

CHAPTER I

INTRODUCTION

In considering the response patterns of an organ system towards the action of different chemicals, the gastrointestinal tract or the gut occupies a key position for pharmacological exploration since antiquity. The gut of an animal is not only most important organ for living but it performs with amazing capability the most intricate tasks of adapting the animal under the altered or adverse circumstances for its survival. The gastrointestinal tract, on one side, with the help of its digestive and absorptive faculties, keeps an intimate relation with the internal environment of the animal, and on the other side, by accepting the exogenous food particles of foreign origin and rejection of noxious by-products of digestion, detoxication and excretion, always continues its relation with the exterior. Lying in mid-position of external and internal environment, it affords a nice adaptability of the animal by supplying the essential nutrients, an emergent step for adaptation towards nutritional deficiencies and by eliminating the toxic or harmful substances coming exogenously along with food stuffs or excreted as by-products of digestion and secretion. Moreover, such a system in one way is in direct contact with the external environment at its oral and anal ends and its contents are considered to be a part of that environment. The functional analysis of such a system has been the object of most fascinating

study from the earliest times, yet physiologists are still much worse informed about this particular organ in comparison with other related systems. It is true that the intestine is perhaps the organ in which the pharmacological methods are most frequently used in elucidating the functional modification of it towards a variety of chemical agents.

The functions of intestine are varied of which the movement or the motility is the most important. Movements of the alimentary canal are consisting of propulsion, segmentation and mixing of contents resulting from the contractile activity of the muscular elements which enabled the organ to function efficiently in various other ways as being needed during homeostatic adaptability of the internal environment in accordance with the demands made on the organism as a whole. This motor activity of the gut can be nicely assessed experimentally both in situ or in isolated condition. The spontaneous or induced movements of intestine are due to the peculiar arrangement pattern of the smooth muscle cells as well as their controlling innervation pattern. The motor action of small gut has been studied since the days of Ludwig (1861). Many of our present-day concepts and much of our knowledge on it were explored during the last century, and many of the research publications of this century are simply elaborations of those observations. Yet "a sort of mystical and obscurantist odour seems to have lingered on even to the 20th century around all studies of the bowel, for it is a strange fact that about few subjects have battles more bitter and persistent been waged than those that have concerned the mechanisms of intestinal movements and the actions of drugs thereon" (Vaughan Williams, 1954). Bayliss and Starling (1899) reported in their classic paper on the movements and innervation of small intestine, "On no subject in physiology do we meet with so many discrepancies of facts and opinion as in that of the physiology of the intestinal movements. Among factors contributing to such discrepancies must doubtless be included the varying behaviour of the gut in different animals, the varying conditions of the animal with regard to feeding or

conditions of experiment, such as exposure and cooling of intestine" and every student working on intestinal motility would agree that the above citations are also applicable today in its strict sense.

The comparative physiology of gut motility encompasses an enormous field of research and considerable amount of information are available concerning the vertebrate gut, although a large part of it is based on mammalian gut, as in most cases the same was used as a reference point for the whole group. During the past fifty years an enormous number of papers has been published showing that the motor activities of the alimentary canal of mammals and higher vertebrates are under the influence of a complex system of nervous and endocrine controls, whereas unfortunately scanty information are available on fish. But the results which have been obtained from the limited studies on fish certainly suggest that while in principle the situation may be the same there are many differences in detail (Barrington, 1957). Although a comprehensive review on the structure and activity of the gut in fishes is available (Von Euler & Ustlund, 1957; Barrington, 1957; Burnstock, 1958), specific studies of gastrointestinal smooth muscle, particularly the effects of various drugs, hormones, ions etc. are sparse. However, the available reports on the motility of intestine of fishes indicate that the gastrointestinal smooth muscle from the lower vertebrate groups like teleost fishes is spontaneously active in a manner comparable to mammalian gut muscle. Further, the sensitivity of the fish gut muscle to hormones, particularly acetylcholine, has often been reported to be much higher than that of the mammalian gut and a group of workers (Gaddum & Szerb, 1961) established a method of estimating acetylcholine, serotonin and other biologically active substances like Substance P etc with the goldfish intestine in a microbath.

The response of gold fish intestine towards acetylcholine and Substance P as reported by Gaddum and his group provoked us to find out such sensitive response of other fish intestine commonly available in India. After preliminary experiments with

the intestines of few commonly known fresh water teleost fishes like, *Ophicephalus punctatus*, *Anabas testudineus*, and *Clarias batrachus*, it was discovered that isolated pieces of intestine of the *Ophicephalus punctatus* contract vigorously in the presence of very small amounts of acetylcholine (10^{-12} gm/ml) or serotonin (10^{-10} gm/ml) without any help of specially designed microbath as used by Gaddum's group. The preliminary communication of this observation (Seal & Maiti, 1967) has been published, and subsequently led us to test the possibilities of using the same material for developing an easier and standard demonstrable method for the bioassay of acetylcholine and serotonin.

In considering the sensitive responses of acetylcholine and serotonin towards the contraction of isolated intestine of *Ophicephalus punctatus*, it became mandatory to explore the action of various other chemicals, which are commonly used for analysing the mode of action of smooth muscles, on the isolated preparation of fish gut. It is true that extensive approaches for studying the comparative aspects of gut smooth muscle pharmacology have already been made, yet fish intestine has so far been ignored. Because of the lack of precise knowledge on the morphology and physiology of the smooth muscle cells of fish gut and their innervation pattern and the mode of action of well known drugs on it, the present investigation has been undertaken. It is admitted that the results so far obtained, in general, have only served to emphasize the complexity of the problem and the inadequacy of some of the earlier explanations.

In the present work, an attempt has also been made to establish a method of estimating acetylcholine and serotonin from biological fluids with the help of isolated fish gut of *Ophicephalus punctatus*. The action of some well known chemical agents have been screened on the particular preparation. In order to throw some light on the mode of action of those agents, the physiological characteristics of spontaneous motility, the morphology and the innervation pattern of the smooth muscles

of the intestine of *Ophicephalus punctatus* have been studied. It is hoped that the present study on in vitro preparations of fish gut may provide some clues on the mechanism of action of some chemicals in a comparative pharmacological basis and further it would help to establish for assaying the neurohormones particularly acetylcholine and serotonin from the tissues under varying physiological conditions which could later on be utilised widely.

Accordingly the present work is arranged :

- 1) The present knowledge on the action of different chemicals on fish intestine so far studied are compiled in the Review Chapter (II). The general organization of the intestine, physiological peculiarities with particular reference to its motility and contractility are included in this chapter.
- 2) Chapter III describes briefly the general morphology, habitat, distribution, availability, life history, systematic position, size, shape, and mode of living of *Ophicephalus punctatus*.
- 3) The general morphology, structural peculiarities, histological patterns, innervation pattern of the different portions of the fish gut are given in Chapter IV.
- 4) Chapter V deals with preliminary study of the actions of acetylcholine and serotonin on the contractile response of isolated fish intestine. An approach has been made in this chapter to evaluate the results in a comparative basis in order to establish the method of assaying the acetylcholine and serotonin with other standard methods.
- 5) A comparative analysis of the action of acetylcholine and epinephrine on the different parts of the intestine

of same species and the action of acetylcholine and serotonin on the foregut of few other allied species of fresh water but partially air-breathing teleost fishes have been made in Chapter - VI.

- 6) Chapter VII deals with the spontaneous motility and the physiological characteristics of the contractility of isolated fish gut under anoxia, altered temperature and pH. The action of some well known chemical agents, like barium chloride, histamine, nicotine, pitocine, epinephrine, acetylcholine, serotonin and sodium, calcium, potassium ions and others have been studied. An attempt has also been made to explore the mode of action of those chemicals by using common ganglion blocking agents or local anaesthetic agents.
- 7) In Chapter VIII an attempt has been made to estimate the acetylcholine content of the brain of rats under particular physiological condition by employing the bioassay technique developed with the fish gut. The results from this study has also been communicated.
- 8) This is followed by Chapter IX under the heading General Discussion. It includes a correlated analysis of the results of the actions of different pharmacological agents on the intestine of *Ophicephalus punctatus* in relation to its structural peculiarity along with the comments arising out of the experimental results of the individual chapter. This ends with conclusion and summary.