QUANTITATIVE EVALUATION OF ADENOSINE TRIPHOSPHATASE IN PROVENTRICULUS AND VENTRICULUS OF DEVELOPING FOWL AND QUAIL AND CERTAIN ADULT BIRDS.

CHAPTER V

QUANTITATIVE EVALUATION OF ADENOSINE TRIPHOSPHATASE IN PROVENTRICULUS AND VENTRICULUS OF DEVELOPING FOWL AND QUAIL AND CERTAIN ADULT BIRDS.

The sequential chemical and physical changes taking place during embryonic development have modifying effects on differentiating cells. The process of organogenesis and histogenesis therefore involve certain basic chemical and physical changes and there is very reason to believe that these changes are brought about under the control of enzymes.

Adenosine triphosphatase (ATPase) is one such key enzyme in the developing systems denoting energy utilizing or energy generating pathways in metabolically active tissues which are engaged in the synthesis and active transport of metabolites across the cell membrane or active mitotic activity. Na^+-K^+ dependent ATPase is widely accepted to be intimately associated with mechanism of transport of ions and metabolites such as glucose and amino acids, etc across the glandular epithelial tissues (Ganong, 1989), whereas Mg^{2+} ATPase could be expected in the tissue or cells engaged in active mitosis or in the organ with high rate of growth (Mazia *et al.*, 1961; Patel, 1978; Schuurmans and Bonting, 1981). The synthesis and utilization of ATP, therefore, occur simultaneously in any living cells and the activity of ATPase is an indication of energy consuming activities of the cell (Silver and Stull, 1982; Hai and Murphy, 1988; Kenney *et al.*, 1988). Bonting *et al.* (1961) in their studies on various tissues from cat concluded domestic? that the tissue concerned with secretory functions have the highest ATPase activity. The relationship between energy release and protein synthesis, and ATPase actions and lipolysis have been reported by many workers (Kielly and Meyerhof, 1950; Martonisi, 1963, 1967; Young, 1970; Burleigh, 1974; Lundholm and Schersten, 1975).

When cellular ATP content is reduced through increased activity of ATPase, mitochondrial respiration is increased in order to produce more ATP, hence more oxygen is consumed. When ATPase activity is decreased the reverse is true. This appears to be closely related to and influence the ability by a wide variety of animal cells to transport other solutes against an electrochemical gradient; these transport processes have considerable physiological importance (Skou, 1965; Schuurmans and Bonting, 1981; Hai and Murphy, 1988).

With this background information concerning the role of ATPase in different physiological functions, a quantitative study was carried out in the stomach complex of developing and adult fowl and quail, and certain other adult birds so as to understand the role of this enzyme in this metabolically active organ and correlate its role in the energy consuming activities.

Materials and Methods

Newly hatched chicks of fowl (RIR variety) and quail were procured from Government Poultry Farm, Baroda and Poultry Project Development, Dahod, Gujarat respectively. They were reared in a well maintained aviary of Department of Zoology, M.S. University of Baroda. Pigeons were procured from a commercial animal dealer. Adult house swift and house sparrow were caught from their nests located in the premises of the Zoology department.

Young ones of fowl and quail falling in the following age groups *viz.*, 0, 5, 10, 20 and 30 days as well as their adult and other birds mentioned above were weighed and then decapitated in the early morning hours. The proventriculus and ventriculus were quickly removed and blotted free of blood and other tissue fluids. The pre-weighed pieces of proventriculus and ventriculus were homogenized separately in cold distilled water and processed further as follows:

Biochemical analysis

ATPase: The Total ATPase activity was measured according to the method of Umbreit *et al.* (1957). The inorganic phosphate released was estimated by the method of Fiske and SubbaRow (1925).

Protein: The total protein content of homogenate was estimated with the help of Folin phenol method as described by Lowry *et al.* (1951) using bovine serum albumin as standard.

RESULTS

Proventriculus (Table 1; Figure 1a)

From a moderate level of ATPase in the proventriculus of fowl on the day of hatching an increase was noted on day 5 followed by a fall in the level on day 10 of development. A three fold increase was noted on days 20 and 30 followed by a drop in the concentration in the adult. In quail a gradual increase was observed from the day of hatching till day 20. After a small decline on day 30 the enzyme registered a still lower level in adult.

On a comparative basis, the ATPase activity was high in the proventriculus of adult fowl followed by quail, sparrow, pigeon and swift in that order (table 2; figure 1b).

Ventriculus (Table 1; Figure 1a)

In the ventriculus of fowl the concentration of ATPase remained characteristically low during the different days of development except on day 20 when a twofold increase was noted. In quail also, the concentration remained the same (though on a higher note) from the day of hatching till day 10 but decreased drastically on day 20 and thereafter.

In the adult birds, sparrow depicted highest activity of ATPase followed by fowl, pigeon, swift and quail (table 2; figure 1b).

DISCUSSION

Every organ must contain an enzyme system that can catalyse the hydrolysis of ATP to ADP and orthophosphate and thus liberate energy. This enzyme system, which is generally regarded as adenosine triphosphatase (ATPase) is known to be located in the microsomal particles that contain lipids and proteins (Skou, 1965). In any metabolically active tissue or organs such as proventriculus and ventriculus the synthesis and hydrolysis of ATP are the characteristics features and run parallel. It is essentially so in the tissues or organs which are active in the synthesis and transport of metabolites or which have high mitotic activity.

Table 1. Levels of ATPase* activity in the proventriculus and ventriculus of developing and adult fowl and quail.

AGE (DAYS)	PROVENTRICULUS		VENTRICULUS	
	FOWL	QUAIL	FOWL	QUAIL
0	58.31 ± 2.15@	6.54 ± 0.32	6.02 ± 0.16	23.92 ± 1.08
5	73.36 ± 3.65	8.25 ± 0.33	4.93 ± 0.14	28.34 ± 0.64
10	14.90 ± 0.99	10.92 ± 0.71	6.84 ± 0.23	24.22 ± 0.84
20	41.03 ± 1.80	22.49 ± 0.96	14.37 ± 0.21	6.65 ± 0.30
30	42.85 ± 1.39	17.04 ± 0.83	4.66 ± 0.25	9.11 ± 0.50
ADULT	20.62 ± 0.67	10.27 ± 0.52	6.97 ± 0.14	4.08 ± 0.19

* μg phosphorus released/mg protein/10 minutes.
@ Values expressed as mean ± SEM of six animals.

n=7

Table 2. Levels of ATPase* activity in the proventriculus and ventriculus of adult birds.

ANIMAL MODEL	PROVENTRICULUS	VENTRICULUS
SWIFT	5.70 ± 0.39[@]	4.54 ± 0.21
SPARROW	9.84 ± 0.40	12.02 ± 0.62
QUAIL	10.27 ± 0.51	4.08 ± 0.19
PIGEON	7.32 ± 0.82	6.24 ± 0.20
FOWL	20.62 ± 0.67	6.97 ± 0.14

* μg phosphorus released/mg protein/10 minutes.
@ values expressed as mean ± SEM of six animals.

Fig 1a. Levels of ATPase in growing and adult birds.

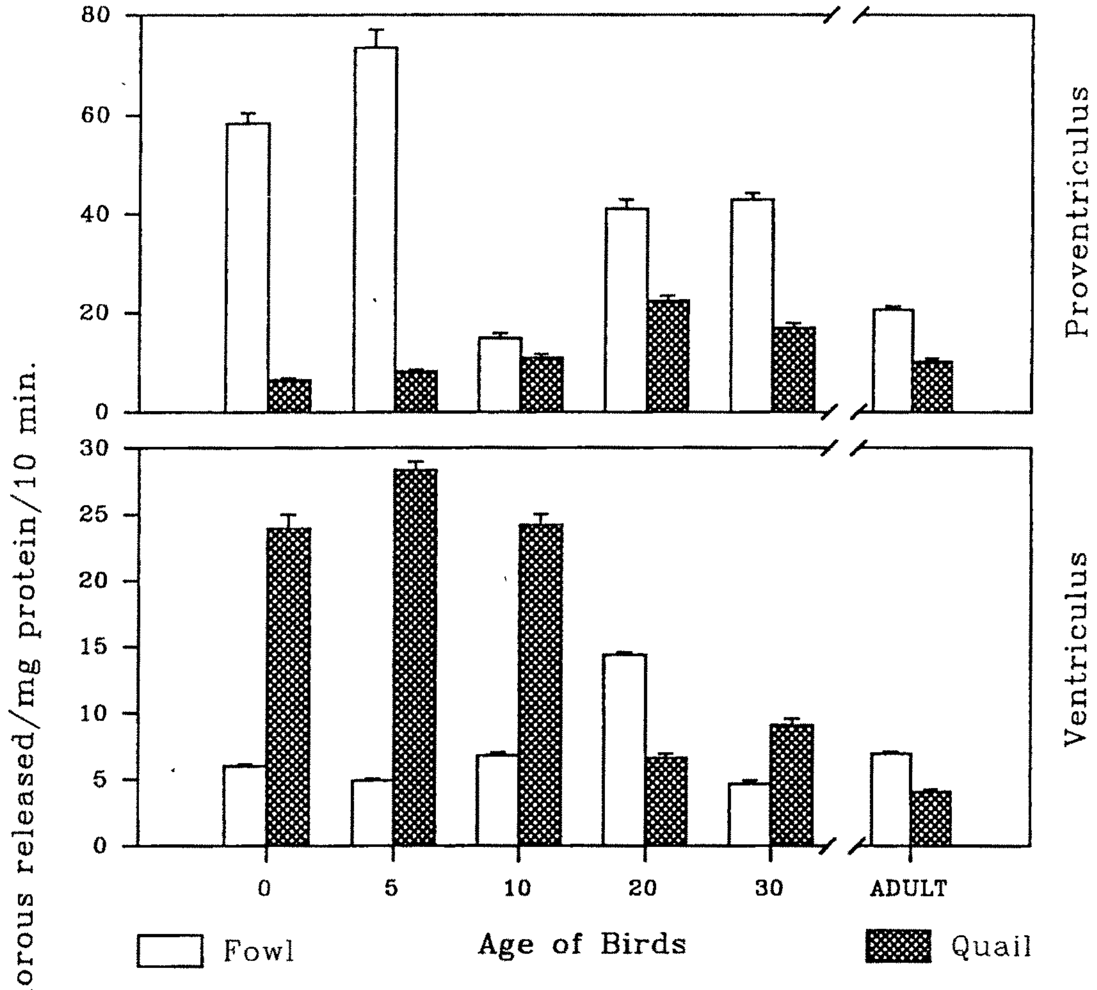
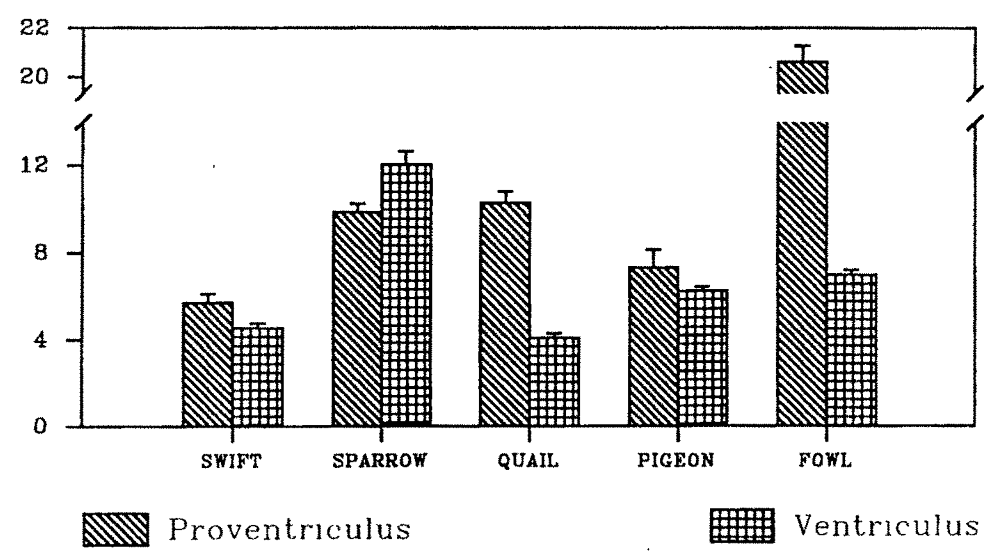


Fig 1b. Levels of ATPase in adult birds.



ATPase system was studied by many workers (Skou, 1957, 1960 and 1965; Dunham and Glynn, 1961; Talesara and Narang, 1979). There are reports on the importance of Na^+-K^+ dependent ATPase in the mechanism of transport of ions and metabolites across the glandular epithelial tissues (Tanaka, 1987). In the wake of the above reports high levels of ATPase in the proventriculus of fowl during the initial days of development is noteworthy and correlates its presence in the glandular organs with the secretory functions. A low but appreciable amount of ATPase present in the ventriculus of fowl and proventriculus of quail and a moderate level of the enzyme in the ventriculus of quail can be explained in terms of energy demand of these organs during different days of development. Various metabolic processes occurring during the initial stages of development may demand high energy which is made available from ATP; and ATPase induced hydrolysis of ATP could be an important feature in the normal functioning of proventricular multiple glands. The availability of nutrients might also determine the rate of operation of metabolic activities. Shah and Panicker (1974) investigated histochemically a few glycolytic and TCA cycle enzymes in the ventriculus of developing pigeon. They inferred that the pigeon ventriculus, during the initial days of development, though dependent on carbohydrate metabolism, also utilize lipids (Shah and Panicker, 1974a, b). Interestingly enough, this very period is also characterized by quick development of connective tissue around the smooth muscle fasciculi and consequently the acid and alkaline phosphatase activities were also noted to be moderate to high (Shah and Panicker, 1971). Elsewhere in the present study (chapters III) a steady decline in the levels of glycogen and fluctuations in the amount of lipids in the developing stomach complex of fowl and quail has been discussed. At the same time non-specific phosphatases varied systematically in their concentration (chapter VI) in both proventriculus and ventriculus of these developing young ones. As far as the proventriculus and ventriculus are concerned, the activity of ATPase is related not only to the need of energy for secretory function of the mucosal tubule, but also to the laying down of the connective tissue around the smooth muscle fasciculi, synthesis of contractile protein, post mitotic autophagosis and formation of collagen and fibrous protein. All these processes require a steady supply of energy which is met by the breakdown of ATP by ATPase. During the different phases of development of the organs, directed mainly at the attainment of functional competence, the increased energy requirement is met by the catabolism of reserve glycogen as evidenced by a decreased glycogen content (chapter III). At the same time there is still an active process of lipid catabolism as evidenced not only from the fact that the ventriculus has a poor capacity to store metabolites but also from the fact that as ventriculus performs a slow and sustained contraction a ready supply of fatty acids made available through blood might be economical and useful (White *et al.*, 1964). As far as the adult birds are concerned the proventriculus depicted a low ATPase concentration when compared with ventriculus in all the adult birds. The enzyme in proventriculus was observed to be lowest in swift and highest in quail with the other three birds *viz.*, sparrow, pigeon and fowl occupying an intermediary position. In case of ventriculus, the enzyme activity was found to be highest in sparrow followed by fowl, pigeon, swift and quail in that order. In general, the food habits of these birds are taken into considerations while ascribing any particular role for the enzyme. It is generally known that large variations in size and muscularity of stomach are found in birds which are known to differ in their diets (Shah and Panicker, 1975). These morphological variation exhibited by the stomach of birds with different diets logically should reflect on the variations in the physiological and biochemical activities of the stomach muscles. Birds with inherent habits of preferring specific type of food cannot change their diets or feeding habits since the growth and size of ventriculus are genetically predetermined. Perhaps the adult birds under investigations here are primarily adapted for grains and seeds (except swift) but have secondarily taken to insects or mixed type of food. Yet they have well developed muscularity but differ from each other in the thickness of the component parts. Hence the variations in the concentration of ATPase as described above in the stomach complex of adult birds are justifiable.