Incidence of fertilization failure and embryo loss in Holstein Friesian heifers and postpartum dairy cows

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Introduction The cost of replacing cows that are culled due to reproductive failure is approximately £18,000 per 100 cow herd per year. It is estimated that fertilisation rates following artificial insemination are >90% (Diskin *et al*., 2006) and yet calving rate to a single insemination is in the order of 31% (Law *et al*., 2009). The majority of this reproductive wastage occurs between insemination and maternal recognition of pregnancy, approximately 16 days after ovulation, with relatively little late embryonic/foetal mortality. The objective of this study was to quantify reproductive wastage prior to day 7 post insemination and to evaluate energetic, metabolic and hormonal effects in early lactation (day 0-42) on embryonic loss.

Material and methods Sixty-three autumn-calving Holstein Friesian cows (mean parity 3.1) and 32 nulliparous Holstein Friesian heifers (mean age 15 months) were used (Study 1). Lactating cows were offered a total mixed ration (TMR) comprising 60% concentrates and 40% forages (60% grass silage and 40% maize silage) on a DM basis. The complete diet contained 185 g CP/kg DM and 12.4 MJ ME/kg DM. An average daily energy balance for each individual cow was calculated for each week of lactation according to Thomas (2004). Heifers were offered grass silage *ad libitum* and 3 kg concentrate per head per day. A subsequent study (Study 2) was conducted on 16 spring calving Holstein Friesian cows (mean parity 2.8) and 18 nulliparous Holstein Friesian heifers (mean age 14 months) to validate the technical aspects of single embryo recovery. Lactating cows were offered a TMR comprising 50% concentrate and 50% forage (65% grass silage and 35% maize silage) on a DM basis. Heifers were offered grass silage *ad libitum* and 3 kg concentrate per head per day. Individual cow intakes were not recorded in Study 2. Oestrous cycles were synchronised to ovulate at days 42, 70 and 98 post calving in Study 1 and 61 d post-calving in Study 2, using a controlled intra-vaginal drug release (CIDR) of progesterone. Animals were bred by AI approximately 56 h after CIDR removal. The uteri of all animals were non-surgically flushed 7 days post-insemination to recover and classify embryos; the proportion of unfertilised oocytes, degenerate embryos and viable embryos (morula/blastocyst stage) was recorded. Data were analysed using logistic regression models via Genstat.

Results Only animals that were detected in oestrus and had normal luteal phase progesterone concentrations were included in the data set. In Study 1, cows had an average milk yield of 38.0 kg/d (100 d), an average DM intake of 21.5 kg/d (100 d), and an average six week energy balance of -28.0 MJ/d**.** In Study 2, cows had an average milk yield of 34.8 kg (61 d). The recovery rate did not differ (P>0.05) between heifers and cows but the proportion of oocytes fertilised (as a proportion of recovered structures) was significantly lower in lactating cows than heifers (32.5 vs. 81.3%, respectively; P<0.001)**.**

Table 1 Recovery and fertilisation rates from single oocyte flushes of Holstein Friesian cows and heifers (Studies 1 & 2)

	Period	Flushes (n)	Nothing recovered (n)	Unfertilised Oocyte (n)	Blastocyst / Morula (n)	Recovery rate (%)	Fertilization rate (%)
Heifers	Study 1	35	25	3	7	28.6	70.0
	Study 2	18	12	0	6	33.3	100.0
	Total	53	39	3	13	30.2	81.3
Cows	Study 1	91	55	24	12*	39.5	33.3
	Study 2	16	12	3	1	25.0	25.0
	Total	107	67	27	13	37.4	32.5

* Two structures were degenerate; one at the 2 cell stage and the other at the 8-16 cell stage.

Cows with fertilised oocytes (Study 1) had a more positive average daily energy balance (EB) in week 1 post calving than cows yielding an unfertilised oocyte (-10.5 vs. -46.8 MJ/d; P<0.01; SED 11.49). Similarly, cows with fertilised oocytes had a more positive cumulative EB in week 1 (-75.7 vs. -326.6 MJ; P<0.01; SED 80.0) and 2 (-252.9 vs. -614.4 MJ; P=0.058; SED 183.6) post calving than cows with unfertilised oocytes. Plasma urea concentrations were lower in cows with fertilised oocytes in week 1 post calving compared to those with unfertilised oocytes (4.30 vs. 5.66 mM; P<0.01; SED 0.48).

Conclusions The results clearly demonstrate that single embryo recovery is difficult with 66.3% of animals flushed yielding no oocyte/embryo. Fertilisation failure occurred in 53.5% of animals (67.5% of cows, 18.8% of heifers) which is greater than 10-20% estimated by Diskin *et al.* (2006). Furthermore, energy status in early lactation significantly affected fertilisation rate. Reproductive wastage during the 7 day period post insemination was lower than anticipated, with 15.4% of fertilised oocytes from lactating cows being degenerate.

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