Available online at www.sciencedirect.com
SCIENCE DIRECT
Theriogenology
ELSEVIER
Theriogenology 64 (2005) 1647–1655
www.journals.elsevierhealth.com/periodicals/the

Fertility in heifers and cows after low dose insemination with sex-sorted and non-sorted sperm under field conditions

M. Bodmer [a,*], F. Janett [b], M. Hässig [b], N. den Daas [a], P. Reichert [a], R. Thun [b]

[a] *Big-X Inc., Seewen, Switzerland*
[b] *Clinic of Reproduction, University of Zürich, Switzerland*

Received 14 September 2004; received in revised form 7 April 2005; accepted 8 April 2005

Abstract

The present study was performed to test fertility after low dose insemination with sexed and non-sexed sperm in dairy cattle under field conditions in Switzerland. Spermatozoa were stained with Hoechst 33342 and sorted by flow cytometry. A total of 132 heifers and cows were inseminated with 2×10^6 X-bearing, frozen-thawed sperm (A) and 91 animals were inseminated with the same dose using non-sorted, frozen-thawed sperm (B). Pregnancy examination by ultrasound was performed twice, 30–40 days (PE1) and 70–90 days (PE2) after insemination. The pregnancy rates after PE1 were 33.3% (9/27) and 59.3% (16/27) in heifers ($P = 0.05$) and 27.6% (29/105) and 28.1% (18/64) in cows ($P > 0.05$) for groups A and B, respectively. Embryonic losses between PE1 and PE2 in heifers were 11.1% (1/9) and 0% (0/16) and in cows 17.2% (5/29) and 5.6% (1/18), the differences between groups A and B not being significant ($P > 0.05$). Calving rates in heifers were 29.6% (8/27) and 57.8% (15/26), whereas in cows 22.1% (23/104) and 23.4% (16/63) gave birth to calves (for both groups $P > 0.05$). The sex ratio was different ($P < 0.05$) between A (85.3%) and B (58.6%). From our results it can be concluded that conception rates of sorted and non-sorted semen are similar using an insemination dose of 2×10^6. Fertility may be increased by improving sexing technology and animal management.

Keywords: Sexed semen; Low dose insemination; Cows; Heifers; Fertility

* Corresponding author at: Mühlehofstrasse 2, 6260 Reiden, Lucerne, Switzerland.
E-mail address: migibodmer@hotmail.com (M. Bodmer).

0093-691X/$ – see front matter © 2005 Elsevier Inc. All rights reserved.
doi:10.1016/j.theriogenology.2005.04.011

1. Introduction

Until now, the only reliable method for separating X- and Y-chromosome-bearing spermatozoa is flow cytometrical cell sorting for DNA content of sperm. The first live offspring from flow cytometrically sorted sperm resulted from rabbits surgically inseminated into the oviduct [1]. From then on, offspring of pre-selected sex have been produced with an accuracy close to 90% in pigs [2–4], cattle [5–8], horses [9,10] and humans [11]. Due to improvement of nozzle design [12] and adaptation to high-speed cell sorting [13] the efficiency of the sexing procedure has increased considerably. Accuracy of the desired sex ranges from 85 to 90% and sorting speed has been established at up to 11×10^6 sperm cells/h [13]. The sexing method has also been combined with in vitro fertilization in pigs [2,3] and cattle [14,15]. To make sorted sperm available for artificial insemination on a commercial basis, experiments using low dose insemination with flow-sorted sperm were performed in horses [9,10], cattle [6,7], sheep [16] and pigs [4,17]. In addition, it is known that during sorting with the flow cytometer, dead sperm cells are not sorted, and therefore not loaded into straws [18].

To increase fertility, uterine horn insemination with sorted frozen-thawed sperm was tested in cattle [6]. Results of these experiments did not show a significant increase of pregnancy rates compared with uterine body insemination, although some authors [19] claim to have better results when performing uterine horn insemination with non-sorted sperm. Seidel et al. [6] combined sorted, liquid semen with very low dose ($(1–2) \times 10^5$ in 0.1 mL) insemination into the uterine horn ipsilateral and contralateral to the ovary bearing the largest follicle. Pregnancy rates for ipsilateral and contralateral inseminations were nearly identical and ranged from 2.6 to 22.4%, depending on the time between the end of sorting and insemination as well as on the bull used. A recent study in pigs [4] shows, that low dose (70×10^6 spermatozoa) insemination with flow cytometrically sorted sperm deep into the uterine horn resulted in pregnancy rates of 45.6 and 35% for induced and spontaneous ovulation, respectively. In horses, hysteroscopic insemination into the uterine horn [10,20] and ultrasound guided deep uterine insemination [9,20] were performed using sex-sorted sperm in low concentrations (5×10^6 sperm cells/dose). Hysteroscopic insemination resulted in more pregnancies than ultrasound guided deep uterine insemination [10,20].

Reduced fertility, when using sorted sperm, has been attributed to damage of spermatozoa caused by the sexing process [18]. This includes staining and incubation of spermatozoa with Hoechst 33342, sperm dilution, exposure to high pressure and laser light, the rapid projection into the collection tube and also centrifugation to concentrate sorted sperm. After sorting, spermatozoa are partially capacitated resulting in a shorter life span and consequently in reduced fertilizing capacity [4]. To increase sperm quality and fertility of sexed sperm, Seidel et al. [21] as well as Suh and Schenk [22] showed that post-thaw motility and fertility were considerably higher when lower pressure was used during sorting. This however, reduces the sorting rate by 2–3% [22]. Embryo survival rates were also impaired with sorted sperm in cattle [6], pigs [4], mares [9] and rabbits [23] when compared with non-sorted sperm but calves produced with sexed sperm were not different from control calves regarding birth weight, mortality and weaning weight [24,25]. Furthermore no differences were observed between cows carrying calves derived from