CHAPTER VI

EARLY INFLUENCE OF TESTOSTERONE PROPIONATE ADMINISTRATION TO NORMAL INTACT MALE ALBINO RATS (Rattus norvegicus albinus) ON CERTAIN ASPECTS OF HEPATIC METABOLIC PATTERNS: III PROTEIN AND NUCLEIC ACID METABOLISM

It is now an accepted fact that regulation of protein metabolism is achieved by mechanisms operating at the sub-cellular level and also by co-ordinated actions between cells and tissues. (Brayson et al., 1965; Malt, 1967; Blakley and Vitols, 1968; Diczfalusy, 1973). Hormones play a major role in regulation of biosynthesis of specific proteins. (Talalay and Williams-Ashman,1958; Williams-Ashman et al., 1958; Mayol and Thayer,1970; Beier and Beier-Hellwig, 1973; Scherrer, 1973; Talwar et al., 1973). Moreover, it is a known fact that different tissues respond differently to the same hormones (Barrington, 1964; Bern and Micoll, 1968). Leathem (1970) has pointed out that certain hormones; under particular experimental set up, have inhibitory influence on protein metabolism in some organs, whereas, they may have stimulatory effect in others. Anabolic influence of male sex hormones on growth and development of reproductive and accessory sex organs is a well documented fact (Kochakian and Harrison, 1962; Kochakian,1964; Doeg et al., 1972; Mainwaring and Wilce,1972;

Coffey, 1974; Liang and Liao, 1975; Rajalakshmi and Prasad, 1976). It is a proven fact that both DNA and RNA are necessarily associated with protein synthesis as well as growth (Wilson, 1962; Vollmer and Kauffmann, 1963; Kidson and Kirby, 1964; Liao et al., 1965; Widnell and Tata, 1966; Willams-Ashman and Shimazaki, 1967; Kosto et al., 1967; Minguell and Sierralta, 1975; Kurtz et al., 1976). Apart from androgens, the oestrogens are also known to influence protein metabolism (Aschkenasy-Lelu and Aschkensasy, 1959), wherein these authors have pointed out dose-dependent actions of oestrogens at low dose levels oestrogens were noted to have slightly anabolic actions, but at higher dosages they were distinctly catabolic. It is also very obvious from several studies (Fuji-Villee, 1968; Mayol and Thayer, 1970; Stormshak et al., 1976, and Engel et al., 1980) that it is not only the dose level of hormone that counts but their effects need also be studied in relation with time. Taking this into consideration; different doses of androgens were selected for the present study. Munro (1970) has suggested that cells/tissues are capable of synthesizing and degrading their own constituent proteins. The liver proteins, together with plasma proteins synthesized by the liver, constitute the major component of labile proteins. Deposition or loss of proteins from liver and pancreas is faster than any other organ. As early as 1946, Kochakian observed that androgens have anabolic effect in

castrated as well as intact animals. However, the present study reports on different pattern of response of hepatic tissue as influenced by TP administration to intact animals.

The earlier work done in this laboratory has shown that, as far as the protein metabolism is concerned, even deprivation of androgens showed an anabolic effect within first 48-hours of orchidectomy and also with subsequent replacement therapy within as short a span as few hours (Ambadkar and Gangaramani, 1981; Ambadkar et al., 1987). It was, therefore, thought necessary to investigate the possible early effects of androgens on the hepatic tissue of the normal healthy intact male albino rats.

MATERIALS AND METHODS :

Adult male albino rats (Rattus norvegicus albinus) weighing 120-160 gms were employed as experimental animals. Testosterone propionate (TP) was injected intramuscularly as a single dose per animal before sacrifice. Three different experimental groups were injected with 0.1, 0.25 and 0.5 mg of TP and these animals were sacrificed after 30, 60, 90, and 120 minutes. Total protein, DNA and RNA contents were quantitatively estimated in the hepatic lobes (M and Sp.). The other details were as described in Chapter-1.

RESULTS :

DNA : With 0.1 mg TP administration it was observed that in the case of M-lobe there was consistant and gradual suppression right from 30 minutes interval to 90 minutes, but at 120 minutes interval signs of recovery were evident (Table 6.2, Fig.6.2), whereas, in the case of Sp.lobe, suppression of DNA level was apparent upto one hour but thereafter there was recovery of DNA content through 90 and 120 minutes. 0.25 mg dose of TP did induce reduction in DNA content of M-lobe as well as that of Sp.lobe. However, there was a striking increase, above normal level, in case of the M-lobe which was also apparent at 90 and 120 minutes. The Sp.lobe also registered a highly significant rise in DNA content by 60 minutes, nevertheless, the recovery to normal level was apparent by 90 as well as 120 minutes intervals. In a marked contrast the higher dose of 0.5 mg TP was noted to suppress significantly the DNA levels in case of both the lobes and at all the intervals studied.

RNA : 0.1 mg TP injection to normal rats showed that there was a consistant suppression in RNA content of M-lobe right from 30 minutes interval to 90 minutes and was maximum at 120 minutes (Table 6.3). In the case of Sp.lobe, initially there was no suppression in the RNA

level, but at later intervals it exhibited a trend similar to that observed in M-lobe, with maximum influence at 120 minutes interval. Higher dosages of 0.25 mg and 0.5 mg of TP revealed highly significant reduction in RNA content of both the liver lobes as early as 30 minutes intervals, and it continued to be so at all intervals studied.

It is apparent from table 6.3 that both the liver lobes respond to TP administration in a dose-dependent manner and also with respect to time lapse; the suppression of RNA levels being greater and quicker with increasing dose.

PROTEIN : From the values represented in Table 6.1 it could be seen that 0.1 mg TP dose induced fluctuations in total protein content of both the lobes with an apparent transitory recovery by 90 minutes interval only. The higher dose of 0.25 mg had a remarkable suppressing influence within first 30 minutes of hormone administration but later on trend of recovery was obvious at all the three intervals studied. It is very remarkable that 0.5 mg dose of TP could induce very significant and consistent suppression of protein content in case of both the lobes of liver (Table 6.1, Fig.6.1).

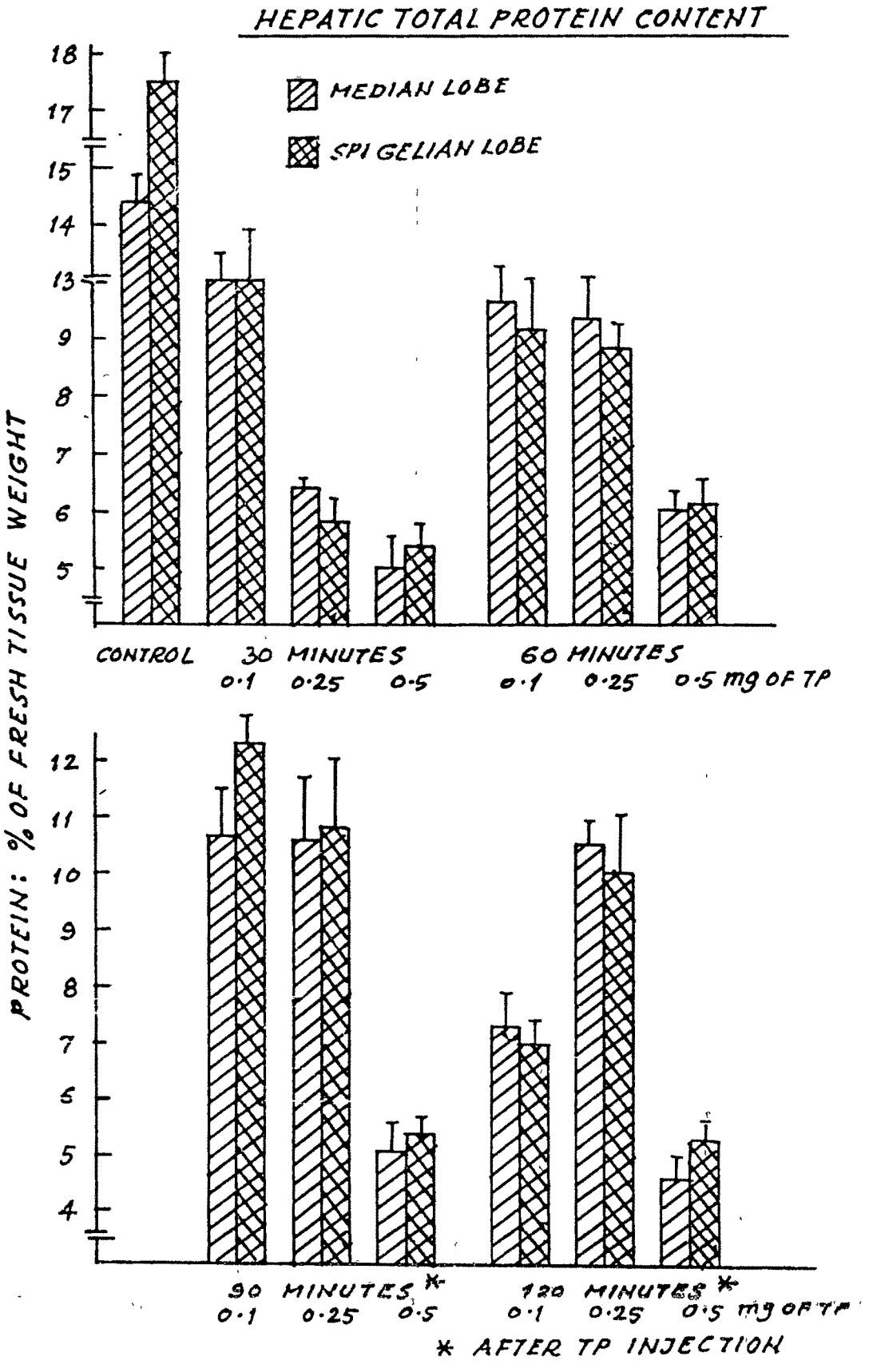
HEPATIC TOTAL PROTEIN CONTENT
MEDIAN LOBE
SPIGELIAN LOBE
PROTEIN: % OF FRESH TISSUE WEIGHT
18
17
15
14
13
9
8
7
6
5
CONTROL
30 MINUTES
0·1 0·25 0·5
60 MINUTES
0·1 0·25 0·5 mg OF TP
12
11
10
9
8
7
6
5
4
90 MINUTES *
0·1 0·25 0·5
120 MINUTES *
0·1 0·25 0·5 mg OF TP
* AFTER TP INJECTION

Table 6.1 : Immediate influence of exogenous TP administration to white rats, with respect to hepatic protein content.

Post operative intervals	Normal animals administered with TP					
	0.1 mg		0.25 mg		0.5 mg	
	M	SP	M	SP	M	SP
30 minutes	12.95* ± 0.5	13.01***** ± 0.9	6.42***** ± 0.1	5.85***** ±0.4	5.08***** ±0.6	5.44***** ±0.49
60 minutes	9.65***** ±0.8	9.12***** ± 1.3	9.33***** ±0.7	8.84***** ±0.4	6.05***** ±0.3	6.17***** ±0.5
90 minutes	10.78***** ± 0.9	12.33***** ± 0.6	10.65***** ± 1.2	10.89***** ±1.3	5.14***** ±0.5	5.43***** ±0.36
120 minutes	7.32***** ± 0.60	6.97***** ±0.41	10.52***** ±0.39	9.99***** ±1.0	4.61***** ±0.4	5.35***** ±0.3

Normal hepatic protein content expressed as % of fresh tissue weight

M (Median lobe)	Sp (Spigelian lobe)
14.41 ± 0.5	17.55 ± 0.8

* $P < .05$ ** $P < .02$ *** $P < .01$ **** $P < .001$ ***** $P < .0005$

± S.E.M. of at least eight animals.

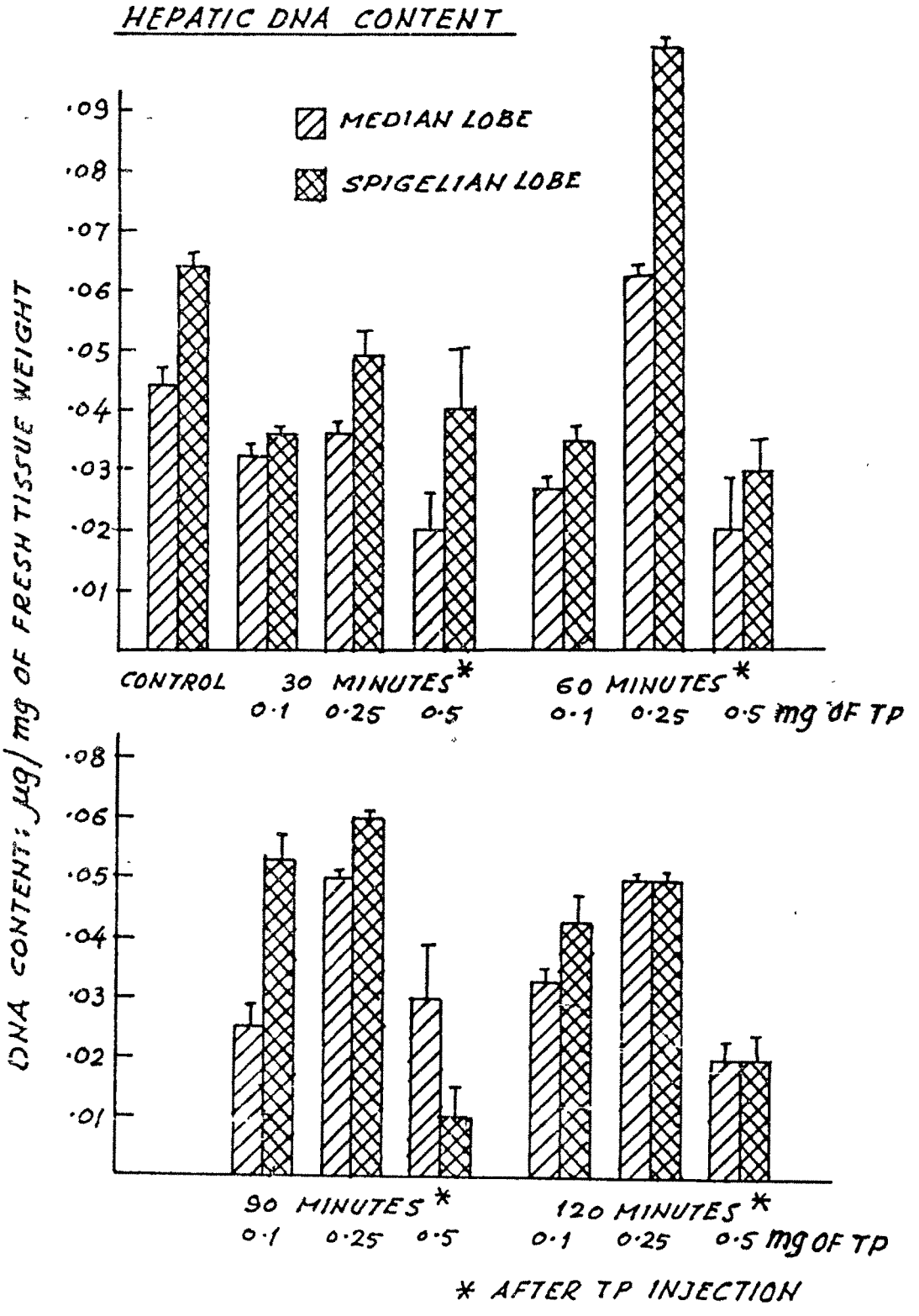
HEPATIC DNA CONTENT
MEDIAN LOBE
SPIGELIAN LOBE
DNA CONTENT; µg/ mg OF FRESH TISSUE WEIGHT
·09
·08
·07
·06
·05
·04
·03
·02
·01
CONTROL
30 MINUTES*
0·1 0·25 0·5
60 MINUTES*
0·1 0·25 0·5 mg OF TP
90 MINUTES*
0·1 0·25 0·5
120 MINUTES*
0·1 0·25 0·5 mg OF TP
* AFTER TP INJECTION

Table 6.2 : Immediate influence of exogenous TP administration to white rats, with respect to hepatic DNA content

Post operative intervals	Normal animals administered with TP					
	0.1 mg		0.25 mg		0.5 mg	
	M	SP	M	SP	M	SP
30 minutes	***** 0.030 ± .001	***** 0.036 ± .001	***** 0.036 ± .002	* 0.049 ± .004	***** 0.02 ± .006	* 0.04 ± .01
60 minutes	***** 0.027 ± .002	0.035 ± .002	**** 0.062 ± .02	***** 0.10 ± .01	***** 0.02 ± .005	***** 0.03 ± .006
90 minutes	***** 0.025 ± .004	*** 0.053 ± .004	**** 0.05 ± .01	0.06 ± .01	***** 0.03 ± .009	***** 0.01 ± .005
120 minutes	***** 0.033 ± .002	* 0.043 ± .004	**** 0.05 ± .004	*** 0.05 ± .003	***** 0.02 ± .003	***** 0.02 ± .004

Normal DNA content of the hepatic tissue expressed as g/mg of fresh tissue weight

M (Median lobe) 0.044 ± 0.003

Sp (Spigelian lobe) 0.064 ± 0.002

* P < .05 ** P < .02 *** P < .01 **** P < .001 ***** P < .0005

± S.E.M. of at least eight animals.

HEPTIC RNA CONTENT

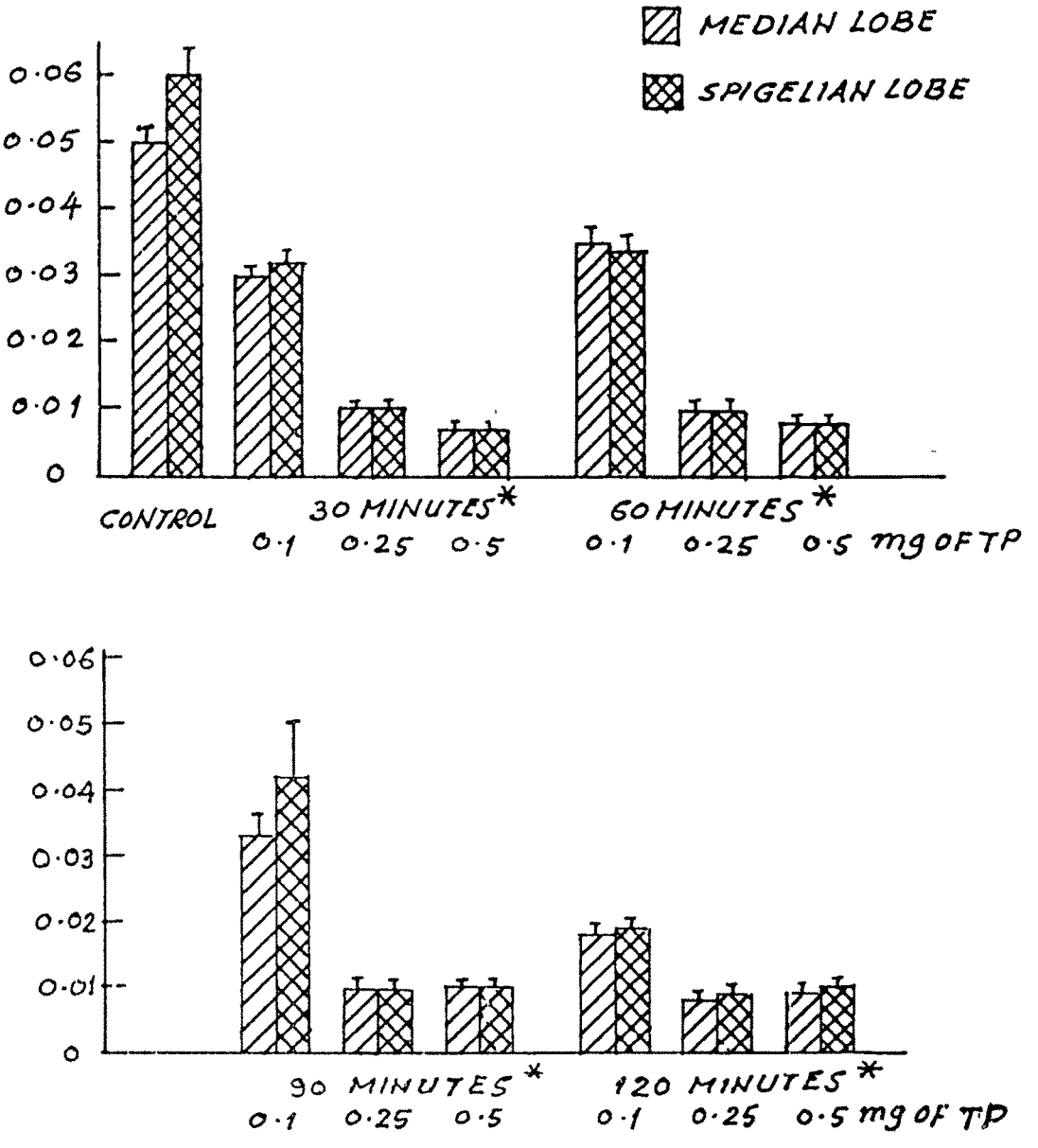

Table 6.3 : Immediate influence of exogenous TP administration to white rats, with respect to hepatic RNA content

Post operative intervals	Normal animals administered with TP					
	0.1 mg		0.25 mg		0.5 mg	
	M	SP	M	SP	M	SP
30minutes	***** 0.03 ± .001	0.060 ± .004	***** 0.01 ± .001	***** 0.01 ± .001	***** 0.007 ± .001	***** 0.007 ± .001
60 minutes	***** 0.035 ± .002	***** 0.033 ± .001	***** 0.01 ± .001	***** 0.01 ± .002	***** 0.008 ± .0007	***** 0.008 ± .002
90 minutes	***** 0.033 ± .003	***** 0.034 ± .002	***** 0.01 ± .001	***** 0.01 ± .002	***** 0.01 ± .002	***** 0.01 ± .002
120 minutes	***** 0.018 ± .001	***** 0.019 ± .002	***** 0.008 ± .0006	***** 0.009 ± .0003	***** 0.009 ± .0003	***** 0.01 ± .001

Normal RNA content of hepatic tissue expressed as g/mg of fresh tissue weight

M (Median lobe) 0.05 ± .002

Sp (Spigelian lobe) 0.06 ± .004

* P < .05 ** P < .02 *** P < .01 **** P < .001 ***** P < .0005

± S.E.M. of at least eight animals.

DISCUSSION :

At the very onset, it could be seen from the results presented here that, in general, administration of TP to normal intact animals led to varying degrees of depletion of protein content in both the hepatic lobes. Further, it could be seen that the first two dose levels (0.1 mg and 0.25 mg) apparently permitted some staggering effect on depletion of protein levels but that of 0.5 mg dose led to a consistantly reduced level of hepatic protein. There was a simultaneous and concommitant fall in the levels of DNA as well as RNA, that of latter being more intense.

Here it would be necessary to add that similar very early influences of hormone administration on protein and nucleic acid contents were obtained under various experimental conditions in respect of different tissues by several workers (Kosto et al., 1967; Fu-Liyu and Felgelson, 1971; Siboyadjiev and Hadjlolov, 1971; Sidransky, 1982; Mukku et al., 1982; Ambadkar et al., 1987).

Interdependence of patterns of nucleic acid metabolism and those of protein metabolism is a well known fact (Kosto et al., 1967; Kurtz et al., 1976; Minguell and Sierralta, 1975; Bryson et al., 1965; Malt, 1967; Blakley and Vitols, 1968). From the results presented here it could be seen that reduced levels of RNA and DNA could

logically lead to corresponding reduction in the rate of protein synthesis. On the other hand, increased levels of such enzymic activities as those of GOT and GPT (Chapter 5), which are known to be present in wasting conditions of mammalian liver (Myron Johnson, 1974), would lead to protein degradation. The combined action of these two factors are probably responsible for reduction in hepatic tissue protein content.

Androgens are normally known to exhert an overall anabolic influence. It is clear from the work of Kosto et al. (1969) that prolonged injections of androgenic steroids to castrates gradually lead to a "wearing off" of anabolic influence and reduction of growth in the accessory sex glands. Lesser and Bruchosky (1973) have also reported that continued injections of dihydrotestosterone (DHT) to castrated rats initially lead to reversal of castration effects but, beyond 5 days the rate of cell proliferation fell and the rate of DNA synthesis also declined; reaching almost negligible levels.

From the work of Aschkenasy-Lelu and Aschkenasy (1959) it is apparent that not only temporal effects are of significance but dose levels also tell upon differences in hormone action. These authors, working on the influence of oestrogen on uterine protein metabolism, have proved

that whereas low dose levels of oestrogen exert a slightly anabolic action while higher doses exhibit distinct catabolic influence.

Literature cited above mostly dealt with effects of hormone administration on protein synthesis in castrated animals, wherein initial reversal effects are self-explanatory but the later effects are in all probability related to regained normalcy of the concerned tissues which were then subjected to further influence of steroid hormones. During the course of present investigation intact animals were subjected to hormone treatment and hence the effects are probably comparable to the later conditions reported in the works cited above. It is, therefore, likely that exogenously administered hormone leads to higher titres in circulation and this immediately leads to the unexpected results reported here. Hence, it could be suggested that early influence of TP administration to intact animals basically differs from that obtainable under various other conditions - temporally as well as dose-wise.