CHAPTER VIII

COMPARATIVE STUDY OF EARLY HEPATIC RESPONSE TO ANDROGEN ADMINISTRATION IN NORMAL INTACT AND 48-HOUR ORCHIDECTOMISED RATS

Influence of androgens within minutes/hours is now a known fact (Birkko, 1981; Ambadkar and Gangaramani, 1982; Max and Toop, 1983; Aruldhas and Govindrajulu, 1985). Earlier work carried out in our laboratory has revealed that injection of 0.1 mg of testosterone propionate was an ideal dose for reverting majority of the metabolic alterations induced after castration in case of hepatic gland. Based on this consideration, during the course of the present investigation, early effects of administration of 0.1 mg dose of TP were studied in case of normal intact albino rats. The purpose was to make a comparative study of immediate response of hepatic tissue to androgen administration in normal and the 48-hour orchidectomised animals. Further, it should be stated here that in organizing this chapter the usual pattern had to be altered to suit the major purpose of comparing the results reported in previous chapters with those from earlier work in case of 48-hour castrated rats.

Different methods employed have already been stated in Chapter-I, and, hence, are not repeated here. Instead of showing the results obtained presently in a separate table, the values have been incorporated in the composite tabulation, which depicts the values obtained earlier as well as those obtained during the course of the present investigation. The comparison represented in this table is for 60 and 120 minutes post administration intervals in case of all the parameters considered. The discussion that follows, therefore, is based on these points.

From the values given in the table it could be seen that mere castration (at 48-hours interval) led to noticeable rise in the plasma glucose level, whereas administration of TP to castrates led to not merely the reversal of castration effect but to a significant supporession of plasma glucose level (almost to 50%). On the other hand, taking into consideration the influence of 0.1 mg TP administration to normal intact rats, one finds that even within as short a span as 2 hours a graded and distinct hyperglycaemia was evident. It is logical to think that if mere castration induces noticeable hyperglycaemia, then TP administration should revert such an influence, however, the values obtained show much more than this kind of effect. Basing the thought on these premises; administration of TP to normal animals could not be expected to lead to distinct

hyperglycaemia around 120 minutes. In these circumstances it could be suggested that the early response of the rats to TP administration basically differs due to orchidectomy. It is, therefore, evident that TP administration to castrates leads to hypoglycaemic state, whereas exogeneous administration of the same hormone to intact animals leads to diametrically opposite influence. To the best of the knowledge of the present author such an early hyperglycaemic response to TP administration has so far not been reported. In these circumstances, the author was compelled to look for underlying biochemical changes such as the levels of hepatic glycogen content as well as enzyme activities viz.- glucose-6-phosphatase, glycogen synthetase and phosphorylase, for possible explanations.

From the data represented in the Table, it could be seen that the response of the hepatic tissue to TP administration in case of 48-hour castrates is basically different from that of the normal intact animals. It is, therefore, clear that circulating androgenic levels somehow influence sensitivity of the hepatic metabolic patterns. This phenomenon expressly needs further investigation and the idea is engaging the thoughts of the present author. The statement made above is borne out by the values reported in the table in case of variations in the levels of hepatic glycogen content as well as the enzymic

activities of glycogen synthetase, phosphorylase, cAMP-specific phosphodiesterase and glucose-6-phosphatase. Under both experimental conditions the hepatic tissue of rats responds to the administration of TP through suppression of phosphorylase activity eventhough a closely associated important enzyme - cAMP-phosphodiesterase registered significant lowering in one case (castrates) and elevation in the other (intact). This corroborates the statement regarding differential metabolic sensitivity under varying androgenic titres. More or less similar trend is evident in case of G-6-Pase activity, and this trend of variation is complimentary to the observed glycaemic levels. Taking into account the variations obtained in case of glycogen synthetase; it can be easily seen that administration of TP to castrates does not lead to any significant effect but it remarkably suppresses this enzyme activity within first 60 minutes of hormone injection in case of normal animals. However, this effect starts waning with the time lapse.

It is clear from these observations that the hepatic tissue responds differently to androgen administration, when the cells are starved of circulating androgen levels and when they are exposed to an excess of androgen level under <u>in vivo</u> condition. The observed hypoglycaemic condition after TP administration to 48-hour castrates is possibly due to the increase in the glucose utilization by

the peripheral tissues rather due to glycogenolysis, while the hyperglycaemia in normal animals after TP administration can be attributed to increased release of glucose into blood by the hepatic tissue despite the fact that phosphorylase activity exhibits subnormal levels. Though there are quantitative differences in the pattern of responses as far as the carbohydrate metabolism is concerned, one can still emphasise a common feature that the mechanism of glycogenolysis and that of glucose release (from liver into blood) are differently and independently influenced by circulating androgen levels.

Total protein content of the hepatic tissue was found to be increased after castration as well as after TP replacement in such castrates with no sign of recovery even after 2 hours of hormone treatment. In contrast, TP administration to normal intact rats caused progressive decline in the protein content, indicating a catabolic trend. This is in stark contradiction to well known anabolic influence of androgens in general. Therefore, to verify these findings further, a few enzyme activities concerned with overall protein metabolism as well as nucleic acid contents were investigated. Inter-relationship between nucleic acid content, RNA synthesis and that of protein content is well established (Ridford 1960; Hay and Fishman, 1961; Osteen and Walker, 1961).

Increased DNA content of the hepatic tissue due to castration and its subsequent status quo condition probably reflects on possible induction of polyploidy in the said gland (Konopkova and Medvidek, 1972). Contrary to this TP injection to intact animals points towards degradation of nucleic acids (DNA as well as RNA) and hence a definite impairment of protein synthesising machinery could be visualized. When one considers the rest of the data on protein metabolism, the picture that emerges is very confusing. The total hepatic protein content registered under all experimental conditions, considerably high levels, even in case of lowered RNA value in TP treated castrates. Under the circumstances the observed increase in protein content could be attributed to an yet early stimulating influence of hormone administration on a comparatively non-labile protein synthesizing enzymic machinery, existing after castration, and its facilitation under increased availability of c-AMP as indicated by significantly reduced c-AMP-specific phosphodiesterase activity level.

The observed alterations in the enzymic activities viz.- 5'-nucleotidase, Aspartate transaminase, and alanine transaminase point towards a similar pattern of response irrespective of the experimental conditions under consideration. Castration led to depletion initially, with

slight improvement after TP administration within first 60 minutes out later on again depletion was evident. Normal rats administered with TP responded only after 60 minutes but here also depletion was noticed. These alterations, with the data on hand, do not permit any definite conclusion regarding the changes in total protein content. However, one could still maintain that they do indicate a changed sensitivity of the hepatic tissue towards levels of androgenic hormones.

The involvement of AA in Steroidogenesis (Szent, 1957; Bacq and Alexander, 1961; Biswas and Deb, 1970; Chinoy, 1972a, b; Chinoy and Seethalakshmi, 1978), its dynamic interrelationship with circulating levels of testosterone (Chinoy and Parmar, 1975) as well as the results reported earlier (not included in thesis) provoked the idea of studying the impact of the experimental regimen on normal intact rats. Surprisingly it was observed that TP administration either radically suppressed AA synthesis or increased its utilization irrespective of either of the experimental conditions. In contrast to this, it is worthy of mention here that castration per se was seen to increase AA content of hepatic tissue. Furthermore, the blood AA levels do not show alteration after castration, and after

first hour of hormone injection, but at second hour, the AA content is depleted in castrates while elevated in intact animals. This indeed suggests that alterations in circulating testicular androgen levels exert some sort of a regulatory effect on the AA synthesis as well as its degradation/ retention by the hepatic tissue. Further, the mechanism of AA synthesis and retention are seemingly influenced in different ways and probably independently of each other.

Additionally, it would be appropriate to restate that the sensitivity of the Spigelian lobe of the rat liver obviously differs in several ways as compared to that of the median lobe (representative of rest of the lobes). This has been amply brought out by the data presented in previous chapter and in the present one.

EXPLANATIONS TO FLOW CHARTS

(1) Part 1 : Experiments and results based on castration and replacement therapy.

(2) Part 2 : Experiments and results based on TP administration to normal intact animals.

a. 30 minutes after TP administration

b. 60 minutes after TP administration

c. 90 minutes after TP administration

d. 120 minutes after TP administration

Abreviations used :

Metab.	-	Metabolism
AA	-	Ascorbic acid
PL Glucose	-	Plasma glucose
Gly.Syn.	-	Glycogen synthetose
Phrylase	-	Phosphorylase
PDE	-	Phosphodiesterase
G-6-Pase	-	Glucose-6-Phosphatase
5'-Nu	-	5'-Nucleotidase
DNA	-	Deoxyribose nucleotide
RNA	-	Ribose nucleotide
Pr.	-	Protein.

Median lobe - 

Spigelian lobe - 

Blood - 

Plasma - 

Increase - 

Decrease - 

No change -

Table : 8.1a Comparison and androgen administration to 48 hours castrates and normal intact animals

Parameters evaluated	NORMAL	48 hrs CASTRATES	48 hrs CAS + 0.1 mg TP 1 HR	NORMAL + 0.1 mg TP. 1 HR	48 hrs CAS + 0.1 mg TP 2 HR	NORMAL + 0.1 mg TP 2 HR
Blood AA	1.16 ±0.23	1.28 ±0.03	0.933 ±0.29	0.99 ±0.09	0.87 ±0.13	1.45 ±0.22
Plesma-glucose	80.12 ± 2.6	96.80 ± 6.52	39.50 ± 3.15	89.83 ± 9.3	46.00 ± 2.68	126.33 ± 3.2

Table : 8.1b Comparision of androgen administration to 48 hrs. Castrates and normal intact animals

Parameters evaluated	NORMAL		48-hrs. CASTRATES		48-hrs CAS+0.1 mg TP 1 hr.	NORMAL + 0.1 mg TP 1 hr.	48-hrs CAS+0.1 mg TP 2 hrs.	NORMAL + 0.1 mg TP 2 hrs.	48-hrs CAS+0.1 mg TP 1 hr.
	M	Sp	M	Sp	Median - lobe				
Glycogen	3.83 ±0.12	3.66 ±0.16	7.77 ±0.50	8.17 ±0.43	8.33 ±0.07	1.75 ±0.2	6.61 ±0.06	1.73 ±0.13	6.51 ±0.06
G-6-Pase	1.93 ±0.13	1.57 ±0.10	1.84 ±0.09	1.74 ±0.09	1.34 ±0.18	3.87 ±0.45	1.01 ±0.05	2.21 ±0.16	1.05 ±0.19
Glycogen synthetase	0.32 ±0.03	0.32 ±0.03	0.23 ±0.05	0.23 ±0.03	0.22 ±0.02	0.02 ±0.002	0.28 ±0.04	0.29 ±0.04	0.19 ±0.04
Phosphorylase	78.82 ±1.7	58.74 ± 1.3	90.0 ±4.49	59.74 ±2.58	17.04 ±2.05	64.54 ± 1.0	21.38 ± 1.69	60.21 ± 7.0	13.36 ± 2.22
5'-nucleotidase	4.60 ±0.34	4.03 ±0.20	1.90 ±0.19	1.75 ±0.16	5.51 ±0.66	4.85 ±0.22	0.98 ±0.05	3.42 ±0.78	3.10 ±0.037
GOT	262.29 ± 9.9	222.57 ± 5.9	205.28 ± 6.4	193.47 ±18.4	236.34 ±20.1	253.25 ± 5.7	156.18 ±29.2	65.49 ±7.7	236.48 ± 17.1
GPT	101.03 ± 7.6	107.40 ± 4.4	82.51 ±3.9	82.75 ±7.0	71.44 ±10.2	126.5 ±11.3	53.08 ±2.6	61.30 ±12.7	82.93 ± 4.6
Phosphodiesterase	2.34 ± 0.04	2.32 ± 0.04	1.69 ±0.052	1.57 ±0.55	1.38 ±0.18	4.9 ±0.41	1.50 ±0.13	7.17 ±0.35	1.88 ± 0.13
Total Protein	14.41 ±1.67	17.55 ± 2.73	18.24 ±0.74	19.11 ±0.58	20.93 ±0.60	9.65 ±0.8	20.44 ±0.1	7.32 ±0.6	21.93 ± 0.45
DNA	0.044 ±0.01	0.064 ±0.009	0.06 ±0.009	0.08 ±0.007	0.048 ±0.001	0.026 ±0.002	0.055 ±0.007	0.033 ±0.002	0.053 ± 0.003
RNA	0.050 ±0.009	0.060 ±0.014	0.055 ±0.003	0.062 ±0.004	0.008 ±0.002	0.03 ±0.002	0.014 ±0.00	0.018 ±0.001	0.008 ± 0.001
Tissue A.A.	31.66 ±3.02	34.53 ± 3.33	46.18 ±0.98	52.28 ± 0.85	24.74 ± 2.24	25.42 ± 2.73	23.90 ±2.52	19.64 ±1.98	22.09 ± 2.27