

## Original Article

Investigating the Pathological Classification of Reproductive System Lesions in Slaughtered Female Water Asian Buffalo (*Bubalus bubalis*)Ameer haider Jawad<sup>1\*</sup> , Ahmed Hamid Al-Azzam<sup>2</sup>

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**ABSTRACT**

**Background:** Water buffaloes, crucial in agricultural systems, are valued for their milk and meat production; however, reproductive inefficiencies, including delayed maturity and long calving intervals, impact productivity. Limited data on reproductive tract pathologies in buffaloes, particularly those culled for abnormalities. Accordingly, this study examines these lesions in slaughtered buffaloes, offering insights into reproductive health challenges.

**Objectives:** This study aims to identify, classify, and quantify reproductive tract lesions in female buffaloes slaughtered at the Al-Najaf abattoir, in Iraq, providing data on the prevalence and types of reproductive abnormalities to improve understanding and management of reproductive health in buffalo populations.

**Methods:** Macroscopic and microscopic examinations were performed on the reproductive tracts of the slaughtered buffaloes to identify and classify the lesions. The reproductive organs, including the uterus, oviducts, and ovaries, were carefully examined, and the prevalence of various lesions was recorded. Statistical comparisons were made between different lesion types and locations within the reproductive tract.

**Results:** Macroscopic screening revealed that 40.5% of the buffaloes had reproductive tract lesions. Uterine lesions were the most prevalent, occurring in 26.88% of the cases, with inflammation and abnormal growths being the most common. Microscopic examination identified various types of inflammation, including mucometritis (2.8%), suppurative metritis (3.7%), pyometra (2.3%), and endometritis (5.66%). Abnormal growths, such as endometrial hyperplasia (3.3%), subserosal fibroids (2.35%), and mucosal polyps (1.88%), were also noted. Vascular disorders like endometrial vascular clotting (1.4%) and vascular sclerosis (1.4%) were less common, while congenital anomalies were rare (0.4%). Ovarian lesions were observed in 13.67% of the buffaloes. The most frequent lesion was ovarian cysts (6.6%), followed by large persistent corpus luteum (2.8%) and luteal cysts (1.4%). Oviduct lesions were less frequent, with an incidence of 5.66%. Common oviduct lesions included congestion (2.3%), hydrosalpinx, and adhesions (1.4% each), with double oviduct being the least common lesion.

**Conclusion:** The uterus is the most affected organ by abnormal changes in female buffaloes, with inflammation being the most prevalent type of lesion in macroscopic and microscopic examinations. These findings highlight the need for improved reproductive health management in buffaloes to prevent and treat such conditions effectively.

**Keywords:** Reproductive tract lesions, Buffaloes uterine inflammation, Ovarian cysts, Hydrosalpinx

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## Introduction

**W**ater buffalo is an essential livestock resource and has ecological importance in agricultural systems, providing milk, meat, and workforce. The most important buffalo breeding-related product in Asia is milk (Coizet, 2014). Buffaloes' main issue is low reproductive effectiveness, which is largely caused by late maturity, long calving intervals, and quiet heat (Zicarelli, 2011). The degree to which reproductive wastage diminishes production proficiency is not well documented. For example, there is currently no data on the proportion of buffaloes with reproductive issues at the time of slaughter or the percentage of culling rate due to genital abnormalities (H Lazim et al., 2008). Buffalo farming has been more popular in recent years as a result of the animal's natural ability to succeed in harsh agro-ecological environments and its unique ability to convert low-quality roughage into animal protein (Rao et al., 2009). Domestic buffaloes (*Bubalus bubalis*) are one of 40 animal species used in food and agriculture production around the world. Domestic buffalo species are classified into two sub-types, namely swamp and riverine-type buffaloes, both of which are regarded as dual-purpose animals. Riverine-type buffaloes are kept primarily for milk and secondary for meat (Perera, 2008). Female animals are usually culled and transported to slaughterhouses because they are either uneconomic to keep or have health problems. As a result, abattoirs are a suitable way to search for pathological lesions of the buffalo reproductive organs that are severe enough to result in infertility or sterility (Purohit et al., 2012). The present study examines the extent of reproductive tract abnormalities present in buffaloes slaughtered in Al-Najaf abattoir, Iraq, and determines and classifies the reproductive tract lesions.

## Materials and Methods

### Study animals

The female buffaloes (*B. bubalis*) were picked up from different regions of Al-Najaf Province, Iraq, to Al-Najaf slaughterhouse, for slaughtering. Firstly, the veterinarians' exam aids this animal to ensure their suitability for slaughtering and respect for general health and non-pregnancy. The slaughtering of these buffaloes regarding Islamic law and regulation. A few minutes after slaughtering, the genitalia (including the ovary oviduct and uterus) were obtained to study their morphological and microscopical characteristics.

### Sampling process

From January to June 2021, regular visits, mainly two visits in a week, were paid to the Al-Najaf abattoir. Repetitive postmortem examination of 212 female buffaloes was carried out with particular attention to the detection of the age of female buffaloes and then the macroscopic and microscopic examination of the reproductive system after slaughtering. The lesions when observed in their reproductive system (ovary, oviduct, and uterus) tissue sample measured as (1×1 cm). The reproductive tract system was examined macroscopically to study the morphological characteristics (size, color, consistency). The lesions were documented grossly through imaging by phone camera (iPhone 12 Pro Max) and were transferred in a plastic container that contained formalin 10%, to the Tissue Processing Unit in the Faculty of Veterinary Medicine, University of Kufa, Iraq. The obtained specimens were used for routine hematoxylin and eosin stains to study the microscopic changes in the female reproductive tract system tissue.

### Routine histopathological technique

The specimens were washed under the running of tap water to eliminate most of the formalin used as fixative the flowing steps were flowed to prepare the histological section of the female reproductive tract depending on (Bancroft & Gamble, 2008).

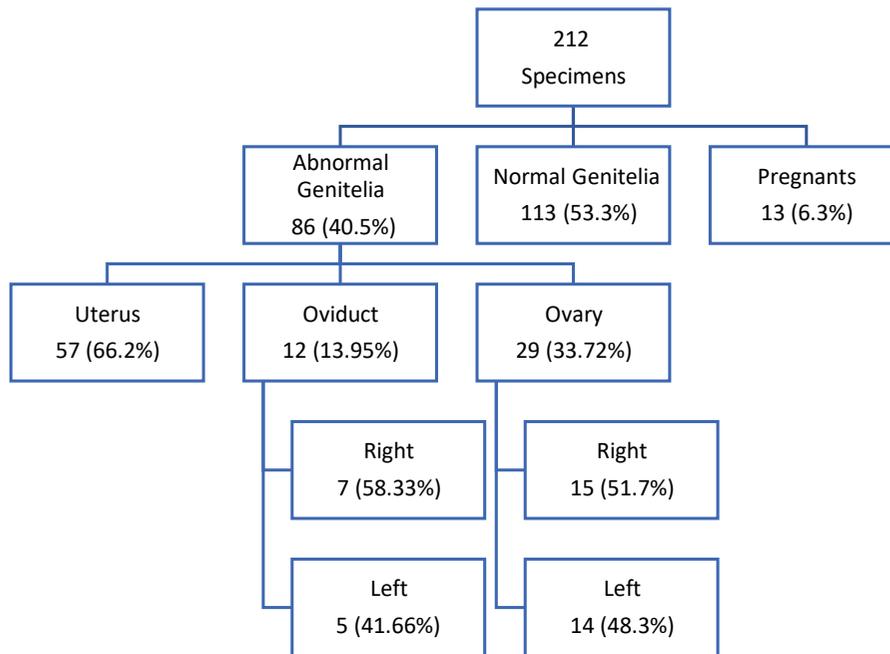
### Study categorization

The gross lesions were scientifically described and classified according to the age group (2-3 years, 3-4 years, 4-5 years, and >5 years). The microscopic description of lesions was carried out and classified into main pathological changes (inflammation, vascular disorders, congenital anomalies, and abnormal growth); then, these lesions were distributed according to the age group. The comparison between lesions appeared microscopically in the left and right ovaries and the oviduct took place.

## Results

### Macroscopic study

Macroscopic evaluation of genitalia of slaughtered female buffaloes in Al-Najaf slaughterhouse revealed that 40.5% was abnormal due to involving lesions in the uterus, ovary, and/or oviduct; some lesions appeared in more than one part of the genitalia. The result also revealed the distribution of lesions among the different parts of the genital tract as 66.2% in the uterus, 33.72% in the



**Figure 1.** Incidence of pathological conditions in genital tract of female buffaloes in total examined animals (n=212)

Notes: Total examined animals=212

ovary, and 13.95% in the oviduct (Figure 1 and Table 1). The results also included the comparison between the numbers and percentages of the lesions in right and left ovaries and oviducts which show no significant differences between them.

Table 2 also revealed the comparison between the occurrences of the lesion in different parts of the genital with the age factor the result illustrated that 4-5 years and 5 years old buffaloes were the most affected age group.

**Gross lesions description**

This pathological survey revealed various reproductive system lesions. The lesions observed through this study were as follows.

**Macerated fetus**

Macerated fetuses found in the uterus of slaughtered buffalo with foul-smelling combined with serous fluid (Figure 2).

**Uterine fibroid**

Small white nodular lesions with firm consistency appeared on the wall of the uterus of more than 5-year-old female buffalo (Figure 3).

**Uterus didelphys**

Uterus didelphys is a congenital anatomical defect of the female genital tract characterized by the presence of a completely separated uterine pocket (Figure 4).

**Table 1.** Percentage of particular lesions in genitalia of slaughtered buffaloes / total female pathological lesions (n=86)

Parts of the Genital Tract	No.		%
	Abnormal Specimens*	Particular Lesions of Total Slaughtered Buffalo (n=212)	
Uterus	57	26.88	66.2
Oviduct	12	5.66	13.95
Ovary	29	13.67	33.7

\*Some specimens showed mixed lesions from different parts of the reproductive system at the same time.

**Table 2.** Gross incidence of abnormalities in different parts of the reproductive system for each age group (n=86)

Parts of the Genital System	No./No. (%)				Total Number of Abnormal Lesions
	Age Group				
	2-3 (y)	3-4 (y)	4-5 (y)	>5 (y)	
Uterus	5	11	14	27	57
Oviduct	1	2	3	6	12
Ovary	2	5	9	13	29
Total	8(8.1)	18(18.3)	26(26.5)	46(46.9)	98(100)*

\*Some specimens showed mixed lesions from different parts of the reproductive system at the same time.

### Pyometra

A post-mortem examination of yellowish creamy purulent exudate fills the lumen of the uterus, an irregularly thickened congested wall is also noted (Figure 5).

### Macroscopic examination of oviduct

Table 3 revealed the statistics of lesions recognized in the oviduct by macroscopic examination. The highest number and the percentage was congestion 2.3%. The hydrosalpinx and adhesions form the second lesion recognized in the examined oviduct represented by 3(1.4%). The double oviduct also forms a considerable lesion during the examination of the oviduct representing 1(0.4%).

The grossest lesions that appeared in the oviduct were congestion, and wall adhesion with surrounding struc-

tures and hydrosalpinx, while the least prominent gross lesion was double oviduct.

### Microscopic study

#### Microscopic examination of the uterus

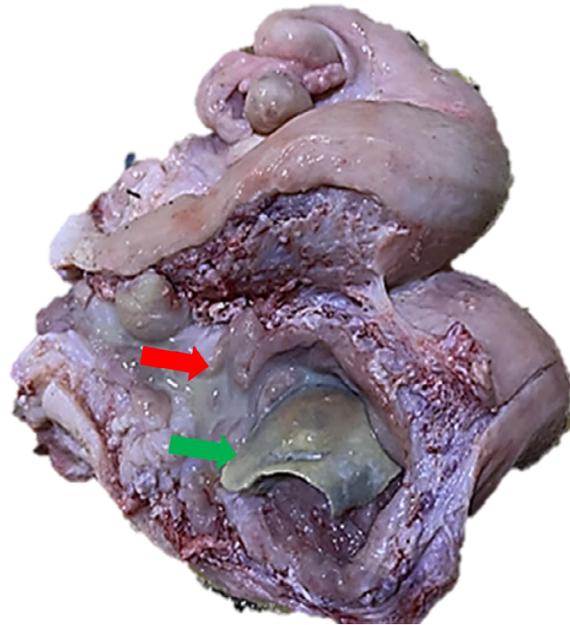
The most common lesions were inflammation and abnormal growths while the vascular disorder, degeneration, and congenital anomalies were the least common prevalent lesions.

Table 4 revealed the statistics of lesions recognized by microscopic examination the highest number and perseverance was a different type of inflammation (mucometritis, suppurative metritis, pyometra, endometritis) which formed 6(2.8%), 8(3.7%), 5(2.3%), 12(5.66%) respectively. The abnormal growth formed the second

**Table 3.** Revealed different lesions that observed during macroscopic examination in right and left oviducts with their numbers and percentages with the age of affected animals (No. of abnormal oviducts=12)

Affection	No.	Age Group								Oviducts Affections\Total Specimen (n=212) (%)
		2-3 (y)		3-4 (y)		4-5 (y)		>5 (y)		
		Right	Left	Right	Left	Right	Left	Right	Left	
Congestion	5	1	0	0	0	0	1	1	2	2.3
Hydrosalpinx	3	0	0	0	1	1	0	1	0	1.4
0 oviduct	1	0	0	1	0	0	0	0	0	0.4
Adhesions	3	0	0	0	0	1	0	1	1	1.4
Total (No. [%])	12(13.95)*	1(8.3)**	0(0)**	1(8.3)**	1(8.33)**	2(16.6)**	1(8.3)**	3(25)**	3(25)**	5.5***

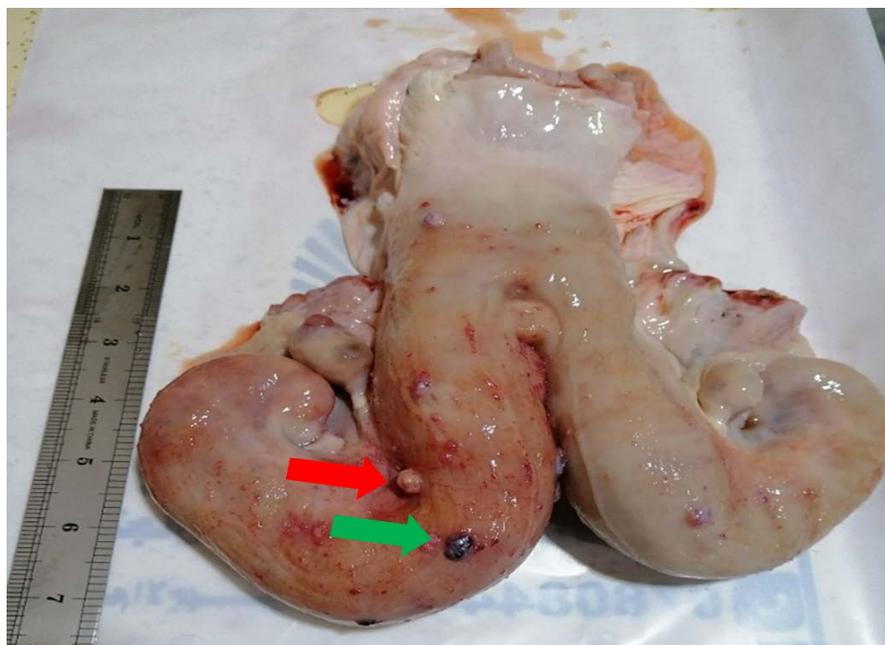
\*Percentage of oviduct lesions /total no. of abnormal genitalia (n=86), \*\*Percentage of oviduct lesion on each side /total no. of the lesion in the oviduct (n=12), \*\*\*Percentage of oviduct lesions/total no. of collected specimens (n=212).



**Figure 2.** Photograph of genitalia 4-5 years female buffalo showing maccratedfoetus, the remaining skull (green arrow) with serous inflammatory fluid (red arrow)

lesion recognized in the examined uterus represented by 7(3.3%) as endometrial hyperplasia 5(2.35%) as subserosal fibroid for 4(1.88%) as a mucosal polyp. The vascular disorder also forms a considerable lesion in examined uteri represented by endometrial vascular clots (1.4%) and vascular sclerosis (1[4%]). The lowest

lesion was identified by microscopic examination with congenital anomalies (0.4%).



**Figure 3.** Photograph of genitalia >5 years female buffaloes showing uterine subserosal fibroid (red arrow) and nodular-like structure in uterine wall (green arrow)



**Figure 4.** Photograph of genitalia of 2-3 years female buffalo showing uterine didelphys

### Degeneration

An endothelial cell that lines the uterus of buffaloes shows vacuolar degeneration formation of nonlipid vacuoles in the cytoplasm, most frequently due to accumulation of water by cloudy swelling (Figure 6).

### Discussion

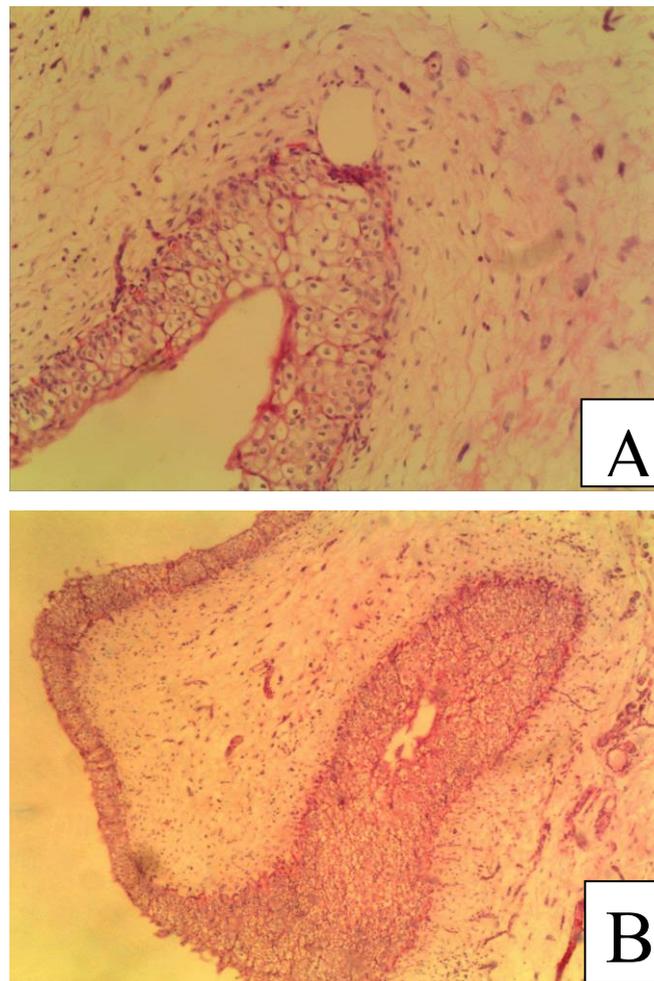
This study investigated the incidence of reproductive system lesions in slaughtered female buffaloes in the Al-Najaf abattoir. Such studies are critical for epidemiological surveys of buffaloes' genital diseases (Ponraj et al., 2017).

The present results indicated the existence of specific and nonspecific reproductive system affections in slaughtered buffaloes. Some of these are economically important, such as ovarian cysts and pyometra. The incidence of various pathological conditions in the reproductive system in our study is considered high. This may be caused by bacterial viral fungal infections as well as the structural and histological properties of the reproductive system (Azawi et al., 2008). Our study results showed a higher prevalence of reproductive system lesions (40.56%) were compared with other studies, such as those mentioned by Azawi and Ali, 2011 (53.30%) and consistent with a study by Saxena et al., 2006 which mentioned that the incidence as 41.8%.

### Uterine lesions



**Figure 5.** Photograph of genitalia of 3-4 years female buffalo showing purulent yellowish creamy exudate in the lumen of uterus (green arrow) with congestion of the wall



**Figure 6.** Microscopic section uterus of 1-2 years old slaughtered buffalo revealed to vacuolar degeneration of endothelial characterized by epithelial cell clear cytoplasm with peripheral nuclei (H&E stain, X400, X100)

The uterine lesions were classified into inflammatory conditions, abnormal growths, vascular disorders, degeneration change, and congenital anomalies. In this study, the incidence of the percentage of pathological conditions in the uterus to whole abnormal collected specimens was 26.88%. It is lesser than the results registered by [Modi et al., 2011](#) in India which reported 49.08%, while [Gautam et al., 2021](#) in Agra mentioned a lower percentage (17.02%).

The results of our study identified 14.62% inflammatory reactions which form the highest incidence among other uterine lesions in contrast to [Hamouda et al., 2020](#) which mentioned that the incidence of inflammation in the uterus constitutes (8.5%). Coincidentally, [Gartma et al., 2006](#) revealed that the incidence of uterine inflammation is 19.5%.

The inflammatory type that was reported in our study includes mucoendometritis (2.8%), suppurative metritis

(3.7%), pyometra (2.3%), and endometritis (%5.66). The increasing incidence of different inflammatory reactions is primarily caused by the decrease in healthcare breed diversity, management, geographic environment, level of nutrition, age advancing as well and immunological status in dairy animals uterine infections are causing sub-fertility in chronic cases and infertility in acute cases ([Sheldon et al., 2009](#)).

Retained placenta, manipulation to fix dystocia, or genital prolapse may cause subsequent inhabitation of the genital tract from healing and involution, thereby delaying the post-calving anestrus. Additionally, the retained tissues in the lumen become a favorable environment for bacterial growth. Therefore, it reduces the uterine immunity by impairing the function of uterine ([Kimura et al., 2002](#)); that is, inequality of this condition that instructs to variety in nutrition, breeds, and administration ([Lazim et al., 2008](#)). Moreover, traditional practice considers one of the influences that lead to endometritis, which is

**Table 4.** Revealed different lesions that were observed during microscopic examination in the uterus with their numbers and percentages with the age of affected animals (No. of abnormal uterus=57)

Microscopic Abnormalities	NO.	Age Group				Uteri Affections\Total Specimen (n=212) (%)	Category of Abnormal Lesion
		2-3 (y)	3-4 (y)	4-5 (y)	>5 (y)		
Mucomitritis	6	0	2	1	3	2.83	Inflammation
Suppurative metritis	8	1	1	2	4	3.77	Inflammation
Pyometra	5	0	1	2	2	2.35	Inflammation
Endometritis	12	2	2	3	5	5.66	Inflammation
Endometrial blood vessels clot	3	0	0	1	2	1.41	Vascular disorders
Sclerosis of myometrial blood vessels	3	0	1	1	1	1.41	Vascular disorders
Endometrial glandular hyperplasia	7	1	1	2	3	3.3	Abnormal growths
Polyps	4	0	1	1	2	1.88	Abnormal growths
Fibroid	5	0	1	1	3	2.35	Abnormal growths
Didelphys uterus	1	0	0	0	1	0.47	Congenital anomalies
Vacuolar degeneration	3	1	1	0	1	1.41	Degeneration
Total	57	5 (8.77)	11(19.29)	14(24.56)	27(47.36)	(26.8)	

that several farmers use non-sterile foreign tools to enter the vagina of buffaloes for induced the letdown of milk through the postpartum period. Finally, that tradition has increased the opportunity to expose the uterine of water buffaloes to different infections (Azawi et al., 2008).

Abnormal growths were reported in female buffaloes' genital system at 7.54%. This was higher than the previous findings by Saxena et al., 2006 in different slaughterhouses in Rajasthan (India); they reported the number as 0.78%. The abnormal growths diagnosed in our study include endometrial glandular hyperplasia (3.3%), polyps (1.2%), and fibroid (2.35%).

This study also showed vascular disorders in some of the genital tract specimens (2.83%) which include endometrial blood vessel clotting (1.41%) and sclerosis of myometrial blood vessels (1.41%). Vascular disorders can vary, depending on the relevant disease. Age is considered the most common risk factor which affects the heart and blood vessels.

The rate of cell degeneration (vacuolar degeneration) was 5.26% in the current study and appeared as large clear vacuoles within the cytoplasm, It is caused by cellular hypoxia, which affects the sodium-potassium mem-

brane pump and is the first sign of cell injury (Timbrell, 2009).

Congenital anomalies in the buffalo reproductive tract were 1.75% compared with Azawi and Ali, 2015 which was 1.13%. In this study, we reported one case of didelphys uterus in a >5-year-old female buffaloes (Iqbal et al., 2017) which was also reported didelphys uterus in Sahala slaughterhouse Islamabad as a case report. Animals having uterus didelphys defects are reported to have normal pregnancy and calving (Roberts, 1986) but some animals may suffer from dystocia due to fetal limbs entering each cervical canal (Noakes et al., 2009). Because of the possible genetic nature of the defect, such animals should not be used for breeding after diagnosis and must be culled.

### Ovary lesions

The pathological conditions of the ovary are classified into persistent graffian follicle, ovarian cyst, persistent corpus luteum, and luteal cyst. In this study, we report the incidence of pathological conditions in the ovary in slaughtered animals was (13.67%). Comparing with Azawi and Ali, 2015 in Mosul slaughterhouse, they reported (16.79%) and Gautam et al., 2021 in Agra, they reported 8%. The frequency of ovarian abnormalities

in the left ovary was 48.27% and the right ovary was 51.72%. Ovary abnormalities in this study including the ovarian cyst (6.6%) form the highest prevalence among other ovarian lesions which are collectively lesser than that recorded by (Azawi & Ali, 2015) in Iraq, they reported 8.8%.

The persistent graffain follicles were identified depending on morphological characteristics reported as 2.8% while Alwan et al., 2001 reported an incidence percentage of 1.5%.

The frequency of persistent corpus luteum was 2.8% which forms a high value when compared with that mentioned by Saxena et al., 2006; they reported 1.3% in different slaughterhouses in Rajasthan (India).

The luteal cyst at a rate of 1.4% while a study (Azawi & Ali, 2015) in Mosul slaughterhouse reported 0.2%. Occasionally the corpus luteum does not regress routinely even if the female is not pregnant. This is considered persistent corpus lutein. The persistent corpus lutein continues to create progesterone to prevent ovulation, follicular development, and estrus (Lashari & Tasawar, 2012)

### Oviducts lesions

In this study, the incidence of pathological conditions in the oviduct was 5.66% while Ananda Raja, 2003 and Gautam et al., 2021 reported 9.56% and 1.4%, respectively. The affections include unilateral abnormalities that result in infertility, while bilateral leads to sterility. Oviduct affections have been identified mainly from slaughterhouse research. Salpingitis is the most incidence. The frequency of oviduct abnormalities in the left oviduct was 41.66% and the right oviduct was 58.33%. The macroscopic pathological conditions of the oviduct include congestion, and hydrosalpinx (1.41%) which is consistent with mentioned by Hassanin et al., 2010 in Egypt who reported 1.47% double oviduct incidence was 0.47%, while Adhesions was 1.4%. This result is consistent with that mentioned by Azawi and Ali, 2015 and Durgesh et al., 2010. A higher occurrence of oviduct abnormalities has been reported for buffaloes species compared with cattle (Iannuzzi et al., 2005). Salinities may occur after uterine washing with solutions of strong antiseptic which may leak into the fallopian tube. A downward infection agent from the peritoneum causes inflammation in the oviduct. The exact etiology of hydrosalpinx continues to be poorly known although an extension of inflammatory exudates from the uterus is considered one possible reason (Lazim et al., 2009), and congenital serous secretions as another possible eti-

ology (Ellington & Schlafer, 1993). In a recent study of buffalo genitalia, the congenital abnormalities included accessory oviducts and aplasia. Mainly occur segmental uterotubal junctions (Shivhare et al., 2012) Adhesions of the oviducts have been recorded in studies on the buffalo state and illustrated the result which presents the inflammatory nearby to the ovarian duct. Adherence to the ovarian duct could grow to the adjacent bursa because of continuous infection or resulting rough rectal manipulations.

## Ethical Considerations

### Compliance with ethical guidelines

This study was approved by the Ethics Committee of the University of Kufa, Kufa, Iraq (D12938).

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### Authors' contributions

All authors contributed equally to the conception and design of the study, data collection and analysis, interpretation of the results, and drafting of the manuscript. Each author approved the final version of the manuscript for submission.

### Conflict of interest

The authors declared no conflict of interest.

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