The importance of recording in establishing the value of sexed semen to dairy farmers.

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Abstract

One of the biggest developments since the advent of artificial insemination over five decades ago is the commercialisation of sexed semen. Semen contains sperm bearing approximately equal numbers of X or Y chromosomes, resulting in female or male offspring, respectively. Sorting bovine sperm by flow cytometry has improved considerably and is over 90% accurate, generating so called ‘sexed semen’. Poorer conception rates are generally noted for sexed semen compared with conventional semen (~80% of conventional), restricting its use to primarily on heifers.

Dairy farmers often rely on circumstantial evidence to assign calf to cow after the calving process and thereby establish parentage of the calf. It is anecdotally reported that mistakes in assigning parentage to calf, circumstantially, is in excess of 15% and with the use of sexed semen this will likely increase. In a well constructed artificial insemination programme, it is not unreasonable to expect around 25 heifer calves for every 100 cows inseminated. In a sexed semen programme, despite lower conception rates, the expected number of heifer calves for every 100 cows inseminated would be around 40 (a 60% increase in heifer calf numbers).

Preliminary results from New Zealand suggest that using fresh sexed semen results in conception rates that are 94% of those achieved with conventional frozen-thawed semen in both heifers and lactating cows. The challenge for recording is to ensure that each insemination event distinguishes the type of semen product used from a given sire (i.e., frozen-thawed vs. fresh; sorted vs. non-sorted) to allow appropriate evaluation of fertility performance of both the sire and the cow. This paper will discuss the technology developments in sexed semen and the challenges that dairy farmers will face with increasing use of this animal multiplication technology.

Introduction

The ability to select the sex of offspring at conception is one of the most sought after reproductive biotechnologies of all time. Female dairy offspring are more desirable than male offspring, particularly with the impending removal of the European Union milk quota regime. Semen contains approximately equal numbers of sperm containing X or Y chromosomes, resulting in female or male offspring, respectively. A major breakthrough in the development of sexed semen came when it was observed that sperm containing X-chromosomes contain more DNA (c. 4.2 %) than sperm containing Y-chromosomes (Moruzzi, 1979). Fluorescence activated cell sorting (FACS), a type of flow cytometry, was then identified as a reliable method of sorting sperm based on their DNA content (Garner et al., 1983), in order to produce semen enriched in either X- or Y-chromosome bearing sperm.

Fluorescence Activated Cell Sorting
Since the initial inception of FACS as a method to distinguish populations of X- and Y-chromosome bearing sperm, numerous refinements to the procedure have taken place in order to facilitate the commercial application of this technology (Sharpe and Evans, 2009), which consistently produces approximately 90% gender bias amongst offspring.

Sperm are stained with a non-toxic, DNA-binding dye (Hoechst 33342) and pumped in a stream in front of a laser beam (Johnson and Welch, 1999). A crystal vibrator is used to break the stream into individual droplets, to facilitate analysis of individual sperm. The illuminated stained sperm emit a bright fluorescence, which is rapidly measured by a photo-multiplier tube as the sperm flow past in single file (Garner and Seidel Jr, 2008). The sperm must be oriented at the appropriate angle to the laser to ensure adequate illumination for subsequent accurate measurement of a 4% difference in fluorescence (Sharpe and Evans, 2009). A high-speed computer is used to analyse the relative fluorescence of the X- and Y- sperm populations, which are then sorted by DNA content by placing opposite charges on droplets containing X-sperm from those containing Y-sperm (Seidel Jr, 2007). The droplets fall past charged deflector plates, and separate into two streams for collection. A third stream of uncharged droplets passes through as waste and is discarded (Seidel Jr, 2007).

**Challenges and limitations of FACS for sorting semen**

The primary limitations of using FACS to sort semen are: (i) the slow speed of the process relative to the number of viable sperm required for artificial insemination in cattle; and (ii) the high proportion of sperm cells that are lost, cannot be oriented for sorting, or cannot be accurately identified as bearing an X or Y chromosome and pass through without being sorted (combined >75% loss) (Seidel Jr and Garner, 2002). Of the remainder that is successfully sorted, only half is the desired gender. Consequently, only 10-15% of the original sperm population entering the flow cytometer are recovered as marketable, sexed semen (Seidel Jr and Garner, 2002). Conventional semen straws contain ~20 million sperm. Sorting speeds are currently inadequate for commercially viable production of semen straws containing 20 million sperm, and consequently sexed semen straws generally contain approximately 2 million sperm (Sharpe and Evans, 2009). As a result of both sperm damage during the sorting process and lower sperm numbers included in each straw, use of sexed semen generally results in poorer conception rates compared with conventional semen. Data from recently published studies conducted in the USA (DeJarnette et al., 2009, Norman et al., 2010) and Denmark (Borchersen and Peacock, 2009) indicate that conception rates achieved with frozen-thawed sexed semen in maiden heifers are approximately 80% of those achieved with conventional semen (e.g. 70 vs. 56%). The reduction in fertility observed when using sexed semen has, to date, restricted its use to inseminations on maiden heifers. Preliminary results from New Zealand suggest that using fresh sexed semen results in conception rates that are 90-95% of those achieved with conventional frozen-thawed semen in both heifers and lactating cows (Unpublished results: LIC field communication). Avoiding the sperm damage and mortality associated with the freeze-thaw process has beneficial implications for fertility performance, and has important implications for sexed semen use in seasonal-calving systems, such as in Ireland.
Benefits of sexed semen use

The principal benefit of using sexed semen is increased numbers of heifer calves born, with approximately 90% of successful pregnancies resulting in a heifer calf. The subsequent increased availability of replacement heifers may be utilised to expand herd size and production. Alternatively these heifers may be sold as calves, which would increase revenues compared with the sale of lower value dairy bull calves. Using female sexed semen may also allow farmers to reduce the incidence of difficult calvings (heifer calves are lighter than male calves), and improve biosecurity by increasing herd size while maintaining a closed herd.

In seasonal calving systems, the use of male sexed semen from beef sires on later calving cows may also be considered to increase the value of beef output from the dairy herd, as male beef calves traditionally command a premium over females.

Impact of using sexed semen on animal recording and genetic evaluations

The genetic evaluation of dairy animals relies on the collection of pedigree information on the population under consideration, as well as trait information on these individuals and their relatives. Accurate parentage assignment is therefore important to reduce the errors that could occur in extensively farmed systems. Depending on the farming systems, errors in assignment of parentage could vary from around 7% in German dairy cattle (Sanders et al 2006) to around 15% in New Zealand dairy cattle (Spelman, 2002). In both New Zealand and Ireland, seasonal compact calving systems are the norm. As the size of dairy herds has increased in New Zealand, anecdotally, the percentage of mismothering has increased considerably (R Spelman, personal communication). This situation will be further exacerbated with the increased use of sexed semen, and the incidence of incorrect assignment of parentage is likely to increase.

Breeding organisations will need to take this into account. When the incidence of incorrect parentage assignment exceeds 15%, the breeding scheme will need to adjust for the population structure and the type of errors that could impact on the validity of the scheme (Oliehoek and Bijma, 2009). It is also an opportunity for organisations that offer a parentage verification service to improve accuracy and become imaginative in the types of products that can be offered to their farmer clients.

There is also the added challenge to distinguish between the various semen products. Fresh and frozen conventional semen will now have the sexed semen product as part of the offering and AI technicians will need to be vigilant in the recording of these different products during insemination.
References