

## Histological structure and hormonal profile of pituitary and thyroid glands affected by castration and iodine supplementation in male rabbits

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### ABSTRACT

This study aims to evaluate the effects of castration and iodine supplementation on the histological structure of pituitary and thyroid glands and their related hormones in NZW male rabbits. Animals were randomly divided into two groups. The first group was supplemented with iodine as potassium iodide at a level of 400 ppm, while the other was control. Each group was divided into two subgroups; the first subgroup was bilaterally castrated (BC) after testicles descended using banding technique, while the other served as an intact (INT) control. Plasma luteinizing hormone (LH) concentrations were increased ( $P < 0.01$ ) in BC compared to INT rabbits, with or without iodine supplementation. Plasma T<sub>3</sub> and T<sub>4</sub> levels were lower ( $P < 0.01$ ) in BC than in INT rabbits, while they were higher ( $P < 0.01$ ) with iodine in BC rabbits than in INT ones. Histological changes observed in the pituitary were as follows: Castration alone induced an increase in basophil number and a reduction in the number of acidophils and chromophobe cells. Many of these cells in BC rabbits had a ring of clear cytoplasm around their nuclei and their pituitaries showed increased vascularisation. In INT rabbits with iodine, basophilicity increased, showing presence of polygonal hypertrophied basophils. In BC rabbits with iodine, the number of basophils of large-sized was greatly increased. Histological alterations in the thyroid were as follows: In BC rabbits, there was a remarkable increase in fibroblasts. Follicular cells appeared less closely packed to each other and their epithelium was cuboidal. The number of parafollicular cells, however, did not change in BC sections. The colloid appeared less homogenous in BC than in INT sections. In INT rabbits with iodine, there was a marked change in follicular cell shape, which was converted to squamous epithelium. They have a marked depletion in parafollicular cells and colloid substance. In BC rabbits with iodine, the follicular epithelium was flattened, resembling that of the INT with iodine, and there was a severe depletion in parafollicular cells and colloid.

**KEYWORDS:** castration, iodine, pituitary and thyroid hormones, histology, rabbits

### INTRODUCTION

Recent studies on male rabbits (Toson & Soliman 2001) and male goats (El-Barordy *et al.* 1996) indicated that castration could be used to improve growth performances such as weight gain, feed conversion rate and carcass traits, without impairing physiological functions in male goats and rabbits. These improvements in rabbits were accompanied by a modification in thyroid activity and secretion of metabolic hormones (Toson & Soliman 2000). Moreover, castration has a profound effect on the appearance and number of acidophils and basophils of pituitary cells and subsequently affects pituitary hormone secretion (Nalbandov 1976). Many studies have shown that the gonadotrophic potency of the pituitary is higher in castrated than in intact animals (Pakarinen & Huhtaniemi 1992; Watanabe *et al.* 1998).

On the other hand, iodine plays an important role in thyroid physiology, resulting from its importance as a requisite substrate for the synthesis of thyroid hormones and from its action as a regulator of thyroid function (Saller *et al.* 1998). Iodine supplementation, at a level of 400 ppm as potassium iodide, was reported to improve productive and reproductive performance of rabbits without harmful effects (Zaki El-Din 1996). Thyroid hormones play an important role in the regulation of pituitary function (Zhu *et al.* 1997) and administration of these hormones increases the gonadotrophic potency of pituitary gland in rats, while

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thyroidectomy decreases the gonadotrophic potency of pituitaries in rats, rabbits and goats (Nalbandov 1976). In male rabbits, the effects of castration and iodine supplementation on some histological and endocrine functions of pituitary and thyroid glands have not been fully clarified. In a recent report on rabbits, Toson & Soliman (2001) showed that castration, but not iodine supplementation, could be used to improve growth performance. It was therefore of interest to study the influences of castration and iodine on the histological structure of pituitary and thyroid glands and to evaluate their related hormone secretion profiles.

## MATERIALS AND METHODS

The present study was carried out at the experimental farm of the Animal Production Department, faculty of Agriculture, Minia University, from December 1998 to March 1999. Twenty four New Zealand White (NZW) male rabbits were used after testicles descended at approximately 12 weeks of age. The animals were individually weighed and divided randomly into two equal groups of 12 males each. Iodine, as potassium iodide (KI), was added to the drinking water at a level of 400 ppm for the first group, while the second group was not supplemented. Each group was randomly divided into two subgroups, the first subgroup served as intact control (INT) and the other one was bilaterally castrated (BC) using banding procedure, as previously described by Toson & Soliman (2000) according to Chase *et al.* (1995). Animals were individually housed in wire cages and fed ad lib. on a commercial pelleted diet containing 17.0 % crude protein, 14 % crude fibre and 2600 ME/Kg. The experiment was conducted under winter conditions and continued for 10 weeks post-treatment. At the end of the experimental period, three animals from each subgroup were randomly chosen, weighed and slaughtered after 12 hours of fasting. Blood samples were collected during slaughter from each animal into heparinized tubes and plasma samples were obtained by centrifugation at 3000 r.p.m for 10 min, and stored at -20°C until assayed for hormonal analysis. Plasma luteinizing hormone (LH), triiodothyronine (T<sub>3</sub>) and thyroxine (T<sub>4</sub>) concentrations were determined using a direct solid-phase <sup>125</sup>I radioimmunoassay technique using (Coat-A-Count) RIA kits purchased from Diagnostic Products Corporation (DPC, Los Angeles, CA, 90045-5597, USA). In addition, T<sub>4</sub>/T<sub>3</sub> ratio was calculated. Immediately after slaughter, thyroid and pituitary glands were removed from each animal of the different experimental subgroups and fixed in Bouin's, as well as formal saline, fixatives. They were then embedded in parablax, sectioned at 5 µm, mounted on microscopic slides and stained with haematoxylin and eosin (H & E), periodic acid Schiff's (PAS) and Azan stains.

The data of hormonal assay were statistically analyzed by 2-way factorial ANOVA design using General Linear Model (GLM) of the statistical analysis system (SAS, 1992). The data were analysed using the following model :

$$Y_{ijk} = \mu + C_i + T_j + (CT)_{ij} + e_{ijk}$$

where :  $Y_{ijk}$  =  $ijk^{\text{th}}$  observation of the trait;  $\mu$  = general mean;  $C_i$  = fixed effect of  $i^{\text{th}}$  castration treatments (1=intact and 2=castrated),  $T_j$  = fixed effect of the  $j^{\text{th}}$  iodine treatment (1=non-treated and 2=treated);  $CT_{ij}$ =effect of interaction ( $ij^{\text{th}}$ ) between castration and iodine treatments;  $e_{ijk}$ =random error of the  $ijk^{\text{th}}$ . Significant differences among means of experimental subgroups were detected using Duncan's Multiple Range test (Duncan 1955).

## RESULTS

**Pituitary hormone (LH, ng/ml):** Data of LH assay, presented in Table 1, indicated that the plasma concentrations of luteinizing hormone (LH) were significantly ( $P<0.01$ ) increased by 144 % in BC rabbits compared to INT ones. With iodine supplementation, the LH levels were also significantly ( $P<0.01$ ) elevated by 98 % in BC rabbits when compared with INT males.

LH levels were not significantly different between iodine INT and non-iodine INT rabbits. Levels of LH showed a significant ( $P<0.05$ ) decrease in iodine BC, compared with non-iodine BC, rabbits. Data presented in Table 2, show that the main effect of castration on LH was significant ( $P<0.01$ ), while the main effect of iodine and the interaction effect of castration and iodine on LH were not significant. Thyroid hormones ( $T_3$  and  $T_4$ , ng/ml): Plasma concentrations of triiodothyronine ( $T_3$ ) and thyroxine ( $T_4$ ) were significantly ( $P<0.01$ ) higher in INT than in BC rabbits (Table 1). There were no significant differences in  $T_4/T_3$  ratio between INT and BC rabbits. With iodine supplementation, in contrast, the concentrations of  $T_3$  and  $T_4$  as well as  $T_4/T_3$  ratio were significantly ( $P<0.01$ ) increased in BC as compared with INT rabbits. Levels of  $T_3$  and  $T_4$ , as well as  $T_4/T_3$  ratio, were significantly higher ( $P<0.01$ ) in non-iodine INT than in iodine INT rabbits. Meanwhile,  $T_3$  and  $T_4$  showed higher ( $P<0.05$ ) levels in iodine BC than in non-iodine BC rabbits.

Data, in Table 2, show that the main effect of castration on  $T_3$  was not significant, while it was significant ( $P<0.05$ ) related to  $T_4$  and  $T_4/T_3$  ratio. The main effect of iodine was not significant on  $T_3$  and  $T_4/T_3$  ratio while it was significant ( $P<0.01$ ) related to  $T_4$ . The interaction effect of castration and iodine treatment was significant ( $P<0.01$ ) on  $T_3$  and  $T_4$ . But, it was not significant on  $T_4/T_3$  ratio.

Items	Thyroid hormones			Pituitary hormone
	$T_3$ (ng/ml)	$T_4$ (ng/ml)	$T_4/T_3$ ratio	LH (ng/ml)
Non-iodine treat.				
Intact	2.48±0.05 a	133.3±3.60 a	53.84±0.98 a	8.53±0.89 cd
Castrated	1.87±0.03 c	105.0±1.20 b	56.13±1.49 a	20.76±0.98 a
Iodine treat.				
Intact	1.42±0.01 d	57.33±1.78 c	40.35±0.94 b	7.76±0.48 d
Castrated	2.23±0.04 b	128.0±1.88 a	57.51±0.60 a	15.37±1.23 b

Table 1: The effects of castration and iodine supplementation on thyroid hormones ( $T_3$  and  $T_4$ ) and pituitary LH secretions in NZW male rabbits (means ± S.E). <sup>a,b,c & d</sup> Means within the same column having different superscripts are significantly different ( $P<0.01$ ).

S.O.V	d.f	$T_3$	$T_4$	$T_4/T_3$ ratio	LH
		F value	F value	F value	F value
Castration treat.	1	0.28 NS	8.02 *	5.80 *	37.39 **
Iodine treat.	1	2.98 NS	12.75 **	2.08 NS	3.61 NS
Interaction	1	11.7 **	43.87 **	4.34 NS	2.04 NS

Table (2) : Analysis of variance (ANOVA) for the data of table1.

\* ( $P<0.05$ ), \*\* ( $P<0.01$ ), NS=not significant.

**Histological structure of pituitary gland:** The pituitary lies in a depression of the sphenoid bone at the base of the skull, the sella turcica and is attached to the base of the brain by a slender stalk. The gland is divided into two major parts, adenohypophysis (glandular portion) and neurohypophysis (neural portion). The adenohypophysis is further subdivided into pars distalis (anterior lobe), pars tuberalis and pars intermedia. The pars distalis is the largest part of the hypophysis and it is composed of two main cell types, chromophobes and chromophils according to their affinities for histological dyes (Fig. 1). Both chromophobes and chromophils are arranged in the form of anastomosing cords and are surrounded by a rich network of sinusoidal capillaries (Fig. 2). Chromophobe cells are the smallest cell type in the pars distalis. They have little affinity to either basic or acidic dyes and their cell boundaries are poorly defined (Figs. 1 & 2). Chromophils are subdivided on the basis of their staining properties into acidophils (alpha cells) and basophils (Beta cells). Acidophils take up acidic stains and are identified easily in either hematoxylin and eosin or Azan preparations. They are larger than chromophobes, their cell boundaries are well defined (Fig. 1) and their cytoplasm is crowded with granules (Fig. 1). Basophils (Beta cells) appear larger than acidophils. Their cytoplasmic granules are less numerous. They stain poorly with hematoxylin and eosin but are stained somewhat deeply blue with Azan (Figs. 1 & 2).

In all mammalian species, different types of acidophils and basophils are distinguished by selective staining methods and by immunocytochemistry using antibodies to specific hormones. There are two distinct types of acidophil; somatotrophs and mammotrophs. Three types of basophil are defined. These are thyrotrophs, gonadotrophs and corticotrophs. In the present study, however, the different types of acidophils and basophils were not distinguishable in the untreated animals with any of the stains used. Non-iodine BC rabbits showed an increased number of basophils, accompanied by a reduction in the number of acidophil and chromophobe cells (Fig. 4). These basophils appeared larger in size compared with the basophilic cells of the control group, and their cytoplasm was somewhat vacuolated (castration cells) (Figs. 3 & 4). Many of these cells had a ring of clear cytoplasm around their nuclei (Fig. 5 & 6). The gland in these animals also showed an increase in its vascularization (Fig. 4).

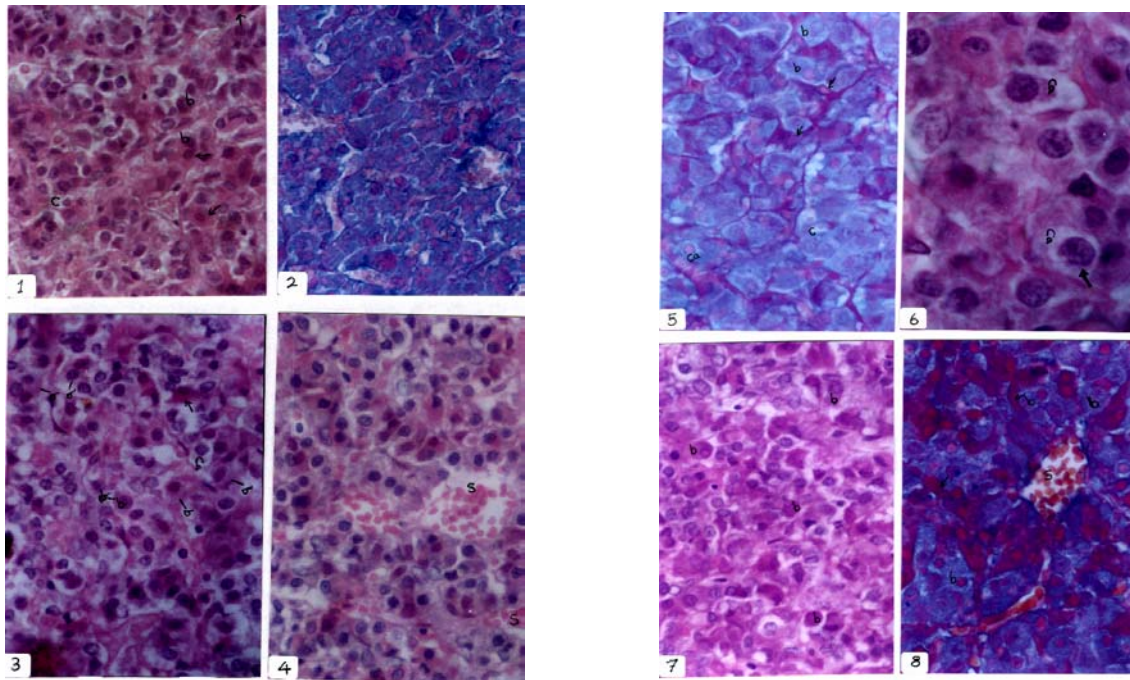


Fig. 5: Another section through the pituitary gland of BC rabbits showing beta cells (b), some of which have a ring of clear cytoplasm around the nucleus (castrated cells, ca, alpha cells (arrows), and chromophobes (c). (Azan, X 500).

Fig. 6: Magnified portion of section in BC rabbits showing castration cells (ca) with a ring of clear cytoplasm (thick arrow) (H&E, X 1250).

Fig. 7: Section through pituitary of intact rabbits supplemented with iodine showing hypertrophied basophils (b). (H&E, X 500).

Fig. 8: Another section through pituitary gland of intact-iodine treated rabbits showing dilated sinusoids (s) congested with red blood cells. Note the hypertrophy of basophils (b), and the considerable increased number of alpha cells (arrows). (Azan, X 500).

In iodine-INT rabbits, the pituitary showed increased basophilicity with the presence of hypertrophied basophils which were polygonal in shape (Fig. 7). The cytoplasm of these cells was filled with colloid substance (Figs. 7, 8 & 9). It was also observed that many basophilic

cells were enriched with granulated cytoplasm. Such granulation may represent different proliferated developing thyrotrophes (Figs. 9 & 10). The vascularization was prominent, with dilatation of congested sinusoidal capillaries as noted in castrated rabbits (Fig. 8). In iodine-BC rabbits, the number of large-sized basophils was greatly increased, showing different degrees of cytoplasmic basophilic affinity (Figs. 11 & 12).

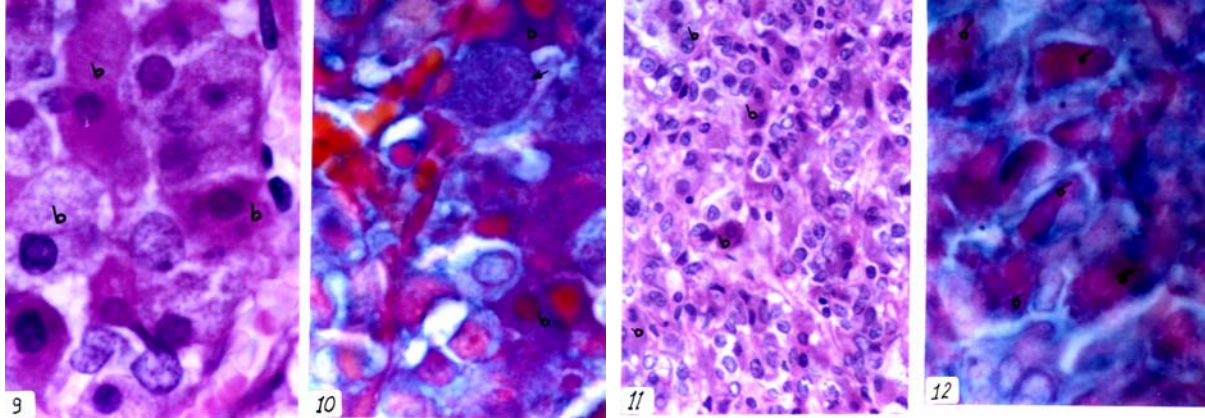


Fig. 9: Magnified portion of Fig. (8) demonstrating hypertrophy of beta cells (b).(H&E, X 1250).

Fig. 10: Another magnified sector of pituitary of intact-iodine treated rabbits. Note the presence of basophils (b), some of which with granulated cytoplasm (arrow).(Azan, X 1250).

Fig. 11: Section through pituitary gland of the BC rabbits supplemented with iodine showing the increased number of basophils (b) with different degrees of basophilic affinity. (H&E, X 500).

Fig. 12: Section of pituitary gland of BC rabbits supplemented with iodine. Note the dominance of basophilic cells (b). (Azan, X 1250).

**Histological structure of thyroid gland:** The gland consists of two lobes joined by a thin band of tissue, the isthmus. The gland is closely attached to the anterior and lateral aspect of the trachea by loose connective tissue. In hematoxylin and eosin preparations, the gland appears to be enclosed in a connective tissue capsule which penetrates it forming trabeculae. The gland is composed of closely packed follicles, which vary considerably in size, and are surrounded by a rich capillary network. The walls of the follicles consist of one layer of simple cuboidal epithelium (follicular cells) which rests upon a thin basement membrane (Fig. 13). The lumen of the follicles contains a clear gelatinous material (colloid) which stains positively with PAS (Figs. 13 & 17). In addition to the follicular cells, the gland contained a small population of parafollicular cells, or C cells. In sections stained with hematoxylin and eosin, these cells appeared polygonal with a granular, weakly eosinophilic cytoplasm, which is paler and larger than that of the follicular cells. They are located individually or in small groups, both within the follicular wall and in the interfollicular spaces. When found in the follicular wall, they are situated immediately adjacent to the basement membrane and without contact with the follicular lumen (Fig. 13). In non-iodine BC rabbits, the follicular cells appeared less closely packed to each other with cuboidal epithelium where some fibroblasts detected inbetween (Fig. 14). In addition, there were obvious differences in the affinity toward staining of the colloid with both H & E and PAS stains, in different follicles and even in the same follicle. Sometimes, different areas of the colloid in the same follicle showed obvious cracking as well as some peripheral vacuolation (Figs. 14 & 18). Such variation was not observed in INT sections in which the colloid appeared more homogenous. On the other hand, the number of parafollicular cells did not change in BC sections. In iodine-INT rabbits, there was a marked change in the shape of the follicular cells. The follicular epithelium was flattened, converted to squamous epithelium (Figs. 15). There was a marked depletion of parafollicular cells and colloid substance. The variation in the density of the staining of

colloid was greatly obvious when compared with the castrated rabbits (Figs. 15 & 19). In iodine-BC rabbits, the follicular epithelium was flattened, resembling that of the iodine treated animals (Fig. 16). There was a severe depletion in the parafollicular cells and the colloid showed a slight improved picture although the cracked appearance was still observed (Figs. 16 & 20). The variation in the stain affinity of the colloid was obvious, especially in section stained with PAS (Figs. 16 & 20).

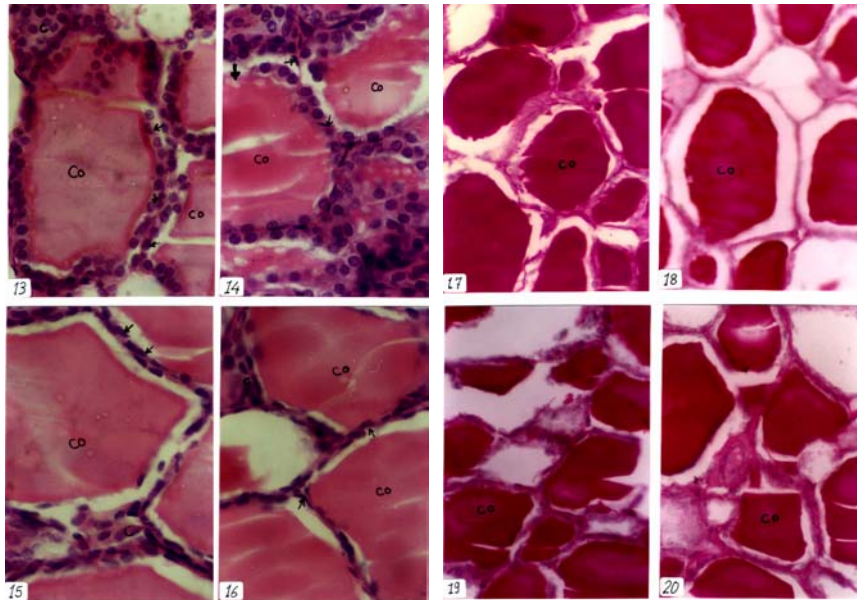


Fig. 17: Section of thyroid gland of INT rabbits showing the intensity and homogenous staining of the colloid (co). (PAS, X 312.5).

Fig. 18: Section of thyroid gland of BC rabbits showing the non-homogenous staining and less intensity of colloid (co). (PAS, X 312.5).

Fig. 19: Section through thyroid gland of iodine INT rabbits showing the depletion and less homogeneity of colloid (co). (PAS, X 312.5).

Fig. 20: Section of thyroid gland of iodine BC rabbits showing the slight improved intensity of colloid staining (co) (PAS X 312.5)

Fig. 13: Section through the thyroid gland of INT rabbits showing cuboidal follicular cells (arrows), and parafollicular cells (c). Note that follicles are filled with colloid (co). (H&E, X 500).

Fig. 14: Section of the thyroid gland of BC rabbits showing follicular cells which are not closely packed to each other, fibroblasts (thin arrows), less intensity of colloid which shows cracked picture (co), and peripheral vacuolation (thick arrows).(H&E, X 500).

Fig. 15: Section through thyroid gland of iodine INT rabbits showing remarkable change of the shape of follicular cells which became squamous (arrows). Note the marked depletion of parafollicular cells (c), and colloid substance (co). (H&E, X 500).

Fig. 16: Section through thyroid gland of iodine BC rabbits showing severe depletion of interfollicular cells (c) and the slight improved intensity of colloid substance (co). The majority of follicular cells have the squamous appearance (arrows).(H&E, X 500).

## DISCUSSION

The present study demonstrated that bilateral castration (BC) of male rabbits with or without iodine supplementation, induced a significant increase in pituitary gonadotrophic secretion of luteinizing hormone (LH). This observation in rabbits coincides with previous reports in which castrated animals exhibited higher levels of LH than intact ones of many species including mice (Lindzey *et al.* 1998); rats (Watanabe *et al.* 1993); and goats (Kanematsu 1990). Thus, the gonadotrophic potency of the pituitary is always higher in castrated than in intact animals (Nalbandov 1976). As far as pituitary gonadotrophic potency is concerned, the present study showed that plasma LH levels were increased by 144 % in non-iodine BC rabbits, and by 98 % in iodine BC rabbits, at 10 weeks post-castration. These results are in accordance with earlier findings that serum levels of LH in male rats increased 6-8 fold after one week of castration (Pakarinen & Huhtaniemi 1992). Such enhanced LH release in BC rabbits seen in the present work could be explained by the observation that the serum LH is released in a tonic or basal fashion in both female and male animals. These tonic levels of LH

are controlled by a negative feedback mechanism from the testes. Therefore, the increased tonic serum LH concentrations in male after castration are due to the lack of a negative feedback mechanism from the testicular steroids on the tonic LH control centre in the hypothalamus (Hafez 1987). However, it was noticeable that such gonadotrophic potency was decreased for iodine BC compared with those of non-iodine BC rabbits. This observation may reflect an interactive effect of castration plus iodine on pituitary activity.

The levels of plasma LH were positively modulated in BC rabbits, with or without iodine treatment. This result may indicate a change in rhythmic secretion of hypothalamic LH-releasing hormone (LHRH) which acts as a neuromodulator to control LH release from adenohypophysis. The mechanisms by which LH and LHRH are released, affected by castration, could be linked to an excitatory noradrenergic synapse which may mediate the increased LHRH following castration, and the evidence for this case is the ability of alpha-adrenergic receptor blocking drugs to prevent the post-castration type of LH release (McCane 1980). In addition, the pituitary sensitivity to LHRH has increased in the absence of gonadal steroids after castration (Kittok *et al.* 1989). Therefore, although the LHRH levels have not been altered with castration, the frequency of LHRH pulses were moderately increased in castrated, rather than in intact rats (Levine & Duffy 1988). However, some evidences indicated that pituitary fragments of castrated bulls released LH synchronously regardless of whether pituitaries were perfused with or without the hypothalamus (Kanematsu 1990). Additionally, there was no consistent change detected in the amount of LHRH resulting from perfused pituitary of castrated rats (Phelps *et al.* 1992). Therefore, it was suggested that most of the increase in serum LH after castration is not mediated at the hypothalamic level as reported by Emanuele *et al.* (1996) who noticed that castration of male rats induces a prompt increase in serum LH levels and a concomitant rise in steady state levels of the mRNAs directing its synthesis. Taken together, the role of castration on the actual delivery of LHRH to the anterior pituitary has not been addressed yet.

Histological examinations in the present study showed that castration induced considerable structural changes in the pituitary gland. BC rabbits showed an increase in basophil number, which was accompanied by a decrease in acidophil (somatotrophs and lactotrophs). The basophils appeared hypertrophied and many of them had a ring of clear cytoplasm around their nuclei (Fig. 6). In agreement with these observations, Fawcett (1986) stated that after castration, the rat hypophysis contains increased amounts of gonadotropic hormones, and basophilic cells became markedly enlarged and vacuolated in a characteristic way (castration cells), which are then called signet-ring cells because of the eccentric nucleus and the large vacuole in the cytoplasm (Nalbandov 1976). The histological changes observed in basophils for BC rabbits may reflect the proliferation and hypertrophy of the gonadotrophs which were accompanied by an increase in their potency to synthesize and release more LH affected by castration. Such results could be clarified by the observation that removal of the target organ (testes) results in hypertrophy of those cells in the adenohypophysis responsible for elaboration of the corresponding tropic hormone (Fawcett 1986). Expansion in the gonadotropic cell area following gonadectomy was found to be correlated with a rise in serum LH (Ibrahim *et al.* 1986). Furthermore, in a study on male rats, gonadotrophs stimulated by castration showed hypertrophy or hyperplasia and plasma LH increased 30-70 fold and the majority of gonadotrophs showed features of castration cells containing many granules (Watanabe *et al.* 1993, 1998).

In the present study, thyroid hormones (T<sub>3</sub> and T<sub>4</sub>) and T<sub>4</sub>/T<sub>3</sub> ratio were significantly reduced in iodine INT rabbits. The effect of iodine on the rate of thyroid hormone synthesis has been shown to depend on the amount and duration of iodine administration (Nagataki, 1974). Subnormal concentrations of T<sub>4</sub> and T<sub>3</sub> in serum denote the presence of thyroid hypofunction. Severe thyroid failure is characteristically associated with decrease in both T<sub>3</sub>

and T<sub>4</sub> concentrations (Ingbar & Woeber 1981); this was the case in the present study indicating that iodine treatment of 400 ppm induced drastic hypothyroidism. Thyroid hormones regulate thyrotrophin (TSH) secretion via a direct negative feedback upon the pituitary thyrotrophs (Hershman & Pekary 1985). More recently, evidence for thyroid hormone inhibition of thyrotrophic releasing hormone (TRH) release has become available (Rondeel *et al.* 1988). The plasma TSH is distinctly elevated in hypothyroidism.

The pituitary of iodine INT rabbits showed increased basophilia with the presence of hypertrophied granulated basophils. Such granulation may represent different proliferated developing thyrotrophs which synthesize and release TSH. In this regard, the pituitary in hypothyroidism may show an increase in the number of actively secreting thyrotrophs as reported by Ingbar & Woeber (1981). The hypertrophied thyrotrophs are easily recognized and are called thyroidectomy cells (Daughaday 1981). Moreover, iodine BC rabbits showed elevated T<sub>3</sub> and T<sub>4</sub> levels as compared with iodine INT ones, reaching approximately the normal values detected in non-iodine INT rabbits. Such results could be due to the interactive effect of castration and iodine treatment on thyroid activity and thus modulating its hormonal secretion rate. The pituitary of iodine BC rabbits exhibited a greatly increased number of basophils with different degrees of basophilic staining affinity which may indicate an increase in TSH secretion. This suggestion may be confirmed by the results of Borges *et al.* (1998) which indicated that castrated old rats have higher concentrations of serum and basal *in vitro* TSH compared with their control. Examination of thyroid histology in BC rabbits showed slight changes, where the colloid appeared less homogenous in BC than in INT sections. In iodine-INT rabbits, the follicular epithelium was flattened converted to squamous and a marked depletion in parafollicular cells and colloid substance was noticed, indicating the hypoactivity of the gland. Also, iodine-BC rabbits resembled to a great extent that of iodine-INT thyroid sections associated with marked depletion of parafollicular cells and colloid. According to Fawcett (1986), the uptake of colloid from the lumen of the follicle has been studied mainly in animals strongly stimulated to release hormone by administration of TSH of the hypophysis. So, the detected depletion of colloid in follicular lumen was presumably due to the increased TSH released by hypertrophied basophils (probably thyrotrophs) detected in the pituitary gland, as well as hypothyroidism resulting from iodine treatment. Meanwhile, the alteration in the thyroid of iodine-INT rabbits paralleled the decrease in their hormonal secretion (T<sub>3</sub> and T<sub>4</sub>) denoting hypothyroidism in these animals. On the other hand, changed thyroid picture in iodine-BC rabbits did not show any obviously improved structure in follicular epithelium, although plasma T<sub>3</sub> and T<sub>4</sub> levels resembled to some extent those of non-iodine INT rabbits. Such elevated levels of thyroid hormones in iodine BC rabbits may explain the slight improvement in colloid appearance (Fig. 16) as compared to non-iodine BC ones (Fig. 14). Similar changes have been reported in individuals treated with iodine prior to surgical resection of the gland (Curran 1985). In these cases, involution is almost complete, and a part of the relatively small follicles close to the fibrous capsule of the gland, the thyroid consists of large follicles lined by flattened epithelium and full of well irregularly stained-colloid. Some peripheral vacuolation of the colloid is however still visible, showing that resorption of the colloid continues, as shown in Figs. (14, 18). It is probable that changes in thyroid hormone profile as a result of castration could be related to indirect mechanisms through pituitary site of action and its secretion of thyrotrophin (TSH) which might be altered and influenced by gonadal manipulation of male rabbits.

Iodine treatment caused hypothyroidism and did not exhibit any improvement in growth performance of male rabbits, corresponding with work by Toson & Soliman (2001) who, in a recent report, used a similar dose of potassium iodide. However, these studies disagree with the results of Zaki El-Din (1996) who recommended such iodine levels to improve productive and reproductive performance using young male Bouscat rabbits at 5 weeks of age. The



differences between the present results and previous study may be due to the different breed and age of experimental rabbits used and/or interactions with environmental conditions and physiological responses of animals.

Conclusively, the present study showed that castration induced an increase in gonadotrophic potency for BC rabbits, with or without iodine supplementation, since they exhibited higher levels of LH than the untreated rabbits. This enhanced LH release in BC rabbits was accompanied by a marked increase in basophils number which may denote hypertrophy of gonadotrophs. The increase in gonadotrophic potency of BC rabbits seen in this study could be explained by the lack of the inhibiting effect of testicular androgens on pituitary function, which are removed by castration. The mammalian reproductive function is under control of the integrate hypothalamic-pituitary gonadal axis (HPG), and the utilization of castration may be an effective tool to investigate hormonal interactions in the mammalian HPG (Shi *et al.* 1998). On the other hand, additional information would be helpful regarding the optimal amount of iodine supplementation, that avoids its negative effects on thyroid status and growth performance in male rabbits.

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