Review on Cloning in Farm Animals

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Abstract: Cloning is one of the breeding technologies like other new technologies. It must be assessed through a wide range of angles such as animal welfare, food safety, precautionary principles and ethical concerns. Cloning of plants (such as growing a plant from a cutting) has been a common practice of mankind for hundreds and perhaps thousands of years. Even cloning of small animals has a long history dated back to the 1960's (HRF, 2014. Cloning is already being used commercially in the livestock industry in some parts of the world for the replication of elite breeding animals. The use of cloning technology is therefore facilitating the development and commercialisation of genetically modified animals for food production purposes. SCNT produces animals that are genetically unlike any animal found in nature. Many species have been cloned since Dolly the sheep, the first mammal to be cloned from an adult cell, was born in 1996. There are now estimated to be around 6000 farm animal clones worldwide.
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Introduction

Cloning is one of the breeding technologies like other new technologies. It must be assessed through a wide range of angles such as animal welfare, food safety, precautionary principles and ethical concerns. There is great need for more clarity about these aspects and about the dangers stemming from misunderstanding. In what concerns food production, benefits of cloning are limited at present, but potentially promising in the long term. Cloning allows the reproduction of animals that already have shown good productivity, a low incidence of disease and ability to cope with the environment (Thomson, 1999) and clone is simply one organism made from another, resulting to two organisms with the same set of genes. When earthworms are cut in half, they regenerate the missing parts of their bodies, leading to two worms with the same set of genes. The purpose of cloning was simply to multiply an organism without waiting for nature to act on it first (EL, 2010). Cloning is considered to be one of a number of assisted reproductive technologies, many of which are at various stages of development and have not necessarily been widely adopted, but which are considered to be technological solutions to problems associated with the declining reproductive efficiency of dairy cattle as a result of advances made in increasing milk production. In addition, the advances that have been made in selecting traits associated with increasing milk production have also caused a substantial reduction in genetic diversity. Thus, advances made in the arts are viewed as potential solutions to maintain and improve the genetic superiority of dairy animal (CPSU, 2010).

Cloning is commercially available, the technology still is considered to be quite inefficient and very costly. Inefficiencies stem from the micromanipulation of oocytes and culture of donor cells and cloned embryos. This is due to the large number of abortions that occur throughout gestation. In addition, when pregnancies actually progress to term, gestation is usually extended and calves are born much larger than average due to large offspring syndrome (LOS) that leads to dystocia, and most animals require cesarean section. These large offspring usually have postnatal weakness, hypoxia, hypoglycemia, metabolic acidosis and hypothermia, all requiring immediate intensive care. Other problems that are sometimes associated with the technique include incomplete reprogrammation, shortening of telomeres (the physical ends of linear eukaryotic chromosomes), hypertension, kidney abnormalities, liver problems, and limb and body defects (Van Arendock, 2003).

The ethical issues surrounding cloning have not been resolved sufficiently to warrant a wholesale endorsement of the practice. The slippery slope of mastering animal cloning could entice some humans to duplicate themselves, or to create living, breathing organ farms within cloned humans. Of all the animal cloning pros and cons, this is the most frightening to most ethicists and religious observers. The safety of cloned animals as food sources has not been established. Although the animals are in theory identical, the same forces that result in so many visibly defective offspring could also result in invisible but dangerous alterations on the cellular level. These changes could be dangerous to human beings

consuming the meat, eggs or milk of the cloned animals (Shugerman, 2014).

Generally, the objective of this paper is

.to review the history, purpose, ethical and welfare issues c.

.To know success rates of cloning in farm animals based on available literature.

.To discuss application of cloning in farm animals.

.To understand the processes of cloning by SCNT.

.To Know Advantages, And Disadvantages Of Cloning.

Review On Cloning In Farm Animals

History of cloning

Cloning of plants (such as growing a plant from a cutting) has been a common practice of mankind for hundreds and perhaps thousands of years. Even cloning of small animals has a long history dated back to the 1960's (HRF, 2014). However, human cloning had not been thought possible until the successful cloning of the first mammal, Dolly the sheep, in 1997. The birth of Dolly is a major scientific and technological breakthrough. However, it alsoraised the possibility that one day humans will be cloned, as well as many medical and ethical issues and concerns associated with this possibility. Following the cloning of Dolly, many other animals, including cows and mice, have been successfully cloned (HAS, 2003).

In 1952, cloning by nuclear transfer first demonstrated in animals. Source of nuclei was the very early embryo of the frog. In1989-90, cloning first achieved in mammals (rabbits, sheep, and cows) by nuclear transfer from very early embryos**.** In 1997, Cloning first achieved Dolly the sheep using adult sheep cell (Twyman, 2003). In 2001, Scientists at advanced cell technology institute announced the birth of a cloned baby bull (a large wild ox) named Noah. Noah was the first endangered animal to be cloned. Although Noah died of an infection unrelated to the cloning procedure, the experiment opened the door to saving endangered species through cloning. Later in 2003, Dolly the sheep was put down by a lethal anesthetic injection. She suffered from lung cancer caused by a virus. She was six and a half years old. Apart from the cancer and her well-publicized arthritis, she was relatively healthy and normal (HAS, 2003).

The scientific community was shocked by Dolly`s birth and continued to investigate further on the topic of human clones. The DNA came from a single cell taken from her mother's egg, which is fused with the mammary cell. After the fused cell develops into an embryo, it is then implanted in a substitute sheep. This embryo then grows into a lamb that obtains the same genes as its donor sheep. Dolly was created as a healthy lamb after more than 277 attempts. That is a part of the reason cloning has become controversial. Scientists fear that applying this technique to humans may lead to malformations or diseases in the human clone. Moreover, human cloning is even more complicated, with greater risks, permanent damage, and potentials for error (EL, 2010).

1996 – Dolly the sheep – first mammal cloned from an adult cell (Wilmut et al, 1997)

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1998 – Cow (Kato et al, 1998); Mouse (Wakayama et al, 1998)

⇩

1999 – Goat (Baguisi et al, 1999)

⇩

2000 – Pig (Polejaeva et al, 2000)

⇩

2002 – Rabbit (Chesne et al, 2002); Cat (Shin et al, 2002); Rat (Zhou et al, 2003)

⇩

2003 – Horse (Galli et al, 2003); Mule (Woods et al, 2003); Deer (Anon., 2003)

⇩

2004 – Buffalo (Shi et al, 2007)

⇩

2005 – Dog (Lee et al, 2005)

⇩

2006 – Ferret (Li et al, 2006)

⇩

2009 – Camel (Wani et al, 2010)

Figure 3.1: Timeline Of Domestic Species Cloned.

Potential applications of animal cloning

Replication of elite breeding animals

Cloning is already being used commercially in the livestock industry in some parts of the world for the replication of elite breeding animals. It has been widely reported in the media that products from the offspring of cloned animals have already entered the human food chain in the United States and elsewhere (Weiss, 2008; Bethge, 2009; Plume, 2009.

Following the decision by the US Food and Drug Administration (FDA, 2008) that products from cloned animals are safe, food from clones and their offspring

can freely enter the marketplace in the US and there is no requirement for these products to be labelled. There remains a voluntary moratorium in place for clones of species other than cattle, pigs and goats until more information is available on these species (FDA, 2010).

A number of companies in the US offer cloning services to the livestock breeding industry, primarily for cattle and also for pigs (Via Gen, 2009; Trans Ova Genetics, 2009; Cyagra, 2009). Bovance, a joint venture between Via Gen and Trans Ova Genetics, states (Bovance, 2009):

The situation in Asia is less clear but it is likely that products from the offspring of clones have entered the food chain in at least some Asian countries. As early as 2002, calves cloned from an elite Holstein dairy bull were sold to China by Australian-based company, Clone International (BBC, 2002). Cloning of livestock is also being undertaken within China by Yangling Keyuan Cloning (People's Daily, 2001).

While Bovance (2009) considers that "cloning will remain a technology suited exclusively for the most elite tier of genetics, and cloned individuals will represent only a fraction of a percentage of tomorrow's cattle breeding foundation", some authors have suggested that there will be a transition from the commercial use of semen and offspring of clones to the production of food products from cloned animals themselves over the next few years (Suk et al, 2007).

Production of transgenic animals

Although the SCNT process is very inefficient (see Section 5), cloning is more efficient than the process of creating transgenic animals by microinjection of foreign DNA (Vajta and Gjerris, 2006). Cloning can be used to increase the efficiency of production of transgenic animals using nuclear transfer of cultured transgenic cells. The foreign DNA is introduced to cultured cells, which can then be screened to identify those cells that have successfully incorporated the foreign DNA. These transgenic cells are used as the nuclear donor in the SCNT process to create cloned transgenic animals. Cloning could also be used to create multiple copies of an existing transgenic animal.

The use of cloning technology is therefore facilitating the development and commercialisation of genetically modified animals for food production purposes. Potential applications include:

The production of animal products with altered characteristics, for example, milk with higher levels of proteins called caseins (to increase the yield of cheese that can be obtained) or lower levels of lactose or lactoglobulin (substances in milk which can cause allergic reactions in some people) (Heyman, 2001);

The production of cloned transgenic male animals who produce mono-sex sperm – the animals are modified to disrupt the development of either X- or Y-chromosome-bearing sperm so that single-sex offspring are produced (Forsberg, 2005).

Advantages of cloning

Food produced from cloned animals is in legislation also called 'novel foods'. The first legislation on novel foods was introduced in May 1997. Novel foods include foods derived from animals obtained by breeding methods not used in Europe before 1997. This is the case for cloning. The novel foods regulation requires a premarketing assessment of such foods and a specific regulation authorising them. Until the present there was not any request from any interested party to trade such products. The reason for this is that cloning is not aimed to produce animals that would go to the food chain, but to preserve a breed or for elite animals mainly used for breeding (EFSA, 2008). On December 28, 2006, the U.S. Food and Drug Administration (FDA) approved the consumption of meat and other products from cloned animals. Cloned animal products were said to be virtually indistinguishable from the non-cloned animals. Furthermore, companies would not be required to provide labels informing the consumer that the meat comes from a cloned animal (FVE, 2009). Cloning also helps infertile couples to have child, can be used to protect endangered species, improves food supply (meat), used to replace dead people, can be used to replace damaged organs or for organ transplant referred as therapeutic advantage of cloning (HAS, 2003).

Disadvantages of cloning

Many births of deformed children, inheriting diseases, can devastate parents if unsuccessful, clone may be teased or bullied about who they really are, change people's thoughts on God and religion, cannot always trust Science Technology, clone may not live very long, selling and buying clones is another threat to humanity, clone would get treated like and object and not a human (Vogel, 2008). Potential problem with cloning is the possibility of loss of genetic diversity. Since unlimited number of identical animals could be produced with cloning, an over population of the same genetic makeup could result in inbreeding and loss of genetic variation, which is not desirable (Benagiano and Primiero, 2002).

The improved genetic gains are expressed by increased levels of milk production. Obviously, as the producer replaces the old herd with the new herd, average milkproduction per cow will increase, thus generating increased revenues. The increased revenues are used to pay for the replacements. The question is: what price would the producer be willing to pay for the genetically improved cows? Gains in this exercise are measured in pounds of milk and are derived from the potential increases in genetic gain. Suppose that a producer decided to invest in cloned animals by

replacing 10% of the milking herd with cloned cows (that had demonstrated higher genetic gains) each year over a period of 10 years. So that after 10 years the entire herd is made up of the improved dairy cattle (Dematawewa and Berger, 1998).

Kinds of cloning

Cloning in nature

Cloning as asexual reproduction is a very common form of multiplication in plants. All plant organs can be sources of asexual reproduction, but stems are the most common ones.

Inanimalsthe reproductive process is also diversified to the point that almost any mechanism we can imagine has already been implemented. The various forms of asexual reproduction co-exist with hermaphroditism and bisexual external and internal copulation (Benagiano and Primiero, 2002) and asexual reproduction includes budding (jellyfish and tapeworms), fragmentation (worms), and parthenogenesis (some fishes, insects, frogs and lizards). However, most of the animals that are able to reproduce asexually reproduce through parthenogenesis only at certain times. Aphids use parthenogenesis in the spring when they find themselves with ample food. Parthenogenesis is more rapid than sexual reproduction (Walmut *et al*., 1999).

In nature, twins form very early in development when the embryo splits in to two. Twinning happens in the first days after egg and sperm join, while the embryo is made of just a small number of unspecialized cells. Each half of the embryo continues dividing on its own, ultimately developing into separate, complete individuals. Since they developed from the same fertilized egg, the resulting individuals are genetically identical (UOU, 2014).

Artificial Embryo Twining

Artificial embryo twinning is carried out in a petridish instead of inside the mother. A very early embryo is separated into individual cells, which are allowed to divide and develop for a short time in the petridish. The embryos are then placed into a surrogate mother, where they finish developing. Again, since all the embryos came from the same fertilized egg, they are genetically identical (Thomson, 2006).

Somatic Cell Nuclear Transfer

Clones of adult animals are created by SCNT. There are two variations of this method. They are the Roslin Technique and the Honolulu Technique. It is important to note that in all of these techniques the resulting offspring will be genetically identical to the donor and not the surrogate, unless the donated nucleus is taken from a somatic cell of the surrogate (Bailey, 2014). The term SCNT refers to the transfer of the nucleus from a somatic cell to an egg cell. A somatic cell is any cell of the body other than a germ (sex) cell. An example of a somatic cell would be a blood cell, heart cell, skin cell etc. In this process, the nucleus of a somatic cell is removed and inserted into an unfertilized egg that has had its nucleus removed. The egg with its donated nucleus is then nurtured and divides until it becomes an embryo. The embryo is then placed inside a surrogate mother and develops inside the surrogate (UOU, 2014).

2.5.3.1 Roslin technique

The Roslin Technique is a variation of SCNT that was developed by researchers at the Roslin Institute. The researchers used this method to create Dolly. In this process, somatic cells (with nuclei intact) are allowed to grow and divide and are then deprived of nutrients to induce the cells into a suspended or dormant stage. An egg cell that has had its nucleus removed is then placed in close proximity to a somatic cell (Bailey, 2014). Then ovum and somatic cell's nucleus are stimulated with a shock and will begin to divide. The egg is now viable and capable of producing an adult organism containing all the necessary genetic information from just one parent. Development will ensue normally and after many mitotic divisions, this single cell forms a blastocyst (early stage embryo) with about 100 cells with an identical genome to the original organism. Stem cells can then be obtained by the destruction of this clone embryo for use in therapeutic cloning or in the case of reproductive cloning the clone embryo is implanted into a surrogate mother for further development and brought to term (Walmut *et al*., 1999).

Honolulu technique

Honolulu technique was developed by Dr. Teruhiko Wakayama at the University of Hawaii. In this method, the nucleus from a somatic cell is removed and injected into an egg that has had its nucleus removed. The egg is bathed in a chemical solution and cultured. The developing embryo is then implanted into a surrogate and allowed to develop (Bailey, 2014).

Cloning in different farm animals

Sheep

All previous cloning experiments used donor nuclei from cells in early embryos. In this experiment, the donor nuclei came from a slightly different source: cultured sheep cells, which were kept alive in the laboratory. Wilmut and Campbell transferred the nuclei from cultured cells into enucleated sheep egg cells. This experiment showed that cultured cells can supply donor nuclei for cloning by nuclear transfer. Because scientists had already learned how to transfer genes into cultured cells, this experiment showed that it might be possible to use such modified cells to create transgenic animals (EFSA, 2008). The famous lamb, named Dolly, brought cloning into the lime

light. Willadsen used a chemical process to separated one cell from an 8-cell lamb embryo. Then he used a small electrical shock to fuse it to an enucleated egg cell. As luck would have it, the new cell started dividing. By this time, in vitro fertilization techniques had been developed, and they had been used successfully to help couples have babies. So after a few days, Willadsen placed the lamb embryos into the womb of surrogate mother sheep. The result was the birth of three live lambs. This experiment showed that it was possible to clone a mammal by nuclear transfer and that the clone could fully develop. Even though the donor nuclei came from early embryonic cells, the experiment was considered a great success (UOU, 2014).

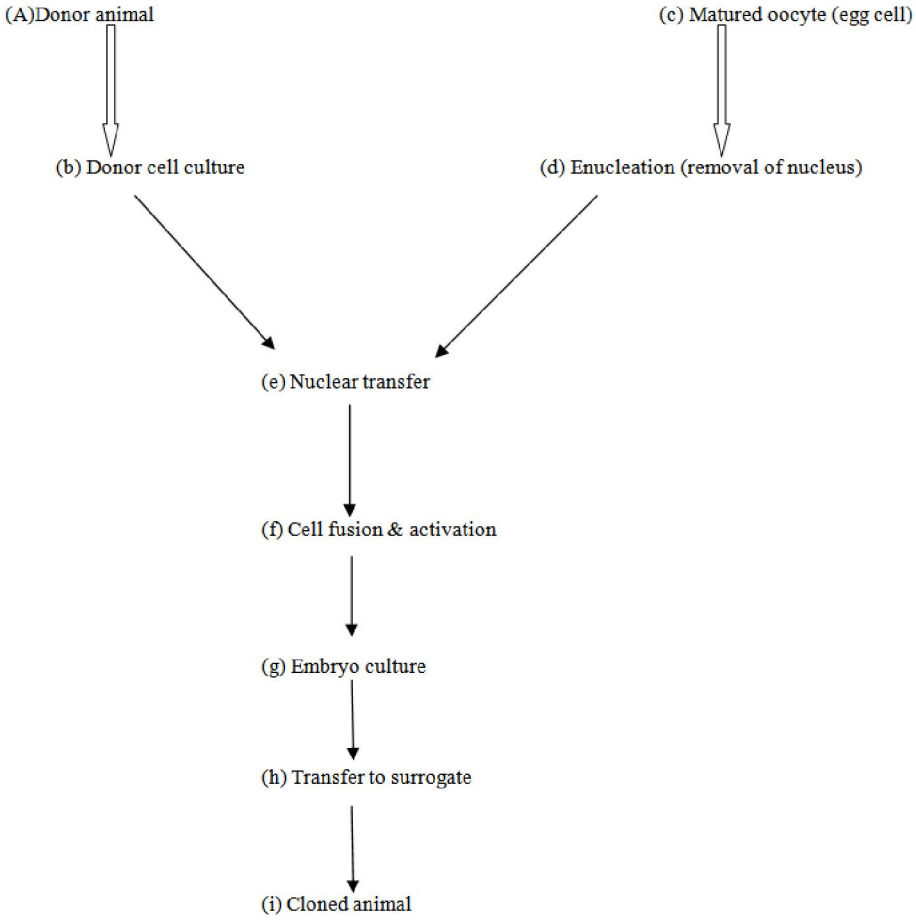

Figure 3.2: The process of cloning by somatic cell nuclear transfer
Source: Adapted from Tian et al (2003) & Campbell et al (2007).

Horses

Equine cloning is possible today, and its value to the industry will be determined over the next few years. Cloning should be viewed as a method for producing a breeding animal rather than as a means to duplicate a performance horse. Late pregnancy losses are rare and this may be because it has a similar type of placenta to the pig, and also be the reason for the difference with ruminants (FVE, 2009).

Cattle

In cattle, only around 6% of the embryos transferred to the reproductive tracts of recipient cows result in healthy, long term surviving clones. Using methods very similar to those used by Willadsen on sheep, First, Prather, and Eyestone produced two cloned calves. Their names were Fusion and Copy. This experiment added cows to the list of mammals that could be cloned by nuclear transfer. Still, mammalian cloning was limited to using embryonic cells as nuclear donors. Cloning using nuclei from differentiated adult somatic cells still wasn't thought possible (UOU, 2014). Overall, up to 40% or more of clones are fatally affected before 6 months of age. While the fetus is unlikely to experience any adverse effect if it dies in uterus, the impact on the surrogate dam has to be considered. This may involve her carrying an overweight fetus and placenta, resulting in dystocia and an eventual Caesarean section. The

welfare of cattle clones that are born alive and survive to weaning may be seriously compromised and they also take longer to reach a normal homeostasis, the impact of which is not known. After weaning there is little evidence that their health and welfare are affected within their production lifetime. Longer term studies that might be relevant for breeding animals are not yet available (FVE, 2009).

Pigs

In the pig, late losses in pregnancy appear to be far less frequent and occur at 35 - 45 days, and also between 60 and 70 days. Retardation of growth in uterus has been recorded. Reduced, litter size is observed (range 1 - 12 with an average of 4 - 6) but can be compensated through transferring more embryos. Neonatal problems are rare (Ortegon *et al*., 2007). The percentage of SCNT piglets born with malformations was higher than in normal animals: 7% for LW clones and 26% for mini pig clones. The piglets were killed when they were malformed for example in their legs, heart, diaphragm, tongue or testes, or when they did not function normally and had for example anorexia or no weight gain (Schmidt *et al*., 2010).

In another report, hematology and clinical chemistry of surviving clones appeared normal. Furrowing rate, litter size, and piglet survival were no different from controls. To evaluate the fertility of the clones' offspring, the authors also produced 14 F2 normal piglets from three F1 females: three F1 female pigs from the cloned mini pig were artificially inseminated with semen from conventional Duroc or Landrace sires, giving rise to 22 piglets, of which in the 3 litters: 13 piglets were born alive, 1 born alive, and in the third litter all 8 were dead; all 14 survivors appeared healthy (Watanabe and Nagai, 2011). It is found that stillbirths and neonatal deaths in the offspring of SCNT clones were 5.6% and 1.4% respectively. All of these piglets were delivered by natural parturition and the birth weight from piglets surviving the prenatal period was similar to that from conventionally bred piglets (EFSA, 2012).

Success rates of cloning

With more than 95% of cloning attempts regularly causing death or severe health problems for cloned animals and their surrogate mothers, there is widespread recognition in the scientific and medical communities that cloning present's serious risks to the animals involved. Yet the FDA has repeatedly glossed over the animal welfare problems raised by animal cloning. In its risk assessment on cloned foods, the US FDA dismisses any concerns by asserting that cloning poses no unique risks to animal health that are not seen with the assisted reproductive technologies already practiced by many large scale livestock operations (Heyman et al., 2007). Somatic Cell.

Nuclear Transfer SCNT is incredibly inefficient. Stresses placed on both the egg cell and the introduced nucleus is enormous, resulting in a low percentage of successfully reprogrammed cells. For example, Dolly the sheep was born after 277 eggs were used for SCNT, which created 29 viable embryos. Only three of these embryos survived until birth, and only one survived to adulthood (Vogel, 2008).

Issues related to cloned animals

Safety of food from cloned animals

Animal cloning may become a commercial venture to help improve the quality of herds. The US Food and Drug Administration (FDA) requested livestock producers and researchers to keep food from animal clones or their offspring out of the food supply. Since then, FDA has conducted an intensive evaluation that included examining the safety of food from these animals and the risk to animal health. Based on a final risk assessment, a report written by FDA scientists and issued in January 2008, FDA has concluded that meat and milk from cow, pig, and goat clones and the offspring of any animal clones are as safe as food we eat every day (FDA, 2008). And also, cloning is often pursued in order to aid in the intensive production of livestock to produce animals those grow faster so they can be slaughtered sooner, and to raise more animals in a smaller space. The rise of factory-farming has already led to serious animal health problems, including animals who grow so big so quickly that their bones break, and animals who are confined to spaces so small they cannot even turn around or stretch. Moreover, the industrialization of agriculture has driven many small farmers out of business, concentrating operations to only a handful of large corporations that are willing to sacrifice welfare and sustainability for profit (Vagita and Gjerris, 2006).

However, EFSA acknowledges that there is limited information on the immune competence of clones and that it is therefore unclear whether there may potentially be an increased public health risk from cloned animal products if clones are more susceptible to infection by pathogens that can also infect humans. EFSA recommends that the susceptibility of clones and their offspring to disease should be investigated further and, if evidence of reduced immune competence of clones becomes available, it should be investigated whether consumption of meat and milk derived from clones or their offspring may lead to an increased human exposure to pathogens (EFSA, 2008).

Other authors have also highlighted the need for further research. For example, Heyman et al (2007) concluded that the quality and safety of milk and meat from healthy adult cloned cattle are broadly similar to those from normal animals but they advise:

Cloned animals, although apparently normal, are however significantly different from contemporary controls maintained in the same conditions, and we feel that more studies on clones and offspring of clones are necessary to evaluate the safety of their use for human consumption."

Ethical issues

Although cloning may eventually become an important technology for livestock production, four ethical issues must be addressed before the practice becomes widespread. First, researchers must establish that the procedure is not detrimental to the health or well-being of affected animals. Second, animal research institutions should evaluate the net social benefits to livestock producers by weighting the benefits to producers against the opportunity cost of research capacity lost to biomedical projects. Third, scientists should consider the indirect effects of cloning research on the larger ethical issues surrounding human cloning. Finally, the market structure for products of cloned animals should protect individual choice, and should recognize that many individuals find the prospect of cloning (or consuming cloned animals) repugnant. Analysis of these four issues is complicated by spurious arguments alleging that cloning will have a negative impact on environment and genetic diversity (Thomson, 2006).

From a Christian Theistic worldview, cloning or intentionally making changes in the human blueprint is playing God. This is something that would not be condoned. Alternatively, from the opposite humanist worldview, human engineering is no more than helping evolution along and it would be negligent not to improve our lot. Our position on this issue like so many other cultural issues is dependent upon our belief on a more fundamental worldview truth. This can be seen in the polarized conflicting views regarding government funding and support or alternatively restrictions and moratoriums animal (Thomson, 2006). Many experts also feel that cloning is not natural because, overall, cloning requires a significantly greater level of involvement and interference with animals' reproductive performance than conventional production methods. Several religious groups, including from Protestant, Catholic, Jewish, Muslim, Hindu, and Buddhist faiths, have rejected animal cloning on ethical grounds. Cloning and genetic engineering are viewed by these groups as tantamount to playing God (FDA, 2005).

Welfare Issues

Welfare of clones

Clones may be born unusually large and with a range of associated health problems, termed "large offspring syndrome" (LOS). This is a common problem in cattle and sheep clones and gives rise to increased perinatal mortality, excess foetal size, abnormal placental development, enlarged internal organs, increased susceptibility to disease, sudden death, reluctance to suckle and difficulty in breathing and standing (EFSA, 2008). LOS was first described in pregnancies with in vitro-fertilized embryos but its incidence is much higher in clone pregnancies (Ibid.). In contrast to cattle and sheep clones, with cloned piglets there is an increased incidence of growth retardation during development in the uterus, resulting in low birth weight (Ibid.).

Panarace et al (2007) reviewed commercial experience of cattle cloning over five years in the US, Argentina and Brazil. Overall, only 9% of transferred embryos resulted in the birth of live calves. Continual losses during gestation were documented, with 37% of recipient cows being pregnant 30 days into gestation, falling to 11% at term. On average, 42% of cloned calves died between delivery and 150 days of age. 18% died during birth, 10% died within the first 24 hours and a further 14% died up to 150 days of age. The most common abnormalities were enlarged umbilical cord (37%), contracted flexor tendons (21%), calves depressed/prolonged recumbency (20%), respiratory problems (19%), hyper/hypothermia (17%) and persistent urachus (failure of the connection between the bladder and the naval to close) (10%). 37% of calves required treatment with antibiotics.

Watanabe and Nagai (2009) reviewed mortality in cloned cattle and their offspring in Japan. 16.4% of cloned calves were stillborn and a further 14.4% died within the first 24 hours. 24.1% of cloned cattle died due to disease during the first 30 days of their life. The main problems identified in dead calves two to three days after birth were respiratory problems (35.3%) and deformed hearts (11.8%). After four days the major cause of death was pneumonia. By 200 days of age, mortality in cloned cattle reached the same level as conventionally bred cattle. Mortality in the offspring of cloned cattle did not differ significantly from mortality in conventionally bred cattle.

Loi et al (2004) reviewed experience of sheep cloning over six years in Europe. While early development of clone embryos appeared similar to control embryos produced by fertilization, the majority of clone pregnancies established at day 30 were lost over the following months and problems were observed in the few pregnancies that were carried to term. Only 13% of clone pregnancies reached term. Around 40% of the surrogate animals carrying clones developed acute hydroallantois (rapid accumulation of fluid in the placenta during the latter stages of pregnancy) and 45% showed a premature placental ageing (post-mature placenta). In both cases, the offspring were born alive but died at various times after delivery. Post-mortem investigations revealed

that the ultimate cause of death was a direct consequence of placental abnormalities. Overall, placental abnormalities were observed in 67% of live clone births. The abnormalities leading to pre- and post-natal mortality in the large majority of clones that developed to term were hardly seen in in vitro-derived fertilized embryos and were totally absent in naturally mated ewes. At the time of the review no cloned sheep remained alive.

In its 2008 Opinion on Animal Cloning, the EFSA Scientific Committee, which advises the European Commission, concludes (EFSA, 2008):

"The health and welfare of a significant proportion of clones, mainly within the juvenile period for bovines and perinatal period for pigs, have been found to be adversely affected, often severely and with a fatal outcome.

In June 2009, the EFSA Scientific Committee examined further evidence and confirmed that these conclusions are still valid (EFSA, 2009).

In order for the SCNT process to be successful, the donor cell must be correctly reprogrammed so that gene expression is properly controlled in order to allow normal development. The major cause of abnormalities in clones appears to be incorrect reprogramming of the donor cell genome. EFSA (2008) states:

"Epigenetic dysregulation [abnormal control of gene expression] is considered to be the main source of adverse effects that may affect clones and result in developmental abnormalities."

The effects of these reprogramming failures are obvious in the majority of cloned animals, resulting in a range of physical abnormalities and illnesses which clearly cause suffering andoften death. Even those clones that survive and appear normal may have underlying abnormalities. During early development in mammal embryos, the majority of genes on the X chromosome are repressed (switched off) in a process called "X inactivation". This is one of the epigenetic processes that may not happen properly in clone embryos. Research by Cao Geng-Sheng et al (2009) suggests that even apparently normal clones may have subtle epigenetic abnormalities.

There is insufficient data available to date to be able to draw firm conclusions about the long-term health of cloned livestock. However, studies of cloned mice suggest that surviving clones may suffer long-term health problems, including obesity and abnormalities in a number of organs and early death (Ogonuki et al, 2002; Tamashiro et al, 2002; Inui, 2003; Shimozawa et al, 2006).

Welfare of surrogate dams

Pregnancy failure and abnormal or difficult delivery (dystocia) are common in clone pregnancies and delivery is often by Caesarean section (EFSA, 2008). Initial pregnancy rates in cattle used as surrogate dams are similar between those carrying clones and those carrying embryos produced by artificial insemination or in vitro fertilization. However, there is a continued pregnancy loss throughout the entire gestation period in those carrying clones which is not observed with other ARTs and embryo survival is only one third of that following in vitro embryo production (Ibid.). The high rate of pregnancy failure is linked to placental abnormalities (Hill et al, 2000; Oishi et al, 2006; Kim et al, 2009).

In its 2008 Opinion on Animal Cloning, the EFSA Scientific Committee, which advises the European Commission, states (EFSA, 2008):

"For surrogate dams, an increase in pregnancy failure has been observed in cattle and pigs and increased frequencies of hydrops [abnormal accumulation of fluid in the foetus] and dystocia have been observed especially in cattle. This together with the increased size of the offspring (large offspring syndrome) makes Caesarean sections more frequent in cattle carrying a clone than with conventional pregnancies... [D] ystocia carries the risk of unrelieved "extra" pain during birth due to the large offspring. If the dam has to have a Caesarean section then that itself carries the risk of pain and anxiety due to the procedures involved, including a failure to provide adequate post-operative pain relief. If the Caesarean section is not planned then there is the added burden of the pain of both the dystocia and the Caesarean section."

Welfare of clone offspring

The use of cloning in commercial livestock breeding is therefore likely to accelerate the spread of genetics that are associated with poor welfare, leading to greater suffering from health and welfare problems connected with fast growth and high yields. It is increasingly being recognised that livestock breeding must pursue different goals, aimed at producing more robust animals, if farm animals are to have an acceptable quality of life (Rauw et al, 1998; Sandøe et al, 1999).

The European Parliament (2008) states:

While the principal purpose of cloning is to produce multiple copies of animals with fast growth rate"s or high yields, traditional selective breeding has already led to leg disorders and cardiovascular malfunction in fast-growing pigs, and lameness, mastitis and premature culling in high-yielding cattle... cloning the fastest-growing and highest-yielding animals will lead to even higher levels of health and welfare problems.

Conclusions And Recommendations

SCNT produces animals that are genetically unlike any animal found in nature. Many species have

been cloned since Dolly the sheep, the first mammal to be cloned from an adult cell, was born in 1996. There are now estimated to be around 6000 farm animal clones worldwide.

Cloning can be used to increase the efficiency of production of transgenic animals using nuclear transfer of cultured transgenic cells. The use of cloning technology is therefore facilitating the development and commercialisation of genetically modified animals for biomedical, research and food production purposes. Compassion in World Farming is opposed to the genetic modification of farm animals.

The welfare of animals used as surrogate dams is also adversely affected because of the high rates of pregnancy failure, birthing difficulties and Caesarean sections.

Although the offspring of clones do not appear to suffer any obvious abnormal effects, the use of cloning to replicate elite high-yielding animals for breeding is likely to accelerate the spread of livestock genetics associated with poor welfare, leading to greater suffering from health and welfare problems connected with fast growth and high yields.

Reduced genetic diversity increases the susceptibility of livestock populations to diseases and other risk factors. This raises the possibility of large numbers of animals succumbing to diseases to which they are susceptible, with potentially serious animal welfare, social and economic consequences.

An efficient animal cloning technology would provide many new opportunities for livestock agriculture, human medicine, and animal conservation. Nuclear cloning involves the production of animals that are genetically identical to the donor cells used in a technique known as somatic cell nuclear transfer (SCNT). However, at present it is an inefficient process in cattle, only around 6% of the embryos transferred to the reproductive tracts of recipient cows result in healthy, long term surviving clones. Of concern are the high losses throughout gestation, during birth and in the postnatal period through to adulthood.

Success rate of cloning is very low. To produce the first cloned animal Dolly, for example, 277 cloned embryos were implanted and only 1 animal was born successfully. Cloning also faced ethical and welfare challenges. Opponents believe that cloning is completely unethical and is against animal welfare.

Generally cloning has many advantages and some disadvantages. If further studies are carried out, the disadvantages can be reduced. So to make use of the advantages, cloning should be accepted and developed. Ethical challenges facing this technology are less important.

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Referencs

1. Bailey, R. (2014): Animal cloning techniques. *Animal care practice,* 23(5):68-92.
2. Benagiano, G. and Primiero, F.M., (2002): Human reproductive cloning. *Introduction journal Gynecology, Obstet*, 79: 265–268.
3. California Polytechnic State University (CPSU) (2010): Economic Analysis of the Impact of Cloning on Improving Dairy Herd Composition. *Express Academic Express*. 13(2):10.
4. Dematawewa, C. M. and Berger, P. J. (1998): Break even cost of cloning in genetic improvement of dairy cattle. *Journal of Dairy Science*, 81(4), 1136-1147.
5. Europe Food Safety Authority (EFSA) (2008): Scientific Opinion of the Scientific Committee on a request from the European Commission on Food Safety. *Nature*, 767: 1-49.
6. Europe Food Safety Authority (EFSA) (2012): Cloning farm animals, *Nature*.10 (7):27.
7. Essays Library (EL) (2010): Essays on cloning history. *http://hotessays.blogspot.com/2010/02/cloning-history essay.html* Accessed date 11/7/2014. FDA (2005): Consider morals, ethics in animal cloning Policy. *Animal reproduction science,* 11(38).47.
8. Food and Drug Administration (FDA) (2008): Animal cloning, UK. New Hampshire Avenue Silver Spring, Pp: 7-27.
9. Federation of Veterinary of Europe (FVE) (2009): Are animals be cloned. *Reproduction fertility and de* ral School (HAS) (2003): cloning. http://*bsp.med.harvard.edu.q=node/1*. Accessed date 12/5/14.
10. Health Research Funding (HRF) (2014): Cloning. *http://healthresearchfunding*. Accessed date 12/15/14.

11. Heyman, Y., Chavatte, P., Berthelot, V., Fromentin, G., Hocquette, J. F., Martignat, L., and Renard, J.P. (2007): Assessing the quality of products derived from cloned cattle. *Theriogenology,* 67(1):134-141.
12. McClintock, A. E. (1998): Impact of cloning on cattle breeding systems. *Reproduction, fertility and development, 10*(8):667-670.
13. Ortegon, H., Betts, D. H., Lin, L., Coppola, G., Perrault, S. D. and Blondin, P. (2007): Genomic stability and physiological assessments of live offspring sired by a bull clone. *Theriogenology,* 67(1):116-126. *velopment*, 52(3):87-101. Harvard Agricultu.
14. Schmidt, M., Winter, K. D., Dantzer, V., Kragh, P. M., Du, Y., Lin, L., Liu, Y., Vajta, G., Sangild, P. T., Callesen, H. and Agerholm, J. S., (2011): Maternal endometrial edema may increase peri-natal mortality of cloned and transgenic piglets. *Reproduction fertility and development*, 23: 645- 653.
15. Shugerman, L. (2014): Cloning in animals. *http://healthresearchfunding.org/pros-cons-animal-cloning/* Accessed date 12/14/14.
16. Thomson, P.B. (1999): Ethical Issues in Livestock Cloning. *Journal of agricultural and environmental ethics*, 11(3): 197-217.
17. Twyman. R, (2003): Animal cloning. *http://genome.wellcome.ac.uk/doc_wtd021034.html*. Accessedate 12/10/2014.
18. University Of Utah UOU, (2014): What cloning and why. *http://learn.genetics.utah.edu/content/cloning/whatiscloning/* Accessed date 11/7/2014.
19. Vajta, G., and Gjerris, M. (2006): Science and technology of farm animal cloning. State of the art. *Animal Reproduction Science, 92*:211-230.
20. Van Arendock, J.A.M., & Bijma, P. 2003). Factors affecting commercial application of technologies in dairy cattle in Europe-A modeling approach. *Theriogenolgy*, 59:635-649.
21. Vogel, G (2008): Breakthrough of the year. Science, 3(2):1766–1767.
22. Watanabe, S. and Nagai, T. (2011): Survival of embryos and calves derived from somatic cell nuclear transfer in cattle. *Animal Science Journal*, 82:360-365.
23. Wilmut, I., Schnieke, A. E., McWhir, J., Kind, A. J., and Campbell, K. H. S. (1997): Viable offspring derived from fetal and adult mammalian cells. *Nature*, 385:810-813.

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