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J.P.Crilly[a], L.Söderquist[b], A.Holmström[c], N.D.Sargison[a]

a. Farm Animal Practice, Royal (Dick) School of Veterinary Studies, University of Edinburgh, Easter Bush, Roslin, Midlothian, UK, EH25 9RG

b. Division of Reproduction, Department of Clinical Sciences, Swedish University of Agricultural Sciences, PO Box 7054, SE-750 07, Uppsala, Sweden

c. Gård & Djurhälsan, Kungsängens Gård, SE-753 23, Uppsala, Sweden

Artificial insemination (AI) by intravaginal deposition of frozen-thawed semen (Fairnie and Wales 1982), is simple, non-invasive and requires little specialised equipment. In Norway and Sweden the technique is widely performed by farmers themselves. Mean conception rates of 50% in Sweden (L.Söderquist, pers.obs.) and 67% in Norway (Paulenz and others. 2005) have been reported. Flocks in Scandinavia are small relative to the UK (Ulvund 2012) and sheep are frequently housed during the mating period. AI is performed to natural oestrus. Heat detection is performed using halter-trained entire rams wearing aprons. Optimal results are obtained with thrice daily heat detection and insemination 12-24 hours after standing oestrus is detected. A dose of $200x10^6$ spermatozoa is used (Paulenz and others. 2005). To the best of our knowledge, there has been no previous trial of this technique in the UK. The object of this study was to test the viability of this method of AI in sheep that are at pasture during the mating period.

A 4 year old Texel ram with no detectable abnormalities (Boundy 1992; Gouletsou & Fthenakis 2010) was submitted to an ovine semen collection centre (AB Europe, Ormiston, East Lothian, UK). Semen was collected using an artificial vagina and a teaser ewe; it was of suitable quality for freezing (>5 x 10^9 spermatozoa per ml, progressive motility > 85%, <5% morphologically abnormal). The semen was extended with Triladyl semen extender (Minitüb, Tiefenbach, Germany) before gradual cooling to 4'C, division into doses and freezing on solid carbon dioxide before storage in liquid nitrogen. 40 0.25ml straws (≤200 x 10^6 spermatozoa per dose) were produced; the decision was made to inseminate each ewe with 2 straws. Straws were stored in liquid nitrogen until the time of AI.

A vasectomised ram was introduced to 40 Cheviot Mule ewes (aged 2.5-4.5 years) for 3 days (20^{th}-22^{nd} October) to induce a degree of oestrous synchronisation (Atkinson and Williamson 1985; Rosa and Bryant 2002). This ram was reintroduced on 2^{nd} November, with keel paint applied (Salamon & Morrant 1963). The ewes were inspected twice daily (7 a.m. and 4 p.m.) and any marked animals removed.. The marked ewes were inseminated the following session (e.g. seen marked at 7 a.m., inseminated at 4 p.m.) and then turned out into a separate paddock. 20 ewes were inseminated between 3^{rd} and 7^{th} November. The remainder were dispersed to the rest of the flock. 2 days after the last ewe had been inseminated a Suffolk ram with a different colour of keel paint was introduced to the AI group, to mate any ewes which did not conceive to AI. The choice of the two breeds of sire ensured that the lambs could be easily identified as having been conceived to AI or natural service due to the obviously different phenotypes .

Insemination was performed as follows. Straws were thawed in water at 35'C for 12 seconds, then dried, the tip removed with a pair of scissors and loaded into a pre-warmed ovine AI gun (Minitüb, Tiefenbach, Germany), a sheath was then applied to the gun. The gun was maintained at 37'C on a

cloth covered electronic hot plate (Hearson, London, UK). Temperature of the water and hot plate were monitored by thermometer (Zeal mercury thermometer, GH Zeal Ltd, London UK and Brannan digital in/out thermometer, S. Brannan & Sons Ltd, Cleator Moor, Cumbria, UK respectively).The ewe was gently restrained by the shepherd. The vulva was cleaned using paper towel, the lips parted and both guns gently inserted until resistance was felt. They were then withdrawn slightly and plungers depressed. Guns were cleaned with surgical spirit and dried between ewes. Ewes were marked, ear tag numbers recorded and the time and date of insemination noted.

At birth the lamb phenotype (Texel X or Suffolk X), litter size and number of males and females was recorded.

10 of the 20 ewes conceived to AI (see Table 1), a proportion similar to that previously described in Norway (Paulenz and others 2005). The mean litter sizes for AI and non-AI offspring were 1.7 and 1.9 , respectively.

While this trial involves a small number of sheep, it provides proof of concept that an acceptable conception rate is possible using this technique under UK sheep-farming conditions. The number of spermatozoa deposited in each ewe in this trial (up to 400 x 10^6) was higher than reported from Scandinavia; higher spermatozoa number have been shown to result in higher conception rates in several studies involving various AI methods (Maxwell 1986; Maxwell & Hewitt 1986; Paulenz and others 2002). However, increasing the dose from $100x10^6$ to $600x10^6$ spermatozoa in vaginal AI at synchronised oestrus with frozen-thawed semen in an Australian study only increased the conception rate from 17.0 to 17.4% (Maxwell & Hewitt 1986).The volume of frozen-thawed semen was higher than previously described for this method (Paulenz and others 2005). Breed differences in ovulation rate may explain the differing conception rates; significantly higher conception rates in more prolific breeds has been reported for cervical AI using frozen-thawed semen (Donovan and others 2004).

Further study is needed to further refine the technique and to determine whether acceptable conception rates are achievable following oestrus synchronisation using exogenous hormone treatments. Confirmation of the efficacy of this technique, which could be performed by farmers themselves, would allow the development of completely closed commercial sheep flocks. A robust and simple method of ovine AI would allow farmers to produce improved sires on farm, for use within their own flock, rather than purchasing rams. This would reduce the risk of the import of disease onto the farm, and also reduce the access cost of genetic improvement, as only semen doses need be purchased, rather than a ram (Evans & Maxwell 1982). Unlike laparoscopic AI, this technique does not raise ethical concerns over a surgical procedure which is not for the animal's benefit (Stafford and others 2006).

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Table 1

Date of AI, date of lambing and the offspring of each ewe.

Ewe ID	Date of AI	Date lambed	No. of lambs	Lamb phenotype	Conception to AI
3685	06/11/2014	31/03/2015	2	Texel X	Yes
3696	07/11/2014	31/03/2015	2	Texel X	Yes
3718	05/11/2014	31/03/2015	1	Texel X	Yes
3723	04/11/2014	31/03/2015	2	Texel X	Yes
3728	06/11/2014	30/03/2015	2	Texel X	Yes
3733	05/11/2014	31/03/2015	2	Texel X	Yes
3759	06/11/2014	18/04/2015	1	Suffolk X	No
4836	06/11/2014	16/04/2015	3	Suffolk X	No
5052	06/11/2014	02/04/2015	2	Texel X	Yes
5064	07/11/2014	01/04/2015	1	Texel X	Yes
5071	03/11/2014	15/04/2015	2	Suffolk X	No
5087	04/11/2014	12/04/2015	3	Suffolk X	No
5089	04/11/2014	18/04/2015	2	Suffolk X	No
5090	05/11/2014	28/04/2015	1	Suffolk X	No
5111	06/11/2014	19/04/2015	1	Suffolk X	No
5115	07/11/2014	23/04/2015	2	Suffolk X	No
5116	03/11/2014	16/04/2015	2	Suffolk X	No
5135	05/11/2014	01/04/2015	1	Texel X	Yes
5143	04/11/2014	13/04/2015	2	Suffolk X	No
5453	06/11/2014	31/03/2015	2	Texel X	Yes