CHAPTER – 10

INCREASED B-CELL REGENERATION AND PROTECTION AGAINST DIABETOGENIC INFLUENCE OF ALLOXAN DUE TO NEONATAL LUZINDOLE ADMINISTRATION.

INTRODUCTION:

Diabetes mellitus is a common metabolic disorder marked by hyperglycemia, altered metabolism of lipids, carbohydrates and proteins and vascular complications (Keen Tang Fui, 1982; Pickup and Williams, 2003). Chronic sustained hyperglycemia is an identifying feature of diabetes mellitus (Keen and Tang Fui, 1982; Ziv *et al.*, 1999). Insulin dependent diabetes mellitus (IDDM) now known as type I diabetes mellitus is characterized by B cell destruction leading to absolute insulin deficiency and ketosis. The other type of diabetes known as non insulin dependent diabetes mellitus (NIDDM), now known as type II diabetes mellitus is the most prevalent form of the disease which exhibits insulin resistance and even leading to insulin secretary defect (Keen and Tang Fui, 1982). Type II diabetes mellitus usually results from a combination of peripheral insulin resistance and impaired insulin secretion. Much evidence suggests that insulin resistance is influenced by different physiological and pathological

conditions such, as a high-fat diet, the dietary fatty acid composition (Storlien *et al.*, 1991), hypertriglyceridemia, obesity and diabetes (Borkman *et al.*, 1993; Vessby *et al.*, 1994). Hepatic insulin resistance correlates with intra cellular lipids (Ryysy *et al.*, 2000; Anderwald *et al.*, 2002) and plasma free fatty acid (Bergman *et al.*, 2000).

Animal models are commonly used to understand the molecular mechanisms and related features of diabetes. These are mainly genetic and spontaneous animal models of type II diabetes mellitus which have been used and which are highly heterogeneous. At one end of the spectrum there is mild hyperglycemia associated with obesity and hyperinsulinemia. At the other extreme animal models of type II diabetes mellitus can develop a severe form of diabetes with extensive B cell degeneration, occasionally resulting in ketosis and the requirement of exogenous insulin to sustain life (Pickup and Williams, 2003). Experimental animal models can be induced by chemical destruction or surgical removal of part or B cell mass. Streptozotocin exerts a selective toxic effect on B cell and induces diabetes mellitus in most laboratory animals (Lown *et al.*, 1979; Doux *et al.*, 1986). High doses of B cells toxins like streptozotocin and alloxan induce insulin deficiency in type I diabetes mellitus with ketosis. But doses employed to cause only partly destruction of B cell mass can be used to produce a mild insulin deficient state of type II diabetes mellitus without ketosis (Portha *et al.*, 1989). Alloxan and the product of its reduction, diluaric acid, establish a redox cycle with the formation of superoxide radicals, these radicals undergo dismutation to hydrogen peroxide thereafter

highly reactive hydroxyl radicals are formed. The action of reactive oxygen species with a simultaneous massive increase in cytosolic Ca^{++} ions causes rapid destruction of B cells (Szkuldelski, 2001).

Melatonin the pineal hormone has been shown to reduce the levels of triglycerides and cholesterols in mammalian species (Rasmussen *et al.*, 1999; Hoyos *et al.*, 2000; Nishida *et al.*, 2002). It has an inhibitory effect on the uptake of plasma fatty acids for lipogenesis as well as fasting induced lipolysis in the inguinal fat pad (Sauer *et al.*, 2001). It is also shown recently that administration of melatonin to rats with type II diabetes mellitus reduces these plasma hyperlipidemia, hyperinsulinemia and hjyperleptinemia and improves the conditions associated with type II diabetes (Nishida *et al.*, 2002). By contrast pinealectomy in the non-obese diabetic mouse resulted in an acceleration of development of type I diabetes mellitus (Conti and Maestroni, 1996). Further pinealectomized rats show increase in plasma and hepatic levels of cholesterol compared to pineal intact animals (Damian *et al.*, 1979). Pinealectomy may therefore worsen defects associated with type II diabetes mellitus. Studies intended to understand the role of neonatal melatonin on adult metabolic homeostasis, by using luzindole, a melatonin receptor antagonist has revealed age specific (i.e. weaning, pubertal, young and adult) effects have been revealed. Whereas melatonin receptor antagonism in the weaning period showed hyperinsulinemia, hyperglycemia, increased insulin sensitivity and glycogenic effect and decreased lipogenesis (Chapter 1,2,3), in the pubertal period it is marked by reduced insulin

sensitivity with decreased glycogenic effect and increased lipid and protein contents (Chapter 4,5,6). In this context it was thought interesting to test the effect of neonatal melatonin antagonism by luzindole treatment (nL) on weaning induced diabetes by alloxan treatment on carbohydrate, lipid and protein metabolic features and histoarchitecture of pancreas at the pubertal age of 45 days. Alloxan was used as streptozotocin sensitivity varies with species, strain, sex and nutritional status in activity (Okamato, 1981).

MATERIAL AND METHODS: See page numbers 18-38.

RESULTS:

Body and Organs weights: The body weight of all the alloxanised rats decreased significantly as compared to control and nLT rats. The relative weight of pancreas of LA(100) rats decreased significantly as compared to the age matched controls. The relative weight of liver of LA(100) and CA(100) rats decreased significantly as compared to LA(75) and age matched controls. The relative weight of spleen of LA(75) rats increased significantly as compared to all the other groups. The relative weight of kidney of LA(100) and LA(75) rats increased significantly as compared to CA(100) and age matched control rats. The relative weight of testes of LA(100) and LA(75) decreased significantly as compared to CA(100) and age matched controls. The relative weight of adrenals of all the alloxanised rats increased

significantly as compared to nLT rats of same age but showed no significant alteration as compared to age matched controls.

Serum glucose and insulin levels: The serum glucose level of LA(100), CA(100) and LA(75) rats decreased significantly as compared to control rats and nLT rats. While the, serum insulin levels of LA(100) and LA(75) rats increased significantly as compared to CA(100) and age matched controls. (Figure and Table; 10.16, 10.35).

Hepatic glycogen content and the activities of glycogen synthetase, glycogen phosphorylase and glucose-6-phosphatase: The hepatic glycogen content of LA(100) and LA(75) rats increased significantly as compared to the CA(100) rats but were still decreased as compared to age matched control rats. The hepatic glycogen synthetase and glycogen phosphorylase activities of all the alloxanised rats decreased significantly as compared to control rats while, the CA(100) rats showed significantly decreased glycogen synthetase activity in liver and significantly increased glycogen phosphorylase activity as compared to LA(100) and LA(75) rats. The glucose-6-phosphatase activity of LA(100) rats increased significantly as compared to the CA(100) and LA(75) rats as well as age matched controls (Figure and Table; 10.17, 10.19, 10.21).

Muscle glycogen content and the activities of glycogen synthetase and glycogen phosphorylase: The glycogen content of all the alloxanised rats increased significantly as compared to both nLT and age matched control. The muscle glycogen synthetase activity increased significantly in LA(100) and CA(100) rats as compared to the

LA(75) and age matched control rats. The muscle glycogen phosphorylase activity decreased significantly in all the alloxanised groups as compared to the nLT rats (Figure and Table; 10.15, 10.18).

`Hepatic and muscle protein content: The hepatic and muscle protein content of LA(100), CA(100) and LA(75) rats increased significantly as compared to control rats (Figure and Table; 10.22, 10.23).

Hepatic total lipid and cholesterol contents: The hepatic total lipid content of LA(75) rats increased significantly as compared to LA(100), CA(100) and age matched control and nLT rats. The hepatic cholesterol content of the alloxanised rats showed no significant alterations as compared to control and nLT rats.(Figure and Table; 10.24, 10.25).

Muscle total lipid and cholesterol contents: The muscle total lipid content of the LA(100) and CA(100) rats decreased significantly as compared to the LA(75) and age matched controls. The muscle cholesterol content of LA(100) rats increased significantly as compared to all other age matched groups (Figure and Table; 10.26, 10.27).

Adipose tissue total lipid and cholesterol contents: The adipose tissue total lipid content of LA(100) and LA(75) rats decreased significantly as compared to CA(100), nLT and control rats while, the adipose tissue cholesterol content of all the alloxanised rats decreased significantly as compared to nLT and age matched controls (Figure and Table; 10.28, 10.29).

Serum lipid fractions: The serum triglyceride, phospholipid and cholesterol levels of all the alloxanised groups decreased significantly as compared to control rats. However the serum total lipid level of LA(100) and LA(75) rats decreased significantly as compared to CA(100), nLT and control rats. The serum free fatty acid level of LA(100) and LA(75) rats decreased significantly as compared to CA(100) rats. (Figure and Table; 10.30, 10.31, 10.32, 10.33 and 10.34).

Histological observations: The histological sections of pancreas reveal increased B cell number and a higher B:A cell ratio with increased neogenesis of the islet cells in the nLT rats subjected to alloxanisation on the 22nd day. The sections of the nLT rats show distinct neogenesis of B cells from acinar cells by transdifferentiation. The islet cells are more compactly packed in the sections from the nLT rats (Plate; 11-14).

Figure 10.1: Body weight of pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

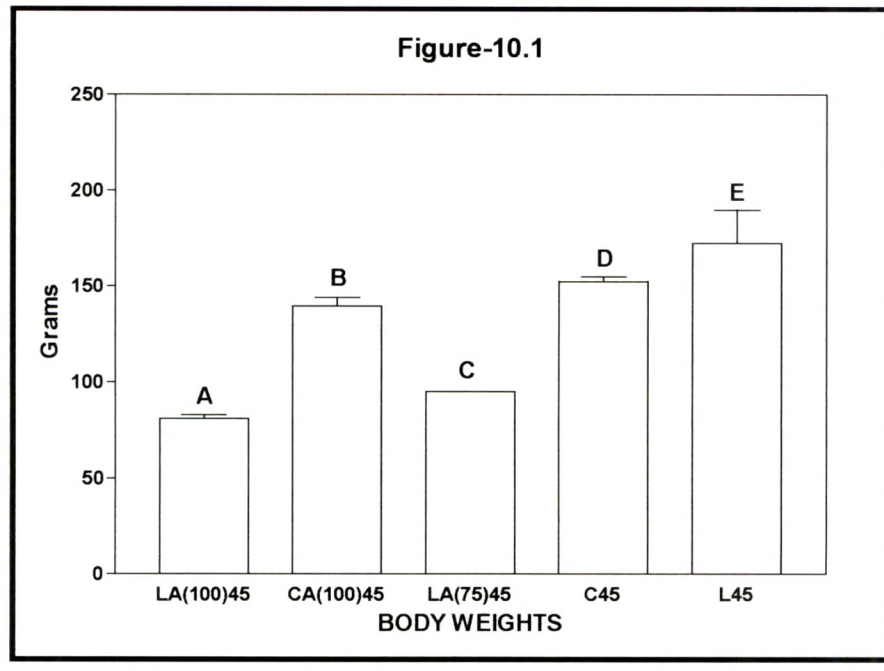


Table 10.1: Body weight of pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

	LA(100)(A)	CA(100)(B)	LA(75)(C)	C(D)	L(E)
BODY WEIGHT	81.00 ±2.00	139.50 ±4.51	95.20 ±0.10	152.50 ±2.5	172.50 ±17.54

Bonferroni's Multiple Comparison Test

	AvsB	AvsC	AvsD	AvsE	BvsC	BvsD	BvsE	CvsD	CvsE	DvsE
p	■	NS	◆	◆	●	NS	NS	■	◆	NS

Values are expressed as mean ± SEM, ◆p<0.001; ■P<0.01; ●P<0.05; NSNon Significant

Figure 10.2: Absolute pancreas weight of pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

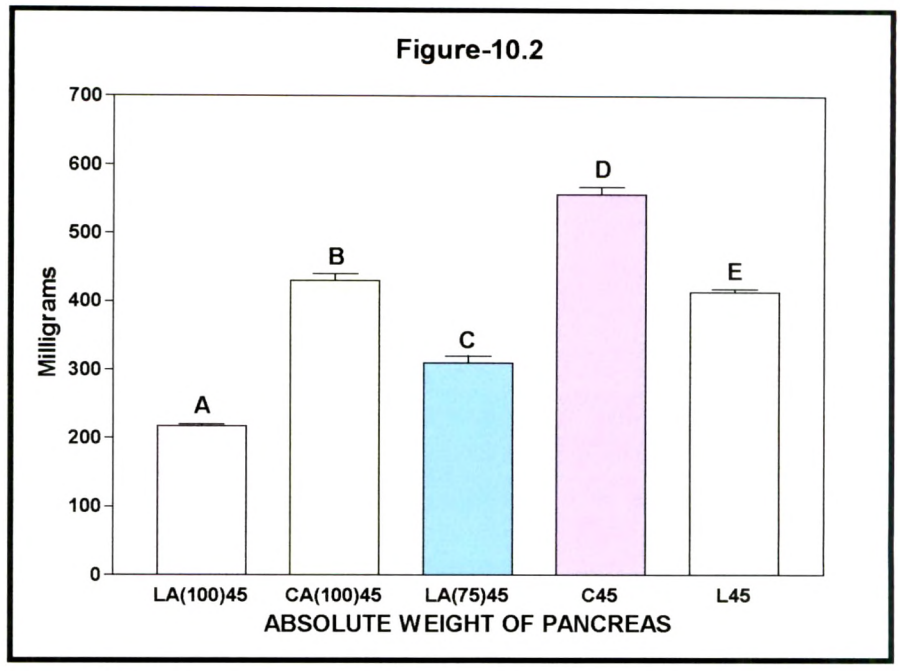


Table 10.2: Absolute pancreas weight of pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

	LA(100)(A)	CA(100)(B)	LA(75)(C)	C(D)	L(E)
PANCREAS	217.50 ±2.50	430.00 ±10.02	310.00 ±10.02	557.00 ±5.01	414.00 ±4.00

Bonferroni's Multiple Comparison Test

	AvsB	AvsC	AvsD	AvsE	BvsC	BvsD	BvsE	CvsD	CvsE	DvsE
p	◆	◆	◆	◆	◆	◆	◆	NS	◆	◆

Values are expressed as mean ± SEM, ◆p<0.001; NSNon Significant

Figure 10.3: Absolute liver weight of pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

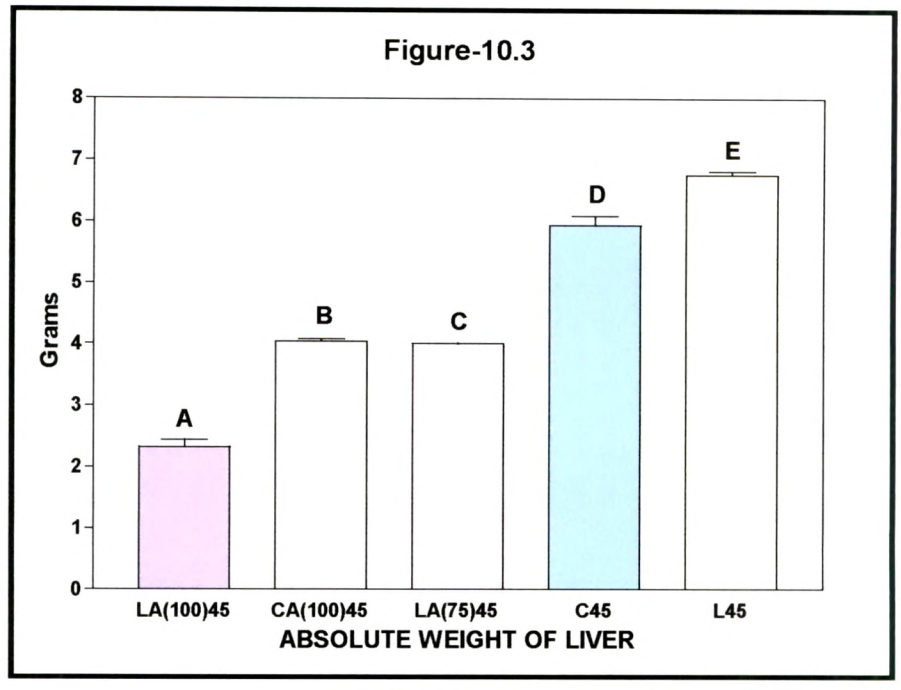


Table 10.3: Absolute liver weight of pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

	LA(100)(A)	CA(100)(B)	LA(75)(C)	C(D)	L(E)
LIVER	2.32 ±0.12	4.04 ±0.039	4.00 ±0.01	5.94 ±0.15	6.76 ±0.059

Bonferroni's Multiple Comparison Test

	AvsB	AvsC	AvsD	AvsE	BvsC	BvsD	BvsE	CvsD	CvsE	DvsE
p	◆	◆	◆	◆	NS	◆	◆	◆	◆	◆

Values are expressed as mean ± SEM, ◆p<0.001; NSNon Significant

Figure 10.4: Absolute spleen weight of pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

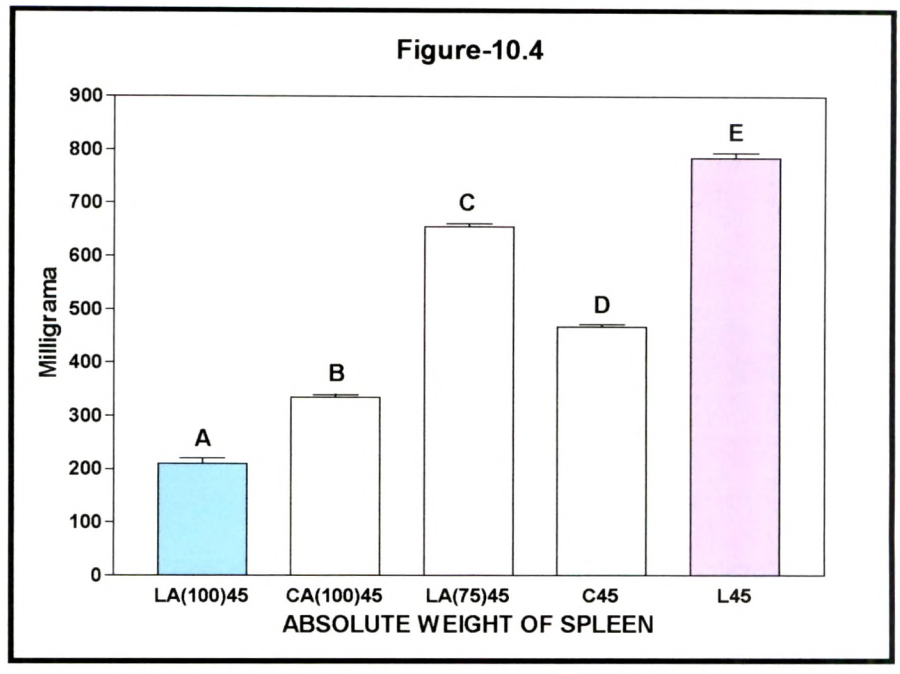


Table 10.4: Absolute spleen weight of pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

	LA(100)(A)	CA(100)(B)	LA(75)(C)	C(D)	L(E)
SPLEEN	210.00 ±10.02	335.00 ±5.01	565.00 ±5.01	467.5 ±4.51	785.5 ±8.52

Bonferroni's Multiple Comparison Test

	AvsB	AvsC	AvsD	AvsE	BvsC	BvsD	BvsE	CvsD	CvsE	DvsE
p	◆	◆	◆	◆	◆	◆	◆	◆	◆	◆

Values are expressed as mean ± SEM, ◆p<0.001;

Figure 10.5: Absolute kidney weight of pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

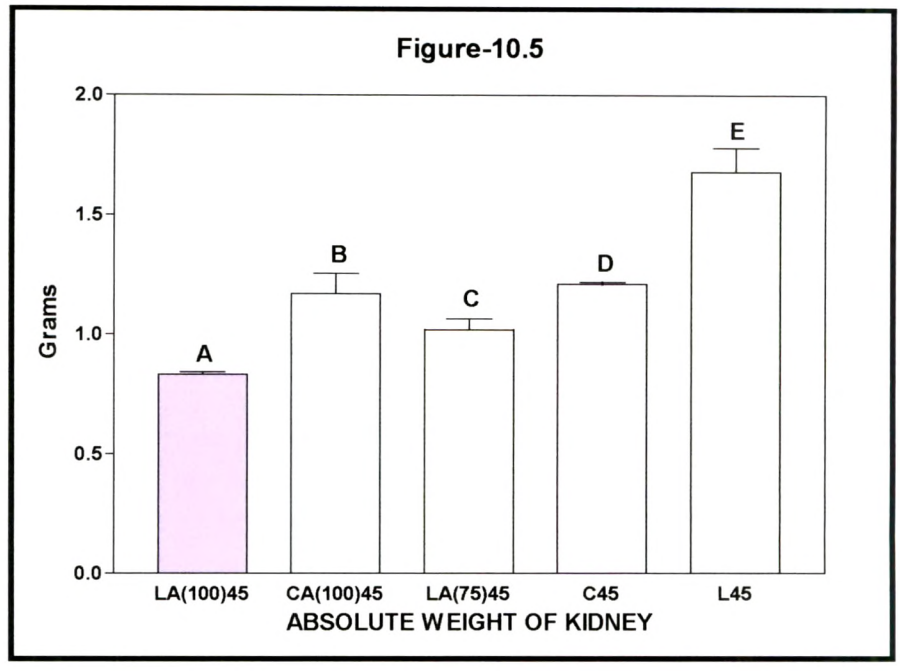


Table 10.5: Absolute kidney weight of pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

	LA(100)(A)	CA(100)(B)	LA(75)(C)	C(D)	L(E)
KIDNEY	0.83 ±0.01	1.17 ±0.085	1.02 ±0.044	1.21 ±0.0099	1.68 ±0.10

Bonferroni's Multiple Comparison Test

	AvsB	AvsC	AvsD	AvsE	BvsC	BvsD	BvsE	CvsD	CvsE	DvsE
p	●	NS	■	◆	NS	NS	◆	NS	◆	◆

Values are expressed as mean ± SEM, ◆p<0.001; ■P<0.01; ●P<0.05; NSNon Significant

Figure 10.6: Absolute testes weight of pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

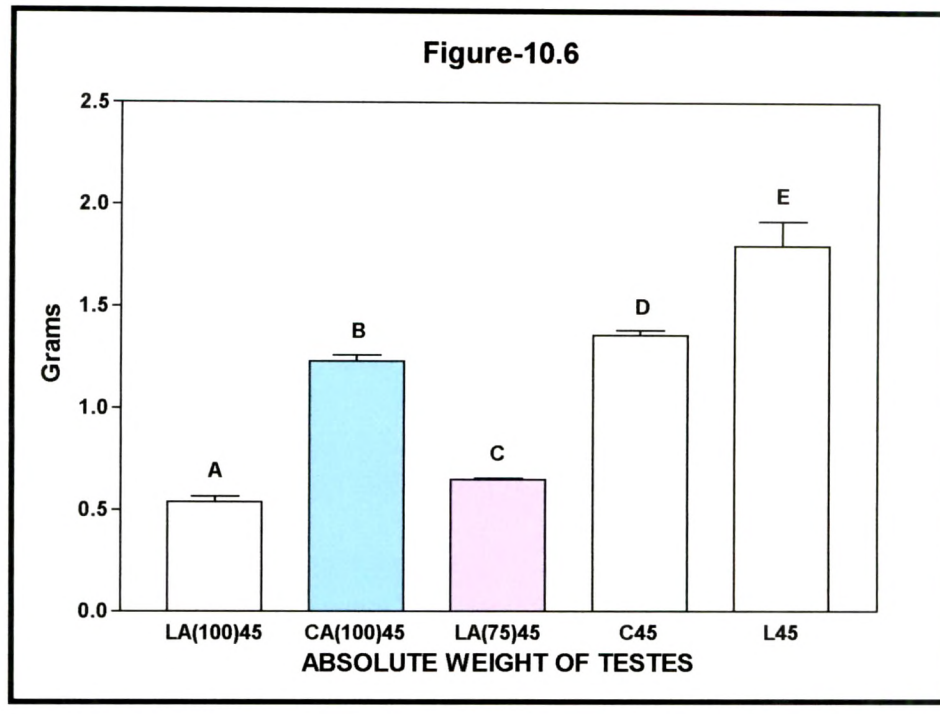


Table 10.6: Absolute testes weight of pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

	LA(100)(A)	CA(100)(B)	LA(75)(C)	C(D)	L(E)
TESTES	0.54 ±0.025	1.23 ±0.028	0.65 ±0.0075	1.36 ±0.024	1.80 ±0.12

Bonferroni's Multiple Comparison Test

	AvsB	AvsC	AvsD	AvsE	BvsC	BvsD	BvsE	CvsD	CvsE	DvsE
p	◆	NS	◆	◆	◆	NS	◆	◆	◆	◆

Values are expressed as mean ± SEM, ◆p<0.001; NSNon Significant

Figure 10.7: Absolute adrenal weight of pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

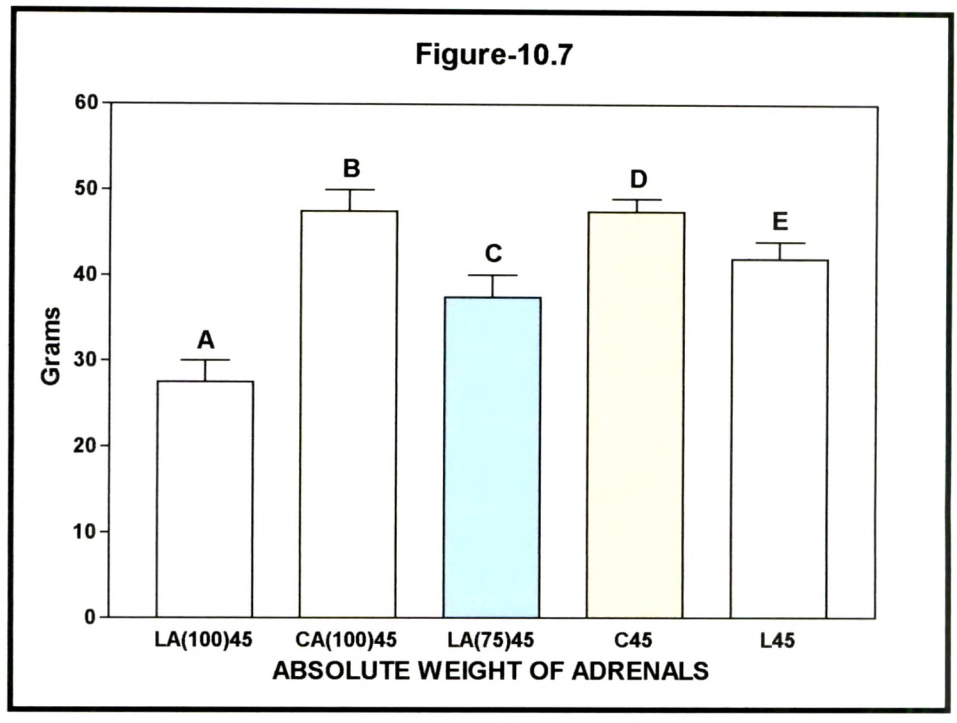


Table 10.7: Absolute adrenal weight of pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

	LA(100)(A)	CA(100)(B)	LA(75)(C)	C(D)	L(E)
ADRENAL	27.50 ±2.50	47.50 ±2.50	37.50 ±2.50	47.50 ±1.50	42.00 ±2.00

Bonferroni's Multiple Comparison Test

	AvsB	AvsC	AvsD	AvsE	BvsC	BvsD	BvsE	CvsD	CvsE	DvsE
p	◆	NS	◆	■	NS	NS	NS	NS	NS	NS

Values are expressed as mean ± SEM, ◆p<0.001; ■P<0.01;
NSNon Significant

Figure 10.8: Relative pancreas weight of pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

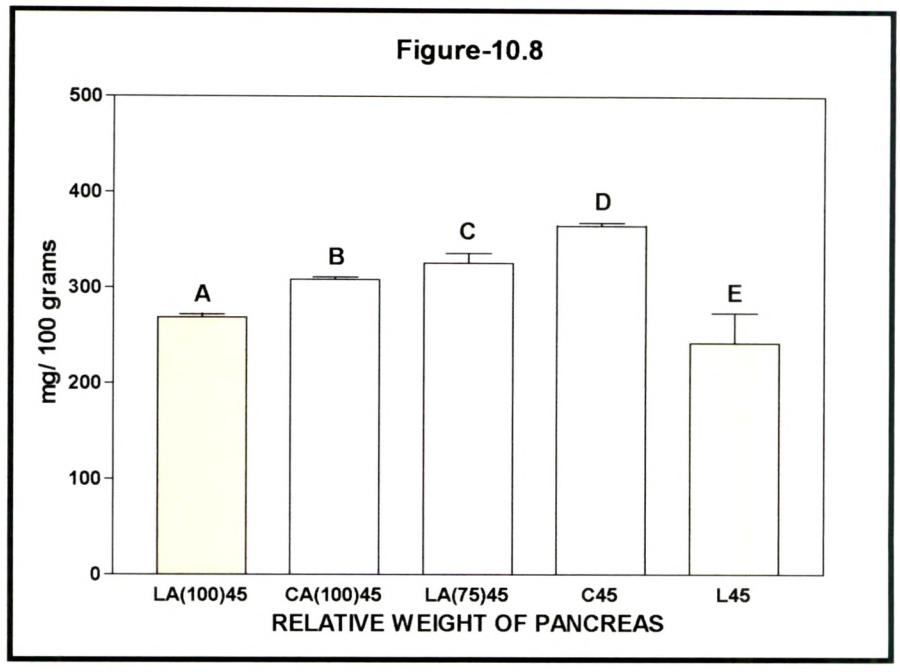


Table 10.8: Relative pancreas weight of pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

	LA(100)(A)	CA(100)(B)	LA(75)(C)	C(D)	L(E)
PANCREAS	268.60 ±3.55	308.33 ±2.78	325.62 ±10.53	365.29 ±2.71	242.25 ±22.31

Bonferroni's Multiple Comparison Test

	AvsB	AvsC	AvsD	AvsE	BvsC	BvsD	BvsE	CvsD	CvsE	DvsE
p	NS	NS	■	NS	NS	NS	NS	NS	●	◆

Values are expressed as mean ± SEM, ◆p<0.001; ■P<0.01; ●P<0.05; NSNon Significant

Figure 10.9: Relative liver weight of pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

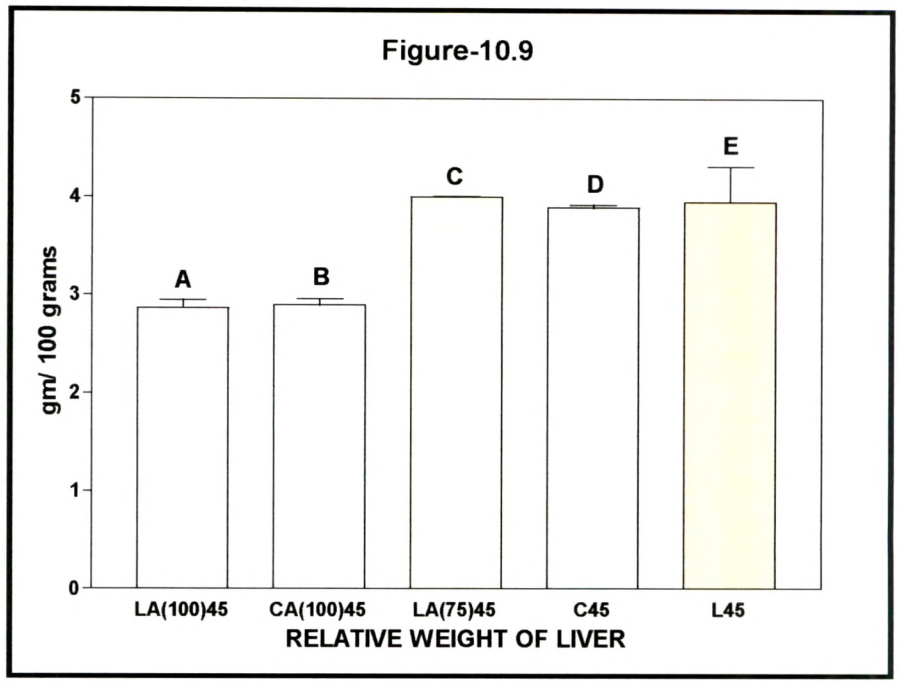


Table 10.9: Relative liver weight of pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

	LA(100)(A)	CA(100)(B)	LA(75)(C)	C(D)	L(E)
LIVER	2.86 ±0.085	2.89 ±0.064	4.00 ±0.004	3.89 ±0.03	3.95 ±0.33

Bonferroni's Multiple Comparison Test

	AvsB	AvsC	AvsD	AvsE	BvsC	BvsD	BvsE	CvsD	CvsE	DvsE
p	NS	■	■	■	■	■	■	NS	NS	NS

Values are expressed as mean ± SEM, ■P<0.01; NSNon Significant

Figure 10.10: Relative spleen weight of pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

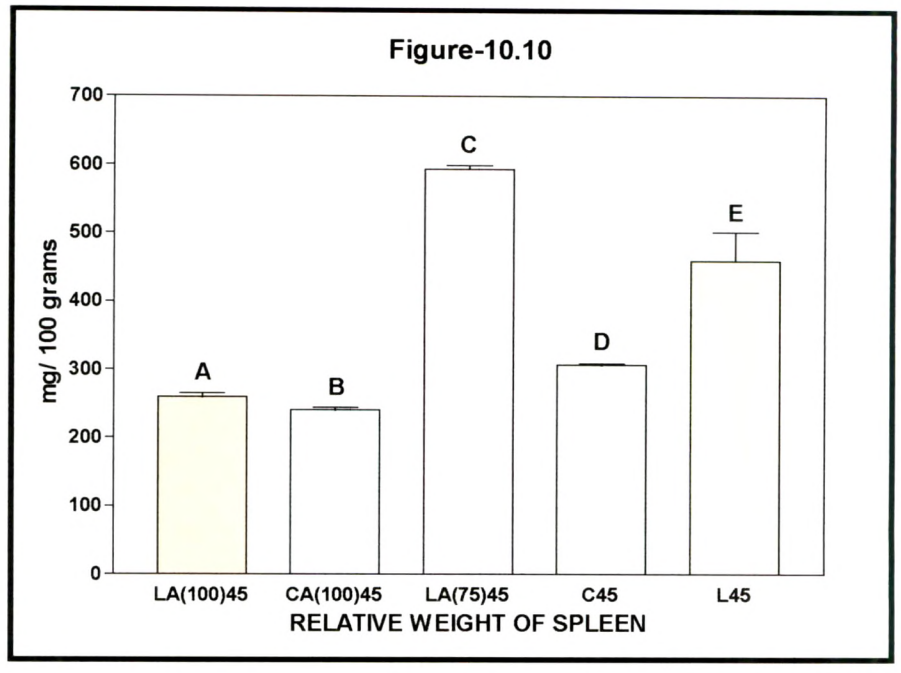


Table 10.10: Relative spleen weight of pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

	LA(100)(A)	CA(100)(B)	LA(75)(C)	C(D)	L(E)
SPLEEN	259.11 ±5.96	240.27 ±4.17	593.48 ±5.26	306.58 ±2.07	459.59 ±41.82

Bonferroni's Multiple Comparison Test

	AvsB	AvsC	AvsD	AvsE	BvsC	BvsD	BvsE	CvsD	CvsE	DvsE
p	NS	◆	NS	◆	◆	NS	◆	◆	■	◆

Values are expressed as mean ± SEM, ◆p<0.001; ■P<0.01; NSNon Significant

Figure 10.11: Relative kidney weight of pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

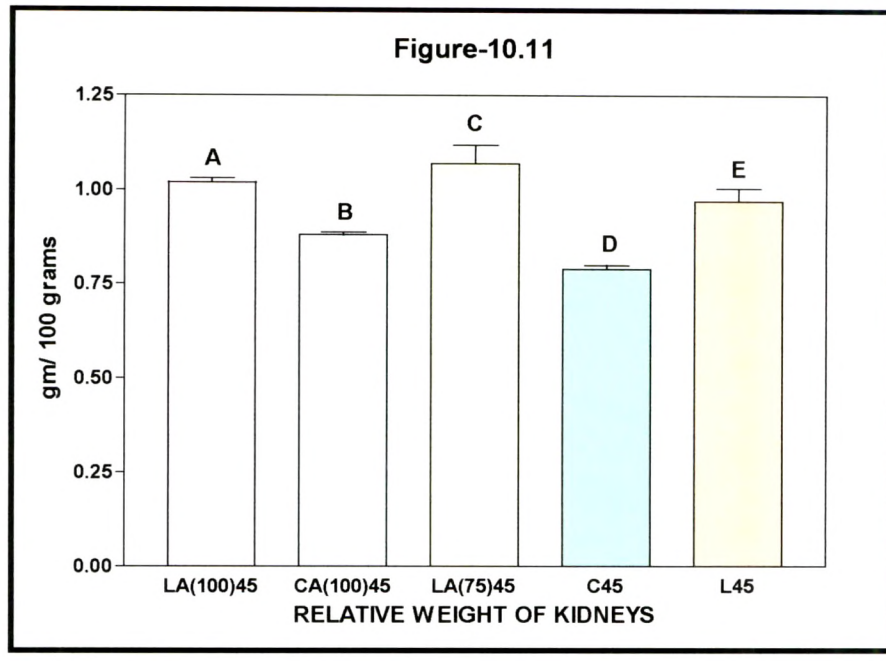


Table 10.11: Relative kidney weight of pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

	LA(100)(A)	CA(100)(B)	LA(75)(C)	C(D)	L(E)
KIDNEY	1.02 ±0.0099	0.88 ±0.0064	1.07 ±0.049	0.79 ±0.0099	0.97 ±0.034

Bonferroni's Multiple Comparison Test

	AvsB	AvsC	AvsD	AvsE	BvsC	BvsD	BvsE	CvsD	CvsE	DvsE
p	●	NS	◆	NS	■	NS	NS	◆	NS	■

Values are expressed as mean ± SEM, ◆p<0.001; ■P<0.01; ●P<0.05; NSNon Significant

Figure 10.12: Relative testes weight of pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

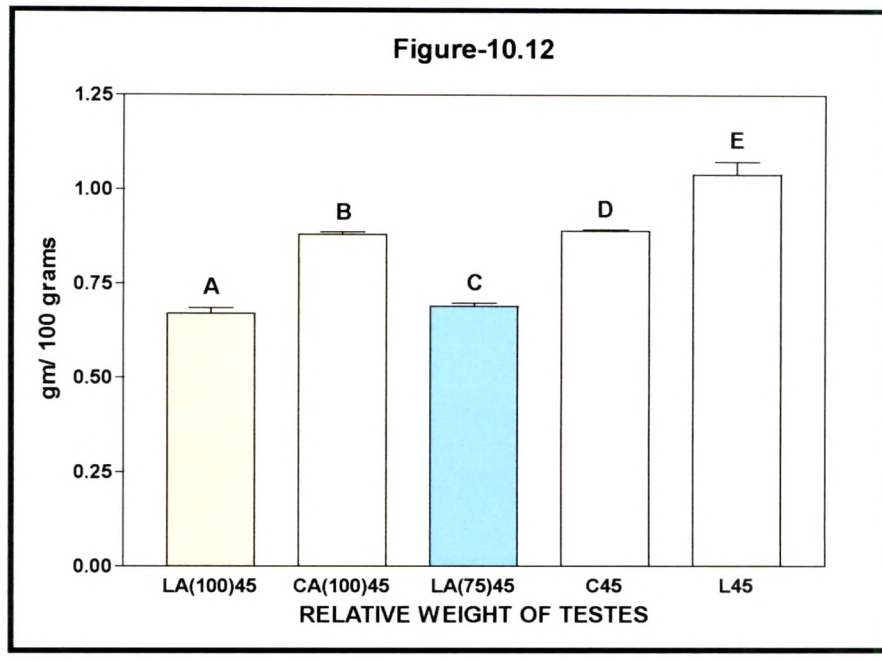


Table 10.12: Relative testes weight of pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

	LA(100) (A)	CA(100) (B)	LA(75) (C)	C (D)	L (E)
TESTES	0.67 ±0.014	0.88 ±0.0064	0.69 ±0.0078	0.89 ±0.002	1.04 ±0.034

Bonferroni's Multiple Comparison Test

	AvsB	AvsC	AvsD	AvsE	BvsC	BvsD	BvsE	CvsD	CvsE	DvsE
p	◆	NS	◆	◆	◆	NS	◆	◆	◆	◆

Values are expressed as mean ± SEM, ◆p<0.001; NSNon Significant

Figure 10.13: Relative adrenal weight of pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

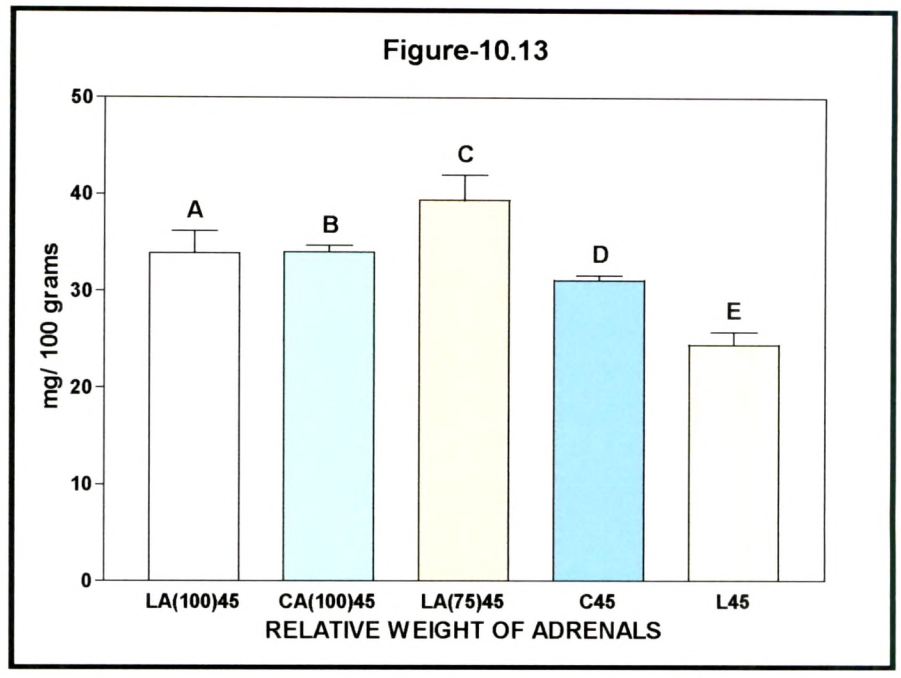


Table 10.13: Relative adrenal weight of pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

	LA(100) (A)	CA(100) (B)	LA(75) (C)	C (D)	L (E)
ADRENAL	33.89 ±2.25	34.02 ±0.69	39.37 ±2.63	31.13 ±0.47	24.47 ±1.32

Bonferroni's Multiple Comparison Test

	AvsB	AvsC	AvsD	AvsE	BvsC	BvsD	BvsE	CvsD	CvsE	DvsE
p	NS	NS	NS	●	NS	NS	●	●	◆	NS

Values are expressed as mean ± SEM, ◆p<0.001; •P<0.05;
NS Non Significant

Figure 10.14: Hepatic glycogen content of pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

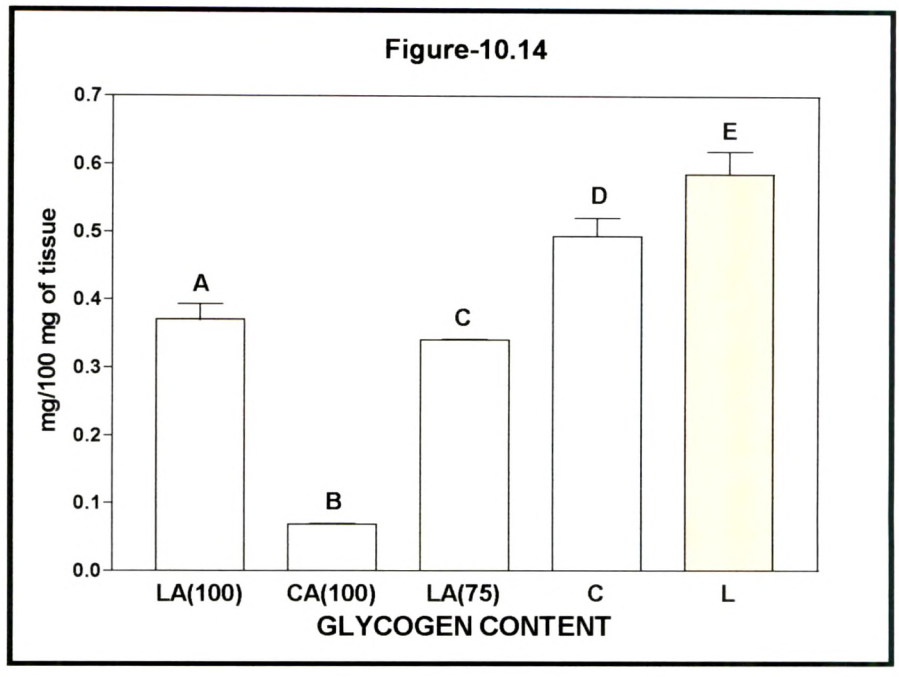


Table 10.14: Hepatic glycogen content of pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

	LA(100) (A)	CA(100) (B)	LA(75) (C)	C (D)	L (E)
GLYCOGEN	0.37 ±0.023	0.069 ±0.0007	0.34 ±0.0007	0.49 ±0.028	0.58 ±0.034

Bonferroni's Multiple Comparison Test

	AvsB	AvsC	AvsD	AvsE	BvsC	BvsD	BvsE	CvsD	CvsE	DvsE
p	NS	NS	NS	NS	NS	NS	•	NS	NS	NS

Values are expressed as mean ± SEM, •P<0.05; NSNon Significant

Figure 10.15: Muscle glycogen content of pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

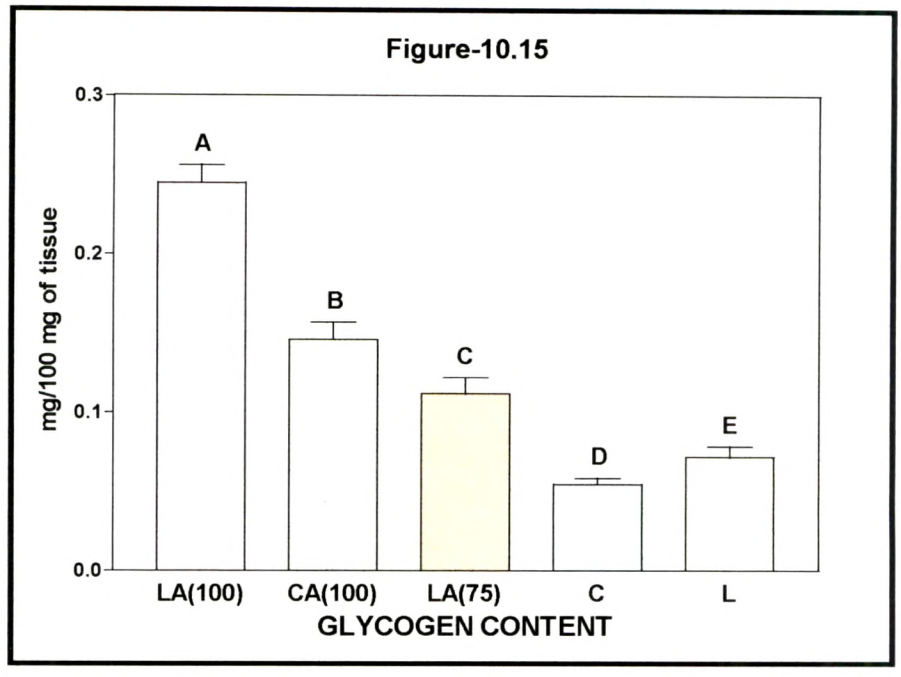


Table 10.15: Muscle glycogen content of pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

	LA(100) (A)	CA(100)(B)	LA(75)(C)	C(D)	L(E)
GLYCOGEN	0.25 ±0.011	0.15 ±0.011	0.11 ±0.010	0.055 ±0.0039	0.072 ±0.0069

Bonferroni's Multiple Comparison Test

	AvsB	AvsC	AvsD	AvsE	BvsC	BvsD	BvsE	CvsD	CvsE	DvsE
p	●	■	◆	◆	NS	●	NS	NS	NS	NS

Values are expressed as mean ± SEM, ◆p<0.001; ■P<0.01; ●P<0.05; NSNon Significant

Figure 10.16: Serum glucose level of pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

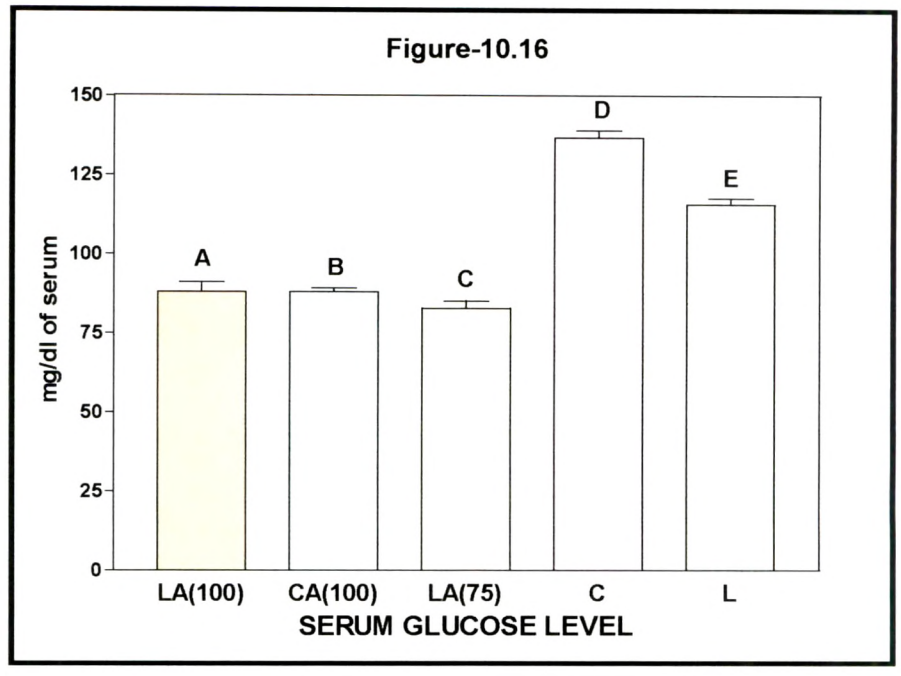


Table 10.16: Serum glucose level of pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

	LA(100) (A)	CA(100) (B)	LA(75) (C)	C (D)	L (E)
GLUCOSE	87.86 ±3.11	87.86 ±1.12	82.75 ±2.34	136.72 ±2.38	115.55 ±2.04

Bonferroni's Multiple Comparison Test

	AvsB	AvsC	AvsD	AvsE	BvsC	BvsD	BvsE	CvsD	CvsE	DvsE
p	NS	NS	◆	◆	NS	◆	◆	◆	◆	◆

Values are expressed as mean ± SEM, ◆p<0.001; NS Non Significant

Figure 10.17: Hepatic protein content of pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

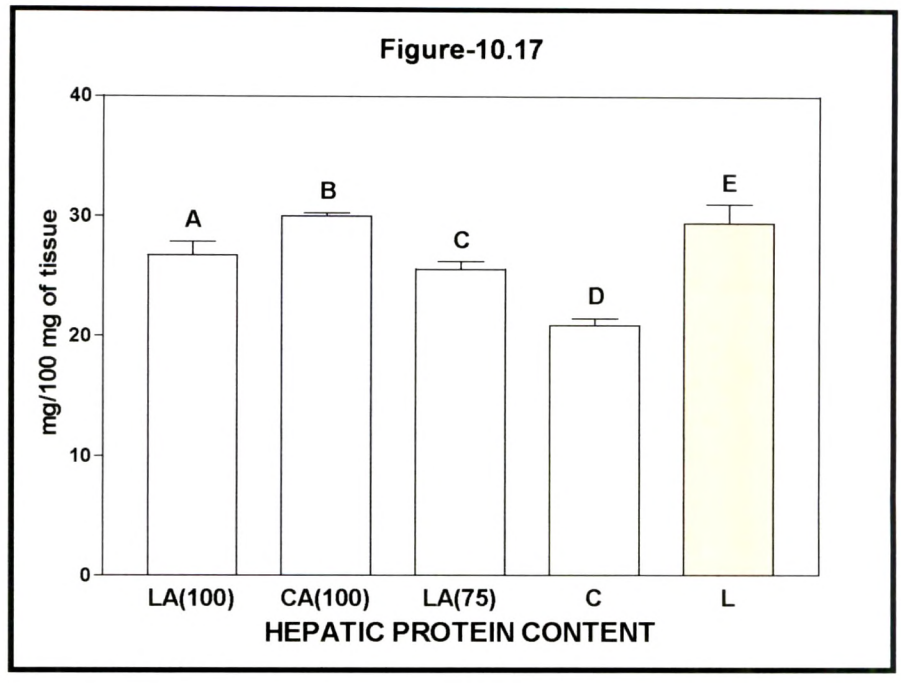


Table 10.17: Hepatic protein content of pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

	LA(100) (A)	CA(100) (B)	LA(75) (C)	C (D)	L (E)
PROTEIN	26.74 ±1.12	29.99 ±0.27	25.58 ±0.64	20.92 ±0.54	29.43 ±1.61

Bonferroni's Multiple Comparison Test

	AvsB	AvsC	AvsD	AvsE	BvsC	BvsD	BvsE	CvsD	CvsE	DvsE
p	NS	NS	■	NS	NS	◆	NS	●	NS	◆

Values are expressed as mean ± SEM, ◆p<0.001; ■P<0.01; ●P<0.05; NS Non Significant

Figure 10.18: Muscle protein content of pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

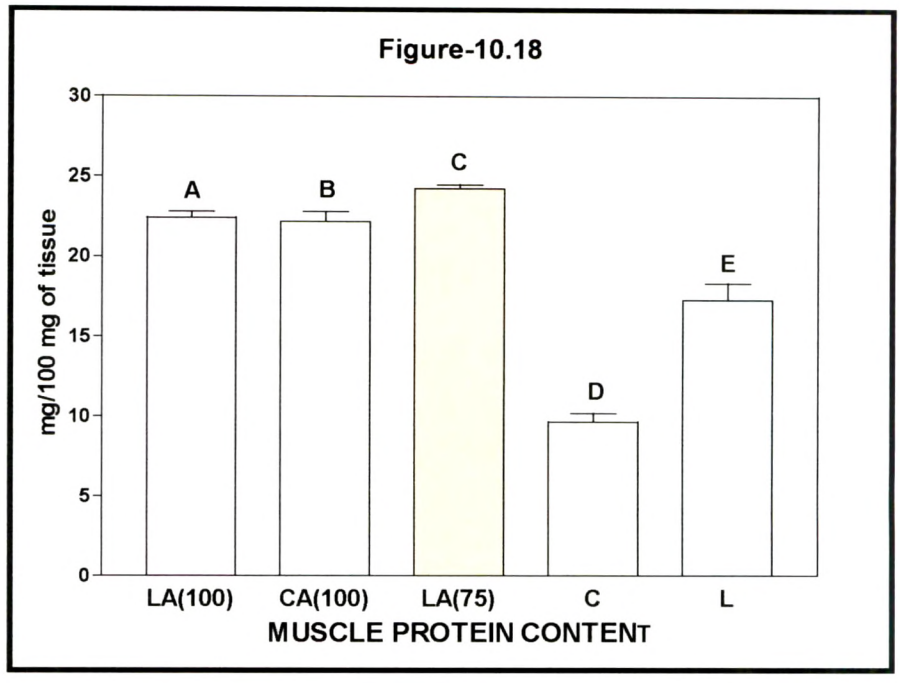


Table 10.18: Muscle protein content of pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

	LA(100) (A)	CA(100) (B)	LA(75) (C)	C (D)	L (E)
PROTEIN	22.41 ±0.37	22.16 ±0.62	24.24 ±0.24	9.66 ±0.54	17.29 ±1.06

Bonferroni's Multiple Comparison Test

	AvsB	AvsC	AvsD	AvsE	BvsC	BvsD	BvsE	CvsD	CvsE	DvsE
p	NS	NS	◆	◆	NS	◆	◆	◆	◆	◆

Values are expressed as mean ± SEM, ◆p<0.001; NSNon Significant

Figure 10.19: Hepatic glycogen synthetase activity in pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

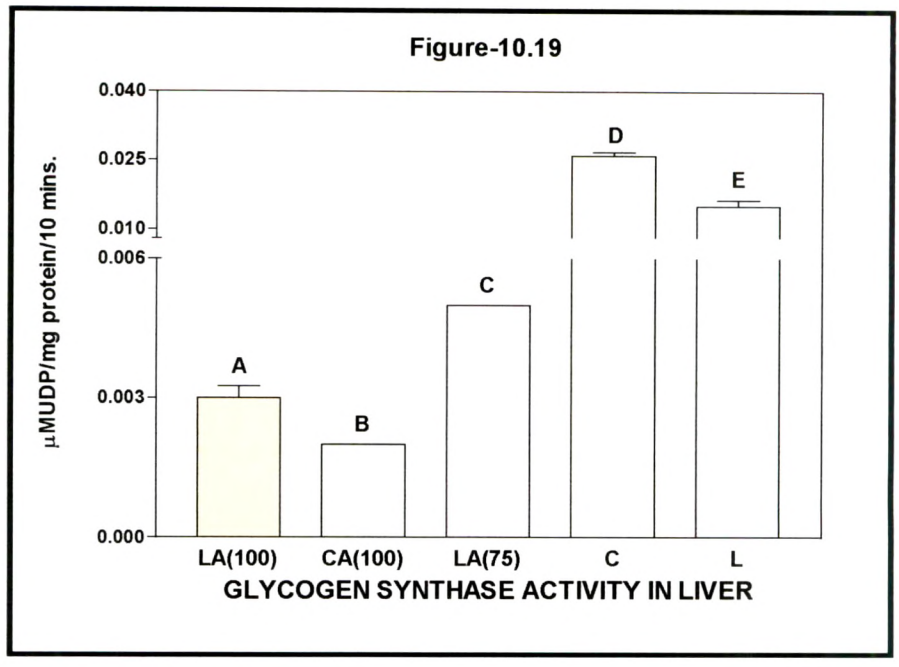


Table 10.19: Hepatic glycogen synthetase activity in pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

	LA(100)(A)	CA(100)(B)	LA(75)(C)	C(D)	L(E)
GLYCOGEN SYNTHETASE	0.003 ±0.00025	0.002 ±0.00	0.005 ±0.00	0.026 ±0.00075	0.015 ±0.0014

Bonferroni's Multiple Comparison Test

	AvsB	AvsC	AvsD	AvsE	BvsC	BvsD	BvsE	CvsD	CvsE	DvsE
p	NS	NS	◆	◆	NS	◆	◆	◆	◆	◆

Values are expressed as mean ± SEM, ◆p<0.001; NSNon Significant

Figure 10.20: Muscle glycogen synthetase activity in pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

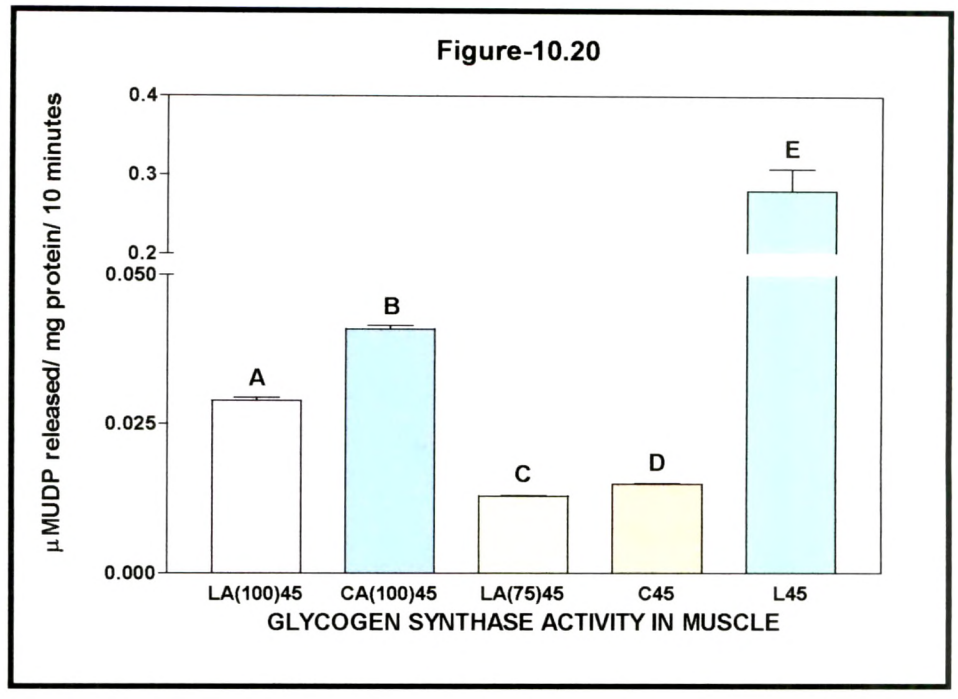


Table 10.20: Muscle glycogen synthetase activity in pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

	LA(100)(A)	CA(100)(B)	LA(75)(C)	C(D)	L(E)
GLYCOGEN SYNTHETASE	0.029 ±0.00047	0.041 ±0.0006	0.013 ±0.000025	0.015 ±0.00085	0.28 ±0.028

Bonferroni's Multiple Comparison Test

	AvsB	AvsC	AvsD	AvsE	BvsC	BvsD	BvsE	CvsD	CvsE	DvsE
p	NS	NS	NS	◆	NS	NS	◆	NS	◆	◆

Values are expressed as mean ± SEM, ◆p<0.001; NSNon Significant

Figure 10.21: Hepatic glycogen phosphorylase activity in pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

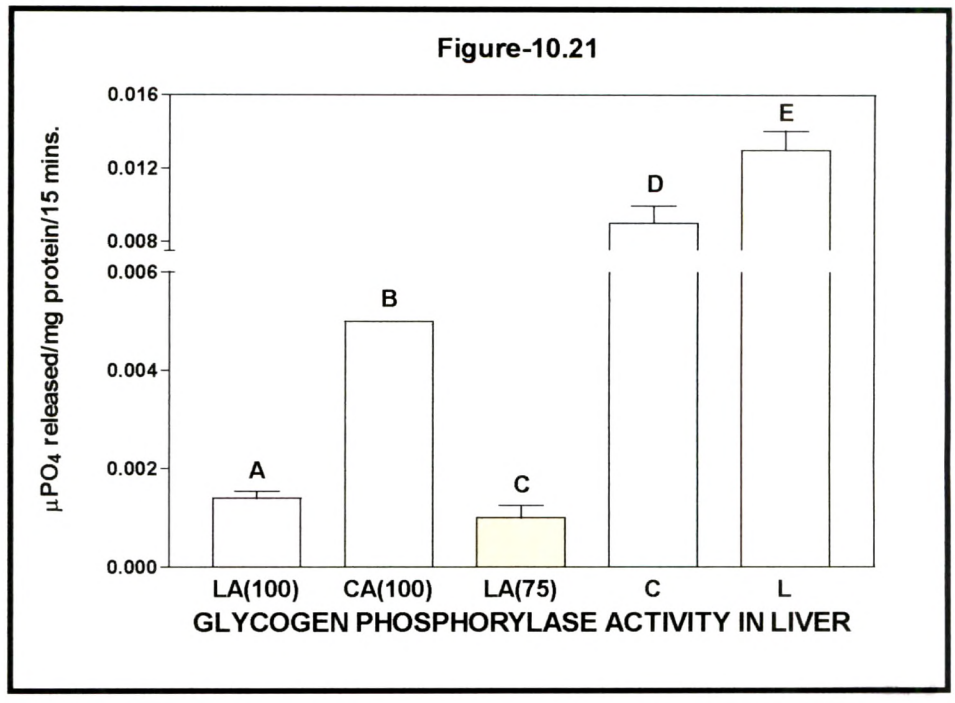


Table 10.21: Hepatic glycogen phosphorylase activity in pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

	LA(100)(A)	CA(100)(B)	LA(75)(C)	C(D)	L(E)
GLYCOGEN PHOSPHORYLASE	0.0014 ±0.00014	0.005 ±0.00	0.001 ±0.00025	0.009 ±0.00095	0.013 ±0.00105

Bonferroni's Multiple Comparison Test

	AvsB	AvsC	AvsD	AvsE	BvsC	BvsD	BvsE	CvsD	CvsE	DvsE
p	●	NS	◆	◆	■	■	◆	◆	◆	■

Values are expressed as mean ± SEM, ◆p<0.001; ■P<0.01; ●P<0.05; NSNon Significant

Figure 10.22: Muscle glycogen phosphorylase activity in pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

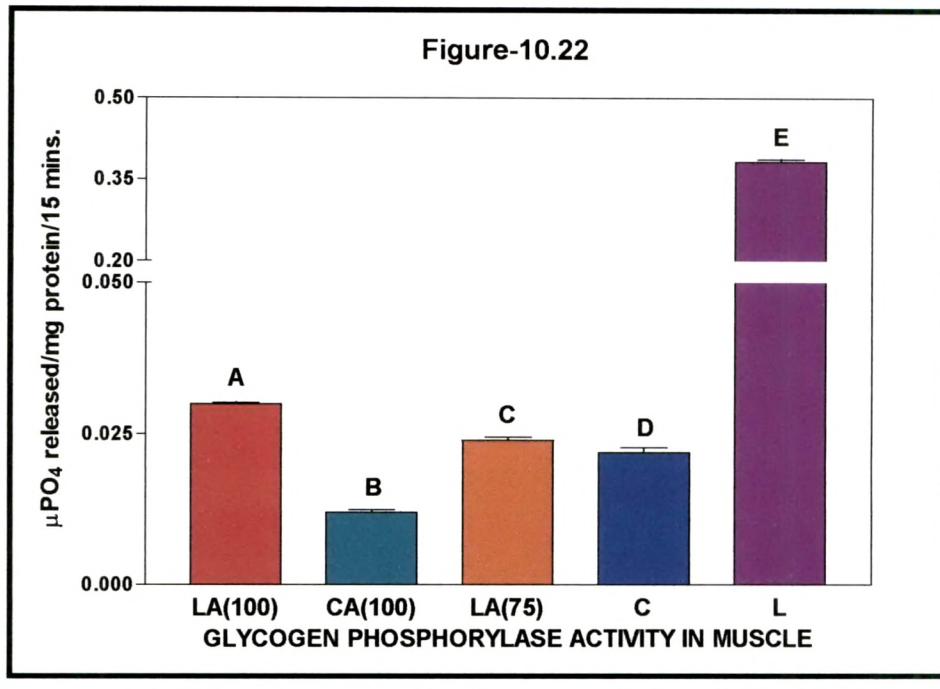


Table 10.22: Muscle glycogen phosphorylase activity in pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

	LA(100)(A)	CA(100)(B)	LA(75)(C)	C(D)	L(E)
GLYCOGEN PHOSPHORYLASE	0.03 ±0.00025	0.012 ±0.0004	0.024 ±0.000475	0.022 ±0.00075	0.383 ±0.0048

Bonferroni's Multiple Comparison Test

	AvsB	AvsC	AvsD	AvsE	BvsC	BvsD	BvsE	CvsD	CvsE	DvsE
p	◆	NS	NS	◆	●	NS	◆	NS	◆	■

Values are expressed as mean ± SEM, ◆p<0.001; ■P<0.01; ●P<0.05;
NS Non Significant

Figure 10.23: Hepatic glucose-6-phosphatase activity in pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

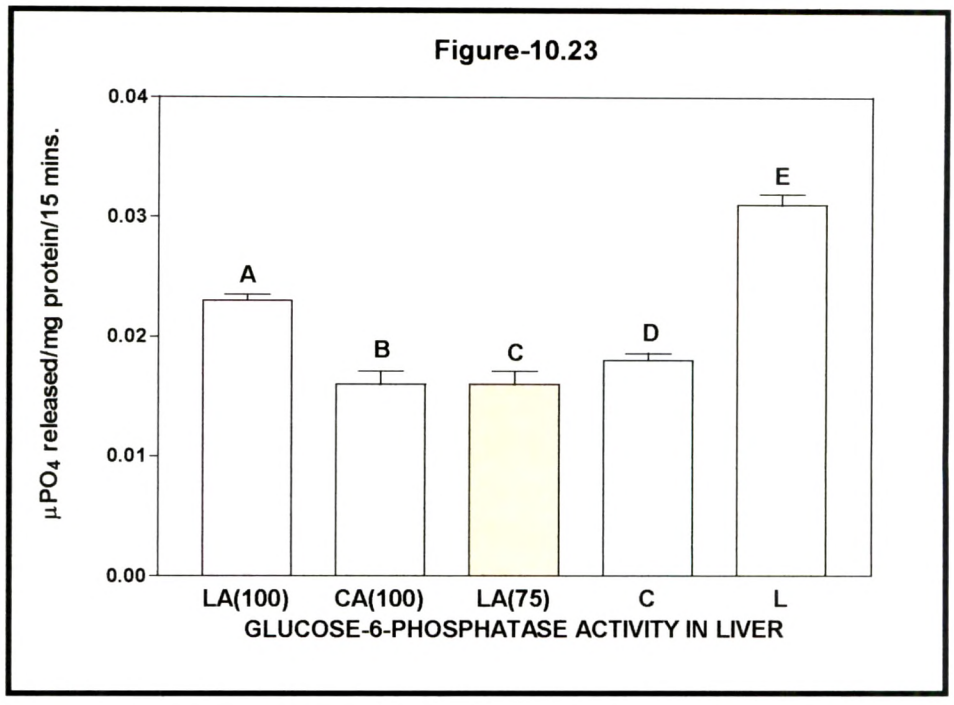


Table 10.23: Hepatic glucose-6-phosphatase activity in pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

	LA(100)(A)	CA(100)(B)	LA(75)(C)	C(D)	L(E)
GLUCOSE-6-PHOSPHATASE	0.023 ±0.000475	0.016 ±0.0011	0.016 ±0.0011	0.018 ±0.0006	0.031 ±0.0009

Bonferroni's Multiple Comparison Test

	AvsB	AvsC	AvsD	AvsE	BvsC	BvsD	BvsE	CvsD	CvsE	DvsE
p	◆	◆	■	◆	NS	NS	◆	NS	◆	◆

Values are expressed as mean ± SEM, ◆p<0.001; ■P<0.01;
NSNon Significant

Figure 10.24: Hepatic total lipid content of pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

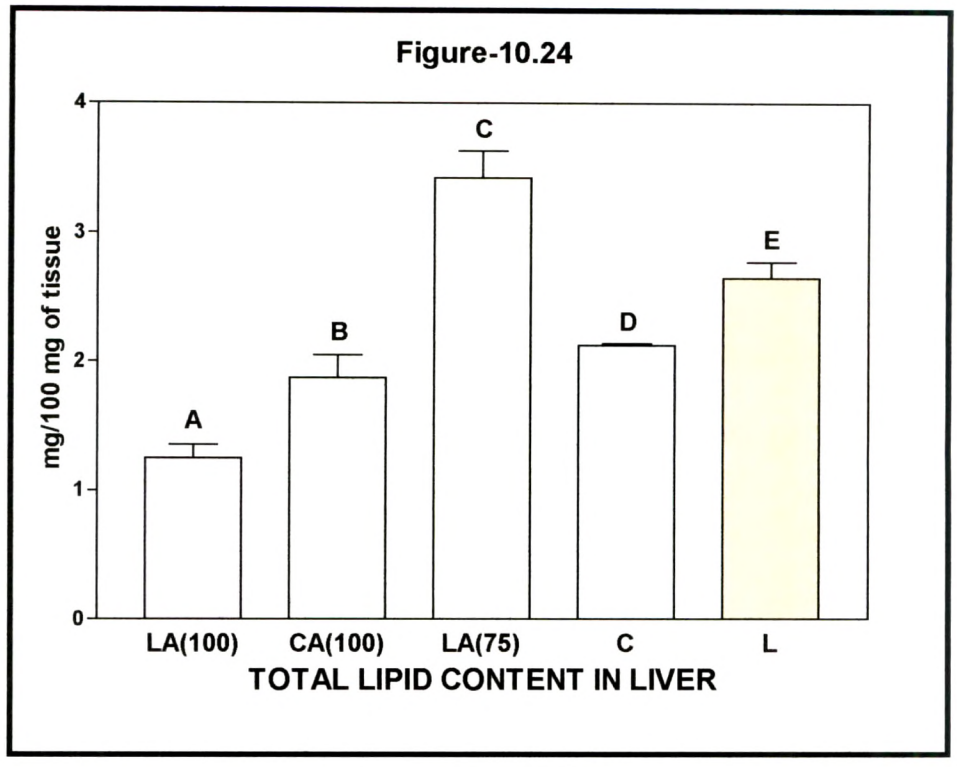


Table 10.24: Hepatic total lipid content of pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

	LA(100) (A)	CA(100) (B)	LA(75) (C)	C (D)	L (E)
TOTAL LIPIDS	1.25 ±0.104	1.875 ±0.175	3.425 ±0.2096	2.125 ±0.017	2.65 ±0.119

Bonferroni's Multiple Comparison Test

	AvsB	AvsC	AvsD	AvsE	BvsC	BvsD	BvsE	CvsD	CvsE	DvsE
p	NS	◆	■	◆	◆	NS	●	◆	●	NS

Values are expressed as mean ± SEM, ◆p<0.001; ■P<0.01; ●P<0.05; NSNon Significant

Figure 10.25: Hepatic cholesterol content of pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

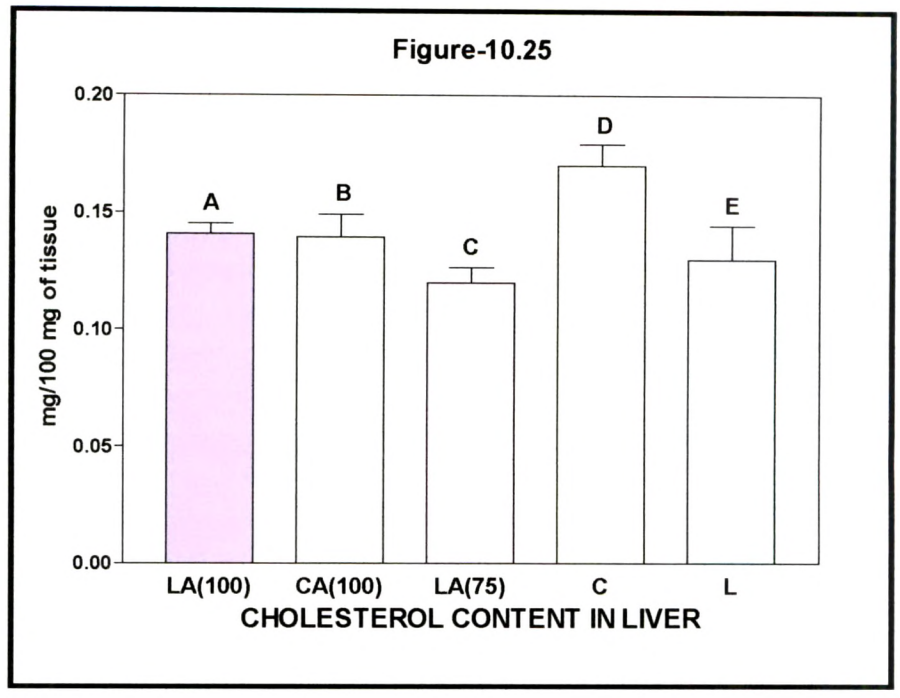


Table 10.25: Hepatic cholesterol content of pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

	LA(100)(A)	CA(100)(B)	LA(75)(C)	C(D)	L(E)
CHOLESTEROL	0.1407 ±0.004435	0.1395 ±0.00955	0.12 ±0.0066	0.17 ±0.0091	0.13 ±0.01435

Bonferroni's Multiple Comparison Test

	AvsB	AvsC	AvsD	AvsE	BvsC	BvsD	BvsE	CvsD	CvsE	DvsE
p	NS	NS	NS	NS	NS	NS	NS	•	NS	NS

Values are expressed as mean ± SEM, •P<0.05; NSNon Significant

Figure 10.26: Muscle total lipid content of pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

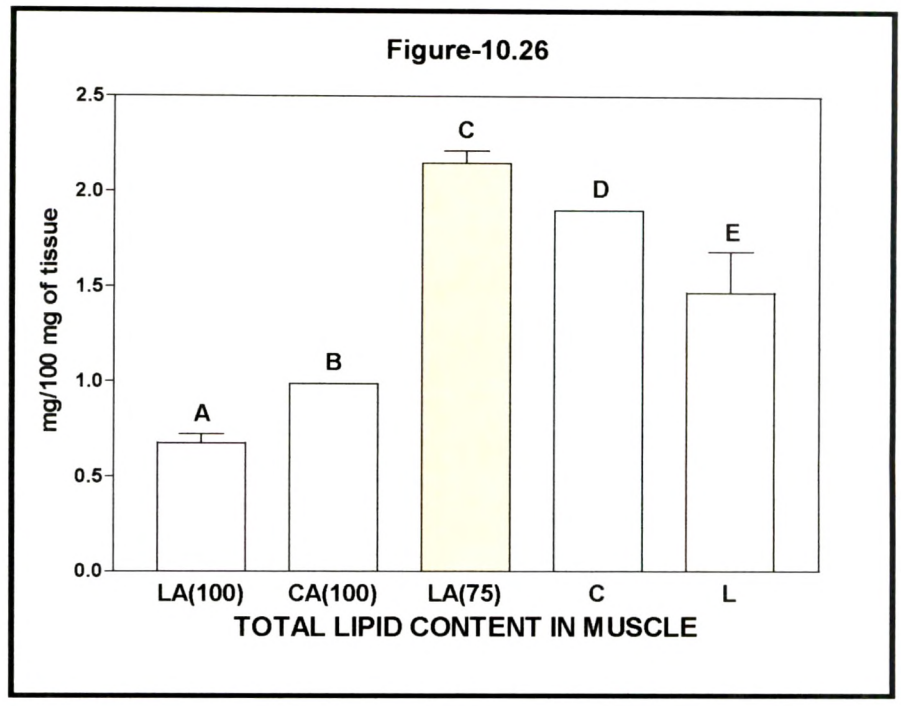


Table 10.26: Muscle total lipid content of pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

	LA(100) (A)	CA(100) (B)	LA(75) (C)	C (D)	L (E)
TOTAL LIPIDS	0.675 ±0.04785	0.987 ±0.00016	2.15 ±0.0645	1.9 ±0.00004	1.47 ±0.2136

Bonferroni's Multiple Comparison Test

	AvsB	AvsC	AvsD	AvsE	BvsC	BvsD	BvsE	CvsD	CvsE	DvsE
p	NS	◆	◆	◆	◆	◆	●	NS	■	NS

Values are expressed as mean ± SEM, ◆p<0.001; ■P<0.01; ●P<0.05; NSNon Significant

Figure 10.27: Muscle cholesterol content of pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

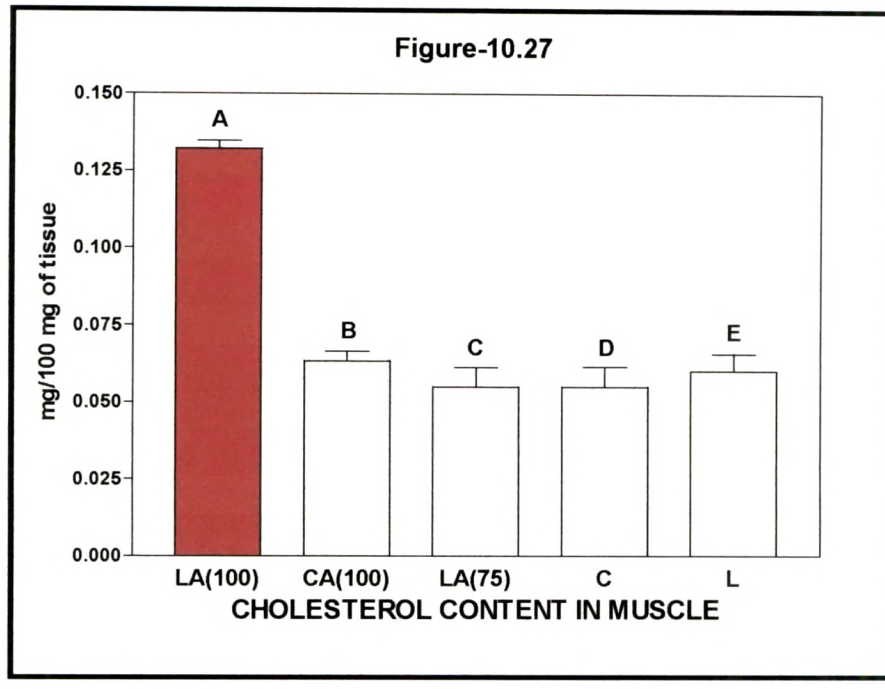


Table 10.27: Muscle cholesterol content of pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

	LA(100)(A)	CA(100)(B)	LA(75)(C)	C(D)	L(E)
CHOLESTEROL	0.1321 ±0.0026	0.0633 ±0.00315	0.055 ±0.0062	0.055 ±0.00645	0.06 ±0.0057

Bonferroni's Multiple Comparison Test

	AvsB	AvsC	AvsD	AvsE	BvsC	BvsD	BvsE	CvsD	CvsE	DvsE
p	◆	◆	◆	◆	NS	NS	NS	NS	NS	NS

Values are expressed as mean ± SEM, ◆p<0.001; NSNon Significant

Figure 10.28: Adipose tissue total lipid content of pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

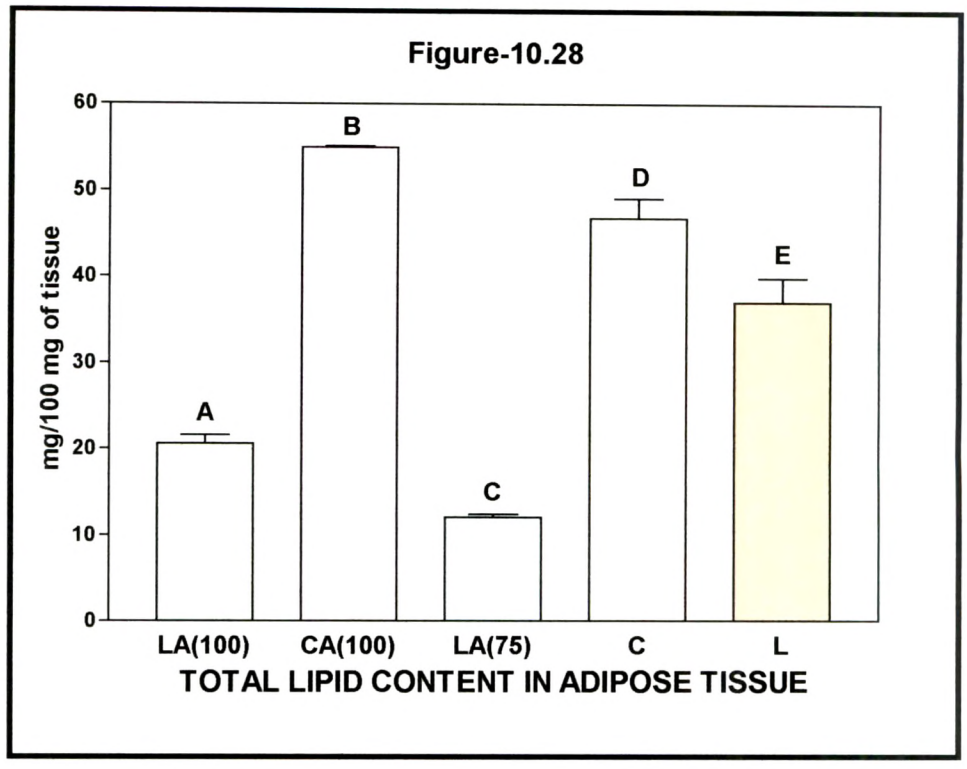


Table 10.28: Adipose tissue total lipid content of pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

	LA(100) (A)	CA(100) (B)	LA(75) (C)	C (D)	L (E)
TOTAL LIPIDS	20.6 ±0.9899	54.95 ±0.1499	12.066 ±0.3511	46.8 ±2.28	37.00 ±2.7967

Bonferroni's Multiple Comparison Test

	AvsB	AvsC	AvsD	AvsE	BvsC	BvsD	BvsE	CvsD	CvsE	DvsE
p	◆	●	◆	◆	◆	●	◆	◆	◆	■

Values are expressed as mean ± SEM, ◆p<0.001; ■P<0.01; ●P<0.05; NS Non Significant

Figure 10.29: Adipose tissue cholesterol content of pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

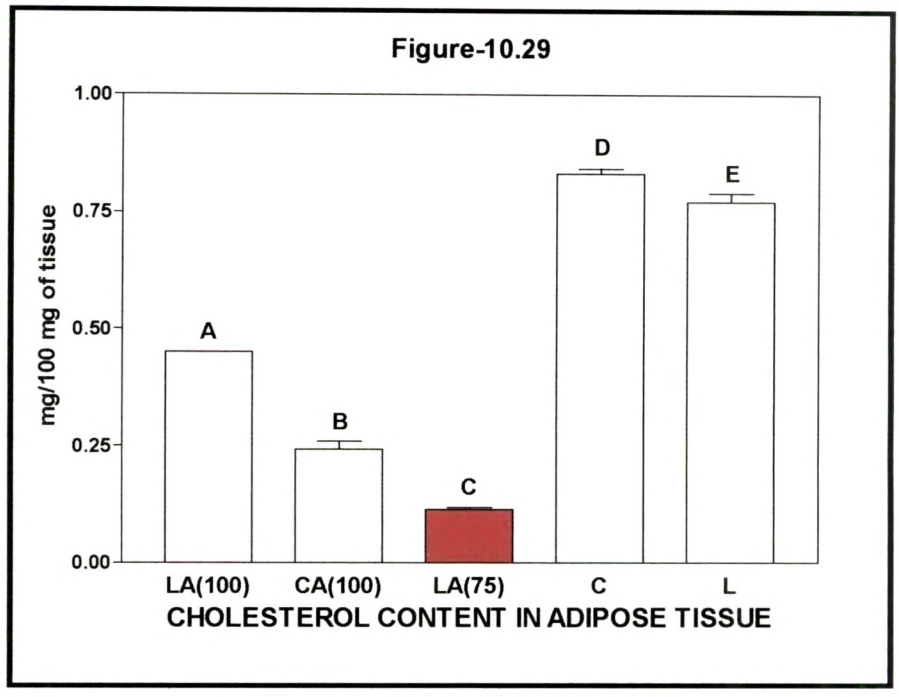


Table 10.29: Adipose tissue cholesterol content of pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

	LA(100)(A)	CA(100)(B)	LA(75)(C)	C(D)	L(E)
CHOLESTEROL	0.4514 ±0.00	0.2429 ±0.0173	0.1144 ±0.0033	0.832 ±0.01105	0.772 ±0.179

Bonferroni's Multiple Comparison Test

	AvsB	AvsC	AvsD	AvsE	BvsC	BvsD	BvsE	CvsD	CvsE	DvsE
p	◆	◆	◆	◆	◆	◆	◆	◆	◆	●

Values are expressed as mean ± SEM, ◆p<0.001; ●P<0.05; NS Non Significant

Figure 10.30: Serum triglyceride level of pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

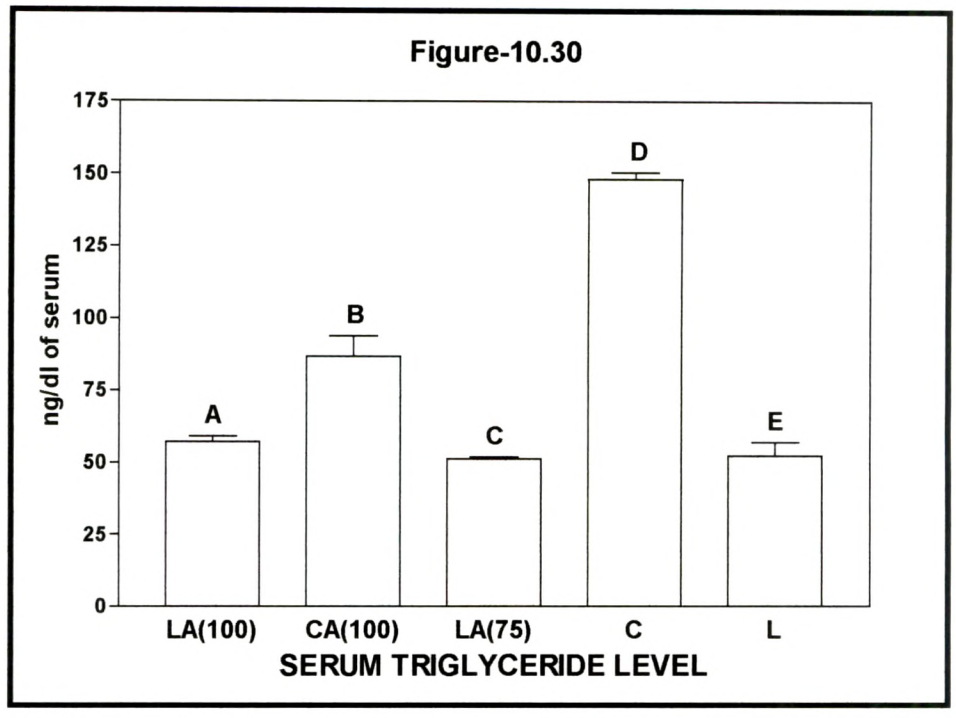


Table 10.30: Serum triglyceride level of pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

	LA(100)(A)	CA(100)(B)	LA(75)(C)	C(D)	L(E)
SERUM TRIGLYCERIDE	57.10 ±1.9985	86.785 ±6.9035	51.30 ±0.5093	148.187 ±2.3246	52.31 ±4.662

Bonferroni's Multiple Comparison Test

	AvsB	AvsC	AvsD	AvsE	BvsC	BvsD	BvsE	CvsD	CvsE	DvsE
p	◆	NS	◆	NS	◆	◆	◆	◆	NS	◆

Values are expressed as mean ± SEM, ◆p<0.001; NSNon Significant

Figure 10.31: Serum cholesterol level of pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

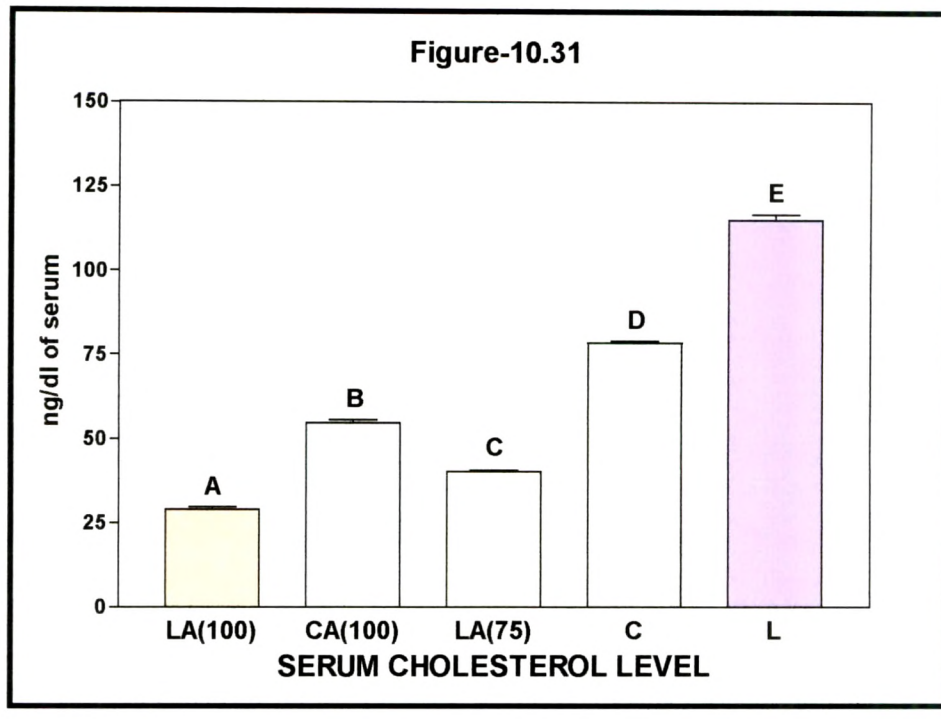


Table 10.31: Serum cholesterol level of pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

	LA(100)(A)	CA(100)(B)	LA(75)(C)	C(D)	L(E)
SERUM CHOLESTEROL	28.90 ±0.7911	54.58 ±1.6534	40.21 ±0.4306	78.492 ±0.5883	115.06 ±1.79

Bonferroni's Multiple Comparison Test

	AvsB	AvsC	AvsD	AvsE	BvsC	BvsD	BvsE	CvsD	CvsE	DvsE
p	◆	◆	◆	◆	◆	◆	◆	◆	◆	◆

Values are expressed as mean ± SEM, ◆p<0.001

Figure 10.32: Serum total lipids level of pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

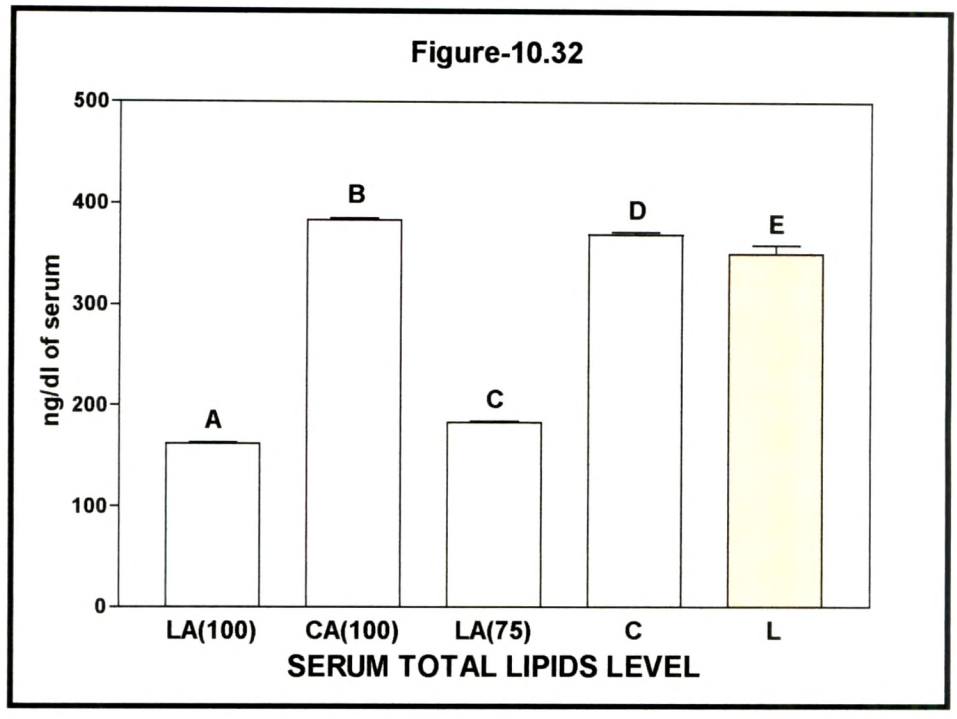


Table 10.32: Serum total lipids level of pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

	LA(100) (A)	CA(100) (B)	LA(75) (C)	C (D)	L (E)
SERUM TOTAL LIPIDS	162.08 ±1.272	383.57 ±1.9725	183.57 ±1.306	369.98 ±2.6205	351.12 ±8.364

Bonferroni's Multiple Comparison Test

	AvsB	AvsC	AvsD	AvsE	BvsC	BvsD	BvsE	CvsD	CvsE	DvsE
p	◆	●	◆	◆	◆	NS	◆	◆	◆	NS

Values are expressed as mean ± SEM, ◆p<0.001; ●P<0.05; NSNon Significant

Figure 10.33: Serum phospholipids level of pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

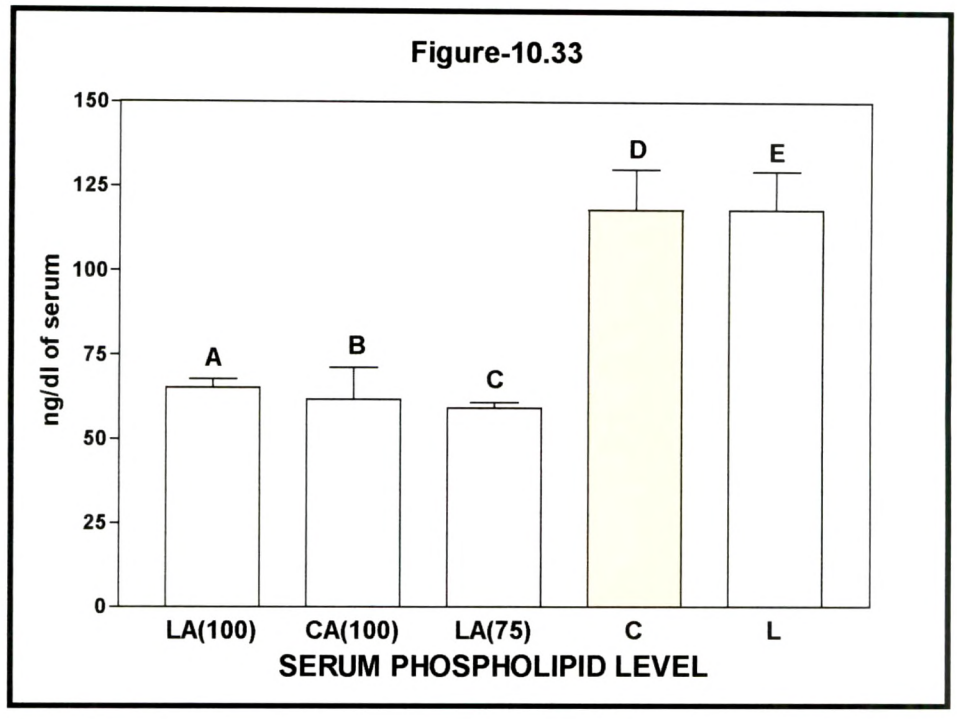


Table 10.33: Serum phospholipids level of pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

	LA(100)(A)	CA(100)(B)	LA(75)(C)	C(D)	L(E)
SERUM PHOSPHOLIPIDS	65.20 ±2.56	61.82 ±9.39	59.17 ±1.62	117.97 ±17.05	118.08 ±11.49

Bonferroni's Multiple Comparison Test

	AvsB	AvsC	AvsD	AvsE	BvsC	BvsD	BvsE	CvsD	CvsE	DvsE
p	NS	NS	■	■	NS	■	■	■	■	■

Values are expressed as mean ± SEM, ■P<0.01; NSNon Significant

Figure 10.34: Serum free fatty acid level of pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

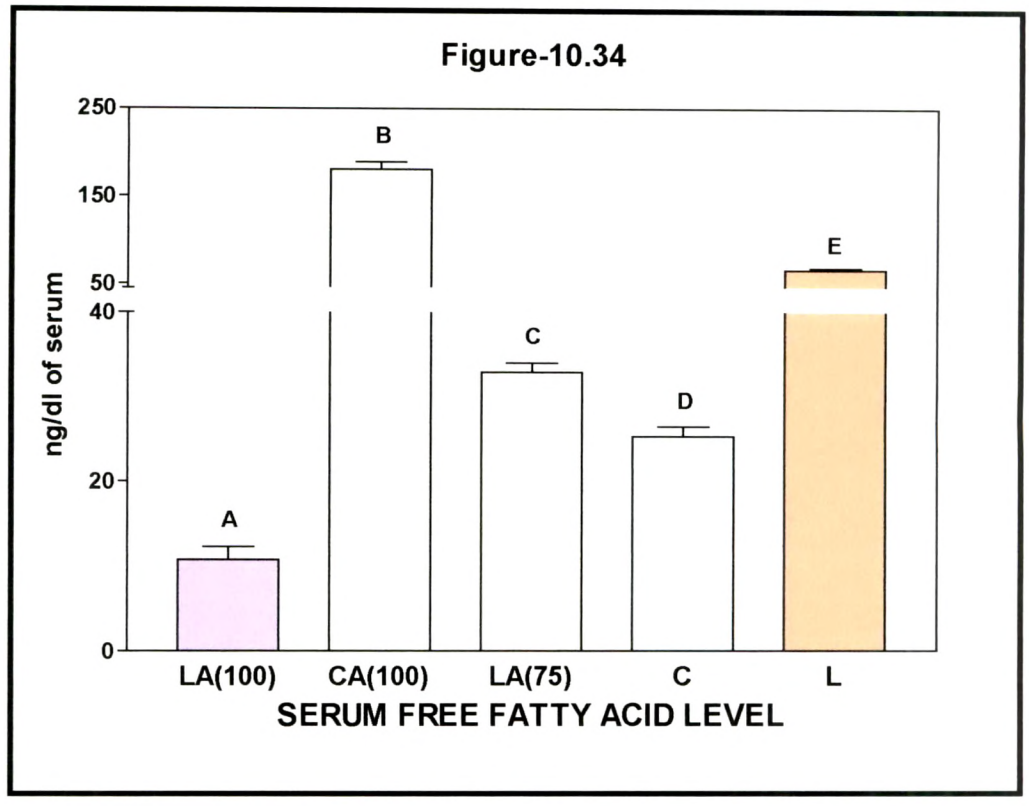


Table 10.34: Serum free fatty acid (FFA) level of pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

	LA(100) (A)	CA(100) (B)	LA(75) (C)	C (D)	L (E)
SERUM FFA	10.82 ±1.50	180.39 ±8.51	32.89 ±1.10	25.33 ±1.16	65.67 ±2.31

Bonferroni's Multiple Comparison Test

	AvsB	AvsC	AvsD	AvsE	BvsC	BvsD	BvsE	CvsD	CvsE	DvsE
p	◆	●	NS	◆	◆	◆	◆	NS	◆	◆

Values are expressed as mean ± SEM, ■P<0.01; NSNon Significant

Figure10.35: Serum insulin level of pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

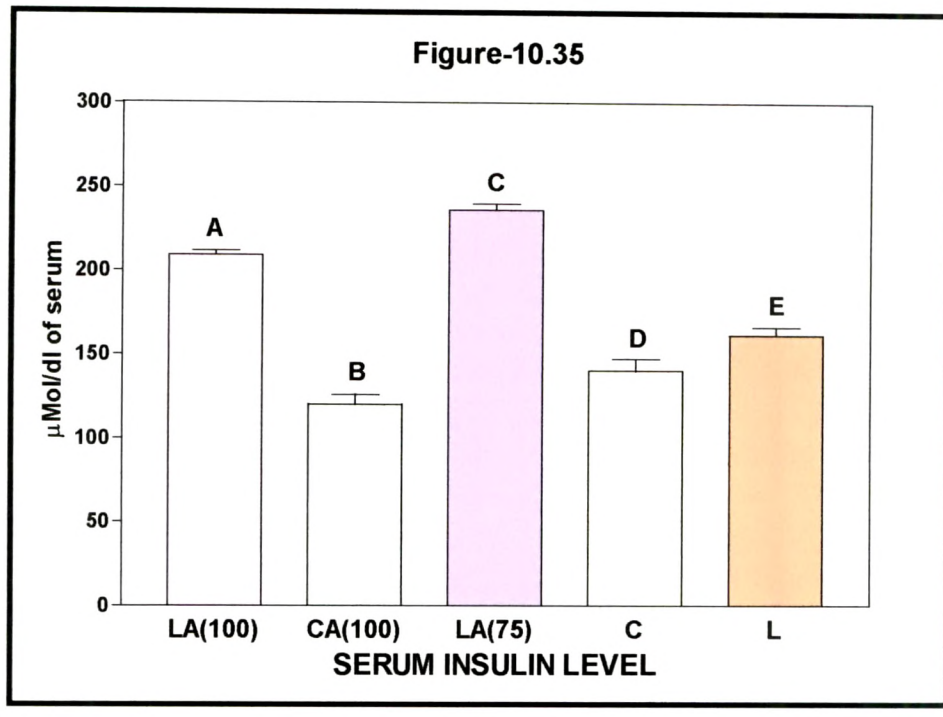


Table10.35: Serum insulin level of pubertal rats on 45th day subjected to neonatal luzindole treatment and weaning alloxanisation on 22nd day:

	LA(100) (A)	CA(100) (B)	LA(75) (C)	C (D)	L (E)
INSULIN	209.19 2.685	120.08 ±5.845	236.26 ±3.458	140.27 ±7.0682	161.65 ±4.8055

Bonferroni's Multiple Comparison Test

	AvsB	AvsC	AvsD	AvsE	BvsC	BvsD	BvsE	CvsD	CvsE	DvsE
p	◆	●	◆	◆	◆	NS	◆	◆	◆	NS

Values are expressed as mean ± SEM, ◆p<0.001; ●P<0.05; NSNon Significant

PLATE – 11

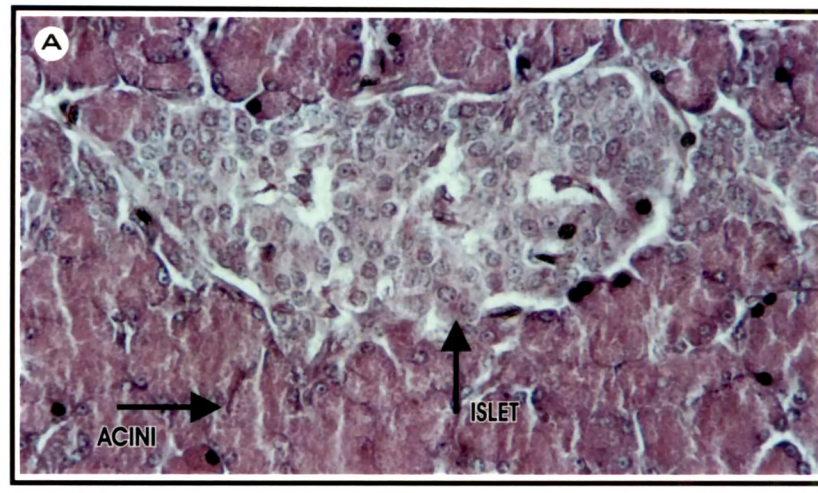
Photomicrographs of sections of pancreas – 450 X

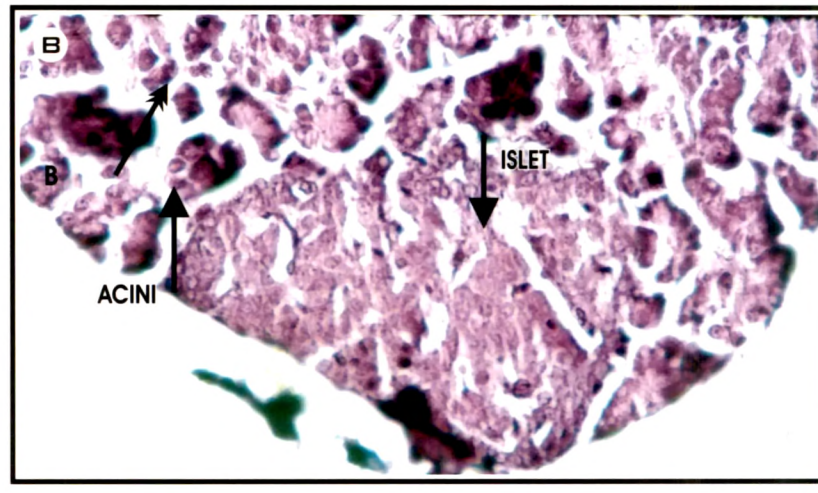
FIGURE (A): Transverse section of the pancreas of male luzindole alloxanised (100) treated rats on the 45th day showing islet and pancreatic acini. There is an increase in the islet size and islet cell number, with an increased B:A cell ratio.

FIGURE (B): Transverse section of the pancreas of male control alloxanised (100) pubertal (45th day) rats showing islet and pancreatic acini. Note the decreased islet size and reduced B as well as A cell number. Also the islet and acinar cells show clear areas due to loss of cells.

FIGURE (C): Transverse section of the pancreas of male luzindole alloxanised (75) treated rats on the 45th day showing islet and pancreatic acini. There is an increase in the islet size and islet cell number, with an increased B:A cell ratio.

PLATE - 11





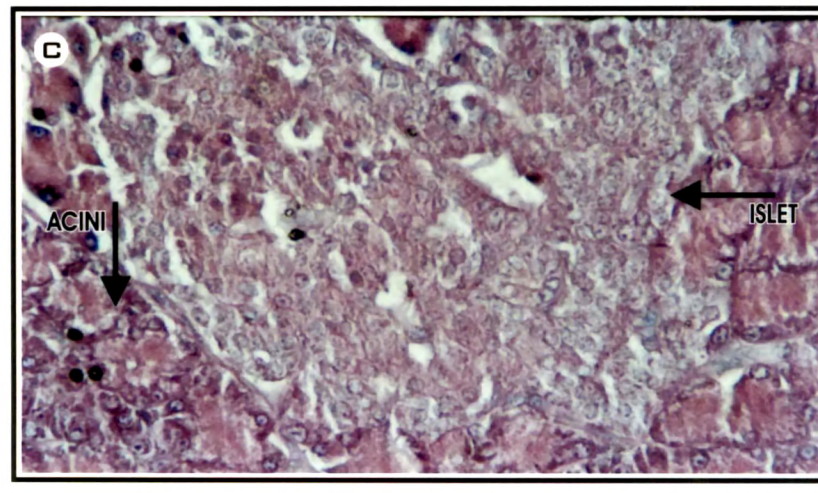


PLATE – 12

Photomicrographs of sections of pancreas – 1000 X

FIGURE (A): Transverse section of the pancreas of male luzindole alloxanised (100) treated rats on the 45th day showing increased transdifferentiation as well as increased B cell population with reduced effect of the damage caused by allxoan.

FIGURE (B): Transverse section of the pancreas of male control alloxanised (100) pubertal (45th day) rats showing islet and pancreatic acini. Note the reduced B as well as A cell population. Also the islet and acinar cells show clear areas due to loss of cells.

FIGURE (C): Transverse section of the pancreas of male luzindole alloxanised (75) treated rats on the 45th day showing islet and pancreatic acini. There is an increase in the islet size and islet cell number, with an increased B:A cell ratio. Also the transdifferentiating cells are visible at the periphery of the islet and at the junction of acini

PLATE - 12

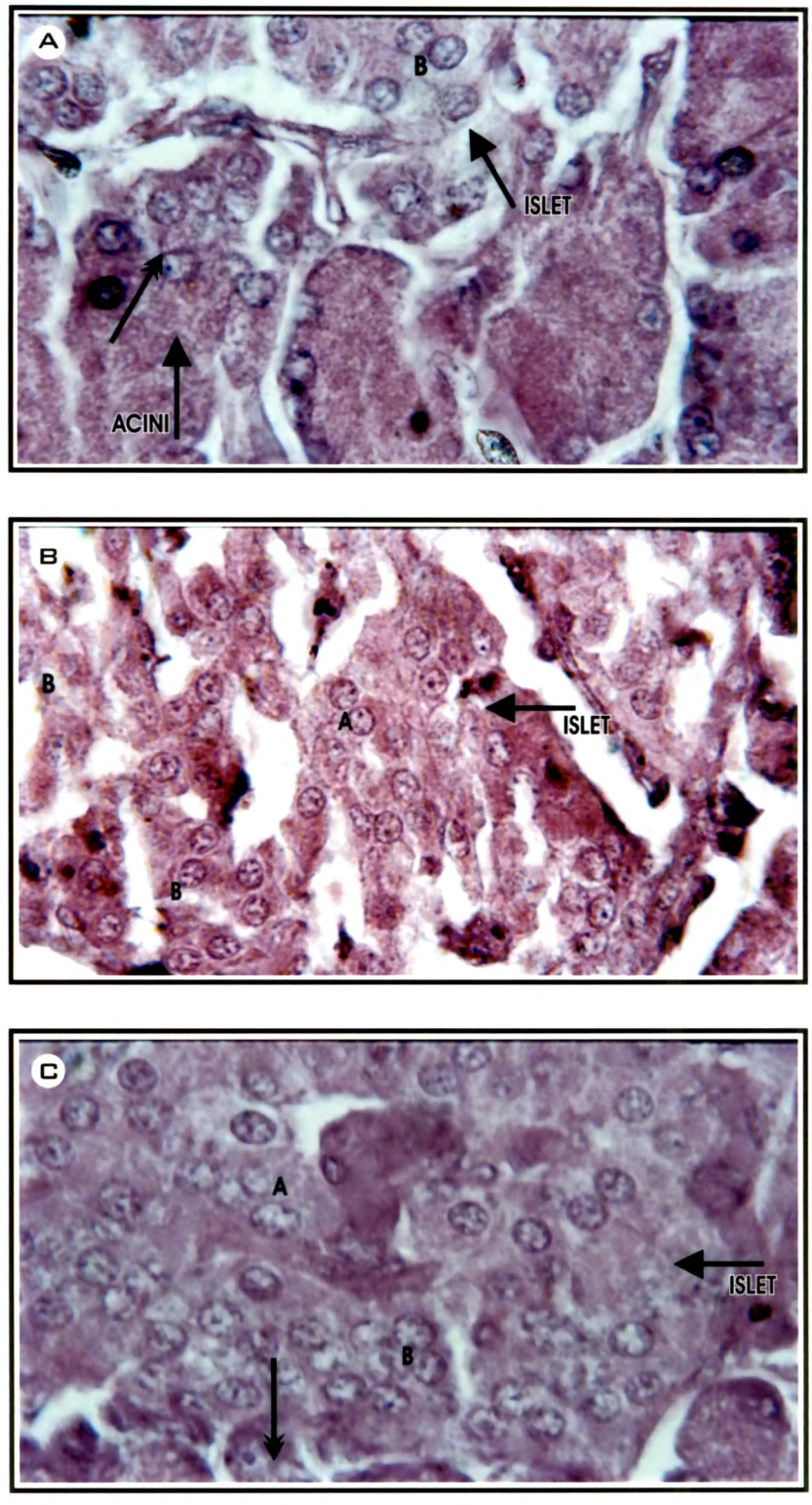


PLATE – 13

Photomicrographs of sections of pancreas – 1000 X

FIGURE (A): Transverse section of the pancreas of male luzindole alloxanised (100) treated rats on the 45th day showing increased transdifferentiation as well as increased B cell population with reduced effect of the damage caused by allxoan.

FIGURE (B): Transverse section of the pancreas of male control alloxanised (100) pubertal (45th day) rats showing islet and pancreatic acini. Note the reduced B as well as A cell population. Also the islet and acinar cells show clear areas due to loss of cells.

FIGURE (C): Transverse section of the pancreas of male luzindole alloxanised (75) treated rats on the 45th day showing islet and pancreatic acini. There is an increase in the islet size and islet cell number, with an increased B:A cell ratio. Also the transdifferentiating cells are visible at the periphery of the islet and at the junction of acini. Some degree of damage is visible in the form of clear spaces (double headed dotted arrow).

PLATE - 13

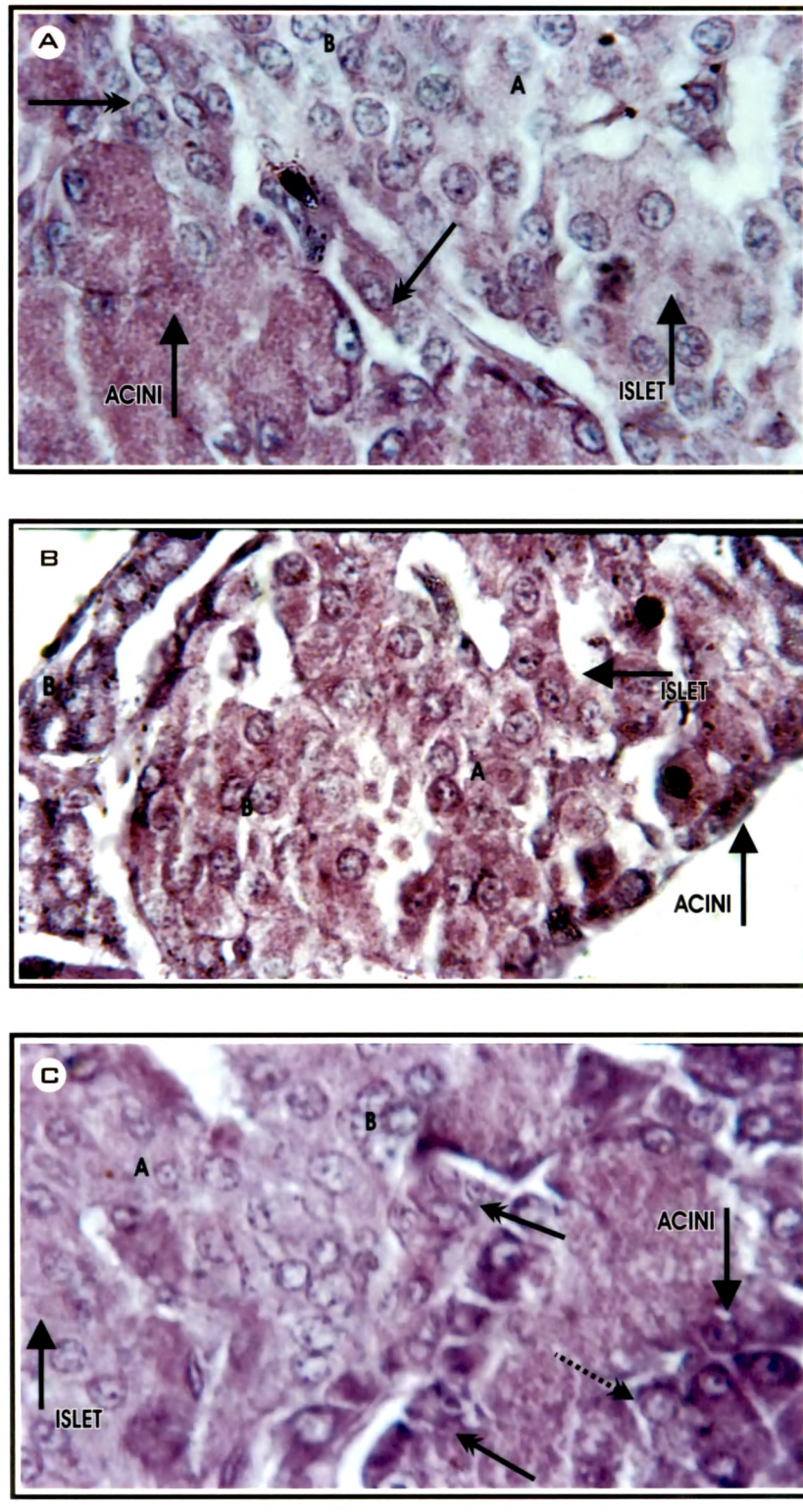


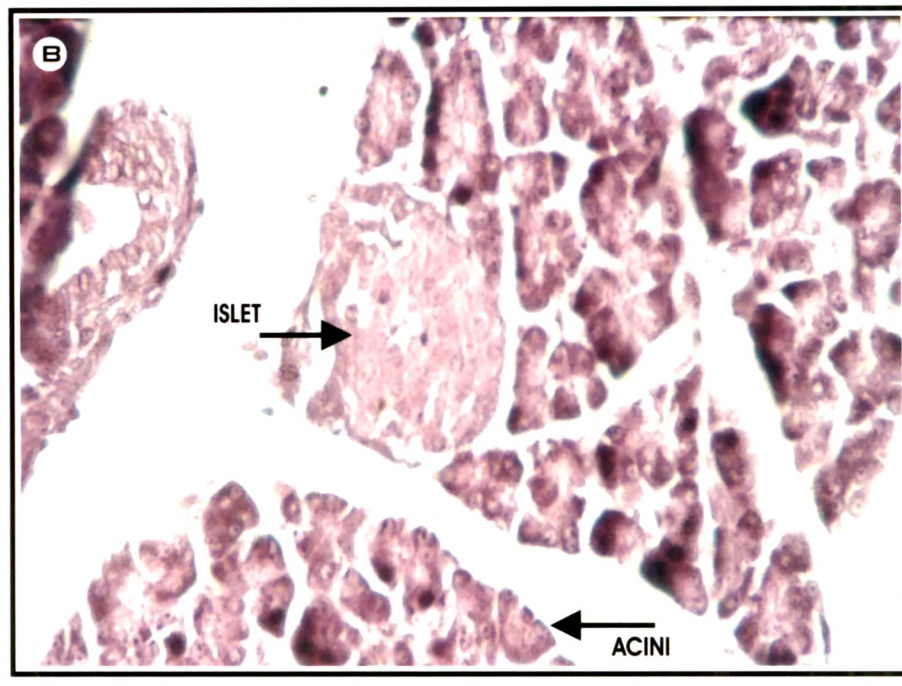
PLATE – 14

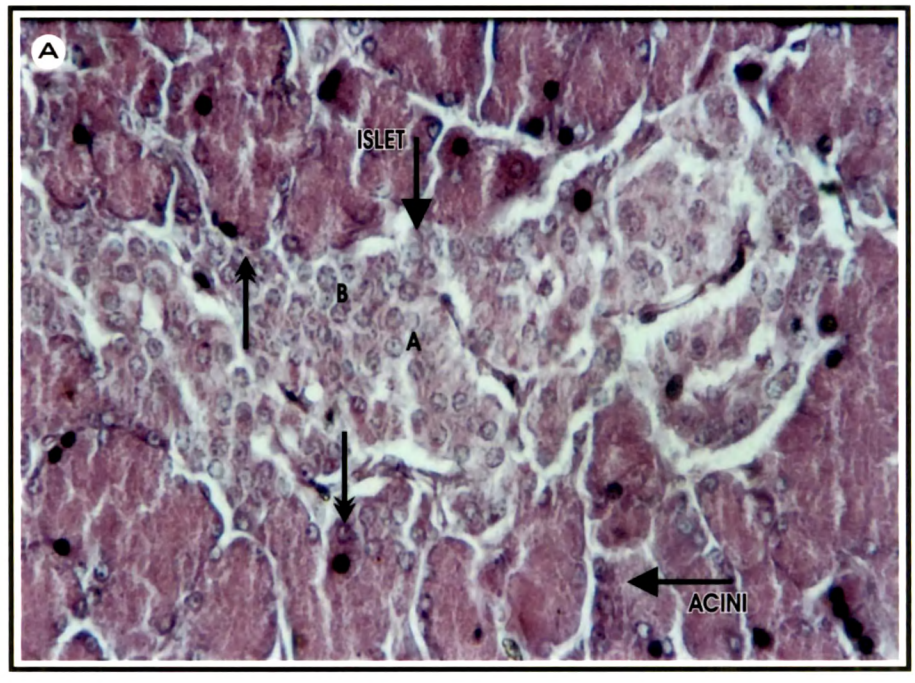
Photomicrographs of sections of pancreas – 450 X

FIGURE (A): Transverse section of the pancreas of male control alloxanised pubertal (45^{th} day) rats showing islet and pancreatic acini. Note the increased B cells as compared to A cells.

FIGURE (B): Transverse section of the pancreas of male luzindole alloxanised (100) treated rats on the 45^{th} day showing islet and pancreatic acini. There is an tremendous increase in the islet size and islet cell number, with an increased B:A cell ratio. Note the islet periphery showing acinar cells transdifferentiating (double headed arrow) into islet cells.

PLATE - 14





DISCUSSION:

The present results show that weaning alloxan treatment has significant effects on body and organ weights as well as on the metabolic features of control nMT animals as well as in nL animals which are quite distinct from those shown by non nL animals. This suggests the effect of nL treatment on alloxan induced changes. The body weight of nL animals are significantly reduced in a dose dependent manner while, there is no significant difference in non nL rats (Fig. and Tab.; 10.1). Whereas the relative weight of pancreas, liver, spleen and adrenals showed a decrease in non nLT rats the nLT(100 mg alloxan) rats showed significant increase in weight except for liver (Fig. and Tab.; 10.8, 10.9, 10.10, 10.11, 10.12, 10.13). The nL(75 mg alloxan) rats showed uniform increase in weight for all these organs. Kidney in general shows an increase in all alloxanised animals more significantly in nL rats. The increase in the relative weight of all the organs and decrease in body weight suggest altered growth kinetic in nL rats whose underlying cause is inexplicable at this juncture. The significant increase in the adrenal weight of alloxanised nL rats suggests the activation of hypothalamo hypophysial adrenal axis.

The significant hyperglycemia seen in the alloxanised control animals suggests poor recovery of B cell mass from alloxan challenge (Fig. and Tab.; 10.16; Plate, 11-14). This is reflected in the depleted hepatic glycogen content and hypoinsulinemia seen in these rats (Fig. and Tab.; 10.14, 10.15, 10.35). However the insulin sensitivity of the muscle of these animals seems to have increased as reflected in the

increased glycogen load, which is more than that seen in the nL rats shown on a percentage basis. Obviously increased insulin sensitivity and glycogen accumulation seems to be a characteristic feature of both control and nL animals subsequent to alloxanised animals. The nL animals however show significant hypoglycemia with relatively higher hepatic glycogen content which would suggest a faster recovery of the B cells after alloxan induced damage. Apparently reduced melatonin action in the neonatal period has a favorable influence on B cell recovery/regeneration. Histological observations reveal the enlarged islets/islet mass with increased number of B cells generated probably by both proliferation of surviving B cells as well as neogenesis by transdifferentiation from acinar cell in nL rats after alloxan induced damage more pronouncedly in the nL(75) rats (Plate, 11-14). It is now realized that islet growth and increase in B cell number are features characteristic of even adult animals (Fernandes *et al.*, 1997; Bonner-Weir, 2000; Lipsett and Finegood, 2002; Paris *et al.*, 2003). This ability of B cell generation is more pronouncedly seen in the pre adult stages extending from neonatal to pubertal age (Bonner-Weir, 2000; Arulmozhi *et al.*, 2004). Based on the present study it is inferable that B cell growth, recovery and regeneration are all heightened under a low neonatal background. Increased islet cell density was a feature reported earlier on rats treated with luzindole in neonatal period (Chapter 1-4 & 7). The favorable influence of reduced neonatal melatonin action on B cell recovery and regeneration subsequent to weaning alloxan challenge is substantiated by the recorded

hyperinsulinemia in these animals. A greater insulin action is seen in nL(75) rats as indicated by not only increased hepatic and muscle glycogen content but also the significantly higher lipid deposition in these organs. Whereas the C(100) and nL(100) rats have shown a decrease in tissue lipid content including the adipose tissue relative to their non-alloxanised counter parts (Fig. and Tab.; 10.28). An interesting observation is the significantly reduced adipose tissue lipid load simultaneous to increased liver lipid load in nL(75) rats which is quite distinct reverse pattern of higher adipose tissue lipid load and lower hepatic lipid load in both C(100) and nL(100) (Fig. and Tab.; 10.24). Hypertriglyceridemia and higher LDL usually seen in diabetic condition are related with a decreased activity of insulin sensitive lipoprotein lipase (Pfeifer *et al.*, 1983; Taskinen *et al.*, 1986) and increased expression of microsomal triglyceride transfer protein (MTP) for protein (Ginsberg and Grundry, 1982; Kuriyama *et al.*, 1998). Except for the increased total lipid content in the nL(75) rats, tissue lipid profiles (hepatic, muscle, adipose tissue, total lipids and cholesterol) as well as levels of serum lipid fractions (Triglycerides, Cholesterol, Total lipids and Fatty acids) have shown significant decrement in both nL(75) and nL(100) rats (Fig. and Tab.; 10.24, 10.25, 10.26, 10.27, 10.28, 10.29, 10.30-10.34). Obviously alloxan induced diabetogenic effects are effectively countered by prior neonatal functional deficiency of melatonin. This antidiabetogenic effects are also substantiated by the relatively lesser protein anabolism seen in these rats as marked by the hepatic and muscle protein contents. This

is in contrast to the reported pinealectomy induced potentiation of type II diabetic features of hyperinsulinemia and hepatic triglyceride content (Nishida *et al.*, 2003).

It is concluded from the present observations that neonatal functional melatonin deficiency has favorable influence in preventing the diabetogenic effects of alloxan on serum insulin, glucose and lipids and tissue metabolite load as well as promotes B cell proliferation and neogenesis.

SUMMARY:

Previous studies on the effect of neonatal melatonin antagonism on weaning age revealed hyperinsulinemia, hyperglycemia, increased insulin sensitivity and glycogenic effect and decreased lipogenesis in the pubertal period and marked by reduced insulin sensitivity with decreased glycogenic effect and increased lipid and protein content in the adult stage. In this context, it was thought interesting to test the effect of neonatal melatonin antagonism by luzindole treatment on weaning induced diabetes by alloxan treatment. Since experimental induction of diabetes has been carried out in neonates and adults, it was thought interesting to see the effects of induction of diabetes at weaning age. To this end, rat neonates have been treated with Luzindole (An MT_2 receptor blocker) (400 µg/Kg body weight) intra peritoneally from day 1 to day 21 to generate a hypomelatonemic status and; a low (100 µg/kg) dose and a high dose (150 µg/kg) of alloxan were given on the 22^{nd} day and the effects assessed on the 45^{th} day. The LA(100) and LA(75) rats showed significantly decreased

body weights as compared to CA(100) and age matched controls. The relative weight of pancreas did not show any alteration in control and alloxanised rats. The relative weight of liver of LA(100) rats decreased significantly as compared control rats of the same age. The relative weight of spleen of LA(75) increased significantly as compared to all other groups. The relative weight of kidney of LA(100) and LA(75) rats increased significantly as compared to age matched controls. The hepatic glycogen content decreased significantly in all the alloxanised groups while, the muscle glycogen content increased significantly. The serum glucose level decreased significantly in all the alloxanised groups as compared to all other groups while the, serum insulin level in LA(100) and LA(75) increased significantly as compared to all other groups. The hepatic protein content did not show any significant alteration while, the muscle protein content increased significantly in all the alloxanised rats. The hepatic glycogen synthetase and glycogen phosphorylase activities decreased significantly in all the alloxanised groups as compared to the controls. The hepatic glucose-6-phosphatase activity increased significantly in the LA(100) rats as compared to all other groups. The muscle glycogen synthetase and glycogen phosphorylase activities of all the alloxanised groups decreased significantly as compared to the nLT rats. The hepatic total lipid content of LA(75) rats increased significantly as compared to all other groups while the hepatic cholesterol content did not show any significant alterations. The muscle total lipid content of LA(100) rats decreased significantly as compared to control rats of the same age

while the muscle cholesterol content of LA(100) rats increased significantly as compared to all other groups. The adipose tissue total lipid content of LA(100) and LA(75) rats decreased significantly as compared to CA(100) and control rats while, the adipose tissue cholesterol content of all the alloxanised rats decreased sitnificantly as compared to age matched control rats. The serum triglyceride, cholesterol and phospholipid levels decreased significantly in the alloxanised rats as compared to control rats of the same age. The serum total lipid and free fatty acid levels of LA(100) and LA(75) rats decreased significantly as compared to CA(100) and control rats of the same age. It can be inferred from the above observations that neonatal melatonin deficiency has favorable influence in preventing the diabetogenic effects of alloxan on serum insulin, glucose and lipids and tissue metabolite load as well as promotes B cell proliferation and neogenesis.