REVIEW OF LITERATURE
II. REVIEW OF LITERATURE

2.1. Incidence of pyometra

Varying percentage of incidence of pyometra ranging from 0.7 to 9 % has been reported by Stone et al. (1988), Wheaton (1995) and Ewald (1961). However, Fakuda (2001) reported an incidence of pyometra in up to 15.2% in a beagle colony and Egenvell et al. (2001) and Hagman (2004) stated that 25% of bitches developed pyometra by 10 years of age.

2.1.1. Age and Parity

Dow (1958) reported that bitches aged between 6 to 16 years were commonly affected with pyometra where as Krook et al. (1960) reported the average age of dogs affected with pyometra as 8.44 ± 0.12 yrs. Maximum incidence being in the age group of 7 to 12 years and mean age around 10 years. Borresen (1979) reported the average age of 119 bitches affected with pyometra as 7.8 years. It was also stated that nulliparity, abnormal estrous cycles and psuedopregnancy increased the risk of pyometra.

In a study of 30 cases reported by Frost (1963) all cases were over seven years of age. In studies of Fidler et al., (1966) the average age of 68 bitches with pyometra was 7.8 years and was close to that of 245 bitches in the control group (7.7 years).

Dirr (1975) also opined that the incidence of pyometra increased with age and in young bitches after estrogen treatment.
Nelson and Feldman (1986) opined that pyometra was primarily a disease of older bitches and its incidence was also found to increase in younger bitches due to increasing use of estrogen and progesterone for treatment of mismating.

Stone et al. (1988) reported that the mean age of 27 dogs diagnosed to be suffering from pyometra as 6.4± 3.6 years in addition a retrospective study involving case records of 100 other dogs with pyometra revealed the mean age as 8.7± 3.00 years.

In a review of medical record of 73 dogs diagnosed as having pyometra, Wheaton et al. (1989) reported the mean age of dogs with pyometra as 7.9 years. The study also revealed that 11 percent (8 bitches) developed pyometra within 3 years of age. Six of the younger bitches in this study had received estrogen within previous six months.

Dolezel (1989) reported that the majority of 24 bitches aged four to twelve years old of various breeds were diagnosed to be suffering from pyometra were nulliparous.

A mean age of seven years (range 1-14 years) was reported by Sevelius et al., (1990) who investigated 103 bitches with a tentative diagnosis of pyometra based on history, physical examination, radiographs and biochemical testing, further a history of whelping was available in only 14 bitches.

Blendinger and Dostedt (1991) in an evaluation of 302 clinical cases of canine pyometra found that 28.5 percent of bitches were aged five to seven years, 42.9 percent were between eight to ten years of age and 15.9 percent were 11 to 13 years old.
Fanti et al (1992) surveyed the clinical records of 942 cases of canine pyometra and reported that the age of the affected bitches ranged between 7 to 10 years.

Okubo et al. (1995) reported that the ages of 50 dogs studied with pyometra ranged from 4 to 17 years with a mean of 9.8 years, 82 percent had not puppies and 9 were nulliparous.

Wheaton et al. (1995) reported that pyometra occurred most frequently in bitches over six years of age, average age being ten years and therefore it was primarily a disease of middle aged cycling bitches.

In a case controlled study of 953 dogs with pyometra and 10660 unmatched control dogs, Niskanen and Thursfield (1998) investigated the relationship between the age, parity, hormonal therapy, breed and pyometra on Finnish dogs. Cases were reported in animals between 9 months and 18 years of age, with a median age at diagnosis of 9 years. Nulliparous bitches had a moderately higher risk of developing pyometra than primparous and multiparous animals.

Dhaliwal et al. (1998) reported that the mean age of 34 bitches of 18 pure breeds or cross breeds suffering from cystic endometrial hyperplasia as 8.5±2.8 years with a range of 1 to 14 years. Further, with the exception of two bitches which had whelped at least three years before, the rest were all nulliparous.

In another study Dhaliwal et al. (1999) reported that the mean age of 13 bitches affected with pyometra was 8.8 years and ranged from 1 to 14 years.
Chastain et al., (1999) has opined that incidence of pyometra is more in nulliparous bitches and bitches greater than 4 years of age.

Johnston et al. (2001) observed that pyometra typically affects mature bitches that have undergone repeated estrus cycling, with a reported mean age of 7.25 years.

Feldmen and Nelson (2004) reported that in a group of 192 bitches that were treated medically for pyometra, the mean age was 2.4 years.

Smith (2006) stated that the average age of onset of pyometra was 9.36± 0.35 years.

Kashinath et al. (2009) screened 446 dogs out of them 18 (4.03%). were suffering from pyometra. The higher incidence of pyometra was found in 10(55.55%) bitches which were in the age group of 6-9 years and out of 18 bitches 14 (77.77 %) were nulliparous bitches suggesting that the incidence of pyometra was very high in middle aged and nulliparous bitches.

2.1.2. Breed

Predisposition to pyometra in certain breeds has occasionally reported in the literature. Ewald (1961) recorded a relatively high breed specific prevalence in Coollie and Belgium spherfered dog and low prevalence in Daschound and poodle.

Krook et al. (1960) compared 487 cases of pyometra with a sample of general dog population in Sweden and stated that Chow-chow, French bull dog, Coollie, Pointer, Great Dane, Rottweiller, Skye terrier, Saint Bernad and Sweedish Hound had a
statistically significant predisposition to the development of pyometra where as Boxer, Fox terrier and Poodle had decreased risks. However in above studies base population was not considered.

Eganvall et al. (2001) reported that the occurrence of pyometra differed with age, breed, and geographic location. The risk of developing pyometra was increased (identified using multivariate models) in rough Collies, Rottweilers, Cavalier King Charles Spaniels, Golden Retrievers, Bernese Mountain Dogs, and English Cocker Spaniels compared with baseline (all other breeds, including mixed breed dogs). Breeds with a low risk of developing the disease were Drevors, German Shepherd Dogs, Miniature Dachshunds, Dachshunds (normal size), and Swedish Hounds. Survival rates indicate that on an average 23-24% of the bitches in the databases will have experienced pyometra by 10 years of age. In the studied breeds, this proportion ranged between 10 and 54%. Pyometra is a clinically relevant problem in intact bitches, and differences related to breed and age should be taken into account in studies of this disease.

Hagman (2004) in his study, by considering the base population in Sweden has recorded high risk of predisposition to pyometra in Collie (rough-haired), Rottweiler, Bernese, Mountain Dog, Cavalier King, and Golden Retriver. Lower risk of developing pyometra has been reported in sweedish hound, Dachshund and German Shefered dogs.

Smith (2006) reported that some of the breeds predisposed to pyometra include the Rottweiler, Saint Bernard, Chow Chow, Golden Retriever, Miniature Schnauzer, Irish Terrier, Airedale Terrier, Cavalier King Charles Spaniel, Rough Collie, and Bernese Mountain dog
Kashinath et al. (2009) in his study of breed wise incidence of pyometra found that majority of the cases were in Spitz, followed by German Shepherd, Pomeranian, Cocker Spaniel, Doberman and Lhasa Apso. It was also noted that smaller breeds were affected more than the large breeds of dogs.

Niskanen and Thurshfield (1998) failed to demonstrate breed specific risk of development of pyometra and concluded that there was no genetic component as an etiological factor in development of pyometra.

Sevelius et al. (1990) opined that the difference in the incidence of pyometra in different breeds could be consequent to population of a particular breed in different geographical location rather than any breed predisposition.

2.1.3. Pyometra in relation to previous estrous cycle

Pyometra is considered a disease of diestrus, although some anestrous bitches can be diagnosed with pyometra Noakes et al. (2001).

Verstegen et al. (2008) reported approximately one-third of the total pyometra cases in the anestrous cases and it was uncertain whether the animals described were observed after the end of a normal luteal phase or whether there had been, at least in some of the animals, a premature shortening of the luteal phase. This premature shortening of the luteal phase may be induced by production of endogenous prostaglandin in response to the uterine inflammation, as observed in other species.

The relevance of the association between pyometra and diestrus was demonstrated by Lesboyries and Berthelon (1936) and reported that ovariectomy without hysterectomy
in bitches with pyometra was followed by clinical cure in 5 to 7 days. Similarly, in a retrospective study of more than 10 years, Janssens and Janssens (1991) confirmed the evidence of the role of ovarian steroids in the pathogenesis of the disease, as bilaterally ovariectomized dogs never developed pyometra.

Sokolowski (1992) stated that pyometra occurred most frequently during dioestrous or met oestrous in mature intact bitches and its development depended on estrogen priming of uterus during proestrus and estrus.

Okubo et al. (1995) while investigating the pathogenesis of pyometra in 50 bitches reported the consistent finding of a corpus luteum on the ovaries which coincided with post estrus period of 1 to 2 weeks as notified by their owners.

Blendinger et al. (1997) concluded that pyometra may occur at any stage of reproductive cycle and most animals were in dioestrus phase.

Dhaliwal et al. (1999) reported that onset of clinical signs of pyometra occurred between 1 to 28 weeks post pro oestrous.

Smith (2006) reported that most bitches with pyometra were presented to the clinician from 4 weeks to 4 months after estrus. The owners of older bitches may not report any recent estrus activity, assuming that their bitch has undergone ‘menopause’.

Pretzer (2008) has reported mean interval of 5.7 weeks from previous estrous to onset of pyometra and most bitches present with pyometra within 12 weeks after the onset of the previous estrus.
2.2. The Pathogenesis of Canine Pyometra

Canine pyometra is a disease affecting the adult intact bitch causing a variety of clinical signs of both genital and systemic disease. The concept of cystic endometrial hyperplasia (CEH) - Pyometra, was introduced by Dow (1957) and stated that hormonal changes lead to CEH which predisposes the uterus to secondary infection, leading to pyometra. Before Dow’s investigations the condition had been described under a variety of names.

2.2.1. Hormonal component

Traditional theories suggest that hormonal changes render the uterus susceptible for infection. This understanding was originally based on work by Teunissen (1952) and Dow (1957, 1958, 1959a, 1959b), who investigated the importance of estrogen and progesterone in the development of endometritis. They found that the cystic and inflammatory changes of the uterus associated with endometritis could be reproduced by injections of progesterone. Estrogen alone seemed to play a less important role, but appeared to enhance the endometrial response to progesterone and exogenous estrogen administration, used to terminate pregnancy, has been reported to increase the risk of pyometra (Bowen et al. 1985).

Cox (1970) opined that Progesterone stimulates endometrial glandular secretion and suppresses contractions of the uterus and thus, creates an intrauterine environment predisposing to bacterial growth
Several authors investigated whether prolonged or excessive progesterone production would be responsible for the development of pyometra, but failed to show such disturbances (Christie et al. 1972; Hadley, 1975; Chauffaux & Thibier, 1978; Austad et al. 1979).

De Bosschere et al. (2001) stated that in the common dogma, CEH-associated degenerative changes are caused by the presence of numerous crypts and cysts where bacteria can proliferate and reduce the local immunity within the uterine tissues (cystic distention of glands, fibrosis, etc.) were suggested to provide opportune conditions for establishment of uterine infections and this association is reflected in the naming of the condition ‘Cystic Endometrium-Pyometra Complex’.

However, while common dogma dictates that CEH usually precedes pyometra, it is also obvious that CEH does not inevitably progress to pyometra in all bitches. This is evident, as all dogs develop CEH with age, whereas only some of them will develop a pyometra. Similarly, pyometra can develop in young animals which do not have prior clinical or pathological evidence of CEH.

Recent work by Nomura and Nishida (1998) and the clinical observations of Koguchi et al. (1995) have suggested that the classical sequence of progesterone leading to CEH and subsequently CEH to pyometra may not be correct, and that the sequence may in fact be reversed with bacteria being the initiating factor. A subtle (sub-clinical) uterine infection or endometrial irritation by foreign bodies may first occur at the end of estrus or during the first half of diestrus, providing the stimulus for an excessive endometrial hypertrophy and hyperplasia, similar to what is observed at the time of
implantation (trophoblastic or decidual reaction). The resulting increase in endometrial
glandular proliferation and luminal epithelial cellular secretions can initiate the
development of a pyometra or a mucometra, depending on whether the promoter is of
bacterial origin or not. This hypothesis is supported by results of experiments done in
dogs which have shown that, at specific periods during the luteal phase of the cycle, a
variety of physical, biological, and chemical substances will cause the endometrium to
proliferate.

More recently, the possibility of an exaggerated endometrial response to
progesterone and/or estrogen has been investigated (Dhaliwal et al. 1999 and De
Bosschere et al. 2002). Presently, there is no conclusive evidence for changes in these
hormone receptors being responsible for the development of pyometra. Interestingly
enough, De Bosschere et al. (2001) found that the expression of estrogen and
progesterone receptors differed significantly between uteri of bitches with CEH and uteri
of bitches with pyometra, and suggested that these differences indicate different
pathogenesis of the two conditions. The expression of androgen receptors in the uterus
was found to be significantly decreased in bitches with pyometra as compared to healthy
bitches (Sauerwein et al. 1998).

2.2.2 Bacteriological component

In the past, investigations of the bacteria associated with canine pyometra have
been few. However, many authors have reported a predominance of Escherichia coli in
canine pyometra samples (Asheim, 1965; Renton et al. 1971; Grindlay et al. 1973;
Sandholm et al. (1975; Kivisto et al. 1977; Vandeplasch et al. 1991 and Dhaliwal et al. 1998).

Sandholm et al. (1975) found that E. coli adheres to receptors in the progesterone stimulated canine endometrium which might be one explanation for the observed predominance of these bacteria.

Arora (2007) found that certain serotypes of E. coli, e.g. 02, 04, 06, and 075, were more commonly associated with pyometra than others and the presence of Cytotoxin Necrotizing Factor (CNF) was associated with more severe endometrial changes.

Dhaliwal et al. (1998) recorded E. coli serotype 032 and 04 as the most commonly prevalent bacteria. In addition, they found CNF in 44 percent serotypes associated with pyometra.

Dow (1959b) suggested that the route of infection of the uterus was hematogenic or lymphogenic as well as ascending. The ascending route was not supported by the work of Meyers-Wallen et al. (1986), who observed that the type of bacteria isolated from the vagina did not necessarily represent the bacterial species isolated from the uterus in pyometra.

Sandholm et al. (1975) showed that cystitis was commonly associated with canine pyometra, and that the E. coli bacteria isolated from the urinary bladder and the isolate from the uterus showed many similarities. It was suggested that the urinary tract may serve as a bacterial reservoir and bacteria ascend into the uterus during a susceptible stage in the estrous cycle.
2.3. Clinical signs.

Common clinical signs of pyometra in bitches are not limited to the genital tract, e.g. vaginal discharge, but include systemic signs such as vomiting, inappetence, polyuria, polydipsia and lethargy. (Dow, 1957; Borresen, 1979; Nelson and Feldman, 1986 and Stone et al. 1988). It has been suggested that the clinical signs are more severe in cases where the cervical canal is occluded (Dow, 1958 and Borresen, 1975). However, the cervix may spontaneously open or close during the disease causing intermittent vaginal discharge or a sudden deterioration in the clinical status of the bitch (Studdert, 1971).

Low (1954) stated that a pendulous distended abdomen assuming a pear shaped was a consistent finding in canine pyometra and expressed the limitation of recording body temperature in diagnosis as it might be sub normal or moderately elevated.

Whitney (1956) observed that temperature recorded in cases of canine pyometra was of prognostic value due to the fact that thermal reaction was extremely variable and abnormal temperature was of significance.

A detailed study on cystic hyperplasia-pyometra complex in the bitch was made by Dow (1958). He classified cystic endometrial hyperplasia-pyometra complex based on clinical, histological and pathological examination into four groups. In Group I cystic hyperplasia-pyometra complex was characterized by cystic glandular hyperplasia of the endometrium without any superimposed inflammatory reaction. In 53 cases, signs of genital disease was absent in every case though a slight mucoid discharge was apparent in a few of the cases. In Group II there were 22 cases in which the characteristic lesion was
diffuse plasma cell infiltration of the endometrium superimposed with cystic glandular
hyperplasia. In most cases, the only symptoms of note at presentation was a vulval
discharge of mucoid fluid varying from clear to dull red in colour. Many of the animals
also showed evidence of malaise and anorexia. In Group III the characteristic lesion
found was an acute inflammatory reaction in the endometrium exhibiting variable degree
of cystic glandular hyperplasia in bitches. Vulval edema was almost a consistent finding.
Vulval discharge was visible in only 52 out of 76 patients. In this group, listlessness and
some degree of inappetence was apparent in all but four cases. Increased thirst had been
noted in 45 cases and 34 had vomited at some time during the course of illness. Diarrhea
was an infrequent symptom being present in only five cases. The body temperature was
within normal limits in 15 cases, higher body temperature of 103 to 105 °F was found in
11 cases and sub normal temperature in 6 other cases.

The Group IV consisted 21 cases in which the cervix was either completely
closed or partially open. The cases with closed cervix were seriously ill and showed gross
abdominal distension, polydipsia, frequent vomiting and marked depression. While the
only symptoms observed in open cases were vulval discharge and partial anorexia.

Asheim (1963) observed that animals with pyometra exhibited signs of
polydipsia, polyuria and less urine concentrating ability.

Fowler (1964) reported the clinical signs of pyometra as excessive bleeding
during proestrous, polyuria and polydipsia at post estrum and pseudo pregnancy with
marked mammary development vaginal hyperplasia or prolapse and vulval edema. He
also observed anorexia, emesis and elevated temperature.
Asheim (1965) reported that bitches affected with pyometra exhibited poor general conditional emesis, anorexia, diarrhea and vulval discharge with body temperature being normal or slightly elevated.

Roberts (1971) stated that body temperature in acute cases of pyometra may be elevated, while it could be normal in chronic and subnormal in severely toxæmic bitches.

Smith (1974) reported the important signs of pyometra as listlessness, weakness of hind limb, discomfort, abdominal fullness, occasional vomiting and reluctance to climb the stairs. Initially the body temperature was 1-2°C less than normal in early toxæmia and became abnormal as toxæmia progressed.

Arthur (1996) reported that closed cases showed severe ill health and abdominal distension. In early stages of closed pyometra the temperature ranged from 103-105°F and became subnormal in later stages. Open cases on the other hand exhibited normal body temperature.

Hardy and Osborne (1974) reported that, vomiting, polyuria, polydipsia, nocturia, diarrhea and vulval discharge were common signs in pyometra.

Borresen (1979) reported that 80% of 119 pyometra bitches showed clinical signs of dehydration and hypoperfusion although only about one third of the cases examined were weak and in poor condition.

Nomura et al. (1994) studied 207 bitches with pyometra and reported that 73.2% were dull and 78.5% anorectic, 70% had polydipsia and 80% had vaginal discharge.
Vomiting was recorded in some cases but the body temperature remained normal in all cases.

Huszenicza et al. (1985a) found that pyometra cases of bitches examined prior to ovario- hysterectomy exhibited polyurea, polydypsia and more variable clinical signs such as anorexia, weakness, fever, dehydration, vomiting, anaemia and icterus.

Nelson and Feldmen (1986) stated that clinical signs of pyometra and abnormalities on physical examination are dependent on the patency of the cervix and ability of the client to recognize the problem quickly.

In studies on renal dysfunction in dogs with pyometra Stone et al. (1988) reported that of 27 cases investigated, 70% had history of diarrhea, vomiting, anorexia and/or lethargy.

In a review of medical records of 80 cases of canine pyometra Wheaton et al. (1989) reported the clinical signs as depression (79%), anorexia (79%), vaginal discharge (88%), polydipsia (63%), polyuria (38%), vomiting (33%), dehydration (15%), diarrhea (26%), pyrexia (43%) and palpable uterus (40%).

The clinical signs and medical history in 83 cases of pyometra was reported by Sevelius et al. (1990). General appearance was normal in only 16% of the patients. It was slightly depressed in 26 patients (31%), moderately depressed in 38 animals (46%) and severely depressed in four other patients (5%). The general appearance was not recorded in 2% of the cases. In 30% of the cases the temperature was significantly elevated and in
18% of the bitches vaginal discharge and polydipsia was common signs nevertheless 30% of the patients lacked polydipsia.

Sokolowski (1992) reported that the clinical signs such as depression, pyrexia, anorexia, polyuria, polydipsia depended on the existence of endotoxameia, patency of cervix and general health.

Leopold (1993) reported that affected bitches exhibited polyuria, polydipsia and vomiting.

Memom and Mickelsen (1993) reported the clinical signs of closed pyometra in a four year old Golden Retriever bitch. Medical history included anorexia, loss of weight and lethargy for about ten days. Polydipsia but not polyuria had been noticed by the owner for a week. The bitch has vomited a few times and a intermittent fever as high as 39.6°C has been recorded. The bitch had signs of depression and pain and walked with stiff gait and was somewhat resistant to palpation.

In studies of Gandotra et al. (1994) out of 24 cases of pyometra, four had closed pyometra. Almost all bitches were off feed, dull and depressed with temperature ranging from 103-104°F.

Pawde and Kumar (1996) reported that affected bitches showed serosanguineous or mucopurulent discharge from the vulva, lethargy, inappetence, polyuria, polydipsia and vomiting.
Johnson (1995) reported the typical signs of pyometra as vulval discharge, lethargy, vomiting, polyuria, polydipsia, weight loss abdominal distension and dehydration.

Dhaliwal et al. (1998) reported that 30 (88.2%) of the bitches diagnosed as cases of pyometra had a vulval discharge which was indicative of an open cervix and only three had a closed cervix.

Feldman and Nelson (2004) were of the opinion that clinical signs of pyometra in dogs depend primarily on whether the cervix is patent enough to allow drainage of purulent fluid. The most common clinical finding in bitches with open-cervix pyometra is a mal odorous, sanguinous to mucopurulent vaginal discharge.

Verstegen et al. (2008) in their review were of the opinion that the most obvious clinical sign in pyometra is vaginal discharge, which may vary from serosanguinous to mucopurulent. In some bitches, the amount of discharge is minimal and fastidious grooming by the bitch makes that discharge difficult to detect. In other cases, signs of vaginal discharge may not be externally apparent, but vaginal smears and vaginoscopic examination reveal the presence of uterine exudate in the cranial vagina. The amount of vaginal discharge is also partially dependent on the degree of cervical patency.

2.4. Hematology

The systemic effects of pyometra in bitch are reflected by several hematological parameters. The most characteristic alteration is an inflammatory leukogram with marked
elevation of the total white blood cell count (WBC) and usually a regenerative left shift in the differentiated WBC count (Dow 1957, Sandholm et al.1975, De Shepper et al.1986, Stone et al.1988, Wheaton et al.1989 and Sevelius et al.1990).

The average leukocyte count in studies of Rechfeld (1954) were found to be 44480 per cmm ranging from 40800 to 166400. It was concluded that the neutrophils were increased, especially the immature forms.

Dow (1958) reported that in 22 cases of cystic hyperplasia pyometra complex in bitches in which the characteristic lesion was a diffuse plasma cell infiltration of the endometrium. The total leucocyte count ranged from 12000-15000 cells/mm$^3$. There was little change in the differential leucocyte count and without evidence of anaemia. In 76 other cases in which the characteristic lesion was an acute inflammatory reaction in the endometrium, the leukocyte count ranged from 19000-145000 cells/mm$^3$. The leukocyte count in closed cases were always over 40000 cells/mm$^3$. The leukocytosis was always due to an increase in neutrophils of which up to 35% were immature forms. The number of cases with a high leucocyte count abnormalities were noted in the morphology of neutrophils. However, there was no evidence of anaemia in any of the cases examined. In 21 cases of chronic endometritis the leukocyte count ranged from 16000-28000 cells/mm$^3$ in open cases and from 31000-84000 cells/mm$^3$ in closed cases. The leukocytosis was largely due to neutrophils. Only one animal showed evidence of anaemia.
Walker (1965) stressed the importance of blood picture as diagnostic aid in animals with pyometra the reaction being extreme neutrophilia with a leucocytosis ranging from 14000 to 180000 cells/mm³.

Whitney (1967) reported on an average total leukocyte count of 20000 to 80000 cells/mm³ with neutrophilia and relative lymphopenia.

Renton et al. (1971) observed that in 90% of bitches with pyometra a leukocytosis was present. In 29 open pyometra cases the total leukocyte count ranged from 8000-79000 with mean of 32500 cells/mm³. The mean of total leukocyte count was slightly greater than 36600 cells/mm³ in closed type.

Roberts (1971) reported leucocytosis in pyometra bitches to over 5000 cells/mm³ and that most of the leukocytes were neutrophils.

Wasecki (1973) described the differences in leucocergy (leucocyte aggregation) red and white cell count, before and ten days after surgery in pyometra. There was a highly significant difference between leucocergy at estrous and pyometra, the degree of aggregation being independent of leucocytosis.

Hardy and Osborne (1974) attributed the anaemia in dogs affected with pyometra either to toxic depression of bone marrow and or loss of red cells into the uterine lumen. Further they reported that the total leucocytosis was higher in close pyometra than that in open pyometra and toxic states led to a non regenerative normocytic, normochromic anemia with a degenerative shift to left.
Sandholm et al. (1975) reported the mean leucocytic count in bitches affected with pyometra as 33000±15900 cells/mm³. Which was about three times the value for clinically normal dogs.

Kivisto et al. (1977) reported that leucocyte count was the useful aid for diagnosis of pyometra along with estimation of gammaglobulin in the blood.

Buckner (1979) reported that WBC count ranged from 18,000 to 1,00,000 per cmm. A regenerative left shift of leucocyte might develop depending on the stage of disease and severity of the bacterial infection.

Borrensen (1980) reported very high leukocyte count in severe cases which developed microcytic hypochromic anaemia consequent to diapedesis of erythrocytes into uterine lumen or a shortened life span of circulating erythrocytes associated with iron deficiency.

Greene (2006) reported that non regenerative microcytic anaemia with a concurrent blood loss of chronic nature. The number of immature forms when exceeds the segmented neutrophils indicates the severity of inflammation and suppurative nature of the disease.

Childers and Bazar (1985) studied a case of atypical pyometra and observed normal WBC count, degenerative left shift and normocytic normochromic anaemia.

Huszencicza et al. (1985b) reported that the values of haemoglobin, haematocrit and erythrocyte count were suggestive of anaemia despite concurrent dehydration in 54
bitches affected with pyometra in comparison to 23 bitches (asymptomatic). Leucocytosis with a relative increase in neutrophyls and decreased lymphocytes also observed with no significant difference between open and closed pyometra.

De Seepper et al. (1986) stated that pyometra was accompanied by normochromic anemia, neutrophilia and manocytosis, indicative of subacute inflammation and endotoxemia.

De Seepper et al. (1987) presented type of anaemia I relation to the total leucocyte count in 112 dogs with pyometra. Thirty one per cent showed a normal blood cell picture without anaemia, 51 percent exhibited a non regenerative normocytic normochromic anaemia while another 12% showed a non regenerative mycocytic hypochromic anaemia.

Stone et al. (1988) studied the hematological parameters in 27 typical cases of canine pyometra and compared with100 other cases studied retrospectively. In prospective studies the mean leucocyte count was 35073 ± 20700 cells/mm and ranged from 10700 to 94000 cells/mm. All dogs with leucocyte count greater than 50000 were aged more than 7 years. The mean PCV was 38 ± 6 percent and ranged from 21 to 48 %. 22 of pyometra cases studied retrospectively were considered anaemic with PCV less than 35 percent.

Wheaton et al, (1989) reportd that in 25 percent of affected bitches there was evidence of marked anaemia (PCV more than 35%), leucocytosis(WBC more than17,000 cells/µl) was observed in 68% of cases while leucopenia (WBC less than 5000 cells /µl) was reported in 8% of affected animals. A differential count revealed neutrophilla in 67%, lymphocytosis in 9% of cases, monocytosis in 57% of cases and eosinophilia in
16% of affected animals. Eighty seven of affected animals exhibited a shift to left with band cells.

Sevelius et al. (1990) observed that 49% of the bitches with pyometra in their study had a normal leukocyte count. Total leucocyte counts were less than 6000 cells/mm³ in 1% of the cases, between 6000 to 17000 in 33% of cases, 17000 to 30000 in 19% of cases and greater than 30000 in 29% of cases. They also reported differential count in 50 cases of pyometra. Fourteen (28%) showed normal leukocyte count and normal differentials, while leucocytosis with shift to left was found in 12 (24%) and 18 (36%) developed leucocytosis with normal differentials. A normal leukocyte count with a degenerative shift to left was found in 6 (12%). Normal haematocrit values (PCV more than 39%) were obtained in 36% of cases. Haematocrit values were between 30 to 39% in 34% of cases and less than 30% in another 16% of cases. They stated that 19% of pyometra cases were markedly anaemic.

Schalm et al. (1991) described haemograms of 50 bitches with pyometra. Some cases were suggestive of regenerative left shift whilst the others a degenerative shift to left. They further stated that the lymphocyte count was depressed and monocyte count elevated in chronic cases and moderate normocytic normochromic anaemia. Packed cell volume (PCV) range of 36 to 40% reflects the existence of border line anaemia being masked by dehydration.

Sokolowski (1992) reported that white cell count in excess of 30000 cells/mm³ was a common finding in bitches with pyometra.
Memon and Mickelsen (1993) in a four year old Golden Retriever bitch with a closed cervix pyometra reported neutrophilic leucocytosis with left shift, monocytosis and thrombocytopenia.

Haematological studies in 24 cases of pyometra by Gandotra et al. (1994) revealed a considerable increase in the total white blood cell count and a marked neutrophilia (76.36±1.45%). The neutrophilic count was even much more in closed pyometra (86.25±0.46) lymphocytes, monocytes and eosinophils were within normal limits. There was also a slight decrease in haemoglobin concentration (10.95±0.37g/dl).

Johnson (1996) reported leucocytosis, neutrophilia, a left shift to immature forms and a substantial monocytosis and total leukocyte count ranged from 25000 to 100000 cells/mm³. Mild to moderate normocytic, normochromic non regenerative anaemia was also reported.

Kochhar et al. (1996) reported marked leucocytosis (32000 cells/mm³) with absolute neutrophilla (79%) and hypo haemoglobin level (8.3g %) were common findings of pyometra in bitches.

A higher total leukocyte count, a more marked shift to left in differential leukocyte count and toxic degeneration of neutrophils were reported in 60 bitches with clinical diagnosis of pyometra by Fransson et al. (1997).

Kaymaz et al. (1999) found that in 18 bitches with a clinical diagnosis of Cystic endometrial hyperplasia-pyometra complex the leucocyte count were increased and in 93.3% of cases the mean corpuscular volume was higher.
Kashinath et al. (2009) in their study reported leucocytosis along with absolute neutrophilia, lymphopenia and monocytosis.

2.5. Haemobiochemistry

Dow (1958) reported the plasma urea nitrogen levels in 76 patients with an acute inflammatory reaction of the endometrium. The plasma urea levels in the affected animals ranged from 2-180 mg/dl with a mean of 38±11 mg/dl. Levels over 65mg/dl warranted a guarded prognosis and were poor surgical risks.

Groulade and Groulade (1959) found high total plasma proteins values in bitches with metritis and pyometra.

Russe and Lewis (1959) used blood urea nitrogen values for prognosis of pyometra and stated that recovery chances were best when blood urea nitrogen values were below normal and prognosis was poor when blood urea nitrogen values above 90 mg percent.

Asheim (1965) reported that toxic pyometra cases with associated nephritis had elevated blood urea nitrogen, hyperproteinemia, hypoalbuminemia and hyperglobulinemia.

Renton et al. (1971) reported blood urea levels in open and closed type of pyometra. In 21 open cases the blood urea nitrogen levels ranged from 11 to 310 mg/dl, while it was between 16 and 240 mg/dl, in closed pyometra. Further they reported that 24 percent in open type had abnormal levels, nevertheless blood urea nitrogen levels in open (141mg/100 ml) and closed (125 mg /100 ml) cases were non significant.
Roberts (1971) reported nephrities with an elevated blood urea nitrogen levels may be noted occasionally in pyometra affected bitches.

Hardy and Osborne (1974) observed mild to marked hyper protenimia and 17.7 percent of 79 cases examined had an elevated blood urea nitrogen values and elevated creatinine values were also found.

Sandholm et al. (1975) reported that gamma globulin content (26.3±3.9 g/dl) was much higher in bitches affected with pyometra than that of clinically normal dogs.

Buckner (1979) found levels of blood urea nitrogen and creatinin to be elevated in dogs with pyometra.

Cox and Joshua (1979) reported that a light increase in blood urea nitrogen values (up to 100 mg %) as indicated by irreversible renal damage. An increase in total serum protein was also reported.

Borrensen (1980) correlated results of Haemoglobin, Packed cell volume, serum protein, blood urea nitrogen , creatinin , cholesterol and some serum enzymes, in bitches with clinical pyometra, with the degree of polydypsia, anaemia and dehydration in the same patients and concluded that the pyometra syndrome includes a liver derangement in addition to bone marrow suppression and renal function.

Hardy (1980) reported moderate to marked hyper protenemia, elevated blood urea nitrogen values and elevated creatinine values in bitches with pyometra.
De Schepper et al. (1986) observed that total blood protein values were increased in half the cases and aspirate amino transaminase, alamine amino transaminase and Lactate amino dehydrogenase were also increased in half of the 96 cases studied.

Capiau et al. (1987) examined a group of 174 bitches with pyometra and reported a significant increase in serum amylase, alkaline phasphatase, aspirate amino transaminase and Lactate dehydrogenase.

Stock and De Schepper (1987) reported that in 83 bitches with cystic hyperplasia pyometra complex serum values showed a decreased alanine amino transaminase and increased aspirate amino transaminase and increased Aspartate amino transaminase and an increase in Aspartate amino transaminase and Alanine amino transaminase ratio, these values were more pronounced in 62 clinically ill bitches. They concluded that the levels of Alanine amino transaminase determination are not influenced by serum components or drugs used.

De Schepper et al. (1987) examined 96 bitches with cystic hyperplasia-pyometra complex, and reported a significant Aspartate amino transaminase & a decrease of Alanine amino transaminase, leading to a significant increase in Aspartate amino transaminase to Alanine amino transaminase ratio, in those bitches which had total leucocyte count greater than 20 x 10 cells and had typical signs of pyometra like depression, vomiting, anorexia, polydypsia, polyurea, radiographically distended uterus with or without vaginal discharges.
Stone et al. (1988) examined the renal function of 27 dogs with pyometra and reported that 26% were azotemic (serum creatinine 1.2 mg/dl) and serum urea nitrogen value greater than 27 mg/dl before surgery. All dogs with azotemia exhibited clinical signs of vomiting, diarrhea, dehydration, anorexia and weakness. The mean serum creatinine values of dogs with azotemia was 2.1 ± 1.2 mg/dl and mean serum urea nitrogen value was 45.1 ± 26.7 mg/dl. When dogs with azotemia were excluded from the calculation, group means for serum creatinine (0.69 ± 0.2 mg/dl) and Serum urea nitrogen (11.3 ± 4.7 mg/dl) values were normal limits.

Colombo et al. (1988) in studies involving 73 bitches found values of gamma glutamyl transaminase, blood urea nitrogen and creatinine to be significantly elevated in a group of bitches with initial endometrial hyperplasia and pyometra with chronic endometritis and closed cervix, while that indicators of liver function viz., asparatate amino transaminase, alanine amino transaminase were elevated but to a lesser extent.

Schalm et al. (1991) reported slight to moderate hyper proteinaemia in excess of 8.0 g/dl accompanied by elevated blood urea nitrogen levels in bitches with pyometra.

Jensen et al. (1992) in a study involving 80 bitches with pyometra concluded that blood urea concentrations had no diagnostic value for cystic endometrial hyperplasia-pyometra syndrome.

Leopold (1993) reported that blood urea and creatinine values were elevated in bitches with pyometra.
Wheaton et al. (1989) noted that in 12% of affected dogs the serum creatinine levels were higher than 1.5 mg/dl. In 27% of affected dogs the serum urea nitrogen levels were over 20 mg/dl, while the serum alanine transaminase concentration was more than 60u/l in 22% of the cases.

Sevelius et al. (1990) reported the blood urea nitrogen levels did not differ significantly from the normal values in 83 cases of pyometra in bitches.

Kaymaz et al. (1999) analyzed blood serum profiles of 18 bitches with cystic endometrial hyperplasia-pyometra complex and reported an increase in Alanine amino transaminase, lactate dehydrogenase and triglycerides and decreased creatinine, urea and serum albumin.

Verstegen et al. (2008) in their review opined that the most consistent clinical blood chemistry finding is elevated serum alkaline phosphatase, present in approximately 50 –75% of cases, occasionally serum alanine aminotransferase concentrations may also be mildly elevated. These changes reflect hepato-cellular damage in response to toxemia, or diminished hepatic circulation due to dehydration. Hyperproteinemia may develop in response to dehydration, and hyperglobulinemia reflects the chronic antigenic stimulation present with this disease and also mentioned renal dysfunction is a feature of canine pyometra. Serum blood urea nitrogen and creatinine concentrations are not usually elevated, unless pre-renal azotemia develops as a consequence of dehydration. Azotemia is generally associated with more severe clinical signs. Even in non-azotemic rehydrated patients, glomerular filtration is usually decreased, indicating that some factors associated with the disease affect renal perfusion either in the presence or absence of azotemia.
2.6. Ultrasonographic Studies

The diagnosis of pyometra is best made with the aid of ultrasonography (Fayrer-Hosken et al. 1991) and findings typically include an enlarged uterus with convoluted, tubular horns filled with anechoic to hypoechoic fluid (Voges and Neuwirth 1996). The luminal contents are usually homogenous, but the contents may also be echodense with slow, swirling patterns (Nyland and Mattoon, 2002).

Wheaton et al. (1989) examined 7 animals suspected to be affected with pyometra and reported that in all cases ultrasonography revealed an enlarged uterus.

Zoldag et al. (1992) reported the diagnostic value of ultrasonography of pyometra in the bitch where in a very close correlation (r = 0.985) was found between the post-operative macroscopic and ultrasound scan measurement of the uterus. They concluded that ultrasonography is an accurate procedure for the qualitative and quantitative examination and diagnosis of canine pyometra.

Memon and Mickelsen (1993) have described the ultrasonic features of close pyometra in a four year old Golden Retriever bitch on transabdominal ultrasonography, they observed a large fluid filled uterus involving both uterine horns, the diameter of left and right horns were 4.0 and 3.2 cm respectively.

Alvarenga et al. (1995) presented the ultrasonicographic findings of uterus affected with pyometra. A B-mode scanner, 5 MHz ultrasonicographic instrument was used in 33 bitches with clinical histories compatible with pyometra. The dilated uterus presented an ultrasonographic image of well defined tubular structure with a diameter between 0.5 to 4
The uterine lumen contents were less echogenic than the wall, with clear echoic spots. There was a correlation between the increase in viscosity of the secretion and echogenicity. Ultrasonographic diagnosis was possible in 31 dogs (94%) and was confirmed at laprotomy. They concluded that B-mode ultrasonography to be an efficient method to diagnose pyometra in bitches.

Tello et al. (1996) scanned 50 bitches with clinical signs of pyometra and compared its diagnostic accuracy with radiography at lateral and ventrodorsal view and concluded that ultrasonography was more efficient than radiography in diagnosing positive cases with 100 percent efficacy.

Renton et al. (1993) measured the uterine horns by ultrasonography before application of PGF2α as medical treatment and reported the mean diameter of the uterine horn around 1.5 cm.

Matton and Nyland (1995) opined that ultrasonographic imaging of the uterus presents a much more specific technique which allows not only the type of pyometra to be recorded (showing whether it is localized, segmental or uniform tubular in nature) but also integrity of uterine wall and content type.

Fayrer et al. (1991), reported that ultrasonography can be used to make diagnosis of pyometra prior to any appearance of clinical signs, when monitoring bitches subjected to hormonal therapy for treatment of mismating or estrus suppression or when monitoring bitches exhibiting ovarian acyclicity.
2.7. Treatment.

The treatment of choice has traditionally been ovariohysterectomy (Hardy and Osborne 1974), but in some cases the bitches can be too severely affected to survive. Supportive treatments such as intravenous fluids are often administered in conjunction with surgical treatment of pyometra (Johnson 1995).

Medical treatment of pyometra with compounds promoting expulsion of the uterine pus, in combination with antimicrobials, is an option in some cases depending on the status of uterus and the general condition of the bitch (Meyers-Wallen et al. 1986; Trasch et al. 2003 and Gobello et al. 2003). Intra-uterine drainage is also a possibility (Funkquist et al. 1983). The fertility may thus be preserved, although decreased, but with high probability of recurrence of the disease (Nelson et al. 1982; Meyers-Wallen et al. 1986; Gilbert et al. 1989; Trasch et al. 2003; Gobello et al. 2003).

The earliest proposed medical therapy employed simply the use of systemic and local single antibiotics (Querol, 1981 and Threlfall, 1995). However, this generally leads to either a worsening or a delay in the worsening of the disease, with need for additional treatment at a later date.

During the last 10 years, new approaches have been proposed and numerous successful results of medical treatment for canine pyometra have been reported. Although these approaches have involved different protocols, they all essentially have the following goals (Verstegen et al. 2008).
(1) Preventing progesterone effects by either inducing luteolysis or preventing progesterone binding to its receptors. New protocols for the use of prostaglandins have been proposed, either alone or in association with either dopamine agonists or progesterone receptor antagonists.

(2) Promotion of cervical relaxation in closed pyometra to allow for the expulsion of the uterine contents. This is generally achieved by the administration of either prostaglandins or progesterone-receptor antagonists.

(3) Induction of uterine contractions and emptying, either directly through the use of \( \text{PGF}_2 \alpha \), or indirectly via progesterone receptor antagonists.

(4) Inhibition of bacterial growth and development through the use of broad-spectrum or specific spectrum antibiotics.

(5) Facilitating uterine regeneration in animals with clear signs of uterine degeneration. This is accomplished by prolonging anestrus. Prolongation of anestrus allows for further apoptosis and regeneration of the endometrium, preparing the animal for a new pregnancy. The androgen-receptor agonist, mibolerone, is used for this purpose.

2.7.1. Prostaglandins

Prostaglandins are important in reproductive biology with many physiological and pharmacological roles and also as mediators in inflammatory events (Kindahl et al. 1976; Kindahl 1980 and Fredriksson et al. 1984). Prostaglandins are derived from arachidonic acid and are produced and released from neutrophils, macrophages, lymphocytes and platelets during inflammation (Kindahl, 1980).
More recent and successful medical treatments have involved the repeated administration of PG F₂α (Meyers-Wallen et al., 1986 and Renton et al., 1993), which causes luteolysis and thus reduces plasma progesterone concentrations. Reduction in progesterone concentrations induces cervical relaxation, a decrease in uterine secretions and, since prostaglandins also have a uterine spasmogenic action facilitates the expulsion of uterine fluid.

Jackson (1979); Hubler et al. (1991) and Berchtold (1997) reported that higher doses of prostaglandins are associated with substantial adverse effects, including salivation, vomiting, straining, diarrhea, pyrexia, some occasional respiratory distress and uterine rupture, especially in cases of closed cervix pyometra leading to shock and death in bitches.

The use of PGF₂α to treat pyometra in dogs has been reported by several authors and the results have generally been positive, except when high doses were used (Fennie et al., 1989; Gilbert et al., 1989; Chen et al., 1996; Romagnoli et al., 1996 and Gabor et al., 1999).

Nelson et al. (1982) treated seventeen bitches with pyometra or postpartum endometritis with prostaglandin F₂α. Response to the treatment included cessation of uterine discharge, decrease in uterine diameter, reduction in plasma progesterone concentration, and return of a normal leukogram. Treatment of 14 bitches was successful, and 13 of these bitches subsequently experienced estrus. Of those 13 bitches, 11 were bred and 9 became pregnant. In 2 bitches, pyometra developed again, within 6 weeks of
the following estrus, illustrating that bitches so affected are susceptible to recurrence of the disease.

Meyers Wallen et al. (1986) studied immediate and long-term outcomes of PGF$_2\alpha$ treatment for canine pyometra in 10 bitches. Dinoprost tromethamine (0.25 or 0.5 mg/kg of body weight, subcutaneously) was given once daily for 3 days. Bitches were bred at the first post treatment estrus and monitored for a minimum of one year. Pretreatment WBC counts often did not reflect the severity of histopathologic findings in the uterus, but post treatment WBC counts were useful in monitoring response to treatment. Four bitches produced a litter within one year of treatment. Four bitches had recurrence of pyometra within one year of treatment, and these same bitches had another recurrence after an additional prostaglandin treatment. Three additional bitches had a recurrence by 27 months after therapy. Results suggested that subclinical disease may persist after treatment, with clinical recurrence during diestrus. Despite the high recurrence rate, it was concluded that this treatment is a practical treatment for canine pyometra when reproduction is desired.

Gilbert et al. (1989) treated bitches with pyometra with dinoprost, a prostaglandin F-2 $\alpha$-THAM salt or luprostiol, a synthetic PG analogue and oral broad-spectrum antibacterial drugs. PGF$_2\alpha$ treatment lasted 2 to 26 days and dosage varied from 26.8 to 258 micrograms/kg. Clinical cure from symptoms was achieved in 33 of the 40 bitches. Of 14 bitches from which the owners opted to breed, nine eventually produced litters. Long-term complications were anoestrus, recurrence of metritis, failure to conceive and abortion.
Renton et al. (1993) carried out a clinical trial comprising of eight bitches diagnosed as having open pyometra, as assessed by ultrasonographic, radiographic, haematological and clinical investigation. Plasma progesterone was assessed before treatment with PGF$_2\alpha$ (Dinoprost) and three of the bitches had low concentrations. Each bitch was given multiple injections of PGF$_2\alpha$ and monitored during and after treatment. All bitches had basal progesterone concentrations after treatment and the uterine diameters were markedly reduced by that time. One bitch subsequently produced a litter of seven pups. Five of six bitches that were subjected to ovariohysterectomy 1 to 2 months later had normal uteri, although one had caseated areas in the uterine wall.

Feldman and Nelson (2004) used natural prostaglandin on day 1 at the dose rate of 0.1 mg/kg body weight SC followed by 0.2 mg/kg on day 2 and 0.25 mg/kg from day 3 to 7. The complete resolution of their uterine infection in 153 of 163 cases observed in ten days post treatment and of the 153, 109 whelped more than once.

Verstegen et al. (2008) in their review opined that, treatment with PGF$_2\alpha$ at doses of 10 to 50 mg/kg, administered three to five times daily for 3 to 7 days, have been used successfully for treatment of canine pyometra, either solely or in combination with other drugs. Extreme care should be exercised in calculating the dose, as the therapeutic index is relatively small (LD$_{50}$ in dogs is approximately 5 mg/kg) and side effects are quite severe when high doses are used (more than 100 mg/kg). The drug should be given SC or IM. Since side effects are dose-dependent and are known to diminish with repetition of treatment, it is recommended that one should start with the lowest dosage to avoid the classic side effect of vomiting, and then to slowly increase the dosage to reach higher doses (50 mg/kg) after 2 to 3 days.
Intra vaginal infusion of natural prostaglandins once or twice daily has also been tried by Gaber et al. (1999) in seventeen bitches with metritis or pyometra. PGF$_2$α (150 micrograms/kg body weight) was administered once or twice daily by infusing 0.3 ml per 10 kg body wt into the vaginal lumen. Bitches were also treated with amoxicillin (15 mg/kg body wt/48 hour) and/or gentamicin (4 mg/kg body wt/day) administered as intramuscular injections. Fifteen bitches were treated successfully with intravaginally administered PGF$_2$ α for 3 to 12 days and with intramuscularly administered antibiotics for 4 to 12 days. Success of treatment was judged by cessation of vaginal discharge, the absence of fluid in the uterus as determined by ultrasonography, and the overall health status of the animal. As two bitches with pyometra showed clinical deterioration in spite of medical treatment, no side effects (salivation, vomiting, diarrhea, hyperpnoea, ataxia, urination, anxiety, pupillary dilatation followed by contraction) were observed after PGF$_2$ α treatment. The disease did not recur during the subsequent oestrous cycles within 12 months after the initial treatment. The results demonstrate that intra vaginal administration of PGF$_2$ α was effective in 13 dogs (86.6%) with metritis or pyometra, and caused no side effects. Although the study was based on a relatively small number of cases, it was concluded that prostaglandin F$_2$α can be a useful means of treating bitches with metritis or pyometra. However, in severe cases of pyometra ovario hysterectomy is needed.

2.7.2. Progesterone-receptor antagonists

Progesterone-receptor antagonists, such as mifepristone or aglepristone bind to the progesterone receptor which they completely block, preventing any biological activity. Progesterone receptor antagonists competitively prevent progesterone from
binding to its receptor to induce transcription and exert all its biological effects at the cellular level. Consequently, the absence of receptor stimulation and activation mimics the effects observed when luteolysis is induced and thereby causes relaxation of the cervix (Hubler and Arnold, 2000; Hoffman et al. 2001; Wherend et al. 2003 and Corrda et al. 2006).

This product has been effectively used in the treatment of uterine infections associated with elevated plasma progesterone concentrations (Hoffman et al. 2001; Blendinger et al. 1997 and Breitkopf et al. 1997)

Breitkopf et al. (1997) conducted a study to know the effectiveness of an antiprogestin treatment on the involution of the pyometric uterus and general health of bitches. In their clinical study comprising seven bitches with pyometra and progesterone concentrations more than or equal to 2 ng per ml were treated with the antiprogestin RU 46534. The dose selected was 5 or 6 mg per kg body weight subcutaneously on the first day of treatment and 3 mg per kg body weight on days 2, 3, 4, 8, 12 and 16. Antibiotics were administered until day 16. A vulval discharge was observed within 12 to 24 h. In one bitch the dose of the antiprogestin had to be increased after day 4. In six bitches the uterine lumen became ultrasonographically undetectable between days 8 to 12, in one bitch some luminal material could still be detected on day 28. The number of blood leucocytes tended to increase after the onset of treatment but had returned to the upper normal range by day 16. In all dogs general condition and feed consumption improved rapidly and were normal within 8 days. No side effects were noted. Two of the dogs were
mated subsequently and produced two healthy litters. These observations confirm that the treatment of pyometra with an antiprogestin may lead to a clinical recovery.

Trasch et al. (2003) carried out a clinical study to determine the therapeutic success of the medical treatment of canine pyometra with the antigestagen aglepristone. In 48 (92.8%) of the 52 treated bitches, healing could be achieved within the first 3 weeks after the treatment had been started. One bitch died as a result of renal insufficiency, in three bitches there was no emptying of the uterus, so ovariohysterectomy became necessary. In these three patients, ovarian and endometrial cysts were present. Forty-one bitches could be followed up for 3 months. Four animals developed a recurrence (9.8%). In three bitches ovarian cysts and cystic endometrial hyperplasia could be found. In 37 animals data on the subsequent sex cycles are available. In 22 bitches next heat started at the expected time, in seven animal, heat started too early. In eight bitches the period of anoestrus was prolonged. Five of the six bred bitches delivered at least one litter. The presented data show that treatment of pyometra by aglepristone resulted in a high healing rate. The recurrence rate can be minimized by the selection of bitches without ovarian cysts and cystic endometrial hyperplasia.

Wehrend et al. (2003b) reported successful treatment of closed cervix pyometra with Antiprogestin on day 1, 2 and 7. On day 7 in all bitches uterine diameter was undetectable under ultrasonography and leucocytosis returned to normal physiological range.

Verstegen et al. (2008) in their review pointed out a controversy regarding the ability of this treatment to induce uterine contractions if used alone. Unlike the action of
PGF<sub>2α</sub>, progesterone antagonists are not expected to induce myometrial contractions. In their experience, uterine contractions associated with the use of progesterone-receptor antagonists have never been obvious enough to allow for the safe use of this type of drug without combination with prostaglandins.

However, some studies hypothesize that uterine contractions are indirectly induced by the local uterine release of endogenous prostaglandins as a consequence of the endometrial inflammatory process associated with the pyometra (Gobello et al. 2003).

2.7.3. Progesterone-receptor antagonists combined with prostaglandins

The use of this combination of medications results in both uterine contractions and induction of luteolysis, which in turn prevents all the effects of progesterone on the uterus and on the immune system.

Gobello et al. (2003) conducted a study in 15 bitches affected with pyometra to compare the efficacy and safety of two protocols using a combination of aglepristone and cloprostenol and to describe the progesterone (P<sub>4</sub>) serum profiles before and during treatments, for group I (n = 8) animals aglepristone was administered at 10mg/kg, s.c., on days 1, 3, 8, and 15 (if not cured), combined with cloprostenol at the dose of 1 microg/kg, s.c., on days 3 and 8, and group II (n = 7) received the same treatment with aglepristone as treatment I but cloprostenol on days 3, 5, 8 10, 12, and 15 (if not cured). Before the beginning of the treatments and then on days 8, 15, and 29 all bitches were evaluated for clinical signs, side effects, hemogram, serum P<sub>4</sub> concentrations, and uterus diameters. Bitches in both treatment groups, with or without initial basal P<sub>4</sub> serum concentrations, achieved treatment success without side effects and no significant differences, either on
day 15 or on day 29. In both treatments groups, clinical signs, blood parameters, and uterine diameters improved to normal values throughout the experiments. A significant interaction between day and treatment was found for percentage change in P₄ when all bitches were considered together. Redevelopment of pyometra in the next estrous cycle occurred in 20 percent of the bitches. One nonrecurrent bitch was mated and whelped a normal litter. It is concluded that these two combined protocols proved to be efficient and safe in reversing clinical signs of open cervix pyometra independently of initial P₄ concentrations and that the number of cloprostenol administrations seemed to have an effect on P₄ serum changes throughout treatments.

A study was under taken by Fienie (2006) to evaluate the efficacy of aglepristone at the dose rate of 10 mg/kg on days 1, 2 and 8 for the treatment of metritis or pyometra in bitches either alone or with or without the addition of low doses of cloprostenol for 5 days. Examinations performed on day 90, in addition to days 8, 14 and 28, determined that treatments had been curative in the long term. Aglepristone alone was curative in bitches with metritis and closed pyometra, cervical opening occurred within 48 h of aglepristone administration. The additional treatment with cloprostenol from days 3 to 7 with open or closed pyometra, significantly improved the overall success rate at day 90, compared to bitches without cloprostenol. The leucocyte count and plasma progesterone concentrations significantly decreased over the course of treatment.

Ucmak and Cagataytek (2008) investigated the effectiveness of ovariohysterectomy operations with aglepristone or aglepristone combined with PGF₂α treatment protocols. Thirty bitches with pyometra were divided into three equal groups
the surgically treated group (OP) and groups treated pharmacologically aglepristone (AL) and combined with PGF2α (AP). A dose of 10 mg/kg of aglepristone was administered subcutaneously on days 1, 2, and 7 and if needed on day 14. The bitches of the AP group received additionally 0.25 mg/kg of dinoprost tromethamine, once every 24 h between days 3 to 7. Eighty percent of the bitches in surgically treated group and fifty percent in the pharmacologically treated groups were recovered. The authors opined that by carrying out frequently repeated examinations, pharmacological treatments using aglepristone or combined with PGF2α constitutes a safe alternative to ovariohysterectomy in bitches in the dioestrus phase with P₄ more than 2 ng/ml and without ovarian cysts.

Gobello et al.(2003) and Fienie (2006) opined that the combination of aglepristone with cloprostenol was more effective in the medical treatment of open and closed pyometra than aglepristone alone.

2.7.4. Prostaglandin E

In many species, normal relaxation of the cervix at the time of estrus and ovulation is probably the result of the peri ovulatory changes in reproductive hormones that occur at this time. The increases in estradiol (and possibly oxytocin) receptor concentrations (Shemesh et al,1997) during the peri-ovulatory period are thought to increase prostaglandin E₂ synthesis and receptors (Schmitz et al.2006), leading to remodeling of cervical extracellular matrix (Stys et al.1981 and Ledger et al. 1983) which allows relaxation of the cervix. In ewes, misoprostol has been demonstrated to improve cervical patency and penetrability at the end of estrus (Leethongdee et al.2007).
This effect appears to be related to misoprostol-induced increased expression of the mRNA for FSH-R in all cervical layers (Leethongdee et al. 2006).

Romagnoli (1996) reported that PGE has been recently tested in dogs with pyometra and appears to be effective for causing evacuation of uterine content, although no information is yet available on its luteolytic properties.

Anecdotal results concerning the successful use of misoprostol intravaginally to promote cervical relaxation have been presented in dogs, but without scientific evidence of its efficacy (Verstegen et al. 2008).

2.7.5. Antimicrobial treatment

Concomitant broad-spectrum antimicrobial therapy should be administered during any treatment protocol. Ideally, identification and sensitivity should be determined from vaginal discharges as soon as possible and before initiating any antimicrobial treatment.

The type of antimicrobial drug chosen for adjunctive pyometra treatment is mainly based on previous knowledge of antimicrobial susceptibility of E. coli. (Gandotra et al. 1994, Pradhan et al. 1999 and Lee et al. 2000).

In canines, most studies on E. coli were performed on isolates from urinary tract infections (Low et al. 1988). Bacterial cultures from urinary tract infections are likely to be biased towards problematic cases, and it is uncertain if their resistance pattern is representative also of E. coli from pyometra (Bywater, 2000 and Kerrn et al. 2002).
Many antimicrobials have been used successfully, but in vitro sensitivity studies and clinical evidence suggest that amoxicillin, amoxicillin plus clavulanic acid, cephalosporins, or potentiated sulfonamides are good initial choices. The final choice should always be based on the culture identification and sensitivity of the bacteria involved (Verstegen et al. 2008).

Coggan et al. (2008) in their study reported antimicrobial susceptibility of the isolates considering the 151 strains of E. coli, 86.1% were resistant to cephalothin, 68.9% to ampicillin, 46.4% to cefoxitin, 34.4% to tobramycin, 32.5% to tetracycline, 29.8% to amicacin, 27.8% to cefalexin, 15.2% to gentamicin, 13.9% to cefotaxim, 13.2% to sulphazotrin, 12.6% to enrofloxacin, 10.6% to aztreonam, 7.9% to chloramphenicol, 6% to neomycin, 2% to norfloxacin and 0.7% to polimixin B. The highest sensitivity was to norfloxacin (94%), polimixin B (82.8%), sulphazotrin (76.8%), enrofloxacin (75.5%) and chloramphenicol (75.5%).

2.8. Disease evaluation.

Romagnoli (1996) stated that the condition of dogs under treatment should be monitored closely. In some animals, due to the increase contractility of the uterus, resorption of toxins may increase and the overall condition of the animal may eventually deteriorate. Supportive treatments are absolutely required and should minimally include perfusion with IV fluids at 1.5 to 2 times the maintenance rate, and eventually renal and hepatic as well as cardiovascular support. The rupture of the uterus, never observed by the authors when prostaglandins are used at low doses but described by others when prostaglandins were used at doses more than 100 mg/kg, spontaneously or during
treatment, may be observed by temporary improvement of the clinical status of the dog before a severe bacterial peritonitis supervenes and an acute abdomen syndrome develops.

When prostaglandins alone, or in association with progesterone-receptor antagonist, are used, the condition of the animals usually improves within the first 48 h after the onset of treatment (Gobello et al. 2003). This is accompanied with a noticeable increase in the amount of discharge 24 h after initiation of treatment. This increase in the amount of the exudate is associated with the decrease in plasma progesterone concentrations or progesterone inhibition. (Feini 2006) The discharge changes from purulent or serosanguineous to serous, and its cessation occurs in most cases in 4 to 7 days. The most commonly affected parameters include changes in the blood profile (leukocytosis with neutrophilia or leukopenia and inhibition of lymphocyte activity). It has been recently shown that following hysterectomy for pyometra treatment, all affected parameters return to normal within 7 days (Bartoskova et al. 2007). In medically treated dogs, the leukogram returns to normal within 10 to 15 days, although leukocytosis may be initially aggravated in some cases (England et al. 2007). Evaluation of the efficacy of the treatment is documented with ultrasonography and is demonstrated by a reduction of the uterine lumen by at least 5 percent within 3 to 5 days after the start of treatment. In cases where such a reduction in the size of the uterus is not noted, the client should be informed of the possibility of unsuccessful medical treatment. The dog should be reassessed after another 2 to 5 days of treatment and if the uterus is no longer responding, either a complementary approach to the medical treatment or an ovario hysterectomy should be considered.
2.9. Recurrence of pyometra

The incidence of pyometra recurrence after medical treatment is still controversial, with contradictory results published. However, the percentage of recurrence is obviously decreasing over time with improvements in therapeutic approaches and treatments. Meyers-Wallen et al. (1986) described therapeutic success in all PGF₂α treated animals with recurrence in 40% of the bitches within 1 year, and 77% within 27 months.

Johnston et al. (2001) gave an overview of success and recurrence rates after conservative treatment of pyometra with prostaglandins and reported that recurrence rates averaged 10%.

Ettinger and Feldman (2004) have stated that pyometra recurs within one year of successful prostaglandin treatment in 26 to 40% of bitches.

Trasch et al. (2003) found that 18.9% of the treated dogs relapsed after treatment with aglepristone alone and they concluded that the recurrence rate can be minimized by the selection of bitches without ovarian cysts and cystic endometrial hyperplasia.

Fieni (2006) reported 19% (4 of 21) of recurrence rate with antiprogestine and prostaglandin treatment after 24 month where as Gobello et al (2003) observed recurrence of the disease in 20% of the dogs treated.

Blendinger (1996) opined that the outcome of several different protocols for conservative treatment of canine pyometra is similar but two considerations may improve the results in the future:
1) Preclusion from treatment of bitches with ovarian diseases (cysts / irregular heats) and cystic endometrial diseases.

2) The addition of prostaglandins to the treatment protocol (at least to bitches with low progesterone levels).

England and Yeager (1993) reported that, although it could be possible to reduce the incidence of CEH and its effects on pyometra, the presence of ovarian cysts is more difficult to assess, even with ultrasonography. Furthermore, it is not possible to differentiate some estrogen producing ovarian cysts from non pathologic para ovarian cysts or corpora lutea with a fluid-filled cavity.

Verstegen et al. (2008) in a case-based study comparing incidence of pyometra in a population of previously treated pyometra dogs (n = 57) and a control age-matched group of dogs not having presented with pyometra earlier (n = 256), they were not able to find any statistical difference in the probability that a dog of any specific age group either with or without prior disease would develop a pyometra). Therefore, when treatment and uterine regeneration is successful, the probability for the bitch to develop pyometra again is the same as the probability for a naive bitch of the same age to develop a pyometra. These conclusions were the same in terms of fertility, which is not affected. It appears that a delayed response to treatment is associated with the increased likelihood of recurrence of symptoms, i.e. dogs that respond rapidly are more likely to breed successfully in the future. Therefore, evaluation of success of treatment be made after 5 days and in case of no apparent improvement, ovario hysterectomy is recommended.