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RESEARCH ARTICLE

IMPACT OF SAGO FACTORY EFFLUENT ON BIOCHEMICAL CONSTITUENTS IN DIFFERENT TISSUES OF MALE LARVAE OF DRAGONFLY *Bradinopyga geminata*

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Abstract

The antepenultimate larvae of dragonfly were treated in sublethal concentrations of sago factory effluent. Under effluent stress, biochemical constituents like total free amino acids, total proteins, total free sugars, glycogen content, reducing sugars, total lipids, triacylglycerol and cholesterol were found to decrease in the haemolymph, fat body and testis thus showing physio-metabolic dysfunction in the larvae. It is implied that the metabolic stress caused by the effluent could alter the reproductive process in the male dragonflies.

INTRODUCTION

The dragonfly larvae are the inhabitants of fresh waters and are predacious and economically very important group of insects. In recent times, the freshwater habitats are found to be severely affected by a variety of pollutants due to increased industrialization, urbanization, population explosion and agricultural activity throughout the world. In India, most of the industries are situated on the banks of watercourses which form the effective means of disposing the wastes. Among the industries, sago factory is one of the wet process industry and cause ecological threat by their wastes.

Insects require various biochemical substances for their normal development and growth (Chen, 1971). Many investigators have made studies on sex peptides, storage proteins, carbohydrates and lipids in various tissues of insect groups (Koopmanschap *et al.*, 1992 ; Nobuyoshi karagiri *et.al.*, 1998). Compared to the depth of data available on the biochemistry of female insects, much less is known in their counterparts that too with regard to the accessary reproductive glands. Attempts have also been made to show the adverse effects of many pollutants on biochemical constituents, fecundity, post-embryonic development and reproduction in a number of insects (Mandal and Chaudhuri, 1992 ; Raina *et al.*, 2001 ; Razdan, 2002 ; Rajathi ef *al.*, 2003). Thus the literature survey indicates that not much work have been made pertaining to the effect of industrial effluents on odonate larvae in relation to sexes. Therefore, the present work has been carried out to evaluate the toxicity of Sago Factory effluent on biochemical constituents in the tissue of the male larvae of dragonfly, *Bradinopyga geminata*.

Materials and Methods

The dragonfly larvae were manually collected from their natural habitat and were acclimatized to laboratory conditions. The male and female larvae were identified by following the method outlined by Ueda (1989) and larval instars by the method given by Dunkle (1985). The static bioassays were carried out by following the procedure given by Sprague (1973) using antepenultimate male larvae and the Sago Factory effluent collected

from a local industry. The LC50 96 hr value of the effluent to the test animals was found to be 6%. Various sub-lethal concentrations of the effluent namely 0.2%, 0.4%, 0.6%» and 0.8% were prepared. In each concentration, a group of 10 animals were introduced. As the antepenultimate larvae required 45 days to become final instars and the effluent-treated antepenultimate larvae showed no moulting, the experimental duration was entended upto 45 days with appropriate controls. Various biochemical constituents such as total free amino acids, total proteins, total free sugars, reducing sugars, glycogen content, total lipids, triacylglycerol and cholesterol were estimated in haemolymph, fat body ad testis using standard methods. The data were tabulated and statistically analysed.

Results and Discussion

From Table 1, it is clear that various biochemical constituents increase in the haemolymph, fat body and testis with advancement of larval development. This observation coincides with the findings of Subramanian (1993) and Paritha Bhanu (2003) in other odonate larvae. In insects, the main function of proteins is the **storage of amino** acids which are used in biosynthetic activities towardsmetamorphosis. The proteins are released into the haemolymph of insects as soon as they are synthesized in the fat body from where they are sequestered into the male organs. In male insects, the sugars serve as energy source for maintaining reproductive activity. The predominant lipids in the testis of insects are phospholipids with smaller triacylglycerol and cholesterol. According to Chen (1985), the secretion of male glands contain glyco-lipo-protein complex present in the fat body and haemolymph.

In the present investigation, the Sago Factory effluent is found to bring out an overall reduction of biochemical substances in the tissues of male larvae (Tables 2 - 4). The effluent stress might have adversly affected the biochemical machinary of the treated larvae with the resultant physico-metabolic dysfunction in them. This corraporates the findings of Claudia (1996) and Singh and Rao (2001) in several hexapods exposed to heavy metals and plant extracts. In general, proteins and lipids are transported from insect fat body to gonads via haemolymph. In male insects, the haemolymph, fat body and testis play a key role to provide precursors for the secretion of male glands (Rockstein, 1978; Basker and Ranganathan, 1987). The decrement of proteins and lipids as well as sugars in the testis of present experimental larvae could imply that the transport of substances within the tissue might have altered, thus arresting its development. This effect of Sago Factory effluent is found to be similar to the influence of apholate and insecticides which are found to inhibit the development of gonads and spermatogenesis in male insects (Cantwell and Henneberry, 1963; Shambu Kumar Paul *et al.,* 1991). Therefore, it is evident that the metabolic stress caused by the sago factory effluent could undoubtedly alter the reproductive processes in the male dragonflies.

Table 1. Biochemical constituents in various tissues of last three male larval instars of *B. geminata.* (Each value is the mean ± SD of 5 observations).

Biochemical constituents (mg)	Larval instars						
	Antepenultimate		Penultimate		Final i	nstars	
	Haemolymph	Fat body	Haemolymph	Fat body	Haemolymph	Fat body	Testis
Total free	353.13 ±31.40	5.91 ± 0.43	380.78 ± 32.67^{NS}	6.*21 ± 0.58^{NS}	384.72 ±33.15^{NS}	6.60 ± 0.38^{NS}	44.41 ± 0.32
amino acids			(+7.82)	(+5.08)	(+8.94)	(+11.68)	
Total proteins	178.55 ± 14.34	8.08 ± 0.67	183.48 ± 18.47^{NS}	9.47 ±0.76*	161.78 ± 14.76^{NS}	10.06 ±0.63*	5.24 ± 0.49
			(+2.76)	(+17.20)	(-9.39)	(+24.50)	
Total free	212.09 ± 17.49	2.07 ± 0.29	250.56 ±20.21*	2.36 ± 0.19^{NS}	272.27 ± 22.03*	2.66 ±0.14*	3.24 ± 0.29
sugars			(+18.13)	(+14.49)	(+28.37)	(+28.50)	
Glycogen	7.89 ± 0.67	11.83 ± 0.85	10.85 ±0.85*	12.82 ± 1.02^{NS}	11.83 ± 0.94*	13.81 ±0.86*	22.48 ± 1.55
content			(+37.52)	(+8.37)	(+49.94)	(+16.73)	
Non-reducing	55.24 ± 4.05	0.45 ± 0.04	69.05 ± 4.94*	0.50 ± 0.04^{NS}	64.12 ±4.06*	0.56 ± 0.03*	0.75 ± 0.05
sugars			(+25.00)	(+11.11)	(+16.08)	(+24.44)	
Total lipids	150.93 ± 12.98	3.74 ± 0.23	156.85 ± 13.87^{NS}	3.94 ± 0.28^{NS}	179.54 ± 12.83*	4.14±0.27^{NS}	2.9.8 ±0.14
			(+3.92)	(+5.35)	(+18.96)	(+10.70)	
Triacylglycerol	20.71 ± 1.88	0.69 ± 0.06	22.68 ± 1.88^{NS}	0.72 ± 0.07^{NS}	24.66 ± 1.85*	0.86 ± 0.03*	0.42 ± 0.03
			(+9.51)	(+4.34)	(+19.08)	(+24.63)	
Cholesterol	8.87 ± 0.45	0.20 ± 0.02	9.86 ± 0.94^{NS}	0.24 ± 0.02*	11.83 ±0.93*	0.29 ±0.01*	0.31 ± 0.02
			(+11.16)	(+20.00)	(+33.37)	(+45.00)	

* Significant at P< 0.05 ; ^{NS} - Not Significant

+ Indicates percent increase over antepenultimate larva - indicates percent decrease over antepenultimate larva

Table 2. Biochemical constituents in the haemolymph of control and effluent-treated male larvae of *B. geminata.* (Each value is the mean ± SD of 5 observations).

Biochemical constituents (mg/ 100ml)	Effluent Concentration (%)				
	Control	0.2	0.4	0.6	0.8
Total free amino acids	384.72 ± 33.15	329.14 ±23.58* (-14.45)	246.85 ±15.86* (-35.84)	176.21 ± 12.23* (-54.20)	120.31 ± 8.38* (-68.72)
Total proteins	161.78 ± 14.76	144.90 ±9.09* (-10.43)	128.80 ± 7.74* (-20.39)	119.14 ±8.80* (-26.36)	112.70 ±7.90* (+30.34)
Total free sugars	272.27 ±22.03	226.66 ± 14.99* (-16.75)	200.00 ±14.14* (-26.54)	124.66 ±8.69* (-54.21)	113.33 ±7.92* (-58.37)
Glycogen content	11.83 ± 0.94	10.89 ±0.65* (-7.95)	9.98 ± 0.62* (-15.64)	9.07 ± 0.59* (-23.33)	8.16 ±0.53* (-31.02)
Non-reducing sugars	64.12 ±4.06	50.28 ± 3.17* (-21.58)	41.14 ±2.62* (-35.83)	41.14 ±2.62* (-35.83)	32.00 ± 2.45* (-50.09)
Total lipids	179.54 ± 12.83	137.23 ± 9.62* (-23.56)	113.36 ±7.39* (-36.86)	107.40 ± 8.26* (-40.18)	59.66 ± 3.61* (-66.76)
Triacylglycerol	24.66 ± 1.85	18.35 ± 1.16* (-25.58)	17.64 ± 1.14* (-28.46)	15.52 ± 0.95* (-37.06)	14.11 ± 0.88* (-42.78)
Cholesterol	11.83 ± 0.93	8.34 ± 0.56* (-29.50)	7.58 ± 0.45* (-35.92)	5.68 ± 0.27* (-51.98)	4.93 ± 0.29* (-58.32)

* Significant at $P < 0.05$

\- Indicates percent decrease from control.

Table 3. Biochemical constituents in the fat body of control and effluent-treated male larvae of *B. geminata.* (Each value is the mean ± SD of 5 observations).

Biochemical constituents (mg / 100mg)	Effluent Concentration (%)				
	Control	0.2	0.4	0.6	0.8
Total free amino acids	6.60 ± 0.38	5.28 ± 0.37* (-20.00)	3.96 ± 0.20* (-40.00)	2.64 ± 0.13* (-60.00)	2.11 ± 0.15* (-68.03)
Total proteins	10.06 ± 0.63	7.80 ± 0.44* (-22.47)	6.20 ± 0.42* (-38.37)'	4.80 ±0.26* (-52.29)	4.80 ± 0.22* (-52.29)
Total free sugars	2.66 ± 0.14	2.45 ±0.13* (-7.89)	2.04 ± 0.14* (-23.31)	1.43 ±0.06* (-46.24)	1.43 ± 0.07* (-46.24)
Glycogen content	13.81 ±0.86	11.04 ±0.70* (-20.06)	9.38 ± 0.58* (-32.08)	8.28 ± 0.52* (-40.04)	7.45 ± 0.45* (-46.05)
Non-reducing sugars	0.56 ± 0.03	0.44 ± 0.02* (-21.42)	0.39 ± 0.02* (-30.35)	0.30 ± 0.02* (-46.42)	0.28 ± 0.01* (-50.00)
Total lipids	4.14 ± 0.27	2.85 ±0.15* (-31.15)	2.63 ± 0.15* (-36.47)	2.63 ± 0.15* (-36.47)	2.34 ± 0.13* (-42.47)
Triacylglycerol	0.86 ± 0.03	0.75 ± 0.05* (-12.79)	0.69 ± 0.05* (-19.77)	0.61 ±0.04* (-29.06)	0.56 ± 0.03* (-33.25)
Cholesterol	0.29 ±0.01	0.24 ± 0.01* (-17.24)	0.23 ± 0.01* (-20.68)	0.18 ± 0.01* (-37.93)	0.15 ± 0.01* (-48.27)

* Significant at P< 0.05

- Indicates percent decrease from control.

Table 4. Biochemical constituents in the testis of control and effluent-treated male larvae of *B. geminata.* (Each value is the mean ± SD of 5 observations).

Biochemical constituents (mg / 100mg)	Effluent Concentration (%)				
	Control	0.2	0.4	0.6	0.8
Total free amino acids	4.41 ±0.32	3.69 ± 0.19*	3.43 ± 0.19*	2.81 ±0.14*	2.70 ± 0.13*
		(-16.32)	(-22.36)	(-36.28)	(-38.77)
Total proteins	5.24 ± 0.49	4.87 ± 0.30*	4.69 ± 0.26*	4.33 ± 0.26*	4.06 ±0.26*
		(-7.06)	(-10.50)	(-17.37)	(-22.52)
Total free sugars	3.24 ± 0.29	2.59 ±0.15*	2.16 ± 0.13*	1.51 ±0.06*	1.08 ±0.08*
		i (-26.23)	(-33.33) «	(-53.39)	(-66.66)
Glycogen content	22.48 ± 1.55	19.48 ± 1.33*	17.98 ± 1.09*	14.98 ± 1.12*	13.48 ±0.84*
		(-13.35)	(-20.18)	(-33.36)	(-40.04)
Non-reducing sugars	0.75 ± 0.05	0.70 ± 0.05*	0.68 ± 0.04*	0.55 ± 0.03*	0.51 ±0.03*
		(-6.66)	(-9.33)	(-26.66)	(-32.00)
Total lipids	2.98 ±0.14	1.73 ± 0.04*	1t60 ± 0.07*	1.33 ±0.06*	1.33 ± 0.06*
		(-41.94)	(-46.30)	(-55.36)	(-55.36)
Triacylglycerol	0.42 ±0.03	0.36 ± 0.02*	0.34 ± 0.02*	0.33 ± 0.02*	0.30 ± 0.02*
		(-14.28)	(-19.04)	(-21.43)	(-28.57)
Cholesterol	0.31 ± 0.02	0.28 ± 0.01*	0.26 ± 0.01*	0.20 ±0.01*	0.19 ±0.01*
		(-9.67)	(-16.13)	(-35.48)	(-38.71)

* Significant at P< 0.05

- Indicates percent decrease from control.

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