Management of twinning cow herds

B. W. Kirkpatrick[1]

Department of Animal Sciences, University of Wisconsin, Madison 53706

ABSTRACT: Beef producers are commonly opposed to twin births owing to the number of detriments associated with the trait. These include increased incidence of calf mortality, dystocia (malpresentation), stillbirth, abortion, calf abandonment, retained placenta, lengthened interval from parturition to conception, and occurrence of freemartin heifers. Some problems can be overcome with changes in management, others lack an obvious management fix, and still others are of little practical significance. Management alterations that may facilitate successful exploitation of twin birth include 1) pregnancy checks to determine twin vs single pregnancy, 2) adequate calving facilities, and 3) early calf weaning. Determining pregnancy status eliminates surprises at calving time in that cows at high risk for malpresentation are identified beforehand. Additionally, cows gestating twins could be fed a higher plane of nutrition in recognition of the higher nutritional requirements both during late gestation and subsequently while nursing twins. Adequate calving facilities are a necessity given the significant fraction of twin births that are malpresentations. Calving pens also permit temporary penning of cows with their twins to address calf abandonment problems. Given the typically longer interval from parturition to conception for twin-bearing cows, early weaning may help maintain adequate reproductive performance. There are no obvious alterations of management that will improve twinning-related problems with stillbirth, abortion, or retained placenta. Freemartinism is an item that is more a perceived than real problem; although the percentage of fertile females is reduced in a twinning system, the absolute number of fertile females produced from a twinning system will differ little from that from a single-birth system.

Key Words: Calves, Cows, Reproduction, Twins

 J. Anim. Sci. 80(E. Suppl. 2):E14–E18

Introduction

Twin birth offers the potential for increased beef production efficiency if suitable changes in management can be made to accommodate problems inherent with twinning. Twin birth is undesired by most beef producers for its association with a number of detriments, including lower perinatal calf survival and poorer cow reproductive performance. Regarding cow performance, twinning is associated with increased dystocia (due to malpresentation), increased incidence of retained placenta, greater frequency of abortion in twin gestation, and longer interval from parturition to first estrus (Table 1). Results cited in Table 1 represent only the most recent reports in the literature. Descriptions in the scientific literature of the association between twin birth and cow or calf performance can be found from prior to the 1930s up to the present. The results of these numerous surveys and studies have generally been consistent in their identification of problems associated with twinning with one exception, that being dystocia. Inconsistent results for incidence of dystocia may reflect two competing dynamics: twinning reduces incidence of dystocia attributable to large calf size but increases incidence of dystocia attributable to malpresentation. Efforts to exploit twin birth in beef production systems will require changes in management to address these problems.

Diagnosing Twin Pregnancy

In typical beef herds, twins are a relatively unusual event. Reviews on the genetics of twinning (Rutledge, 1975; Morris and Day, 1986) suggest that twinning rate in beef breeds is typically less than 5%. As a consequence, most twin births in beef cattle herds are unanticipated events. Because they are unanticipated, most producers are not prepared to address complications, such as malpresentation, that may arise. One of the most important changes in management that should accompany efforts to exploit twinning is determination of pregnancy status with regard to single vs twin preg-

[1]Correspondence: phone: 608-263-4323; fax: 608-262-5157; E-mail: kirkpat@calshp.cals.wisc.edu.

Received July 27, 2001.

Accepted December 3, 2001.

Table 1. Detrimental association of twinning with calf and cow performance

Item	Single birth	Twin birth	*P*-value
Perinatal calf mortality, %			
Mee, 1991	6.2	16.5	<0.001
Day et al., 1995	3.2	15.7	<0.005
Gregory et al., 1990	5	22	<0.01
Cady and Van Vleck, 1978	5.9	22.4	<0.05
Retained placenta, %			
Anderson et al., 1979	0	14	NS[a]
Gregory et al., 1990	2.8	21.5	<0.01
Eddy et al., 1991	2	16	<0.001
Echternkamp and Gregory, 1999a	1.9	27.9	<0.01
Dystocia, %			
Anderson et al., 1979	20	21	NS
Gregory et al., 1990	23	35	<0.01
Berry et al., 1994, 1st lactation	30.6	34.5	NS
Berry et al., 1994, 2nd lactation	11.8	30.0	<0.05
Eddy et al., 1991	5	7	NS
Mee, 1991	46.4	45.6	NS
Echternkamp and Gregory, 1999a	20.6	46.9	<0.01
Abortion frequency, %			
Mee, 1991	1.81	1.20	NS
Day et al., 1995	12.0	29.3	<0.05
Echternkamp and Gregory, 1999b	3.5	12.4	<0.01
Stillborn calves, %			
Mee, 1991	4.1	12.9	<0.001
Interval, parturition to conception, d			
Gregory et al., 1990b	83	93	<0.01
Eddy et al., 1991	92	125	<0.001

[a]NS = not significant.

nancy. Available methods for diagnosing pregnancy status include transrectal palpation of the reproductive tract or transrectal ultrasonography to visualize the reproductive tract. Palpation can be used to determine pregnancy status but may provide only modest sensitivity. Day et al. (1995), in a report utilizing data from practicing veterinarians in commercial California dairy herds, reported a sensitivity for diagnosis of twin pregnancy of 49.3% (meaning that in just under half the cases when a cow was actually pregnant with twins was a diagnosis of twin gestation made). Ultrasonography represents a more sensitive means of determining pregnancy status as documented by several studies (Davis and Haibel, 1993; Dobson et al., 1993; McMillan et al., 1994). Davis and Haibel (1993) reported more accurate diagnosis of twin pregnancy when ultrasonography was performed between 49 and 55 d of gestation relative to earlier dates, with no improvement in accuracy by examination at later dates. From the data provided by the Davis and Haibel (1993), sensitivity of ultrasonic diagnosis of twin pregnancy between averages of 49 to 55 d of gestation was 93%. McMillan et al. (1994) reported lower accuracy (40 to 62%) when making a diagnosis at 45 or 60 d of gestation, which may in part reflect a higher rate of subsequent embryonic or fetal loss. As in the two previously mentioned studies, Echternkamp and Gregory (1991) reported greater accuracy (65%) in diagnosis of twin pregnancy at 46 to 79 d of gestation vs 28 to 45 d (57%). They also noted difficulty in performing ultrasonic evaluation after 80 d of gestation. Examination at later stages of gestation may be complicated by physical size and location of the uterus. As a consequence, a herd of beef cows might need to be pregnancy-checked by ultrasound on two different occasions to ensure that all animals are at one time or the other in a stage of gestation that permits accurate ultrasonic evaluation. A likely constraint to the application of this technology for many producers is the availability of equipment or ultrasonography services at a justifiable cost.

A future, alternative approach for determining pregnancy status may be evaluation of hormone or protein levels associated with luteal, fetal, or placental tissues. Among factors that display differing levels between single- and twin-gestating cows are progesterone (Dobson et al, 1993), estrone sulfate (Dobson et al., 1993; Takahashi et al., 1997), cortisol (Patel et al., 1996), bovine pregnancy-specific protein B (Patel et al., 1995; Dobson et al, 1993), bovine pregnancy serum protein 60 (**PSP60**) (Patel et al., 1998; Chauvin et al., 1999), and pregnancy-associated glycoprotein (Zoli et al., 1991). Variability in some of these factors is too great for practical application as predictors of twin vs single pregnancy; however, others such as PSP60 show potential value. Chauvin et al. (1999) in work with a twinning beef cattle population found that a diagnostic test based on a single measurement of PSP60 had a sensitivity of 77.2% (percentage of cows carrying twins correctly diagnosed) and specificity of 75.9% (percentage of cows carrying singles correctly diagnosed). An assay of this

type provides an advantage over ultrasound of being applicable at a later stage of gestation. However, further work is needed to attain an assay with acceptable sensitivity, specificity, and cost.

Dealing with Dystocia

Twin-bearing cows are at higher risk than single-bearing cows for dystocia due to malpresentation. Echternkamp and Gregory (1999a) in a study of 3,370 single and 1,014 twin births reported an incidence of malpresentation of 4.5% for single births and 37.0% for twin births. Therefore, a prior diagnosis of twin pregnancy is valuable to producers in alerting them to cows that are more likely to encounter problems at calving. In addition to diagnosing pregnancy status, as described above, producers who wish to exploit twinning in cattle would be well served by investing in some basic calving facilities. Calving pens with a headgate and a gate that doubles as a crowding gate are relatively simple and inexpensive to construct and reduce stress on both animal and handler. Facilities should be designed so that it is easy to move cows into the calving pens.

In the event of a malpresentation with twin calves, it will be necessary to pen the cow and reposition the calves prior to delivery. Although this is not a trivial issue, adequate facilities greatly simplify the task, and compared with singletons, twins are easier to reposition due to their lighter weight.

Nutritional Requirements of Twinning Cows

In addition to giving producers foreknowledge of potential calving problems, diagnosis of twin pregnancy would also permit producers to modify feeding programs for twin-bearing cows during the last third of gestation. Twin pregnancy has a higher metabolizable energy (ME) requirement than does single pregnancy, and dams gestating twins lose maternal weight during the last weeks of gestation, whereas dams of singles gain weight during the same period if fed equivalent diets (Koong et al., 1982). Koong et al. (1982) found no difference in maintenance energy requirements for dams carrying twin vs single fetuses, only a difference in pregnancy energy requirement (6.49 ± 0.26 and 6.09 ± 0.28 Mcal ME/d for twin-bearing cows and heifers, 3.80 ± 0.29 and 3.89 ± 0.22 Mcal ME/d for single-bearing cows and heifers). Using the values reported by Koong et al. (1982) and assuming a 600-kg cow, daily dietary energy would need to be increased about 13% for twin-bearing cows relative to single-bearing cows. Bunge et al. (1997) reported a similar value of 11% greater energy requirement for heifers gestating twins in a separate study.

Early Weaning of Calves

Most data suggest that twinning cows are at risk for longer intervals from parturition to conception. In a study using several thousand cumulative records from the USDA twinning cattle population, Echternkamp and Gregory (1999b) reported a greater than 12-d longer interval from parturition to conception for cows giving birth to and suckling twins vs cows birthing and suckling singles. Corresponding differences in conception rate were observed between the two groups; 79.2% of single-bearing cows conceived in a 60- to 70-d breeding period vs 66.3% for twin-bearing and twin-suckling cows. Turman et al. (1971), in a much smaller study, used superovulation to create multiple births. All dams suckling single calves rebred in a 90-d breeding period but 25% (5/20) of cows suckling twins failed to rebreed during the same period. Wheeler et al. (1982) used embryo transfer to create twin births in heifers and cows. Averaged across age groups, twin-bearing and -rearing females experience an interval from parturition to first ovulation that was approximately 17 d longer than that for single-bearing and -rearing females; however, this difference was not statistically significant. Differences between these same groups in interval from parturition to conception were less for females bearing and rearing twins, but again differences were not statistically significant. Other studies have simulated twinning postnatally by forced adoption of an additional calf on a dam of a single calf. In one such study, suckling two calves vs one caused a significant delay in return to estrus; at 90 d postpartum, 71.4 and 88.8% of cows suckling a single natural or foster calf, respectively, had returned to estrus vs 42.8% for cows suckling two calves (Wyatt et al., 1977; Wettemann et al., 1978). Single- and twin-suckling cows in this study were fed differing amounts of concentrate to achieve similar weight changes in both groups over the suckling period. In contrast, a more recent study with a very similar design failed to detect an effect of single vs twin suckling on percentage of females expressing estrus, interval from parturition to first estrus, or first-service conception rate (Small et al., 2000). One aspect of the studies that may have contributed to the different outcomes was use of beef $\times$ dairy cross cows in the former (Holstein $\times$ Hereford) and beef cows (primarily Hereford breeding) in the latter. Assuming that suckling two calves does predispose a cow to delayed conception, one answer would be to remove the suckling stimulus by early calf weaning.

Early weaning (i.e., at 6 to 8 wk of age or younger) has been well documented to improve postpartum reproductive performance. Laster et al. (1973) weaned calves 1 wk prior to the start of the breeding season at an average age of 55 d. The percentage of 2-, 3-, and 4-yr-old and older cows conceiving in a 42-d breeding season was increased by 25.9, 15.6 and 7.9%, respectively, for early-weaned calves vs those weaned after the breeding period. Similarly, Lusby et al. (1981) compared calf weaning at 6 to 8 wk of age with weaning at 7 mo of age for first-calf heifers and reported an increase in the percentage of females conceiving in a 64-d breeding season of 37.4%. Correspondingly, the

interval from parturition to conception was 17.5 d less for the early-weaned group. Houghton et al. (1990) reported a significant 24-d reduction in postpartum interval to first estrus for early-weaned vs normally weaned cows in a study using mature beef cows. However, no effect on conception rate was observed despite the reduced postpartum anestrus interval. Bell et al. (1998) compared early weaning, once-daily milking, and normal weaning using first-calf heifers and reported a reduction in days to first estrus of 17 d for early-weaned vs normally weaned cows among those that were less than 85 d postpartum at initiation of the study. Considering results from these various studies, early weaning of twin calves may be a way to improve postpartum reproductive performance of twin-bearing and twin-suckling cows, particularly those of younger age.

Maternal Behavior and Calf Acceptance

A common concern among producers is the propensity for cows that bear twins to accept only one of the twins and to abandon the other. This problem is seldom encountered in the USDA Meat Animal Research Center (**MARC**) twinning herd (Gordon Hayes, personal communication), and the lack of a problem is attributed to their management practice of routinely penning twins with their mother for 24 h or longer before they are moved to pasture with other cows with twins. Likewise, in the author's experience it has not been a significant problem. In the author's twinning herd only cows requiring assistance during delivery are routinely penned with their calves for 24 h. Cows giving birth to twins without assistance who show signs of not accepting both calves are penned with their twins for approximately 24 h and that is usually sufficient to correct the situation (personal observation).

Other Issues

Several problems that accompany twin birth have no easy management fix. Included among these are the greater frequency of abortion, stillbirth, and retained placenta. These are important issues that are detrimental to the reproductive efficiency of a beef cow herd.

Some authors have suggested that the incidence of retained placenta may be affected by nutritional regimen (Studer, 1998), particularly in the context of dairy cattle managed for high production levels. Dairy cows that are in excessive condition at calving or are subject to imbalanced rations at the transition from the dry period to lactation are thought to be at higher risk for retained placenta (Studer, 1998). Whether this is a suitable comparison with twinning beef cattle is unclear. Gestation length in twinning cattle is roughly 5 to 7 d shorter in duration than for cattle bearing singles (Cady and Van Vleck, 1978; Anderson et al., 1982; Echternkamp and Gregory; 1999a). As a consequence, retained placenta in twinning cattle may more likely result from the slightly premature parturition. Several studies with dairy cattle have suggested that vitamin E administration 1 to 2 wk prior to parturition can reduce the incidence of retained placenta (Trinder et al., 1973; Julien et al., 1976), although results have varied (Batra et al., 1992). Again, whether this would be an effective approach for preventing twinning-related incidences of retained placenta is unknown. There is no clearly effective treatment for retained placenta (reviewed by Peters and Laven, 1996), so in twinning cows that do experience retained placenta the most cost-effective approach to treatment may simply be no intervention.

It is difficult to unambiguously distinguish stillborn calves that are delivered near full-term from perinatal calf death losses, unless personnel are present at birth. Gregory et al. (1996) in examining extensive records from the MARC twinning herd report survival rates at birth for single and twin calves of 96.4 and 83.4%, respectively. The authors report that this accounts for prematurely delivered calves, which would presumably include stillbirths, so that higher mortality of twins in this report probably reflects both the influence of dystocia and stillbirth combined. Although calf losses due to dystocia can probably be reduced by providing assistance at parturition, there is no clear means of preventing early-term abortions, late-term abortions, or stillbirths associated with twinning cows. It is possible that different individuals or genetic lines of cattle may have varying predisposition to carry multiple fetuses successfully to term. In the longer term, current efforts to identify specific genetic loci controlling ovulation rate (Blattman et al., 1996; Kappes et al., 2000; Kirkpatrick et al., 2000) may facilitate introgression of specific genes into different populations to test this hypothesis. Unfortunately, in the short term there is no management strategy that will clearly improve this aspect of reproductive performance.

Implications

Exploitation of twin birth presents a potential means of dramatically improving efficiency of beef production. Successful use of twinning in beef cattle production will require changes in management to address problems of increased dystocia and calf mortality and poorer postpartum reproduction. Key changes in management may include diagnosis of twin vs single pregnancy, modification of nutrition for cows bearing or suckling twins, and use of early weaning.

Literature Cited

Anderson, G. B., P. T. Cupps, and M. Drost. 1979. Induction of twins in cattle with bilateral and unilateral embryo transfer. J. Anim. Sci. 49:1037–1042.

Anderson, G. B., R. H. BonDurant, and P. T. Cupps. 1982. Induction of twins in different breeds of cattle. J. Anim. Sci. 54:485–490.

Batra, T. R., M. Hidiroglou, and M. W. Smith. 1992. Effect of vitamin E on incidence of mastitis in dairy cattle. Can. J. Anim. Sci. 72:287–297.

Bell, D. J., J. C. Spitzer, and G. L. Burns. 1998. Comparative effects of early weaning or once-daily suckling on occurrence of postpartum estrus in primiparous beef cows. Theriogenology 50:707–715.

Berry, S. L., A. Ahmadi, and M. C. Thurmond. 1994. Periparturient disease on large, dry lot dairies: Interrelationships of lactation, dystocia, calf number, calf mortality, and calf sex. J. Dairy Sci. 77 (Suppl. 1):379 (Abstr.).

Blattman, A. N., K. E. Gregory, and B. W. Kirkpatrick. 1996. A search for quantitative trait loci for ovulation rate in cattle. Anim. Genet. 27:157–162.

Bunge O., H. J. Papstein, and K. Ender. 1997. Studies on the growth, gain and carcass value of primipara during singleton and twin pregnancies. 2. Gain of body substances and energy, birth weights and courses of calving. Zuechtungskunde 69:20–30.

Cady, R. A., and L. D. Van Vleck. 1978. Factors affecting twinning and effects of twinning in Holstein dairy cattle. J. Anim. Sci. 46:950–956.

Chauvin, C., S. Camous, P. Gillard, P. Maugrion, C. Robert, and J. L. Foulley. 1999. PSP60 profile and its variation factors and application to twinning diagnosis in a beef suckling herd. Rev. Med. Vet. 150:143–150.

Davis, M. E., and G. K. Haibel. 1993. Use of real-time ultrasound to identify multiple fetuses in beef cattle. Theriogenology 40:373–382.

Day, J. D., L. D. Weaver, and C. E. Franti. 1995. Twin pregnancy diagnosis in Holstein cows—discriminatory powers and accuracy of diagnosis by transrectal palpation and outcome of twin pregnancies. Can. Vet. J. 36:93–97.

Dobson, H., T. G. Rowan, I. S. Kippax, and P. Humblot. 1993. Assessment of fetal number, and fetal and placental viability throughout pregnancy in cattle. Theriogenology 40:411–425.

Echternkamp, S. E., and K. E. Gregory. 1991. Identification of twin pregnancies in cattle by ultrasonography. J. Anim. Sci. 69 (Suppl. 1):220 (Abstr.).

Echternkamp, S. E., and K. E. Gregory. 1999a. Effects of twinning gestation length, retained placenta, and dystocia. J. Anim. Sci. 77:39–47.

Echternkamp, S. E., and K. E. Gregory. 1999b. Effects of twinning on postpartum reproductive performance in cattle selected for twin birth. J. Anim. Sci. 77:48–60.

Eddy, R. G., O. Davies, and C. David. 1991. An economic assessment of twin births in British dairy herds. Vet. Rec. 129:526–529.

Gregory, K. E., S. E. Echternkamp, and L. V. Cundiff. 1996. Effects of twinning on dystocia, calf survival, calf growth, carcass traits and cow productivity. J. Anim. Sci. 74:1223–1233.

Gregory, K. E., S. E. Echternkamp, G. E. Dickerson, L. V. Cundiff, R. M. Koch, and L. D. Van Vleck. 1990. Twinning in cattle: III. Effects of twinning on dystocia, reproductive traits, calf survival, calf growth and cow productivity. J. Anim. Sci. 68:3133–3144.

Houghton, P. L., R. P. Lemenager, L. A. Horstman, K. S. Hendrix, and G. E. Moss. 1990. Effects of body composition, pre- and postpartum energy level and early weaning on reproductive performance of beef cows and preweaning calf gain. J. Anim. Sci. 68:1438–1446.

Julien, W. E., H. R. Conrad, and A. L. Moxon. 1976. Selenium and vitamin E and incidence of retained placenta in parturient dairy cows. II. Prevention in commercial herds with prepartum treatment. J. Dairy Sci. 59:1960–1966.

Kappes, S. M., G. L. Bennett, J. W. Keele, S. E. Echternkamp, K. E. Gregory, and R. M. Thallman. 2000. Initial results of genomic scans for ovulation rate in a cattle population selected for increased twinning rate. J. Anim. Sci. 78:3053–3059.

Kirkpatrick, B., B. M. Byla, and K. E. Gregory. 2000. Mapping quantitative trait loci for bovine ovulation rate. Mamm. Genome 11:136–139.

Koong, L. J., G. B. Anderson, and W. N. Garrett. 1982. Maternal energy status of beef cattle during single and twin pregnancy. J. Anim. Sci. 54:480–485.

Laster, D. B., H. A. Glimp, and K. E. Gregory. 1973. Effects of early weaning on postpartum reproduction of cows. J. Anim. Sci. 36:734–740.

Lusby, K. S., R. P. Wettemann, and E. J. Turman. 1981. Effects of early weaning calves from first-calf heifers on calf and heifer performance. J. Anim. Sci. 53:1193–1197.

McMillan, W. H., A. P. Oakley, and D. R. H. Hall. 1994. Determining the number of calves in early pregnancy using real-time ultrasound imaging in beef cows induced to twin. Proc. N. Z. Soc. Anim. Prod. 54:353–355.

Mee, J. F. 1991. Factors affecting the spontaneous twinning rate and the effect of twinning on calving problems in 9 Irish dairy herds. Irish Vet. J. 44:14–20.

Morris, C. A., and A. M. Day. 1986. Potential for genetic twinning in cattle. In: Proc. 3rd World Congr. Genet. Appl. to Livest. Prod., Lincoln, Nebraska. 11:14–29.

Patel, O. V., S. Camous, N. Takenouchi, T. Takahashi, M. Hirako, N. Sasaki, and I. Domeki. 1998. Effect of stage of gestation and foetal number on plasma concentrations of a pregnancy serum protein (PSP-60) in cattle. Res. Vet. Sci. 65:195–199.

Patel, O. V., I. Domeki, N. Sasaki, T. Takahashi, M. Hirako, R. G. Sasser, and P. Humblot. 1995. Effect of fetal mass, number and stage of gestation on pregnancy-specific protein-B concentrations in the bovine. Theriogenology 44:827–833.

Patel, O. V., T. Takahashi, N. Takenouchi, M. Hirako, N. Sasaki, and I. Domeki. 1996. Peripheral cortisol levels throughout gestation in the cow: Effect of stage of gestation and foetal number. Br. Vet. J. 152:425–432.

Peters, A. R., and R. A. Laven. 1996. Treatment of bovine retained placenta and its effects. Vet. Rec. 139:535–539.

Rutledge, J. J. 1975. Twinning in cattle. J. Anim. Sci. 40:803–815.

Small, J. A., E. Charmley, and A. D. Kennedy. 2000. The performance of primiparous and multiparous beef cows rearing single and simulated-twin calves. Can. J. Anim. Sci. 80:569–576.

Studer, E. 1998. A veterinary perspective of on-farm evaluation of nutrition and reproduction. J. Dairy Sci. 81:872–876.

Takahashi, T., M Hirako, H. Takahashi, O.V. Patel, N. Takenouchi, and I Domeki. 1997. Maternal plasma estrone sulfate profile during pregnancy in the cow: Comparison between singleton and twin pregnancies. J. Vet. Med. Sci. 59:287–288.

Trinder, N., R. J. Hall, and C. P. Renton. 1973. The relationship between the intake of selenium and vitamin E on the incidence of retained placenta in dairy cows. Vet. Rec. 93:641–646.

Turman, E. J., D. B. Laster, R. E. Renbarger, and D. F. Stephens. 1971. Multiple births in beef cows treated with equine gonadotropin (PMS) and chorionic gonadotropin (HCG). J. Anim. Sci. 32:962–967.

Wettemann, R. P., E. J. Turman, R. D. Wyatt, and R. Totusek. 1987. Influence of suckling intensity on reproductive performance of range cows. J. Anim. Sci. 47:342–346.

Wheeler, M. B., G. B. Anderson, R. H. BonDurant, and G. H. Stabenfeldt. 1982. Postpartum ovarian function and fertility in beef cattle that produce twins. J. Anim. Sci. 54:589–593.

Wyatt, R. D., M. B. Gould, and R. Totusek. 1977. Effects of single vs simulated twin rearing on cow and calf performance. J. Anim. Sci. 45:1409–1414.

Zoli, A. P. L. A. Builbault, P. Delahaut, W. B. Ortiz, and J. F. Beckers. 1991. Radioimmunoassay of a bovine pregnancy-associated glycoprotein in serum: Its application for pregnancy diagnosis. Biol. Reprod. 46:83–92.