Abortifacient Response and Plasma Vasoconstrictive Activity After Feeding Needles from Ponderosa Pine Trees to Cattle and Sheep[1]

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ABSTRACT: Consumption of needles from *Pinus ponderosa* (PN) during late pregnancy causes cattle, but not sheep, to abort. This differential response may be caused by differences in ruminal microflora or postabsorptive metabolism. Pine needles were fed (2 $kg \cdot cow^{-1} \cdot d^{-1}$ or .4 $kg \cdot ewe^{-1} \cdot d^{-1}$) mixed with corn silage. In Exp. 1, cows were assigned at 250 d of pregnancy to feed treatments (T): 1) silage, 2) PN + silage, or 3) pretreated with sheep ruminal fluid and fed PN + silage. Interval to parturition was 34.3, 11.3, and 8.3 d for the T1, T2, and T3, respectively (T1 vs T2 + T3, $P < .01$; T2 vs T3, $P > .5$). Inoculation with sheep ruminal fluid did not alter activity of the abortifacient agent of PN. In Exp. 2, pregnant and nonpregnant ewes and cows were fed silage or PN mixed with silage, and plasma was analyzed for uterine vasoconstrictive activity in an in vitro placentome perfusion bioassay. Consumption of PN decreased interval to parturition in cattle ($P < .01$) but not in sheep ($P > .5$) and increased vasoconstrictive activity ($P < .05$) in plasma from nonpregnant and pregnant cows and ewes. The PN-fed ewes had a greater incidence of dead lambs at parturition (0/8 vs 5/8 for control vs PN-fed, $P < .01$). We conclude that pregnancy is not required for increased vasoconstrictive activity induced by pine needles, that sheep and cattle do not differ in ruminal metabolism of the abortifacient compounds in PN, and that species differences are subtle and due to postdigestive differences in response to the abortifacient agent.

Key Words: Cattle, Sheep, *Pinus ponderosa*, Pine Needles, Abortion

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Introduction

Needles from Ponderosa pine trees (*Pinus ponderosa*) cause cattle and bison, but not sheep, to abort if the needles are consumed during late pregnancy (Short et al., 1992). The mechanism of action in cattle involves constriction of the maternal cotyledonary arterial bed (Christenson et al., 1992b) that results in a profound decrease in uterine blood flow, stress to the calf, and an induced parturition (Christenson et al., 1992a). Cross-species inoculation of ruminal fluid has alleviated toxic effects of some plant materials (Hammond et al., 1989; Wachenheim, et al., 1992; Kronberg and Walker, 1993). We do not know whether this is possible with the abortifacient compound found in pine needles. It is also possible that the active material gets into the blood stream but does not exert the same physiological effect in sheep that it does in cattle. Effect of pregnancy status on vasoconstrictive activity also is not known. These experiments were conducted to determine the effect that sheep ruminal microorganisms have on response of cows to pine needles, whether sheep abort and(or) have vasoconstrictive activity in blood after they consume pine needles, and whether animals must be pregnant for the vasoconstrictive activity to be present.

Materials and Methods

General experimental protocols were as described by Short et al. (1992). Pine needles (**PN**) were collected from mature Ponderosa pine (*Pinus ponderosa*) trees in Custer County, MT. Live trees were

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cut during the winter. Needles were collected by stripping at the time the trees were cut or by cutting branches and stripping needles after allowing them to dry. Air-dried needles were ground through a hammermill with a 2.25-cm screen and mixed (2 $kg\cdot cow^{-1}\cdot d^{-1}$ for cattle and .4 $kg\cdot ewe^{-1}\cdot d^{-1}$ for sheep) with the base diet at the time of feeding. Breeding dates of cows and ewes were known so that experiments could be started at specified stages of pregnancy. Cows that aborted were injected with 20 mL of an antibiotic (Penstrep®, Durvet, Blue Springs, MO) each day for 3 d after calving to prevent complications from retained placentae.

In Exp. 1, 21 cows were assigned to one of three treatments at 250 d of pregnancy. Treatments for these cows were 1) fed only corn silage, 2) fed PN mixed with corn silage, and 3) pretreated with sheep ruminal fluid and fed PN mixed with corn silage. Pine needles were fed mixed with a corn silage diet for 2 wk to six ruminally cannulated wethers. Ruminal fluid was collected from these wethers, filtered through a double layer of cheesecloth, and pooled. One liter was transferred by gavage to each Treatment 3 cow on d −8, −5, and −2 from the start of the experiment. During transfer, ruminal fluid was maintained in an anaerobic environment and at body temperature in insulated, capped containers.

In Exp. 2, pregnant and nonpregnant ewes and cows were fed corn silage or PN mixed with corn silage (Table 1). Pregnancies were at 260 ± 3 d for cows and 128 ± 2 d for sheep at the start of PN feeding (d 0).

Plasma from blood samples taken on d 0 and 7 were analyzed for uterine vasoconstrictive activity (tone) in a bovine in vitro placentome perfusion bioassay (Christenson et al., 1992b). This assay uses a perfused placentome from late-pregnant cows to measure plasma-induced changes in pressure exerted by the caruncular arterial vascular bed (i.e., maternal cotelydonary vasculature) against a constant intra-arterial perfusion flow. Effects on vasoconstrictive activity were measured as the difference in contractile response measured after a depolarizing bolus dose of KCl during perfusion (mm Hg pressure after KCl − baseline mm Hg pressure before KCl = vasoconstrictive activity) of plasma from d 0 vs d 7 for each animal (expressed as percentage change of d 7 vs d 0 response).

Statistical analyses were done by ANOVA for continuous data and by chi-square for discrete data. The models included species (only Exp. 2), treatment, and error. Orthogonal contrasts were used to separate treatment effects (Exp. 1, T1 vs T2 + T3 and T2 vs T3; Exp. 2, control vs pregnant fed PN + nonpregnant fed PN and pregnant fed PN vs nonpregnant fed PN).

Results and Discussion

In Exp. 1, interval to parturition was 34.3, 11.3, and 8.3 d for Treatments 1, 2, and 3, respectively (pooled SE = 1.97; T1 vs T2 + T3, $P < .01$; T2 vs T3, $P > .5$). There was no evidence that microflora from sheep ruminal fluid altered ruminal fermentation in cows sufficiently to modify abortifacient effects of PN.

Data from Exp. 2 are summarized in Table 1. Pine needle feeding decreased the interval to parturition in cattle ($P < .01$), but not in sheep ($P > .5$). Percentage change from d 0 to d 7 in vasoconstrictive activity of plasma was increased by feeding PN in nonpregnant and pregnant cows and ewes, but the increase was not as great for nonpregnant cows ($P < .05$). Variation in bioassay response was greater than expected (SE = 4.5) based on data from earlier studies. Pine needle-fed ewes had a greater incidence of dead lambs at parturition (0/8 vs 5/8 for control vs PN-fed, $P < .01$).

Pine needles consumed during late pregnancy caused abortions in cattle, but not in sheep, in this

Table 1. Effect of species and pregnancy status on response to pine needle (PN) feeding

Species and pregnancy status	Treatment	n	Days to parturition[a]	Change in vasoconstrictive activity, %[bc]
Cattle				
Pregnant	Control	8	22.9[d]	−1.3[d]
Pregnant	Fed PN	7	5.0[e]	24.9[e]
Nonpregnant	Fed PN	4	—	10.2[f]
Sheep				
Pregnant	Control	8	23.9[d]	−1.5[d]
Pregnant	Fed PN	8	22.0[d]	17.6[e]
Nonpregnant	Fed PN	4	—	19.5[e]

[a]Pooled SEM for cattle = 1.4, pooled SEM for sheep = 1.2.
[b]Pooled SEM = 4.5.
[c]Percentage change from d 0 to d 7 in perfusion pressure response in a placetome perfusion bioassay of plasma (see text for more details).
[d,e,f]Comparisons within column with different superscripts differ ($P < .05$) based on ANOVA and orthogonal comparisons described in the text.

study as well as in studies reported earlier (James et al., 1989; Short et al., 1992). Although sheep do not abort, there is some effect of the PN, as evidenced by an increased incidence of dead lambs and a level of plasma bioassayed vasoconstrictive activity that is similar between cattle and sheep. Previous studies (Short et al., 1992) did not report live/dead or vasoconstrictive activity data.

Pine needles cause a disturbance in ruminal digestibility (Adams et al., 1992; Pfister and Adams, 1992), but there is no evidence that digestive changes related to the abortifacient agent are different between species. Transfer of sheep ruminal fluid to cattle had no effect on abortion response. That finding agrees with the fact that we now know that sheep do not destroy the activity in PN in the rumen because activity exists in plasma.

We have previously found that there were no deleterious reproductive effects (estrous cycle length, progesterone concentration) of feeding PN to nonpregnant cows (Short et al., unpublished data). Results from the study reported here show that nonpregnant cows and ewes have assayable vasoconstrictive activity (although activity in nonpregnant cows was not as great as that in nonpregnant sheep). Therefore, the activity must be specific for the pregnant uterus. The basis for the differential response between cattle and sheep (both pregnant and nonpregnant) is not known but may involve differences in control of blood flow to the uterus, differences in fetal response to decreased blood flow, or differences in signaling mechanisms involved with initiation of parturition.

Implications

Producers that graze cattle in areas with Ponderosa pine trees risk losses caused by abortions if cows eat pine needles. Sheep do not abort after eating pine needles but do have a greater incidence of dead lambs and an increase in plasma activity of the abortifacient material. Sheep apparently do not digest the abortifacient agent in pine needles differently from cattle but do metabolize it differently or have a different physiological response after digestion. These experiments help us understand the differences between sheep and cattle and the physiological basis of the problem of pine needle-induced abortion.

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