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CLOSE HOUSE	Effect of Furadan on Protein, Reduced Glutathione, Lipid Peroxidation And Ascorbic Acid Content of Muscle of Psammophilus Blanfordanus			
KEYWORDS	Psammophilus blanfordanus, furadan, muscle, protein, ascorbic acid, lipid peroxidation, reduced glutathione			
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ABSTRACT Psammophilus blanfordanus were divided into four groups as A, B, C and D. Eeach group comprising of five				

animals. Animals of group A (control) were given orally 3 µl of acetone/g body weight. The experimental animals of group B, C and D were administered orally with 3 µl of furadan (0.005gm of furadan dissolved in 1ml of acetone)/g body weight. The animals were sacrificed after different time intervals such as 0hr (group A), 24hr (group B), 48hr (group C) and 72hr (group D). The protein, reduced glutathione (GSH), lipid peroxidation (LPX) and ascorbic acid (ASA) content of muscle of the animal were measured and compared at different time intervals.

INTRODUCTION

India has achieved self-sufficiency in food production mainly due to introduction of modern agricultural practices, chemical fertilizers and pesticides (Kumar and Singh, 2012). Currently about 153 pesticides have been registered and 35 are widely used in India. Among them 33 pesticides have been banned based on their persistence and toxicity towards non targeted organisms (Shivakumar 2000). The pesticides can be circulated into different ecosystems by different agents (Weber et al., 1997) after entering in to the environment like water, air, soil and other agents (Farmer et al., 1972). Carbofuran (2, 3-dihydro-2, 2-dimethyl-7-benzofuranol methyl carbamate), which is an organocarbamate pesticide and commonly known as furadan, has broad spectrum of action and short half life in the environment. It is therefore widely used as an insecticide, nematicide and acaricide to protect the agricultural and industrial products (Gupta, 1994; Osten et al., 2005; Gera et al., 2011). The application of carbofuran is preferred over the organophosphates (OPs) and organochlorines (OCIs) due to its lower toxicity in comparison to OPs and OCIs (Agrawal and Sharma, 2010).

Lizards and other reptiles have been reported to be more sensitive to the effects of persistent insecticides than are birds and mammals. This apparent sensitivity may result from their low metabolic rate and resultant inability to quickly detoxify contaminants (Hall 1980). This study was designed to see the toxic effects of carbofuran on skin of Psammophilus blanfordanus by measuring GSH, LPX, Ascorbic acid content of muscle at different time intervals of (control) Oh and experimental (24h,48h and 72h).

MATERIALS AND METHODS

Psammophilus were caught locally from Baripada, Mayurbhanj, Odisha from the month of January 2012 to August 2012. The animals were divided into four groups as A, B, C and D. Eeach group comprising of five animals. Five animals of each group were kept in four different terrarium. They are acclimatized for 7 days in laboratory condition before the experiment. Animals of group A (control) were given orally 3 μ l of acetone/g body weight. The experimental animals of group B, C and D were administered orally with 3 μ l of furadan (0.005gm of furadan dissolved in 1ml of acetone)/g body weight. The animal after administered orally with acetone or furadan were separated into labeled, perforated plastic bottle. The animals of control group (0h) were sacrificed immediately, whereas the animals of experimental group B, C and D were sacrificed after 24hr, 48hr and 72hr of

treatment. Immediately muscle from hind limb was dissected out and kept at 0°C. The tissue homogenate was prepared with phosphate buffer (pH 7.4) and then centrifuged at 4000 rpm for 10 minutes in a cold centrifuge machine.

Measurement of protein content

Protein estimation of samples was made according to the method of Lowry et al. (1961). The data were expressed in mg/g tissue.

Measurement of Lipid Peroxidation

Lipid peroxidation of the sample was estimated as thiobarbituric acid reacting substance (TBARS) by thiobarbituric acid (TBA) according to the method of Ohkawa et. al. (1979). The data were expressed as nmoles of TBARS/mg protein.

Measurement of reduced glutathione (GSH)

Glutathione content (GSH) was estimated by the method of Ellman (1959) and the amount of glutathione is expressed as mg/g tissue.

Measurement of Ascorbic Acid (ASA)

Ascorbic acid of the sample were estimated by Jagota and Dani (1982) method.

All the solution were prepared by using Millipore distilled water. The above experiments were repeated for 5 times.

RESULTS AND DISCUSSION

Protein content

Protein content (mg/g tissue) in muscle tissue of Psammophilus blanfordanus treated with furadan (0.005mg/ ml of acetone) were 28.794±0.527 mg/g at 0 hr (control), 29.228±0.914 mg/g tissue at 24 hr, 38.014±1.01 mg/g tissue at 48 hr and 35.462±1.1 mg/g tissue at 72 hr. The protein content (mg/g tissue) of Psammophilus blanfordanus exposed to furadan was highest at 48hr, then decreased at 72 hr but higher than control. The protein content was lowest at 24 hr in comparison to control (Fig. 1).

The correlation analysis was used for the measurement of the linear association between variables. Pearson's correlation coefficients (r^2) among the analytical variables showed that the time interval is highly significant with protein content in muscle tissue of Psammophilus blanfordanus. [Time interval – protein content (0.797; P \leq 0.05)]. One way ANOVA showed that incubation period has significant effect on protein content of liver tissue. Post-hoc analysis revealed that only 48 hr

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is significant [F(3,8)=3.091, P<0.001)]. All the combinations are different from each other (P< 0.001, LSD).

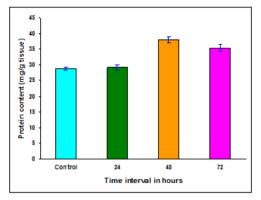


Fig. 1: Comparison of protein content in muscle (mg/g tissue) of Psammophilus treated with furadan

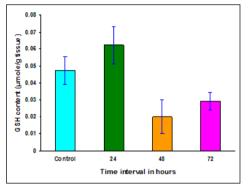


Fig.2: Comparison of GSH content in muscle μ mol/g tissue) of Psammophilus treated with furadan

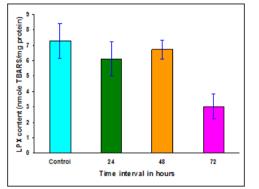


Fig.3: Comparison of LPX content in muscle (nmol TBARS/ mg protein) of Psammophilus treated with furadan

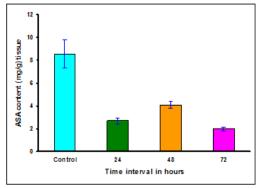


Fig.4:Comparison of ASA content in muscle (mg/ g tissue) tissue) of Psammophilus treated with furadan

GSH content

GSH content (µmol/g tissue) in muscle tissue of Psammophilus blanfordanus treated with furadan (0.005mg/ml of acetone) were0.0472 \pm 0.008 µmol/g tissue at 0 hr (control), 0.0622 \pm 0.0111 µmol/g tissue at 24 hr, 0.02 \pm 0.01 µmol/g tissue at 48 hr and 0.0292 \pm 0.0052 µmol/g tissue at 72 hr. The GSH content (µmol/g tissue) was highest at 24hr and lowest at 48 hrs but GSH content was lower at 72 hr in comparison to control (Fig. 2).

Pearson's correlation coefficients (r²) among the analytical variable showed that the time interval is significant with GSH content in muscles tissue of Psammophilus blanfordanus. [Time interval – GSH content (-0.604; P \leq 0.05)] One way ANOVA showed that incubation period has significant effect on GSH content of liver tissue. Post-hoc analysis revealed that 24 hr is significant [F(3,8) = 13.503, P< 0.001)]. All the combinations are different from each other (P< 0.001, LSD)

LPX content

LPX content (nmol TBARS/mg protein) in muscle tissue of Psammophilus blanfordanus treated with furadan (0.005mg/ml of acetone) were 7.286 \pm 1.143 nmol TBARS/mg protein at 0hr (control), 6.106 \pm 1.099 nmol TBARS/mg protein tissue at 24 hr, 6.726 \pm 0.595 nmol TBARS/mg protein tissue at 48 hr and 3.036 \pm 0.815 nmol TBARS/mg protein tissue at 72 hr. It was estimated that the concentration of LPX was found highest in 0 hr and decreases from 72 hrs to 48hrs to 24 hrs in muscle tissue of Psammophilus blanfordanu. (Fig. 3).

Pearson's correlation coefficients (r²) among the analytical variables are present. The time interval is highly significant with LPX content in muscles tissue of Psammophilus blanfordanus. [Time interval – LPX content (-0.748; P \leq 0.05)]. One way ANOVA showed that incubation period has significant effect on protein content of liver tissue. Post-hoc analysis revealed that only 48 hrs is significant [F(3,8)=12.228, P< 0.001)]. All the combinations are different from each other (P< 0.001, LSD).

Ascorbic acid

Ascorbic acid content (mg/g tissue) in muscle tissue of Psammophilus blanfordanus treated with furadan (0.005mg/ml of acetone) were 8.54 ± 1.22 mg/g at 0 hr (control), 2.696 ± 0.262 mg/g tissue at 24 hr, 4.1 ± 0.3 mg/g tissue at 48 hr and 1.984 ± 0.173 mg/g tissue at 72 hr. It was estimated that the concentration of Ascorbic acid was found highest in 72 hr and lowest in 48 hr. The ascorbic acid content at 0 hr was more than 24 hr in muscle tissue of Psammophilus blanfordanus. (Fig-4).

Pearson's correlation coefficients (r²) among the analytical variables are present. The time interval is significant with protein content in muscles tissue of Psammophilus blanfordanus. [Time interval – Ascorbic acid content (-0.790; P \leq 0.05)]. One way ANOVA showed that incubation period has significant effect on ascorbic acid content of muscle tissue. Posthoc analysis revealed that only 24 hrs is significant [F(3,8) = 62.936, P < 0.001)]. All the combinations are different from each other (P< 0.001, LSD).

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