HISTOLOGICAL AND HISTOCHEMICAL STUDY OF BRUNNER'S GLANDS IN LOCAL ADULT COWS

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ABSTRACT

The study was conducted to add information to the database for concentrate on the Brunner's glands of the duodenum in adult local cows. The histological study showed that Brunner's glands present in the tunica submucosa of the duodenum in local adult cows, the glands were branched tubuloalveolar secreting mucous, extended from the end of the pylorus to the first and middle third of duodenum, and absent at the last third of the duodenum. The excretory ducts of Brunner's glands opened at the crypts of lieberkuhn at the base of intestinal villi penetrating the muscularis mucosa layer.

The statistical analysis showed there was a significant variation in the diameter and lumen of Brunner's glands between the first and middle third of duodenum at ($P \leq 0.05$). The diameter and lumen of Brunner's glands at the first third of duodenum were lower than that of the middle third of duodenum. The statistical study showed , that there was no significant variation of the lining epithelium of Brunner's glands between first and middle third of duodenum, and there was no significant variation in the thickness of Brunner's glands aggregation between first and middle third of the duodenum.

The histochemical study revealed that there were different types of glycoprotein in Brunner's glands and excretory duct in local adult cows, at first third of duodenum Brunner's glands contain a large amount of neutral glycoprotein and little amount of acidic glycoprotein, while at middle third of duodenum the Brunner's glands contain the same amount of neutral and acidic glycoprotein. At the first third of duodenum, the excretory ducts of Brunner's glands contain neutral glycoprotein more than acidic glycoprotein, while at the middle third of duodenum the excretory ducts of Brunner's

glands contain a large amount of acidic glycoprotein and little amount of neutral glycoprotein.

INTRODUCTION

Brunner's glands are branched tubulalveolar coiled glands present in the submucosa of the duodenum as a collection (clusters) extended from duodenopyloric junction to different distance in the submucosa of the duodenum(1,2). The excretory ducts of Brunner's glands extend caudally in most animals toward jejunum and opened between the bottom of the crypts of lieberkuhn (3). The main function of Brunner's glands in the production of alkaline mucin which protects the duodenal mucosa from the effect of hydrochloric acid comes from the stomach (4,5). There is a scarcity of up to date works concentrated on the Brunner's glands in adult local cows. Accordingly, the current study aimed to elucidate the morphological and histochemical features of these glands.

MATERIALS AND METHODS

Ten samples of the duodenum from clinically healthy adult native cows have been collected from Mosul abattoir, determine the average length of the duodenum about one meter, starting from the pyloric sphincter and end to duodenojejunal flexure. For conducting the histological and histochemical study of the mucosa of the duodenal wall, the duodenum was divided into three equal parts (first third, middle third and last third), The samples cleaned from the surrounding fatty tissues, then the samples fixed in alcoholic formalin about (48 hours), the routine histological preparation used for these samples using gradual concentration of ethyl alcohol for dehydration and chloroform for clearing and paraffin wax for embedding (6). The paraffin blocks cut with rotary microtome then the histological slides stained with Harris hematoxylin and eosin stain for the general histo-morphological feature , alcoholic PAS stain for the detection of neutral glycoprotein and Alcian blue PH 2.5 for the detection of the acidic glycoprotein.

All micro morphometric measurements were done by using the color USB 2.0 digital camera (Scope Image 9.0) which is provided with image processing software and

connected to a light microscope. Data were presented as means ± SD (standard deviation) and were analyzed using (t-test) with a significant level set on P≤0.05.

RESULTS

Histological study

The present study showed that the Brunner's glands were present in the submucosa of the duodenum, these glands were composed of mucous secreting cells (Fig 1). Glands were tubuloalveolar branched started from the pylorus-duodenal junction and extending to first and middle thirds of duodenum and absent at the last third of duodenum (Fig 2). The present study showed that the excretory ducts of Brunner's glands opened at crypts of lieberkuhn at the base of intestinal villi where they penetrated the muscularis mucosa (Fig 3).

Morphometric study

The morphometric study showed presence of a significant variation in the diameter of Brunner's glands between first and middle third of duodenum at ($p \leq 0.05$) where the mean diameter of the glands at first third was (39.76±1.96)µm lower than the mean diameter of the glands at middle third (54.45±1.76)µm (Tables 1), while the lumen of Brunner's glands showed significant variation between first and middle third of duodenum at ($p \leq 0.05$) , where the mean diameter of the lumen of Brunner's glands at first third was (19.60±2.53)µm lower than the mean at middle third (33.85±0.91)µm (Table 1).

The statistic study showed that there was no significant variation between the lining epithelium height of Brunner's glands at the first and middle thirds of the duodenum at ($p \leq 0.05$). The present study showed there was a significant variation in the excretory ducts diameter of Brunner's glands between first and middle thirds of duodenum at ($p \leq 0.05$) where the mean diameter of excretory duct at first third was (27.50±2.62) µm lower than that of the excretory ducts (59.73±9.42) µm at middle third of duodenum (Table 1).

Histochemical study:

The present study showed that there were different types of glycoprotein in Brunner's glands in local adults cows , Brunner's glands appeared strong reaction with alcoholic PAS at first thirds of duodenum, while they showed mild reaction with alcian blue PH 2.5 (Table 2) (Fig 4), these results showed that Brunner's glands

contain a large amount of neutral glycoprotein and little amount of acidic glycoprotein at the first third of duodenum. At the middle third of duodenal Brunner's glands showed moderate reaction with alcoholic PAS and alcian blue PH 2.5 respectively (Table 2) (Fig 5). These results indicate that Brunner's glands contain equal amounts of neutral and acidic glycoprotein. The excretory ducts of Brunner's glands at first third showed strong reaction with alcoholic PAS and mild reaction with Alcian blue PH 2.5, oppositely the middle third of duodenum showed strong reaction with Alcian blue PH 2.5 and mild reaction with alcoholic PAS , this result indicates that the excretory ducts of Brunner's glands in the first third of duodenum contain little amount of acidic and large amount of neutral glycoprotein, while in the middle third of duodenum contain large amount of acidic and little amount of neutral glycoprotein (table 3).

Table(1): Histological measurements of brunner's glands and their secreting

Grouping	Mean diameter of gland	Mean diameter of lumen's gland	Mean height of cells	Mean diameter of ducts
First third	39.76± 1.96 *	19.60± 2.53*	11.36± 0.89	27.50± 2.62*
Middle third	54.45±1.76	33.85± 0.91	10.66± 0.90	59.73± 9.42
Last third	There are no Brunner's glands			

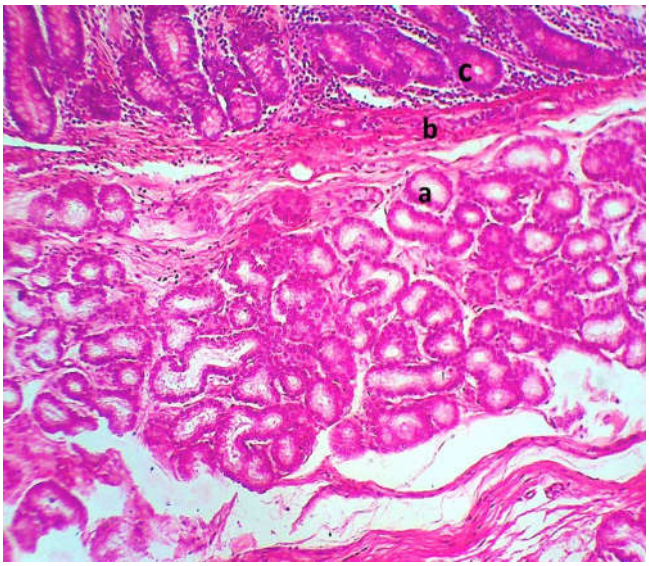
* mean there is a significant difference between first and middle thirds at $p \leq 0.05$

Table (2) Histochemical reactions of brunner's glands in local cows.

Grouping	PAS stain	Alcian blue stain 2.5
First third	+++	+
Middle third	++	++

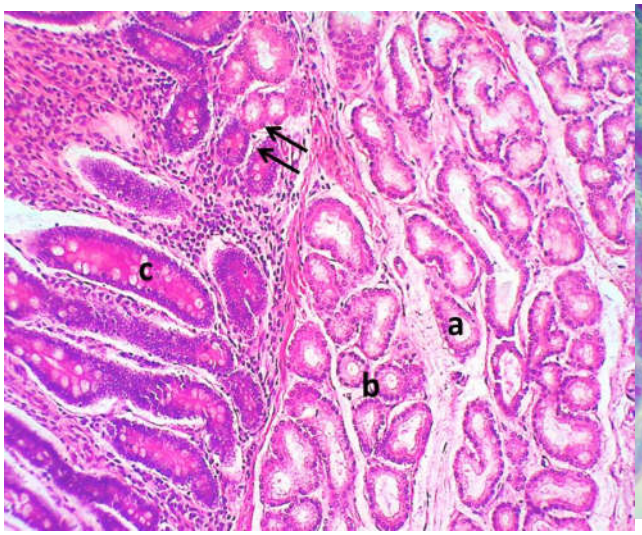
Table (3) Histochemical reactions of the ducts of brunner's glands in local cows

Grouping	PAS stain	Alcian blue stain 2.5
First third	+++	+
Middle third	+	++

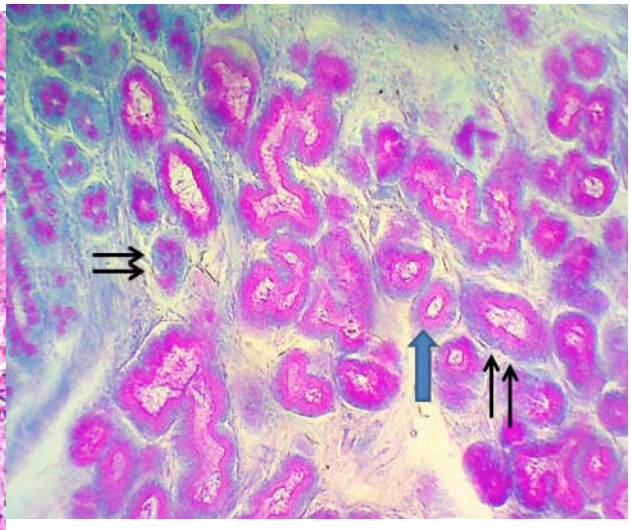


(Figure 1) Photomicrograph of first third of duodenum showing, **a.** brunner's gland, **b.** muscularis mucosa, **c.** crypts of lieberkuhn, H&E stain (X220).

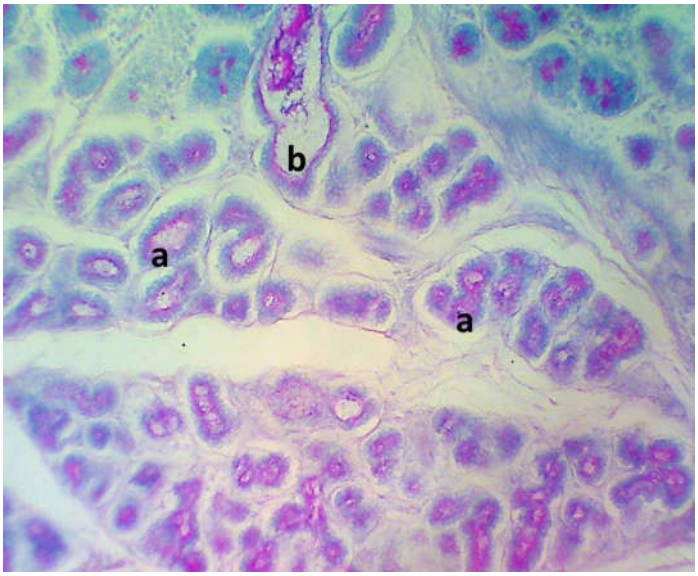
(Figure2) Photomicrograph of last third of duodenum, **a.** muscularis mucosa, **b.** crypts of lieberkuhn. Note absence of Brunner's glands in submucosa(**c**), H&E stain (X200).



(Figure3) Photomicrograph of first third of duodenum, **a.** Brunner's gland, **b.** Excretory ducts of Brunner's glands, **c.** Crypts of lieberkuhn. Noted excretory ducts of Brunner's glands opened at crypts of lieberkuhn (**arrows**), H&E stain (X250).



(Figure 4) Photomicrograph of first third of duodenum, note that brunner's glands and excretory ducts strong reaction with alcoholic PAS **thick arrow**, while they showed mild reaction with Alcian blue PH 2.5 stain small arrow (X250).



(Figure 5) Photomicrograph of middle third of duodenum, note that brunner's glands, **a.** and excretory ducts, **b.** showed mild reaction with alcoholic PAS and Alcian blue PH 2.5(X250).

DISCUSSION

The study showed that Brunner's glands present in submucosa of duodenum, which appeared to be mucous secreting glands, this result agree with (1) in most species, (7) in local buffalo, (8) in local sheep and black goat, (9) in Iranian buffalo and dromedary camels (10). These glands reported being mixed glands in horse and serous glands in rabbits(11). The study showed that Brunner's glands appeared branched tubuloalveolar acini coiled and these results come to in agreement with (1) in most species of animals. The study showed that Brunner's glands extended directly from pyloro-duodenal junction to the first and middle thirds of duodenum, but absent at the last third of duodenum, these results agreement with (7) in local buffalo and in Iranian buffalo (9), while in local sheep and black goat, Brunner's glands extended from a distance from the pylorus (8) whereas in camels Brunner's glands extended proximally two meters away from the pylorus while in horse Brunner's glands extended caudally to proximal third of Jejunum (11). The present study showed that excretory ducts of Brunner's glands opened at crypts of lieberkuhn at the base of intestinal villi penetrated the muscularis mucosa, these results are similar to that of (3) in most species and (7) in local buffalo and with Iranian buffalo (9) and (10) in camel, while in local sheep and black goat either open directly to the duodenal lumen or at crypts of lieberkuhn (8), while our results were different from that in cat, where at the excretory ducts of Brunner's glands opened directly to the duodenal lumen (1). The statistical results showed a significant variation in diameter and lumen of Brunner's glands between first and middle third of duodenum at ($p \leq 0.05$) where the diameter and lumens of Brunner's glands in middle third were larger than these in first third, and these results agreed with (7) in local buffalo and in local sheep and black goat (8). The statistical analysis indicated that there was no significant variation between the lining epithelium of Brunner's glands between first and middle third of duodenum at ($p \leq 0.05$), this result came in agreement with (7) in local buffalo and local sheep and black goat (8). This study showed a significant variation in the diameters of excretory ducts of Brunner's glands at ($p \leq 0.05$) between first and middle third of duodenum, where the diameter of excretory ducts of Brunner's glands in middle third larger than these in first third.

The histochemical study revealed the presence of different types and concentrations of glycoprotein in Brunner's glands and excretory ducts in the first and

middle third of duodenum. The amount of neutral glycoprotein in Brunner's glands at first third of duodenum was more than in middle third of duodenum, this result agreed with (7) in local buffalo and (12) in local cow, while the amount of acidic glycoprotein were the same in Brunner's glands at first and middle third of duodenum , this result came in consistency with (7) in local buffalo and disagree with (13) on local cows.

The present study showed that the excretory ducts of Brunner's glands contained different types and amount of glycoprotein at the first and middle third of duodenum, and the amount of neutral glycoprotein in excretory ducts of Brunner's glands at first third of duodenum was more than that in the middle third of duodenum, while the amount of acidic glycoprotein is present in the middle third of duodenum more than that in the first third of duodenum and that is agreement with (7) in buffalo.

دراسة نسيجية ونسيجية كيميائية لغدد برونر في الابقار البالغة المحلية

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الخلاصة

بينت الدراسة النسيجية ان غدد برونر تتواجد في الابقار المحلية البالغة في الطبقة تحت المخاطية من العفج ، وهي غدد انبوبية سنخية متفرعة ذات افراز مخاطي ، وتمتد من بداية العفج عند المنطقة البوابية الى نهاية الثلث الوسطي، وتختفي مع بداية الثلث الاخير من العفج، كما اظهرت الدراسة ان القنوات الافرازية لغدد برونر تفتح على طويقات ليبركهن عند قاعدة الزغابة مخترقة عضلة المخاطية للعفج.

اظهرت الدراسة الاحصائية ان هناك فرق معنوي في اقطار وتجاويف غدد برونر بين الثلث الاول والوسطي من العفج عند مستوى معنوية ($P\leq 0.05$). حيث كانت اقطار وتجاويف غدد برونر في الثلث الاول من العفج اقل منها في الثلث الوسطي منه، في حيث لم تظهر الدراسة اي فرق معنوي في ارتفاع خلايا غدد برونر بين الثلث الاول والثلث الوسطي من العفج مما يدل ان الفرق هو في تجويف الغدد، كما لم تظهر الدراسة الاحصائية اي فرق معنوي في تجمعات غدد برونر بين الثلثين الاول والوسطي.

كشفت الدارسة الكيميائية النسيجية ان هناك انواع مختلفة من البروتينات السكرية في غدد برونر والقنوات الافرازية للغدد في الابقار المحلية البالغة، ففي الثلث الاول من العفج تحتوي غدد برونر على كمية كبيرة من البروتين السكري المتعادل في غدد برونر وكمية قليلة من البروتين السكري الحامضي ، بينما احتوت غدد برونر في الثلث الوسطي على كميات متساوية من البروتين السكري المتعادل والحامضي. احتوت القنوات الافرازية

لغدد برونر في الثلث الاول على كمية من البروتين السكري المتعادل اكبر من الحامضي، بينما ف ي الثلث الوسطي من العفج احتوت قنوات غدد برونر على كمية كبيرة من البروتين السكري الحامضي وكمية قليلة من البروتين السكري المتعادل .

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