ISSCR INTERNATIONAL SOCIETY FOR STEM CELL RESEARCH

Human Fetal Tissue: A Critical Resource for Biomedical Research

Fetal tissue research has made major contributions to our understanding of biology and the development of new medical technologies, including vaccines for many diseases, that have saved millions of lives.

Fetal tissue is an essential "gold-standard" resource that enables laboratory-based research into how human tissues and organs develop. With the consent of donors, this unique and valuable tissue can be used for research into basic biological processes and human development, as well as creating new treatments for life-threatening diseases.

Fetal tissue is obtained from spontaneous miscarriages and legal abortions. In each case, the fetal tissue would be discarded if not donated by patients for medical research. Ongoing access to human fetal tissue that has been obtained legally and with donor consent is required to address many important questions in biomedical research and for the development of new therapies.

The International Society for Stem Cell Research (ISSCR) endorses fetal tissue research as essential to the prevention and treatment of life-threatening diseases.

Below, we outline examples of how the use of fetal tissue has led to therapies that have saved lives as well as ways in which fetal tissue research continues to be necessary for medical advances.

1. **Parkinson's disease**
2. **Huntington's disease**
3. **Blindness**
4. **Pregnancy**
5. **Zika Virus**
6. **HIV**

About the International Society for Stem Cell Research (ISSCR)
The International Society for Stem Cell Research (ISSCR) is an independent, nonprofit organization established to promote and foster the exchange and dissemination of information and ideas relating to stem cells, to encourage the general field of research involving stem cells and to promote professional and public education in all areas of stem cell research and application.

1. PARKINSON'S DISEASE AND FETAL TISSUE

Parkinson's disease involves a progressive loss of midbrain dopamine-producing neurons, leading to problems with movement and cognition. The symptoms of Parkinson's disease can be ameliorated by transplanting dopamine-producing neurons into the brain, to replace those lost as a result of the disease. The dopamine-producing neurons that have been used in past and pending clinical trials have been obtained from human fetal brain tissue.

The use of human fetal midbrain tissue as a source of replacement dopamine-producing neurons in the treatment of Parkinson's disease goes back to the late 1980s and continues to the present day. This work (Lindvall et al., 1990; Freed et al., 1992; Kefalopoulou et al., 2014) showed that developing human neurons from fetal tissue could:

- survive being transplanted into the adult human brain;
- make and receive connections;
- release dopamine; and
- make a subset of patients much better for years.

These findings opened the whole field of regenerative medicine in Parkinson's disease and laid the foundations for all of the subsequent work now underway exploring different sources of neurons for this devastating condition (Barker et al., 2013, 2015).

Multiple clinical trials are planned to start in the next few years to test the effectiveness of transplanting dopamine-producing neurons obtained from human fetal tissue, embryonic stem cells, or induced pluripotent stem cells into the brains of patients with Parkinson's disease. An EU-funded clinical trial (TRANSEURO) is now underway, seeking to minimize side effects and maximize benefit using refined protocols for patient selection, tissue preparation and implantation, and immunosuppressive treatment.

The use of human fetal tissue to study the development of the human midbrain and the dopamine-producing cells within it has provided new insight into disease pathogenesis and has profoundly improved our ability to generate dopamine-producing neurons from other stem cells. Improved production of dopamine-producing cells is thought to be one major advance that could improve outcomes in the current generation of clinical trials by:

- ensuring that new stem cell-derived dopamine cells function like normal dopamine cells of the type lost in Parkinson's disease (Grealish et al., 2014); and
- delineating the different types of dopamine cell that exist in the human midbrain, only some of which are lost in Parkinson's disease (DDPDGENES consortia 2011-2015).

This work has also clearly shown that normal human brain development is not the same as that seen in the mouse, the commonly used alternative model system.

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2. HUNTINGTON'S DISEASE AND FETAL TISSUE

Huntington's disease is marked by a loss of striatal projection neurons in the brain, leading to problems with movement and cognition. The disease is caused by inheriting a mutated version of a specific gene. Drugs are available to manage the symptoms but they offer only limited relief.

In the 1990s, attempts were made to treat the disease by transplanting striatal neurons from human fetal tissue to replace the neurons that are lost due to the disease. These studies showed some benefits in some patients (Bachoud-Lévi et al., 2006, 2000; Reuter et al., 2008), although not in all cases (Barker et al., 2013; Hauser et al., 2002). Nevertheless, this approach has shown that human fetal striatal neurons can survive in the brains of patients with Huntington's disease, leading to ongoing research into the development of new stem cell-based therapies.

The use of fetal tissue has been critical for understanding the normal development of the human striatum. This has two important implications:

- It enables us to understand how this neural network is normally generated and this instructs us as to how to make better stem cell-derived striatal neurons (Delli Carri et al., 2013; Onorati et al., 2014); and
- It provides insights into developmental processes that might be recapitulated in the disease and by so doing opens up new therapeutic strategies.

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3. BLINDNESS AND FETAL TISSUE

The retina is the nerve tissue lining the inside of the eye. The purpose of the retina is to receive light that the lens has focused, convert the light into neural signals, and send these signals on to the brain for visual recognition. The retina plays an essential role in vision; retinal malformation, damage, or degeneration can cause permanent blindness. For example:

- *Retinopathy of prematurity.* Retinopathy of prematurity is a leading cause of blindness in babies that are born early. To prevent this disease, researchers must study how the human retina develops normally, so that they can understand how this process is perturbed when babies are born prematurely (Ma et al., 2015). This can only be done accurately using human fetal retinal tissue (Maminishkis et al., 2006).

- *Age-related macular degeneration.* There are numerous degenerative diseases that lead to blindness, including age-related macular degeneration, which affects approximately 1 in 5 people over age 75. Studies of fetal human retinal pigment epithelium have helped scientists understand the disease process and identify new potential therapeutic approaches (Zhou et al., 2015).

For individuals who have already suffered retinal cell loss due to traumatic injury or diseases such as age-related macular degeneration or an inherited condition known as retinitis pigmentosa, cell transplantation offers the possibility of restoring sight. Human fetal retinal tissue was used to pioneer retinal cell transplantation for blinding disorders (Seiler et al., 2012), and this tissue is now being used in early stage clinical trials around the world (for example, Lawley et al., 2015).

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4. PREGNANCY AND FETAL TISSUE

The placenta is an active, complex organ that plays a critical role in pregnancy to keep the baby alive and healthy. Problems with the placenta are common causes of pregnancy complications, and many of these remain difficult to predict and treat. While much has been learnt about the placenta in recent decades, there is much more that needs to be understood.

There are many reasons why studying human embryonic and fetal tissue are critical for research about the placenta's role in normal and abnormal pregnancy. Key examples include:

- *Species variation.* Everything about the placenta, from the cell types it contains to how it interfaces with maternal cells of the blood and uterus, varies among species, including between humans and nonhuman primates. The mouse placenta, which is implanted in the uterus for a little over two weeks, appears to have a very different way of engaging the mother's immune system as compared to its human counterpart, which must avoid rejection for nine months.

- *Differences in function between the early pregnancy- and term-placenta*. The placenta's lifespan matches that of pregnancy. Thus, the placenta at term exhibits features of aging. To understand how it forms and functions, researchers need access to early gestation samples, which allow us to study how it changes over the course of pregnancy and how disruption of these changes can lead to complications during pregnancy.

- *Impact of early-stage placental development.* Many of the most common pregnancy complications (*e.g.,* preeclampsia, a subset of preterm births) are thought to be the result of defects in the early stages of placental development, making it imperative to investigate normal placental development during the first and second trimesters of pregnancy.

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5. ZIKA VIRUS AND FETAL TISSUE

Fetal tissue has a unique and important role in our ability to tackle the global emergency embodied in the Zika virus outbreak. The Zika virus, which is usually relatively benign in adults, can have devastating effects on the developing human fetus. In otherwise healthy pregnant women, the Zika virus can cross the placenta and infect the growing fetus, where it can destroy the developing brain, resulting in microcephaly as well as a host of related malformations.

Scientists are trying to better understand how the virus infects the fetus, how it causes cell death in the developing brain, and how targeted therapies can be designed.

Animal and cell culture models are insufficient:

- The effects of the Zika virus on the developing human brain have been hard to reproduce in animal models such as mice or rats; and
- Human stem cell-derived cerebral or brain organoids (small, three-dimensional models of embryonic human brains that form some of the structures of the brain) have been used to try to address this limitation, but it is not clear that these *in vitro* models accurately reflect the disease. One significant limitation of these systems is that they are incomplete, and lack essential cell types that are involved in infectivity and spread of the virus within the human brain, such as the microglia and cells that line blood vessels in the brain.

The study of human fetal tissue is driving new treatment strategies:

- The use of donated fetal tissue, including placental tissue, has provided the best understanding of how Zika viruses behave in the body. These tissue samples have taught us how the virus is able to cross the placenta and infect human brain cells to produce the malformations observed in affected infants (Mlakar et al., 2016; Nowakowski et al., 2016; Tabata et al., 2016); and
- Insights gained through the study of fetal tissue samples are already guiding the development of drugs that may protect the unborn baby from the ravages of the Zika virus (Retallack et al., 2016).

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6. HIV AND FETAL TISSUE

Human Immunodeficiency Virus (HIV) attacks the body's immune system, destroying T cells that are critical for fighting disease and infection. More than 1.2 million people in the US are living with HIV (http://www.cdc.gov/hiv/statistics/overview/index.html). While HIV can be controlled with access to good medical care, in 2013, HIV was the 8th leading cause of death for those aged 25-34.

A humanized mouse is a mouse that carries functioning human cells, tissues, or organs. "BLT" humanized mice are created by transplanting human fetal liver and human fetal thymus into immunocompromised mice that naturally lack a functioning immune system, to create a human blood-forming system in these mice. This leads to the generation of human immune system cells, including T cells, in mice. This is a powerful and widely used model for studying HIV as well as other human viruses. Studies using "BLT" mice have:

- Provided insights into HIV biology not possible in others systems (Adoro et al., 2015; Murooka et al., 2012);
- Allowed the development of novel approaches to HIV prevention that could not have been studied in other systems (Balazs et al., 2014; Klein et al., 2012; Lu et al., 2016); and
- Allowed for the testing of drugs in human cells *in vivo* in a way that could not have been done in other preclinical systems (Olesen et al., 2016).

In addition, "BLT" humanized mice are being used to explore how other viruses, including the dengue virus, infect human cells and for the testing of antiviral drugs (Frias-Staheli et al., 2014).

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