MECHANISMS AND THERAPY FOR RETAINED FETAL MEMBRANES AND UTERINE INFECTIONS OF COWS: A REVIEW

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Received for publication: *March 27, 1985*
Accepted: *January 29, 1986*

ABSTRACT

Literature pertaining to retained fetal membranes and postpartum uterine infections of cows was reviewed. Many therapeutic failures are explained by the effects of the uterine environment on antibacterials, the effects of therapy on the uterine defense mechanism, bacterial resistance patterns and the failure to recognize normal physiological processes in the postpartum period. Prostaglandin $F_{2\alpha}$ and its analogues provide effective alternatives to antibiotic and antibacterial therapy for most postpartum disorders.

Key words: retained placentas, uterine infections, therapy

INTRODUCTION

Treatment of postpartum disorders in cows is a controversial subject. There is little agreement among experts about the proper methods, perhaps because insufficient attention is paid to the normal involution process and uterine defense mechanism during the postpartum period. The problem is further complicated by the published results of treatment trials that, particularly due to absence of untreated control groups, have a questionable experimental design.

It is well documented that postpartum uterine disorders play a role in bovine infertility (1-40). However, the economic impact of the disorders may be overestimated and the role of therapy on the outcome of the diseases needs to be critically examined. In addition, the economic losses resulting from unnecessary therapy and milk withdrawal after antibiotic therapy must be weighed against any gains resulting from therapy.

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DISCUSSION

The Normal Postpartum Period

The postpartum period of the cow begins at parturition and continues until uterine involution is complete and normal estrous cycles and estrus behavior resume (41). The end of the postpartum period could be defined as "the first estrus at which pregnancy can be re-established (42)." The length of the postpartum period is affected by a variety of factors including age, breed, season, nutrition, parity, periparturient diseases, and dystocia with its various ramifications (3, 14-16, 19-21, 23, 24, 28-40). The postpartum period is often lenghthened as a result of delayed involution of the uterus, delayed resumption of estrous cycles or both.

Estimates of the average interval from parturition until completion of involution vary widely. Some early studies suggest that this time is 18 to 25 days (7, 43). However, more recent reports indicate that an interval of 30 to 50 days may be more common (41, 44, 46). Return to a histologically normal state takes up to 20 days longer than return to a clinically normal state (44).

Uterine involution begins with reduction of uterine size by vaso-constriction and myometrial contractions. The reduction in size is relatively slow during the first ten days, but is followed by a period of markedly increased uterine tone and reduction in size during days 10 to 14 (7). During this interval, which in dairy cows often coincides with the first postpartum follicular growth and/or ovulation, most of the lochia is discharged. The main reduction in size, to that similar to the pregravid state, is completed by 25 to 50 days (7, 41, 44).

Involution of the maternal placenta involves 1) Necrosis and sloughing of the superficial layer and stalk of the caruncle. This process begins at 5 to 7 days and is complete by 10 to 12 days postpartum. The necrotic tissue contributes to the lochia. 2) Shrinkage of the caruncles to their prepartum size. This is complete by two to three weeks post-partum. 3) Re-epithelialization of the caruncles. Involution is considered by some authors to be complete when epithelium covers the caruncles, usually by 25 to 40 days postpartum (43, 45, 46).

Ovarian function plays an important role in the postpartum period; however, its effect on the rate of uterine involution is not clear. Although the re-establishment of ovarian cyclic activity usually occurs simultaneously with uterine involution, it does not appear to be a pre-requisite. Ovariectomy does not delay uterine involution, estradiol injections do not accelerate it, but progesterone injections slightly delay it (44). Uterine involution in suckled cows, which often have greatly delayed returns to normal estrous cycles, is not retarded (45). Primiparous heifers, which frequently have delayed onset of normal estrous cycles, have more rapid uterine involution rates than pluri-parous cows (7, 44). Uterine infections, which delay involution, are more readily eliminated in cows with regular estrous cycles (29, 32).

The re-establishment of normal estrous activity is, however, dependent upon the involution of the uterus. In dairy cows, follicular growth begins as early as four to ten days after calving (7), but the occurence of the first postpartum ovulation varies widely, with ranges of 4 to 65 days reported (41). The interval until the first postpartum ovulation is influenced by many factors including breed, milking vs. suckling, nutrition, and periparturient diseases (3, 7). The interval between the first and second postpartum ovulation is usually shorter than the succeeding intervals which are approximately 21 days (7, 41). These short interestrus intervals may be a result of aberrations in either luteinizing hormone (LH) production or release, which lead to abnormal corpus luteum (CL) development and/or maintenance (7). Prostaglandin $F_2\alpha$ ($PGF_2\alpha$) is released for up to 20 days during normal uterine involution and may be responsible for the premature lysis of the first postpartum CL (47).

The role of $PGF_2\alpha$ in uterine involution is not well understood. Studies have shown that there is a massive postpartum release of $PGF_2\alpha$ and this release is related to the degree and rate of uterine involution; i.e., cows with a normal puerperium, that complete uterine involution in a shorter time have a larger and longer postpartum release of $PGF_2\alpha$(47, 48). It is concluded that the release of $PGF_2\alpha$ reflects the degree of endometrial damage and/or repair (47, 48). Uterine involution can be accelerated by exogenous $PGF_2\alpha$ injections, but the large and repeated dosages required are not feasible for field application (47, 48). Cows with retained placentas or abnormal uterine function have a prolonged release of $PGF_2\alpha$. This may be due to a damaged uterus requiring more extensive repair or to bacterial toxins which stimulate $PGF_2\alpha$ release (47). Ovarian activity, determined by detection of increased serum progesterone levels, does not start until the postpartum $PGF_2\alpha$ release either ceases or attains levels which are very close to baseline levels (47). The sustained release of $PGF_2\alpha$ in cows with abnormal uterine involution often results in short luteal phases. This may be a mechanism by which the effect of a prolonged progesterone influence on the uterus is avoided. Progesterone severely inhibits the ability of the uterine defense mechanism (UDM) to eliminate infections (49).

Contrary to an early report (43), involution of the uterus is almost always a septic rather than an aseptic process (3, 5, 9, 12, 32, 36, 50-55). The most common organisms isolated are facultative pathogens, hemolytic streptococci, corynebacteria, staphyloccoci, coliforms, and Gram negative anaerobes. The postpartum uterus provides ideal conditions for bacterial growth; however, under normal circumstances the bacteria are cleared from the uterus in a few days or weeks (5, 12). The bacteria are eliminated by the contraction of the myometrium which forces the lochia out through the cervix (7, 12) by the phagocytic activity of leucocytes in the uterine fluids and endometrium (56-58) and by the antibacterial substances produced by the uterine glands (49, 60).

It has been shown that aerobic and anaerobic bacteria can act synergistically to enhance the growth and/or pathogenicity of each other (60-63). Experimental models have shown that Fusobacterium

necrophorum produces a leucotoxin (61) and Bacteriodes melaninogenicus and B. fragillus produce and release a substance which prevents bacterial phagocytosis (63). These anaerobes, along with Corynebacterium pyogenes which produces a growth factor for F. necrophorum, are implicated as the major causes of postpartum metritis and pyometra (51, 60). Purulent endometritis during the postpartum period can be caused by bacteria such as streptococci, staphylococci, pasteurella, bacilli, and others, but these infections rarely persist, cause permanent endometrial damage, or lead to impaired fertility (5, 13, 17, 18, 22, 49-55).

Although the bacterial invasions by organisms, other than the aforementioned C. pyogenes and the Gram-negative anaerobes, have little or no permanent effect on fertility, they may be responsible for acute, septic metritis which is often associated with retained fetal membranes and for some of the mortalities that occur (51, 64).

The Uterine Defense Mechanism

The initial uterine defense against bacterial infection is phagocytosis and killing by uterine leucocytes (56-58, 65, 66). Several studies have demonstrated that cows with puerperal disturbances (retained placenta and metritis) have decreased leucocytic activity (56-58, 66). It has been shown that cows which retain fetal membranes after normal parturition have decreased leucocytic activity prior to parturition (65, 67). This suggests that decreased leucocytic activity is a cause, not an effect, of retained placentas (68, 69). However, cows suffering acute puerperal metritis without placental retention also have decreased phagocytic activity of the uterine neutrophils (56-58, 65).

The role of immunoglobins in the defense against nonspecific bacterial infection has not been extensively investigated in cattle. Specific antibodies against Streptococcus hemolyticus and Corynebacterium pyogenes were found in the serum of sexually mature heifers, but not in the cervicovaginal mucus until several weeks after the first parturition (59). In another study, C. pyogenes was isolated from the uterus of primiparous cows at ten days postpartum in 30% of the cases; whereas in pluriparous cows isolations were made in only 6%. The lack of antibodies against C. pyogenes in primiparous cows may explain why postpartum metritis is both more common and often more severe in primiparous than in pluriparous cows (5).

Ovarian activity has a profound effect on the ability of the uterus to resist or eliminate bacterial infections. The uterus is highly resistant to infection during the estrogenic phase but very susceptible during periods of progesterone dominance (49).

The uterine defense mechanism (UDM) is inadequate during the progesterone dominated phase because 1) the intrauterine pH is low, creating a more favorable environment for those bacteria regularly isolated from the bovine uterus; 2) the uterine epithelium is less permeable to bacteria and as a result the leucocytic system is stimulated at a later stage; 3) the leucocytes' appearance in the endometrium and uterine lumen is

delayed; 4) the activity of leucocytes is decreased; and 5) the uterine secretion has no detoxicating effect (49). A recent study examined the relationship between polymorphonuclear (PMN) leucocyte functions and the serum levels of estradiol, progesterone, and cortisol (70). High physiologic values of estradiol did not alter the PMN functions which were directly related to bactericidal activity, but physiological levels of progesterone inhibited two of the five parameters used to evaluate PMN functions (70). The researchers suggested that the "increased resistance of the uterus to bacterial infection when estradiol values are high may be related to the fact that progesterone blood values are relatively low at this time (70)." That is, the supposed beneficial effects of increased estradiol production associated with follicular growth are probably the result of a lack of detrimental effects being produced by progesterone secreted by the corpus luteum. Further, low estradiol values in cows with inactive ovaries in the postpartum period may be less detrimental to PMN function than are high progesterone values associated with corpus luteum function. The following studies support this hypothesis.

One study showed that in cows which developed pyometra, ovulation occured on an average of 15.4 days postpartum vs 21.8 days in cows without pyometra and 18.6 days in cows with retained fetal membranes (RFM). Pyometra occurs when increased levels of progesterone coincide with the period when pathogenic organisms such as C. pyogenes and Gram negative anaerobes are in the uterus in high numbers (51). This occurs during the first two weeks postpartum. These organisms cause severe inflammatory reactions that inhibit normal repair of the endometrium, decrease PGF release and, therefore, favor retention of the CL and high progesterone production indefinitely (51). Attempts to stimulate the early return to normal ovarian cyclic activity by injecting gonadotropin releasing hormone (GnRH) early in the postpartum period resulted in increased numbers of pyometras (71, 72). However, some of the GnRH treatments reduced the incidence of ovarian abnormalities such as follicular cysts, improved reproductive performance in herds with good management and benefited cows with retained placentas, but were not recommended as routines for all cows (71-75).

The postpartum period in cows is characterized by dynamic changes in uterine and ovarian function, but the end of the postpartum period is often delayed by failure of either or both to return to normal function. Efforts to speed the return of normal function by using chemotherapy have been mostly unsuccessful. The reasons for the poor response to therapy can in many cases be elucidated, if the effects of therapy on the normal postpartum events are examined. Many of the therapeutic failures are due to the misinterpretation of clinical signs or physiologic events, the failure to consider the pathophysiology of the disease condition, or are the result of the inhibitory effects of the therapy itself.

Retention of Fetal Membranes

One of the most common postpartum disorders encountered is the retention of fetal membranes (RFM). It is generally accepted that the lack of dehiscence and expulsion of the fetal membranes by 12 to 24 hours postpartum constitutes retention (4, 5, 20, 23, 64). The incidence of RFM is quite variable, with morbidity estimates ranging from 1.96% to 55% (23); however, except in circumstances such as Brucella-infected herds, dystocia, or nutritional deficiencies the range is reported to be only 3% to 12% with an average of 7% (23). Mortality resulting from RFM is reported to be 1% to 4% (23). Temporary impairment of appetite and reduction in milk yield occurs in 55% to 65% of affected cows (23).

The impact of RFM on bovine fertility has been examined by several investigators (4, 6, 20, 21, 23, 26, 37, 40). There seems to be agreement that uncomplicated cases of RFM have minor adverse effects on future fertility. However, complications, especially the metritis complex, occur in more than half of the RFM cows (21).

The goal of therapeutic regimens should be to prevent the complications that are associated with RFM. The presently accepted therapeutic approaches, involving efforts to speed expulsion of the RFM or prevent uterine infections, have produced very inconsistent results (6, 23, 26, 37, 38, 40, 64, 74-82).

Retained fetal membranes are usually a result of either a disturbance in the loosening mechanism in the placentomes or of uterine inertia (23, 83, 84). The physiological processes that influence separation and expulsion of the placenta begin weeks or months before parturition so that by the time RFM is diagnosed, the effect - not the cause - is treated and the chances for response to therapy are few.

The physiological separation of the bovine placenta requires: 1) the prepartum maturation of the placenta; 2) the intrapartum mechanical detachment by uterine pressure; 3) anemia of the fetal villi after fetal expulsion; and 4) reduction of the size of the caruncle during postpartum uterine contractions (83).

Pathological factors that affect the loosening process in the placentomes include the following: 1) Immature placentomes, associated with shortened gestation periods (abortions, premature parturitions). These are believed to result from insufficient hormonally-induced changes in the maternal connective tissue and crypt epithelium (83). Studies have shown that cows with RFM have higher blood progesterone and lower $PGF_{2\alpha}$ concentration in the placentomes than cows without RFM (85, 86). 2) Edema of the chorionic villi associated with caesarean sections or uterine torsions mechanically prevents detachment from the maternal caruncles. Little can be done to prevent or treat these conditions. 3) Necrotic areas between the chorionic villi and cryptal walls, possibly due to allergic reactions, suggest prepartum changes induced by a more generalized disease, and are usually unresponsive to specific therapy. 4) Advanced involution of placentomes, occurring cases of prolonged gestation, is accompanied by proliferative changes

in the maternal caruncles that mechanically inhibit loosening. 5) Hyperemia of the placentomes and the associated swelling, perhaps occurring antepartum or resulting from premature closure of the umbilical vessels, may also incarcerate the fetal villi in the crypts. 6) Placentitis and cotyledonitis associated with infections, abortions or placental infections and having various degrees of inflammatory changes, can lead to mechanical inhibition of the loosening process or immaturity of the placentomes. 7) Uterine atony, which is often associated with dystocia, hypocalcemia, hydropic and other pathological conditions, contributes to the failure of the placenta to be expelled; however, RFM resulting from atony without any disturbance in the loosening process in the placentomes only occurs in 1% to 2% of the cases (83). This agrees with other studies which showed that uterine contractility in cows with RFM is similar to or greater than in cows without RFM (87, 88). 8) Acute metritis, caused by organisms invading the uterus at parturition or soon after, can result in uterine inertia, placentitis and RFM (15, 64).

Considering the different mechanisms involved in the etiology of RFM it then becomes understandable that chemotherapy, which is directed at speeding the loosening and expulsion of the placenta after the condition has developed, has little chance for altering the course of events, unless the RFM is due to uterine atony. If the normal loosening process has not occurred by the time of parturition, RFM and its sequela are further complicated by the rapid decrease in myometrial activity beginning about 24 hours post-partum and disappearing entirely in about 48 hours (23). If expulsion of the fetal membranes has not occurred by this time the placenta undergoes progressive liquifactive putrefaction until spontaneous expulsion occurs six to ten days later. A massive invasion of microorganisms occurs during this period, resulting in the associated complications. Therapy, heretofore, has been directed toward reducing the bacterial colonization of the placenta, lochia, and uterus.

Manual removal of the fetal membranes was one of the earliest and most widely practiced forms of therapy and it is still advocated by some authors (23, 56, 83). Unfortunately, most of the literature supports the conclusion that manual removal of RFM, with or without supportive chemotherapy, leads to impairment rather than improvement of fertility (6, 23, 26, 49). Roberts (1971) stated: "if the placenta is allowed to drop away of its own accord, or if it is gently withdrawn from the uterus 10 to 12 days after parturition, involution of the uterus occurs sooner, and cessation of uterine discharge ceases more quickly than when the placenta is removed in a rough manner and portions are left in the uterus (64)". Arthur (1979) stated: "few veterinarians are not familiar with the cow the well being of which was distinctly reduced after manual removal of the membranes (23)". In a study of 134 cows with RFM, manual removal of the placenta, with or without intrauterine terramycin, resulted in lower first-service conception rates, total conception rates and longer calving-to-conception intervals when compared either to non-treated controls or to cows that received intrauterine therapy only (6). It is also reported that manual removal should not be attempted when there is evidence of systemic illness (26, 64).

Some European studies help clarify why manual removal of RFM is contraindicated. After manual removal of the placenta was attempted, the uteri of cows were examined at necropsy (34). Even when removal was accomplished with no external evidence of trauma, hemorrhages, hematomas, and vascular thrombi were found in the uteri. In many cases, when removal was thought to be complete, many of the caruncles had portions of the fetal cotyledons attached, macro- or microscopically. In addition, it was reported that phagocytosis by uterine leucocytes was completely inhibited for several days following attempts at manual removal. All types of intrauterine antiseptics had a similar inhibiting effect on uterine phagocytosis (34, 58). These changes could easily enhance invasion by microorganisms and enhance the production of the common sequela of RFM: metritis, peritonitis, abscesses, and adhesions (58).

The role of leucocytes in the eventual expulsion of retained fetal membranes and the UDM is not well understood. Chemotaxis and leucocyte presence in the placentomes are required for normal loosening of the placenta. When both of these requirements were met the incidence of RFM was approximately 1.4%, when either one was decreased or absent the incidence was 6.8% to 9.6% and when both were absent the incidence approached 100% (68). An unidentified leucocyte chemotaxis inhibiting factor (LCIF) was demonstrated in the cotyledons of retained placentas (69). Polish workers (65) and others (57) reported that phagocytosis was impaired in cows with RFM and metritis. One can speculate that introduction of chemotherapeutic agents into the uterus may interfere with the UDM (58). The use of chemotherapeutic agents will reduce putrefaction but may also inhibit lysis of the villi, thereby actually prolonging the retention of the membranes (23).

When the retained placenta is not manually removed, several alternate forms of therapy have been advocated. Drugs that increase uterine motility - including oxytocin, estrogens, ergot derivatives, calcium preparations, $PGF_2\alpha$ and its analogues - have shown limited benefit (23, 64, 78, 79). The lack of effectiveness is not surprising, considering the low incidence of RFM due to uterine atony without a disturbance in the loosening process in the placentomes (3). Intrauterine (IU) insertion of antibacterials and antibiotics has been a common form of therapy for many years. The reported effects of therapy are very inconsistent. It has been reported that intrauterine tetracycline therapy results in RFM cows having conception rates that approximate the rates of nonaffected herd mates (6, 80-82). However, other authors report either no beneficial effects (37, 76, 79) or detrimental effects of IU therapy (40). In light of controlled studies revealing that eliminating uterine infection with massive (56) and/or repeated doses of intrauterine antibiotics (55) was impossible prior to either expulsion of the placenta (56) or before three weeks postpartum (55), the reported beneficial effects of IU therapy for RFM must be questioned. In spite of repeated IU treatments, chronic endometritis or pyometra are common sequelae to RFM (2, 6, 21, 38, 64, 71, 76). Intrauterine antibacterials, however, do decrease putrefaction and the disagreeable odor associated with RFM (2, 6, 23, 76). Although this may

be an important aesthetic consideration, it is economically unsound when the cost of treatment, the loss of milk for human consumption, and the possible deleterious effects of the treatment on fertility are considered.

Nonantibiotic antibacterials have been advocated for the intrauterine treatment of RFM because of the absence of proven benefit of antibiotics, to avoid the possibility of antibiotic adulteration of milk and to avoid development of antibiotic-resistant organisms (23, 26). Results equal or superior to those achieved with antibiotics have been claimed (26). The use of Lugol's solution is not usually recommended for use on RFM because of its irritating effects on the uterus (64).

The use of estrogens to increase myometrial activity, phagocytosis, and the immune response of the uterus has been widely practiced (2, 23, 37, 40, 64). However, the long-acting estrogens and stilbestrol have been associated with more severe infections of the oviducts and myometrium (90), the development of cystic ovarian disease (64) and depressed fertility from unknown causes (40). After postpartum day 6, 5 mg estradiol benzoate injected intramuscularly has the ability to increase uterine phagocytosis without the undesirable side effects (57). Oxytocin also increases phagocytosis by uterine leucocytes; this effect can be seen for up to eight days postpartum if involution of the uterus has been delayed. Levamisole, one of several known immune potentiators, exerts a stimulating effect on uterine phagocytosis if the pretreatment phagocytic index is low (58). Freunds incomplete adjuvant stimulates the phagocytic index for about eight days (58).

The primary detrimental effect of RFM is that it is a predisposing factor in the development of acute or chronic postpartum metritis (21, 26). Therefore, therapy should be directed toward preventing the adverse effects of postpartum metritis. The common practice of intrauterine antibiotic or antibacterial therapy for RFM has shown little hope for meeting that goal. de Bois (26), summarizes antibiotic treatment as follows: "it is generally accepted that broad spectrum antibiotics are quite capable of controlling putrefaction, but freedom from infections is quite often not achieved despite repeated IU treatments; it is likely that the fertility rate of cows which calved normally, retained their placentas and were treated IU with antibiotics, is less than cows which calved normally and dropped the fetal membranes promptly."

Poor response to treatment is likely due to the following: 1) drugs commonly used to treat RFM do not accelerate the loosening process in the placentome and, in fact, may delay it; 2) intrauterine manipulations (manual removal, antibacterial infusions, etc.) are traumatizing and inhibit the phagocytosis that is necessary for placental detachment and clearance of bacterial infections; 3) drugs used for intrauterine therapy are often rendered ineffective because of the nature of the uterine environment, types of organisms present, and the pharmacological characteristics of the drugs; 4) the infertility in an animal or herd may not be due to the effects of RFM or may be caused by other factors and could not be expected to benefit from this type of therapy.

Postpartum Metritis

Ball et al. (1984) defined the postpartum period as "the period from parturition to complete involution (92)." They subdivided the postpartum period into the "early postpartum period," the "intermediate period" and the "postovulatory period." This classification is useful because it distinguishes the postpartum interval, endocrine status, and clinical findings which are factors that influence prognosis and response to therapy.

The "early postpartum period" is the period following parturition until the pituitary gland becomes sensitive to gonadotrophin releasing hormone (GnRH). During this period, which lasts from 8 to 14 days, the ovaries produce minimal amounts of estrogens or progesterone. Acute puerperal metritis occurs during this period. Puerperal metritis following parturition occurs with or without the retention of the fetal membranes (64). It can be a sequel to emphysema of the fetus, prolonged dystocia, fetotomy, uterine prolapse, manual removal of RFM or unsanitary conditions at parturition (51, 56, 64). Clinical signs usually appear within one to ten days postpartum and last from two to six days but may remain from one to two weeks. The condition is characterized by a fetid, red to reddish-brown, watery uterine fluid and is often accompanied by uterine inertia or atony. In severe cases, peritonitis may occur by extension through the uterine wall. Typical signs of septicemia including depression, anorexia, weak rapid pulse, rapid shallow respirations, gastrointestinal atony and agalactia are often present (64). Treatment should be directed at overcoming the effects of septicemia and toxemia (64, 92).

The intermediate period begins when the pituitary becomes responsive to GnRH and lasts until the first ovulation (92). The length of this period varies greatly because times to the first postpartum ovulation of 4 to 65 days have been reported (41). Infections with bacteria are reduced, eliminated or become chronic during this period (92).

The postovulatory period extends from the first ovulation until uterine involution is complete (92). Uterine involution may be completed during the intermediate period if the first postpartum ovulation is delayed for more than 40 to 50 days. Chronic metritis or endometritis and pyometra are commonly diagnosed during this period (92).

Ideally, therapy for uterine infections should: 1) eliminate bacteria from the uterus, 2) not inhibit the normal UDM, 3) not cause further adulteration of milk or meat for human consumption. Most of the present forms of therapy fail to meet one or more of these criteria. Intrauterine (IU) antimicrobial therapy, as it is usually practiced, fails in most categories.

Optimal pharmacologic properties of intrauterine formulations are rapid dissolution and even distribution of an active drug throughout the uterine cavity, good penetration into the subendometrial layers, limited systemic absorption, lack of irritation, and the maintenance of

antibacterial activity in the environment of the uterus (93). Many factors contribute to the diminished efficacy of most of the commonly used IU drugs and recent reviews on uterine antimicrobial therapy contribute to understanding this (89, 92-94). The postpartum bovine uterus is an anaerobic environment, making the aminoglycoside group of antibiotics (gentamycin, kanamycin, streptomycin, neomycin) ineffective because they require oxygen for their activity (95). During the early postpartum period, many organisms in the uterus are capable of producing enzymes that inactivate or degrade antibiotics (i.e., penicillinase) (95). The presence of pus and organic debris in the uterine fluids could potentially inhibit drugs such as the sulfonamides, aminoglycosides, (95) and nitrofurazone, (92). During the early postpartum period and in cows with endometritis, the absorption of many drugs is greatly diminished (90, 94). When absorption of drugs is low, therapeutic levels in the deeper layers of the uterus and other parts of the genital tract are not likely to be achieved (90). Those uterine infections which might benefit from antibiotic therapy are rarely confined to the uterine cavity and surface of the endometrium (91).

Perhaps the most important reason for the apparent failure of IU therapy to rid the uterus of bacteria is the alteration of leucocyte function which occurs with IU manipulations. All types of tested IU antiseptics destroyed phagocytosis for several days after IU application (58). Many antibiotics, applied locally and perhaps systemically, have the same effect (96-98). IU treatment is more likely to achieve concentrations sufficient to depress or destroy leucocytic activity than systemic treatment (96-98). The potentially disastrous effects of depressed leucocyte activity on the UDM would be magnified if the pathogens present in the uterus were resistant to the antibiotic or antiseptic used. Organisms resistant to the usual IU antibiotics are often isolated from the uterus of postpartum cows (32, 55).

Irritating IU antibacterials should not be used in postpartum cows (2, 64) or cows with pyometra (99). Irritating drugs include nitrofurazone, (92), streptomycin (93), tyrothricin (64), oxytetracycline (100, 101) and Lugol's solution (100, 101). Intrauterine drugs can alter corpus luteum lifespan through irritating effects (100, 101). Infusions given at day 4 or 5 post estrus induce estrus within four to seven days (101). Irritating solutions also cause a necrotizing endometritis and leucocytic exudiation in cows with normal uteri or with mild endometritis (101). This may be beneficial, although experimental evidence is lacking (89). Irritating solutions have no effect on the interval between cycles if they are infused during midcycle or at estrus, but they prolong the interval if given on days 16 through 19 (101).

Considering that most, if not all, IU treatments result in some absorption of the drug, adulteration of meat and milk becomes a problem (10, 25, 94, 102-110). Absorption is less in early postpartum cows, cows with endometritis and during the diestrus period but it varies with the type of drug, dosage, and vehicle used (10, 25, 102-110). It must be recommended that milk and meat for human consumption be withheld after treatment with antibacterials. Unfortunately, no

official guidelines for minimum milk withholding time after IU therapy exist. The only drugs approved by the Food and Drug Administration for IU use in cattle are chlorhexidine tablets and chlorhexidine suspension[a]. No withdrawal time has been established for these products[a]. Antibiotic residues in milk can be detected for 36 to 120 hours after one to three infusions of tetracycline HCl, procaine penicillin G, Chloramphenicol, ampicillin trihydrate, kanamycin sulfate, gentamycin sulfate, and erythromycin by the Bacillus stearothermophilus disc-assay procedure (102). Logic suggests that milk must be withdrawn after intrauterine therapy to avoid the serious ethical or legal consequences of adulteration of milk for human consumption. Testing milk from treated cows for antibiotic residues on the farm may be the only effective method to avoid contamination (109). Withholding milk for human consumption after antibiotic therapy results in considerable economic losses to producers.

Systemic antibiotic administration has several potential advantages over IU applications. Systemic administration results in antibiotic concentration in the uterine tissue and lumen that are similar to blood and plasma concentrations (89, 90, 94). The concentrations achieved would not likely interfere with uterine leucocyte function (96, 97), but may not reach minimal inhibitory concentrations (89, 92). Absorption and clearance of the systemically administered drugs is more rapid than from IU administration (101), therefore, repeated doses two to three times a day may be required to maintain therapeutic levels of antibiotic (89, 90). This, however, can be carried out easier than with IU therapy and the risk of introducing new infections, injuring the endometrium, and depressing uterine phagocyte activity can be avoided. Withholding times for milk from systematically treated cows have been established for many of the antibiotics. However, the commercially available antibiotics that could be expected to be effective for uterine infections are not labeled for that use (91). The "extra-label" use of antibiotics in food-producing animals have come under intense scrutiny by the Food and Drug Administration (111). Much has been written about the pharmacology of the bovine uterus (89, 90, 92-94) but the efficacy of systemic antibiotic therapy for uterine infections has not been demonstrated.

Criteria for drug selection for systemic therapy of uterine infections include rapid and complete absorption from the injection site, a large volume of distribution, minimal binding or inactivation by endometrial tissue or secretions and slow elimination rate from the body (93). Broad spectrum antibacterial activity is also desirable. The fact that parenteral antibiotic therapy provides levels of antibiotic in all parts of the genital system similar to those found in serum would indicate that

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(a) Gable, D.A. Director, Division of Therapeutic Drugs for Food Animals, Center for Veterinary Medicine. FDA, Rockville, MD. personal communication (1984).

parenteral therapy might be superior to IU therapy. However, in the case of oxytetracycline, intravenous dosages of 11mg/kg given twice daily maintained a serum concentration of only 5 µg/ml. This is far below the reported average minimum inhibitory concentration (MIC) of 20.4 µg/ml for C. pyogenes (92). Pharmacologic studies suggest that chloramphenicol should be the most effective available antibiotic for treatment of uterine infections (93). However, chloramphenicol is banned for use in food-producing animals in the United States (111). Ampicillin, gentamycin and kanamycin are also likely to be effective systemically for puerperal metritis cases with systemic involvement (93), but they are not "labeled" for treatment of uterine infections of cows; furthermore it has been reported that uterine secretions severely inhibit the antibacterial activity of penicillin G, ampicillin, neomycin, dihydrostreptomycin, gentamycin, oxytetracycline, and chloramphenicol in both aerobic and anaerobic conditions (112).

Systemic penicillin is reported to be the parenteral "drug-of-choice" for puerperal metritis with systemic involvement, because antibiotic sensitivity tests reveal that most of the organisms responsible for causing "septicemic-metritis" are susceptible to penicillin (51, 82). Cows with puerperal metritis, without systemic involvement, are unlikely to benefit from parenteral penicillin treatments. Although adequate dosages will produce high drug levels in the endometrium and uterine fluids, most cases of puerperal metritis are caused by mixed bacterial infections and penicillinase-producing organisms may inactivate the drug (51, 92, 94).

Adequate field trials comparing the efficacy of parenteral vs intrauterine therapy have not been reported. It is unlikely that trials will be run because of the restrictions on the "extra-label" use of antibiotics in food animals and the requirement for longer milk-withdrawal times following parenteral treatment.

Fortunately for producers and veterinarians, there appears to be an effective alternative to antibiotic therapy for most postpartum uterine infections. Prostaglandin $F_2\alpha$ and its analogues (PGF) have many properties that provide effective therapy for postpartum uterine infections, excluding those with generalized septicemic involvement.

Prostaglandin Therapy for Postpartum Uterine Disorders

Prostaglandin $F_2\alpha$ and several analogues (PGF) have been widely used in reproductive management programs. PGF has been used to cause lysis of a mature CL for synchronizing estrus, inducing estrus, terminating pregnancy, and treating luteal cysts, postpartum endometritis, and pyometra (113-124). Administrating PGF to cows with a CL, between days 5 to 16 post ovulation, or to those with a mature CL or luteal tissue associated with pregnancy, pyometra or luteal cysts results in luteolysis and a rapid decrease in blood progesterone levels, usually within 24 hours post-injection (113, 124). The decrease in progesterone levels is followed by follicular growth and estrogen secretion. Estrus and ovulation usually occur within three to seven days post-injection in

nonpregnant animals (113). In pregnant animals, lysis of the CL results in abortion or induction of parturition during much of the gestation period (124).

The rationale of using PGF for postpartum uterine infections is 1) PGF-induced luteolysis decreases progesterone inhibition of the UDM; 2) estrogen production, which follows luteolysis, stimulates the UDM; 3) PGF may stimulate myometrial contractions that aid in the expulsion of uterine lochia, pus or other contents; and 4) PGF may have a stimulatory effect on phagocytosis by uterine leucocytes (125).

Evidence from experimental and field trials demonstrates the effectiveness of PGF treatments, showing it to be the most effective therapy for pyometra (116, 123). One PGF injection resulted in negative bacterial cultures and normal clinical findings in approximately 90% of cows with chronic endometritis (pyometra) (116). Cows that failed to respond completely after one PGF treatment often recovered after a second injection 10 to 14 days later (116, 124). Any advantage from supplemental intrauterine or systemic antibiotic therapy has been only rarely demonstrated (118). In fact, one study showed that infusing nitrofurazone after evacuating the uterus not only failed to prevent relapses but also had a negative effect on conception rates following clinical recovery (123).

The argument favoring the use of PGF prior to the formation of the first postpartum corpus luteum is not strong. However, benefits have been claimed (29, 77, 119, 120). It has been reported that cows with delayed uterine involution have higher serum progesterone levels than normal cows even when a CL cannot be palpated (91). It was hypothesized that the source of progesterone was the adrenal or luteinized follicles. If the progesterone source were luteinized follicles, PGF would be expected to be luteolytic and decrease the serum progesterone. PGF would have no effect if the progesterone was adrenal in origin. The weak uterotonic effect (78, 120) and the stimulatory effect on phagocytosis (66, 125), theoretically, should be beneficial.

Following the development of the first postpartum CL, PGF would be expected to have beneficial effects resulting from luteolysis and the decreasing progesterone levels and the benefits of the induced estrus. It has been observed that cows which ovulate and develop a CL early in the postpartum period are more prone to develop pyometra (51). This observation supports the hypothesis that "increased progesterone levels, coinciding with high numbers of organisms in the uterus, especially C. pyogenes, contribute to the development of pyometra (51)." It also supports the hypothesis that prevention of early luteal formation or induction of premature lysis of the CL are effective, both prophylactically and therapeutically for postpartum uterine infections. Several reports support this hypothesis (90, 114-122).

In a Yugoslavian trial (115) involving 209 cows that had either delayed uterine involution or uterine discharge at 18 to 19 days postpartum, the

animals received either a cloprostenol[b] (an analogue of $PGF_2\alpha$) injection or an intrauterine infusion of a popular proprietary antibacterial containing metacresolsulphone and formaldehyde (other trials have reported this infusion product to be "superior to" or "as beneficial as" intrauterine antibiotic therapy (36, 128)). All cows were re-examined at 3 and 11 days after PGF or IU treatment and retreated on day 11. A final clinical examination was conducted 14 days after the initial treatment or three days after the second treatment. No detectable differences in the rate of uterine involution or restoration of vaginal discharge to normal were found between the cloprostenol-treated cows and those receiving intrauterine therapy. However, cows receiving cloprostenol had 22% better first-service conception rates(34.6% vs 16.2%), 21% fewer inseminations per pregnancy (2.42 vs 2.90) and a15% (89.5 vs 105.9 days) improvement in calving-to-conception interval. When comparing the percentage of treated cows that became pregnant within 85 days of calving, a "must" if a 365-day calving interval is to be maintained, it was found that 60.9% of the cloprostenol-treated group achieved this goal vs 38.2% of the intrauterine-treated group. This was an improvement of 59.4%. Improvements in these parameters are the "economically important" parameters. Clinical recoveries without corresponding improvements in fertility are of little value.

Although the rates of apparent clinical recoveries following either PGF or intrauterine therapy are similar in all reports that were reviewed, PGF treatment resulted in equal or superior fertility than that obtained after intrauterine therapy, regardless of the chemotherapeutic agent used (114, 118, 122). In one trial, intrauterine antibiotic therapy at 5 to 16 days after PGF treatment resulted in better fertility than two PGF treatments or two IU antibiotic treatments. Unfortunately, the antibiotic used was not identified (118).

Uterine Infections after the Postpartum Period

Uterine infections, either clinical or subclinical, have long been touted as major causes of conception failures or symptomless "repeat-breeders" (1-34). Early reports of improved conception rates observed following IU therapy after breeding (8, 127) have not been confirmed by other studies (2,32,128-130). In fact, postbreeding infusions in repeat-breeding cows often result in conception rates lower than in untreated controls (129, 130). Routine IU treatments in the postpartum period do not improve fertility (36, 38, 131-136) and in some cases they decrease fertility (131, 135). It is not at all surprising that beneficial effects of intrauterine therapy for conception failures are difficult to document, considering 1) the characteristics of the organisms most often

(b) Estrumate, Imperial Chemical Industries PLC: Pharmaceuticals Division, Adderly Park, Macdesfield, Cheshire, England SK10 4TF.

responsible for persisting uterine infections (C. pyogenes and Gram negative anaerobes (11-13, 51, 92)); 2) the likelihood of permanent endometrial damage resulting from either infections (11) or the use of irritating solutions (2, 64); 3) the adverse effects that IU therapy exerts on uterine phagocytosis (56-58); and 4) the failure to demonstrate a clear relationship between "repeat-breeders" and the presence of endometritis (13, 18, 22).

As stated earlier, many of the chemotherapeutic agents commonly used for intrauterine therapy are irritating to the endometrium. The local irritation and necrosis may stimulate the UDM (89) and/or cause release of PGF, which results in luteolysis and induction of estrus (100, 101, 122, 129). It is probable that any observed beneficial effect of intrauterine therapy would be the result of either one or a combination of these actions, considering that most of the commonly used antibiotics are ineffective in the uterus. Since PGF injections are nearly always effective in inducing luteolysis and estrus, the use of intrauterine irritants can no longer be justified on the basis that they may cause luteolysis. Beneficial effects of shortening the interestrus intervals with PGF have not been documented in cases of mild-to-moderate endometritis; however, based on the curative effects observed following one or two PGF treatments in pyometra cases. (116, 123), it is logical to assume that PGF also would be effective in the less-severe cases. Any expected benefits of uterine irritation, or "chemical curettage," have to be weighed against the possible unwanted effects of iatrogenic endometrial and oviductal damage plus the economic loss sustained when withholding milk after intrauterine chemotherapy. This plus the finding that endometritis is not a significant cause of repeat breeding in clinically normal cows makes intrauterine therapy a very questionable and expensive procedure.

Other Factors Affecting Results of Therapy

There is much confusion concerning the proper management of postpartum uterine disorders. It is often a result of misinterpretation of physiologic events. Some of the erroneous assumptions are that the discharge of reddish brown fluid from the vagina during the first 10 to 14 days postpartum is a sign of puerperal metritis, when it is, in fact, the discharge of lochia - a desirable physiological event (7); and that purulent vaginal discharge is a sign of metritis or endometritis, when, in the postpartum period, it may be a sign of an active UDM (57, 58, 89). Purulent endometritis or metritis, which often recover spontaneously, are often misdiagnosed as "pyometra" if the diagnosis is made before the end of the expected lifespan of the first postpartum CL. In cases of pyometra the CL is retained for a prolonged period. Bacteriological cultures of uterine fluids can be misleading. Because of the constantly changing microflora during the postpartum period, a result of clearance and recontamination, a single culture is of little diagnostic value (12, 13). Further, C. pyogenes seems to be the only organism that is consistently associated with pathological lesions in the uterus (11). Cloudy mucous at estrus is often interpreted as a sign of endometritis. However, one study reported that clear mucus contained pathogenic

organisms in 23% of the cases, the same percentage as in opaque mucous (53). The wide variation in the rate of uterine involution makes a single clinical examination of little diagnostic or prognostic value prior to 30 days postpartum (41).

SUMMARY

The preponderance of literature reviewed supports the conclusion that there is only an occasional beneficial effect from using antibiotic and antibacterial therapy for uterine disorders in the postpartum period. Cows with systemic involvement often respond to antibiotic therapy as a life-saving procedure or for reducing the deleterious effects of toxemia or septicemia on milk production. However, any gains obtained in the ensuing increase in milk production would, because of residues, probably be negated by the losses incurred from withholding the milk from human consumption. Routine intrauterine antibiotic therapy in cows without histories of postpartum disorders cannot be justified (131-136), nor can the use of intrauterine antibiotics, postinsemination, in repeat-breeding cows (127-130). The prophylactic application of intrauterine tetracycline drugs in cows with RFM has been reported to be beneficial (6, 80) or detrimental (40) to fertility. Despite treatments with a variety of intrauterine drugs, the fertility of cows with RFM is often less than that of unaffected cows (26). The efficacy of systemic antibiotic therapy for uterine infections without systemic involvement has not been documented.

Antibiotic or antibacterial therapy for postpartum uterine infections has been practiced for many years (1-3). In spite of several decades' experience with a variety of treatment regimens, it is clear that antimicrobial therapy has done little to alleviate the infertility associated with uterine infections. Recent research on the pharmacology of the uterus and the effects of antibacterial therapy on the uterine defense mechanism help explain the often disappointing results.

In terms of fertility, the therapeutic and prophylactic use of $PGF_2\alpha$ and its analogues in the postpartum period consistently show results equal to or better than IU antibacterial therapy. Because PGF therapy has the advantages of systemic vs intrauterine administration and no requirement for milk withdrawal, it should be recommended as the initial treatment regimen for postpartum uterine disorders. It is likely that extended intervals from parturition to insemination, either through choice or because of estrus detection failures, are more responsible for long parturition-to-conception intervals than are uterine infections (15, 16, 19, 138, 139). Inducing estrus with PGF not only has therapeutic value for uterine infections, but it also shortens the interval from parturition to each successive estrus following the induced estrus (140). Shortening the interval from parturition to first or later insemination by five to ten days with PGF therapy would decrease the calving-to-conception interval by a similar time regardless of any beneficial effect on uterine infections or increased estrus detection efficiency.

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