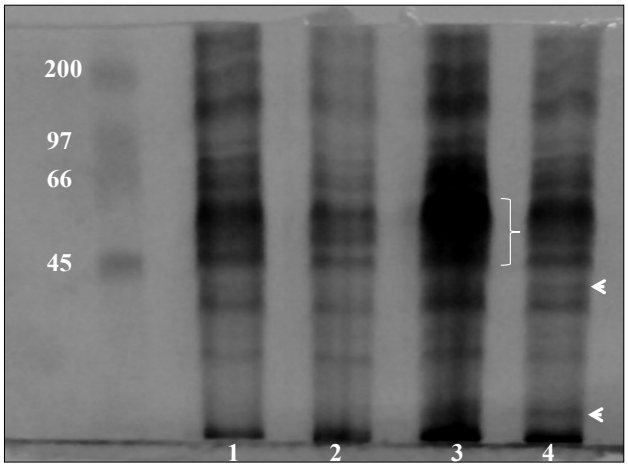

Fig (1): Effects of lambda cyhalothrin on the hepato mitochondrial proteins pattern.

The mitochondrial proteins, 42µg, were electrophoresed in 12% SDS polyacrylamide slab gels together with 10 µg prestained protein markers (Bio-Rad). Lane 1: control, lane 2: propolis, lane 3: LC toxication, shows a coalescence and indistinct bands around 60 kDa and lane 4: propolis supplementation with LC shows two minor bands around 30 and 14 kDa (head arrow), missed in other mitochondrial preparations.

4.Discussion

During the last years, new pesticides were developed and were found to interfering with mitochondrial electron transport, most of them with the proton-translocating NADH :ubiquinone oxidoreductase complex I (**Motoba *et al,* 1992, Hollingworth *et al,* 1994, Hollingworth and Ahammadsahib 1995).** Proton-translocating NADH :ubiquinone oxidoreductase is the first electron transport

Table (2): SOD activity, GSH content and LPO level in rat liver mitochondria exposed to 0.5 mg/kg bw lambda cyhalothrin, 100 mg / kg bw propolis and their combination after six weeks

Groups	SOD (U / mg Protein)	GSH (µmol / mg Protein)	LPO (nmol MDA / mg Protein)
(I) Control	45 ± 1.3^{c}	5.5 ± 0.02^{c}	2.9 ± 0.12^{a}
(II) Propolis	46 ± 1.1^{c}	5.1 ± 0.04^{c}	2.7 ± 0.14^{a}
(III) Lambda cyhalothrin	18 ± 0.9^{a}	1.8 ± 0.03^{a}	5.6 ± 0.08^{c}
(IV) Their combination	34 ± 0.5^{b}	3.9 ± 0.01^{b}	$3.\ 5\pm0.10^{b}$

Values are expressed as means (5 rats) ± standard deviation (SD).
Values in column with different letters are significantly different at ($p \leq 0.05$).

complex of the mitochondrial respiratory chain. It oxidizes NADH and transfers the electrons via a flavin mononucleotide cofactor and several iron–sulfur clusters to ubiquinone Q. 4 protons per 2 electrons stoichiometry is the widely accepted figure for the coupled vectorial proton translocation. So, complex I contribute to the protonmotive force that drives ATP synthesis. The energy- transducing function is maintained by the mitochondrial inner membrane and over 95 % of total cellular ATP is supplied by mitochondrial phosphorylation **(Weiss, 1988).** Therefore cellular activities can be adversely affected by damage to the mitochondrial energy-transducing functions **(Nishihara and Utsumi, 1986).** In the present study the reduction of mitochondrial NADH dehydrogenase by LC toxication may depend on the intricate structure of this enzymatic complex, which consists of at least 40 different polypeptides strongly embedded in the inner mitochondrial membrane. This unique feature explains the mitochondrion's great vulnerability to lipophilic molecules **(Degli Esposti 1998, Miyoshi 1998, Don and Hogg 2004).** Regarding potential toxicity, complex I is a secondary target of nitric oxide (NO) in general and of nitrogen radical species (RNS) in particular. **Gassner *et al,* (1997)** found that submicromolar concentrations of pyrethroid inhibit the respiratory complex I of rat liver mitochondria which in accordance with present study. This decrease in the activities of complex I in turn affected the activities of ATPase adversely. The inhibition of ATPase leads to reduce ATP production and generation of ROS. In line with other studies stated that some pesticides alter the Mg^{2+}-ATPase which may be a causative factor of cellular dysfunction **(Rahman *et al,* 2000 and Mehta *et al,* 2005).** Also this effect was observed in different cell models **(Vidau *et al,* 2009 and Vidau *et al,* 2011).** The reduction of ATPase activity can induce metabolic consequence, due to the acid-base properties of ATP, ADP, and inorganic phosphate the pH of the reaction medium was decreased **(Kraut *et al, 2014).*** Under LC intoxication, the reduction of ATPase activity can be attributed to lactic acidosis. Oxidative damage accumulates more in mitochondria than in the rest of the cells because electrons continually leak from the respiratory chain to form damaging ROS. This oxidative damage may modify mitochondrial proteins, DNA and lipids which may lead to mitochondrial bioenergetics failure leading to necrotic or apoptotic cell death **(Smith *et al,* 1999).** LC in certain doses and conditions is a potent inducer of oxidative stress **(Piner and Uner, 2012).** The SODs are enzymes that catalyze the conversion of $O_2^{\cdot-}$ to H_2O_2 and prevent the build-up of toxic $O_2^{\cdot-}$**.** The reduction in SOD activity observed in the present study agrees with the result of **(Wheeler *et al*, 2001)** who reported that, decrease of mitochondrial SOD by about 50% resulted in a functional decline of oxidative phosphorylation, an increase in oxidative stress and an increase in the rate of apoptosis. The data suggest that SOD plays an important role for balances of mitochondria redox status. Propolis supplementation attenuate the LC toxication, relating to its ability to scavenge the free radicals that cause lipid peroxidation **(Ferrali *et al,* 1997).** Glutathione (GSH) is the main non-protein thiol in cells whose functions are dependent on the redox-active thiol of its cysteine moiety that serves as a cofactor for a number of antioxidant and detoxifying enzymes. While synthesized exclusively in the cytosol from its constituent amino acids, GSH is distributed in different compartments, including mitochondria where its concentration in the matrix equals that of the cytosol. This feature and its negative charge at physiological pH imply the existence of specific carriers to import GSH from the cytosol to the mitochondrial matrix, where it plays a key role in defence against respiration-induced ROS and in the detoxification of lipid hydroperoxides and electrophiles. The present study demonstrated a significant reduction in the GSH content which may relate to either the inhibition of GSH synthesis or increased utilization of GSH for detoxification of toxicant induced free radicals. The decrease in mitochondrial GSH suggests that the oral administration of LC may lead to excessive free radical generation. These free radicals might be attacking the thiol group of cysteine residues and polyunsaturated fatty acids of biological membranes **(Raina *et al,* 2009)**. These findings run parallel with that previously reported by **Omotuyll *et al,* (2006)** who recorded a significant decrease in the GSH content after oral administration of cyfuthrin for 15 weeks. Also, **Raina *et al,* (2009)** recorded a significantly decline in blood glutathione after 30 days of cypermethrin dermal application. However **Kale *et al,* (1999)** recorded an increase in GSH content in erythrocyte after oral administration of a single dose of cypermethrin and or fenvalerate to rats. **Sharma *et al,* (2014)** showed that cypermethrin treated group (at the dose of 3.83 mg / kg bw for 7 days) showed elevation in lipid peroxidation and inhibition in glutathione in Wister rat brain. In addition, **Abbassy *et al,* (2014)** observed significant decrease in GSH after treatment of rats with lambda cyhalothrin in a dose equal 2.6 mg / kg bw, for 6 weeks (3 doses / week). Reduced mitochondria GSH content lead to H_2O_2 accumulation that can cause lipid peroxidation and cell injury **(Martin *et al*, 2000).** Oxygen free radical induced lipid peroxidation, which damaged to cell membranes and developed tissue injury **(Sener *et al,* 2005).** The present results pointed out high elevation of lipid peroxidation in rat liver mitochondria while propolis supplementation reduced the lipid peroxidation induced by LC. Propolis treatments related to scavenging free radicals, elevate GSH content in the cell and normalizing the membrane effects by protecting the lipid constituent of cell membrane of treated rats **(Abdel- Mobdy and Abdel-Rahim 2015).** The present data confirmed by study of the mitochondrial subunit composition using SDS-PAGE, which reveals conformational changes in mitochondrial protein pattern; coalescence and indistinct of the LC toxicated subunit bands indicates a protein with lot of hydrophobic region, suggestion the presence of LC. Change in the band intensity may be related to the level of enzymes (GST isomers appear usually between 24 to 30 kDa range) Also, may be due to the pesticide property and time elapse

after treatment. However, mitochondrial preparation of rats treated with propolis supplemented with LC is characterized by a distinct subunit bands and two minor bands of the kDa 30 and 14 molecular weight proteins, each one of them appears on the gel as one polymeric structure entailing that the protein retains its activity and showing the propolis hepatoprotective role.

Conclusion

The decrease in NADH dehydrogenase and ATPase activities and increased ROS suggest an overall perturbation of the electron transfer pattern leading to a mitochondrial dysfunction as a result of LC toxicity. Propolis attenuates the LC- induced liver injury by blocking the oxidative stress due to its high content of antioxidant ingredients. So the present work recommends the use of propolis as antioxidant food supplement that improves liver function

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الإخلال بالطاقة الحيوية والضغط التأكسدى للامبدا سيهالوثرين والدور الوقائى للبروبوليس فى ميتوكوندريا كبد الفئران

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الملخص العربى

تعتبر الميتوكوندريا مصدر الطاقة واستهلاك الأوكسجين فى الخلية وهدفا محتملا للتسمم بمبيد لامبدا سيهالوثرين (LC) ، النوع الثاني من مبيدات البيريثرويد الحشرية المخلقة والمستخدمة في كلا من المجالات الزراعية و الصحة العامة. و قد اجريت الدراسة الحالية بهدف تقدير تأثيرات التعرض لمبيد LC على سلسلة نقل الإلكترونات فى ميتوكوندريا كبد الفئران، حيث تم معاملة ذكور الفئران البيضاء بجرعات تحت مميتة عن طريق الفم (١/٥٠ من LD_{50} = ٠,٥ ملجم / كجم) لمدة أربعة أسابيع (٣ جرعات / أسبوع) وأسبوعين للملاحظة. تم تقييم اثنين من مؤشرات الطاقة الحيوية فى الميتوكوندريا، نيكوتينميد ادينين داى نيكلوتيد ديهيدروجينيز او NADH ديهيدروجينيز (المعقد الأول)، الأدينوزين ثلاثى الفوسفاتيز (ATPase) ومؤشرات الاكسدة الحيوية فى الميتوكوندريا، انزيم السوبر اوكسيد ديسميوتيز (SOD) ، مستوى الجلوتاثيون المختزل (GSH) ومستوى اكسدة الدهون (LPO) في كبد الفئران. وتم ايضا دراسة تأثير LC على المؤشرات الحيوية السابقة من خلال تحليل البروتين باستخدام تقنية الهجرة الكهربائية للكشف عن التغير الحادث فى تتابع وحدات بروتين ميتوكوندريا الكبد كنتيجة للتأثير السلبى لمبيد LC. وأخيرا تم دراسة الدور الوقائي للبروبوليس ضد التأثيرات السلبية لـ LC.

وأظهرت النتائج:

- انخفاض نشاط كلا من انزيم NADH ديهيدروجينيز، الأدينوزين ثلاثى الفوسفاتيز (ATPase)، السوبر اوكسيد ديسميوتيز (SOD) و مستوى الجلوتاثيون المختزل (GSH) ، بينما زاد مستوى اكسدة الدهون (LPO) بشكل ملحوظ فى ميتوكوندريا كبد الفئران المعاملة بالـ LC مقارنة بالمجموعة الضابطة.
- تمكن البروبوليس بالجرعة المستخدمة (١٠٠ ملجم / كجم) من تقليل الضرر الناتج عن المبيد على المؤشرات الحيوية المختبرة فى الحيوانات المعاملة بمبيد لامبداسيهالوثرين.
- أظهرت نتائج الهجرة الكهربائية تغيير في نمط تتابع وحدات بروتين ميتوكوندريا كبد الفئران المعاملة بمبيد لامبداسيهالوثرين.

اوضحت الدراسة أيضا أن التسمم باللامبداسيهالوثرين أضعف الطاقة الحيوية للميتوكوندريا وحث على الفسفرة التأكسدية مما قد يؤدى إلى خلل وظيفي في الكبد و أن البروبوليس بمحتوياته العالية من الفلافونويد ومركبات الفينول تمكن من تخفيض التأثير السلبي للامبداسيهالوثرين مما يدعم استخدامة كمكمل غذائى مضاد للأكسدة.