GENERAL CONSIDERATION

Avian kidney differs from mammalian kidney in several respects, morphologically as well as physiologically. Anatomical pecularities are in terms of (1) arrangement of renal tissue into lobules, (2) presence of two types of nephrons (3) arrangement of intralobular and perilobular collecting tubules (4) presence of renal portal system draining into cortical areas and (5) presence of a peculiar medullary cone draining into secondary branches of ureter. Physiologically, the avian kidney differs in terms of nitrogenous waste product excreted and mechanism by which urine is concentrated.

Metabolically also avian kidney has distinct features. The presence of inducible cytosolic form of phosphoenolpyruvate carboxykinase (absent in avian liver) equip the avian kidney in utilizing precursors such as alanine, aspartate and other amino acids and pyruvate to synthesize glucose. Because of this reason, avian kidney has higher capacity of gluconeogenesis than avian liver. Gluconeogenesis is an adaptive mechanism, mainly concerned with glucose homeostasis. Production of glucose from lactate or non-carbohydrate sources is initiated when the animals are under stress of starvation or when the food contains very little carbohydrate. In this light a survey of gluconeogenic capacity of kidney of three different birds with different dietary specializations revealed interesting

information (Chapter II). The three birds studied were pigeon (a graminivore), sparrow (an omnivore) and swift (an insectivore). Of these three birds, only swift consumes a low-carbohydrate food as their diet mainly consists of insects. The kidney of swift showed a very high capacity of gluconeogenesis judging from the activities of various enzymes. In contrast, omnivorous sparrow kidney showed the least gluconeogenic capacity, obviously due to the fact that the food they consume brings in adequate supply of carbohydrate, lipid and protein. The adaptive high rate of gluconeogenesis in the swift kidney may also be due to the fact that hormones involved in promoting such metabolic activities are also high in the circulating blood. This needs further investigation, and attempts to establish correlation between hormone levels in plasma and dietary specialization are underway in our laboratory.

Invariably metabolic adaptations are regulated by various hormones. Metabolic hormones such as glucocorticoids, glucagon, catecholamines, thyroxine and insulin, in one way or other, are capable of regulating metabolic adaptation. Gluconeogenesis is one such process that is regulated on a need basis. Both long-term and short-term regulation of gluconeogenesis are encountered in tissues such as liver and kidney. Metabolic regulations in a short-term basis are carried out by activating and inactivating enzymes already existing in the tissue. Long-term regulations on the other hand involve induction or

repression of enzyme synthesis. In other words, long-term regulations are carried out by recruiting more enzyme molecules or down regulating the existing number of enzyme molecules. Enzymes which are allosterically influenced are regulated by supply of substrates. Some key enzymes are activated or inhibited by phosphorylation/dephosphorylation mechanism. Protein kinases (which are required for activating the phosphorylating/dephosphorylating enzymes) are activated by either cAMP or Ca^{++}. cAMP and Ca^{++} act as second messengers to a large number of hormones. By increasing or decreasing the cytosolic concentrations of cAMP and Ca^{++}, most of the hormones could influence metabolic pathways.

Hormonal action on metabolic events, initially involve plasma membrane receptor activation and the ensuing cytosolic messenger formation (cAMP) or release (Ca^{++}). Some of the hormones could act on nuclear membrane and thereby act on induction or repression of enzyme synthesis. Activation or inhibition of gluconeogenesis by hormones is also accomplished by regulating the concentration of cAMP or Ca^{++} or by transcriptional methods. Gluconeogenesis is activated by glucocorticoids, glucagon and catecholamines while it is inhibited by insulin. Although glucocorticoids, glucagon and catecholamines can stimulate gluconeogenesis there could be subtle differences in the degree of activation by each hormone in a given tissue. All of them need not also act at the same control points of

entry of precursors into gluconeogenic pathway. Gluconeogenic precursors join the pathway at different points. Some like lactate undergoes oxidation to join the pathway, others like glycerol join the pathway at the triose phosphate level. Amino acids such as alanine and aspartate are converted into keto-acids and ultimately form oxaloacetate which then joins the gluconeogenic pathway. Some hormones activate the pretriose reactions while others act at the triose level. Sometimes tissues also specialize to use one or two gluconeogenic precursor (s) more than others.

Avian kidney can utilize precursors such as lactate, pyruvate, oxaloacetate, alanine, aspartate, glutamine and glycerol to form glucose while avian liver predominantly converts lactate into glucose. This difference is due to the presence or absence of inducible cytosolic or mitochondrial enzymes concerned with gluconeogenesis. Action of hormone on tissue also differ accordingly. Since most of the hormones are present in the circulating blood at any given time, each one can act only concertingly. Agonists may act synergistically while antagonists can act only when the ratio tips in each ones favour.

In this light of forgoing account, one should bear in mind that administration of any one hormone in <u>in vivo</u> condition may not produce the anticipated, characteristic action. Some of the results obtained in the present investigation exemplify

this fact. Limitations experienced in explaining some of the data are due to cross reaction by native hormones already present, or compensatory release of some antagonists, or complementary release of agonists. Starved or fed condition add another dimension altogether and have to be taken into consideration while explaining the observed effects of extraneously introduced hormones.

Glucocorticoids are the most effective gluconeogenesis promotors. Corticosterone, which is the glucocorticoid found generally in birds, when administered in pigeon, produced a mild increase in gluconeogenic activities in the kidney (Chapter III). Since pigeons were not subjected to starvation, the effect of corticosterone administration did not produce a maximum gluconeogenic activity in the kidney.

Starvation could induce the release of glucagon and catecholamines. These hormones are also known to enhance gluconeogenic activities . Administration of glucagon combined with 48 hr starvation (Chapter VII) produced only a reduction in glycolytic activities in the kidney of pigeon. Some of the glycolytic enzymes, infact, should undergo inhibition in order to prevent 'futile cycles' from being operative during gluconeogenesis. Catecholamine administration enhanced the activities of some enzymes denoting enhanced gluconeogenesis, while others were inhibited, indicating general reduction in glycolytic

activities (Chapter VIII). It was observed that in the avian kidney, epinephrine stimulated glucose production from precursors such as amino acids, while norepinephrine induced synthesis of glucose from lactate (Chapter VIII). An increase in gluconeogenesis could also be brought about by increasing the supply of precursors. In fact thyroid hormone administration (Chapter IV) increased the gluconeogenic rate, probably by promoting release of precursors such as lactate, pyruvate, alanine and aspartate from extrahepatic tissues. Hyperthyroidic condition elevates the plasma concentration of these percursors in mammals (Muller et al., 1983) along with an enhanced extraction of these precursors by splanchnic tissues (Wahren et al., 1981).

In contrast to the actions of corticosterone, glucagon, catecholamines and thyroid hormones; all of which, in one way or other, increased gluconeogenesis in the kidney; insulin effected an inhibition of such activities in the kidney (Chapter V). Normally, insulin would induce tissues to take up more metabolites such as glucose and amino acids while inhibiting glycogenolysis and gluconeogenesis. In the kidney of pigeon, insulin may not be inducing such enhancement in the rate of uptake of metabolites. However, the role of insulin in the metabolic activities in the kidney of birds needs further thorough investigation.

Metabolic activities in the liver are now known to be regulated by autonomous nervous system. Previous studies in this laboratory (Verma et al., 1983; Pilo et al., 1984) on vagotomized pigeon indicated that gluconeogenesis in the kidney was activated when vagal denervation was made. This fact is further substantiated by the present study with acetylcholine administered pigeon. Administration of acetylcholine caused a reduction in gluconeogenic activity in the kidney (Chapter VI). Whether this effect of ACh is a direct action on renal tissue or brought about through an increased release of insulin is difficult to envisage.

Since catecholamines, especially norepinephrine caused an increased gluconeogenic activity in the kidney (Chapter VIII) it could be assumed that sympathetic activation would also lead to similar enhancement. Chemical sympathectomy (through 6-OHDA treatment) caused no apperent changes in the kidney metabolic activities (Chapter IX). The main reason for this may be that the adrenergic fibres present in the kidney are preganglionic fibres and thereby secrete acetylcholine. The fact that 6-OHDA treatment produced an intense reactivity of AChE in the kidney of pigeon probably indicate that both sympathetic (adrenergic) and parasympathetic (cholinergic) fibres are present in the kidney of pigeon (Chapter X).

The studies presented herein are at best to be considered as preliminary, and data raise more questions than solving any.

However, the data obtained from these studies indicate that all the metabolic hormones and autonomic nervous system can exert some influence on the kidney gluconeogenic activities probably to bring in short-term or long-term adaptations. Detailed studies are necessary to understand how and what metabolic reactions are regulated by various hormones and neural activities. Measurements of activities of key gluconeogenic enzymes such as PEPCK and Fructose 1,6-diphosphatase and their isoenzymic forms and enzymes in the 'futile cycle' such as phosphofructokinase and pyruvate kinase are already being carried out in our laboratory.