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Sudan University of Science and Technology  
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**Prenatal Morphological and Morphometric Studies on the  
pancreas of the camel foetus (*Camelus Dromedarius*)**

دراسات شكلية وقياسية شكلية في بنكرياس أجنة الجمال وحيدة السنام قبل الولادة

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## **Dedication**

To my father's spirit (Eltayeb), mother (Bakheeta), to my Wife (Abeer), to my son (Mohammed), to my brothers, sister, Clan and All friends.

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I render May thanks to Al Mighty Allah who gave me the courage, patience and strength to conduct this study.

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## Abstract

This study aimed at investigating the development of the gross anatomy, histology and morphometry of the pancreas of the camel foetus (*Camelus Dromedarius*). A total of thirty camel fetuses were used in this study. Specimens were collected from Tamboul Slaughterhouse, Sudan. Foetuses were divided into three trimesters according to the gestation period. The pancreas in the first trimester (Crown Vertebral Rump Length (CVRL) 8-17cm, 87-111 days of gestation) had gray colour but in the second (Crown Vertebral Rump Length (CVRL) 45-71cm, 188-259 days of gestation) and third trimester (Crown Vertebral Rump Length.

(CVRL) 75-110cm, 270-366 days of gestation) the colour was grayish pink. The pancreas was found to be divided into a body, a right lobe and a left lobe at all trimesters. These divisions could be described as a wide body, a quadrate right lobe and a long tongue-shaped left lobe in the second and the third trimesters. The body of the pancreas was related dorsally to the visceral surface of the liver and caudally to the stomach. It was related cranially to the curvature of the duodenum and ventrally to the spleen. The right lobe was related dorsally to the visceral surface of the liver, medially to the portal vein and laterally to the descending duodenum and ventrally to the transverse colon. The left lobe was related caudodorsally to the stomach, ventrally to the kidney, spleen and cranially to the colon. The pancreas in the second and third trimesters was clearly tubuloacinar and containing a single row of similar pyramidal epithelial cells converging to a central narrow lumen. However, in the first trimester the gland appeared as incomplete tubuloacinar with only acini. These cells were characterized by spherical or sometimes oval nuclei near the base or centrally located. The gland was covered by a connective tissue capsule rich in collagenous and reticular fibers in all trimesters. The connective tissue septa divided the parenchyma into lobules. No elastic fibres were observed except around the walls of the blood vessels. The ducts and islets of Langerhans were not observed in the first trimester. However, they were clearly distinguished in the second and third trimester. The duct system consisted of intralobular ducts

which were lined by low cuboidal cells in the second and third trimesters. The interlobular ducts located between the lobules in the connective tissue septa, were lined by stratified cuboidal cells in the second trimester, however, they were lined by columnar epithelium in the third trimester. The islets of Langerhans first appeared at the second trimester as pale areas among the secretory units. These structures were containing cells with rounded, oval or irregular nuclei.

Morphometric study was carried out using the points-counting technique. The volume of the pancreas was determined by the water displacement method. The mean absolute volumes of the pancreas were  $1.66 \text{ cm}^3$ ,  $2.7 \text{ cm}^3$  and  $4.18 \text{ cm}^3$  in the first, second and third trimester respectively. The first trimester revealed that the acini occupied about 44.63% ( $0.74 \text{ cm}^3$ ), the connective tissue occupied about 38.82% ( $0.64 \text{ cm}^3$ ). The blood vessels were about 16.55% ( $0.28 \text{ cm}^3$ ). The total volume of the pancreatic components in the second trimester were as follows; the acini accounted for about 54.98% ( $1.48 \text{ cm}^3$ ), islets of Langerhans about 1.82% ( $0.05 \text{ cm}^3$ ), ducts about 3.30% ( $0.10 \text{ cm}^3$ ), connective tissue about 27.70%, ( $0.75 \text{ cm}^3$ ) and blood vessels about 12.20% ( $0.32 \text{ cm}^3$ ). In the third trimester the acini accounted for about 60.95% ( $2.55 \text{ cm}^3$ ), islets of Langerhans about 2.52% ( $0.11 \text{ cm}^3$ ), ducts about 2.44% ( $0.10 \text{ cm}^3$ ), connective tissue about 26.75% ( $1.12 \text{ cm}^3$ ) and blood vessels about 7.34% ( $0.30 \text{ cm}^3$ ).

## المستخلص

هدفت هذه الدراسة لتقصي تطور التركيب التشريحي العياني، والنسجي والقياسات الشكلية في بنكرياس أجنة الجمال وحيدة السنام قبل الولادة. تم استخدام ثلاثون من أجنة الإبل وحيدة السنام في هذه الدراسة. جلبت العينات من مسلخ تمبول بالسودان. قسمت العينات إلى ثلاث مجموعات عمرية: الأثلوث الأول (17-8 سم، 111-87 يوم) و الأثلوث الثاني (71-45 سم، 259-18 يوم) و الأثلوث الثالث (110-90 سم، 366-270 يوم). يتصف البنكرياس باللون الرمادي في الأثلوث الأول أما في الأثلوث الثاني والثالث فلونه وردي رمادي. وجد أن البنكرياس ينقسم إلى جسم وفص أيمن وفص أيسر في كل المجموعات العمرية. يمكن وصف هذه الأقسام بجسم عريض وفص أيمن شبه رباعي وفص أيسر طويل لساني الشكل. يتموضع جسم البنكرياس ظهرياً مع السطح الحشوي للكبد وذليلاً مع المعدة وقهفياً مع الإنشاء الأول للإثني عشر (العفج)، وبطنياً مع الطحال والكلية اليسرى. بينما يتموضع الفص الأيمن ظهرياً مع السطح الحشوي للكبد في المنطقة الوسطى أعلى الوريد البابي، جانبياً للعفج النازل وبطنياً للقولون المستعرض. أما الفص الأيسر فيقع ظهرياً ذليلاً مع المعدة وبطنياً مع الطحال والكلية اليسرى وقهفياً مع القولون. أوضحت الدراسات النسيجية أن الوحدات الإفرازية للبنكرياس أنبوبية سنخية تحتوي على صف واحد من الخلايا الهرمية الظهارية تنتهي بتجويف مركزي ضيق في الأثلوثين الثاني والثالث أما في الأول فتبدو غير مكتملة تحتوي فقط على الأسناخ. تتميز هذه الخلايا بانوية كروية أو بيضاوية تتموضع مركزياً أو بالقرب من القاعدة. تغطي الغدة بمحظة من النسيج الضام غنية بالألياف الشبكية، والكولاجينية في كل المجموعات العمرية. ينقسم متن الغدة إلى فصيصات بواسطة فواصل النسيج الضام. لم تشاهد الألياف المرنة إلا في جدار الأوعية الدموية. في الأثلوث الأول لم تشاهد القنوات وجزر لانجرهانز بينما ظهرت في الأثلوثين الآخرين. تشكل نظام القنوات من قنوات داخل الفص، وقنوات بين الفصوص، لم تظهر هذه القنوات في الأثلوث الأول ولكن ظهرت في الأثلوثين الثاني والثالث. تبطن القناة داخل الفص بنسيج طلائي مكعباني بسيط أما القناة بين الفصوص فتبطن بنسيج مكعباني مطبق في الأثلوث الثاني بينما تبطن بنسيج عمودي بسيط في الأثلوث الثالث. وجدت جزر لانجرهانز كمناطق باهتة اللون بين الوحدات الإفرازية في الأثلوثين الثاني والثالث وتحتوي على أنوية دائرية وبيضاوية وأخرى غير منتظمة وتحاط بنسيج ضام رفيع. تمت الدراسات القياسية باستعمال تقنية عد النقاط الواقعة على مكونات العضو و شملت المكونات كل من التالي: الأسناخ والقنوات وجزر لانجرهانز والأوعية الدموية و النسيج الضام. و تم قياسها بعد معرفة متوسط الحجم الكلي للبنكرياس في كل الأطوار عن طريق إزاحة الماء. وكان الحجم الكلي للبنكرياس ( 1.66 سم<sup>3</sup> في الأثلوث الأول، 2.7 سم<sup>3</sup> الثاني و 4.18 سم<sup>3</sup> في الأثلوث الثالث). كشفت الدراسة أن نسب المكونات في الأثلوث الأول كالتالي: الأسناخ 44.63% وبحجم مطلق 0.74 سم<sup>3</sup> والنسيج الضام حوالي 38.82% والحجم المطلق 0.64 سم<sup>3</sup> و الأوعية الدموية 16.55% والحجم المطلق 0.28 سم<sup>3</sup>. بينما نتيجة الأثلوث الثاني كالتالي: الأسناخ 54.98% والحجم المطلق 1.48 سم<sup>3</sup>، جزر لانجرهانز حوالي 1.82% والحجم المطلق 0.05 سم<sup>3</sup> والقنوات حوالي 3.3% والحجم المطلق 0.10 سم<sup>3</sup> والنسيج الضام حوالي 27.70% والحجم المطلق 0.75 سم<sup>3</sup> والأوعية الدموية حوالي 12.20% والحجم 0.32 سم<sup>3</sup>، أما نتيجة الأثلوث الثالث كالتالي: الأسناخ 60.95% والحجم المطلق 2.55 سم<sup>3</sup> وجزر لانجرهانز حوالي 2.52% والحجم المطلق 0.11 سم<sup>3</sup> والقنوات حوالي 2.44% والحجم المطلق 0.10 سم<sup>3</sup> والنسيج الضام حوالي 26.75% والحجم المطلق 1.12 سم<sup>3</sup> والأوعية الدموية حوالي 7.34% والحجم المطلق 0.30 سم<sup>3</sup>.

## Table of contents

No.	Topics	Page No.
	Dedication	I
	Acknowledgments	II
	Abstract	III
	المستخلص	V
	Table of Contents	VI
	List of Tables	IX
	List of Figure	X
	<b>INTRODUCTION</b>	<b>XIII</b>
<b>CHAPTER ONE: LITRETURE REVIEW</b>		
1-1	<b>Gross anatomy</b>	2
1-1-A	Colour, shape and lobation	2
1-1-B	Topography of the pancreas	2
1-1-C	Weight and dimensions	3
1-1-D	The blood supply	4
1-2	Histological studies	4
1-2-A	The exocrine portion	5
1-2-A-1	The secretory units	5
1-2-A-2	The duct system	5
1-2-A-2-1	The intercalated duct	6
1-2-A-2-2	The intralobular duct	6
1-2-A-2-3	The interlobular duct	6
1-2-A-2-4	The main pancreatic duct	6
1-2-B	The endocrine portion (Islets of Langerhans)	7
1-2-B-1	The beta cells	7
1-2-B-2	The alpha cells	7
1-2-B-3	The delta cells	8

1-3	Development of the pancreas in various species	8
1-4	Morphometry	9
<b>CHAPTER TWO: MATERIAL AND METHODS</b>		
2-1	Gross anatomy	12
2-2	Histology	12
2-3	Morphometric study	13
2-3-1	Sampling	14
2-3-2	Determination of the volume densities	14
2-3-2-1	Point-counting methods	14
	Text Figure	15
2-3-3	Calculation of the volume densities	16
2.3.4	Calculation of the absolute volumes	17
2.3.5	Statistical analysis	17
<b>CHAPTER THREE: RESULTS</b>		
3-1	<b>The first trimester</b>	19
3-1-A	Gross anatomy	19
3-1-B	Histology	19
3-1-C	Morphometric study	20
3-2	<b>The Second trimester</b>	20
3-2-A	Gross anatomy	20
3-2-B	Histology	21
3-2-B-1	The exocrine portion	21
3-2-B-2	Endocrine portion	21
3-2-C	Morphometric study	21
3-3	<b>The Third trimester</b>	22
3-3-A	Gross anatomy	22
3-3-B	Histology	22



3-3-B-1	The exocrine portion	22
3-3-B-2	Endocrine portion	23
3-3-C	Morphometric study	23
<b>CHAPTER FOUR: DISCUSSION</b>		
4-1	Anatomical studies	64
4-2	Histological studies	65
4-2-A	The exocrine portion	65
4-2-B	The endocrine portion (islets of Langerhans)	66
4-3	<b>Morphometric study</b>	66
4-3-A	First trimester	66
4-3-B	Second trimester	67
4-3-C	Third trimester	67
	Conclusion	68
	Recommendations	69
	<b>References</b>	70

## List of Tables

No		Pages
1	<b>Table 1.</b> the procedure used for sampling	16
2	<b>Table 2.</b> Points falling on each parameter, the volume densities and the absolute volumes of the different components of the pancreas at the first trimester for 5 sections shown as means $\pm$ Standard Deviation.	24
3	<b>Table 3.</b> The volume of fresh pancreas, volume densities and absolute volumes of the main components in 5 camel's foetuses at the first trimester expressed as Means $\pm$ Standard Deviations (SD).	24
4	<b>Table 4.</b> The data obtained by point-counting of fields on seven sections of the pancreas at the second trimester.	25
5	<b>Table 5.</b> The data obtained by point-counting of fields on twelve sections of the pancreas at the third trimester.	26
6	<b>Table 6.</b> Points falling on each parameter, the volume densities and the absolute volumes of the different components of the pancreas at the second trimester for 5 sections shown as means $\pm$ Standard Deviation.	27
7	<b>Table 7.</b> Total data obtained by point-counting of fields of five histological sections at the third trimester.	27
8	<b>Table 8.</b> The volume of fresh pancreas and volume densities of their main components in 5 camel foetuses at the second trimester expressed as mean and standard deviations, Mean absolute volumes of components indicated in cm <sup>3</sup> .	28
9	<b>Table 9.</b> The volume of fresh pancreas and volume densities of their main components in 5 camel foetuses at the third trimester expressed as mean and standard deviations Mean absolute volumes of components indicated in cm <sup>3</sup> .	28
10	<b>Table 10.</b> Summary of result shown in Table 8 and Table 9.	29

## List of Text Figures

No		Pages
1	<b>Fig.1</b> The first trimester, Gross anatomy.	31
2	<b>Fig.2</b> The first trimester Gross anatomy.	32
3	<b>Fig.3</b> The first trimester, Histology, H&E staining.	33
4	<b>Fig.4</b> The first trimester, Different shapes of nuclei, H&E staining	34
5	<b>Fig.5</b> The first trimester, Different shapes of nuclei, H&E staining, A high magnification of Fig.4.	35
6	<b>Fig.6</b> The first trimester, reticular fibers surrounding the acini, Silver staining.	36
7	<b>Fig.7</b> The first trimester, reticular fibres in the capsule, Silver staining.	37
8	<b>Fig.8</b> The first trimester collagenous fibres surrounding the acini, Van Gieson staining.	38
9	<b>Fig.9</b> The first trimester, collagenous fibres in capsule, Van Gieson staining.	39
10	<b>Fig.10</b> The second trimester, Gross anatomy.	40
11	<b>Fig.11</b> The second trimester, Gross anatomy.	41
12	<b>Fig.12</b> The second trimester, Histology, H&E staining.	42
13	<b>Fig.13</b> The second trimester, Different shapes of nuclei, H&E staining.	43
14	<b>Fig.14</b> The second trimester, intralobular duct, H&E staining.	44
15	<b>Fig.15</b> The second trimester, islets of Langerhans and intralobular, duct H&E staining.	45
16	<b>Fig.16</b> The second trimester, interlobular duct, H&E staining.	46
17	<b>Fig.17</b> The second trimester, A high magnification of Fig. 16 H&E staining.	47
18	<b>Fig.18</b> The second trimester, collagenous fibres, Van Gieson staining.	48
19	<b>Fig.19</b> The second trimester, reticular fibres, Silver staining.	49
20	<b>Fig.20</b> The second trimester, islets of Langerhans, H&E staining.	50

21	<b>Fig.21</b> The third trimester, Gross anatomy.	51
22	<b>Fig.22</b> The third trimester, Gross anatomy.	52
23	<b>Fig.23</b> The third trimester, Histology, H&E staining.	53
24	<b>Fig.24</b> The third trimester, intralobular duct and different shapes of nuclei, H&E staining	54
25	<b>Fig.25</b> The third trimester, interlobular duct, H&E staining.	55
26	<b>Fig.26</b> The third trimester, reticular fibres, Silver staining.	56
27	<b>Fig.27</b> The third trimester, reticular fibres surrounding the interlobular duct Silver staining.	57
28	<b>Fig.28</b> The third trimester, collagenous fibres, Van Gieson staining.	58
29	<b>Fig.29</b> The third trimester, collagenous fibres, Van Gieson staining.	59
30	<b>Fig.30</b> The third trimester, elastic fibres in the wall of a blood vessel, Ver hoeff staining	60
31	<b>Fig.31</b> The third trimester, groups of islets of Langerhans, H&E staining.	61
32	<b>Fig.32</b> The third trimester, A high magnification of <b>Fig 31</b> , H&E staining.	62

## Introduction

Camels are multipurpose animals serving people specially nomads for many decades. They belong taxonomically to the order *Artiodactyls* (even-toed ungulates), sub order *Tylopoda* (pad-footed), and Family Camelidae (Wilson, 1995). They are pseudo-ruminants that possess a three-chambered stomach, lacking the omasum (Sonfada, 2008). The dromedary camel (*Camelus dromedarius*) is a main source of meat, milk and hides and wool especially for communities living in the desert. (Williamson and Payne, 1978). The Camels have unique metabolic traits that enabled them to live in the desert without food and water for few days (Haghkhah and Madjlesi, 1999). Compared to other domestic animals the camel research showed little attention. Although, the anatomy of many parts of the camel was studied thoroughly little research was directed to the development of its various organs.

The pancreas is both exocrine-endocrine gland that produces digestive enzymes and hormones. The pancreas is a necessary digestive gland whose exocrine secretions include essential enzymes for digestion along with many electrolytes. Its hormonal secretions are produced by pale staining cell clusters of islets of Langerhans which have various activities such as control of blood sugar concentration (Dyce and Wensing, 1971; Nickel, Schummer, and Seiferle, 1973; Bloom and Fawcett, 1986; Dyce, Sack, and Wensing, 1987; Gartner, 2006).

The pancreas in the adult camel was studied by many authors (Adeghate, 1997; AL-Ajlan and Bailey, 1999; Masaad, 2007; Baragob, Mohammed, Hana, Somia, Khojali, and Alkari, 2011; Elamin, Al-Malki, Ismael and Ayoub, 2014; Hafez, Zaghoul, and Caceci, 2015; Zghair, 2016).

Recently, the endocrine portion had been studied by few investigators (Hafez, *et al.*, 2015; Zghair, 2016; Hafez and Zaghoul, 2017).

Hyttel, Sinowatz, Vejlsted, and Betteridge, (2010) had studied embryological development of pancreas in cattle and pig and Murtaugh (2007) had studied embryological development of pancreas in mouse.

From the available literature, studies on the development of the pancreas in camel foetus were virtually lacking. Hence the current research was undertaken to give insight on the development of pancreas of the camel foetus.

### **General objectives**

- To investigate the morphological development and morphometry of the pancreas of the camel foetus (*Camelus dromedarius*) at different stages of pregnancy.

### **Specific objectives**

- To study the gross anatomical features of the camel foetus pancreas at different stages of gestation period.
- To study the histological structure of the camel foetus pancreas at different stages of gestation period.
- To study the morphometry of the camel foetus pancreas at different stages of gestation period.

**CHAPTER ONE**  
**LITERATURE REVIEW**

# CHAPTER ONE

## LITERATURE REVIEW

### 1-1: GROSS ANATOMY

#### 1-1- A: Colour, shape and lobation

The colour of pancreas varied in domestic animals. It was described as pinkish yellow 'grey' light or dark yellow red or reddish cream in equines (Sisson, 1975), pinkish yellow brown in bovines (Dyce *et al.*, 1987), grey pinkish in the camel (Sultan, 1999).

Variations in the shape were reported in the pancreas of different domestic animals. In the horse, Indian donkey and sheep the shape was irregular but triangular in outline (Bradley, 1946; May, 1970; Sisson, 1975; Dyce *et al.*, 1987; Dhoolappa Ashok, Ramakarishna and Gadre, 2004), while in ruminants it was irregular (Dyce and Wensing, 1971; Sultan, 1999).

Regarding the lobation of the pancreas, it was clear that there was a general agreement that the pancreas consisted of a left lobe, a right lobe and a body. However; differences were seen within these lobes. A long right lobe and a short left lobe were reported in ruminants (Dyce and Wensing, 1971; Nickel *et al.*, 1973; Habel 1975; Dyce *et al.*, 1987) and in the Indian donkey (Dhoolappa *et al.*, 2004). On the contrary, the equine pancreas showed a long left lobe and a short right lobe (Nickel *et al.*, 1973; Sisson 1975). Although the camel is a ruminant yet the lobation of the pancreas more or less, resembled that of the horse (Mustafa, Aly, Amar and Aly, 1983; Smuts and Bezuidenhout, 1987; Taha and Abdel-Magied, 1998; Sultan, 1999).

#### 1- 1- B: Topography of the pancreas

The ruminant pancreas was located almost in the mesoduodenum and the root of the greater omentum entirely to the right of the median plane (Habel, 1975, 1989). In the horse the pancreas lay ventral to the aorta and the caudal vena cava, at the level of the 16<sup>th</sup>, 17<sup>th</sup> and 18<sup>th</sup> thoracic vertebrae, the bulk of the gland being to the



right of the median plane (Bradley, 1946; Nickel *et al.*, 1973; Sisson, 1975; Dyce *et al.*, 1987). It was related dorsally to the right kidney, the caudal vena cava, portal vein, the stomach and the right caudate lobe of the liver (Sisson, 1975).

The left lobe of the pancreas in bovines extended to the spleen and was linked by connective tissue to the rumen and the left crus of the diaphragm. The body of the pancreas was lying between the liver and the omasum ventral to the portal vein, the right lobe was enclosed in the mesoduodenum descendens and extended to the plane of the right kidney (Simoens, 2003). The pancreas in humans was divided to head, body, and tail. The head was situated near the duodenum and the tail extended to the hilum of the spleen (Longnecker, 2014).

The pancreas in camel was lying at the level of the first five lumbar vertebrae (Mustafa *et al.*, 1983; Sultan, 1999). The portal vein was related ventrally to the body of the pancreas. The left crus of the diaphragm and the transverse colon were dorsal to the body of the pancreas (Mustafa *et al.*, 1983; Sultan, 1999). The right lobe was related dorsally to the right crus of the diaphragm, the sublumbar muscles and visceral surface of the liver. Ventrally it was related to the hepatic lymph node, the transverse colon, mesenteric node and the second duodenal flexure. Cranially it was related to the descending duodenum and bulb of the duodenum, caudally to the transverse and descending colons (Mustafa *et al.*, 1983; Smuts and Bezuidenhout, 1987; Taha and Abdel-Magied, 1998; Sultan, 1999). The left lobe was related cranially to the transverse and descending colon, dorsal sac of the rumen while caudally and laterally it was related to the spleen, left kidney and the left adrenal gland (Mustafa *et al.*, 1983; Smuts and Bezuidenhout, 1987; Taha and Abdel-Magied, 1998; Sultan, 1999).

### **1- 1- C: Weight and dimensions**

The weight of the pancreas in the small ruminant was about 60g and in the ox it was 425g (Habel, 1975). The pancreas weighed in the horse about 350 g (Bradley, 1946; Sisson, 1975), and in the Indian donkey its weight was about 95 g (Dhoolappa, Pawar, Ramakarishna, and Gadre, 2004).

In the camel its weight was about 500 g (Smuts and Beziudenhout, 1987; Sultan, 1999). However, Mustafa et al. (1983) stated that the pancreas weight was more than 300 g in camel.

The dimensions of the pancreas of the camel regarding the length, width and thickness of the right lobe, left lobe or the body. The length of the right lobe was about 13cm (Mustafa *et al.*, 1983; Sultan, 1999) and 15cm (Taha and Abdel-Magied, 1998); the length of the left lobe was about 28cm (Mustafa *et al.*, 1983; Sultan, 1999) and 29cm (Taha and Abdel-Magied, 1998). The width of the right lobe was about 5cm (Mustafa *et al.*, 1983) and 8cm (Sultan, 1999); the width of the left lobe was about 5cm (Mustafa *et al.*, 1983; Sultan, 1999). The thickness of the right and left lobes was more or less similar about 1.5cm (Mustafa *et al.*, 1983; Sultan, 1999). Although there was no variation between the right and left lobes concerning width and thickness, yet the difference in the length between the right and left lobes was clearly obvious; it was twice as much in the left lobe compared to the right lobe.

### **1- 1- D: The blood supply**

Generally in almost all domestic animals the blood supply of the pancreas was mainly from the celiac artery and partially from the cranial mesenteric artery (Sisson, 1975). The ruminant pancreas received direct branches from the coeliac artery and also from each of its three branches; the left gastric, the hepatic and the splenic arteries. The cranial pancreatoduodenal artery arose from the gastroduodenal artery, whereas the cranial mesenteric artery furnished the caudal pancreatoduodenal artery (Habel, 1975). The pancreas of the camel received its blood supply from the hepatic and the splenic arteries, as branches of the celiac artery. The intestinal and pancreatoduodenal arteries, which were branches of the cranial mesenteric artery, also contributed to the blood supply of the pancreas (Mustafa *et al.*, 1983; Sultan, 1999).

## **1- 2: THE HISTOLOGICAL STUDIES**

The pancreas was a complex tubuloacinar gland consisted of endocrine and exocrine secretary portions in different animals (Stinson and Calhoun, 1981). The human pancreas was covered by a thin layer of connective tissue that did not form a definite capsule (Bloom and Fawcett, 1986).

The pancreas of the camel consisted of both endocrine and exocrine portions and it was enclosed by a thick connective tissue capsule which was rich in adipose tissue, blood vessels and nerve fibers (Sultan, 1999). The gland was divided into lobules by the connective tissue septa. These septa contained adipose tissue, blood vessels, nerve fibers, ducts and groups of lymphatic cells (Stinson and Calhoun, 1981; Bloom and Fawcett, 1986; Sultan, 1999).

## **1-2- A: The exocrine portion**

This portion consisted of the secretory units and duct system.

### **1-2-A-1: The secretory units**

The secretory units of the pancreas were tubuloacinar with the tubular portion more prominent in ruminants (Stinson and Calhoun, 1981). In the horse, the pancreas was tubuloalveolar and the alveoli were long, like those of the duodenal glands (Sisson and Grossman, 1964). In the pig pancreas, the secretory units showed different shapes: rounded, oval or irregular (Singh and Singh, 1980).

In the camel, they were tubuloacinar with the acinar portion more prominent (Sultan, 1999). The acinar cells in the camel were pyramidal in shape with spherical basal nuclei and their lumina were narrow. These acinus cells rested upon a basal lamina and supported by a network of reticular fibres (Sultan, 1999; Dhoolappa *et al.*, 2004). The acinar cells of sheep were mononucleated but a few were binucleated and the nuclei were mostly spherical or oval in shape (Mukherjee, Singh, Roy, Barnwal and Sharan, 1986). Three types of acinar cells were recognized in the pancreas of sheep, buffaloes, ox, goats, horses, dogs, cats, pigs and fowl. These types were active acinar cells, exhausted acinar cells and resting acinar cells (Singh, 1980; Singh and Singh, 1980; Mukherjee *et al.*, 1986).

### **1-2-A-2: The duct system**

The duct system began as flattened centroacinar cells in the lumen of the acini (Bloom and Fawcett, 1986; Lone, Prasad and Sinha, 1988; Sultan, 1999; Dhoolappa *et al.*, 2004). All of these authors agreed on the presence of intercalated, intralobular, interlobular and the main pancreatic ducts.

#### **1-2-A-2-1: The intercalated duct**

The intercalated ducts continued in the small ducts, still intralobular, which were lined by a cuboid epithelium. In the interlobular connective tissue septa, the ducts were termed interlobular ducts (Cormack, 2001). In sheep intercalated duct was lined by spindle-shaped cells and the nuclei were oval and directed along the duct wall (Lone *et al.*, 1988). Gemmel and Heath (1973) in sheep and Dhoolappa *et al.* (2004) in the Indian donkey reported that this duct was lined by cuboidal cells. In the camel it was lined by simple cuboidal cells which were supported by a basal lamina (Sultan, 1999).

#### **1-2-A-2-2: The intralobular duct**

The intralobular duct was lined by low cuboidal epithelial cells in the goat (Stinson and Calhoun, 1981). Gemmel and Heath (1973) reported that this duct was lined by columnar cells while Lone *et al.* (1988) reported that it was lined by low to tall cuboidal cells in sheep. In the camel, it was lined by simple cuboidal cells (Sultan, 1999).

#### **1-2-A-2-3: The interlobular duct**

The interlobular duct of the pancreas of sheep was surrounded by a thick coat of connective tissue. It was lined with tall cuboidal to columnar cells with goblet cells interspersed among them in sheep (Lone *et al.*, 1988) and the Indian donkey (Dhoolappa *et al.*, 2004). In addition to the lining columnar cells and goblet cells, small mucous glands were also observed (Bloom and Fawcett, 1986). In the camel, the interlobular duct started as small duct which was lined by simple cuboidal epithelium which then changed into stratified cuboidal epithelium which was supported by a basal lamina and thick dense connective tissue layer (Sultan, 1999).

#### **1-2-A-2-4: The main pancreatic duct**

These large ducts in humans were lined with simple columnar epithelium containing some goblet cells (Cormack, 2001). In sheep, the main pancreatic duct was lined by columnar cells with goblet cells and mucous glands (Gemmel and Heath, 1973; Lone *et al.*, 1988). Lone *et al.* (1988) reported that this duct was

surrounded by connective tissue in sheep. In the camel this duct was similarly lined by columnar cells supported by a basal lamina and connective tissue (Sultan, 1999).

### **1-2-B: The endocrine portion (Islets of Langerhans)**

The endocrine portion was incorporated among the exocrine portion of the pancreas. It was in the form of small masses of endocrine cells which were highly vascular and were known as islets of Langerhans. The endocrine portion of sheep pancreas was organized in irregular clumps of cells which were dispersed intralobularly. These clumps showed no distinct capsule but they were separated from the pancreatic acini by a thin layer of reticular tissue (Mukherjee, Singh, Barnwal and Sharan, 1988).

In the camel, the islets had different shapes which varied from round, oval, elongated to irregular (Al-Ani, 1987; Sultan, 1999).

#### **1-2-B-1: The beta cells**

The beta cells had oval nuclei and arranged in cords in the camel (Sultan, 1999). The central location of the beta cells in the islets seemed to be the general rule for the vast majority of domestic animals (Erlandsen, Hegre, Parsons, Mcevoy, and Elde, 1976; Bonner-Wier and Like, 1980; Khatim, Gumaa, Petersson, Lundqvist, Grimelius, and Hellerstrom, 1985; AL-Ani, 1987; Mukherjee *et al.*, 1988; Sultan, 1999). However, the horse seemed to be the exception since the beta cells were located peripherally in the islets (Helmstaedter, Feurle and, Frossmann,, 1976; Dellmann, 1981; Furuoka, Ito, Hamada, Suwa, Satoh, and Itakura, 1989).

#### **1-2-B-2: The alpha cells**

The alpha cells were few in number compared to the beta cells and they were located at the periphery of the islets, but their central location was not uncommon. Their nuclei were generally ovoid in shape; although spherical nuclei were sometimes present (Mukherjee *et al.*, 1988). Peripheral location of the alpha cells in the islets seemed to be the general rule for the majority of domestic animals again the horse seemed to be the exception since the alpha cells were located centrally in the islets (Helmstaedter, Feurle, and Frossmann, 1976; Furuoka *et al.*, 1989).

### **1-2-B-3: The delta cells**

The delta cells location could be peripheral or central in the camels (Khatim *et al.*, 1985; Sultan, 1999). Most of these cells were located in the periphery of the islets in bovines (Bonner-Wier and Like, 1980), the horse (Helmstaedter *et al.*, 1976 and Furuoka *et al.*, 1989), and the camel (Alani, 1987).

## **1.3 DEVELOPMENT OF THE PANCREAS IN VARIOUS SPECIES**

According to (Hyttel *et al.*, 2010) the pancreas in the domestic animals developed from dorsal and ventral endodermal buds of the caudal end of the foregut. The two buds were clearly recognizable by 19 days of gestation in the pig. In cattle they appeared by day 26. With the first rotation of the stomach around a longitudinal axis, the ventral pancreatic bud was moved dorsally, close to the dorsal bud, and with further development the two buds fuse to form a single organ. The dorsal bud developed into the major portion of the pancreas including the left and right lobes as well as a portion of the body; the ventral bud developed into a portion of the body of the pancreas and also the liver (as described above). The dorsal bud gradually grew into the mesoduodenum in an arboreal fashion as endodermal cell cords that later developed a lumen. The main duct of the dorsal bud developed into the accessory pancreatic duct which was later obliterated in the cat and small ruminants. The main duct of the smaller ventral bud, which developed into a portion of the pancreatic body, developed into the pancreatic duct which was later obliterated in the ox and pig. The budding endoderm gave rise to both the exocrine acini and the endocrine islets of Langerhans.

The pancreas in the human was anlagen appeared at five week of the gestation. Fusion of the dorsal and ventral anlagen occurred during the seven week of gestation. Full development of acinar tissue extended into the postnatal period. (Lee and Lebenthal, 1993).The pancreas in the mouse was recognizable by approximately 9 days of development (Wessells and Cohen, 1967). Retaining luminal continuity with the gut tube, these structures evaginated into the

surrounding mesenchyme as dense epithelial buds, which subsequently expanded branched and differentiated to yield a fully functional organ system prior to birth. Gut rotation brings the two lobes into close apposition. In humans, their ductal systems underwent partial fusion, although this process was less obvious in rodents. (Murtaugh, 2007).

The pancreas in zebra fish developed from the posterior foregut endoderm (Field, Dong, Beis, and Stainier, 2003). Origin of endocrine and exocrine structures was uniquely separated in zebrafish. While dorsal bud exclusively gave rise to endocrine pancreas, ventral bud formed mostly exocrine tissue besides very little endocrine cells. The posterior dorsal bud evaginates by 24 hours post-fertilization (hpf), and the anteroventral bud was formed by 32–40 hpf. The two buds fused by 52 hpf to form a single primary islet consisting of beta, alpha, delta and epsilon cells (Field *et al.* 2003; Argenton , Zecchin and Bortolussi, 1999; Biemar, Argenton, Schmidtke, Epperlein, Peers, and Driever , 2001).

The pancreas in alligator rose from dorsal and ventral pancreatic buds that developed at 8 to 13 of embryonic days, respectively (Jackintell and Lance, 1994). Two anlagen fused at 14 days and became similar to adult pancreas at 22-23 days. The number of cells in clusters increased during later stages, and pancreas become fully mature at 60-63days. In alligator and crocodile, alpha and beta cells development preceded by delta and PP cells, and similarly in rat and mouse. (Rhoten, 1987).

The pancreas in buffalo appeared at 308–312 days. Beta, alpha and delta cells were first observed as scattered small clusters in 2nd month, followed by pancreatic polypeptide (PP) cell one month later (Lucini, Castaldo, Lai, and De Vico, 1998). By 8th month, islets appeared to be formed with a beta cells core and alpha and delta cells in periphery.

## **1-4: MORPHOMETRY**

Taga, R., Bispo, L. B., Bordin, R. A. and Hassunuma, R. M. (1998) stated that the body mass of the pancreas in mouse grew linearly by 2201% ( $P < 0.01$ ) from 1.58

to 36.36 g. During the period of 2 -70 days of age. Whereas the pancreatic mass significantly increased by 10246% ( $P < 0.01$ ) from 3.17 mg to 327.96 mg, corresponding to a mean growth of 4.78 mg/day. Volume density of the acinar morphologic compartment exhibited a statistically significant growth only from 7 to 21 days of age ( $P < 0.05$ ), with stabilization during the remaining periods. On the other hand, stroma volume density showed a significant fall from 2 to 21 days ( $P < 0.01$ ). Absolute acinar volume showed a marked 13,384% increase ( $P < 0.01$ ) from 2 to 70 days of age (Taga *et al.*, 1998).

Dean, (1973) reported that in humans the morphometric parameters for the ultrastructure of islet beta cells Comprised 88% of the volume of a single islet of Langerhans, no further subdivision of extra beta cellular space was attempted. Cell volume was calculated from the mean area enclosed within cell membrane profiles and found to give a mean value of  $1434\mu\text{m}^3$ .

Zharkov, V.P., Yarygin, V.N. and Dolzhikov, A. A. (1996) reported that the pancreas of guinea pigs showed two types of islets; giant islets and small islets. The total count of the giant islets was ( $3.8\pm 0.1\%$ ) while that of large islet was ( $13.7\pm 1.2\%$  compared to  $3.6\pm 0.3\%$  in the control) which reached the maximum on day 30 after partial resection of the pancreas.



**CHAPTER TWO**  
**MATERIAL AND METHODS**

# CHAPTER TWO

## MATERIAL AND METHODS

Pancreatic specimens of 30 apparently healthy camel foetuses of both sexes and different ages were collected from Tamboul slaughterhouse. The collected specimens were then taken to the Veterinary Anatomy laboratory of College of Veterinary Medicine, Al Butana University. The approximate age of the foetuses was estimated by using the following formula adopted by El-wishy, A. B., Hemeida, N. A., Omar, M. A., Mobarak, A. M., El Sayed, M. A. (1981).

$$GA = \frac{CVRL + 23.99}{0.366}$$

Where GA is age in days and CVRL is the Crown Vertebral Rump Length. Crown Vertebral Rump Length (CVRL) was measured (cm) as a curved line along the vertebral column from the point of the anterior fontanel or the frontal bone following the vertebral curvature to the base of the tail.

Fetuses less than 111 days were designated as first trimester, 188-259 days as second trimester and 270-366 days as third trimester (Bustinza, 1979).

### **2-1: GROSS ANATOMY:**

Sample of camel foetuses at different trimesters used in this study were fixed in 10% formalin to study the gross external features of the pancreas. Ten foetal samples of all trimesters were used to study the topography, blood and nerve supply of the pancreas.

### **2-2: HISTOLOGY**

Pancreatic tissue from 20 camel foetuses was collected to study the microscopic structure. The tissues were fixed in different fixatives; 10% formalin, neutral formalin, Bouin's solution and Gender fluid (Culling, 1974). Formalin at 10% concentration was found to be the most suitable fixative. Tissues were dehydrated in ascending grades of alcohol (70%, 80%, 90% and 100%). Then they were cleared in xylene and embedded in paraffin wax. Sections, 3-5 $\mu$  thick, were cut

using 820 rotary microtome (Leica model Rm 2125 Rt, China). Sections were spread in water path at 40°C and mounted on clean slides. The slides were then cleared in xylene and rehydrated in descending grades of alcohol (100%, 90%, 80% and 70%). After that the slides were rinsed in distilled water and finally stained with haematoxyline and eosin (H&E) for studying the histological structure of the tissue (Culling, 1974). The following special stains were used to study certain structures according to Bancroft and Gamble (2008)

1. Van Gieson for collagen fibers.
2. Verhoeff for elastic fibers.
3. Silver impregnation for reticular fibers.

## **2-3 MORPHOMETRIC STUDY**

A total of 15 camel foetuses were used for the morphometric studies, 5 for each trimester. The entire pancreases were removed by blunt dissections. Then weighed using normal digital balance. Their weights immersed in water were also measured. The difference between the two measured weights for each pancreas resembled its volume Weibel, E. R., Kistler, G. S. and Scherle, W. F. (1966).

Tissue Samples were taken randomly from the different parts of the pancreas (4 regions for the first trimester, 7 regions for the second trimester and 12 regions for the third trimester representing the different lobes of the gland). They were fixed in 10% formalin and processed similar to the histological samples. Paraffin sections were prepared and stained with haematoxylin and eosin. The sections were examined under the light microscope to choose the one best sections from each block.

The microscopic fields were selected blindly by moving the stage of the microscope. Chosen fields were photographed at two magnifications (X10 and X40). The obtained images were saved as PowerPoint slides in the computer. Later, quantitative analysis was performed by the stereological methods of the point-counting techniques.

### **2-3-1: Sampling**

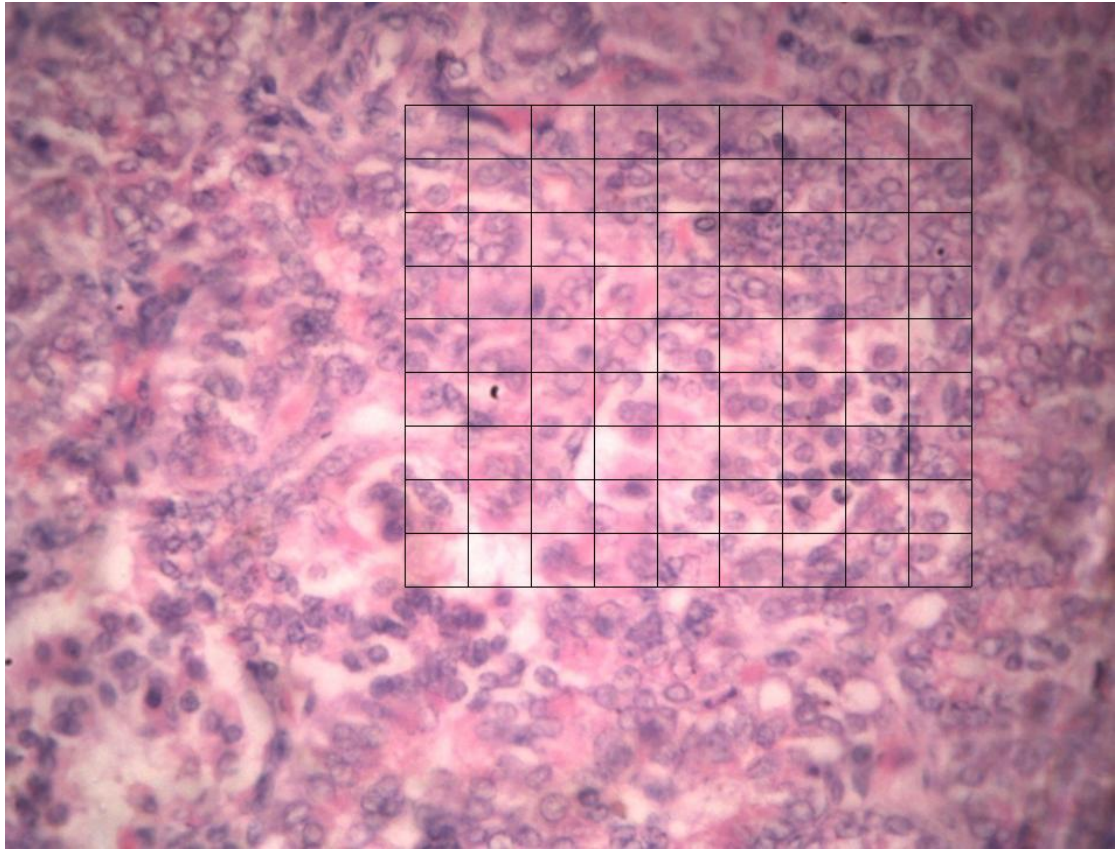
As it is impossible to examine the histological sections of the entire organ, sampling is being necessary and should represent the whole pancreas. Hence, the accuracy of the results obtained by the point-counting technique depends on the sample size. The larger the samples size the better values for each selected parameter to be obtained. A high degree of accuracy can be attained by the procedure of systematic random sampling (Weibel *et al.*, 1966). This method of sampling was applied in the present investigation as it was considered more precise and efficient in the examination of the selected parameters (Mayhew, 1983). The sampling process adopted is shown in Table1. The sampling procedure gave a final size of 230 microscopic fields that were saved as PowerPoint images and analyzed by stereological methods to determine the volume densities ( $V_v$ ) of the components of the parenchyma of the pancreas. These parameters were used to calculate the absolute volumes.

### **2-3-2: Determination of the volume densities**

#### **2-3-2-1: Point-counting methods**

The determination of the volume density  $V_v$  (hence the percentage volume) of the tissue component and derivation of the formula to be applied was used according to the method reported by Weibel *et al.* (1966) and Dunnill (1968). According to the point-counting principle the percentage number of points falling on a given parameter is equivalent to the percentage volume occupied by the component.

To determine the  $V_v$  (percentage volume), the microscopic fields were photographed at X40 as the components of the pancreas were easily identified at this magnification. A grid of 1 cm square lattice was installed and superimposed randomly on the PowerPoint image of each of the 230 fields of the parenchyma of the pancreas (Text Fig). The intersections of the lines at the corners of the squares constituted the points for counting, the total number of points on the grid being 100 points.



**Text Fig:** Showing the grid superiposed on the images.

**Table1:** Showing the procedure used for sampling.

No.	Stage	Specimens number
1	5 pancreases (animals)	5
2	4,7 and12 tissue blocks (slide) per pancreas for the 1 <sup>st</sup> , 2 <sup>nd</sup> and the 3 <sup>rd</sup> trimester respectively	5x4 5x7 5x12
3	Each sections 2 fields (images) per slide (block). 5x4x2= 40 fields (images) 1 <sup>st</sup> trimester. 5x7x2= 70 fields (images) 2 <sup>nd</sup> trimester. 5x12x2= 120 fields (images) 3 <sup>rd</sup> trimester.	40+70+120= 230
4	100 lattice (grid) points per field	100x230=23000

The number of points for each component was counted directly on the computer screen and the volume density ( $V_v$ ), hence the percentage volume was calculated for each of the following parameters of the parenchyma of the pancreas; acini, islets of Langerhans, ducts, connective tissue and blood vessels.

Samples processed by haematoxylin and eosin (H&E) were examined under light microscope at different magnifications to assess the quality of the islets of Langerhans, Connective tissue, Blood vessels, Acini. The microscopic fields were chosen by blind random displacement of the stage of the microscope without looking through the tube. The fields were photographed at two levels of magnification (X10 and X40). The images of the fields were saved in computer as PowerPoint slides which were subjected to quantitative analysis by stereological methods of point-counting and intersection-counting.

### **2-3-3: Calculation of the volume densities**

The volume density ( $V_v$ ) was calculated for each of these components using the formula given by Weibel *et al.* (1966) as follows:

$$V_v = P/PT$$

where P is the number of points falling on the component and PT is the total number of grid points. The number of points to be counted for each component depends on the volumetric proportion itself (i.e.  $V_v$ ).  $V_v$  is volume density (volume fraction). The smaller the  $V_v$ , the larger the total number of points that must be counted for any given standard error (Anderson and Dunnill, 1965; Dunnill, 1968).

#### **2.3.4 Calculation of the absolute volumes**

The absolute volumes of the pancreas components were calculated from the volume densities ( $V_v$ ) of the components and the total volume (V) of the fresh pancreas (i.e. Absolute volume =  $V_v \cdot V$ ) according to Weibel *et al.* (1966).

#### **2.3.5 Statistical analysis**

In this investigation the standard deviation (SD) was calculated for the  $V_v$  of each component of the pancreas as recommended by Weibel *et al.* (1963).

**CHAPTER THREE**  
**RESULTS**



# CHAPTER THREE

## RESULTS

### 3-1: THE FIRST TRIMESTER

#### 3-1- A: Gross anatomy

The pancreas at the first trimester of CVRL 8-17 cm (87-111 days) was gray in colour. It was situated between the liver and stomach in median plane at the region of porta hepatis (Figs. 1 and 2). The pancreas was very small in this trimester, it weighed about 1-3gm and the division to a body, right lobe and left lobe (Figs. 1 and 2). It was lying at the level of the first five lumbar vertebrae. The body of pancreas was related dorsally to the visceral surface of the liver and ventrally to the spleen and mesonephros, caudally to the stomach and cranially to small intestine (Fig. 1).

The right lobe was related dorsally to the visceral surface of the liver, medially to the portal vein and laterally to the descending duodenum and ventrally to the large intestine (Fig. 1).

The left lobe was related caudodorsally to the stomach, ventrally to the mesonephros, spleen, cranially to the large intestine (Figs. 1 and 2).

#### 3-1-B: Histology

The pancreas in this trimester consisted of incomplete or very small secretory units (Figs. 3 and 4). In this trimester the pancreas was covered by a thin connective tissue capsule (Fig. 3). The ducts and islets of Langerhans were not observed in this stage. The secretory units were made of single row of similar pyramidal epithelial cells converging to a central narrow lumen (Figs. 3 and 4). The nuclei of these cells were spherical in shape, and they were usually located near the base of the cell although some of them were centrally located (Figs. 4 and 5). Nuclei which were oval in shape were observed occasionally (Figs. 4 and 5). The connective tissue was rich in reticular fibres, usually surrounding the acini and connective tissue septa. (Fig. 6). They were abundant clearly in capsule (Fig. 7). Collagen fibres were

also observed surrounding the acini and connective tissue septa. (Fig. 8) They were very clear in the capsule (Fig. 9). No elastic fibres were encountered at this trimester. Endocrine portion was not observed in this trimester.

### **3-1-C: Morphometric study**

At this trimester only the acini, connective tissue and blood vessels could be observed in the pancreas of the camel foetus. Hence, only these parameters were considered in this study. The results obtained from analysis of histological sections by point-counting technique for this stage were shown in Tables 2 and 3.

The mean volume of the pancreas was calculated to be  $1.66 \text{ cm}^3$ . The total number of points falling on the acini and connective were more or less similar, the acini were slightly higher (357 for the acini and 310 for the connective tissue). Accordingly, in this stage the acini accounted for about 44.63% of the total volume of the pancreas, whereas the connective tissue occupied about 38.82%; the absolute volumes being  $0.74 \text{ cm}^3$  and  $0.64 \text{ cm}^3$  respectively (Tables 2 and 3). The number of points falling on the blood vessels were less compared with the other two parameters, being 132. Hence, giving only 16.55% of the total volume of the pancreas and an absolute volume of  $0.28 \text{ cm}^3$  (Tables 2 and 3).

## **3-2: THE SECOND TRIMESTER**

### **3-2- A: Gross anatomy**

The pancreas of the camel in second trimester of CVRL 45-71cm (188-259 days) was grayish pink in color. It was lying at the level of the first five lumbar vertebrae. Its weight ranged from 3-5gm. It consisted of a wide body, quadrate right lobe and long tongue-shaped left lobe (Figs. 10 and 11).

The body of the pancreas was related dorsally to the visceral surface of the liver and caudally to the stomach. It was related cranially to the curvature of the duodenum and ventrally to the spleen (Figs. 10 and 11). The right lobe was measured about 2cm in length, it was related dorsally to the visceral surface of the liver, medially to the portal vein and laterally to the descending duodenum and ventrally to the transverse colon (Figs. 10 and 11). The left lobe was measured

about 5.5cm in length, it was related caudodorsally to the stomach, ventrally to the Metanephros, spleen and cranially to the colon (Figs. 10 and 11).

### **3-2-B: Histology**

#### **3-2-B-1: The exocrine portion**

The pancreas in this trimester consisted of secretory units and the ducts were also observed. The secretory units were tubuloacinar and contained a single row of similar pyramidal epithelial cells converging to a central narrow lumen (Fig. 12). In this trimester the pancreas was covered by connective tissue capsule (Fig. 12). These cells contained spherical and sometimes oval nuclei near the base and centrally located, (Fig 13). Connective tissue septa extended into the pancreas dividing it into lobes and lobules usually rich in blood vessels (Fig. 12). The duct system consisted of intralobular duct (Figs. 14 and 15). These ducts were lined by low cuboidal cells (Figs. 14 and 15). Interlobular ducts were noticed located between the lobules in the connective tissue septa. They were lined by stratified cuboidal cells (Figs. 16 and 17). Collagenous fibres were found in the septa and stroma, surrounded the acini and around the ducts (Fig. 18). Similarly, reticular fibres were surrounding the acini and blood vessels and in the septa (Fig. 19). As in the first trimester no elastic fibres were observed.

#### **3-3-B-2: Endocrine portion**

Endocrine portion which was represented by the islets of Langerhans appeared as pale areas among the acini. The shape of islets varied from oval, rounded or irregular surrounded by connective tissue (Figs. 14, 15, 18 and 20). Islets of Langerhans were first appeared at this trimester.

#### **3-2-C: Morphometric study**

In this trimester the following parameters were calculated for morphometry (Acini, Islets of Langerhans, ducts, Connective tissue and Blood vessels). The mean volume of the pancreas was 2.7cm<sup>3</sup>. The number of points falling on the acini increased at the expense of the connective tissue, being 931 and 306 respectively (Table 4). Hence, the acini occupied about 54.98% and the connective tissue about

27.70% of the volume of the pancreas which represented  $1.48\text{cm}^3$  and  $0.75\text{cm}^3$  of the total volume of the pancreas, respectively (Table 6 and 8). The ducts and islets of Langerhans were first observed in this trimester. The points falling on them were only 35 and 30 respectively. These gave volume densities of about 3.30% for the ducts and 1.82% for the islets of Langerhans. Accordingly the absolute volumes were calculated as  $0.1\text{cm}^3$  and  $0.05\text{cm}^3$  respectively (Table 6). The number of points falling on the blood vessels showed an increment compared to the first trimester and calculated to be 142. Hence, giving a volume density of about 12.20% and absolute volume of about  $0.32\text{cm}^3$  (Table 6 and 8).

### **3-3: THE THIRD TRIMESTER**

#### **3-3-A: Gross anatomy**

The pancreas in the third trimester of CVRL 90-110cm (311-366 days) was grayish pink in colour and covered by great amount of fat (Fig. 21). It was lying at the level of the first five lumbar vertebrae. Its weight was about 5-7gm. It consisted of a wide body, a wide quadrate right lobe and long tongue-shaped left lobe (Figs. 21 and 22).

The body of the pancreas was related dorsally to the visceral surface of the liver and caudally to the stomach. It was related cranially to the curvature of the duodenum and ventrally to the spleen (Fig. 21). The right lobe was measured about 3cm in length, it was related dorsally to the visceral surface of the liver, medially to the portal vein and laterally to the descending duodenum and ventrally to the transverse colon (Fig. 21). The left lobe was measured about 6.5cm in length; it was related caudodorsally to the stomach, ventrally to the Kidney, spleen and the splenic vessels and cranially to the colon (Figs. 21 and 22).

#### **3-3-B: Histology**

##### **3-3-B-1: The exocrine portion**

The pancreas at this trimester consisted of abundant secretory units and clear duct system. The secretory units were tubuloacinar (Fig.23). In this trimester the pancreas was covered by a connective tissue capsule and connective tissue septa

were dividing the parenchyma into lobes and lobules. These septa were rich in blood vessels and adipose tissue (Fig. 23). Similar to the second and first trimesters these cells contained spherical and sometimes oval nuclei near the base and centrally located (Fig. 24). The duct system consisted of intralobular ducts which were lined by cuboidal epithelium (Fig. 24). The interlobular ducts were found between the lobules in connective tissue septa and they were lined by columnar epithelium (Fig. 25). The pancreas in this trimester was rich in reticular fibres surrounding the acini (Fig. 26) and ducts (Fig. 27). Collagen fibres were encountered in the connective tissue septa, in the capsule and surrounding acini (Fig.s 28 and 29). Elastic fibres were seen only in wall of blood vessels (Fig. 30) and were not observed elsewhere in the pancreas at this trimester.

### **3-3-B-2: Endocrine portion**

Islets of Langerhans appeared as pale areas scattered among the acini. Their shape varied from oval, rounded or irregular surrounded by connective tissue, some islets were small and others were large. The islets were separated from the acini by a thin layer of connective tissue (Fig.s 31, 32).

### **3-3-C: Morphometric study**

The mean volume was  $4.18\text{cm}^3$ . Similar parameters to those of the second trimester were calculated in this stage. The numbers of points falling on the acini and the connective tissue were found to be the highest. They were found to be 1300 and 785 respectively. The volume density for the acini was about 60.95% and that for the connective tissue was about 26.75%. Hence the absolute volumes calculated for the two parameters were  $2.55\text{cm}^3$  and  $1.12\text{cm}^3$  respectively (Tables 5, 7 and 9). The number of points falling on ducts and islets of Langerhans showed further increase being 54 and 40 respectively. The calculated volume densities and absolute volumes for the ducts were 2.44% ( $0.10\text{cm}^3$ ) and those for islets of Langerhans were 2.52% ( $0.11\text{cm}^3$ ).

**Table 2.** Showing points falling on each parameter, the volume densities and the absolute volumes of the different components of the pancreas at the first trimester for 5 sections shown as means  $\pm$  Standard Deviation.

Section No.	No. of fields counted	Acini	Connective Tissue	Blood Vessels	Total No. of Points
1	8	369	361	70	800
2	8	262	483	55	800
3	8	425	111	264	800
4	8	352	318	130	800
5	8	377	280	143	800
Total	40	1785	1553	662	4000
Mean	357		310.6	132.4	
Volume density	44.63%		38.82%	16.55%	100%
Absolute Volume	0.74 cm <sup>3</sup>		0.64 cm <sup>3</sup>	0.28 cm <sup>3</sup>	

**Table 3.** Showing the volume of fresh pancreas, volume densities absolute volumes of the main components in 5 camels foetuses at the first trimester expressed as Means  $\pm$  Standard Deviations (SD).

Animal No.	Volume of fresh pancreas	Acini		Connective tissue		Blood vessels	
		Vv %	Abs V cm <sup>3</sup>	Vv %	Abs V cm <sup>3</sup>	Vv %	Abs V cm <sup>3</sup>
1	2 cm <sup>3</sup>	46	0.92	45	0.9	8.75	0.17
2	1.5cm <sup>3</sup>	32.75	0.49	60.77	0.91	6.87	0.1
3	1.9 cm <sup>3</sup>	53.12	1	13.87	0.26	33	0.62
4	1.7 cm <sup>3</sup>	44	0.74	39.75	0.67	16.25	0.27
5	1.2 cm <sup>3</sup>	47.12	0.56	35	0.42	17.87	0.21
<b>Mean</b>	1.66 $\pm$ 0.32	44.63 $\pm$ 7.44	0.74 $\pm$ 0.22	38.82 $\pm$ 17.01	0.64 $\pm$ 0.29	16.55 $\pm$ 10.33	0.28 $\pm$ 0.20

**Table 4.** Showing the data obtained by point-counting of fields on seven sections of the pancreas at the second trimester.

Section No.	Number of points falling on						
	No. of fields counted	Acini	Islets of Langerhans	Ducts	Connective tissue	Blood vessels	Total
1	1	80	0	0	19	1	100
	2	69	0	0	31	0	100
2	1	48	1	5	34	12	100
	2	86	3	7	3	1	100
3	1	70	1	0	20	9	100
	2	59	1	1	35	4	100
4	1	54	0	5	34	7	100
	2	62	0	3	20	15	100
5	1	80	4	0	16	0	100
	2	71	4	1	9	15	100
6	1	72	2	4	13	9	100
	2	56	5	1	30	8	100
7	1	34	6	8	36	16	100
	2	90	3	0	6	1	100
<b>Total</b>	<b>14</b>	<b>931</b>	<b>30</b>	<b>35</b>	<b>306</b>	<b>142</b>	<b>1400</b>

**Table 5.** Showing the data obtained by point-counting of fields on twelve sections of the pancreas at the third trimester.

Section No.	Number of points falling on						
	No. of fields counted	Acini	Islets of Langerhans	Ducts	Connective tissue	Blood vessels	Total
1	1	70	5	0	20	5	100
	2	40	1	2	44	13	100
2	1	59	0	1	31	9	100
	2	32	0	0	50	18	100
3	1	57	4	3	26	10	100
	2	33	2	2	60	3	100
4	1	60	6	4	21	9	100
	2	40	5	2	35	18	100
5	1	51	2	1	10	36	100
	2	80	0	6	12	2	100
6	1	36	4	2	52	6	100
	2	80	1	0	19	0	100
7	1	42	2	3	51	2	100
	2	50	3	5	39	3	100
8	1	80	4	0	10	6	100
	2	36	0	2	60	2	100
9	1	50	4	1	30	15	100
	2	48	2	2	36	12	100
10	1	74	1	0	22	3	100
	2	39	0	3	50	8	100
11	1	54	1	0	33	12	100
	2	66	3	0	30	1	100
12	1	90	2	1	2	5	100
	2	33	2	0	42	23	100
<b>Total</b>	<b>24</b>	<b>1300</b>	<b>54</b>	<b>40</b>	<b>785</b>	<b>221</b>	<b>2400</b>



**Table 6.** Showing points falling on each parameters, the volume densities and the absolute volumes of the different components of the pancreas at the second trimester 5 sections shown as Means  $\pm$  Standard Deviation.

Section No.	No. of fields counted	Acini	Islets of Langerhans	Ducts	Connective tissue	Blood vessels	Total of points
1	14	659	50	80	305	306	1400
2	14	714	13	20	552	101	1400
3	14	931	30	35	306	98	1400
4	14	705	19	55	369	252	1400
5	14	840	16	40	407	97	1400
<b>Total</b>	70	3849	128	230	1939	854	7000
<b>Mean</b>		769.8	25.6	46	387.8	170.8	100%
<b>Volume densities</b>		54.98%	1.82%	3.30%	27.70%	12.20%	
<b>Absolute volumes</b>		1.48cm <sup>3</sup>	0.05cm <sup>3</sup>	0.10cm <sup>3</sup>	0.75cm <sup>3</sup>	0.32cm <sup>3</sup>	

**Table 7.** Total data obtained by point-counting of fields of five histological sections at the third trimester.

Section No.	No. of fields counted	Acini	Islets of Langerhans	Ducts	Connective tissue	Blood vessels	No. of points
1	24	1300	54	40	785	221	2400
2	24	1654	67	55	502	122	2400
3	24	1504	49	80	579	188	2400
4	24	1486	62	60	602	190	2400
5	24	1370	70	58	742	160	2400
<b>Total</b>	120	7314	302	293	3210	881	12000
<b>Means</b>		1462.8	60.4	58.6	642	176.2	100%
<b>Volume densities</b>		60.95%	2.52%	2.44%	26.75%	7.34%	
<b>Absolute volumes</b>		2.55c <sup>3</sup>	0.11cm <sup>3</sup>	0.10cm <sup>3</sup>	1.12cm <sup>3</sup>	0.30cm <sup>3</sup>	

**Table 8.** Showing the volume of fresh pancreas and volume densities of their main components in 5 camel fetuses at the second trimester expressed as mean and standard deviations, Mean absolute volumes of components indicated in cm<sup>3</sup>.

Animal number	Volume of pancreas	Acini	Islets of Langerhans	Ducts	Connective tissue	Blood vessels
	Abs V cm <sup>3</sup>	Abs V cm <sup>3</sup>	Abs V cm <sup>3</sup>	Abs V cm <sup>3</sup>	Abs V cm <sup>3</sup>	Abs V cm <sup>3</sup>
1	5.70	0.20	21.70	0.76	22.00	0.80
2	1.40	0.03	39.40	0.98	7.20	0.18
3	2.50	0.037	21.80	0.32	7.00	0.10
4	3.90	0.10	26.30	0.73	18.00	0.50
5	2.85	0.09	29.00	0.92	6.92	0.20
<b>Mean</b>	3.30±	0.10±	27.70±	0.75±	12.20±	0.32±
	1.62	0.06	7.26	0.25	7.23	0.29

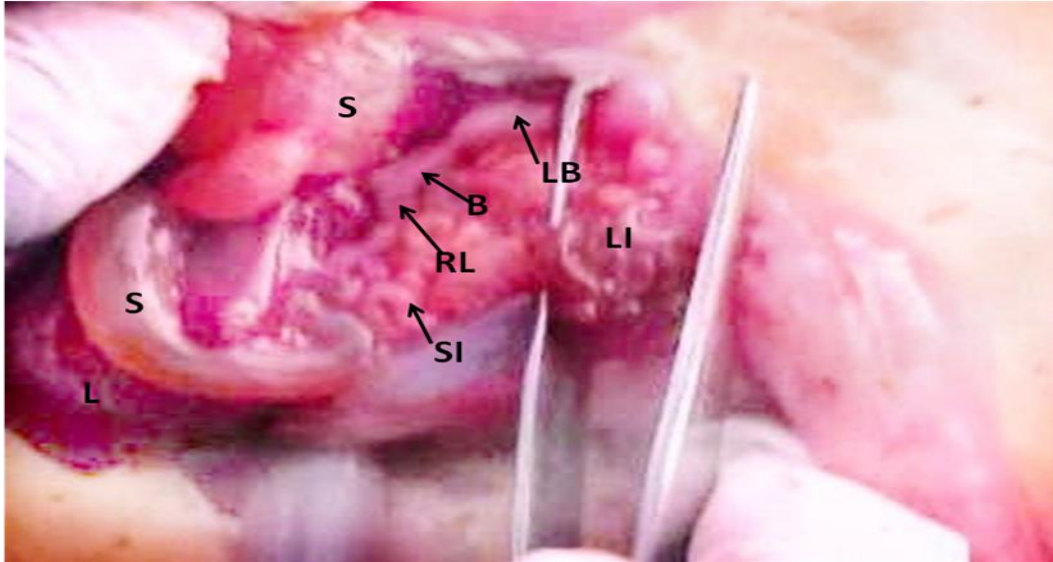
**Table 9.** Showing the volume of fresh pancreas and volume densities of their main components in 5 camel fetuses at the third trimester expressed as mean and standard deviations, Mean absolute volumes of components indicated in cm<sup>3</sup>.

Animal number	Volume of pancreas	Acini		Islets of Langerhans		Ducts		Connective tissue		Blood vessels	
		%	Cm <sup>3</sup>	%	Cm <sup>3</sup>	%	Cm <sup>3</sup>	%	Cm <sup>3</sup>	%	Cm <sup>3</sup>
1	6.1 cm <sup>3</sup>	54	3.20	2.20	0.13	1.60	0.10	32.7	1.90	9.20	0.60
2	5 cm <sup>3</sup>	68.90	3.40	2.70	0.13	2.30	0.10	20.90	1.00	5.00	0.25
3	4 cm <sup>3</sup>	62.60	2.50	2.00	0.08	3.30	0.13	24.10	0.96	7.80	0.30
4	3.5 cm <sup>3</sup>	61.90	2.16	2.58	0.09	2.50	0.08	25.00	0.87	7.90	0.24
5	2.3 cm <sup>3</sup>	57.00	1.30	2.91	0.06	2.40	0.05	30.90	0.71	6.60	0.15
<b>Mean</b>	4.18±	60.95	2.55±	2.52	0.11±	2.44	0.10±	26.75	1.12±	7.34	0.30±
	1.44	±	0.84	±	0.03	±	3.54	±	0.47	±	0.17
		5.71		0.37		0.61		4.92		1.58	

**Table 10.** Summarizing the result shown in Table 8 and Table 9.

Stage of gestation	Volume of fresh pancreas	Acini		Islets of Langerhans		Ducts		Connective tissue		Blood vessels	
		%	Cm <sup>3</sup>	%	Cm <sup>3</sup>	%	Cm <sup>3</sup>	%	Cm <sup>3</sup>	%	Cm <sup>3</sup>
<b>Second trimester</b>	2.7±	54.98	1.48	1.82	0.05±	3.3	0.1	27.7	0.75	12.22	0.32
	0.77	±	±	±	0.04	±	±	±	±	±	±
		8.05	0.34	1.03		1.62	0.06	7.26	0.25	7.23	0.29
<b>Third trimester</b>	4.18±	60.95	2.55	2.52	0.11±	2.44	0.10	26.75	1.12	7.34	0.30
	1.44	±	±	±	0.03	±	±	±	±	±	±
		5.71	0.84	0.37		0.61	3.54	4.92	0.47	1.58	0.17

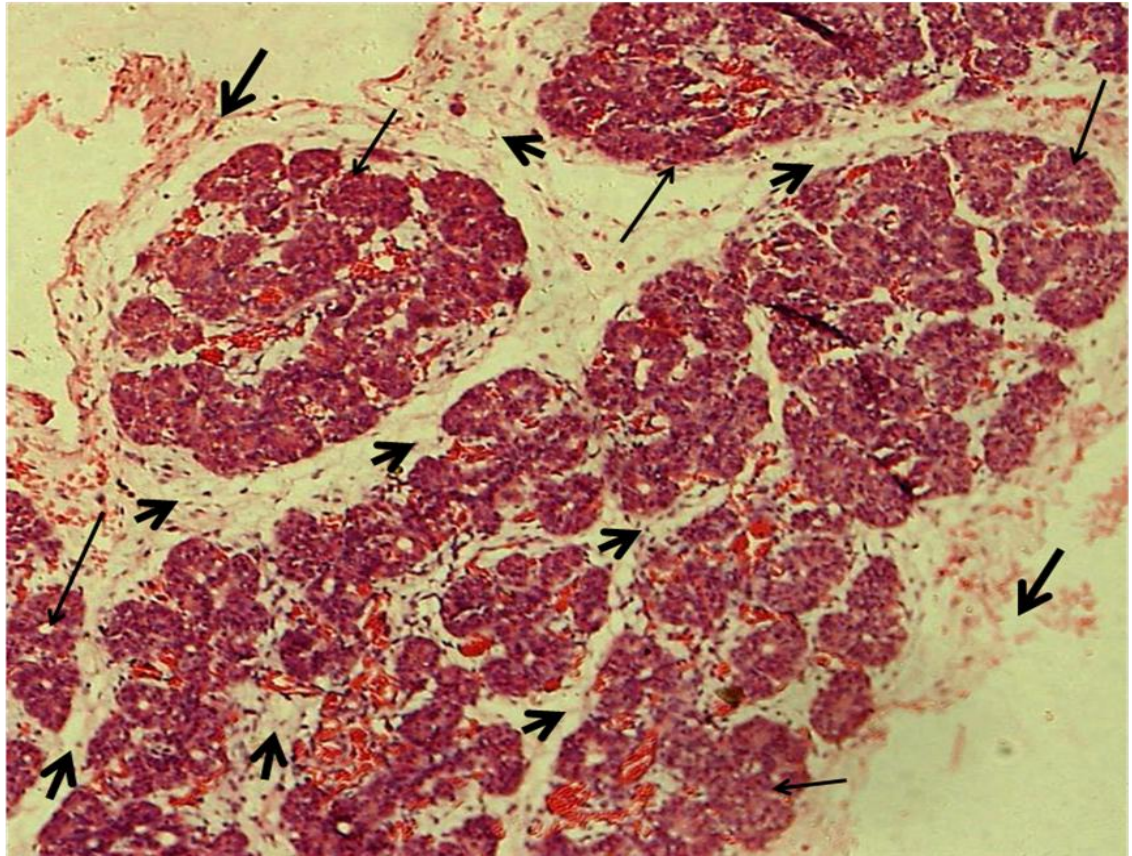
## **FIGURES**



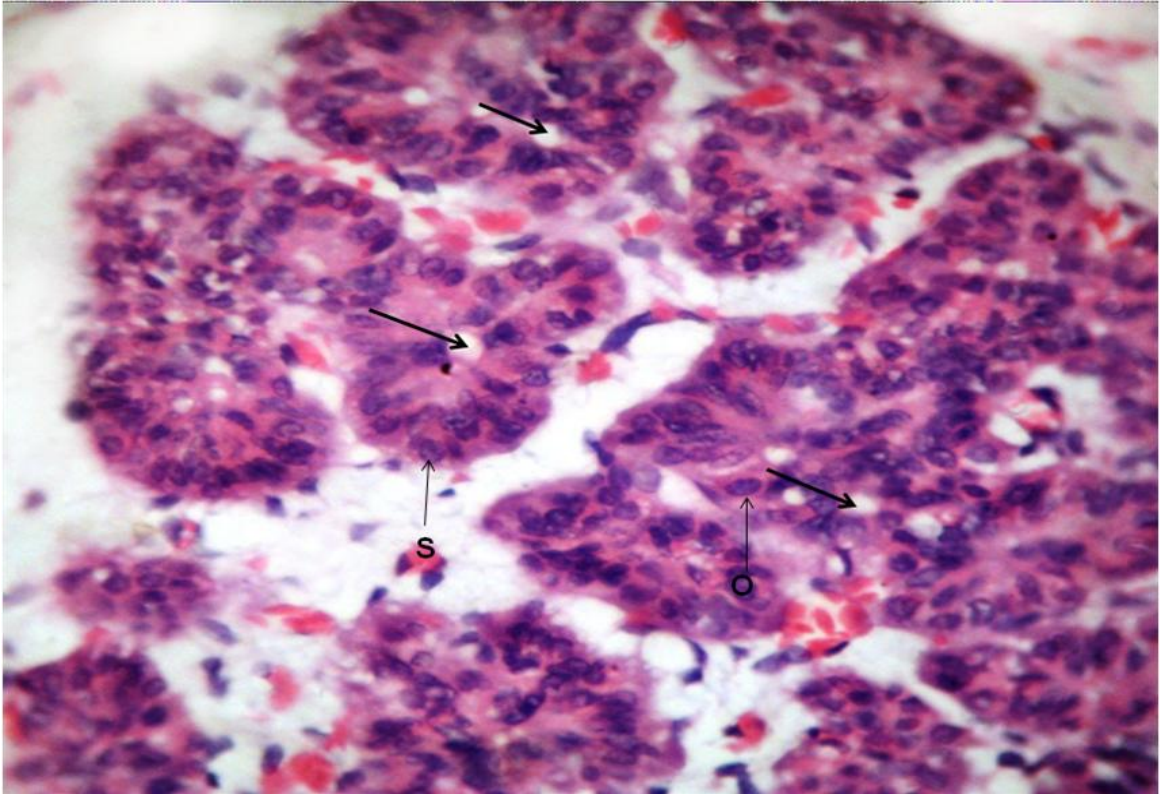
**Fig. 1.** A photograph showing the pancreas of a camel fetus (Left view) at the first trimester of CVRL 8cm (87 days), S, stomach: LB, left lobe of pancreas, B, body of pancreas, R, right lobe of pancreas, SI, small intestine (duodenum), LI, large intestine (colon) L, right lobe of liver.



**Fig. 2.** A photograph showing the pancreas of a camel fetus (Left view) at the first trimester of CVRL 11cm (95 days), stomach: S, pancreas (arrow), L, right lobe of liver, IN, Intestine, SP, Spleen, K, mesonephros.

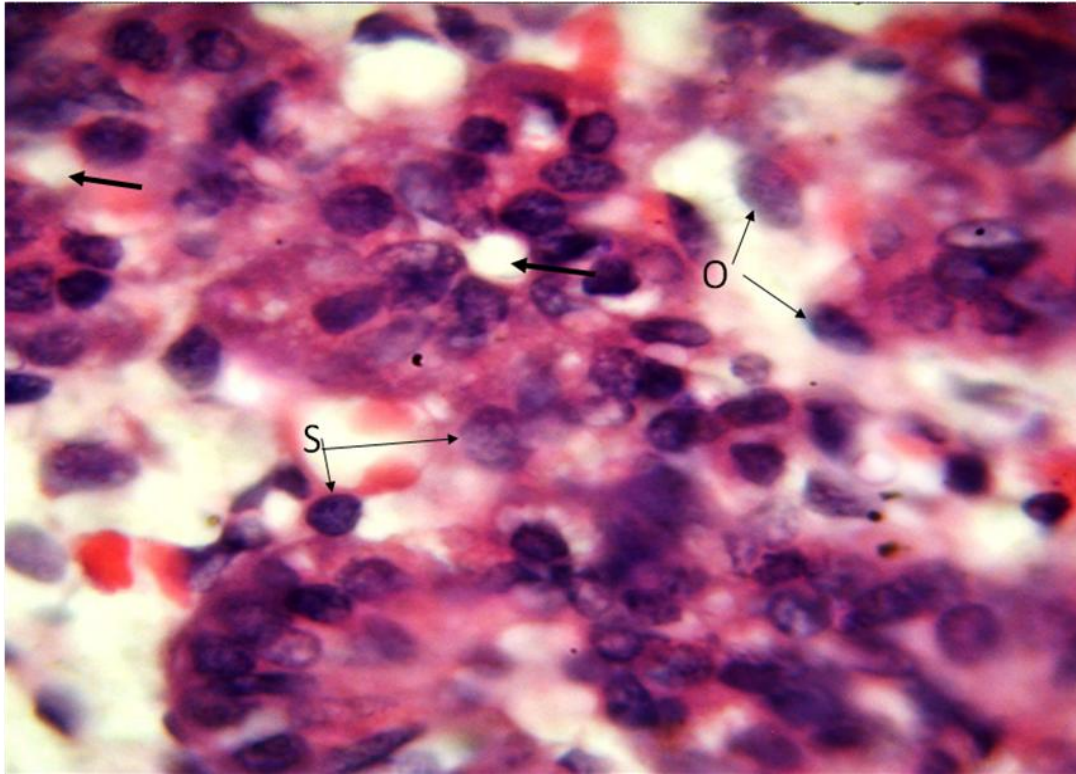


**Fig. 3.** A micrograph showing the pancreas of the camel foetus at the first trimester of CVRL 15cm (106 days), showing lobules ( thin arrows) surrounded by connective tissue septa (arrowheads) and capsule (thick arrows). H&E staining, (X100).

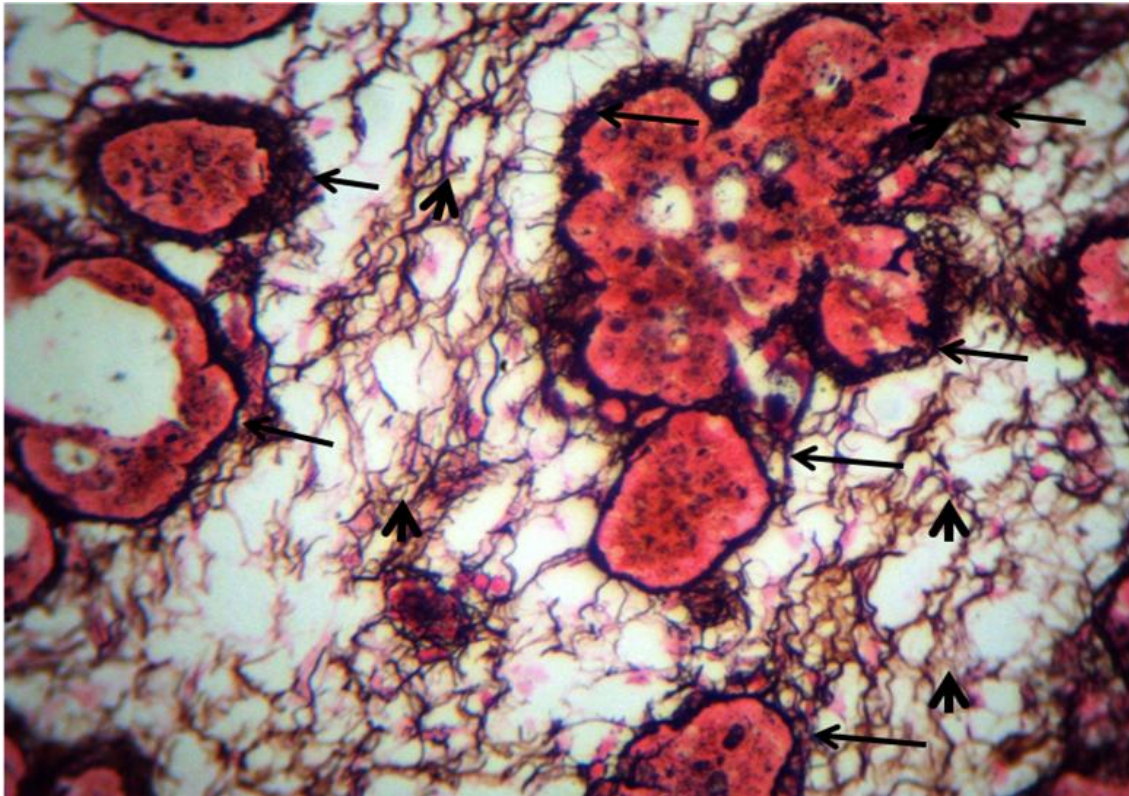


**Fig. 4.** A micrograph of the pancreas of the camel foetus at the first trimester of CVRL 17cm (111 days) showing narrow lumina (arrows) of the acini with different shapes of nuclei, O, oval ,S, spherical. H&E staining, (X200).

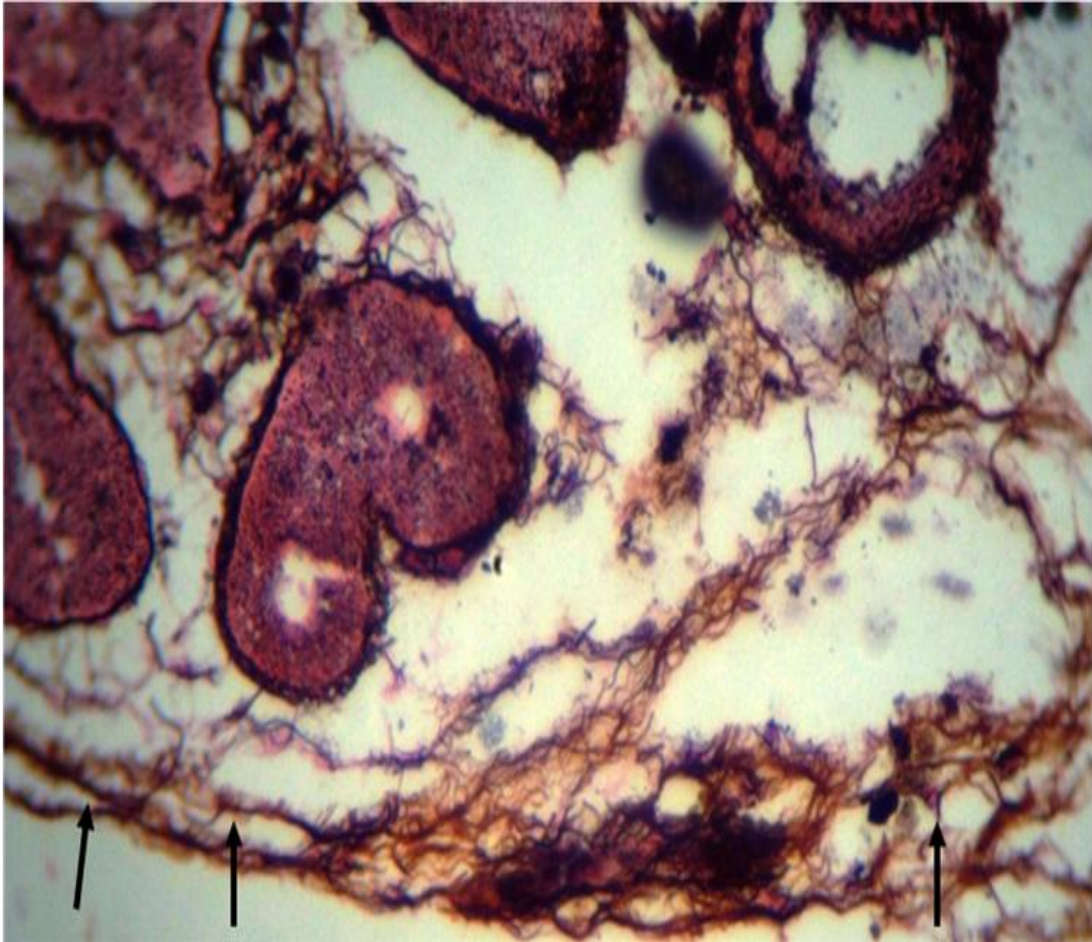




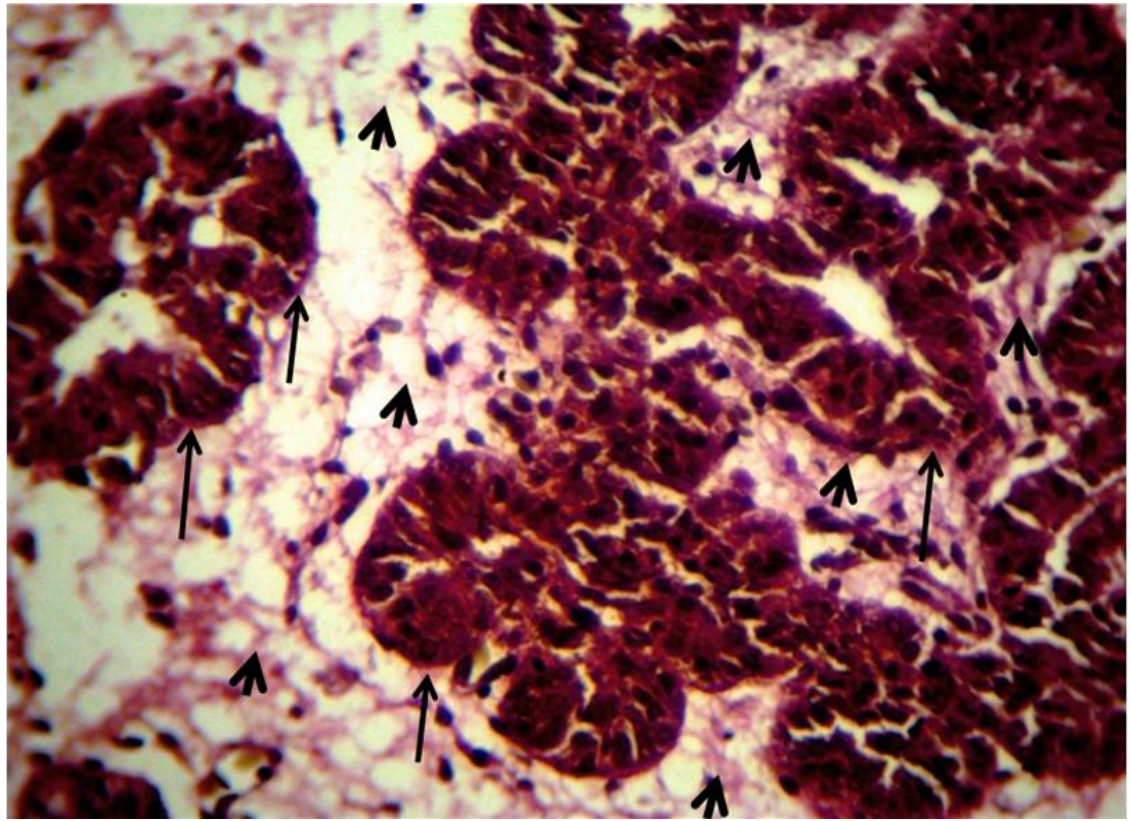
**Fig. 5.** A high magnification of Fig. 4 of the pancreas of the camel foetus of CVRL 17cm (111 days), Showing narrow lumen (thick arrows) of the acini with different shapes of nuclei O: oval nuclei, S: spherical nuclei. H & E staining (X1000).



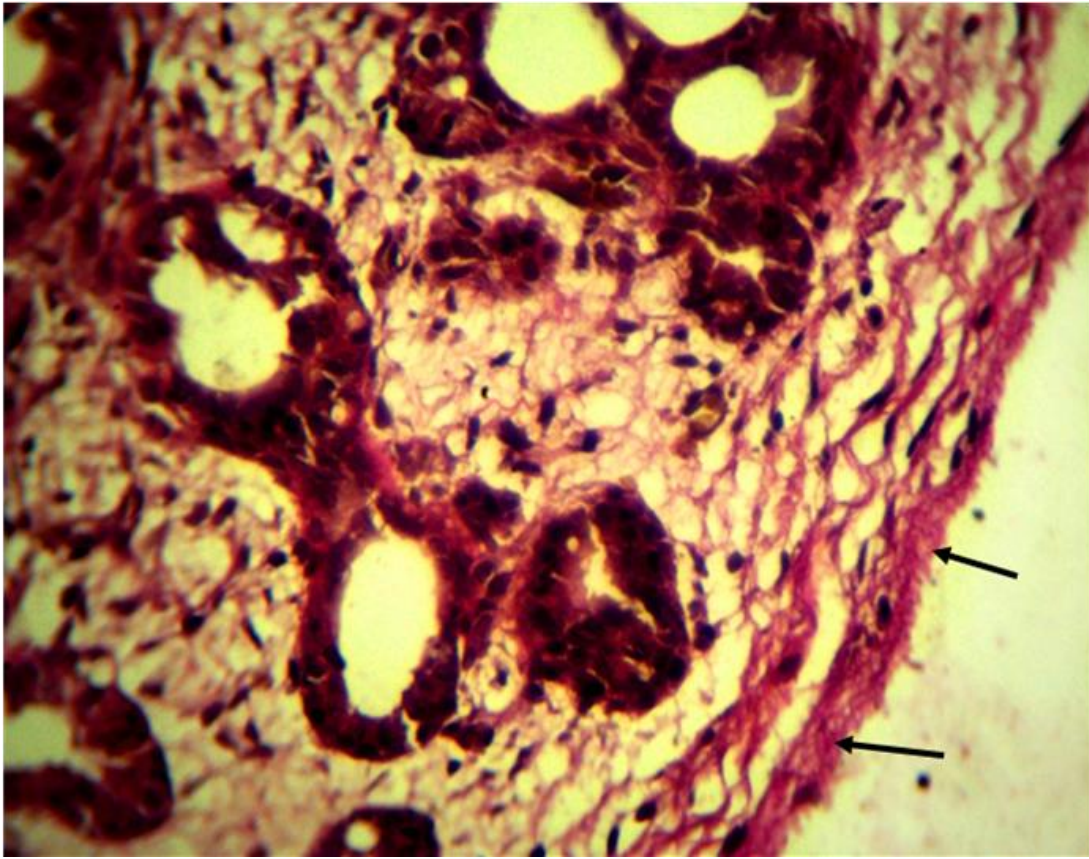
**Fig. 6.** A micrograph of the pancreas of the camel foetus first trimester of CVRL 12 cm (98 days) showing reticular fibers surrounding the acini (arrows) and in the connective tissue septa (arrowheads). Silver staining, (X400).



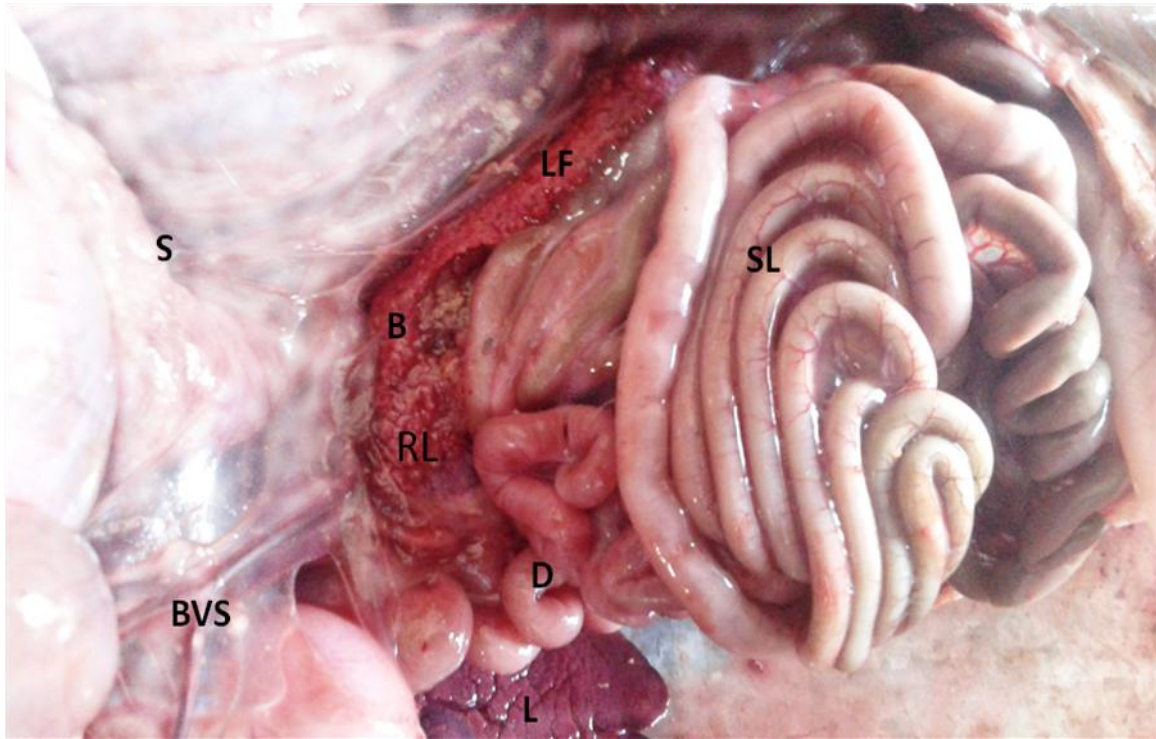
**Fig. 7.** A photomicrograph showing the pancreas of the camel foetus of first trimester CVRL 12 cm (98 days) showing reticular fibres in the capsule (arrows). Silver staining, (X400).



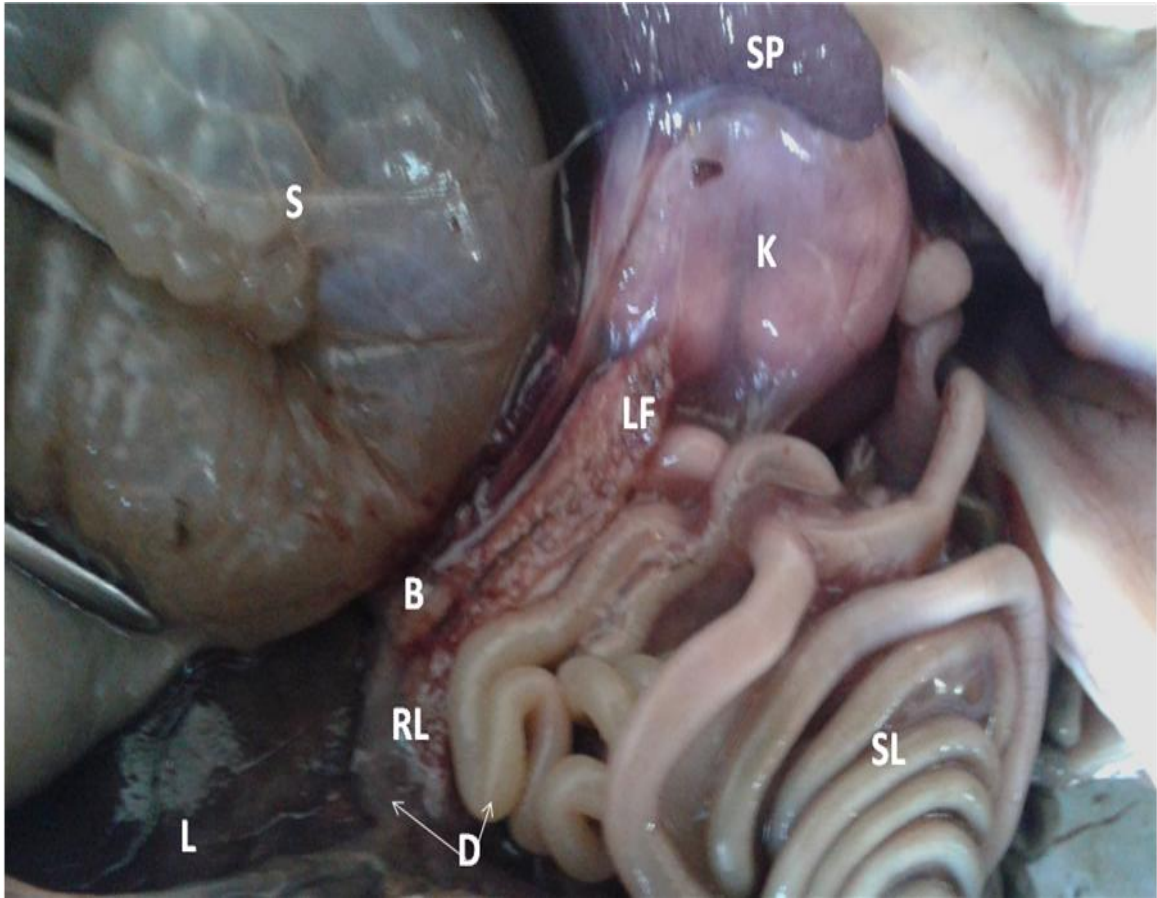
**Fig. 8.** A micrograph of the pancreas of camel foetus in the first trimester of CVRL12 cm (98 days) showing collagenous fibres surrounding the acini (arrows) and in the connective tissue septa (arrowheads). Van Gieson staining (X400).



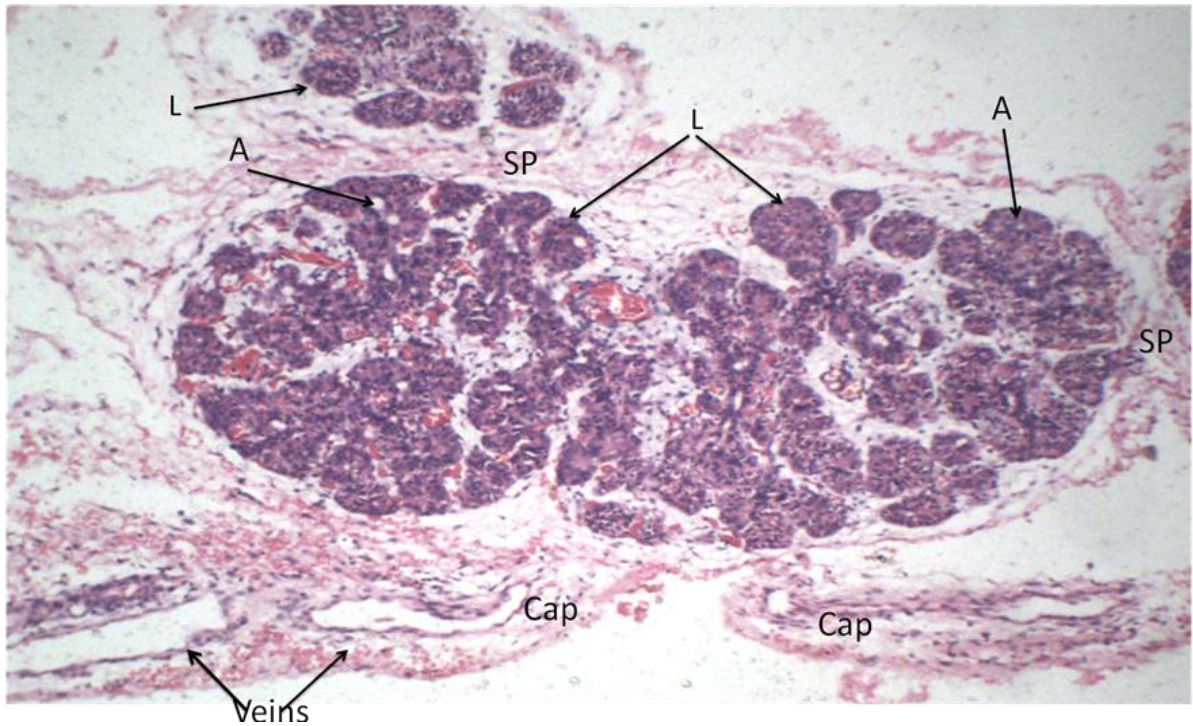
**Fig. 9.** A micrograph of the pancreas of camel foetus in the first trimester of CVRL12 cm (98 days) showing the collagenous fibres in the connective tissue capsule. Van Gieson staining (X400).



**Fig. 10.** A photograph showing the pancreas of a camel fetus (Left view). at the second trimester, CVRL 57cm (221 days). S, stomach, LF, left lobe of the pancreas, B, body of the pancreas, RL, right lobe of the pancreas, L, right lobe of liver, D, duodenum, SL , spiral loop of the colon, BVS, Blood vessels (caudal pancreatoduodenal artery).

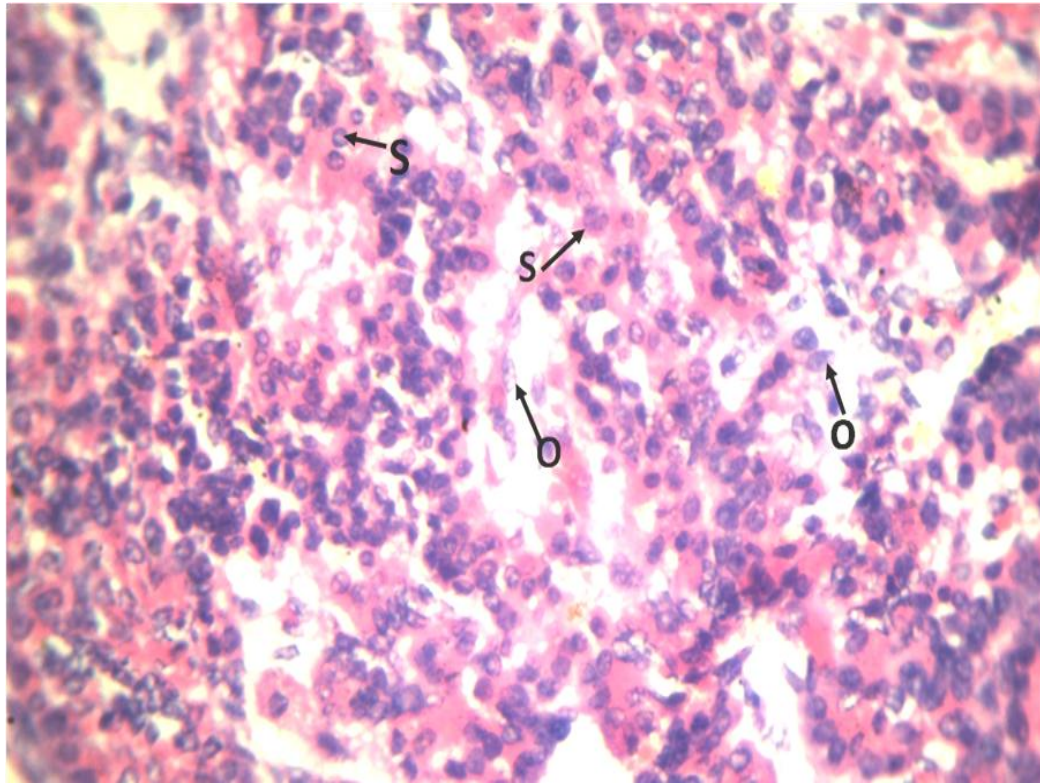


**Fig. 11.** A photograph of the pancreas of a camel fetus (Left view) in the second trimester of CVRL 45cm (188 days). S, stomach, : LF, left lobe of the pancreas, B, body of the pancreas, RL, right lobe of the pancreas, L, right lobe of liver, D, duodenum, SL, spiral loop of colon, SP, spleen, k, Metanephros.

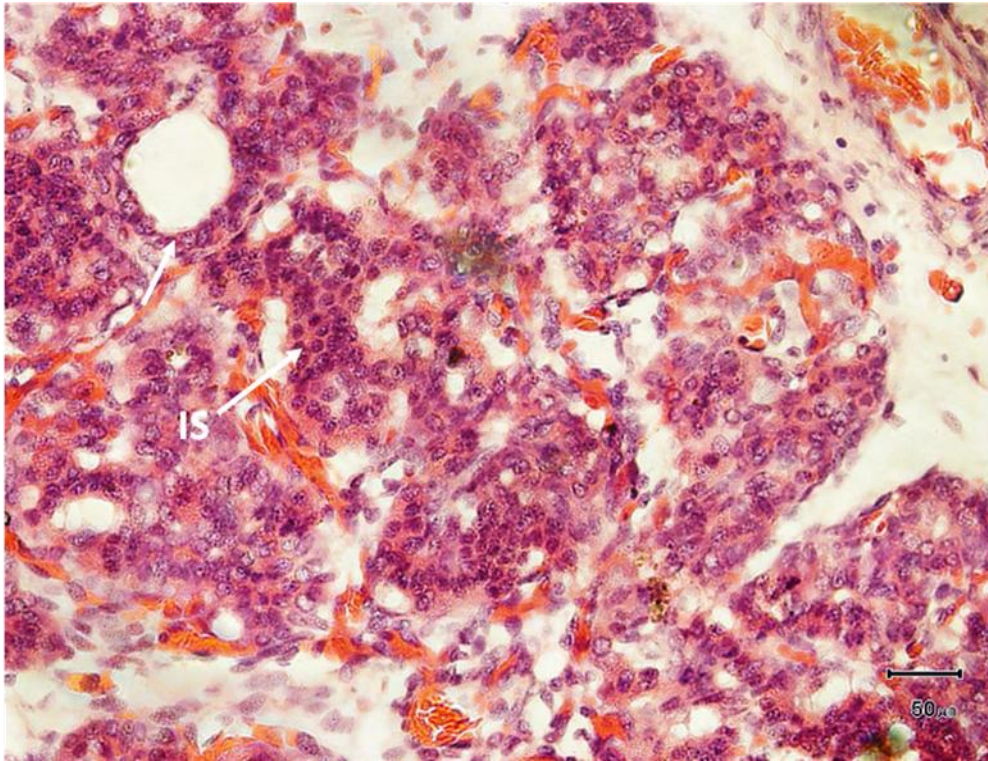


**Fig. 12.** A micrograph of the pancreas of a camel foetus of CVRL 50cm (202 days) at the second trimester, lobules: (L), (Cap) connective tissue capsule, (SP) Connective tissue septa, (A), acini and blood vessels (Veins). H&E staining, (X100).

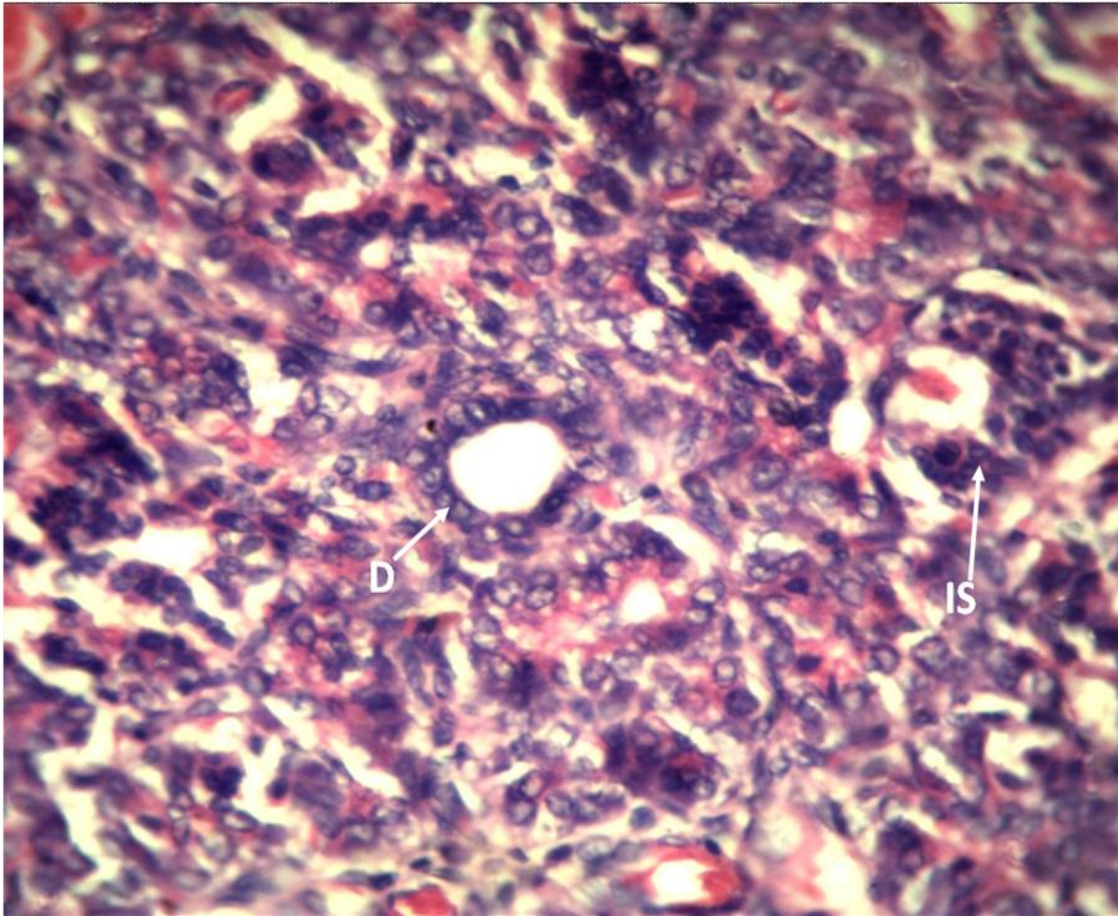




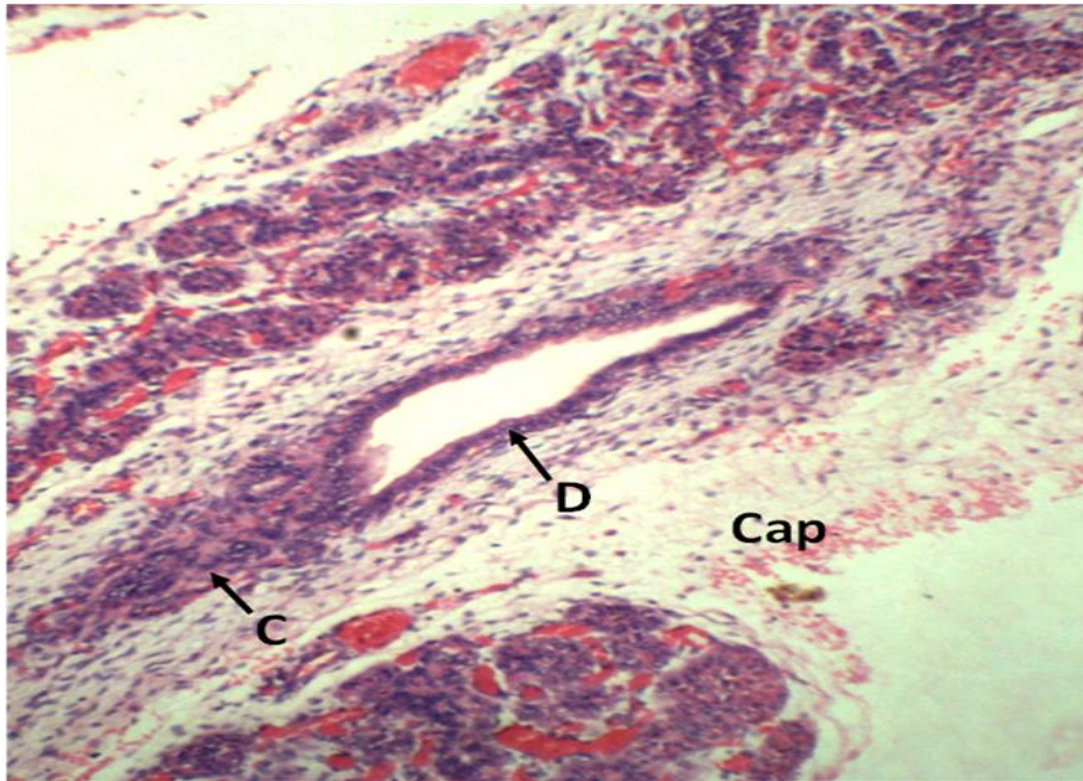
**Fig. 13.** A micrograph of the pancreas of a camel foetus of CVRL70cm (256 days) second trimester, O, oval nuclei, S, spherical nuclei. H&E staining, (X400)



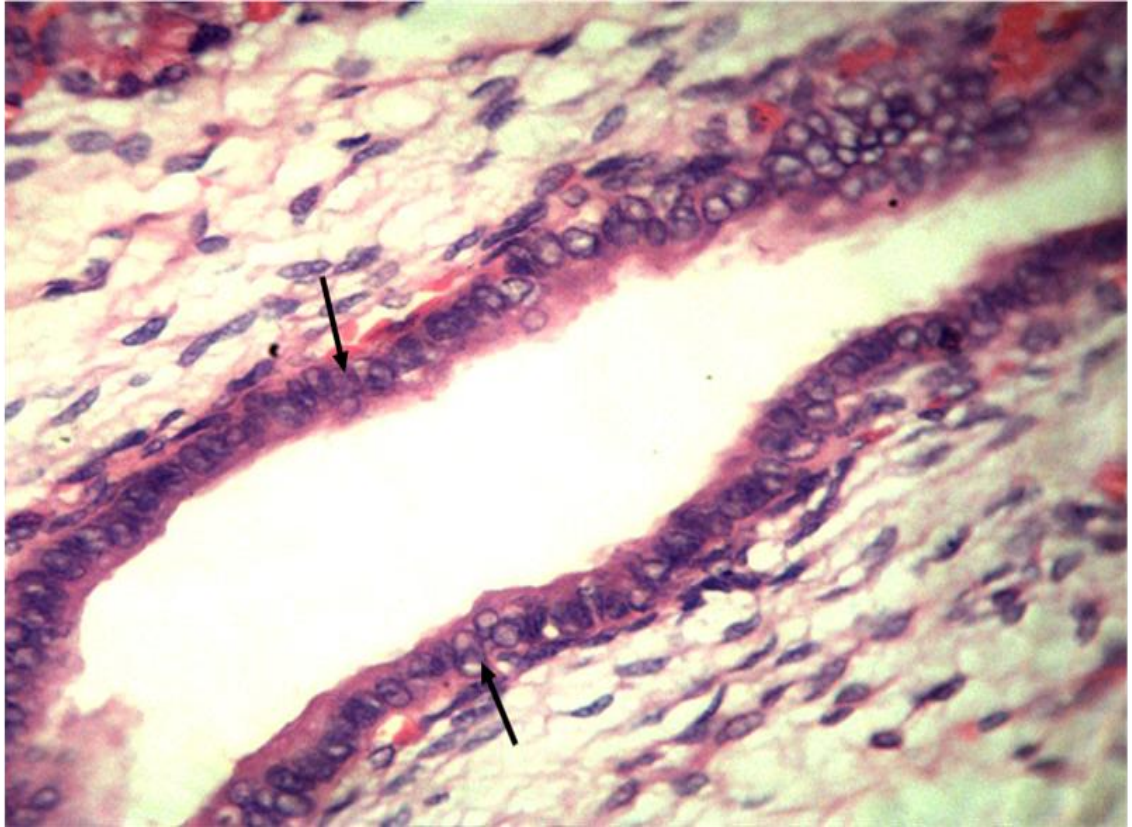
**Fig. 14.** A micrograph showing the pancreas of a camel foetus CVRL 60cm (229 days) second trimester, showing an intralobular duct (arrow) and islets of Langerhans IS. H&E staining, (X400).



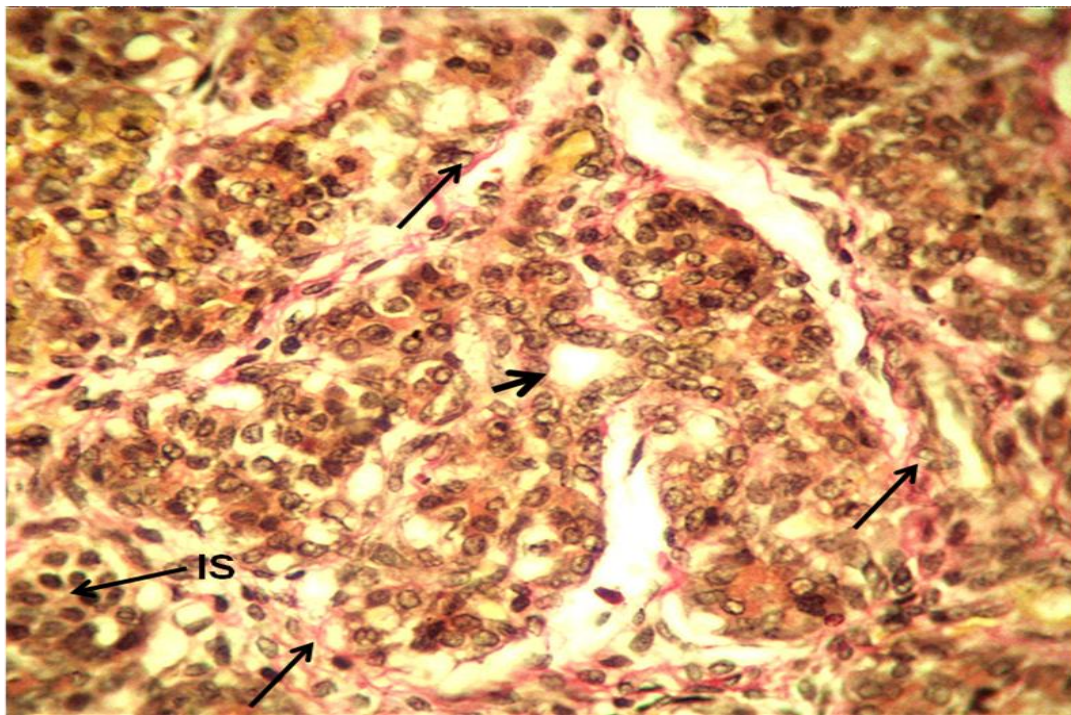
**Fig. 15.** A micrograph of the pancreas of a camel foetus of CVRL70cm (256 days) second trimester, showing islets of Langerhans IS, D, intralobular duct lining by cuboidal cells, H&E staining, (X400).



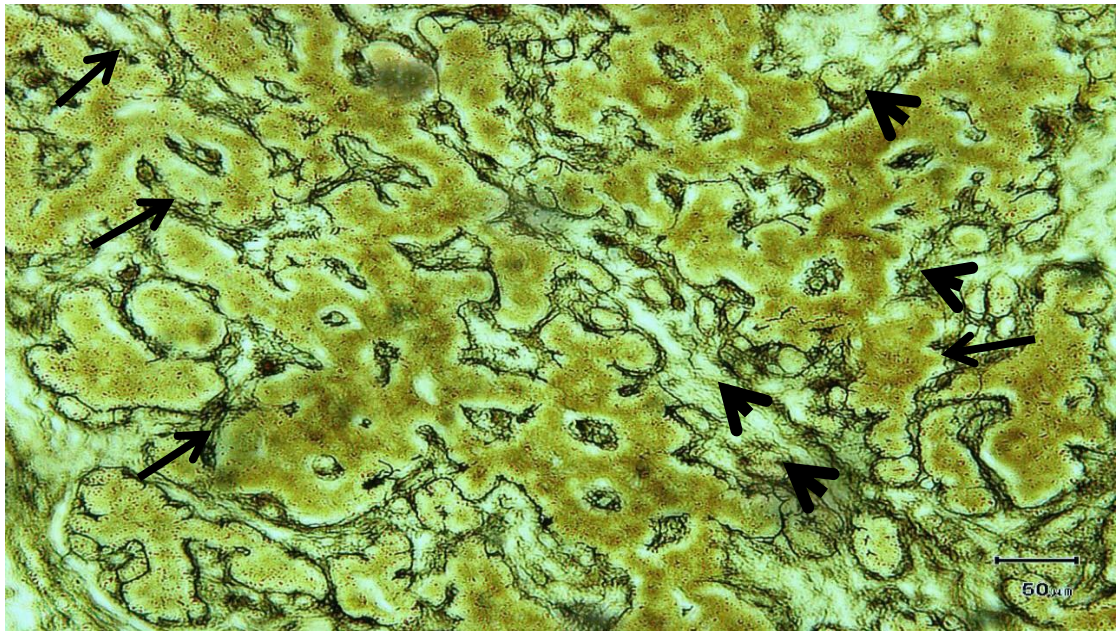
**Fig. 16.** A micrograph of the pancreas of a camel foetus of CVRL60cm (229 days) second trimester, showing D, interlobular duct lined by stratified cuboidal epithelium , C, thick connective tissue, Cap, connective tissue capsule. H&E staining, (X100).



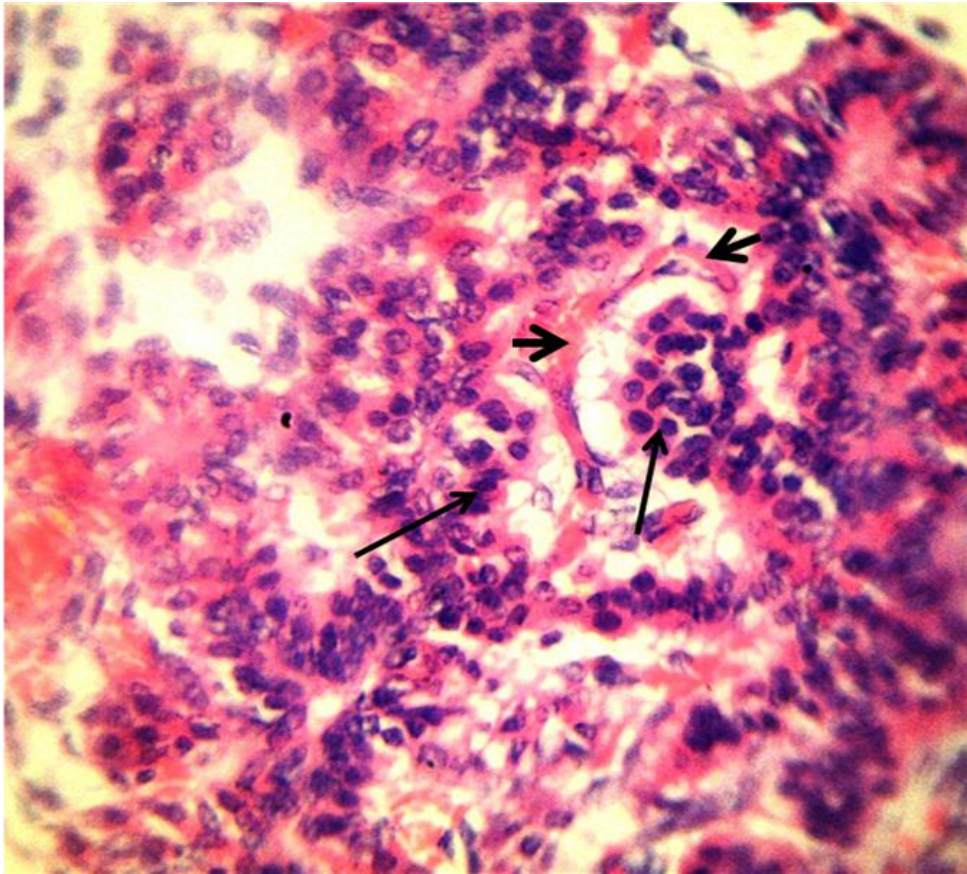
**Fig. 17.** A high magnification of Fig. 16 of the pancreas of the camel foetus of CVRL 60cm (229 days) showing the stratified cuboidal epithelium of the interlobular duct (arrows). H&E staining (X1000).



**Fig. 18.** A micrograph of the pancreas of a camel foetus at the second trimester of CVRL68 cm (251 days) showing collagenous fibres surrounding the acini (arrows), islets of Langerhans IS and intralobular duct (arrowhead). van Gieson staining, (X400).

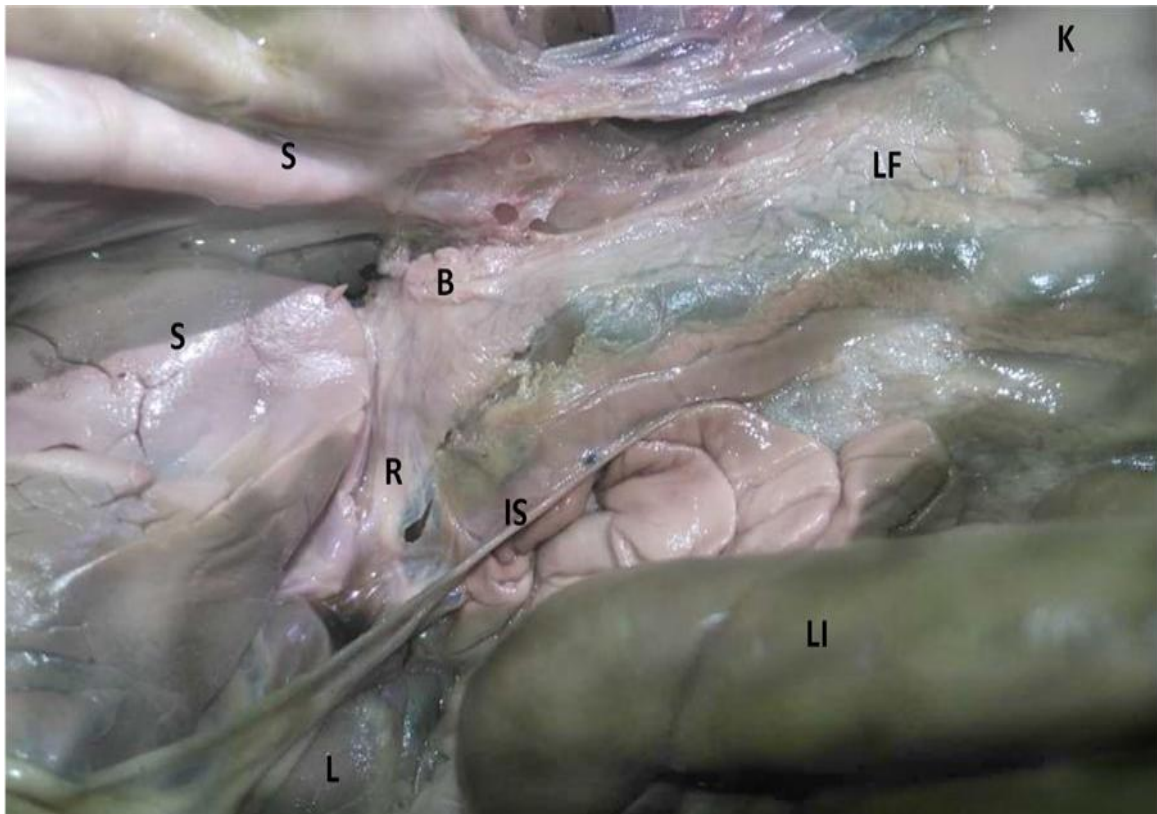


**Fig. 19.** A micrograph showing the pancreas of a camel foetus at the second trimester of CVRL 57cm (221 days). Reticular fibres surrounding the acini (arrows) and in connective tissue septa (arrow heads). Silver staining, (X400).

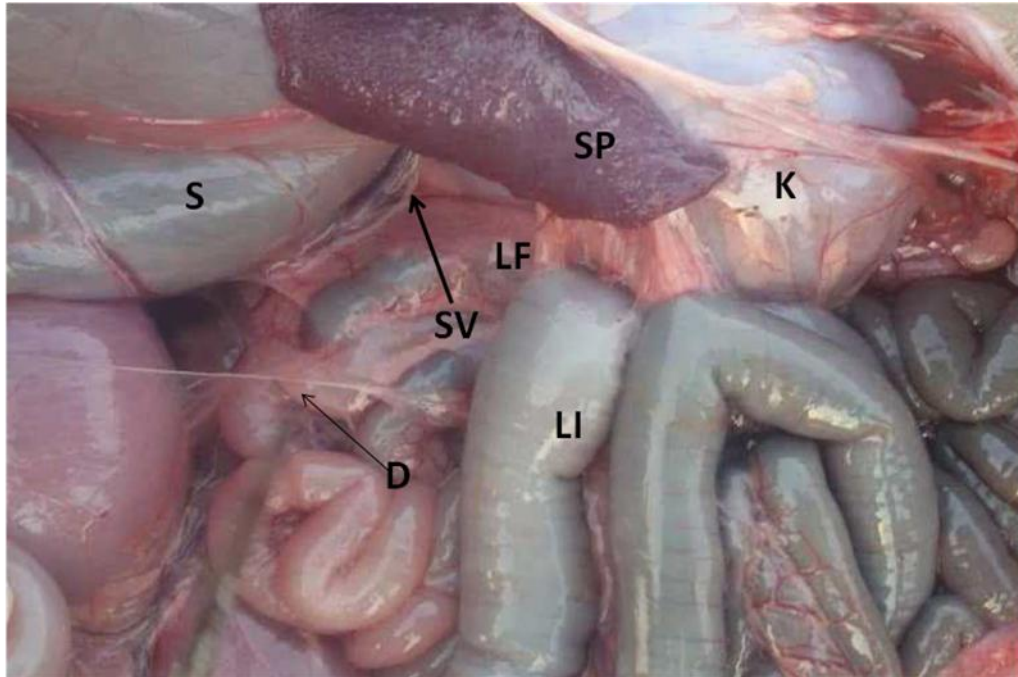


**Fig. 20.** A photograph of the pancreas of the camel foetus at the second trimester of CVRL 60cm (229 days), showing the islets of Langerhans (arrows) surrounded by thin connective tissue (arrowheads). H&E staining, (X400).

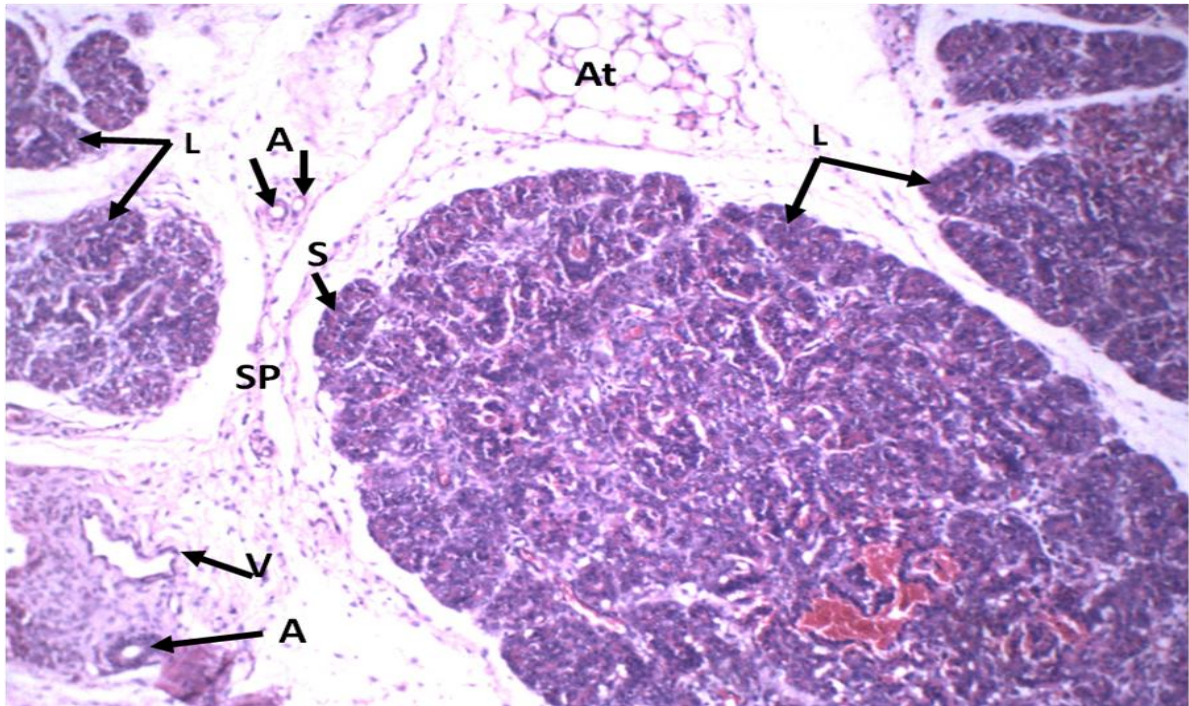




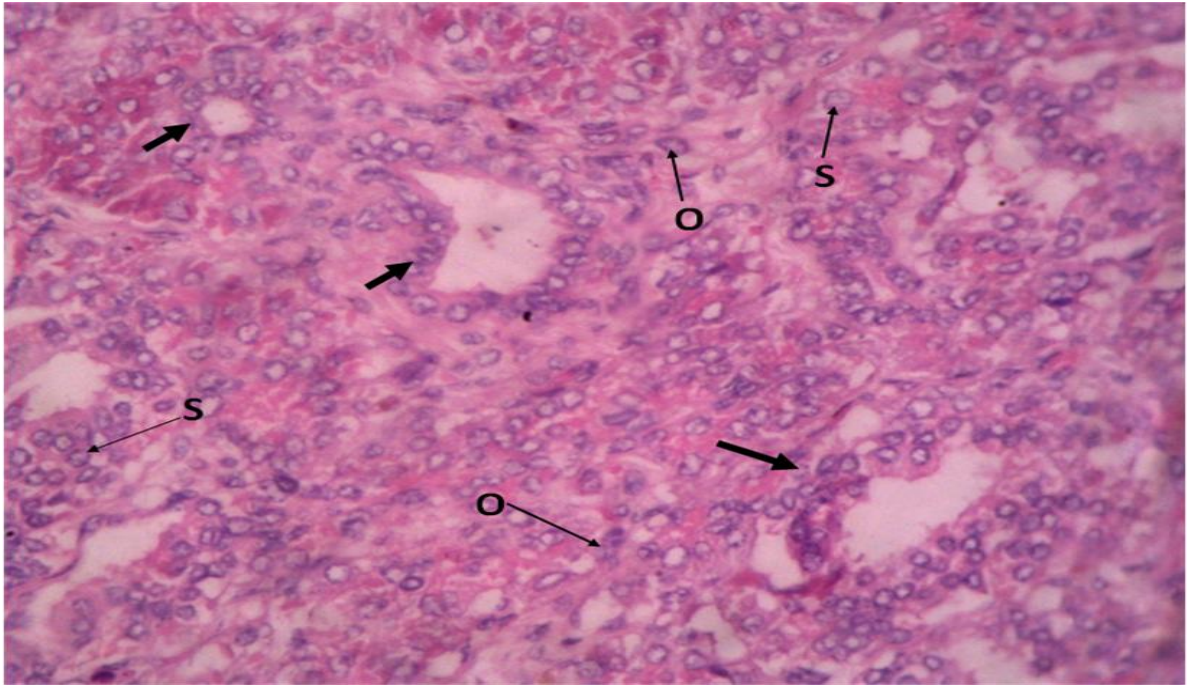
**Fig. 21.** A photograph showing the pancreas of a camel fetus. covered by great amount of fat (Left view). At the third trimester, CVRL 90cm (311 days). S, stomach, LF, left lobe, B, body of the pancreas, R, right lobe of the pancreas, L, right lobe of liver, SI, small intestine(Duodenum), LI , Large intestine(colon), K, kidney.



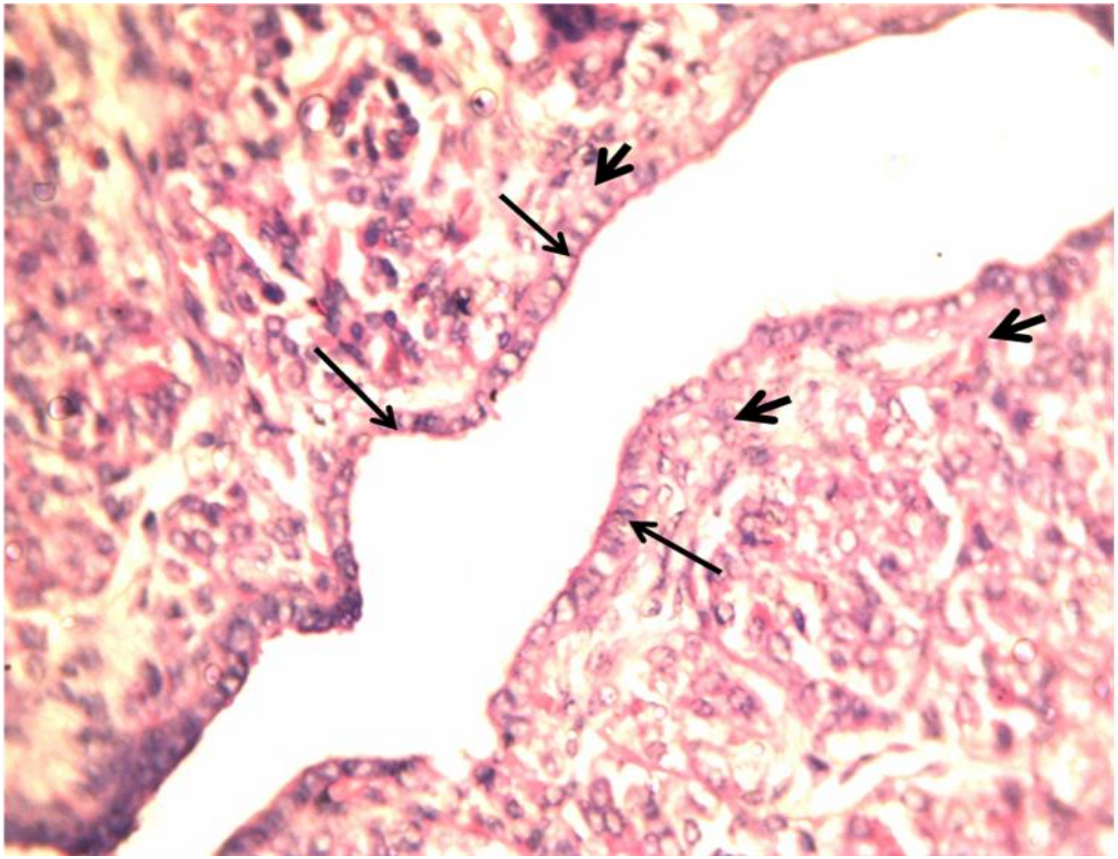
**Fig. 22.** A photograph of the pancreas of a camel foetus in the third trimester (Left view), CVRL 110 cm (366 days) showing, S, stomach, LF, left lobe of pancreas, , D duodenum flexure, LI, large intestine, SP spleen, SV, splenic vessels, K, left kidney.



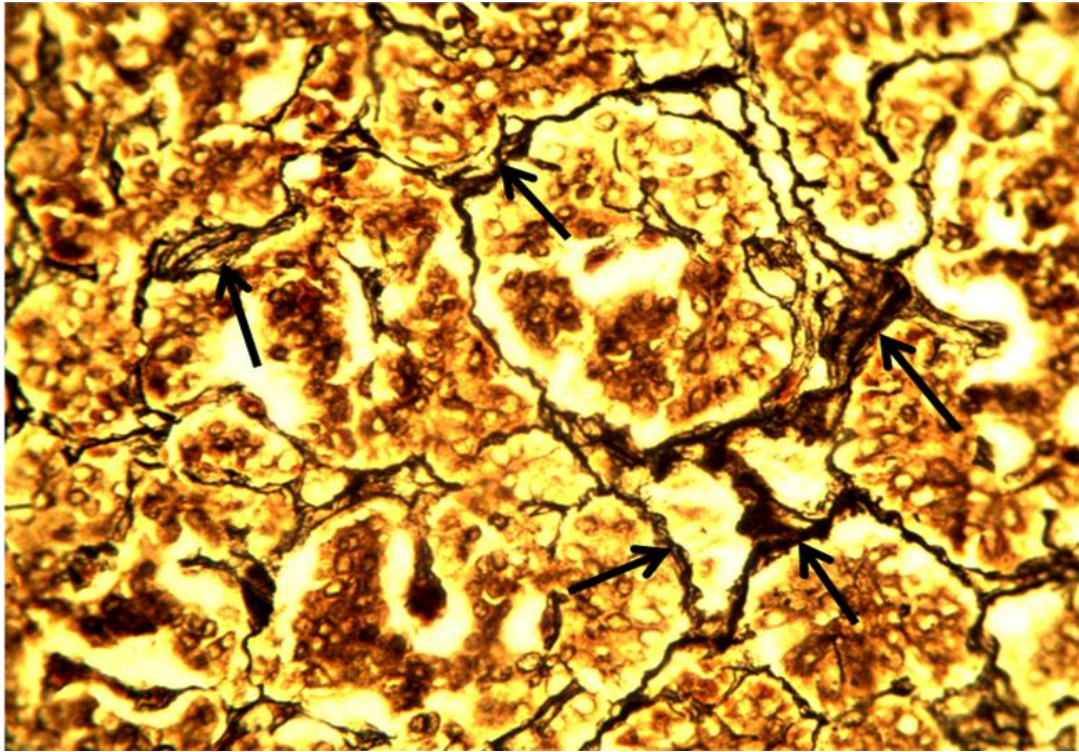
**Fig. 23.** A micrograph of the pancreas of the camel foetus at the third trimester of CVRL 100cm (338 days), At, adipose tissue, A, arterioles, V, vein, SP, connective tissue septa, L, lobules and S, acini. H&E staining, (X100).



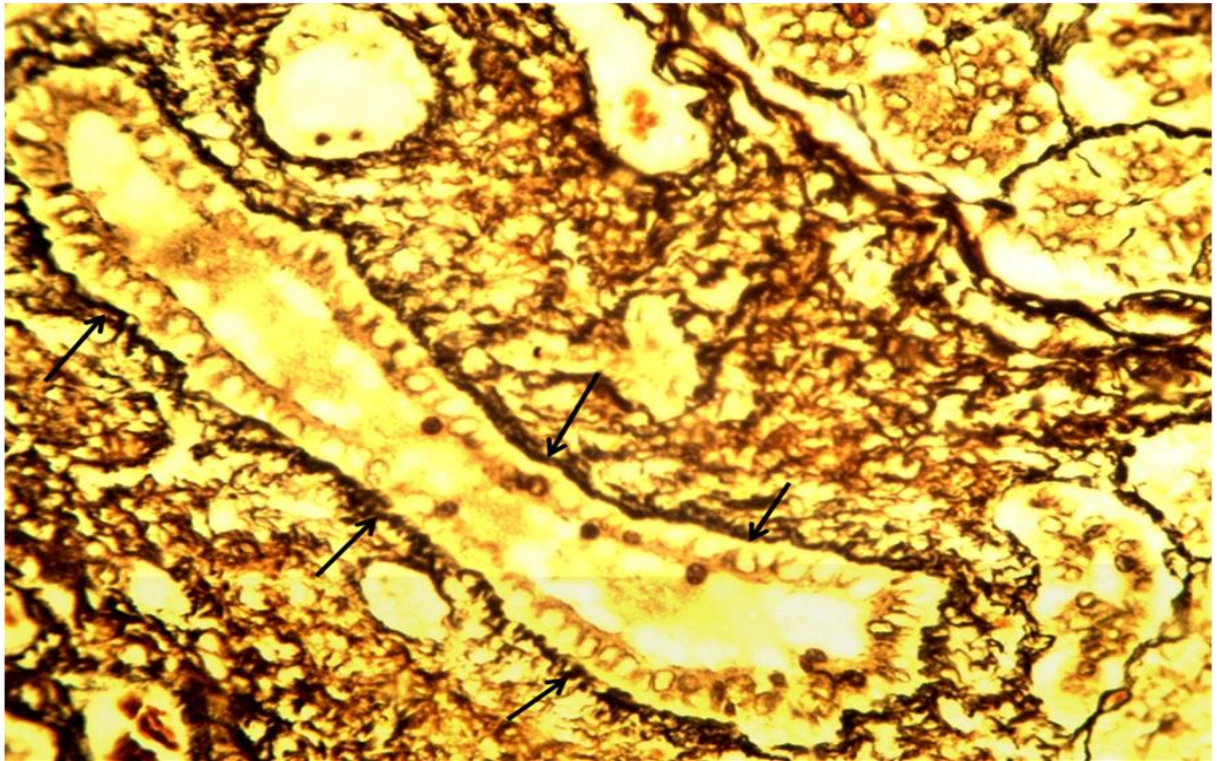
**Fig. 24.** A micrograph showing the pancreas of a camel foetus at the third trimester of CVRL 97cm (330 days). Intralobular duct lined by cuboidal epithelium (thick arrows), S, spherical nuclei, O, oval nuclei. H&E staining, (X400).



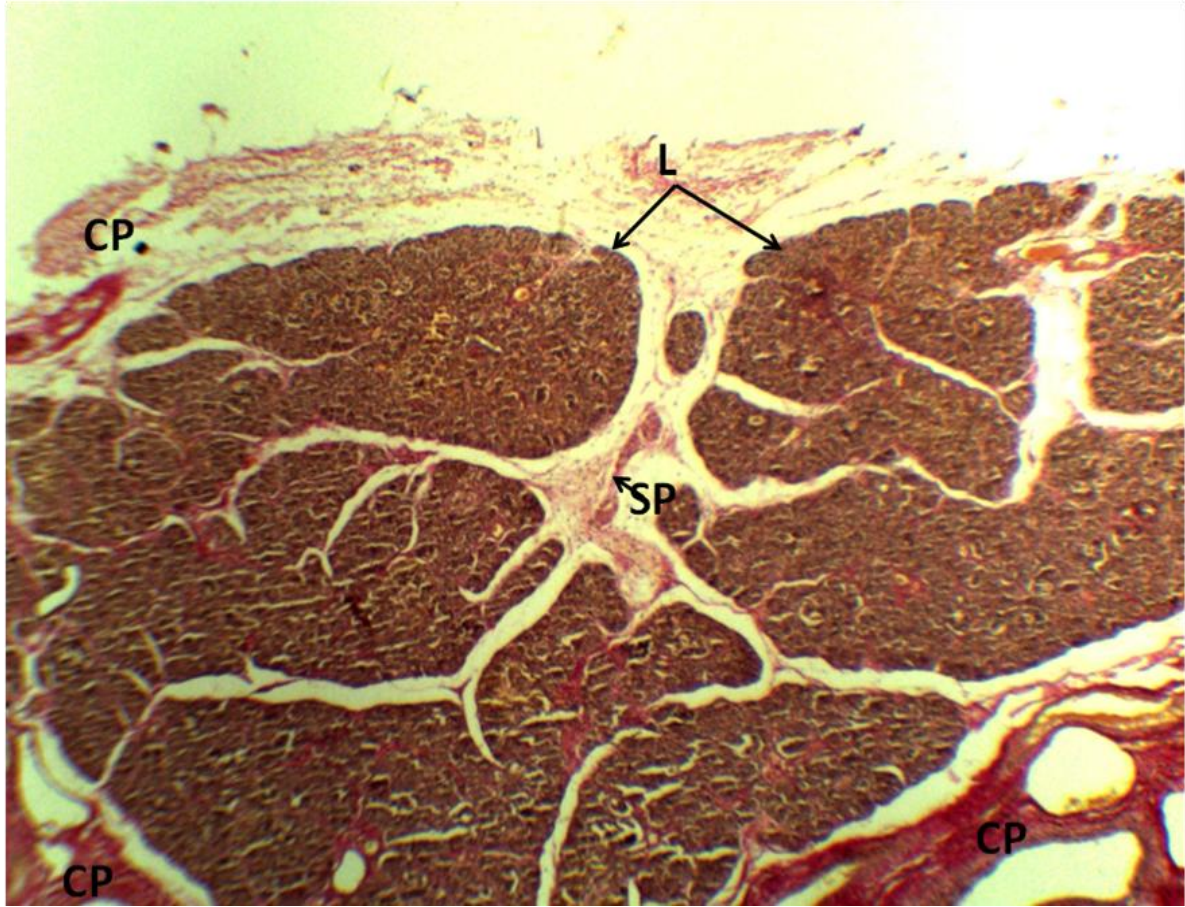
**Fig. 25.** A micrograph of the pancreas of a camel foetus at the third trimester of CVRL 100cm (338 days) showing Interlobular duct lined by columnar cells (arrows) surrounded by thick connective tissue (arrow heads). H&E staining, (X400).



**Fig. 26.** A micrograph of the pancreas of a camel foetus at the third trimester of CVRL 95cm (325 days) showing reticular fibres surrounding the acini (arrows). Silver staining, (X400).

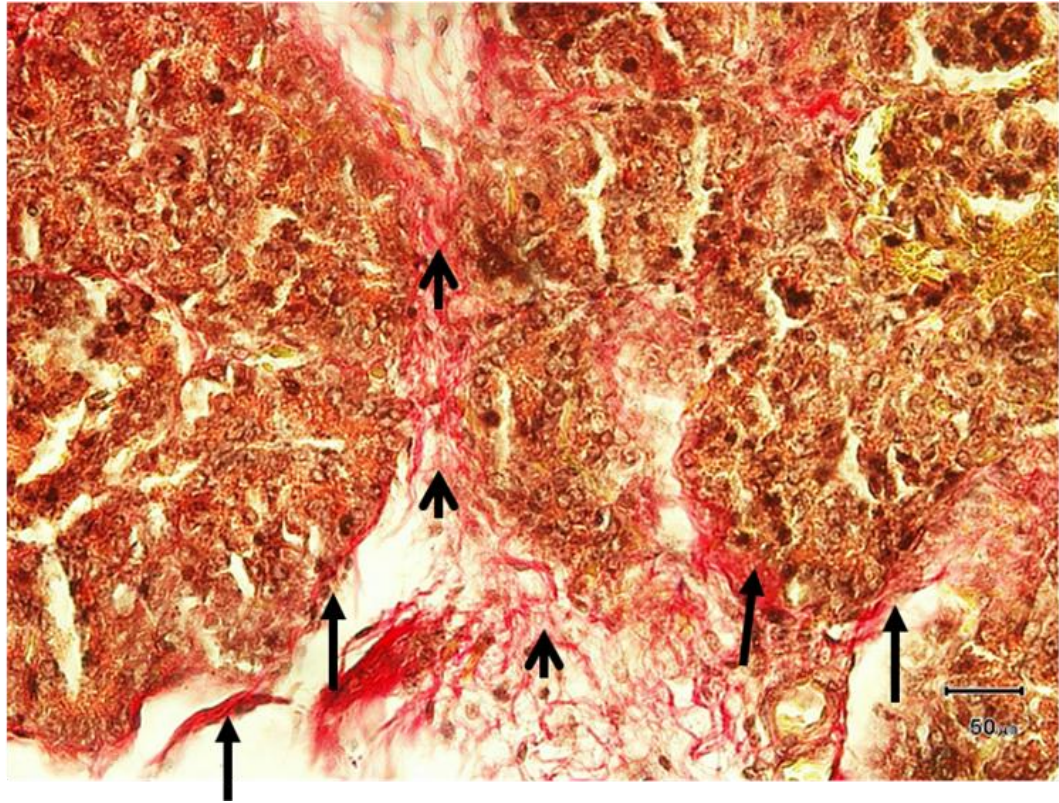


**Fig. 27.** A micrograph of the pancreas of a camel foetus at the third trimester of CVRL 100cm (338 days) showing reticular fibres surrounding the interlobular duct (arrows). Silver staining, (X400).

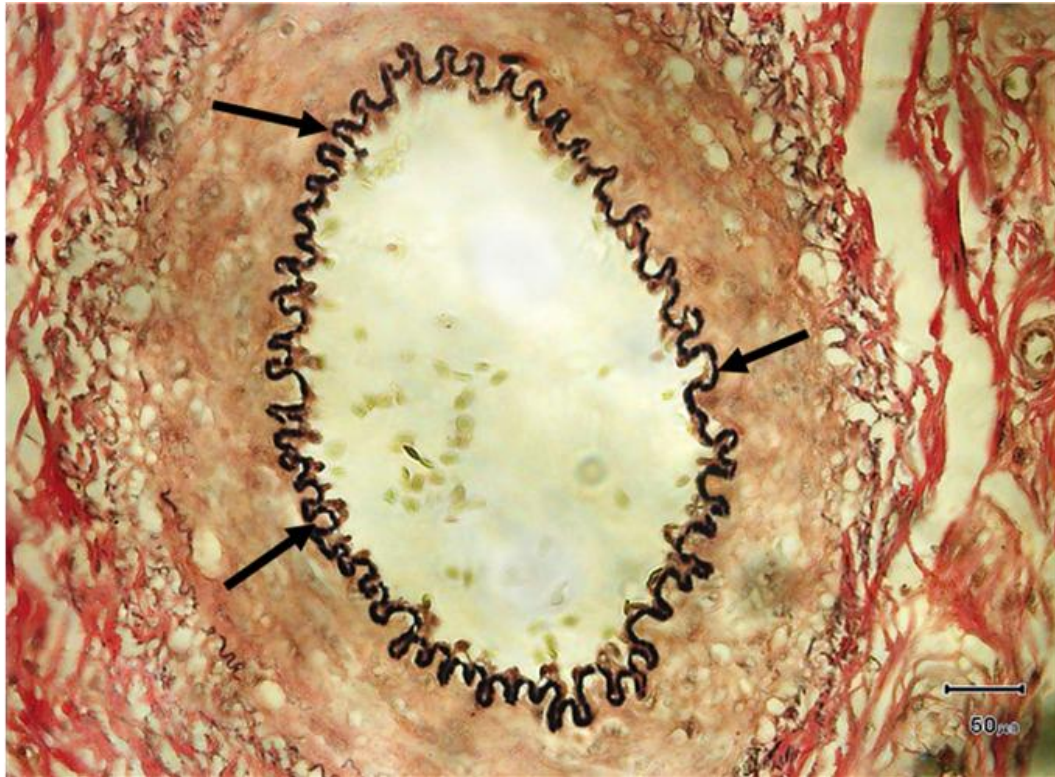


**Fig. 28.** A micrograph of the pancreas of a camel foetus at the third trimester of CVRL 110 cm (366 days) showing the collagenous fibres, L, Lobes, (CP) capsule and (SP) connective tissue septa. Van Gieson staining, (X100).

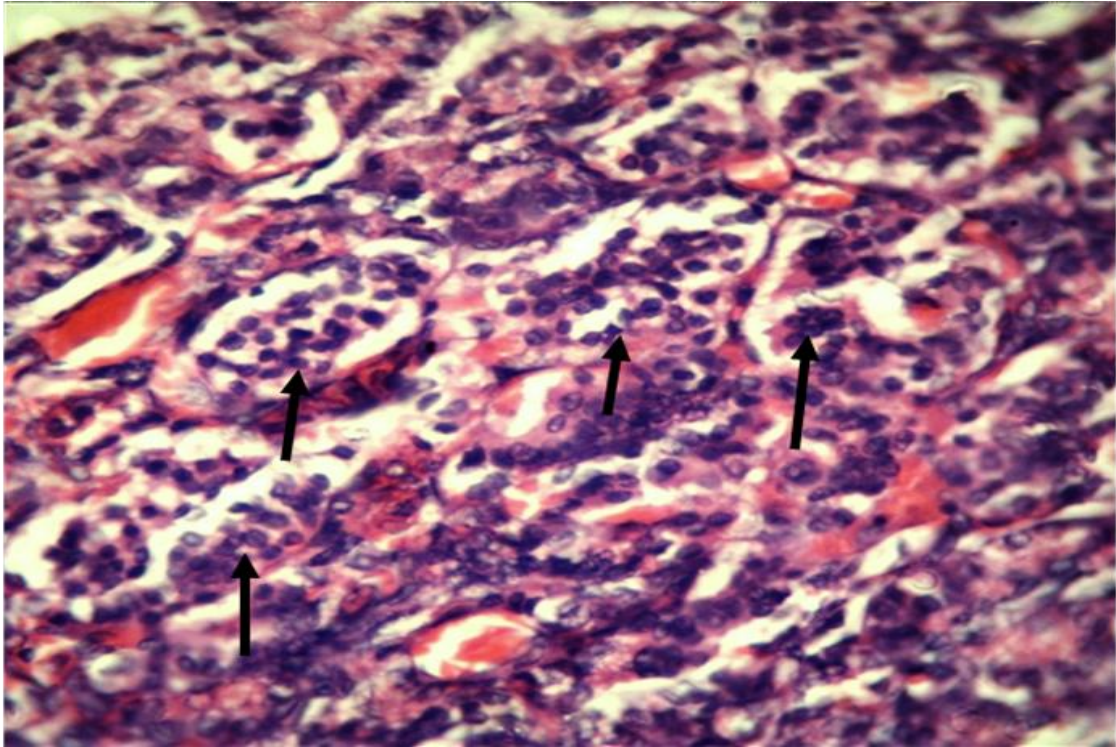




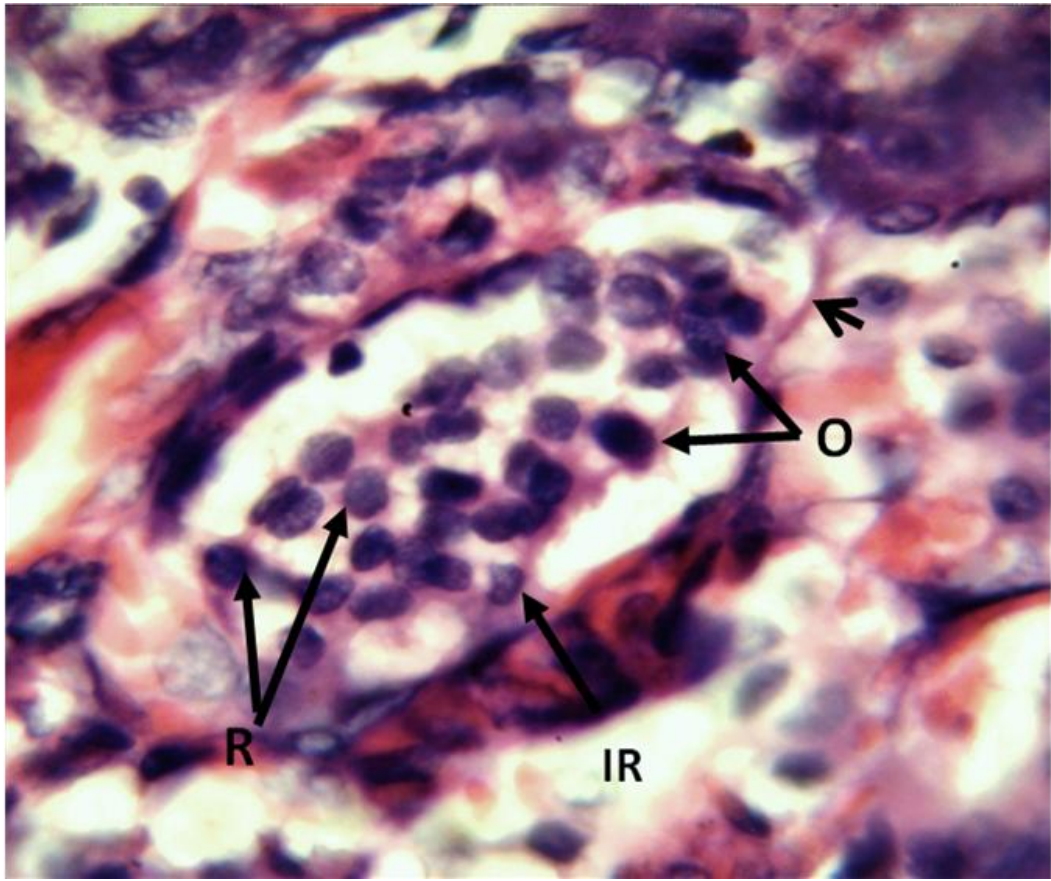
**Fig. 29.** A micrograph of the pancreas of a camel foetus at the third trimester of CVRL 102 cm (344days) showing collagenous fibres surrounding the acini (arrows) and in the connective tissue septa (arrowheads). Van Gieson staining (X400).



**Fig. 30.** A micrograph of the pancreas of a camel foetus at the third trimester of CVRL 105cm (352 days) showing elastic fibres in the wall of a blood vessel (arrows). Ver hoeff staining (X1000).



**Fig. 31.A.** A micrograph of the pancreas of a camel foetus at the third trimester of CVRL 90cm (311 days) showing groups of islets of Langerhans (arrows). H&E.



**Fig. 32. B.** A high magnification of **Fig 31 .A**, O, oval nuclei, R, round nuclei, IR, irregular nuclei, and thin connective tissue surrounding the islet of Langerhans. H&E staining, (X400).

**CHAPTER FOUR**  
**DISCUSSION**

# CHAPTER FOUR

## DISCUSSION

### 4-1: ANATOMICAL STUDIES

It was stated that the pancreas of the camel was greyish pink in colour Sultan (1999) and Masaad (2007). This is also true for the camel foetus in the present study at the second and third trimester however, at the first trimester the pancreas was greyish in colour. In contrast Singh *et al.* (2017) stated that the pancreas in Prenatal Goat was creamy white in colour. The current research indicated that the pancreas of the first, second and third trimesters had no definite shape. A similar observation was mentioned by Taha and Abdel-Magied, (1998), Sultan, (1999) and Masaad (2007) in the adult camel.

The pancreas of all trimesters consisted of a left lobe, a right lobe and a body. But it differed in that the left lobe was longer than the right lobe. This observation was similar to what had already been described by Mustafa *et al.* (1983), Smuts and Bezuidenhout (1987), Taha and Abdel-Magied (1998), Sultan (1999) and Masaad (2007). In the camel, and in the horse (Nickel *et al.*, 1973 and Sisson, 1975).

In the present investigation the pancreas of the first second and third trimesters was situated at level of the first five lumbar vertebrae. This is similar to the findings of Mustafa *et al.*(1983), Sultan (1999), and Masaad (2007) in the adult camel. However, Singh *et al* (2017) stated that the pancreas of goat foetus lied in the abdominal cavity partly on right and partly on left side of median plane. The relationships of the pancreas in the second and third trimesters with the stomach, duodenum, liver, hepatic lymph node, kidneys and the large intestine were generally in agreement with the observation of Bradley (1946) and Sisson (1975) in the horse, Nickel *et al.* (1973) in ruminants, and Bradley (1948) in the dog, Mustafa *et al.* (1983), Smuts and Bezuidenhout (1987), Taha and Abdel-Magied (1998) and Sultan (1999) in the camel.

## **4-2: HISTOLOGICAL STUDIES**

### **4-2-A: The exocrine portion**

In the present investigation the pancreas in the first and second and third trimesters was covered by a connective tissue capsule rich in blood vessels and adipose tissue in third trimester. This was also true in the adult camel and humans (Arey, 1974; Stinson and Calhoun, 1981; Bloom and Faweett, 1986; Sultan 1999, Masaad, 2007).

In the present study the pancreas in second and third trimesters was clearly a tubuloacinar gland. A similar result was previously observed by Masaad (2007) in the adult camel.

The current investigation showed that, the acinar cells of all trimesters had spherical or oval nuclei in the center or near the base of the cell respectively. This is similar to the observation of Stinson and Calhoun (1981) in goats, Mukherjee *et al.* (1986) in sheep, Masaad, (2007), Sultan (1999) in the camel and Dhoolappa *et al.* (2004) in the Indian donkey.

In this research the intralobular duct was lined by cuboidal cells. A similar result was previously observed by Stinson and Calhoun, (1981), Sultan (1999) and Masaad, (2007), in adult camel. However, Gemmel and Heath (1973) had found that the intralobular duct was lined by columnar cells in the sheep.

In the present study the interlobular duct was lined by stratified cuboidal cells in the second trimester whereas in the third trimester it was lined by columnar cells. In contradiction in the adult camel it was lined by cuboidal cells (Masaad, 2007)

Masaad (2007) had reported few elastic fibers in the hepatopancreatic duct of the pancreas in the adult camel. However, in this investigation, the elastic fibers in the pancreas of the foetus were observed only in the wall of the blood vessels.

The current investigation reported that the collagenous fibers were abundant in the pancreas of the camel foetus at the three trimesters. Similar findings were reported in the pancreas of the adult camel (Masaad, 2007).

Masaad (2007) indicated that reticular fibers were only noticed in the basal lamina of the secretary units. In contradiction this study revealed that reticular fibers were numerous and surrounded the acini, ducts and even blood vessels.

#### **4-2-B: The endocrine portion (islets of Langerhans)**

In present study the islets of Langerhans in the second and third trimesters were appeared of different sizes and shapes and were made up of irregular clumps of cells. These findings confirmed those of AL-Ani (1987), Sultan (1999), Masaad (2007) and Zghair (2016) in the adult camel.

### **4-3: MORPHOMETRIC STUDIES**

In this part the morphometric data on the pancreas of the camel foetus were investigated with the view of estimating the volume of the pancreas and the volume densities of its main components namely acini, ducts, islets of Langerhans, connective tissue, and blood vessels. The results of this stereological study of the pancreas of the camel foetus presented in this thesis are the first report regarding morphometry in the camel foetus. Stereological data obtained in this investigation are prerequisite to understand the structure and function correlation. The plots of Weibel *et al* (1963) and Dunnill (1968) were used to insure the sufficiency of the number of points counted for each component studies.

To the best of our knowledge data on the camel foetus pancreas is virtually lacking. Even in other animals the data found in the literature was only estimation of the biometric parameters namely weight, length and width (Singh *et al.*, 2017).

#### **4-3-A: First trimester**

As ducts and islets of Langerhans were not observed in this stage they were excluded from the study. The acini occupied approximately half the volume of the pancreas in this trimester (44.63% which was  $0.71\text{cm}^3$  of the total volume) whereas the connective tissue occupied about (38.82% of the whole pancreas. The blood vessels percentage was to be 16.55% of the total volume of fresh pancreas. No comparative data was available on the camel or other animals. Taga (1998) studied the body mass of the pancreas in the mouse. Numerous studies on the morphometry



of the islets of Langerhans were reported (Dean, 1973; Zharkov, 1996; Yorde and Kalkhoff, 1986; Elayat *et al.*, 1995).

#### **4-3-B: Second trimester**

In this stage the presence of ducts and islets of Langerhans was first observed. Hence, they were included in the parameters (components) studied in this trimester. The acini also occupied half of the volume of the pancreas (54.98% and about 1.48cm<sup>3</sup> absolute volume). An accompanied decrease in the size of the connective was observed (27.70% and absolute volume of 0.75cm<sup>3</sup>). The ducts and islets of Langerhans showed results of 3.30% and 1.82% respectively. The blood vessels exert slight decrease as a result of the development of the other parameters, being 12.20% and absolute volume of 0.32cm<sup>3</sup>. Similarly, no comparative data was available concerning the camel foetus.

#### **4-3-C: Third trimester**

The current research showed that the acini underwent further increase in size and showed a percentage of 60.95% and absolute volume of 2.55cm<sup>3</sup>. The connective tissue presented more or less similar data to the previous trimester showing a sort of consistency, occupying about 26.75% of the volume of the pancreas with absolute volume of 1.12cm<sup>3</sup>. Further increase was shown by islets of Langerhans which occupied 2.52% of the volume of the pancreas giving absolute volume of about 0.11cm<sup>3</sup>. No increase was noticed on the volume of the ducts. This may suggest that the ducts reached their maximum growth at the end of the second trimester trimmer and at beginning of the third trimester. The volume density for the ducts was 2.44% and absolute volume of about 0.1cm<sup>3</sup>. A marked decrease on the percentage of the volume occupied by the blood vessels was noticed. They occupied 7.34% of the volume of the pancreas and showed an absolute volume of 0.3cm<sup>3</sup>.

## CONCLUSION

1. The pancreas was gray in colour in the first trimester but grayish pink in the second and third trimester.
2. The right lobe of the pancreas is shorter than the left lobe.
3. The pancreas of all trimesters was located at the level of the first five lumbar vertebrae
4. The secretory units of the pancreas in all trimesters were tubulo-acinar
5. The islet of Langerhans and ducts first appeared in the second and third trimesters
6. The acini represented the biggest part of the total volume of the pancreas
7. The duct system and islet of Langerhans represented the smallest part of the total volume of the pancreas.

# Recommendations

## **Further studies are needed**

1. To estimate the volume density ( $V_v$ ) of the islet of Langerhans in the body and lobes of foetal pancreas.
2. To Study Immunohistochemistry of the pancreas of camel fetus
3. To Study the ultrastructure of exocrine and endocrine portion of the foetal pancreas.

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