The Armed Forces Institute of Pathology
Department of Veterinary Pathology

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Moderator:

Dr. Don Schlafer, DVM, DACVP, DCVM, DACT, PhD

CASE I – A02-388X (AFIP 3038528).

Signalment: 31-year-old, female, Rhesus macaque (*Macaca mulatta*)

History: Used in an aging study examining neuropathological and behavioral changes related to age. Euthanized at end of study.

Gross Pathology: The right ovary is enlarged (5 x 6 cm) with multiple, large multiloculated cysts, the largest measuring approximately 3.5 to 4cm in diameter. On cut section, the cysts are demarcated by thin walls and contain an orange to red, proteinaceous material. Vagina, cervix and uterus are normal. The left ovary is twice enlarged and contains a solid mass.

Histopathologic Description: The right ovary contains two distinct tumors (a cystadenoma and a granulosa cell tumor) that replace the normal ovarian tissue. The cystadenoma is composed of numerous large cysts filled with eosinophilic homogenous material (proteinaceous fluid), lined by flat cuboidal epithelium with small basophilic nuclei, minimal eosinophilic cytoplasm, separated by and growing along dense collagenous connective tissue. The neoplastic cells form papillary projections into the cysts and in the surrounding interstitium. Mitoses are rare. The granulosa cell tumor is composed of a population of neoplastic cells arranged in stratified layers of polygonal cells with small basophilic nuclei and scant cytoplasm lining spaces with flocculent amphophilic material and growing along a fibrovascular stroma. This population of cells occasionally forms rings of pallisading cells at the center of which is deeply eosinophilic material **(Call-Exner bodies) (fig. 1-1)**. Nests and packets of these cells infiltrate the thin rim of dense ovarian stromal tissue at the periphery. Primary follicles are rare consistent with the monkey's age. Deeply basophilic irregular material (mineral) is multifocally present. Mitoses are rare.

Contributor's Morphologic Diagnosis: Ovary: Papillary serous cystadenoma and Granulosa cell tumor, macrofollicular (coincident in the same ovary)

Contributor's Comment: Ovarian masses can be separated into cysts and neoplasms. Cysts within the ovary are identified by dilatations not involving gonadal or stromal tissue. Serous inclusion cysts have been reported in the bitch and cystic rete tubules have been reported in the bitch and queen. Cysts involving the gonadal stroma are typically cycle dependent and include: cystic graffian follicles, follicular cysts, anovulatory luteinized cysts, and cystic corpus luteum. These are frequently associated with cows and sows.[1] In humans, cystic follicles are so frequent they can be considered physiologic originating from graffian follicles.[2]

Ovarian neoplasms account for 6% of all cancers in the female and are the fifth most common form of cancer in women in the United States (excluding skin cancer). Due to the difficult rate of early detection, these neoplasms are responsible for almost half of the deaths from cancer of the female genital tract. There are numerous types of ovarian tumors, both benign and malignant. About 80% are benign, and these occur mostly in young women between the ages of 20 and 45 years. The malignant tumors are more common in older women between the ages of 40 and 65 years.[2]

Ovarian neoplasms are generally broken into four categories—germ cell, sex cord stromal, surface epithelial, and mesenchymal. A summary of diagnostic criteria can be found in the *Histological Classification of Tumors of the Genital System of Domestic Animals, AFIP Second Series, Volume VI.*[3]

We classified this ovary as having a component of a granulosa cell tumor **(fig. 1-1)** based on its morphologic appearance of cells containing spherical-to-oval, hyperchromatic nuclei, distinct nucleoli, and scant eosinophilic cytoplasm. The Call-Exner bodies supported this diagnosis. The other cystic component of this ovary is composed of infoldings and papillary projections of subsurface small cuboidal epithelium, scant connective tissue stroma, and rare mitoses consistent with papillary serous cystadenoma. Immunohistochemistry for the cystic tissue was strongly positive for cytokeratin, confirming its **epithelial origin (fig. 1-4)**. The granulosa cell component was positive for **inhibin (fig. 1-2)**.

Dual ovarian tumors in non-human primates have not been reported.[5] Dual tumors in ovaries have been reported in humans, but are generally rare.[6,7] The pathogenesis for dual tumors is unknown, but postulated theories include: collision neoplasm, in which two tumors develop spontaneously; a heterologous differentiation within a granulosa cell tumor; or a teratomatous neoplasm with a bidirectional differentiation. Recently, a

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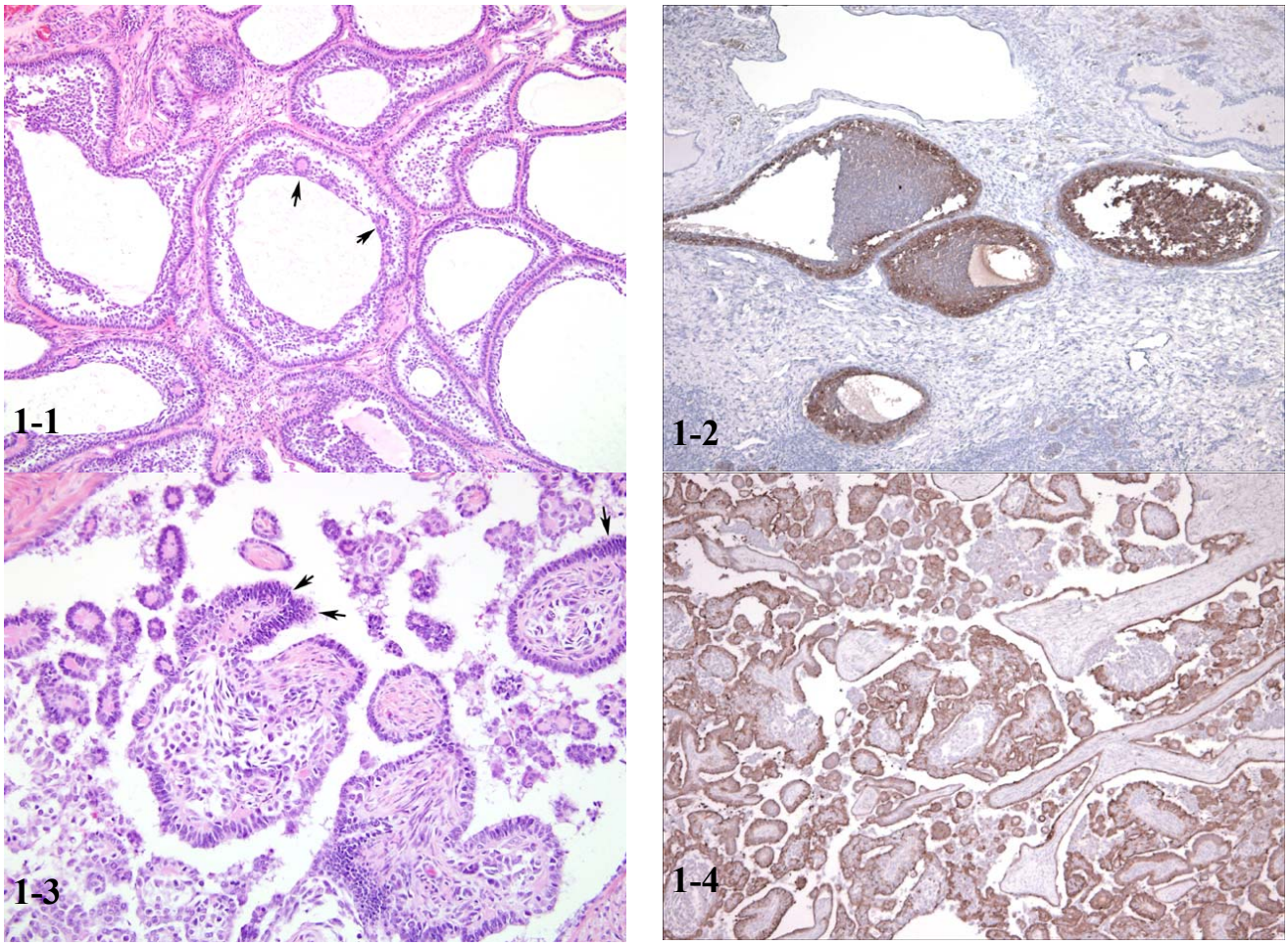
Table 1-1. Key ovarian neoplasms documented in veterinary species. (Courtesy New England Primate Research Center, Harvard Medical School, Division of Comparative Pathology, Southborough, MA, 01772)

Equine	Granulosa cell tumor (almost always unilateral, slow growing, and benign; elevated inhibin in 90%) Cystadenoma—most common tumor of surface epithelium
Bovine	Granulosa cell tumor
Feline	Malignant granulosa cell tumor
Canine	Papillary cystadenocarcinoma, malignant granulosa cell tumor; malignant teratoma
Murine	typically irradiation induced, all types; spontaneous cystadenoma and granulosa cell tumors have been reported.
Rat	Osborne-Mende strain 330, 33% of rats > 18 months develop granulosa cell tumors; Sprague Dawley predisposed to a variety of histological subtypes
NHP	Granulosa cell tumor, teratoma, and cystadenocarcinoma have been reported in baboons, recently choriocarcinomas have been reported in macaques
Ferret	Sex cord stromal resulting in alopecia (Comp Med 2003)
Fish	Ovarian carcinoma in a koi carp (Aus Vet J, 2006)
Poultry	Adenocarcinoma of turkeys and chickens in intensive-laying conditions. Cornell has C strain genetically predisposed to epithelial cancer
Snake	Granulosa cell tumor (especially garter snakes)
Lizard	Teratomas (especially iguanas)



1-1. Ovary, Rhesus macaque. The granulosa cell tumor is composed of stratified trabeculae of polygonal cells as well as rings of pallisading neoplastic cells surrounding small cores of eosinophilic, acellular, homogeneous material (Call-Exner bodies) (arows) (H&E 200X)
1-2. Ovary, Rhesus macaque. The neoplastic cells of the granulosa cell tumor are multifocally immuno-positivite for inhibin. (Inhibin 40X)
Photomicrograph courtesy of the New England Primate Research Center, Harvard Medical School, Division of Comparative Pathology, Southborough, MA 01772
1-3. Ovary, Rhesus macaque. The cystadenocarcinoma displays features of low grade malignancy to include karyotypic polymorphism as well as piling of the neoplastic cells (arrows) (H&E 200X)
1-4. Ovary, Rhesus macaque. Cystadenocarcinoma. The epithelial component is immunoreactive for cytokeratin (cytokeratin 40X). Photomicrograph courtesy of the New England Primate Research Center, Harvard Medical School, Division of Comparative Pathology, Southborough, MA 01772

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case of mucinous cystadenoma and granulosa cell tumor was reported in a 57-year-old woman.[7] The immunohistochemical profile of this tumor demonstrated diffuse positive CK7 and focal weak CK20 within the mucinous component. The granulosa cell component was strongly alpha-inhibin positive and diffuse calretinin positive while being negative for epithelial membrane antigen (EMA) and anticytokeratin antibody AE1/3.

Mixed ovarian tumors in the veterinary literature are also extremely rare. A brief literature review shows key ovarian neoplasms documented in veterinary species (Table 1-1):

For more information, the reader is directed to previous ovarian neoplasm submissions to AFIP as well as the

following websites:

- http://radiology.uchc.edu/eAtlas/nav/msOvary.htm
- http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=PubMed&list_uids=16309432&dopt=Abstract

AFIP Diagnosis: 1. Ovary: Papillary serous cystadenocarcinoma, Rhesus macaque (*Macaca mulatta*), primate. 2. Ovary: Granulosa cell tumor.

Conference Comment: Granulosa cell tumors are the most common ovarian tumor in large animals. They are generally benign in the cow and horse but are often malignant in dogs and cats.[1] Sex cord-stromal tumors may be hormonally active and produce varying amounts of progesterone, estrogen, testosterone, and inhibin.[4] Anestrus, nymphomania, or stallion-like behavior are often seen in the mare, while the bitch may develop prolonged estrus or pyometra.

Epithelial tumors of the ovary generally arise from the surface epithelium, rete ovarii, and from the subsurface epithelial structures (SES) of the bitch. The bitch is unique in that the canine is the only domestic animal to contain SES, resulting in tumors of the ovary being common only in the bitch. This case was studied in consultation with pathologists in the Department of Gynecologic and Breast Pathology of the Armed Forces Institute of Pathology who agree that the epithelial portion of the neoplasm exhibited areas of low-grade carcinoma **(fig. 1-3).** Multifocally neoplastic epithelial cells exhibited moderate cellular atypia and pile up to 5 cell layers thick. Features of malignancy without evidence of metastasis or vascular invasion include a larger size, necrosis, hemorrhage, cellular atypia, piling up of neoplastic cells, increased mitotic index, and stromal invasion.[4]

Contributor: New England Primate Research Center, Harvard Medical School, Division of Comparative Pathology, Southborough, MA, 01772

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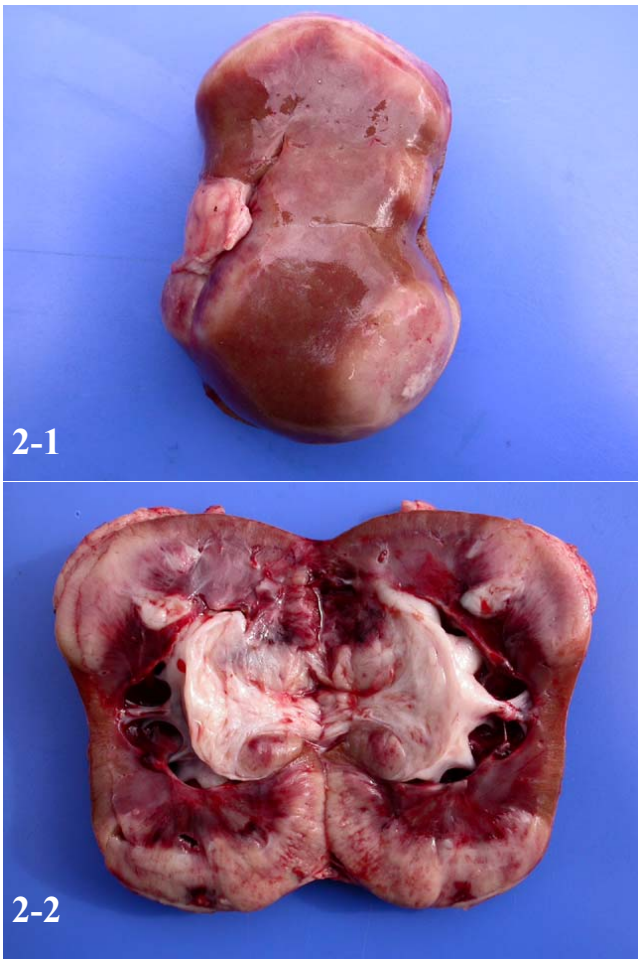
Classification of ovarian tumors[3]

Sex cord-stromal (gonadostromal) tumors	• Granulosa cell tumor (granulose-theca cell tumor) • Thecoma (theca cell tumor) • Interstitial cell tumor (luteoma, lipid cell tumor, steroid cell tumor)
Germ cell tumors	• Dysgerminoma • Teratoma • Embryonal carcinoma
Epithelial tumors	• Papillary adenoma, papillary cystadenoma • Papillary adenocarcinoma • Rete adenoma
Mesenchymal tumors	• Hemangioma • Leiomyoma

2-1. Kidney, Labrador Retriever. Grossly the kidney is enlarged with multiple, randomly distributed masses elevating the capsule.
2-2. Kidney, Labrador Retriever. On cut surface, the renal parenchyma is expanded and disrupted by 3 coalescing masses replacing the cortex, medulla, pelvis and ureter.

Photographs courtesy of Claudio S.L. Barros of the Departamento de Patologia, Universidade Federal de Santa Maria, Santa Maria, 97105-900, RS, Brazil



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CASE II – UFSM-2 (AFIP 2992242).

Signalment: 5-year-old, male, Labrador retriever, canine

History: On June 5, 2004 the dog was presented with a serosanguineous discharge form the prepuce. It was reported by the owner that these signs had started a week ago. On physical examination, a large mass (10 cm in diameter) could be palpated in the prepuce. The owner reported that the growth was noticed two years ago. Extrusion of the penis revealed an 8x10 cm lobulated and verrucous mass involving the caudal aspects of the penile shaft. On cytologic examination of samples obtained of fine needle aspiration the mass, a diagnosis of transmissible venereal tumor (TVT) was made. The dog was placed on chemotherapy (vincristine, 4 weekly injections of 0.5 mg/m^2 body weight). The tumor had regressed somewhat but an enlargement was noticed in the inguinal lymph node one month after the start of the therapy. The penis was amputated and the lymph node excised. At the histopathological examination TVT was confirmed as the primary tumor and metastatic TVT was diagnosed in the lymph node. The dog was sent home with no further treatment. Two months later it returned to the Veterinary Teaching Hospital for check-up when "a mass in the abdominal cavity" was palpated. An ultrasound of the abdominal cavity revealed an enlarged left kidney which was excised and sent to the pathology laboratory. The dog's condition seemed to have somewhat improved and it was sent home again. A couple of weeks after surgery the dog started to progressively loose weight and after two months it was euthanatized. The owner did not permit a necropsy.

Gross Pathologic Findings: The surgical specimen was that of an enlarged kidney (12.0x7.0x5.5 cm) which presented large multifocal white masses protruding from the **capsular surface (fig. 2-1)**. At cut surface the pelvis and calices were marked dilated and large amounts of clear fluid oozed from the pelvis. Three large homogenous white **coalescing masses (fig. 2-2)** involved cortex, medulla pelvis and ureter. In some places sparse hemorrhagic and necrotic foci could be observed in the neoplastic mass.

Laboratory Results: Cytology performed in sample collected by fine needle aspiration of the penile tumor revealed clusters of round cells with high nuleus: cyto-

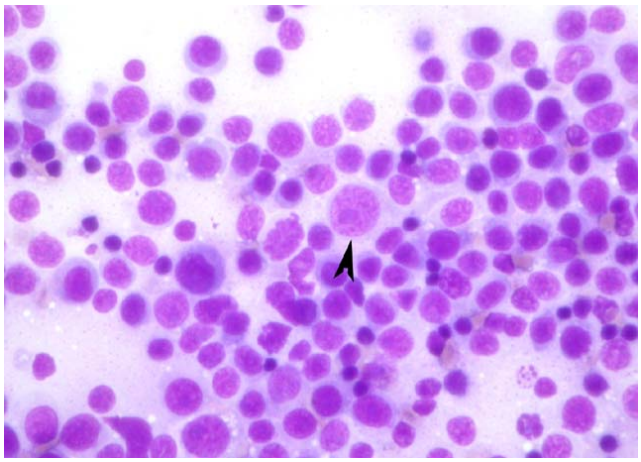
plasm ratio and moderate anisocytosis. The nucleus of these cells was round or oval and formed by loose chromatin with one irregularly shaped conspicuous nucleolus. The cytoplasm of these cells was scant, slightly basophilic, and occasionally revealed multiple small **vacuoles (fig. 2-3)**. There were multiples cells in mitosis, occasionally binucleated cells and moderate numbers of small lymphocytes.

Contributor's Morphologic Diagnosis: Kidney, metastatic transmissible venereal tumor, 5-year-old, male, Labrador retriever, canine

Contributor's Comment: Canine transmissible venereal tumor (TVT), also referred to as Sticker tumor, infectious sarcoma, venereal granuloma, canine condyloma, transmissible lymphosarcoma, transmissible tumor of reticular cells, transmissible histiocytoma and hemoblastoma was described in the 19th century and reportedly is one of the major canine diseases in underdeveloped countries, mainly in those with temperate, tropical or subtropical climates.[13] The tumor affects exclusively dogs and is considered always potentially malign;[12] the cell of origin of the tumor is still unknown, although iummunohistochemical studies support an histiocytic lineage.[9] Attempts on viral isolation from TVT tissues have consistently failed in the past; more recently viral particles have been isolated from TVT fragments but the inoculation of theses particles in dogs failed to reproduced the disease;[8] despite of these negative findings, the viral etiology is favored by some researchers.

In one study, the neutering of bitches affected by the neoplasm resulted in rapid regression of TVT suggesting that the tumor is somewhat hormone-dependant. Additionally, TVTs reportedly tend to be benign in males and frequently metastatizing in females.[5,8,12] While the number of chromosomes in somatic canine cells is 76, the number of chromosomes in the cells of TVT is consistently 58. Such a disparity led, in the past, to the belief that the neoplasm had been acquired from another animal species. The analysis of the MHC molecules on cells of TVT from dogs of different geographic origins revealed that all have the same surface antigens.[8]

The transmission of TVT occurs by allogenic transplantation of viable neoplastic cells from an affected dog to a susceptible one, normally during copulation; however other means of transmission are possible and include licking, biting, and scratching.[5,8,12] The neoplasms affect the external genitalia (penis and vagina) and the skin adjacent to these areas.[5,8,13] Less commonly affected sites include nasal cavity, eyes, lips and other skin regions.[8,12]



2-3. Kidney, Labrador Retriever. Cytological preparation, fine needle aspirate. Neoplastic cells have a small amount of palely basophilic cytoplasm which is microvacuolated. (arrowhead). Photomicrograph courtesy of Claudio S.L. Barros of the Departamento de Patologia, Universidade Federal de Santa Maria, Santa Maria, 97105-900, RS, Brazil

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Dogs of both sexes and all ages are affected but the disease is more prevalent in sexually active dog (average age 4-5-year-old) living in areas with large populations of stray dogs. [12,13]

Gross aspects of TVT are variable but most are either firm or friable verrucous papillary or nodular masses protruding from the surface of penis or vulva.[5] The tumors can be small single nodules or multilobulated masses up to 15 cm in diameter.[5] The surface of these neoplasms are smooth, granular and commonly ulcerated from where bleeding is frequent.[8]

Histologically, TVTs consist of round to oval cells which are closely similar to macrophages and are arranged in ribbons or pallisades; their nuclei are large, round, centrally located in the cell displaying clusters of chromatin and a single prominent, centrally located nucleolus. The cytoplasm of TVT cells is moderate in amount, faintly basophilic and vacuolated. Mitotic index is high and the tumor parenchyma is infiltrated by variable numbers of lymphocytes, plasm cells and macrophages. In regressing TVTs, inflammation, necrosis and fibrosis are frequently seen. [5,8]

The prognosis for TVTs is guarded as these tumors are self limiting in most of the cases,[8] but not uncommonly metastasize to regional lymphnodes, spleen, liver, kidney, peritoneum, lungs and central nervous system.[8,12] Furthermore, surgical excision only results in recurrence

which in some reports is close to 60% of the cases.[12] In a study carried out in a dog colony, the neoplasm was transmitted through 40 generations in a total number of 564 dogs, 68% of which developed the disease and 87% had spontaneous regression of the tumor within 180 days. When dogs are submitted to specific chemotherapy, the prognosis is good in the majority of cases. Dogs recovering from this neoplasm acquire cellular and humoral immunity.[1]

In the case of this report, necropsy was not allowed and thus was not possible to determine the route of metastasis. It is not possible to ascertain if metastasis foci were present in other organs but the surgeon reported only the tumor in the left kidney. The presence of neoplastic tissue in the pelvis and ureter raises the possibility that dissemination of the tumor might have occurred by ascending the urinary tract.

AFIP Diagnosis: Kidney: Transmissible venereal tumor, metastatic, Labrador retriever (*Canis familiaris*), canine.

Conference Comment: The contributor gives an excellent description of canine transmissible venereal tumors (TVT). Currently the TVT is the only known naturally transmitted neoplasm. However, it has been proposed that the recently described Devil facial tumor disease may be transmitted in a similar manner.[7]

It is not known how the tumor evades the host immune system. Class I and II major histocompatibility antigens are not expressed by the tumor cells until regression occurs.[11] Cells of TVTs secrete TGF-β1 and IL-6 which suppress the expression of major histocompatibility antigens.[14]

The histologic appearance of the neoplasm can vary greatly depending on the stage of growth or regression. During regression, TVT cells express major histocompatibility complex class II antigens, and therefore the neoplasms are often infiltrated by inflammatory cells, particularly T lymphocytes. The effect of vincristine administration on the cytomorphology and level of regression in this tumor is difficult to assess.

Contributor: Claudio S.L. Barros of the Departamento de Patologia, Universidade Federal de Santa Maria, Santa Maria, 97105-900, RS, Brazil

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CASE III – MK0605183 (AFIP 3069157).

Signalment: 16-year-old, female, Rhesus macaque (*Macaca mulatta*)

History: The animal had been infected with Simian immunodeficiency virus (SIV) and was being treated with Depo Provera for endometriosis. The animal had a history of several episodes of dehydration despite adequate access to water. Previous bouts of dehydration had responded to fluid therapy. On the day of presentation, the animal was found down in the cage, hypothermic, dehydrated and weak with a distended abdomen. Intravenous fluid therapy, including IV dextrose, was initiated prior to the collection of blood.

Laboratory results: The serum chemical findings were consistent with chronic renal disease (see values table 3-1). The animal was euthanized after failing to respond to IV fluid therapy.

Gross Pathology: The animal was in lean body condition with adequate hydration. Multifocally within the abdomen, the omentum was attached loosely to the peritoneal wall. In between the loops of intestine and on the capsular surface of the kidneys, there were multifocal, small, white, fibrous adhesions. Within the caudal abdomen, surrounding and invading the body of the uterus and effacing the ovaries, and compressing the colon and bladder was a 3 x 3.5 x 6 cm^3 cystic mass. The mass contained approximately 10ml of clear fluid and had an irregularly thickened, lobular inner surface. The lungs were congested with moderate edema and the kidneys were pale. No other significant lesions were noted in the heart, liver, spleen, gastrointestinal tract, or brain.

Histopathologic Description: Submitted sections were from the uterus, ovary, and oviduct. Multifocally, the serosal surfaces of these organs were irregularly expanded by a thick band of homogenous, eosinophilic material with numerous, widely spaced, 15 – 25µm polygonal cells and few glands lined by cuboidal epithelium. The polygonal cells had distinct cell borders with ample, fibrillar cytoplasm. Nuclei were round – oval, with reticular chromatin and a single nucleolus [decidualized stromal cells]. No mitotic figures were seen within the polygonal cells. In some sections, there were small lymphoid aggregates in the stroma. Some glands contained cellular and necrotic debris, but RBCs and hemosiderophages were not a common finding. Few sections contained small foci of mineralization. On some slides, the endometrium was expanded by similar decidualized stromal cells and ample eosinophilic stroma.

Special stains/Immunohistochemistry: The eosinophilic matrix was not birefringent with Congo red staining and few fibrils were seen with trichome stain. The polygonal cells failed to stain with macrophage [HAM56], muscle [Desmin], and epithelium [AE1/AE3] markers.

Contributor's Morphologic Diagnoses: Caudal abdominal mass: Endometriosis with decidualized stromal cells.

Contributor's Comment: AFIP confirmed the diagnosis of endometriosis with decidualized stromal cells. In addition to the uterus and ovaries, endometriosis affected the intestines, kidneys, colon, and bladder in this animal. Although endometriosis is not an unusual lesion in macaques, the presence of the large polygonal decidualized stromal cells with abundant eosinophilic, homogenous stroma was not a change we had commonly encountered. The endometriotic stromal cells we have seen in the past have been small, spindloid, contained little to no cytoplasm, had spindle-shaped nuclei and were not widely separated by stroma.

Endometriosis, the presence of endometrial tissue outside the uterus, is a progressive disease that occurs in women and old world primates, primarily during their reproductive years. [5,10,12,17] Clinical signs associated with endometriosis include dysmenorrhea, dyspareunia, pelvic pain, interference with intestinal and urinary bladder function and reduced fertility.[10] In rhesus, there may be similar signs with irregular vaginal bleeding or heavy menses and signs of pain include lying down in the cage, anorexia, grimacing, decreased grooming, restlessness, and vocalization.[5,10]

On gross examination or laparotomy, endometriosis may have a varied appearance but is most easily recognized by red/brown [chocolate] cysts on the serosal surface of pelvic and abdominal organs. The cysts are endometriotic glands containing viable and degenerate RBCs and hemosiderophages.[5,15] Endometriotic lesions may also exist as clear cysts and variably-sized adhesions between organs.[15] In women with endometriosis, the organs most likely affected (in descending order) are: ovaries, uterine ligaments, rectovaginal septum, pelvic peritoneum, as well as laparotomy scars.[4] Endometriosis may also occur outside of the peritoneum and, rarely, in men.[12]

Endometriosis is definitively diagnosed by laparoscopic surgery to biopsy/remove suspected lesions with histologic examination of the samples. [11] The diagnosis of endometriosis is based upon the presence of endometrial

Table 3-1. Laboratory results, case 3.

Analyte	Value	Normal	Units
BUN	175↑	5 - 25	mg/dL
Creatinine	4.4↑	0.5 - 1.1	mg/dL
Total protein	4.7↓	6 - 8.5	g/dL
Sodium	119↓	145 - 152	mmol/L
Chloride	92↓	105 - 115	mmol/L
Potassium	7.3↑	3 - 4.5	mmol/L
Calcium	1.92↓	2.1 - 2.55	mg/dL
Phosphorous	>12↑	3 - 4.5	mg/dL
Glucose	470↑	60 - 120	mg/dL
WBC	20.8↑	5 – 13.5	K/μL
Polys - calc	18.7 ↑	1.6 – 7.4	K/μL
HCT	34	33 - 45	%

glands and stroma outside the uterus.[4,12] In women, stromal changes that may be associated with endometriosis are fibrosis, numerous small vessels, aggregates of foamy and pigmented [hemosiderin] macrophages, smooth muscle metaplasia, and myxoid change.[3] Progestin treatment, which this macaque received, can cause decidual change in the endometrial stromal cells so that stromal cells appear large and polygonal rather than small and spindeloid. The appearance of endometriotic glands can range from normal-appearing glands lined by cuboidal epithelial cells to glands lined by flattened epithelium that may be mistaken for ectatic vessels.[3] Hormone treatment, pregnancy, and menopause will change the appearance of both the stroma and the glands. For suspected cases of endometriosis, the immunohistochemical marker CD10, which stains both normal and endometriotic stromal cells, can be used to assist in making the diagnosis.[3]

Historically, endometriosis has been thought to develop and progress via one of three theoretical pathways:

1. Regurgitation/implantation: Retrograde menstruation, the backflow of uterine contents, including epithelial cells, and debris through the fallopian tubes into the peritoneal cavity[10], may cause endometriosis.[4]
2. Metaplasia: The peritoneum of the pelvis, which arises from the same embryonic coelomic epithelium as the endometrium, undergoes metaplastic change to develop into endometriosis.[4,8,12]
3. Vascular or lymphatic dissemination: Endometriotic tissue may be transported through pelvic veins and lymphatics to cause endometriosis in tissues distant from the uterus such as lungs and lymph nodes.[4]

The current understanding is that the first step in the pathogenesis of endometriosis is the regurgitation of endometrial contents into the peritoneal cavity.[2] However, retrograde menstruation occurs in 76-90% of women, most of who never develop endometriosis.[5] How the ectopic endometrial tissue develops into endometriosis is still being defined. Although the disease estrogen-dependent,[5] the progression is likely multifactorial in-

volving hormonal, immune, genetic and environmental influences.[10] Significant factors associated with the development of endometriosis are: kinship with affected individuals; abdominal surgeries such as cesarean sections, fetal instrumentation, ovarian follicle aspirations and embryo transfers; age; and with environmental factors such as exposure to dioxin, polychlorinated biphenyls (PCBs) and ionizing radiation.[1,17] Additional risk factors for the development of endometriosis in humans are short menstrual cycle length, prolonged menses, low parity number and increased serum estrogen.[15]

Recent work using gene expression analysis to evaluate the uterine endometrium of women with endometriosis and those without the disease has identified numerous genes that are both up and down regulated in comparison to non affected women. In addition, the uterine endometrium of women with moderate/severe endometriosis is less sensitive to the effects of progesterone. In normal endometrium, progesterone has an antiproliferative effect and, after ovulation, leads to the onset of secretory phase and deciduization of stroma. In the endometrium of affected women, progesterone resistance leads to enhanced cellular survival, as well as decreased regulation of DNA synthesis and cellular mitosis.[2]

Nonprimate and primate animal models have been developed to study the genesis and progression of endometriosis. Endometriosis can be induced in rodents and rabbits by implanting autologous or human uterine tissue within the peritoneum. These models have been helpful in the study of early events involved in the attachment of endometrial stroma and glands after implantation into the abdomen. In addition, the influence of immune and inflammatory components in the development of endometriosis can be more easily studied in a sequential manner in smaller animal models. However, rodents and rabbits lack a true menstrual cycle and the endometriotic lesions they develop differ from those seen in women.[15]

Endometriosis occurs spontaneously and can be induced in primates.[15] In colonies of captive macaques, spontaneous endometriosis has been seen in up to 25% of the population.[10,17] Rhesus macaques have been most often studied and are a good model for human endometriosis in that their menstrual cycle is approximately 28 days long, with a 4 day menstrual bleeding period, they tend to develop the disease during their reproductive years, and have disease that progresses with time. In addition, the clinical signs, gross and histologic lesions in the macaques are similar to those seen in women.[10,17]

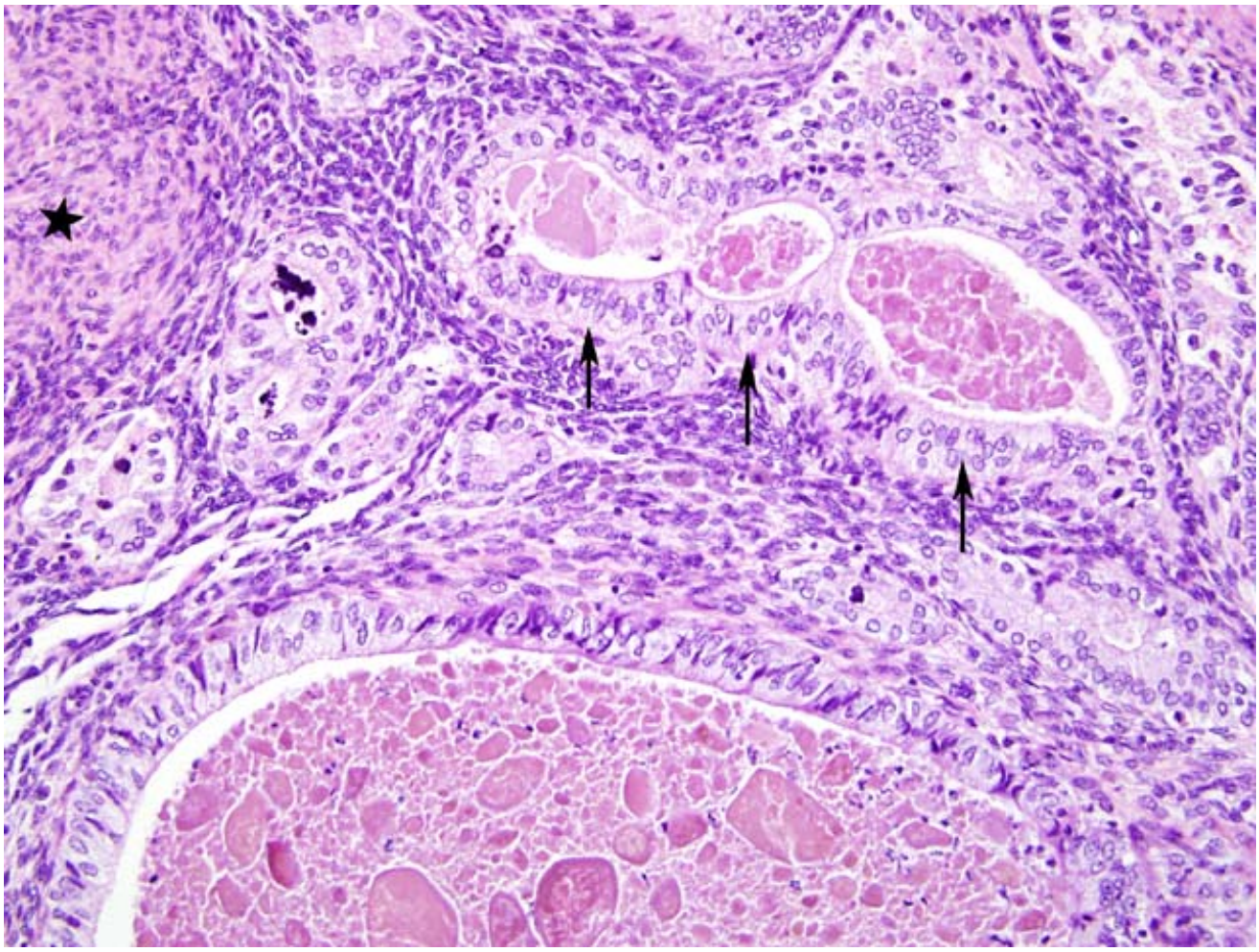
The treatment goals for endometriosis are to decrease pain, decrease the size of endometriotic lesions, remove associated obstructions, and restore fertility.[11] Treatment may involve surgical removal of endometrial explants within the abdomen or medical treatment to cause the ectopic tissue to atrophy or both. Unfortunately, clinical signs may recur after surgery or after hormonal treatment is discontinued.[12,11] Medical therapies target the hypothalamic-pituitary-gonadal axis, by selective modulation of estrogenic and progestogenic pathways, by inhibiting angiogenesis, or by modulating inflammatory and immunological responses.[5,11] Depo Provera, a commonly used progestin-based treatment, will cause endometrial tissue to atrophy, but also may reduce bone density. Other hormone treatments include gonadotropin-releasing hormone agonists, progestogens, androgenic agonists, combined oral contraceptive therapy, and antiprogestogens may also lead to osteoporosis.[11] More recent therapies include selective progesterone-receptor modulators (SPRMs), selective estrogen-receptor modulators (SERMs) which reduce estrogen but have a bone-sparing effect, immunomodulatory drugs, angiogenesis inhibitors which may interfere with implantation and vascularization of ectopic implants, aromatase inhibitors (AIs) which decrease non-ovarian sources of estrogen, and statins which reduce the growth of endometrial stroma *in vitro*.[5]

Renal changes included multifocal, moderate chronic infarcts with mild chronic interstitial nephritis as well as moderate acute tubular necrosis with tubular proteinosis. Adverse renal effects from Depo Provera treatment have not been reported.[13] Clinical veterinarians did not find a cause for the episodes of dehydration and no other animals in colony developed unexplained dehydration. A specific agent for the renal changes was not identified. No opportunistic infections or changes associated with SIV infection were seen in the other tissues of this animal.

AFIP Diagnosis: Ovary and uterus: Endometriosis, with decidualized stromal cells, Rhesus macaque (*Macaca mulatta*), nonhuman primate.

Conference Comment: The contributor gives an excellent overview of endometriosis.

Endometriosis is defined as **endometrial glands or stroma (fig. 3-1)** explanted to abnormal locations within and outside the uterus.[3] The explanted tissue responds to hormonal stimulation similar to normal endometrium.[4] It has been reported in humans, old world monkeys, and apes.[15] Key histologic features include endometrial glands, endometrial stroma, and pigment-containing histiocytes. These features may vary depending on hormonal stimulation. In longstanding cases of endometrio-



3-1. Ovary and uterus, Rhesus macaque. Endometrial glands (arrows) and stroma (star) expanding the ovary and abnormal locations within the uterine wall.. (H&E 200X)

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sis, the endometrial glands or endometrial stroma may be obscured by fibrosis or an infiltrate of histiocytes that contain hemosiderin, ceroid, or lipofuscin.[3]

Adenomyosis is endometrial stroma and/or glands within the myometrium of the uterine wall and has been reported in humans and domestic animals.[4,14]

Contributing Institution: Division of Veterinary Resources, National Institutes of Health, Bethesda, MD

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CASE IV - Case 1 (AFIP 3066305).

Signalment: Heart tissue and fetal membranes are from a male, mixed-breed goat fetus, with a crown to rump measurement of 28cm and weighing 911g.

History: A group of pregnant does at various stages of gestation were co-mingled with three, BVDV (Type 2) persistently infected heifers.

Gross Pathologic Findings: Examination of fetal tissues revealed no significant gross lesions.

Laboratory Results:
Tests performed on fetal tissues
Microbiology:
Bacterial cultures, negative
Fluorescent Antibody
BVDV, positive
Immunohistochemistry
BVDV, positive
Polymerase chain reaction (PCR)
BVDV, positive (typed as Type 2)
Chlamydia, negative
Bluetongue virus (BTV), negative
Virus Isolation
Negative on tissues from this case submission; however, BVDV was isolated from 2 other aborted fetuses with similar histological lesions in this study.

Histopathologic Description: From tissues submitted: Placenta: Creating a thin band along the superficial chorionic stroma, there are scattered necrotic cells admixed with abundant cellular debris. The overlying trophoblastic epithelium is often absent within the more affected regions. The vessels deep to the more affected areas are similarly peppered with nuclear debris, which occasionally obscures the endothelium and muscular layers of the vessel wall. In the deeper stroma, there is a mild, multifocal infiltrate of individually scattered mononuclear cells.

Heart: The heart exhibits a mild infiltrate of mononuclear inflammatory cells forming scattered infiltrates within the epicardium and forming perivascular cuffs within the myocardium.

Contributor's Morphologic Diagnosis: 1. Placenta: Marked, acute, multifocal to coalescing, necrotizing placentitis and vasculitis
2. Heart: Mild, multifocal, non-suppurative epicarditis and perivascular myocarditis

Contributor's Comment: Based upon the histological lesions and ancillary testing (FA, IHC and PCR), the abortion syndrome in these goats is blamed on BVDV infection.

Although BVDV most commonly infects cattle, the virus can also be found in other domesticated and wild ruminants.[1] It was hypothesized that these species may serve as a reservoir for the disease.[1] Seroprevalence to BVDV has also been recognized in many other domesticated and wild ruminant species, the geography of which continues to expand.[1,6,7,12]

Experimental and natural intraspecies transmission of ruminant pestiviruses has been confirmed.[8] This is also supported by the observation that higher seroprevalence rates of BVDV in goats occurs in regions/countries where goats are more likely to be co-mingled with cattle or wild ruminant species.[7] Goats are thus infected by exposure to other persistently infected species.

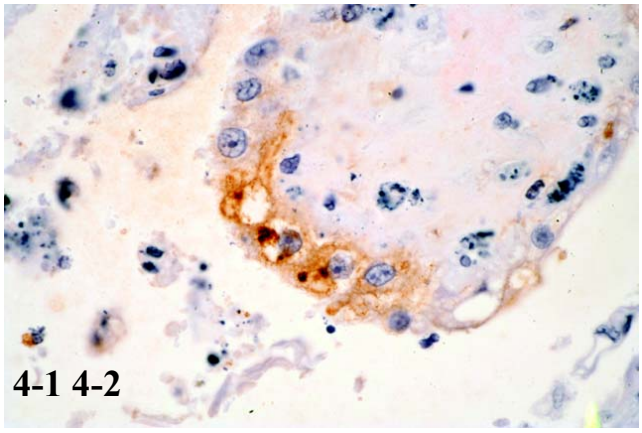
The reproductive consequences of BVDV infection in cattle have been reviewed.[5] BVDV disease in goats, reproductive failures or otherwise, have been less characterized. Experimental infection of adult goats with BVDV results in seroconversion with formation of neutralizing antibodies that persisted for up to 4 years.[9] Infection of kids similarly showed development of neutralizing antibodies accompanied by an impaired growth rate, but otherwise, no significant clinical symptoms.[10] Field outbreaks and experimental inoculation of BVDV in goats has produced reproductive failures.[8] These have been characterized by barreness, abortions, stillbirths or births of weak kids and birth of kids exhibiting clinical signs similar to border disease accompanied by histological lesions in the CNS.[8,11] Also, kids born to does experimentally infected with BVDV during gestation were highly contagious for goats and other susceptible ruminant species.[10]

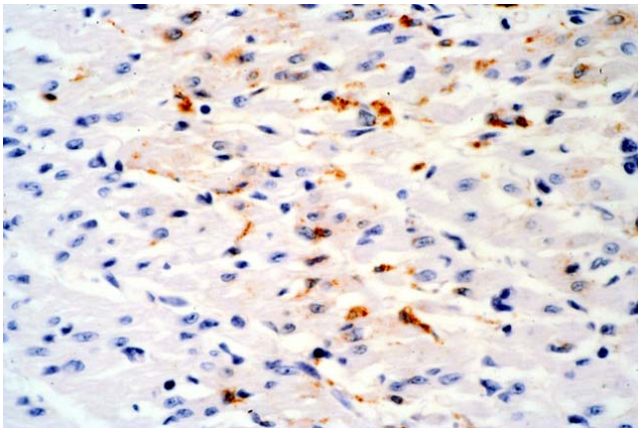
BVDV should be considered as a cause of abortion in goats or perinatal deaths in goats with or without CNS disease. This is especially true in geographic regions where goats and cattle (persistently infected BVDV cattle) are in close proximity.

AFIP Diagnosis: Placenta: Placentitis, necrotizing, multifocal, moderate, goat (*Capra hircus*), caprine.

Conference Comment: Diffuse autolysis obscured many histologic features and complicated the assessment of necrosis in this case. Pathologic lesions of BVD infection in fetal tissues are not considered characteristic and are rarely seen due to fetal autolysis.[13] Although the Bovine Viral Diarrhea Virus was not isolated from this particular animal, a PCR, **immunohistochemistry (figs. 4-1 and 4-2)**, and fluorescent antibody were all positive, making diagnosis highly probable. Convincing vasculitis was not present in the slides reviewed during the conference, but this is possibly due to slide variation.

BVDV is a RNA virus of the Pestivirus genus in the family Flaviviridae.[2] BVDV is known to naturally and subclinically infect pigs, sheep, goats, and several wild Afri-





4-1 Placenta, goat. Multifocally placental trophoblasts are immunoreactive for BVDV.
4-2 Heart, goat. Multifocally within the myocardium, cells are immunoreactive for BVDV.

Photomicrographs courtesy of the Department of Veterinary Pathobiology and the Oklahoma Animal Disease Diagnostic Laboratory, Oklahoma State University, Stillwater, OK

can ruminants.[6] Other pestiviruses in animals include Porcine Pestivirus (classical swine fever virus/hog cholera virus) in swine, and Ovine Pestivirus (Border disease virus) in sheep.[13]

BVDV infection may result in three main disease syndromes: embryonal/fetal disease (transplacental infection), mucosal disease (infection in immunotolerant animal), or bovine viral diarrhea (infection in immunocompetent animal).[13]

The outcomes of transplacental infection with BVDV are presented in table 4–1 below.

Mucosal disease results when an immunotolerant cow is infected with a cytopathic strain of BVDV either from an exogenous source, or through a mutation of the endogenous non-cytopathic strain.[13]

Postnatal infection with BVDV in an immunocompetent animal predominantly result in enteritis primarily of the ileum and proximal colon. Multifocal erosions may occur in oral and esophageal areas.[13]

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Table 4 –1. Outcomes of transplacental infections with BVDV[13]

Cytopathic Strain of BVDV	Prior to 100 days of gestation	embryo and fetuses resorption or expulsion days to months following infection.
	100 -150 days of gestation	teratogenic effects on fetal organs that may result in microencephaly, cerebellar hypoplasia, hydraencephaly, hydrocephalus, microphthalmia, thymic aplasia, hypotrichosis, alopecia, brachygnathism, growth retardation, and pulmonary hypoplasia
Non-Cytopathic Strain of BVDV	Prior to 100-125 days of gestation	Immunotolerance results in birth of persistently infected calf
	After 150 days of gestation	Fetuses mount relatively normal immune response and are born with circulating antibodies

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Notes: