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Review article

The dilemma of twin pregnancies in dairy cattle. A review of practical prospects

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ABSTRACT

Carrying twins has been extensively described as an emerging principal non-infectious factor jeopardizing pregnancy maintenance and reducing the lifespan of dairy cows. The risk of pregnancy loss during the first trimester of gestation for cows carrying twins may be from three to seven times higher than for cows carrying singletons. Longer calving to conception intervals, higher culling rates and a shorter mean production lifespan of 200 days have been reported for cows delivering twins compared to cows delivering singletons. There is therefore a need accurately to detect twin embryos at the time of pregnancy diagnosis to follow twin pregnancies carefully or, alternatively, to remove one of the embryos. Therapeutic approaches for the problem of twin pregnancies include GnRH treatment or induced embryo reduction. With similar results, manual rupture of the amniotic vesicle or transvaginal ultrasound-guided aspiration of allanto-amniotic fluid have been proposed as methods of choice to perform twin reduction in cows on Day 28–41 of gestation. However, benefits and risks of induced twin reduction should be quantified. This report reviews various aspects concerning control of twin pregnancies and the practical implications at herd level. Special attention is paid to timing of spontaneous twin reduction. Prospects for induced embryo reduction are also discussed.

1. Introduction

Bos taurus females are generally considered as monovular. However, the incidence of multiple ovulation and thus twinning are increasing under certain conditions. Although during the 1970s and 1980s some studies sought to increase the twinning rate to improve milk production and progeny per cow (Rutledge, 1975; Van Vleck et al., 1991; Echternkamp, 1992), most authors today would agree that twin births are not desirable in dairy herds (Echternkamp and Gregory, 1999b; Bicalho et al., 2007; Andreu-Vázquez et al., 2012a). Higher incidences of dystocia, freemartins, stillbirths, retained placenta and calf mortality have been related to twin births (Mee, 1991; Beerepoot et al., 1992; Echternkamp and Gregory, 1999a; Andreu-Vázquez et al., 2012a). Thus, longer calving to conception intervals, higher culling rates and a shorter mean production lifespan of 200 days have been reported for cows delivering twins compared to cows delivering singletons (Nielen et al., 1989; Eddy et al., 1991; Bicalho et al., 2007; Andreu-Vázquez et al., 2012a). In effect, twin pregnancies reduce herd profitability, with an average loss of income attributed to cows delivering twins estimated at 74–108$ (Eddy et al., 1991; Beerepoot et al., 1992). From a national perspective in the US, costs related to twinning amount to $55 million per year (Johanson et al., 2001). However, the real economic impacts of twinning are probably on the rise since twinning rates have increased considerably over the past three decades (Nielen et al., 1989; Kinsel et al., 1998; Silva del Rio et al., 2007). Estimates currently run up to 12% in some herds (Silva del Rio et al., 2007) and 10% of cows delivering twins at least once during their life (Andreu-Vázquez et al., 2012a). This increased twinning rate has been linked to increased milk productivity in an epidemiological study on 52,362 lactations (Kinsel et al., 1998). In high producers, the rate of double ovulation may be over 20% (Fricke and Wiltbank, 1999) and 25% for cows in their third or later lactation (López-Gatius et al., 2005). Genetics appears to be a major regulatory factor for twinning rates. In an analysis of data related to 1,324,678

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offspring of 37,174 sires, it was shown that sires born after 1990 had a higher incidence of twins than sires born before 1980 (Johanson et al., 2001). It is reasonable to think that increased twinning is a consequence of selection for milk yield. However, not only genetic progress but also improvements in nutrition and management practices have led to a continuous increase in milk yield. Probably improvements at the farm level related to increased milk productivity have diminished the risk of embryo loss in twin pregnancies and thus raised the twinning rate. Finally, breeding synchronization protocols for fixed-time artificial insemination are becoming standard components of the current breeding management of lactating cows, and some of them can increase the twin pregnancy rate (Andreu-Vázquez et al., 2012b). It is therefore foreseeable that over the years to come, the twinning rate will continue to increase along with milk productivity. Obviously, the incidence of twin births is closely related to the twin pregnancy rate.

In the modern high producing dairy herds, the parturition-calving interval is a key factor for herd survival. During the postpartum period, cows not only have to get pregnant but are also required to maintain their pregnancy until parturition. The embryonic period of pregnancy runs from conception until the end of the differentiation stage (about 45 days), and the fetal period spans from Day 45 of pregnancy to parturition (Committee on Bovine Reproductive Nomenclature, 1972). Pregnancy loss during the late embryonic/early fetal period is an important factor for herd economy, especially in high production systems in which losses of 10–12% are a common figure (Kidder et al., 1954; Santos et al., 2004; López-Gatius, 2012). Carrying twins is a main non-infectious factor compromising pregnancy maintenance following a positive pregnancy diagnosis in dairy cows (López-Gatius et al., 2002, 2004a; Silva-del-Río et al., 2009), exceeding 18% the twin pregnancy rate in some herds (Andreu-Vázquez et al., 2012b). The risk of pregnancy loss during the first trimester of gestation for cows carrying twins may be from three to seven times higher than for cows carrying singletons (López-Gatius et al., 2002, 2006; García-Ispierto et al., 2006; Silva-del-Río et al., 2009) and from five to nine times higher for unilateral than for bilateral twins (Hanrahan, 1983; López-Gatius and Hunter, 2005; Andreu-Vázquez et al., 2011). Although they tend to be cases of twin pregnancies, triplet or even quadruple ones are not exceptional.

Previous studies have addressed the factors affecting the double ovulation and twin birth rates (Nielen et al., 1989; Kinsel et al., 1998; Fricke and Wiltbank, 1999; Lopez et al., 2005; López-Gatius et al., 2005). However, neither the incidence of twins at calving nor double ovulation are a true reflection of twin pregnancy. A very high frequency of double ovulations (13.1%) compared with their corresponding twin births (1.9%) has already been described in the 1950's (Kidder et al., 1952). This enormous difference suggests a high incidence of fertilization failure and/or embryonic mortality in multiple ovulating cows. Evaluating for pregnancy during the first trimester of gestation, multiple gestations and fetal viability are ideal mechanisms for dealing with large herds of animals (López-Gatius, 2012). This article reviews aspects concerning control of twin pregnancies following pregnancy diagnosis. Most of the data were obtained by our research group from research conducted on commercial high-producing Holstein-Friesian dairy herds in North-Eastern Spain. Special attention is paid to timing of spontaneous twin reduction and to practical prospects of twin pregnancy management at herd level. Approaches for induced embryo reduction are also discussed.

2. Diagnosing twin pregnancies

Although plasma concentrations of pregnancy-associated glycoproteins have been recently used to discriminate between singleton and twin pregnancies (Szelényi et al., 2015; García-Ispierto et al., 2016), ultrasound imaging is routinely used for twin pregnancy diagnosis in dairy cattle (López-Gatius and García-Ispierto, 2010). The first problem in diagnosing twin pregnancies is that the two embryos must be clearly observed. Evaluation of ovarian structures opposed to uterine contents is particularly important for twin pregnancy diagnosis. The detection of two or more corpora lutea in a cow can lead to both types of error: a single pregnancy can be diagnosed in a cow carrying twins (false positive), or on the contrary, a single pregnancy often is diagnosed as a twin pregnancy (false negative). In the latter case, which usually involves two corpora lutea located in the same ovary, the observed second embryo could be the first one which has moved from the initial screen site. Thus, a too fast uterine examination can lead to false negative diagnoses. Special efforts should be made to find twins in cows with two or more corpora lutea. Further, in cows carrying twins with a single corpus luteum, probably monozygotic twins, a single pregnancy is often diagnosed. Upon detecting the presence of an embryo, efforts are not usually made to search for a possible second embryo. Although the proportion of monozygous twinning has been reported as very low in dairy cattle over the total births, its incidence of 5.5% among all twin births has a clear interest at herd level (Silva-del-Río et al., 2006).

The major difficulties for twin pregnancy diagnosis are unilateral twin pregnancies, involving cows carrying twins with a single corpus luteum (possibly monozygotic twins). Since an embryo found in the uterine horn contralateral to the corpus luteum is a very rare phenomenon in the cow, with rates from 0% to 1.2% (Boyd et al., 1944; Perkins et al., 1954; Scanlon, 1972; Serrano et al., 2009), it is best always to carefully inspect the uterine horn ipsilateral to the corpora lutea for the possible presence of an embryo or twins. Finally, although the use of ultrasound ensures that the embryo can be detected in all cases between Days 25 and 30 of gestation (Ginther, 1998), the accuracy to detect twins is lower until Day 30, especially in the case of unilateral twin pregnancies (López-Gatius and García-Ispierto, 2010). A useful indicator of the presence of unilateral twins is the common presence of an easily traceable hyperechoic line from embryo to embryo (we can “follow” the line). This echoic line represents the area of apposition between the two chorionic membranes (López-Gatius and García-Ispierto, 2010).

3. Spontaneous reduction of twin embryos

There are clearly situations in which the number of embryos in the early stages of a multiple pregnancy becomes reduced, this usually being termed “spontaneous reduction” (López-Gatius and García-Ispierto, 2010; López-Gatius, 2012). In reality, it is not spontaneous at all. An embryo may die due to expression of an inadequate gene program, itself primarily due to post-ovulatory ageing of a secondary oocyte before fertilization (Hunter, 1989). An alternative route to loss is inadequate progesterone support of the placental membranes including specialized modifications of the endometrium. This can occur in a retarded embryo or be due to an imbalanced vascular delivery of systemic progesterone to two or more embryos under certain types of stress (López-Gatius et al., 2010). However, since embryo reduction in multiple pregnancies escapes often clinical control, we maintain here the term of spontaneous reduction of twin embryos.

Spontaneous embryo reduction has been described in natural twin pregnancies in dairy cattle, with incidences from 11.2% to 28.4% (López-Gatius and Hunter, 2005; Silva-del-Río et al., 2009; López-Gatius et al., 2010; Andreu-Vázquez et al., 2011). Loss of a corpus luteum was noted in more than 25% of cows which suffering embryo reduction remained pregnant (López-Gatius et al., 2010). In the latter study it was demonstrated that both corpora lutea and embryos are vulnerable to the effects of stress factors such as heat stress in cows maintaining their pregnancies (López-Gatius et al., 2010). Embryo reduction is also common (33%) in twin pregnancies following embryo transfer of two or three embryos in beef cattle (Izaike et al., 1988, 1991). Although cows experiencing a single embryo death can remain pregnant, more than 60% of them can suffer pregnancy loss 1–4 weeks later (López-Gatius et al., 2009). Interestingly, most cases of single