Institution: University of Edinburgh and SRUC, Scotland's Rural College

Unit of Assessment: 6

Title of case study: Dolly the sheep – the first cloned adult animal leading to multiple cloning business enterprises

1. Summary of the impact (indicative maximum 100 words)

Impact: Economics. The first cloned mammal to be created from an adult somatic cell and subsequent production of thousands of cloned animals and their progeny.

Significance: The first evidence that adult specialized cells are still capable of driving the development of a complete and fertile animal which has been translated to preserve genetic characteristics of exceptional value (e.g. competitiveness in horses)

Beneficiaries: Agriculture, livestock and equine industry, commerce.

Attribution: Essential improvements to the Somatic Cell Nuclear Transfer (SCNT) technique by Prof. Wilmut (Roslin Institute, now part of UoE) were used to clone Dolly the sheep.

Reach: Worldwide: SCNT technology has been adopted around the world, being used to clone multiple animal species.

2. Underpinning research (indicative maximum 500 words)

The productivity and profitability of animal farming is directly linked to the quality of the livestock available. This in turn depends on the underlying genetics of farm animals. For a long time, Ian Wilmut (employed by Roslin Institute (Group Leader) and UoE (Inaugural Director of MRC Centre for Regeneration and Professor) 1974-2011) and colleagues worked on new methods to create genetically improved livestock by manipulation of stem cells using nuclear transfer.

Research published in 1994 from the Roslin Institute (now part of the University of Edinburgh), established that development of embryos reconstructed by nuclear transfer is related to interactions between the donor nucleus and the recipient cytoplasm at the time of fusion and during the first cell cycle following reconstruction [3.1]. This observation led to two protocols the team then followed for maintenance of normal ploidy, using enucleated oocytes at metaphase II (MII) stage of cell division as recipient cytoplasms (cytoplasts). The first was based on the transfer of nuclei in G1, S or G2 phase of the cell cycle into MII cytoplasts devoid of the so-called maturation-promoting factor (MPF) activity. The second protocol used nuclei in G1 phase of the cell cycle and MII cytoplasts containing high activity of MPF. A study in mice showed that it was possible to use microtubule inhibitors such as nocodazole to hold cells in mitosis before releasing them and using them as nuclear donors as they were expected to be passing through G1 phase [3.2, 3.3].

The practical value of the second protocol was confirmed by the set of experiments published in 1996 in which cells from sheep late blastocysts were either taken at any stage of the cell cycle or were synchronised in G0/G1 by serum starvation (as alternative to nocodazole). Whereas no lambs were obtained without synchronisation, four live lambs were obtained following transfer from late-passage cells at G0/G1 [3.4]. This was followed by a final key experiment the following year, in which nuclei arrested in G0/G1 were derived from a blastocyst, a foetus and, critically, mammary tissue of an adult ewe. Live lambs were obtained from all three populations, with the nuclei from mammary tissue creating Dolly – the first cloned mammal made from an adult somatic cell [3.5].

3. References to the research (indicative maximum of six references)

3.1) Campbell KH, Loi P, Cappai P, Wilmut I. (1994) Improved development to blastocyst of ovine nuclear transfer embryos reconstructed during the presumptive S-phase of enucleated activated oocytes. Biology of Reproduction 50: 1385-1393. http://tinyurl.com/nc5lu36

3.2) Otaegui PJ, O'Neill GT, Campbell KH, Wilmut I. (1994) Transfer of nuclei from 8-cell stage mouse embryos following use of nocodazole to control the cell cycle. Molecular Reproduction

and Development 39: 147-152. http://tinyurl.com/okjdowx

3.3) Otaegui PJ, Waddington D, Wilmut I. (1994) Nuclear transfer of 4-cell mouse embryos: synchronisation with cytoplast partially overcomes nuclear donor cell-cycle effect. Journal of Reproduction and Fertility Abstract Series 13 Abstract 67. (Copy of paper available on request.)

3.4) Campbell KH, McWhir J, Ritchie WA, Wilmut I. (1996) Sheep cloned by nuclear transfer from a cultured cell line. Nature 380: 64-66. http://dx.doi.org/10.1038/380064a0

3.5) Wilmut I, Schnieke AE, McWhir J, Kind AJ, Campbell KH. (1997) Viable offspring derived from fetal and adult mammalian cells. Nature 385: 810-813. http://dx.doi.org/10.1038/386200a0

4. Details of the impact (indicative maximum 750 words)

Somatic Cell Nuclear Transfer (SCNT) cloning is the only technology available that enables generation of 99.8% genetically identical offspring from selected individuals of adult animals (including sterilized animals). As such, it is being exploited as an efficient multiplication tool to support specific breeding strategies of farm animals with exceptionally high genetic value.

Impact on commerce and agriculture

The SCNT technology has been widely adopted and used to create clones of other animals (e.g. cows, deer, ferrets, goats, horses, mice, mules, pigs, wolves, rats, rabbits, monkeys and water buffalo). Multiple new companies have been established (e.g. Korean firm RNL Bio and US firm BioArts in 2008, and Kheiron in Argentina in 2012) and cloning plays increasingly important role in the animal-breeding industry. For example in 2010 the world's first cloned Polo pony was born [5.1]. Over 60 clones have been generated since then, and one of the cloned ponies fetched $800,000 at an auction in Argentina, which was attended by more than 1000 people (2010) [5.1]. In June 2012, the Fédération Equestre Internationale (FEI) - the group that governs international equestrian events, has lifted its ban on cloned horses participating in competitions [5.1]. Consequently, cloned horses will be allowed to participate in the Olympics beginning in 2016. The FEI estimates that more than 110 horses were cloned in 2010 only. First orders for commercial pet dog cloning were completed by the Korean company RNL Bio and US firm BioArts in 2008 [5.2].

In January 2008, the US Food and Drug Administration (FDA) decided that meat produced from clones and their progeny is acceptable for human consumption [5.3]. Similar conclusions have been drawn by European Food Safety Authority and published on 15 July 2008 (updated in 2012) [5.4]. There is already some use of cloned livestock in Europe, for example several hundreds of cattle that are progeny of cloned ancestors live in Switzerland [5.5], but animal cloning for food production in Europe meets with considerable opposition. Use of cloned animals is more widespread in Asia and America, where people are more accepting of consuming cloned animal products [5.6].

Preserving diversity of livestock

The use of cloning from frozen somatic tissue is a much more cost-effective strategy for bio-banking of endangered breeds than the storage of sperm or egg cells. Sample collection and storage is easier and cheaper, the technique can be deployed quickly in emergency situations such as a disease outbreak, and cloning recovers all the genetic variation of the donor – not just half of it as when using sperm. Consequently, SCNT cloning of farm animals is a common new service in some countries including the USA, Argentina and Brazil [5.7], and it is covered by all major veterinary genetics and animal biotechnology textbooks [5.8, 5.9].

Cloning has been used to conserve several animal breeds in the recent past. For example in 2012 an increasingly rare Himalayan pashmina goat breed was successfully cloned. This important achievement, which was performed in the frame of the National Agricultural Innovation Project (NAIP) of the Indian Council of Agricultural Research, offers hope to the people of Kashmir of increasing production of pashmina; a type of cashmere wool. In Kashmir more than 10 million people are associated with $85 million shawl industry that depends on the availability of the exceptionally fine wool produced by the rare animal [5.10].

5. Sources to corroborate the impact (indicative maximum of 10 references)

5.1) Cloned ponies. http://tinyurl.com/om7nor5

5.2) "Dead dog's owner creates FIVE cloned puppies of her beloved pet" Daily Mail August 5, 2008

http://tinyurl.com/nldstku

5.3) CVM GFI#179 "Use of Animal Clones and Clone Progeny for Human Food/Animal Feed" U.S. Department of Health and Human Services, Food and Drug Administration, Center for Veterinary Medicine, January 15, 2008. http://tinyurl.com/pjwan9e

5.4) Barlow S, Chesson A, Collins JD, Flynn A, Hardy A, Jany K, Knaap A, Kuiper H, Le Neindre P, Schans J, Schlatter J, Silano V, Skerfving S and Vannier P. "Food Safety, Animal Health and Welfare and Environmental Impact of Animals derived from Cloning by Somatic Cell Nucleus Transfer (SCNT) and their Offspring and Products Obtained from those Animals" The EFSA Journal (2008) 767, 1-49. http://dx.doi.org/10.2903/j.efsa.2008.767

European Food Safety Authority. "Update on the state of play of Animal Health and Welfare and Environmental Impact of Animals derived from SCNT Cloning and their Offspring, and Food Safety of Products Obtained from those Animals" The EFSA Journal (2012);10(7):2794. http://dx.doi.org/10.2903/j.efsa.2012.2794

5.5) Schweizerische Eidgenossenschaft, Federal Office of Public Health "Food from cloned animals" (updated August 15, 2012) http://tinyurl.com/p4nb4s5

5.6) "In Europe and the US, consumer views on cloned products breed different results" – news release from the Kansas State University (June 21, 2011) http://tinyurl.com/nn34hka

5.7) "Welfare implications of cloning of farm animals" – letter of Prof. Christopher Wathes, Chairmen of the Farm Animal Welfare Committee (FAWC) to Lord Taylor (Parliamentary Under Secretary, Department for Environment and Rural Affairs) dated 11 April 2012. http://tinyurl.com/p2u36u4

5.8) Nicholas FW "Introduction to veterinary genetics" 3rd edition Willey-Blackwell 2010. [Available on request.]

5.9) Singh B, Gautam SK, Chauhan S "Textbook of animal biotechnology", Unit 4, The Energy and Resources Institute (TERI) 2013. [Final publication due 2014]

5.10) "Pashmina goat clone boost for Kashmir's shawl makers" by Athar Parvaiz, Asia Times Online, Apr 26, 2012. http://tinyurl.com/oeqbqn8