#### CHAPTER XI

# SEASONAL ALTERATIONS IN TISSUE AND SERUM IONIC CONTENTS IN NORMAL AND ADRENAL MANIPULATED PIGEONS, COLUMBA LIVIA

Importance of cations in several physiological functions of tissues and cells is an accepted fact in biolical dictum and the divalent cation (ca<sup>++</sup>) has been found to play important roles in tissue functions and therefore has been documented well. Whereas Na<sup>+</sup> is the major constituent of extracellyr fluid, K<sup>+</sup> forms the major constituent of intracellar fluid. Ca<sup>++</sup> has been found to be essential as a coupling factor between excitation and contraction in muscle cells and between stimulus and secretion in various neuroendocrine secretory processes (Hasselback and Mackinose, 1963; Hasselback, 1964; Weber, et al., 1966; Weber, 1966). Apart from these, they are also known to be associated with a number of enzymic reactions. Worth mentioning is the activity of adenosine - triphosphatase (ATPase): Na<sup>+</sup>-K<sup>+</sup> dependant, Ca<sup>++</sup> dependant, and Mg<sup>++</sup> dependant have all been reported in different tissues (Ebashi et al., 1965; Diel and Jones, 1966; Colombo and Marcus, 1973). These ions are also known to effect the water balance in tissues which in turn influences tissue functioning.

In the seasonal breeders the requirement for metabolites do vary according to the reproductive status of the gonads. This has been found during the present course of study where metabolites and enzymes have been observed to undergo season specific alterations (Previous Chapters). Changes in plasma Ca<sup>++</sup>, Phosphorus, lipids and estrogens have been reported to undergo alterations with reference to the reproductive state of turkeys (Wayne <u>et al.</u>,1980). Plasma haematokrit values, protein(), and Na<sup>+</sup> and K<sup>+</sup>, have all been observed to undergo alterations during oviposition  $\frac{the}{in_{L}}$  domestic hen (Rzasa <u>et al.</u>,1982). These observations prompted the present study to evaluate the season specific alterations in Na<sup>+</sup> and K<sup>+</sup> in gonads and serum along with the water content of gonads. To have a general idea regarding the role of adrenals if any, this study was also extended to the adrenal manipulated experimental group of birds (\_\_\_\_\_) on a seasonal basis.

## MATERIALS AND METHODS

As Outlined in Chapter 1

#### RESULTS

Results obtained are  $\bigcirc$  depicted in tables and figures (1-5 and 1-5). The tables and figures represent cumulative values of testis and ovary put together as no remarkable sex difference in gonadal ionic concentration was discrenible.

## Changes in Normal Birds

Tissue and serum Na<sup>+</sup> contents have in general been

found to strike a reciprocal relation. Gonads exhibited high Na<sup>+</sup> content during the non-breeding phase and least during the recrudescent phase, while the serum Na<sup>+</sup> content was maximum during breeding and least during non-breeding. Contrary to this, gonadal and serum K<sup>+</sup> content showed parallel changes with the highest recorded values being during the breeding season and the least during the non-breeding phase. However tissue K<sup>+</sup> content was found to be more than that of Na<sup>+</sup> while the reverse was true for serum.

On a percentage basis gonad Na<sup>+</sup> content exhibited an increase of 122% from breeding to nonbreeding and a decrease of 71% from breeding to recrudescence. On the contrary, K<sup>+</sup> content exhibited a reverse change in the form of reduction of about 62% and an increase of about 38% from breeding to non-breeding and from non-breeding to recrudescence respectively. Serum Na<sup>+</sup> content too exhibited tremendous season specific alterations. It depicted a decrease of about 65% from breeding to non-breeding and a build up, 55% from the non-breeding to the recrudescent phase. Serum K<sup>+</sup> content also exhibited a similar change and the alterations were more pronounced. Accordingly there was about 18% decrease in serum K\* content from breeding to non-breeding and an increase of about 206% from non-breeding to recrudescence. Water content of the gonads was higher during recrudescence and breeding and reduced during non-breeding.

DRY A
(mg/gm LIPID FREE AL PIGEONS, C.LIVI
OF GONADAL IN NORMAL
SEASONAL ALTERATIONS OF GONADAL SODIUM CONTENT TISSUE WEIGHT <u>+</u> S.D.) IN NORMAL AND EXPERIMENT,
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TABLE-1

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REPRODUCTIVE	NORMAL		DEXAMETHASON	ы	ACTH	CORTICO	STERONE
PHASES	1	Boyg	120µg	160µg	0.5 I.U.	1µgM 1µgE	1µgE
RECRUDESCENT	2.27	3.30 <sup>0</sup>	3 <b>.</b> 37 <sup>@</sup>	3.17	ł	ß	ł
	<u>+0.51</u>	<u></u> 40.85	77.0±	±1.22	, 1		
BREEDING	. 3.47	6.58	7.81 <sup>0</sup>	5.42*	<b>I</b>		ł
-	+0•69	+0.815	+3.50	+1.74		,	,
REGRESSION	7.69	, I	ı	ı	*61,•1	3.97*	2•69 <sup>0</sup>
	±3.06	-			<u>+</u> 0•42	±1.17	86°0+
			`				
	@ P<0.022	++ P<0.01	* P< 0.05				
	M - MORNING	E - EVENING	INĠ		-		

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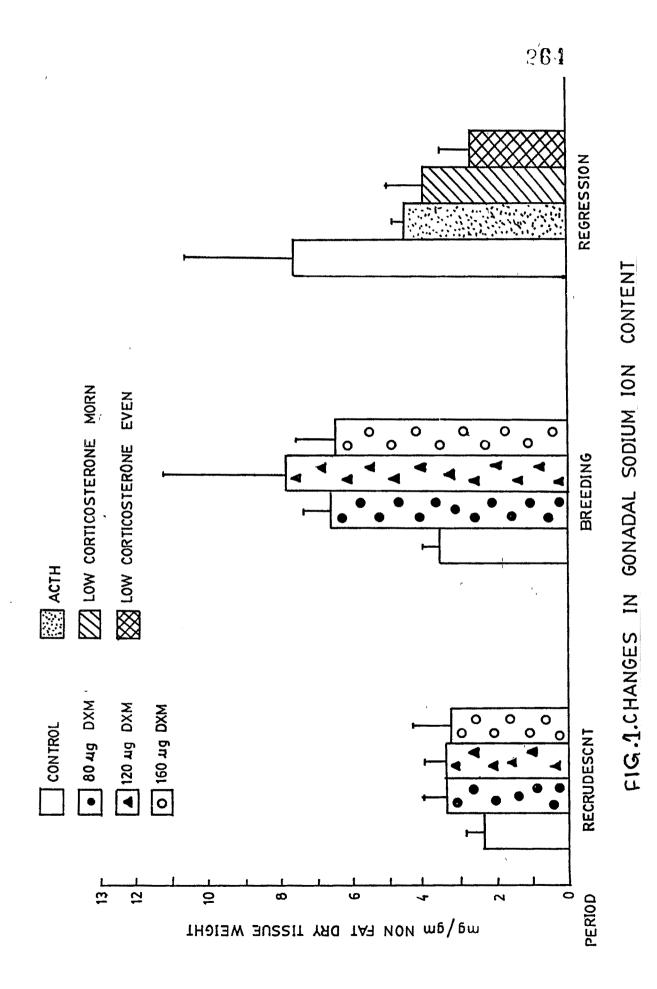
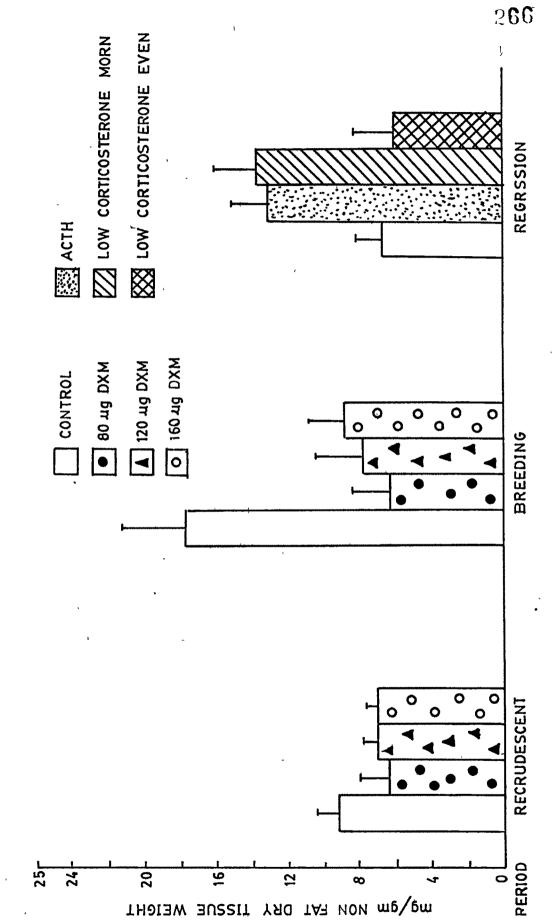


TABLE-2	••	VAL ALTERATIO ISSUE WEIGHT	SEASONAL ALTERATIONS OF GONADAL POTASSIUM CONTENT (mg/gm LIPID FREE DRY TISSUE WEIGHT <u>*</u> S.D.) IN NORMAL AND EXPERIMENTAL PIGEONS, <u>C.LIVIA</u>	L POTASSIUM ( DRMAL AND EXI	content (mg/g Perimental Pi	gm LIPID FREI GEONS, C.LI	Z VIA
REPRODUCTIVE	NORMAL	воре	DEXAMETHASONE 120µg	压 160µg	ACTH 0.5 I.U.	CORTICOSTERONE 1µgM 1µgE	STERONE 1µgE
RECRUDESCENT	9•29 ±1•31	6.27 <sup>@</sup> <u>+</u> 1.85∋	6.95** +0.78	6.95 <sup>0</sup> +0.64		1	1
BREEDING	17.62 +3.64	6.21++ <u>+</u> 2.3	7.72 <sup>++</sup> <u>+</u> 2.62	8.72++ <u>+</u> 2.08	1	ł	, J
REGRESSION	6 <b>.</b> 74 <b>±1</b> .48	I	ı	ı	13.05 <sup>++</sup> <u>+</u> 2.0	13.60 <sup>++</sup> <u>+</u> 2.37	5.96 +2.35
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@ P<0.02 ++ P<0.001 \*\* P<0.005

E - EVENING M - MORNING

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FIG.2. CHANGES IN GONADAL POTASSIUM ION CONTENT

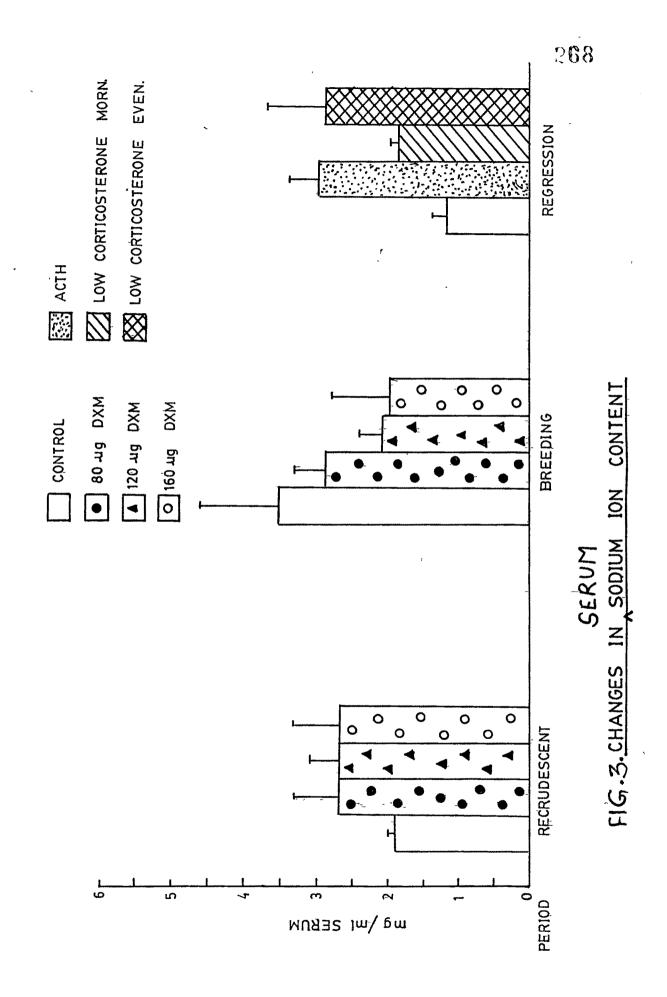
SEASONAL ALTERATIONS OF SERUM SODIUM CONTENT (mg/ml; ± S.D.) IN NORMAL AND EXPERIMENTAL PIGEONS, C. LIVIA	U. Them The The	8	1	++ 1.90 <sup>**</sup> 2.90 <sup>**</sup> <u>+</u> 0.18 <u>*</u> 0.82
m/gm) TNI	ACTH 0.5 I.U.	I	ı	2.96 <sup>++</sup> <u>+</u> 0.55
SODIUM CONTE	NE 1604g	2.70 <sup>+</sup> +0.68	2.05 <sup>+</sup> <u>+</u> 0.860	1
NS OF SERUM IGEONS, C. L	DEXAMETHASONE 120µg	2.70 <sup>++</sup> <u>+</u> 0.47	2.10 <sup>+</sup> +0.303	8
IAL ALTERATIO TPERIMENTAL P	Boyg	2.70 <sup>+</sup> <u>+</u> 0.62	2.80 ±0.41	I
	NORMAL	1.955 <b>+</b> 0.13	3.50 +1.10	1.26 <u>+</u> 0.29
TABLE-3	REPRODUCTI VE PHASES	RECRUDESCENT	BREEDING	REGRESSION

+ P<0.01 ++ P<0.001 \*\* P<0.005

M - MORNING E - EVENING

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	NORMAL A	NORMAL AND EXPERIMENTAL PIGEONS, C. LIVIA	PIGEONS,	, C. LIVIA	• ·		
REPRODUCTIVE PHASES	NORMAL	BOJLE .	DEXAMETHASONE 120µg	з 160у£	ACTH 0.5 I.U.	CORTICOSTERONE 1µgM 1µgE	TERONE 1µgE
RECRUDESCENT	1.38 +0.16	3.13 <sup>++</sup> +0.50	3.46 <sup>++</sup> <u>+</u> 0.73	3.02 <sup>++</sup> ±1.26		ŧ	ĩ
BREEDING	2•31 ±0•167	1.96* <u>+</u> 0.34	1.94* <u>+</u> 0.30	1.60 <sup>++</sup> +0.30	t	1	1
REGRESSION	0•45 <b>±</b> 0•11	F	ł	ł	0.972 <sup>++</sup> <u>+</u> 0.13	0.43 +0.13	1.23++
	+ P<0.01 M - MORNING	++ P<0.001 E - EVENING	* P∠ 0.05	10			

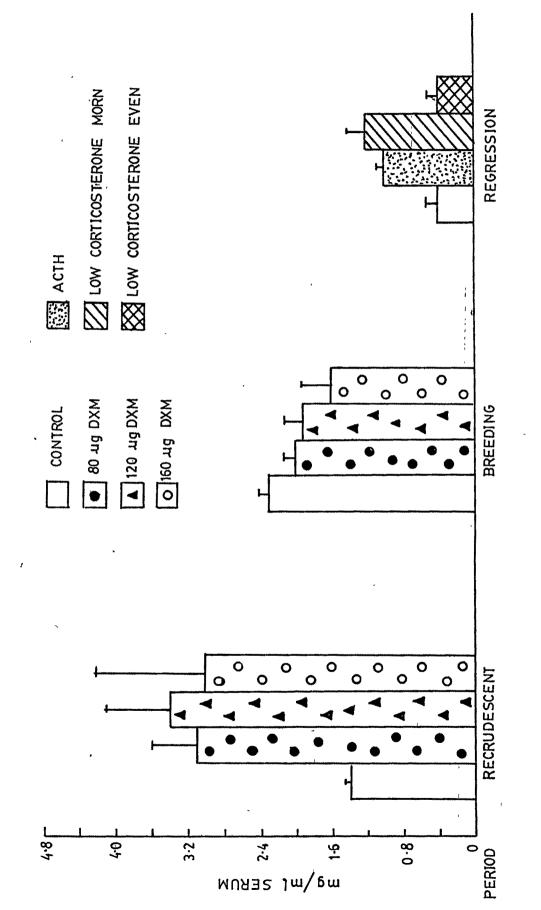
TABLE-4 : SEASONAL ALTERATIONS OF SERUM POTASSIUM CONTENT (mg/ml ± S.D.) IN NOPMAL AND EXDERTMENT DIFFORMS C ITUIA

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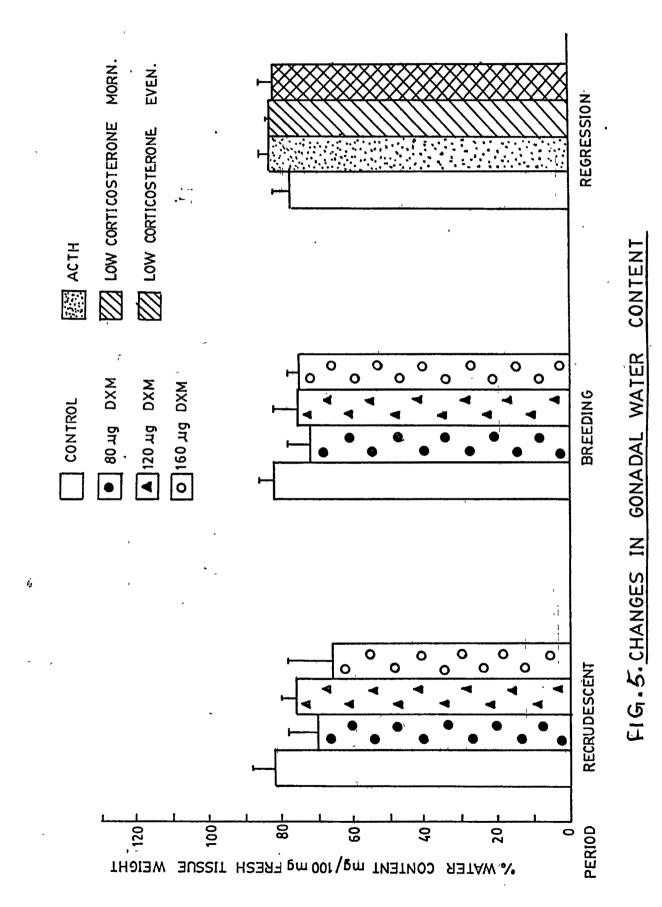
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CONTENT SERUM POTASSIUM ION FIG.4. CHANGES IN

REPRODUCTVE	NORMAT.		DEXAMETHASONE	JNF.	АСТН	CORTICOSTERONE	CORTICOSTERONE
	TRAINON	Bolug	120µg	160Jug	0.5 I.U.	1 Jugh	1µ6E
RECRUDESCENT	81.50	*60°02	75.64	65 <b>.</b> 73 <b>+</b>	× 8°	t	ı
	±6.23	<u>+</u> 9•63	<u>+</u> 3•87	+12.16			
BREEDING	81.61	72.07	75.10*	74 <b>.</b> 86	1	ŧ	ı
	+4.20	±6.61	±7•48	+2.56			
REGRESSION	77.33	I	I		82 <b>.</b> 85 <sup>*</sup>	80 <b>•</b> 56 <b>*</b>	82.40
	<b>44.</b> 94				+2.12	+3.15	<u>+</u> 3.48

+ P<0.01 \* P<0.05 \*\* P<0.005

M - MORNING E - EVENING



Changes Under Experimental Conditions.

The gonads of the adrenal suppressed birds exhibited increased Na<sup>+</sup> content and decreased K<sup>+</sup> content relative to the controls. These values were very much comparable with the values obtained during the non-breeding phase in normal birds. Relative to the controls, the serum Na<sup>+</sup> content was increased during recrudescente and decreased during breeding. Serum K<sup>+</sup> content also exhibited similar set of changes in the adrenal suppressed pigeons during recrudescence and breeding. The water content of the gonads decreased to levels very much comparable with the non-breeding level in the control birds.

In the birds injected with either ACTH or corticosterone, the Na<sup>+</sup> content of gonads decreased and the K<sup>+</sup> content increased, with the values tending to be similar to the ones charactestic of breeding in control birds. In the case of serum Na<sup>+</sup> and K<sup>+</sup>, ACTH/corticosterone administration increased their contents to levels charecteristic of the breeding phase. LCE was ineffective Contents. in the case of serum and gonadal K<sup>+</sup>. Similarly the water content of the gonads also increased to the breeding level on ACTH/corticosterone administration.

#### DISCUSSION

Adrenocortical steroids are known to exert a fundamental influence on water and electrolyte balance in vertebrates. Though aldosterone is the principal adrenal hormone accredited with this function in mammals. even glucocorticoids are reported to have aldosterone like activity in birds (Holmes and Adam, 1963; Nevsakaya et al, 1977). Viewed in this context of a functional overlap, changing titers of glucocorticoids can be expected to have a definite influence on ionic and water balance of submammalian vertebrates. Hence in a seasonal breeder like the tropical wild pigeon, which has been shown to have a parallel adrenal-gonad axis (Chapters I and II), seasonal variations in serum concentration of corticosterone and, resultant effects on tissue and serum ionic contents can be presumed. The present evaluation seems to strengthen the concept as definite alterations in tissue and serum Na<sup>+</sup> and K<sup>+</sup> contents have been obtained under adrenal insuffiency and adrenal excess, which are in keeping with the normal seasonal changes. Normal birds, during the recrudescent and breeding periods showed reduced gonadal Na<sup>+</sup> content and increased K<sup>+</sup> content while the serum cationic content was elevated. The non-breeding period was marked by a reverse set of changes. Apparently decreased Na\* content

coupled with increased K<sup>+</sup> content leading to a reduced Na<sup>+</sup>/K<sup>+</sup> ratio is favourable for gonadal activation and functioning. Similar changes have been recorded by Patel and Ramachandran (1986) in the gonads of domestic pigeons. This is evident not only from the increased Na<sup>+</sup> content and decreased  $K^+$  content with an increase in Na/K<sup>+</sup>ratio during the non-breeding season but also from the similar pattern discernuble in dxm suppressed birds during the recrudescent and breeding periods (See Figs. 1-4 and tables 1-4). Similarly, ACTH/corticosterone administration during the non-breeding, also brought about an increased gonadal K<sup>+</sup> content with decreased Na<sup>+</sup> content and hence a reduced Na<sup>+</sup>/K<sup>+</sup> ratio; a picture characterstic of the breeding phase. All these changes portend adaptive alterations in gonadal Na<sup>+</sup> and K<sup>+</sup>ion content in relation to the breeding activities and the favourable influence of adrenocortical stroids threat in mediating such changes either directly or indirectly. Favourable influence of K<sup>+</sup> in gonadal activity can be surmised from the observation of Setchell et al. (1965) of doubled respiratory rate of testis homogenates in high K<sup>+</sup> containing medium with no exogenous substrate. Further, increased K<sup>+</sup> content in the ovary of immature rats treated with gonadotropins has also been reported by Neauport and Emmerich(1979).

They had alsocobserved increased  $K^+$  and  $\frac{42}{4K^+}$  uptake in the PMSG primed ovaries as well as after FSH and LH administration in hypophysectomized rats. The Na<sup>+</sup>/K<sup>+</sup> ratio could be considered to have favourable influence on transport processes associated with active spermatogenesis. Pertinently, increased activities of alkaline phosphatase and ATPase observed during breeding phase have also been correlated with increased transport of metabolites and other substances into the active gonads (Chapters V and VII). Pilo and Patel (1978) based on their study on pigeon liver have in this context opined that the entry of K<sup>+</sup> into tissues can facilitate entry of glucose and metabolites.

Parallel elevation in serum Na<sup>+</sup> and K<sup>+</sup> levels has been observed during the breeding phase. Converse change was recorded during the non-breeding phase. Apparently hypernatrinemia and hyperkalemia seem to be the feature during the breeding months. From the inferred parallel adrenal-gonad relationship (Chapters II and III), these changes in the serum Na<sup>+</sup> and K<sup>+</sup> contents occurring in relation to gonadal cyclicity in the form of high during breeding and low during non-breeding were mimicked by ACTH/corticosterone administration during the non-breeding season respectively. These observations once again underscore the influence of corticosterone in modulating

ionic balance as well as gonadal cyclicity. Though the serum Na<sup>+</sup>/ K<sup>+</sup> contents showed parallel changes in dxm treated birds, there was however a seasonal difference in terms of the functional status of the gonads. This is exemplified by the increased serum Na<sup>+</sup> and K<sup>+</sup> contents during the recrudescent phase and decreased contents during the breeding phase. Since the increased contents during the recrudescent phase in adrenal suppressed birds were nearly identical to those observed in normal birds during the breeding season, it is quite likely that the hormonal balance prevailing during the recrudescent phase is slightly different from that during the breeding phase, which has given rise to the differential change in serum ion content under dxm treatment. In the light of the suggested aldosterone like activity of glucocorticoids in birds, dxm treatment is expected to lower the serum Na<sup>+</sup> ion content associated with an increase in serum K<sup>+</sup> ion content. However in the present study simultaneous lowering of both Na<sup>+</sup> and K<sup>+</sup> has been obtained during the breeding season. This parallel changes in the serum Na<sup>+</sup> and K<sup>+</sup> contents is further emphasized by the changes obtained either by dxm treatment in the recrudescent phase or by ACTH/corticosterone administration during the non-breeding phase. Some credibility to the present observations is provided by the report of Thomas and Phillips (1971) of simultaneous lowering

the of Na<sup>+</sup> and K<sup>+</sup> contents after adrenalectomy in duck. On a different note, Eiler <u>et al.</u> (1979) obtained a 20% increase in plasma K<sup>+</sup> with no change in Na<sup>+</sup> in dxm treated horses. Taken together, all these observations suggest differential involvement of adrenal steroids (in terms of species and seasonal differences) in regulating the monovalent cationic content of serum. Though there is paucity of information on this line, the present study tends to suggest adaptive modulations in serum and gonadal ionic content in relation to parallel adrenal-gonad relationship. The observations of Rzasa <u>et al</u>. (1982) of significant increase in plasma Na<sup>+</sup> and non-significant increase in serum K<sup>+</sup> in duck during oviposition is relevant in this context.

Finally, the evaluation of the water content of the gonads has revealed relatively higher content during the breeding as compared to the non-breeding and this pattern could be reversed by either adrenal suppression during the recrudescent and breeding phases or by ACTH/corticosterone administration in the non-breeding phase. Apparently, increased functional competence of the gonads is marked by higher water content and vice versa. Similar seasonal changes in gonadal hydration has been reported by Kapur and Toor (1978) in a teleost fish, <u>Cyprinus carpio</u> as well as by Patel and Ramachandran (1986) in domestic pigeons. The involvement of adrenal steroids in the regulation of tissue water content can be presumed from the reports of Ioan (1978) of modification of tissue fluid content after adrenalectomy, and treatment with Nacl and hydrocortisone in rats and also of Whiting and Wiggs (1977) of alteration of hepatic water the content after cortisol administration in brook trout, Salvelinus fontinates (Mitchill).

It can be surmised that the adrenocortical steroids have a regulatory influence on water and electrolyte balance and that the avian corticosteroid has a definite influence in the favourable and adaptive seasonal modulations associated with annual gonadal cyclicity.

### SUMMARY

Tissue and serum ion content (Na<sup>\*</sup> and K<sup>+</sup>) and water content have been studied on a seasonal basis in normal and adrenal manipulated pigeons. High K<sup>+</sup> and water contents and low Na<sup>+</sup> content in the gonads could be discerned during *the* breeding phase while the converse was true during the non-breeding phase. Serum Na<sup>+</sup> and K<sup>+</sup> tended to show somewhat parallel changes and hence their levels were higher in the breeding season and low in the regression phase. The ability of the adrenal gland to modulate these seasonal changes was inferred by the increased Na<sup>+</sup>/K<sup>+</sup> ratio after ACTH or corticosterone administration in the non-breeding phase. These results suggest elevated K<sup>+</sup> and water contents to be associated with gonadal activation and its functioning and the ability of corticosterone to modulate their normal seasonal changes in the pigeon.