

STUDIES ON
Histological Structures Of
Small Intestine Of
Indian Buffalo (Bos Indus)

Thesis

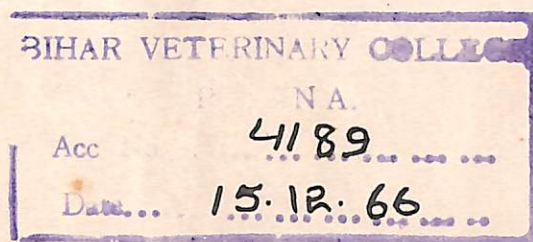
REPORTED TO THE FACULTY OF MEDICAL SCIENCE
OF THE UNIVERSITY
IN FULFILLMENT OF THE REQUIREMENTS
FOR THE DEGREE OF
MASTER OF SCIENCE (M.Sc.)

BY
LAKSHMI KUMAR BARNAL
B.A. (HONORS) DEPARTMENT OF ANATOMY
BIRMINGHAM NAVY COLLEGE
PATNA
1956-57

STUDIES ON
**Histological Structures Of
Small Intestine Of
Indian Buffaloes** (*Bos bubalis*)

Thesis

SUBMITTED TO THE FACULTY OF VETERINARY SCIENCE
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IN PARTIAL FULFILMENT OF THE REQUIREMENTS
FOR THE DEGREE OF
MASTER OF SCIENCE (Veterinary)



By
Awadhesh Kumar Barnwal
B.V.Sc. & A.H.
Post-Graduate Department of Anatomy
BIHAR VETERINARY COLLEGE
PATNA
1966

I certify that this thesis entitled "Studies on the his^tological structures of small intestine of Indian buffaloes (Bos bubalis)" has been prepared under my supervision by Sri A.K. Barnwal, a candidate for the M.Sc. (Vet) degree, with Anatomy as major subject, 1966 and that it incorporates the results of his independent study.

R. C. P. Yadva
30/11/66

Dr. R. C. P. Yadva,
M.S., Ph.D. (Mich., U.S.A.)
Professor and Head of the Department of
Anatomy
Bihar Veterinary College,
PATNA.

Dedicated
To
MY PROFESSOR
Dr. R. C. P. Yadava ,
M.S., Ph. D., (Mich., U.S.A.)

V I T A.

AWADHESH KUMAR BARNWAL .
Candidate for the degree of
Master of Science (Veterinary)

Final Examination: December, 1966
Department of Anatomy
Bihar Veterinary College, Patna .

Thesis: Studies on the histological structures of small
intestine of Indian Buffaloes (*Bos bubalis*).

Outline of studies

Major Subject : Anatomy
Minor subjects: Surgery (including Radiology) and
Gynaecology.

Biographical Items.

Born : January 12, 1939, Hajipur, Muzaffarpur, Bihar .

Undergraduate studies: Central Hindu College, Kanachha,
Banaras Hindu University, Banaras and Bihar Veterinary
College, Patna, Bihar. Obtained the degree in Bachelor
of Veterinary Science and Animal Husbandry, from Bihar
University in 1960.

Experience : Served as Animal Husbandry Supervisor in C.D.
Block, Kurtha, Gaya, from July 1960 to January 1961. A
member of the teaching staff of Bihar Veterinary College,
Patna, from January 1961 till date. Deputed for post-
graduate studies in Anatomy by the Department of Animal
Husbandry, Government of Bihar.

A member of the Bihar Veterinary Association.

A C K N O W L E D G E M E N T S.

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(A.K.BARNWAL).

ABSTRACT

This study is concerned with the results of the investigation of the physiological changes in the human body during the process of aging. The study was conducted over a period of five years, during which time a large number of subjects were observed and their physiological functions were recorded. The results of the study show that there is a significant change in the physiological functions of the human body as it ages. The changes are most marked in the cardiovascular system, which shows a marked decrease in the rate of circulation and a marked increase in the blood pressure. The respiratory system also shows a marked decrease in the rate of respiration and a marked increase in the volume of air breathed. The digestive system shows a marked decrease in the rate of digestion and a marked increase in the volume of food consumed. The results of the study are of great importance in the study of the human body and its functions.

ABSTRACT

The purpose of this study is to determine the effect of aging on the physiological functions of the human body. The study was conducted over a period of five years, during which time a large number of subjects were observed and their physiological functions were recorded. The results of the study show that there is a significant change in the physiological functions of the human body as it ages. The changes are most marked in the cardiovascular system, which shows a marked decrease in the rate of circulation and a marked increase in the blood pressure. The respiratory system also shows a marked decrease in the rate of respiration and a marked increase in the volume of air breathed. The digestive system shows a marked decrease in the rate of digestion and a marked increase in the volume of food consumed. The results of the study are of great importance in the study of the human body and its functions.

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ABSTRACT.

This thesis incorporates the result of investigations on the histological structure of small intestine of Indian buffaloes. Materials were taken from six Indian adult buffaloes and six buffalo-calves. Sections were cut every tenth distance irrespective of the overall length of small intestine. They were fixed in 10% formalin solution. Dehydration and infiltration were done, blocks were prepared and sections were cut and stained in different stains.

A general survey of the histological sections has been done and studied. Measurements of the villi, muscularis mucosae, submucosa, lamina muscularis, diameter of lumen of Brunner's glands and height of its epithelium has been taken.

The external surface of the small intestine is highly vascular and it appears reddish brown. The internal surface presents plicae circularis and longitudinal folds and are absent towards the mesenteric border. Peyer's patches are grossly present in the jejunum and ileum and sometimes in the duodenum of young animals when examined microscopically. There are well developed patches in the ileum of young animals whereas they are in the form of nodules in adults.

Shape and height of the villi varies with age, their location in the small intestine and individual to individual. In adults, the villi are longer in the duodenum and ileum than

young ones. The villi of jejunum are larger in young animals, than adults.

The Goblet cells are very few or even sometimes absent in both villus and crypt epithelium. Enterochromaffin cells have been observed only in the crypts of Lieberkuhn of duodenum in adults. Paneth cells have not been observed in this investigation.

Intracellular lymphocytes into the villus epithelium and lymphocytes penetrating the epithelium from the core of the villi have been observed. Cells under mitotic division are present in the crypt epithelium.

A large number of lymphocytes are present in the lamina propria. Central lacteal and smooth muscle fibers have been observed in the core of the villi. Brunner's glands in duodenum and lymph nodules in all portions of small intestine are present.

Muscularis mucosae is represented by a single layer of circular fibers in young animals whereas in adults there are two definite layers. This layer is thickest in the ileum and thinnest in the ~~duodenum~~, jejunum.

Submucosa is thickest in the ileum of young animals. It is thinnest in the duodenum of adults. Submucosa extends into the plicae circularis and longitudinal folds along with the muscularis mucosae. Meissner's plexus and ganglion cells are also present.

Lumen of Brunner's glands are larger ~~in adults than~~ in young ones whereas its epithelium is higher in adults.

Lamina muscularis is comprised of two layers and a connective tissue layer known as lamina intermuscularis in between them. Inner circular layer contains empty spaces, probably lymph spaces. The outer longitudinal layer is in the form of bundles.

Auerbach's plexuses are present usually in the lamina intermuscularis but sometimes they are in the outer longitudinal layer of lamina muscularis also.

Serosa is composed of loose connective tissue with blood vessels and nerves and sometimes it contains lymph nodules.

Some of the notable peculiarities observed in the histology of small intestine of Indian buffaloes are as follows -

- (1) Greater height of villi in the jejunum of young animals than the adults.
- (2) Greater thickness of submucosa in the ileum of young ones than in adults.
- (3) Greater diameter of lumen of Brunner's gland in young ones than in adults.
- (4) Presence of Peyer's patches in the duodenum of young animals.

- (5) Presence of plexus of smooth muscle fibers in the submucosa.
- (6) Presence of Auerbach's plexus in the outer longitudinal layer of lamina muscularis.
- (7) Presence of lymph nodules in the serosa.

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TABLE OF CONTENTS

INTRODUCTION	1	2	3
MATERIALS AND METHODS	4	5	6
RESULTS	7	8	9
DISCUSSION	10	11	12
CONCLUSION	13	14	15
REFERENCES	16	17	18
APPENDIX	19	20	21
INDEX	22	23	24

STUDIES ON HISTOLOGICAL STRUCTURES OF SMALL INTESTINE OF INDIAN BUFFALOES (*Bos bubalis*)

INTRODUCTION	1	2	3
MATERIALS AND METHODS	4	5	6
RESULTS	7	8	9
DISCUSSION	10	11	12
CONCLUSION	13	14	15
REFERENCES	16	17	18
APPENDIX	19	20	21
INDEX	22	23	24

TABLE OF CONTENTS.

INTRODUCTION	Page 1-3
REVIEW OF LITERATURE	4-31
Villi	4
Lamina Propria	17
Lamina Subglandularis	22
Muscularis Mucosae	22
Submucosa	24
Lamina Muscularis	26
Serosa	29
MATERIALS AND METHODS	32-35
RESULTS AND DISCUSSION	36-49
Microscopic Anatomy			
Mucosa			
Villi	37
Lamina propria	41
Muscularis Mucosae	42
Submucosa	43
Lamina Muscularis	46
Serosa	48
SUMMARY AND CONCLUSION	50-54
LITERATURES CITED	55-67

LIST OF TABLES.

	<u>Page</u>
Table No.1. Comparison of Structures of the small intestine of Domestic Animals-	30
2. Measurements of Structures of Duodenum-	43
3. Measurements of Structures of Jejunum-	46
4. Measurements of Structures of Ileum-	43

NUMBER OF PLATES - - 24.

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INTRODUCTION.

The domesticated water buffalo (*Bos bubalis*) has been known in the far east before written history and has migrated from the original area probably eastern India, to Burma and other countries of the far east (Ainsworth, 1958). The F.A.O. estimates (1958) puts the total buffalo population of the world at 96.4 millions, of which 45 millions or nearly half are in India. Buffaloes are most numerous in India, Pakistan, China, Malaya and South east Asian countries. Small numbers are also found in Egypt, Iraq, Turkey, Italy, Greece and elsewhere. The average annual milk yield per buffalo is 1100 lbs.

INTRODUCTION.

Buffalo's milk being 15 times as rich in fat content as cow's milk, is most valuable for butter and ghee production. They are also better utilizers of scrap fodder (Mottaccherya, 1958). Its capacity to convert feeding stuffs into milk is probably matched by another species of farm animals (Baker, 1958).

According to a folder issued by National Dairy Research Institute, Karnal, in the occasion of World Agricultural Fair (1954-55), in India buffalo milk forms 55% of 598 million pounds of milk produced in a year, whereas a research paper that 50% of the total milk production is contributed by buffaloes (Mottaccherya, 1958). Besides the milk the buffaloes also give skin and horn which are also form a major portion of the dairy products.

INTRODUCTION.

The domesticated water buffalo (*Bos bubalis*) has been known in the far east before written history and has emigrated from its original area probably eastern India, to Burma and other countries of the far east (Agarwala, 1962). The F.A.O. statistics (1958) puts the total buffalo population of the world as 95.4 millions, of which 45 millions or nearly half are in India. Buffaloes are most numerous in India, Pakistan, China, Malaya and South east Asian countries. Small numbers are also found in Egypt, Iraq, Turkey, Italy, Greece and Bulgaria. The average annual milk yield per buffalo is 1100 lbs. as against 413 lbs. given by an Indian cow, goes in favour of buffalo. Buffalo's milk being $1\frac{1}{2}$ times as rich in fat content as cow's milk, is more valuable for butter and ghee production. They are also better utilizers of coarse fodder (Bhattacharyya, 1954). Its capacity to convert feeding stuffs into milk is probably unequalled by another species of farm animals (Hafez, 1952).

According to a folder issued by National Dairy Research Institute, Karnal, in the pavillion of World Agricultural fair (1959-60), in India buffalo milk forms 55% of 528 million maunds of milk produced in a year, whereas a recent estimate puts that 52% of the total milk production is contributed by buffaloes (Makhijani, 1962). Besides its milk the buffaloes meat, hide, hoof and horn also form a major portion of the total production

by other animals.

The physiological value of small intestine is not a matter of controversy. The food taken by the animal comes in contact with pancreatic juice, bile and secretion of intestinal glands in the small intestine. The enzymes present in these secretions help in the digestion of protein, fat, carbohydrates and nucleoproteins and ultimately the absorption of the products of digestion takes place mainly from the small intestine.

Considering the importance, it is imperative to investigate the different aspects of Indian buffalo. So far, much attention has not been paid towards the microscopic structure of small intestine. Therefore, in order to acquire the precise knowledge and understanding about this organ in buffalo, it was considered worthwhile to study its histological structure. In most of the domestic animals histology of intestine and other organs is well known since long time but among the literature available, the author has seen none on small intestine of buffaloes, except one on Brunner's glands by Vaidya and Mariappa (1965).

This work has been done on adult buffaloes and buffalo-calves between one and two years of age. The study comprises the histological structure of small intestine in general and measurements of the villi, muscularis mucosae, submucosa, and lamina muscularis in particular. The height of the epithelium of

Brunner's gland and diameter of its lumen has also been measured. A comparison of structures between two age groups and also between the three portions of small intestine viz. Duodenum, Jejunum and Ileum has been made.

Although a detailed investigation is needed in this field, the author hopes that this study may prove of some help to the research workers, teachers and students of Anatomy.

*

REVIEW OF LITERATURE.

REVIEW OF LITERATURE

VILLI

Jordan (1940) found that the intestinal villi vary much in form, in different mammals and in different portions of the canal in the same individual. They were most highly developed in dog, where they formed long projections with expanded or club shaped extremities and a constricted base or neck.

Foust (1947) found that the villi were long and slender in carnivores, short and plump in ruminants and intermediate in horses and swine.

Blank (1950) found that in cotton rat the villi of the Jejunum were longer than those of duodenum and ileum.

Jacobson and Boer (1952) found gradual transition from closely packed, broad, blunt villi in jejunum to sparsely packed, flattened, leaf like roughly triangular villi in the lower ileum of rabbit. He also found ribbon like variety of villus in upper part of the bowel. In the dog, the villi were more uniform. They were cylindrical with rounded ends and were lying close together. On the antimesenteric border and occasionally in other portions of the circumference, short relatively broad villi were seen lying between the larger and more typical forms. In man the villi of ileum resembled to those of rabbit and were sparsely distributed and ~~were~~ usually conical in shape

with an occasional broad stubby variant.

Calhoun (1954) found that the villi in the small intestine of chicken sometimes branched twice. The villi of the duodenum were longest. She stated that Zietschmann, Batt and Clara had found that the villi in the chicken were tongue shaped, longer and more numerous in the duodenum.

Demke (1954) in a study of the histology of the intestine of Turkey Poult found that the villi were tallest in the duodenum and were somewhat shorter in the jejunum and ileum. Many of the villi were branching in the duodenum but not so in the jejunum and ileum.

Titkemeyer and Calhoun (1955) found that the villi of cat were much longer than in other species. The villi of dog were next in length, followed in order by those of the goat, sheep, pig, horse and cow. (Table No.1) He found that the villi of cat were very close together, quite slender and somewhat constricted at their base. The villi of dog were numerous and showed some branching. The distal end were expanded and club shaped. The villi of goat were wider at the base than those of carnivores. They were somewhat expanded at both ends, with a slight constriction between. In sheep and hog the villi were cylindrical and were arranged in rows. The horses had large broad villi rounded on their distal ends. The villi in the ox were slender and markedly constricted at their base.

Ham (1957) stated that in human the villi of duodenum were broader than those elsewhere and many were leaf like. Villi in the upper part of jejunum were tongue shaped. Further down they became longer and finger shaped. In the ileum they were fewer and still narrower.

Trautmann and Fiebiger (1960) stated that the carnivores possessed the longest and most slender villi and the ruminants had the shortest and thickest ones. The shape, size and number of villi varied in different sections of small intestine.

Baker et al. (1963) studied biopsy material of small intestine in healthy South Indian adults. They found that there were no finger like villi, though such were present in human foetuses. They also found that at birth the finger like villi were present throughout the small intestine of rat. Within 10 days, the villi in the upper jejunum were seen to be slightly leaf like in shape. These villi gradually became broader and broader as the animals became older. Finally, in duodenum and upper jejunum of the adult the villi were represented by a series of parallel ridges.

Suzuki et al. (1963) could see no villi in the lower straight intestine of loach but goblet cells were present.

Copenhaver (1964) stated that in man the villi

differ in shape in different parts of small intestine, being leaf shaped in duodenum, rounded in jejunum and club shaped in ileum.

Creamer (1964) examined twenty specimens of small intestinal mucosa, showing variations in villus shape from finger to leaves and to convolutions. He found that as the villus shape became more abnormal the adult epithelial cells covering them were greatly reduced in number. He suggested that the villus shape is plastic and remoulds to a more simple and economical shape as adult cells become fewer.

Vodovar (1964) found that the villi in the small intestine of pig were cylindrical at birth and during the first stage of their formation in the course of their growth, rapidly became conical and later assumed different shapes. The thickness of mucous and serous layer were more or less uniform throughout, increased with age.

Sehgal et al. (1966) found that there were three types of villi in the small intestine of Rhesus monkey, which could be encountered on dissection microscopy. Under first type, were classical finger like villi, the second one leaf like and the third type extra broad leaves curved at places. The duodenum showed a distinctive villus pattern, with little more than half showing broad curving branched villi. Finger like villi were not found in duodenum. The jejunum and ileum showed with a prominence of leaf-like villi. Many intermediary stages connecting one to

the other were also discernible.

Blood and Lymph Vessels of Villus -

Jacobson and Noer (1952) found that the circulatory pattern in all species bore a definite relationship to the morphology of the intestinal villus. They have studied the vascular pattern of the villus in rabbit, dog and man and found that there was a central vessel present in each animal which proceeded without branching from submucosal vessel to the tip of the villus. It divided there, one branch going to the capillary network and the other to form a precapillary arterio-venous anastomosis with the vein. In dog, precapillary anastomosis was not demonstrated. The capillary network was derived from submucosal vessels and formed arterial branch at the tip of the villus. The venous drainage was by one vein starting from the tip of the villus to the submucosal plexus. In dog, the venous drainage was by one or two veins, starting at the tip of villus and draining into the submucosal plexus.

Papp et al. (1962) were able to demonstrate the existence of a central chylous vessel in the intestinal villus of cat. The wall of the lymphatic capillary was formed by a continuous layer of flattened and irregular endothelial cells.

Kakharov (1963) described the peculiarities of blood and lymph vessels in jejunum and ileum of man. The

centre of each villus was occupied by a lymph sinus which was surrounded by a subepithelial blood capillary network. Each villus was supplied by one arteriole arising from mucosal or submucosal layer. Sometimes two arterioles were supplying one villus. The arteriole divided at middle or upper third of the villus to form a dense capillary network just beneath the epithelium. The venules were situated slightly deeper than arterial network. The central lymph sinus beginning at the tip of the villus represented the commencement of the lymphatic system of intestine. One to three lymph sinuses may be present in one villus depending upon the shape of the villus. At the bases of the villi the lymph sinuses broke up into 2 to 3 lymph capillaries which formed a network in the mucosa.

Villus Epithelium-

Machino et al. (1963) found that at the base of intermicrovillus space the cell membrane sometimes dipped into the underlying cytoplasm, presenting the form of a shallow vase in the intestinal epithelium of rat. Small vesicles appeared from just beneath this vase to the supranuclear and lateral region of the cytoplasm. Endoplasmic reticulum were always rough surfaced. The lateral cell membrane ran from the terminal bar to the basement membrane forming well developed folds or interdigitations but no intercellular spaces were detectable. Fat droplets were found enclosed by a limiting membrane.

Mitochondria of various shape were seen in the apical cytoplasm.

Suzuki et al. (1963) studied the histological structure of intestinal epithelium of loach. He found that the epithelium was composed of single layer of columnar cells. These cells were slender and had a striated border. The nucleus was round or oval being placed on the basal part of the cell. Mitochondria often assembled at the apical portion of the cell.

Sjostrand (1963) found that the plasma membrane of columnar cells of mouse intestinal epithelium was triple layered with a thicker opaque layer at its periphery. Two types of attached zones were found. Apical vesicles in the cytoplasm were smooth surfaced.

Shestopalova et al. (1963) found needle shaped structures with a crystalline type of internal structure in the cytoplasm of epithelial cells of the intestine of newborn cotton rat. Masses of crystals were present in the cytoplasm close to the nucleus.

Trier et al. (1963) found that apparently there were only two types of epithelial cells that covered the villus proper : the absorptive cells with their obvious cuticular border and columnar shape and the goblet cells in between the columnar absorptive cells. The brush border of the columnar absorptive cells had many slender long finger like microvilli.

The lateral margins of adjacent cells had undulating interdigitating plasma membranes, closely apposed to one another. Regions of specialized attachments- desmosomes, were present along these lateral membranes. The mitochondria were rod or filament shaped and endoplasmic reticulum with granular or agranular membranes.

Yamamoto (1965) found that the simple columnar epithelium of lamprey (*Lampetra japonica*) intestine were composed of three different kinds of cell : the striated border cells, the secreting cells and ciliated cells. The first type were most numerous and common throughout the entire intestine and frequently showed numerous small vesicles. The secretory cells were seen sporadically among the other cells and in all cases showed numerous small granules. The ciliated cells were found sporadically or grouped between other cells.

Micro-villi :

Blank (1950) found that the columnar cells losted their striated border near the opening of the crypt of Lieberkuhn in the intestine of cotton rat.

Granger and Baker (1950) found that the striated border on the free surface of the absorptive cells of the intestinal epithelium in rat was composed of very minute processes projecting vertically into the lumen. They were

columnar in form with rather blunt and rounded tips and were packed very closely together. It was calculated that a single cell bore nearly 3,000 processes and this increased the absorptive surface of the intestine about thirty times.

Dalton (1951) found that the height of striated border might change under varying conditions. The epithelial cells of the villi of duodenum showed filamentous mitochondria, to be very numerous in basal cytoplasm. The background of the cytoplasm was organized in the form of vertical striations, especially in the peripheral region of the cells. These striations in some instances appeared to be continuous with filaments of striated border.

Gol'din (1956) found that the cuticular membrane of small intestine in rat was made up of sticklike elements which appeared as full cylinders or pipes. The lumen consisted of consecutive enlargement and constrictions. The pipes appeared in groups of 3 to 5 and more, between them there was a somewhat larger lumen.

Oberti (1960) found that the microvilli, in the intestine of man, had a double limiting membrane which penetrated into the cytoplasm at the base of some of the microvilli. The microvilli showed a central axis running throughout their whole length.

Shestopalova et al. (1960, 1961) located fine

canaliculi or microtubules within the microvilli of epithelial cells, connecting with the canaliculi of adjacent microvilli and with the system of ergastoplasm. They found different types of microvilli. These microvilli were thought to play a role in secretion elaboration, vacuole formation and metabolism.

Brown (1962) found that the microvilli were short, wide and relatively few in crypts, became progressively higher, thinner and more numerous in the intervillus areas and in the villus crest of human jejunum.

Gottlieb et al. (1962) found that the microvilli in the jejunal crypt epithelium of rat were short, wide, sparse, loosely and irregularly distributed. A large number of elongated and closely packed microvilli appeared in columnar epithelium only, from the mid third to the tip of the villus.

Millington and Finean (1962) found that the core of the microvillus contained either paired strands or tubular structures. Lateral cross-fibers extended from the core to the microvillus membrane and it may be an essential part of the supporting structure.

Navarro-Berastegni et al. (1964) observed in man and other animals that the height and thickness of the microvilli changed, depending on their place in the intestine (apex, intervillus spaces and crypts).

-14-

Ito (1965) found that the microvilli of cat, bat, and man were coated with a conspicuous layer composed of fine filaments radiating from outer dense leaflet of plasma membrane. This surface coat was prominent on absorptive cells but was not so thick on the goblet and undifferentiated crypt cells. In other species the surface coat was poorly developed or inconsistent, but all intestinal microvilli had traces of such a coating over their tips and sides.

Intra cellular lymphocyte :

Andrew and Collings (1946), Andrew and Sosa (1947) observed intracellular position of lymphocytes in the distal end of epithelial cells in man and white mice. Definite change occurred in the lymphocytes and ultimate fate appeared to be extrusion in the lumen of intestine.

Kelsall (1946) found lymphocytes, usually between the nucleus and basement membrane of intestinal epithelium of hamsters.

Bloom and Fawcett (1962) stated that the irregularly distributed lymphocytes were seen penetrating from the lamina propria into the epithelium of the villi.

Andrew (1963) found lymphocytes penetrating the intestinal epithelium in man at all ages studied but few signs of disintegration of such cells were observed.

Renewal of epithelial cells :

Leblond and Stevens (1948), Leblond and Messier (1958), Bertalanffy (1960), Creamer et al. (1961) and Padykula (1962) found that there were continuous renewal of epithelial lining of the villi of small intestine. The cell find their way up along the sides of the villi and were extruded into the lumen, presumably from the villus tips.

Kaku et al. (1962) also found that the cells moved to the top of the villus and then fell into the lumen. Three types of cells were seen in the intestinal epithelium : the villus cells, Paneth cells and generative cells. Only the generative cells proliferated to supply cells to other zones. They differentiated into either a villus cell or Paneth cell. The environmental factor had dicisive influence on cellular differentiation.

Fabiny (1959) found that the improper fixation caused a contraction of the central core of the villi and formed a subepithelial space. A delay between death and fixation did not show marked difference.

Fell (1961) found that the epithelial cells became detached from the mucous membrane of the intestine, soon after death in sheep and rats.

Goblet cells :

Shearmann and Muir (1960) observed that the area of mucus synthesis was limited to the base of the goblet cells. The large goblet cells being only concerned with storage and transport.

Blerring (1962) and Freeman (1962) recognized the Golgi apparatus as the site of mucigen accumulation. Mucigen was formed within the granular lamellar membrane of the Golgi. The mucigen enlarged and coalesced with loss of individual mucigen droplet membranes. The cell bulged into its characteristic goblet appearance and subsequently microvilli plasma membrane ruptured with discharge of mucin.

Karwar (1962) found that mitochondria in the goblet cells of rat were filamentous with surface granules attached. Lipid granules and detached surface granules of the mitochondria became duplex as the secretory cycle proceeded. This served as a site for condensation of the mucus. The discharge of mucus from goblet cells was not explosive or continuous but gradual and intermittent.

Gopenhaver (1964) stated that the number of goblet cells were small in the duodenum and progressively greater in jejunum and ileum.

Freeman (1966) found that the endoplasmic reticulum

synthesized a protein which was transported to Golgi apparatus. There the protein was combined with acid mucopolysaccharides and glycoproteins, that are synthesized by Golgi apparatus, to form the mucus droplets.

LAMINA PROPRIA

Trier et al. (1963) identified four types of cells in the crypt region of small intestine of man : Paneth cells, goblet cells, undifferentiated crypt cells and enterochromaffin cells. Paneth cells were at the base of the crypts. Trier (1963) found that the undifferentiated cells at the base of the crypts contained lobulated nuclei and paranuclear accumulation of mitochondria. Argentaffin cells were also seen at the basal region of the crypt.

Paneth cells :

Blank (1950) found that the Paneth cells were generally present in the basal ends of the glands of Lieberkuhn all along the intestine.

Castro et al. (1959) found that very large number of Paneth cells were distributed throughout the small intestine of *Tamandua tetradactylus*. A functional cycle was demonstrated between the Paneth cells and goblet cells, whereby the latter originate from the former. An enzyme chitinase was found in

-13-

the granules of Paneth cells.

Trautmann and Fiebiger (1960) stated that the fundus of intestinal glands contained the specialized markedly granular cells of Paneth, which were absent in carnivores and swine.

Kaku et al. (1962) found that the Paneth cells in the bottom of the crypt, fell into the lumen of the crypt where they discharged their secretory granules.

Copenhaver (1964) stated that in man the Paneth cells were serozymogenic with characteristic striated chromophilic material at the base and large refractile acidophilic granules apically. In fasting animals the secretory granules accumulated and the basal striations became prominent, whereas during active digestion the cell was depleted of both its granules and its chromophilic material.

Staley and Trier (1965) observed actively secreting Paneth cells in preparation from fasting mice. The secretory activity was increased after feeding and after stimulation with pilocarpine. The actively secreting Paneth cells discharged their granules in the crypt lumen by merocrine secretion. The finer structures were the same in resting and in actively secreting cells.

Argentaffin cells :

Blank (1950), Finerty and Cowdry (1960), Coupland and Heath (1961) and Singh (1963) found that the argentaffin cells were more numerous in duodenum, few in jejunum and fewer in ileum.

Romeiser (1953) studied the argentaffin and argyrophile cells in pig, sheep, cow, cat, guinea pig, white mouse and white rat. In all animals argyrophile cells were found in all sections of stomach- intestinal tract. Argentaffin cells were found in all animals, but in some sections they were rare or quite absent.

Schofield (1953) suggested that in the small intestine in man the argentaffin cells were involved in a secretory cycle culminating in the formation of a mucous goblet cell, although not all mucous cells may necessarily arise in this manner.

Demke (1954) found argentaffin cells in the epithelium and tunica propria of the intestine in Turkey Poults.

Ham (1957) and Copenhaver (1964) stated that the argentaffin cells were present in small numbers among the crypt cells and occasionally among the surface epithelium.

-20-

Ghiringhelli and Mira (1960) found that in rats fed with bananas exclusively for seven days, there was an increase in the number of enterochromaffin cells in the duodenum. Cells localized in the lower blind end of the intestinal glands were particularly increased.

Trautmann and Fiebiger (1960) stated that the argentaffin cells were also present between the duodenal gland cells in all the domestic animals.

Coupland and Heath (1961) found argentaffin cells in the deeper layer of liver capsule and duodenum of ox, cow and sheep.

Gerzeli (1961) found that in amphioxus the argentaffin cells were slender elements occupying the total height of intestinal epithelium.

Singh (1963) found that the bodian positive argyrophile cells were present in the epithelium of cardiac end of stomach, duodenum and proximal part of jejunum at 23 mm. stage of human fetuses. Argyrophile cells represented a stage in the maturation of argentaffin cells. Following their first appearance the density of the enterochromaffin cells in the epithelium increased to a peak level at about 120-140 mm. stage and thereafter decreased.

Blank (1950) found that in cotton rat the lamina propria of duodenum contained deffuse lymphoid tissue. In many areas of the tunica mucosa the lymphatic tissue were dense, forming solitary ovoid nodules.

Guelve (1961) found a nerve plexus located between the muscularis mucosae and the base of the glands or crypts. Ganglia and solitary nerve cells were found along the course of bundles of the plexus in all sector of the gastro intestinal tract of cow, horse and pig. In small intestine the dense network of plexus were formed by finer bundles.

Trier et al. (1963) found that in addition to the lymph and blood vessels the lamina propria or the connective tissue core of the villus normally contained smooth muscle, small unmyelinated nerve fibers, lymphocytes, plasma cells, eosinophils, mast cells and connective tissue elements.

Andrew (1963) found that the cell population in the lamina prpria of young individuals were dense. Plasma cells were found with many stages in development.

Deane (1964) found in mouse that the lamina propria were highly cellular. A continuous layer of attenuated fibroblasts were seen below the basement membrane of the epithelium. Fibroblasts in mitotic division were seen. Plasma cells lying in direct contact with macrophages and eosinophils were seen

in large numbers.

Honjin et al. (1965) were able to demonstrate fine nerve bundles between the blood vessels, intestinal glands and proper cells of the lamina propria in man. The fine axons in the bundle lost their sheath and passed into grooves on the surface of the intestinal cell of Cajal, to form slightly swollen expansions which contained many agranular synaptic vesicles.

LAMINA SUBGLANDULARIS.

Titkemeyer and Calhoun (1955) found in dog an extra connective tissue layer between the muscularis mucosae and crypt of Lieberkuhn. This zone of connective tissue was 30 microns in thickness and completely encircled the intestine.

Trautmann and Fiebiger (1960) stated that a thin lamina subglandularis was present in horses and carnivores, lying between the blind endings of gland of Lieberkuhn and muscularis mucosae. In carnivores this layer was subdivided into stratum granulosum with many leucocytes and a nearly homogeneous stratum compactum bordering on the muscularis mucosae.

MUSCULARIS MUCOSAE

Blank (1950) found in cotton rat that the lamina

muscularis mucosae was broken in the region where Brunner's gland was present, because these glands extended through it into the tela submucosa. In places where Brunner's gland were lacking, this layer was lying along the bases of the gland of Lieberkuhn.

Calhoun (1954) stated that the muscularis mucosae in chicken was comprised of an outer circular and an inner longitudinal layer. The latter sent fibers into the villi. In places the outer circular layer appeared to fuse with the circular layer of the lamina muscularis.

Titkemeyer and Calhoun (1955) found that in all cases viz. cow, horse, pig, sheep, dog, cat and goat, muscularis mucosae comprized of two layers- an inner circular and outer longitudinal one.

Trautmann and Flebiger (1960) stated that in domestic animals muscularis mucosae was composed of smooth muscle fibers arranged in two sheets perpendicular to one another. In areas of duodenal glands and in some cases in the area of Peyer's patches this layer was discontinuous and even absent in part because it splits in seperate strands which dip into the glandular layer.

Lane and Rhodin (1964) found that in mouse this smooth muscle element was a three-dimensional network of flattened cells with elongate processes. The processes of

each cell were containing those of its neighbours. The gland of Lieberkuhn were enmeshed in the net of muscle cells.

S U B M U C O S A.

Denke (1954) found in Turkey Poult that the submucosa was poorly developed. No Brunner's gland were noted in the sections examined. A few nodules and diffuse areas of lymphoid tissue were found in the duodenum.

Berry (1962) found in dog that the submucosa of small intestine was composed of interlacing collagenous fibers, which in the neutral and relaxed state consisted of spiral coils. Half of the collagenous strands ran clockwise and half counter-clockwise.

Kakherov (1963) found that in man the lymphatics of different sizes formed lymphatic plexus in the submucosa. The lymphatic vessels of submucosa formed anastomosis with capillaries and vessels of serosa. Arterial plexus of submucosa gave origin to mucosal, muscular and serosal branches.

Alcantara and Oliveira (1964) found that there were a greater number of neurons in duodenum and jejunum in comparison with other segments of intestine of wister rat. Neuron population was found to decrease as the more distal portion of the digestive canal was reached.

Norris et al. (1963) found that the mast cells were more numerous in the mucosal and submucosal layers of stomach, duodenum and ileum and much less frequent in the large bowel of man. No appreciable number of mast cells were observed between fourth and ninth hours after death.

Astaldi et al. (1964) found that the number of mast cells, in the submucosa of human intestine, varied from one case to another. In the lamina propria mast cells were in large numbers in some cases while in others they were totally absent.

Brunner's gland : and Peyer's patches :

Blank (1950) found that in cotton rat the Brunner's glands were extremely abundant near the junction of pylorus and duodenum. The terminal portions of the Brunner's gland were highly branched and coiled and were invested in fibrous connective tissue. The secretory portions of the gland were composed of low columnar cells with their nuclei at the base.

Calhoun (1954) found that there were no Brunner's gland and Peyer's patches in the intestine of chicken.

Moe (1960) observed that the cells of the Brunner's gland in rat were provided with microvilli and had a triple layered plasma membrane. Their cytoplasm contained mitochondria, RNA particles, a Golgi apparatus and secretory granules. The

-26-

secretory granules seemed to be produced by Golgi apparatus.

Cochrane et al. (1964) found that in guinea pig the aciner cells of the Brunner's gland were lying on the basement membrane and were closely interlocked laterally, a few desmosomes were also present. The luminal border of the cells were devoid of microvilli. The apical part of the cell was packed with secretion droplets. The nucleus was situated basally and there was extensive Golgi apparatus.

Vaidya and Mariappa (1965) found in Indian buffaloes that the Brunner's gland extended upto 35 cm. beyond the pylorus.

Trautmann and Flebiger (1960) stated that the Peyer's patches and solitary nodules were larger and more in number in young animals than in adults.

Cornes (1965) examined specimens of normal small intestine from patients between 15 and 95 years of age. Lymphoid follicles were over 200 before 20 years of age to about 100 between 70 to 95 years of age. With increasing age, there was a loss of follicles giving patches of unusual shape and appearance.

LAMINA MUSCULARIS.

Titkemeyer and Calhoun (1955) and Trautmann and

-27-

Fiebigler (1960) found that in dog there was an extra oblique layer of muscle fibers in between the submucosa and inner circular layer. In other animals there was only two distinct layers.

Yamagishi (1960) found that there were slight differences in the fine structure of the small blood and lymph vessels of the muscular and serous coats of the small intestine of cat fish, frog, snake, domestic fowl and dog, the basic structure being the same.

Taxi (1961) observed intimate contacts between nerve fibers and smooth muscle cells in the intestine of mice. Sometimes such nerve fibers were found in a depression of the muscle cell surface. Numerous contact zones between muscle cells were also seen.

Kakharov (1963) found that in man the intra muscular network of lymphatic capillaries gave origin to lymphatic vessels of the lateral aspect of the intestine. The vascular supply of lamina muscularis were by straight and recurrent branches as well as by a number of arterioles coming directly from the mesentery.

Dupont and Sprinz (1964) found that in guinea pigs the granular polydendrocytes, identical to Cajal's interstitial neurons, formed a wide spread fine network

-23-

throughout the mucular coat. The network crossed the primary fiber plexus (Auerbach's plexus) in all directions. There appeared a point of junction of these two networks.

Oosaki and Ishii (1964) found that there were two kinds of contact zones between smooth muscle cells in the lamina muscularis of small intestine in rat, those with fusion of adjoining plasma membranes and those with intercellular attachments. The pattern of the former was characterised by fusion of adjoining plasma membranes resulting in obliteration of intercellular space while that of the latter by the presence of an intercellular space occupied by a homogeneous and amorphous material.

Auerbach's plexus :

Richardson (1958) described the structure of Auerbach's plexus in the small intestine of rabbit. It was found that the Auerbach's plexus took the form of a wide network of ganglia and primary nerve bundles. The interstices of this network were traversed by thinner secondary nerve bundles and close tertiary plexus of continuous strands. Another network of less well defined pattern intermingling with the tertiary plexus were seen. This comprised of cells with extremely long processes intricately interwoven among themselves and occuring at various levels with respect to the secondary

-29-

and tertiary bundles. These star shaped cells appeared to constitute the network of interstitial cells (Described by Cajal, 1893). The remaining occupants of the narrow interval, between the two muscle coats were blood capillaries and occasional connective tissue cells. The nerve bundles from tertiary plexus entered into the muscle coats accompanied by blood capillaries and interstitial cells. These three structures formed a plexus among the muscle fibers.

Leaning and Cauna (1961) found that the nerve cells in the Auerbach's plexus of jejunum of cat were much more than in the duodenum. The ratio between deeply and lightly staining nerve cells were 1 : 1.9 in both duodenum and jejunum.

Kakharov (1962) after injecting a dye in the muscular layer of small intestine of fetuses, children and adults of both sexes could observe not only the lymphatic capillaries but also the peripheral spaces surrounding the Auerbach's plexus. The perineural spaces of the tiny nerve filament emerging from Auerbach's plexus were also seen, which formed direct connection with the lymphatic system. These findings confirmed that the humeral connections were present at the periphery of the body between autonomic and lymphatic systems.

S E R O S A.

Finerty and Cowdry (1960), Bloom and Fawcett (1962)

TABLE NO.1
Comparison of Structures of the small Intestine of Domestic Animals *

	<u>Horse</u>	<u>Cow</u>	<u>Sheep</u>	<u>Goat</u>	<u>Pig</u>	<u>Dog</u>	<u>Cat</u>
Height of villi (μ)	405	363	483	615	470	645	963
Thickness of muscularis mucosae (μ)	26	19	24	15	23	71	15
Lumen diameter of Brunner's gland (μ)	7	25	40	50	3	35	20
Height of epithelium of Brunner's gland. (μ).	15	12	20	35	25	35	10
No. of layers in Tunica muscularis.	2	2	2	2	2	3	3
Thickness of Tunica muscularis.	1,153	583	239	175	520	701	555
Location of Peyer's patches.	Jejunum, Ileum.	Jejunum, Ileum.	Jejunum, Ileum.	Jejunum, Ileum.	Jejunum, Ileum.	Duodenum, Jejunum, Ileum.	Jejunum, Ileum.

*Titkemeyer and Calhoun (1955).

and Copenhaver (1964) stated that the serosa in the small intestine in man comprised of loose connective tissue, covered by a layer of mesothelium.



MATERIALS AND METHODS.

MATERIALS AND METHODS

SOURCE OF ANIMALS

For this study, the small intestine from 12 buffaloes were used. Included in this group were six he-buffalo/-calves from one to two years of age and six adult she-buffaloes.

The materials from buffalo-calves were collected from the Departments of Anatomy and Surgery, Bihar Veterinary College, Patna and from adult she-buffaloes, from slaughter house, Sahganj, Patna. Because of the non-availability of the material from adult he-buffaloes the investigation could not be made on this group of animal.

These animals were in good nutritional condition and appeared to be free from disease.

TECHNIQUE

Fixation and Processing of the Tissues.

Sections were taken from the tract, every tenth distance irrespective of the over all length. The first section was taken as near the pylorus of the stomach as possible and the last one was taken next to the ileo-caecal junction. The other eight sections were taken equidistant between them. It was impracticable to remove the materials immediately after death of the animal at slaughter house or the materials which

-33-

were taken from Surgery or Anatomy Departments. The materials which were obtained from Department of Surgery were fixed usually within two and half hours after the death of the animal. The materials from Department of Anatomy were collected from embalmed animals meant for dissection. However, they were fixed, upon their removal from carcasses, in 10% formaldehyde solution (1 part commercial formalin and 9 parts water).

Small blocks of one centimeter width and about 1.5 centimeter length were cut to show the cross section of small intestine. The blocks were cut in such a fashion that the length of the blocks corresponded to the length of the small intestine, for convenience at the time of embedding.

The blocks were washed in running tap water for 3 to 12 hours and were then put into 70% ethyl alcohol for 12 hours. Dehydration and infiltration were done step by step in the following manner.

1. Normal butyl alcohol and 95% ethyl alcohol (equal parts) 4 hours.
2. Normal butyl alcohol No.1. 4 hours.
3. Normal butyl alcohol No.2. 16 hours.
4. Normal butyl alcohol and 52° c Paraffin (equal parts). 2 hours.
5. 52° c Paraffin No.1. 2 hours.

- | | |
|-------------------------|-----------|
| 6. 52° c Paraffin No.2. | 2 hours. |
| 7. 60° c Paraffin No.3. | 13 hours. |

The infiltrated tissues were then embedded in 60° c Paraffin (B.D.H.).

Sectioning

All sections were cut at seven microns.

Staining and Mounting.

The following stains were used-

1. Harri's Hematoxylin^{and Eosin}- Routine stain.
2. Weigert and Van Gisson's stain (Mallory, 1942)- for collagenous tissue and muscle fibers.
3. Gallego Iron Fuchsin stain (Lillie, 1948)- for collagenous tissue, elastic fibers, muscle fibers and mast cell granules.
4. Fontana -Masson stain (Lillie, 1948)- for granules of enterochromaffin cells.
5. Gridley's reticulum stain (Gridley, 1951)- for reticular fibers.

The stained sections were mounted in canada balsam.

Methods of Measurement.

Measurements of the villi were made from the base

to the tip of the villus. Muscularis mucosae, sub mucosa and lamina muscularis were measured at their thickness. The diameter of the lumen of alveoli of Brunner's gland were measured at the centre and the height of its epithelium were also measured.

All the measurements were made from random samples of those structures seen under the light microscope.

Twenty measurements of same structure in the same animal in different slides were taken. An average of twenty measurements of a particular structure in each animal were taken. The mean of the averages of all measurements in different animals gave the final average measurement of that particular structure.

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RESULTS AND DISCUSSION

RESULTS AND DISCUSSION

Small intestine, as a general rule, is divided into three portions viz. duodenum, jejunum and ileum. Although grossly there is no demarcation between the jejunum and ileum but the last 3 to 4 feet is considered as ileum as stated by Sisson and Grossman (1959).

The external surface is highly vascular and a large number of Blood vessels and capillaries are visible, so much so that this surface appears reddish brown in colour. The internal surface presents longitudinal and circular or oblique folds. Titkemeyer and Calhoun (1955) observed longitudinal folds in all the domestic animals studied, except the ox. The circular folds or plicae circularis are parallel to each other and becomes faint or are altogether absent towards the mesenteric border. Longitudinal folds run longitudinally, are interrupted at places and lie parallel to each other. These folds are also absent towards the mesenteric border of the small intestine. The surface of the mucous membrane over and between the folds is covered with projections- the villi. Peyer's patches are present in the jejunum and ileum only. They are elongated in shape. No Peyer's patches are grossly visible in the duodenum but are sometimes present microscopically in buffalo-calves.

MICROSCOPIC ANATOMY.

The wall of the small intestine is divided into mucosa, submucosa, lamina muscularis and serosa. Plicae

circularis and longitudinal folds of mucous membrane are present. The mucosa is composed of projections of different shape and size called as villi which is lined with epithelium. The second layer of mucosa is lamina propria which contains the intestinal glands or glands of Lieberkuhn and also forms the core of the villi. The third layer is lamina muscularis mucosae which consists of a continuous layer of smooth muscle fibers and forms a boundry between lamina propria and submucosa.

M U C O S A.

Villi

A microscopic study of histological sections prepared from various levels of intestinal tract has revealed numerous variations. The shape and height of the villi varies with age, different portions of small intestine and individual to individual.

In duodenum the villi of buffalo-calves are leaf-like or tongue shaped. Some of the villi are pointed at their tips. The bases are usually wider than the tips. Very few of them are finger like. In adults, the villi become blunt at their tips and are mostly tongue shaped.

In jejunum and ileum of buffalo calves the villi are mostly finger like but some are pointed at the tips. In adults, the villi are constricted at the base. They become

-33-

progressively wider upto the upper third and are pointed and slightly curved at the tips. Some are typical finger like. In distal portions of small intestine the villi are mostly finger like with their blunt ends.

The average height of the villi in the duodenum and ileum is greater in adults than in buffalo-calves whereas in jejunum villi are smaller in adult buffaloes than in the buffalo-calves. They are on average 519 /u and 527 /u in duodenum, 737 /u and 607 /u in jejunum and 436 /u and 559 /u in ileum of buffalo-calves and adult buffaloes respectively. The villi of jejunum are longest among the villi of all the portions of small intestine.

Each villus consists of a loose connective tissue core and an epithelial covering. The connective tissue core of the villus is composed of collagenous and reticular fibers and connective tissue cells. Fibroblasts, plasma cells, macrophages, eosinophils and large number of lymphocytes are present. Mast cells have not been observed under this investigation.

A single lymph vessel- the central lacteal, with endothelial lining is present at the centre of the each villus. Besides these, smooth muscle fibers are also present in the core of the villus.

Epithelium-

The epithelium which covers the villi is composed

-39-

of a single layer of columnar cells. The basement membrane and the cell membrane are not clearly demarcated. The epithelium consists of two types of cells, the columnar absorptive cells and the goblet cells. The free surface of the epithelium bears a striated border which looks like a homogeneous layer under light microscope.

The cytoplasm of the absorbing columnar cells looks finely granular under high magnification. The Granules are aggregated at the apical portion of the cells and take deeper stain. The nuclei are oval or rounded in shape and lie at the base of the cells. The nucleus contains one or two nucleoli and they lie at one corner of the nucleus or may sometimes be in close contact with the nuclear membrane. Chromatin granules are irregularly arranged and are thickly situated at the zone of nuclear membrane.

Intracellular lymphocytes are present at the apical portion of the absorbing cells in buffalo-calves and adult buffaloes. Lymphocytes penetrating into the epithelium from the core of the villus have been observed in some of the sections, and this tallies with the observations of Andrew et al. (1946, 1947), Kalsall (1946), Bloom and Fawcett (1962) and Andrew (1963).

Goblet cells are situated between the columnar absorbing cells and are very few in number as compared to other animals or are even sometimes absent.

It is observed that the delay in fixation of the materials causes detachment and shedding off of the epithelium from the mucous membrane. The minimum time interval between death of the animal and fixation during which this type of change occurs in this species have not been ascertained. A more detailed investigation is needed on this aspect. Such changes were observed by Fell (1961) in the intestine of sheep and rat.

Crypt of Lieberkuhn-

The epithelium covering the villi continues into the glands of Lieberkuhn. The majority of the cells which form the wall of the gland is composed of undifferentiated columnar cells. The nuclei are basally situated and are oval in shape. Cells under mitotic division are present in the crypts of Lieberkuhn in both young and adult animals.

Goblet cells are distributed in between the undifferentiated columnar cells of the crypts. They are fewer among the crypt cells also as compared to other animals or are even sometimes absent.

Cells of Paneth have not been observed under this investigation.

Argentaffin or enterochromaffin cells have been observed by using special stains (Fontana Masson stain). These cells are numerous in the crypts of Lieberkuhn in duodenum of adult animals but are very rarely present in buffalo-calves.

In jejunum and ileum these cells have not been observed among the sections examined. They are rarely present in the villus epithelium. These cells lie singly among the crypt cells. The granules of the enterochromaffin cells stain deep black and are aggregated usually at the basal region of the cell. Sometimes these granules surround the nucleus.

Lamina Propria.

Lamina propria of the mucous membrane fills the space between the glands of Lieberkuhn and forms the core of the villi. It consists of loose connective tissue and a network of reticular fibers. Among the connective tissue cells fibroblasts, macrophages, plasma cells and eosinophils are present. A large number of lymphocytes are infiltrated among the connective tissue cells. Both small and large lymphocytes have been seen. Mast cells have not been observed. Blood capillaries are always present in all the portions of small intestine. A central lacteal lined with endothelium have been observed in the centre of each villus. Smooth muscle fibers are also present in the core of the villi. Blood vessels penetrating the lamina muscularis mucosae and extending upto lamina propria are sometimes present.

Lamina propria in the duodenum presents Brunner's gland which has extended from the submucosa. Solitary lymph nodules are occasionally present in this layer in all parts of

small intestine. Some times these nodules extend into the core of the villi.

Muscularis Mucosae.

Muscularis mucosae forms a boundry between lamina propria and submucosa. In adults, this is in the form of a continuous layer of smooth muscle fibers arranged in two rows. The fibers of inner layer are arranged circularly and the outer one longitudinally. In young animals lamina muscularis mucosae is not well developed and is interrupted at various places. Two layers are not always present in calves. Some times in adults also this is represented by a single layer of circular fibers.

Muscularis mucosae extends into plicae circularis and longitudinal folds of the mucous membrane, along with the submucosa. In most of the cases it is interrupted at places where the Brunner's glands or Peyer's patches are present. These structures extend into the lamina propria through interruptions of lamina muscularis mucosae. In some cases muscularis mucosae, at the region of longitudinal folds of mucosa, sends muscle fibers into the submucosa and forms a network of smooth muscle fibers.

This layer is thickest in the ileum and thinnest in the jejunum of both young and adult animals. It is 26 μ and 42 μ in duodenum, 17 μ and 41 μ in jejunum and 41 μ and

59/u in the ileum of buffalo-calves and adult buffaloes respectively.

T A B L E NO.2

Measurements of structures in Duodenum.

	<u>Buffalo-calves</u>		<u>Adult Buffaloes</u>	
	<u>Range</u>	<u>Mean</u>	<u>Range</u>	<u>Mean</u>
Villi (/u)	444-693	519	339-598	527
Muscularis mucosae (/u)	15- 33	26	23- 54	42
Submucosa (/u)	190-670	370	235-485	377
Diameter of lumen of Brunner's gland (/u)	9- 31	17	9- 14	12
Height of Epithelium of Brunner's gland (/u)	9- 16	13	13- 19	13
Lamina Muscularis (/u)	590-2203	1070	1931-2723	2331

S U B M U C O S A

Submucosa consists of loosely arranged connective tissue, larger blood vessels and nerves. This layer extends into the plicae circularis and longitudinal folds of the mucous membrane along with the muscularis mucosae. It is highly supplied with blood vessels, capillaries and nerves. Meissner's plexus along with the ganglion cells have been observed. In the meshes of collagenous and elastic fibers connective tissue cells are distributed. No gland has been observed in the submucosa except the Brunner's gland in duodenum.

Submucosa is thickest in the duodenum of adult buffaloes and the ileum of buffalo-calves. It is thicker at places where Brunner's glands or Peyer's patches are present. Thickness of submucosa is 370 μ and 377 μ in duodenum, 167 μ and 302 μ in jejunum and 741 μ and 340 μ in the ileum of buffalo-calves and adult buffaloes respectively. In the ileum of young animals its thickness is greatest and is due to the presence of larger Peyer's patches, some times occupying the whole of the thickness of submucosa.

Peyer's Patches-

Peyer's patches are well developed in the ileum of buffalo-calves. They are aggregation of large number of lymphoid follicles and in some cases almost occupies the whole of the thickness of submucosa. Some times they penetrate into the muscularis mucosae and extend upto the lamina propria. In adults, they are rarely in the form of patches. They are usually in the form of solitary follicles or are diffused through a wide space of sub mucosa. Microscopically they are present in the jejunum and ileum and sometimes in the duodenum of young animals.

In one buffalo-calf the follicles of the Peyer's patches have been observed surrounded by smooth muscle fibers.

Brunner's Gland-

Brunner's glands have been observed in the first

section and upto 45 cm. extending beyond the pylorus. Vaidya and Mariappa (1965) observed Brunner's gland, in the duodenum of Indian buffaloes, extending upto 85 cm. beyond the pylorus. Sisson and Grossman (1959) stated that these glands extend throughout the first 4.0 to 4.5 m. in the small intestine of ox. It is probable that they are present but may be distributed thinly enough to be missed when sections are taken every tenth distance only.

These glands are distributed in the submucosa and also extend into the lamina propria. The alveoli of the gland can be differentiated from the gland of Lieberkuhn by its faint staining. Loose connective tissue surrounds the gland and its alveoli. Alveoli are lined with low columnar cells. Cells are wider at the base and narrower at the apex. No striations lining the epithelium of alveoli were visible. Cell membrane is clearly demarcated. The nuclei are oval or rounded in shape and lie close to the basement membrane. Ducts of the gland are lined with simple columnar epithelium. They open into the crypts of Lieberkuhn or between them.

In buffalo-calves alveoli are loosely distributed with a wide space between them but in adults they lie close to each other. The diameter of the lumen of alveoli is larger in young animals than in adults, whereas the height of its epithelium is greater in adults than in young ones. The average diameter of the lumen of Brunner's gland is 17 μ in buffalo-

calves and 12 μ in adults. The height of epithelium is 13 μ in buffalo-calves and 13 μ in adults. It seems that as the height of the epithelium increases, the lumen becomes progressively smaller with the increase in the age of the animal.

No Paneth, goblet or enterochromaffin cells have been observed in the Brunner's glands.

Trautmann and Fiebiger (1960) stated that Paneth cells were present in horse, goblet cells in ox, horse and sheep and enterochromaffin cells in all the domestic animals.

TABLE NO.3

Measurements of structures in Jejunum.

	<u>Buffalo-calves</u>		<u>Adult Buffaloes</u>	
	<u>Range</u>	<u>Mean</u>	<u>Range</u>	<u>Mean</u>
Villi (μ)	700-900	737	359-764	607
Muscularis Mucosae (μ)	10- 23	17	35- 46	41
Submucosa (μ)	103-268	167	170-396	302
Lamina Muscularis (μ)	411-364	602	951-1785	1343

L A M I N A M U S C U L A R I S

The lamina muscularis consists of two well defined layers of smooth muscle fibers- an inner circular and outer longitudinal. Outer longitudinal layer is thinner than the inner circular layer. There is a definite connective tissue layer called lamina intermuscularis, in between these two

layers. Lamina muscularis is thickest in the duodenum of both young and adult animals. Its average thickness is 1070 μ and 2331 μ in duodenum, 602 μ and 1343 μ in jejunum and 661 μ and 1582 μ in the ileum of buffalo-calves and adult buffaloes respectively.

In the inner circular layer in almost all the sections of small intestine and in both young and adult animals, empty spaces are present. These spaces run parallel and cross-wise to the fibers of the circular layer. Trautmann and Flebiger (1960) and Kakharov (1963) reported the presence of lymphatic network in the lamina muscularis which connect with the lymphatics of the submucosa and serosa. It is probable that the spaces observed under this investigation are lymph spaces which have emptied during the processing of the tissue.

Fibers of the outer longitudinal layer are arranged in bundles with loose connective tissue between them.

Lamina intermuscularis contains collagenous fibers and some connective tissue cells. Blood vessels and nerves are usually present. This layer also presents the plexus of Auerbach and ganglion cells.

Large blood vessels and capillaries have been observed in both the inner circular and outer longitudinal layers. Vessels entering the lamina muscularis from serosa have been observed in some of the sections.

Auerbach's Plexus-

Auerbach's plexuses are usually situated in the lamina intermuscularis but in many cases they lie at various depths of the outer longitudinal layer of lamina muscularis. They are abundant and in one view even 3 to 4 of them are visible under low magnification. The Auerbach's plexus, as seen under light microscope, is composed of deeply and faintly staining neurons enmeshed in a network of fine nerve filaments. Large ganglion cells are present in some of the plexuses and are situated at one corner of them. Sometimes very small blood capillaries are visible in the plexus. The plexus is enclosed in a capsule of regularly arranged flat cells internally and a layer of fibroblasts externally.

TABLE NO.4

Measurements of structures in Ileum.

	<u>Buffalo-calves</u>		<u>Adult Buffaloes.</u>	
	<u>Range</u>	<u>Mean</u>	<u>Range</u>	<u>Mean</u>
Villi (A)	312-639	486	425-668	559
Muscularis Mucosae (A)	33- 73	41	45- 79	59
Submucosa (A)	86-2192	741	252-339	340
Lamina Muscularis (A)	516-898	661	1155-1733	1582

S E R O S A.

Serosa is composed of loose connective tissue, covered. Connective tissue consists of collagenous fibers and connective

-49-

tissue cells. Fibroblasts are very few in number. Macrophages and plasma cells are also less in number. Some granular leucocytes are also visible. The lymphocytes are infiltrated among these cells but are fewer in number than lamina propria and submucosa. Collagenous fibers are loosely and irregularly arranged. Large number of blood vessels are present in this layer and some of them penetrate into the lamina muscularis. In one case a solitary lymph follicle have been observed which was lying close to the external layer of lamina muscularis.

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THEORY OF THE EXPERIMENT

The purpose of this experiment is to determine the effect of the concentration of a solution on the rate of reaction. The reaction studied is the reaction between hydrogen peroxide and potassium iodide in the presence of a catalyst. The rate of reaction is measured by the volume of oxygen gas evolved over a fixed period of time. The concentration of the hydrogen peroxide solution is varied, while the concentration of the potassium iodide solution and the temperature are kept constant. The results show that the rate of reaction increases with increasing concentration of hydrogen peroxide.

SUMMARY AND CONCLUSION.

The results of the experiment show that the rate of reaction increases with increasing concentration of hydrogen peroxide. This is in agreement with the theory that the rate of reaction is proportional to the concentration of the reactants. The experiment also shows that the rate of reaction is not affected by the concentration of the potassium iodide solution or the temperature.

The overall purpose of the experiment is to determine the effect of the concentration of a solution on the rate of reaction. The reaction studied is the reaction between hydrogen peroxide and potassium iodide in the presence of a catalyst. The rate of reaction is measured by the volume of oxygen gas evolved over a fixed period of time. The concentration of the hydrogen peroxide solution is varied, while the concentration of the potassium iodide solution and the temperature are kept constant. The results show that the rate of reaction increases with increasing concentration of hydrogen peroxide.

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SUMMARY AND CONCLUSION.

1. For this study materials from six adult buffaloes and six buffalo-calves were taken. Sections were cut every tenth distance irrespective of the overall length of the small intestine. The first section was taken near the pylorus and the last one next to ileo-caecal junction. They were fixed in 10% formalin solution. Dehydration and infiltration were done by the method described in the text and blocks were prepared. Sections were cut and stained in Hematoxylin - Eosin stain. Special stains were also used for differential staining.

2. A general survey of the histological sections were made and measurements of the villi, muscularis mucosae, submucosa, lamina muscularis, diameter of the lumen of alveoli of Brunner's glands and the height of its epithelium were taken.

3. The external surface of the intestine is highly vascular and it appears reddish-brown. Internal surface presents plicae circularis and longitudinal folds which are absent towards the mesenteric border. Peyer's patches are grossly present in the jejunum and ileum only but sometimes it is also present in the duodenum of young animals when examined microscopically.

4. The shape and height of the villi varies with age, their location in the small intestine and individual to individual. In the duodenum of young animals they are leaf-like

or tongue shaped. In adult, they become blunt at their tips and are mostly tongue shaped. In jejunum and ileum of young animals villi are mostly finger like but some are pointed at their tips. In adults, they are constricted at their base, become progressively wider towards the upper third and are pointed and slightly curved at their tips. In adults, the villi of duodenum (527 μ) and ileum (559 μ) are higher than in young ones (519 μ) and 436 μ respectively), whereas in jejunum they are higher in young animals (787 μ) than in adults (607 μ). Villi of jejunum are highest among the three portions of small intestine. Central lacteal and smooth muscle fibers are present in the core of the villi.

5. The villus epithelium consists of single layer of columnar cells and goblet cells. Crypt of Lieberkuhn contains undifferentiated crypt cells, goblet cells and enterochromaffin cells. Paneth cells have not been observed in this investigation. Enterochromaffin cells are usually present in the crypts of Lieberkuhn in the duodenum of adults and have not been observed in other portions of small intestine. Goblet cells are very few in number or even absent among both the villus and crypt epithelium.

6. Intracellular lymphocytes are present among the villus epithelium of both young and adult animals. They are also seen penetrating the epithelium from the core of the villus.

7. Cells under mitotic division have been observed in the crypts of both young and adult animals.

8. Delay in fixation of the tissues has caused shedding of the epithelium from the core of the villi.

9. The lamina propria contains large number of lymphocytes and other connective tissue cells in the meshes of collagenous and recticular fibers. Solitary lymph nodules are present throughout the small intestine. Brunner's glands are present only in the duodenum.

10. In young ones muscularis mucosae is usually represented by single layer of circular fibers but in adults it is mostly in the form of two layers. This layer is thickest in the ileum (41 μ and 59 μ) and thinnest in the jejunum (17 μ and 41 μ) of young and adult animals respectively. In duodenum it is 26 μ and 42 μ in young and adult animals respectively.

11. The submucosa is thickest in the ileum (741 μ) of young animals. In adults it is thickest in the duodenum (377 μ). It is 370 μ in duodenum of calves, 167 μ and 302 μ in jejunum of young and adults respectively and 340 μ in the ileum of adult animals.

The submucosa extends into the plicae circularis

and longitudinal folds along with the muscularis mucosae. This layer is supplied with large blood vessels and nerves. Meissener's plexus and ganglion cells are also present.

12. No glands other than Brunner's glands in the duodenum have been observed. Lumen of alveoli of Brunner's gland are larger ($17/\mu$) in young animals than in adults ($12/\mu$) whereas the height of its epithelium are larger in adults ($13/\mu$) than in young ($13/\mu$) animals.

13. Peyer's patches are well developed in the ileum of young animals and are in the form of nodules in adults. They are present in the jejunum and ileum of both and sometimes in the duodenum of young animals.

14. Lamina muscularis consists of two definite layers with a connective tissue layer known as lamina inter-muscularis in between. Inner circular layer presents empty spaces, probably lymph spaces. The outer longitudinal layer is in the form of bundles. Blood vessels and reticular fibers have been observed in between the muscle fibers. The lamina muscularis is thickest in the duodenum of both young and adult animals. Its average thickness is $1070/\mu$ and $2331/\mu$ in duodenum, $602/\mu$ and $1343/\mu$ in jejunum and $661/\mu$ and $1532/\mu$ in the ileum of young and adult animals respectively.

15. Auerbach's plexuses are found in large numbers

and are usually present in the lamina intermuscularis . Sometimes they are present in the outer longitudinal layer.

16. Serosa consists of loose connective tissue containing blood vessels and nerves. Sometimes lymph nodules are also present. Lymph vessels have not been detected under light microscope.

From the results of this investigation it is concluded that there is no sex difference between the two age groups studied. The small intestine is richly supplied with blood vessels and nerves. The structures develop and become more definite with the increase of age of the animals. Except the height of villi in jejunum, thickness of submucosa in the ileum and lumen of Brunner's glands in duodenum, all the structures are found to be thicker or larger in adults than in young animals.

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Awadhesh Kumar Barnwal.