ABSTRACT.- Silva R.M.M., Macêdo J.T.S.A., Lacerda M.S.C., Azevedo J.P.M.V.B., Ferreira Júnior J.A., Cerqueira R.B. & Pedroso P.M.O. 2020. Lesions of the sheep reproductive system found in a slaughterhouse in Bahia state, Brazil. Pesquisa Veterinária Brasileira 40(12):955-962. Laboratório de Patologia Veterinária, Fundação Universidade de Brasília, Campus Universitário Darcy Ribeiro, Via L4 Norte s/n, Brasília, DF 70910-970, Brazil. E-mail: pedrosovet@yahoo.com.br

The present study aimed to evaluate and diagnose pathologies of the reproductive system of slaughtered sheep in slaughterhouses in the state of Bahia since there are few data on the subject as well as it is important to note the diagnosis of these diseases to apply appropriate control and prophylaxis measures. The research was carried out in slaughterhouses in the state of Bahia according to the “Serviço de Inspeção Federal” and “Serviço de Inspeção Estadual” for sheep slaughter. From July 2018 to February 2019, visits and monitoring of 1,072 slaughtered sheep were carried out. The animals came from 22 municipalities in the state of Bahia, aged from six to 18 months. During slaughter, the sheep reproductive systems were sectioned for evisceration and lesion collection. Additionally, epidemiological surveys related to origin, age, and race were obtained. For bacteriological examination, collections were performed with sterile scalpel slides and swabs in Stuart medium sterile tubes and refrigerated in a thermal box. For histopathological analysis, fragments were fixed in 10% formaldehyde and routinely processed for histology, stained with Hematoxylin and Eosin (HE). A study of the sheep’s main pathological changes in slaughterhouses in the state of Bahia was carried out, in which 1,072 genital systems were evaluated, and 211 alterations were identified. The most affected reproductive organs were the ovaries (65.3%), followed by the uterus (29.4%) and uterine tubes (5.3%). In the ovaries, the most frequent lesion was a follicular cyst (34.1%); in the fallopian tubes, the cysts represented 3.3% of the lesions, and in the womb, endometritis was observed in 9% of the animals. Other pathologies identified in the ovaries were: luteinized cyst (2.3%); cystic granulosa cell tumor (0.5%); benign lesion (0.5%); agenesis unilateral (0.5%), in addition to other changes of little clinical significance, such as corpus luteum cysts (11.8%) and paraovarian cysts (15.6%). In the fallopian tubes, hydrosalpinx was observed (1%), as well as adenoma (0.5%), agenesis (0.5%), and cysts (3.3%). Uterine lesions included hydrometra (2.3%); Cystic Endometrial Hyperplasia (CEH), abscess, and pyometra (1.4% each); adenomyosis, womb sera petechiae, and total segmental aplasia (1% each); two pregnant uterus, in early pregnancy, presented pyometra (0.9%), however, no change was observed in the fetuses; and uterine polyp (0.5%). Ectopic pregnancy with fetal maceration (0.5%) was observed; vaginitis occurred in 0.5% of the animals, and endometrial melanosis in 8.5%. The high incidence of follicular cysts and endometritis are characterized as diseases that reduce the reproductive efficiency in herds, causing infertility and economic losses in production.

INDEX TERMS: Sheep, reproductive system, slaughterhouse, Brazil, ovaries, pathology, follicular cysts, endometritis.
RESUMO.- [Lesões do sistema reprodutor em ovelhas de abatedouros frigoríficos no estado da Bahia.] O objetivo do presente trabalho foi avaliar e diagnosticar alterações do sistema reprodutor de ovelhas abatidas em abatedouros frigoríficos no estado da Bahia. A pesquisa foi realizada em abatedouros frigoríficos do estado da Bahia com Serviços de Inspeção Federal e Estadual no abate de ovinos. No período de julho de 2018 a fevereiro de 2019 foram realizadas visitas e acompanhamento do abate de 1.072 ovelhas. Os animais eram provenientes de 22 municípios do estado da Bahia, com idade entre 6 a 18 meses. Durante o abate, na evisceração foram seccionados os sistemas reprodutores das ovelhas para avaliação e coleta das lesões. Adicionalmente foram obtidos inquéritos epidemiológicos relacionados à procedência, idade e raça. Para o exame bacteriológico, as coletas foram realizadas com lâminas de bisturi estéreis e swabs em tubos estéreis com meio Stuart e refrigeradas em caixa térmica. Para análise histopatológica, os fragmentos foram fixados em formol a 10% e processados de forma rotineira para histologia e corados pela hematoxilina e eosina (HE). Entre 1.072 sistemas genitais avaliados, identificou-se 211 alterações. Os órgãos do sistema reprodutor mais acometidos foram os ovários (65,3%), útero (29,4%) e tubas uterinas (5,3%). Nos ovários, a lesão mais frequente foi o cisto folicular (34,1%); no útero a endometrite (9%) e nas tubas uterinas, os cistos representaram (3,3%). Outras lesões identificadas nos ovários foram: cisto luteinizado (2,3%); tumor de células da granulosa (0,5%); adenoma (0,5%); agenesia unilateral (0,5%), além de outras alterações de pouco significado clínico, como cistos parovarianos (15,6%) e corpo lúteo cístico (11,8%). Nas tubas uterinas observou-se, além dos cistos tubo-ovarianos (3,3%), hidrossalpinge (1%), adenoma (0,5%) e agenesia (0,5%). As lesões uterinas foram endometrite (9%), hidrometra (2,3%); hiperplasia endometrial cística, abscesso e piometra (1,4% cada); adenomiose, petéquias na serosa do útero e aplasia segmentar (1% cada); dois úteros grávidos, em início de gestação, apresentaram piometra (0,9%), porém os fetos não apresentaram alterações; e pôliputo uterino (0,5%). Observou-se uma gestação ectópica com maceração fetal (0,5%); a vaginite ocorreu em 0,5%, e melanosodermia (0,5%). Destaca-se a elevada incidência de cistos foliculares e endometrite que são doenças que reduzem a eficiência reprodutiva dos rebanhos, provocando infertilidade e perdas econômicas na produção.

TERMOS DE INDEXAÇÃO: Sistema reprodutor, ovelhas, abatedouro, frigorífico, Brasil, ovários, patologia, cistos foliculares, endometrite, ovinos.

INTRODUCTION

Brazil has the largest flock of sheep in Latin America, with approximately 18.9 million head, with the Northeast region and the state of Bahia occupying both the 1st place (IBGE 2018). In Brazil, sheep farming is extensive, with low technological investment and reduced zootecchnical indicators, formed mainly by mixed or native sheep (Santos 2001, Osório & Rassi 2011). However, the exploration of small ruminants is a growing activity and important for the Northeast region, mainly due to its adaptability to the arid and semi-arid regions of the country, characterized by long periods of drought, high temperatures, and shallow soils that are difficult to drain (Santos 2001, Leite & Simplício 2005, Holanda Júnior & Martins 2007, Monte 2008). It is essential to highlight that some studies have been carried out in slaughter sheep in Brazil to diagnose the main parasitic and infectious lesions, demonstrating that these studies are also important to reduce errors in slaughter condemnations in animals and guarantee the quality of products for consumers (Souza et al. 2011, Higino et al. 2013, Panzieri et al. 2018). Reproductive efficiency is one of the most critical factors that promote productivity of sheep farming and economic viability, being essential for developing the herd. However, injuries to the reproductive system have significant limitations, which causes a drop in production (Agrawal 2010, Carvalho Júnior et al. 2010). Thus, diagnosis is necessary to prevent and control the main diseases that affect the sheep reproductive system.

The objective of this study was to evaluate the main changes found in the reproductive system of slaughtered sheep in slaughterhouses in the state of Bahia to identify the main injuries that cause losses in sheep farming and facilitate the development of prevention and control measures.

MATERIALS AND METHODS

The research was carried out in three slaughterhouses for sheep and goats in the state of Bahia according to the “Serviço de Inspeção Federal” and “Serviço de Inspeção Estadual”. In the period from July 2018 to February 2019, 1,072 emaciated sheep were slaughtered. During slaughter, the evisceração (removal of internal organs) begins, i.e., the sheep reproductive system was sectioned to evaluate and collect material for anatopathological and bacteriological examinations. In addition, epidemiological information related to the origin, age, and race of the animals was obtained. The changes were grouped into six categories based on macroscopic and microscopic descriptions: inflammatory, degenerative and proliferative changes, developmental abnormalities, and others.

Reproductive systems with lesions were analyzed macroscopically; then fragments were collected and fixed in 10% buffered formaldehyde, cleaved, processed routinely for histology, and stained with Hematoxylin and Eosin (HE). For the bacteriological examination, collections were performed with sterile scalpel slides and swabs in sterile tubes with Stuart medium and refrigerated in a thermal box. In addition, they were sent to the laboratory for isolation and identification of the etiological agent. The culture media used for bacterial identification were Rugai medium, citrate, malonate, and Primary Seeding of uterine secretion were Blood agar, chocolate and Agar Brain Heart Infusion (BHI) Agar, NaCl, and mannitol.

The statistics were based on descriptive analysis as a percentage of the frequency of macroscopic and histopathological changes in the results obtained.

RESULTS

During the studied period, 196 (18.3%) samples of sheep with alterations in the reproductive system were analyzed, with 2.2% of the animals showing concomitant lesions; the total number of injuries corresponded to 19.6% (211/1,072). The animals were consisted of Dorper, Santa Inês, Mestiço, and no defined breeds, aged from six to 18 months and came from 22 municipalities in the state of Bahia, mainly in the North Central region.
Lesions of the sheep reproductive system found in a slaughterhouse in the state of Bahia, Brazil

The most affected organs of the reproductive system were the ovaries 65.3% (138/211), followed by the uterus 29.4% (62/211) and fallopian tubes 5.3% (11/211). In the ovaries, the most evident lesion was the follicular cyst 34.1% (72/211); in utero, endometritis with 9.0% (19/211) and regarding the uterine tubes, cysts with 3.3% (7/211). The classification and number of the identified injuries are shown in Table 1.

**Ovarian changes**

In addition to the follicular cyst, the following lesions were diagnosed in the ovaries: paraovarian cysts 15.6% (33/211) and corpus luteum cysts 11.8% (25/211). Findings of little clinical significance were also diagnosed, such as: 2.3% luteinized cyst (5/211); granulosa cell tumor 0.5% (1/211); adenoma 0.5% (1/211) and agenesis 0.5% (1/211).

In the follicular cyst, this cyst consisted of cystic dilatations filled with translucent fluid in the cortical region of ovary and absence of corpus luteum (Fig.1A). It was observed in the microscopic evaluation of follicular cysts a condition of ovaries with the absence of corpus luteum, follicles without visualization of oocytes; and it could also be described fibrosis and vacuolization in the cells of the thek layer or its absence; as well as a layer of cells of the degenerate, flattened, thin granulosa and, some folded cells and cellular debris.

Regarding the paraovarian cysts, which were located close to the ovaries (Fig.1B), under the microscopy, they were covered by simple cuboidal epithelium and their wall composed of smooth muscle cells; and corpus luteum cysts, which, when cut, identified a cavity in the center filled with translucent material (Fig.1C). The luteinized cyst, also evaluated, showed areas with an absence of the granular layer, a layer of leutein cells in the cyst wall, with yellowish color.

Regarding proliferative changes, one case of granulosa cell tumor was diagnosed. It did not show macroscopic changes. Microscopically, the neoplastic proliferation of granulosa cells was observed, forming structures similar to follicles. Ovarian adenoma, a benign neoplasm, was characterized by the absence of the right ovary, occurred in a sample analyzed simultaneously with the absence of the right uterine tube.

**Table 1. Distribution of 211 lesions in the reproductive system of 1072 sheep in slaughterhouses in the state of Bahia**

| Alterations                          | No. of cases | Frequency (%) |
|-------------------------------------|--------------|---------------|
| **Ovarian changes**                 |              |               |
| Follicular cyst                      | 72           | 34.1          |
| Paraovarian cyst                    | 33           | 15.6          |
| Corpus luteum cyst                  | 25           | 11.8          |
| Luteinized cyst                     | 5            | 2.3           |
| Agenesis                            | 1            | 0.5           |
| Granulosa cell tumor                | 1            | 0.5           |
| Agenesis                            | 1            | 0.5           |
| **Uterine changes**                 |              |               |
| Endometritis                        | 19           | 9             |
| Melanosis                           | 18           | 8.5           |
| Hydrometra                          | 5            | 2.3           |
| Pyometra                            | 5            | 2.3           |
| Abscess                             | 3            | 1.4           |
| Cystic Endometrial Hyperplasia (CEH)| 3            | 1.4           |
| Adenomyosis                         | 2            | 1.0           |
| Petechiae in the serosa             | 2            | 1.0           |
| Segmental aplasia                   | 2            | 1.0           |
| Ectopic pregnancy with fetal m.     | 1            | 0.5           |
| Vaginitis                           | 1            | 0.5           |
| Polyp                               | 1            | 0.5           |
| Changes in uterine tubes            | 11           | 5.3           |
| Ovarian tube cyst                   | 7            | 3.3           |
| Hydrometra                          | 2            | 1.0           |
| Agenesis                            | 1            | 0.5           |
| Adenoma                             | 1            | 0.5           |
| **TOTAL**                           | 211          | 100           |

Uterine inflammatory changes were the most common lesion. In addition, the endometritis was the most observed lesion in the uterus with 9% (19/211). The macroscopic characteristics of endometritis were organ distension, in some cases, from the uterine horns to the cervix, and when cut, with a yellowish-white and paste-like content (Fig.2A). Microscopic evaluation showed a marked inflammatory infiltrate composed of plasmocytes, macrophages, and neutrophils in the submucosa. In one case, in addition to endometritis, dilatation and an increase in the number of endometrial glands were observed, which characterizes Cystic Endometrial Hyperplasia (CEH). Pyometra was diagnosed in 2.3% (5/211). Microscopically, it could be observed an enlarged uterus consisting of yellowish and paste-like content, and of more than 50% purulent exudate.

Among the evaluated cases, two pregnant uteri presented pyometra, where the fetus was located in one of the uterine horns, and the other had content formed by whitish striations. Microscopic analysis showed marked inflammation areas composed of plasma cells, macrophages, and neutrophils in the submucosa. In the bacteriological examination, of the nine samples sent for bacteriological examination, *Escherichia coli* was the most isolated microorganism (6/9), in addition to the following bacteria: *Streptococcus sp.* (1/9) and *Pseudomonas aeruginosa* (1/9). In a sample sent, there was no bacterial growth. The cases of abscesses were located in the uterine wall (Fig.2B) 14% (3/211), both presented 1cm in diameter, characterized by purulent or caseous material that varied from yellowish-white or greenish, with odor or odorless, surrounded by a fibrous capsule. Under the microscopy, the abscess was located in the muscular and serous tissue of the uterine wall. A fibrous connective tissue capsule was observed, with moderate inflammatory infiltrate composed of lymphocytes, macrophages, and plasmocytes, and around the area of inflammatory infiltrate necrosis formed by degenerated...
macrophages and neutrophils. In the vagina, it could be observed vaginitis in 0.5% (1/211) of the animals. Microscopic evaluation showed moderate mixed inflammatory infiltrate composed of lymphocytes, plasmocytes, and neutrophils located under the Basement Membrane (BM).

In hydrometra 2.3% (5/211) of degenerative lesion and the second major pathology were diagnosed, the uterus was dilated and cut, and it was replete with clear fluid accumulation in the lumen. Microscopy showed dilation of the endometrial glands.

With respect to the CEH (1.4%, 3/211), among the proliferative lesions, there was an intense proliferation of endometrial glands with preservation of the morphological pattern. Adenomyosis in 1% (2/211) was identified in the microscopic analysis by distributing the endometrial glands among the myometrial muscle fibers without macroscopic changes. The uterine polyp (0.5%, 1/211) was diagnosed by a pedunculated formation. Microscopically, it had a digitiform shape with the center filled with fibrous connective tissue.

The developmental abnormality was related to total segmental aplasia in 1% (2/211), absence of the formation of a segment of the uterus, the animals presented only one uterine horn. Melanosis in 8.5% (18/211) was characterized by pigmentation distributed in the endometrium areas (Fig.2C). Microscopy showed the distribution of black pigment in the submucosa and loose connective tissue of the endometrium.

Regarding other changes, ectopic pregnancy with fetal maceration was observed in one case, 0.5% (1/211). A whole normal uterus was found, but it was accompanied and connected to the organ removed from the abdominal cavity, a bag of encapsulated fatty tissue, and in its interior bone structures, a macerated fetus.

**Fallopian tubes changes**

The pathologies in the uterine tube were as following: tube-ovarian cyst in 3.3% (7/211), hydrosalpinx in 1% (2/211), adenoma in 0.5% (1/211) and agenesis in 0.5% (1/211). Macroscopy of the cysts was located on the external surface of the wall (Fig.3A), close to the insertion of mesosalpinx. The walls were composed of simple cylindrical ciliated epithelium, basement membrane, and smooth muscle on microscopic examination. Hydrosalpinx, an inflammatory disease, was identified by the unilateral dilation of 0.9cm in diameter of the lumen of uterine tube by liquid content (Fig.3B), under microscopy, absence of folds in the mucosa and, due to distension, the cells exhibited a flattened morphology. The developmental abnormality observed in one case was agenesis of the right uterine tube (Fig.3C). A proliferative lesion found in this organ was the adenoma. Microscopically, cuboid cells were observed grouped in cords and formed the shape of arboriform papillae.
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DISCUSSION

In this study, injuries occurred in 19.5% (211/1,072) of the cases, similar to that observed in other studies of the same nature carried out in the state of Ceará (20.35%) (Santa Rosa et al. 1987), in Iran (25.8%) (Moghaddam & Gooraninejad 2007) and 16.6% (Khodakaram-Tafti & Davari 2013), India (14.98%) (Agrawal 2010) and Himalayas (19.47%) (Sharma et al. 2014).

The ovaries were the most affected organs, with the follicular cyst being the most relevant lesion in this study, presenting results similar to the research by Sharma et al. (2014) as following: 13.56% of lesions in the ovaries was registered (Santa Rosa et al. 1987), in the studies by Moghaddam & Gooraninejad (2007), the highest rates of abnormalities were also in the ovaries consisting of 20.1%, with a predominance of follicular cysts. The etiopathogenesis of follicular cysts may be related to disorders in the Hypothalamic-Pituitary-Gonadal (HPG) axis. If the gonadotropin-releasing hormone (GnRH) is not released from the hypothalamus due to estradiol stimulation, or the anterior pituitary does not release the Luteinizing Hormone (LH), they will cause cyst formation and an increase in progesterone (Silva et al. 2015). The absence of LH receptors in the mature follicle or deficiency of GnRH receptors in the pituitary gland is also related to cyst formation (Santos et al. 2016). Similar results were identified in the ovaries of cows in the microscopic evaluation, such as the absence of oocyte, pelucid zone, and granulosa cells, in addition to internal teak fibrosis and partial luteinization of the cystic wall (Ramos et al. 2010).

The granulosa cell tumor is an ovarian neoplasm of the sexual cords, initially formed by cells similar to the follicular granulosa cells (Santos et al. 2016), and it could be observed in a sheep in this study. Similar data were described in three cows, in which in one case, no macroscopic changes were observed. However, microscopy showed rosette formation in the granulosa cells (Hatipoglu et al. 2002).

The adenoma, a lesion identified in one case in this research, is characterized as an epithelial neoplasm derived from the germinal epithelium, from the Subsurface Epithelial Structures (SES), or the rete ovarii. It did not occur in similar studies by other authors (Santa Rosa et al. 1987, Moghaddam & Gooraninejad 2007, Agrawal 2010, Khodakaram-Tafti & Davari 2013, Sharma et al. 2014). Under the microscopic examination, they tend to have cuboid cells grouped into cords, with a pyknotic nucleus, a sparse cytoplasm grouped in the shape of arboriform papillae (Akihara et al. 2007).

Among the findings with little clinical significance, the paraovarian cyst represented 15.6% and the corpus luteum cysts 11.8%. In a study conducted by Khodakaram-Tafti & Davari (2013), it could be found 0.9% paraovarian cysts in sheep, and in a research performed with cows, this cyst was identified in eight cases (Hatipoglu et al. 2002). The corpus luteum cyst is characterized by the formation of a cystic cavity in the center of the lutein tissue, present in 25% of the corpus luteum (Santos et al. 2016). In studies with cows, 0.74% (Ramos et al. 2010) and 2.51% (Hatipoglu et al. 2002) of corpus luteum cysts were identified.

Endometritis was the most identified uterine pathology in the present study, comprising 9% of changes in the uterus characterized by yellowish-white mucus. In other studies carried out with sheep, endometritis was predominant in 2.93% (Khodakaram-Tafti & Davari 2013) and 0.34% of cases (Agrawal 2010). In the macroscopic evaluation, the presence of mucus, which can vary from serous to purulent and also from varying degrees of purulent striations (Schlafer & Miller 2007), hyperemia and endometrial edema (Santos et al. 2016). Microscopically, an inflammatory infiltrate formed by lymphocytes, histiocytes, and neutrophils in periglandular and perivascular regions could be identified, and it was possible to verify fibrosis in these areas as well (Bonnett et al. 1993, Bondurant 1999). It is a frequent alteration in the uterus of several domestic females that reduces reproductive efficiency of herds, increases the occurrence of estrous repetition and birth interval, and causes a drop in milk production (Nascimento & Santos 2011).

Pyometra, acute or chronic infection of the uterus, with deposition of purulent material and persistence of a corpus luteum, can occur due to endometritis or metritis (Dow 1960, Sheldon et al. 2008, Schlafer & Foster 2015). When it forms purulent exudate, infection is usually caused by Staphylococcus sp. and Streptococcus sp. (Johnson 2006, Foster 2013, Schlafer & Foster 2015). In this work, the bacteria Escherichia coli and Streptococcus sp could be detected.

Pyometra was the second pathology diagnosed in this research with 2.3%. It is described as the accumulation of clear and aseptic fluid in the lumen of the uterus (Nascimento & Santos 2011, Foster 2013), which may be concomitant to the development of endometrial hyperplasia (Santos et al. 2016), as well as being characterized as widespread pathology in...
A case of adenoma was also diagnosed in the fallopian tube. Epithelial and mesenchymal neoplasms of this organ are rare in domestic animals. There are reports of benign epithelial neoplasms in dogs (Pierozan et al. 2016, Gelberg & McEntee 1986, Marino et al. 2007). The principal differential diagnosis of adenoma is related to the adenocarcinoma, and implantation in other organs and peritoneal cavities should be considered (Gelberg & McEntee 1986, Pierozan et al. 2016).

CONCLUSION

Infertility and reduction in reproductive efficiency of sheep can be caused mainly by follicular cysts, endometritis, and cysts in the uterine tubes. These important pathologies interfere in the productivity of sheep farming. Early diagnosis of these changes prevents probable losses in production.

Conflict of interest statement.- The authors have no competing interests.

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