Introduction

The primary purpose of statutory regulations is to serve as a common framework for ensuring with confidence the current state of the art on the quality and safety of tissues and cells for therapeutic benefit. Equally, the regulations and linked guidance should be compatible on a wider level to encourage equitable distribution between countries, where regulations may be similar and well established, in early development, or in their infancy. Many countries have implemented or are refining their healthcare services to provide a better standard of care to patients and to enhance the use of tissues and cells for clinical applications. The steps involved in the processing of tissues and cells are critical activities and require the application of specific controls to prevent contamination and cross-contamination, as well as to maintain quality and safety. This chapter gives an overview on the status, history, and scope of key regulations; the practical aspects of implementation; the interface with advanced therapy medicinal products (ATMPs); medical devices; biologics; and some global perspectives.

The therapeutic application of tissues or cells is preceded by a series of complex and inter-related activities, from donor selection and screening, infectious disease testing, tissue and cell recovery, processing, temporary or long-term storage, and distribution for use in the clinical setting. The organization and delivery of healthcare systems are structured and operate quite differently, according to resources and health programs, to address epidemiological characteristics of the endemic population. To encompass these
diverse organizations, and their inter-linked activities, a tissue establishment can be defined as:

a tissue bank or a unit of a hospital or another body where the activities of processing, preservation, storage or distribution of human tissues and cells are undertaken. It may also be responsible for the procurement or testing of human tissues and cells [1].

Organizations in healthcare services or the commercial sector performing one or typically more of these activities should be authorized by their national regulating body and are expected to verify compliance with appropriate requirements, so governing the quality and safety of tissues and cells.

Professionals working in the tissues and cells sector have not been wholly amenable to “allografts” being referred to as “products” or “devices;” and some have reservations regarding the use of the term “manufacturing” being applied in the context of human tissues and cells donated altruistically for the benefit of others. However, regulatory preferences and established terminology of other healthcare sectors often over-ride the human dimension in this donation-related work and such terms are commonly applied. This chapter discusses the requirements and language that affect professionals involved in the processing of tissues and cells for transplantation.

To assist all countries where cell and tissue banking activities are developing, and some without regulatory oversight, the World Health Organization (WHO) convened meetings in 2004 and 2005 with the participation of numerous experts and regulators from across all of the WHO regions. Global standards necessary for the development of safe tissues and equitable and ethical access to donation and transplantation of tissues and cells were discussed. This resulted in two useful aide-memoires for use by emerging health authorities, published in 2005 [2] and 2006 [3]. Among other key elements, both documents promote the benefits of quality systems and quality programs. The concept of a supplementary aide-memoire on the generic principles of inspection for tissue and cell establishments, including processing controls, was initiated in late 2009. Following this, in May 2010, the 63rd World Health Assembly endorsed a resolution on the WHO guiding principles for human cell, tissues and organs transplantation [4], which urged Member States to formulate and enforce their own policies, laws and, regulations on this subject.

**Regulations – development, scope, and principles**

**Europe**
The forerunner to the Tissue and Cell Directives (see below) was a comprehensive guidance document published by the European Health Committee
of the Council of Europe which defined the standards required, and quality assurances to be achieved, for the transplantation of organs, tissues, and cells. It was updated again in 2010 [5] and is a valuable source of scientific information and clinical practice guidance. The first legislative proposal to set binding requirements on the quality and safety of human tissues and cells by the European Commission was presented in 2002. Its objective was to facilitate the cooperation and collaborative activities between the healthcare systems of the Member States. The regulation of substances of human origin, such as blood, tissues, cells, and organs, at a European Union (EU) level became legally possible when the Treaty of the EU was amended in 1995 by Article 152 of the Treaty of Amsterdam. This article extended the legal competence of the EU to certain aspects of healthcare stating that “Measures setting high standards of quality and safety of organs and substances of human origin, blood and blood derivatives” would be adopted. The legal basis would allow individual Member States to adopt more stringent requirements if they considered it appropriate. National representatives of the Member States, together with their experts, then negotiated a technical and political approach for the regulatory framework to control these activities for the greater protection of public health.

The entry into force for the principal Directive 2004/23/EC [1] was April 2006, which preceded Directive 2006/17/EC [6] in November 2006 and Directive 2006/86/EC [7] in September 2007. Collectively these are the “Tissue and Cell Directives” which address the standards of quality and safety for the donation, procurement, testing, processing, preservation, storage, distribution, and import/export of human tissues and cells. Since then the Member States have transposed the Directives into national laws and put in place the implementation measures for their application. Aspects of these Directives apply also to other manufactured products that are regulated as medicines but are wholly or partially derived from human tissues and cells. Specifically, donation, procurement and testing of the tissues and cells used for the manufacture of such products are regulated by the Tissue and Cell Directives. Tissues and cells used as an autologous graft in the same surgical procedure and those used for research studies not involving application to the human body, are excluded, as are organs, blood, and blood products. The Tissue and Cell Directives therefore regulate tissues for transplantation such as bone, skin, heart valves and corneas, cells such as hematopoietic stem cells from bone marrow, peripheral blood, or cord blood as well as other cells that are not extensively manipulated, plus gametes and embryos.

**United States**

As a comparison, tissue regulations in the USA were first introduced in the 1980s and aimed at processing requirements for the manufacture of two specific human tissue types, corneal lenticules and, separately, human dura mater. By mid-1991, cryopreserved allograft (replacement) heart
valves were singled out as a “product,” like man-made replacement heart valves, requiring data submission to demonstrate safety and effectiveness. These allografts were classified as medical devices by the Food and Drug Administration (FDA) and regulated by their Center for Devices and Radiological Health (CDRH). Although enforcement of requirements related to the Class III medical device designation for allograft heart valves was rescinded due to a decision in late 1994 [8], other federal regulations were published in 1993 and were applicable to a variety of conventional tissue allografts. This federal oversight of human tissues for transplantation advanced further after two specific events. In 1991, there was realization of HIV transmissions from the transplantation of human tissue from one donor reported by the Centers for Disease Control and Prevention (CDC). Soon thereafter, the FDA received reports from US tissue banks of brokers selling unprocessed tissue from improperly screened and tested donors from Russia, eastern Europe, and Central and South America [9]. In response to the concern for public health, the FDA published an Interim Final Rule entitled “Human Tissue Intended for Transplantation” [10]. With this publication, the FDA’s Center for Biologics Evaluation and Research (CBER) was assigned responsibility for oversight of tissue establishments that screen donors, and recover, process, store, and/or distribute tissue for transplantation. This Interim Rule, codified in 21 CFR Part 1270, included minimum requirements for screening and testing tissue donors, and maintaining procedures and records with specific emphasis on preventing the transmission of viral hepatitis and HIV. The American Association of Tissue Banks (AATB) and the Eye Bank Association of America (EBAA) actively promoted communication between industry professionals and CBER to ensure further development of regulations would be effective. Various public workshops and meetings were held and in June 1997 the FDA published a Final Rule and Guidance Document that amended parts of the Interim Rule. These focused on considerations involving the eligibility of deceased donors such as criteria to be used when screening, infectious disease tests that must be performed, plasma dilution evaluation of the blood sample used for testing, and the physical assessment of the donor.

FDA also announced their “Proposed Approach to the Regulation of Cellular and Tissue-based Products” which outlined a tiered, risk-based strategy for regulating traditional tissues as well as new cell/tissue allograft products. This oversight was aimed at the control of contamination and cross-contamination throughout all manufacturing steps. The “Tissue Action Plan” (TAP) was developed to guide this proposal to fruition. Soon after the start of the new millennium, the FDA described a more comprehensive regulatory framework, codified it in 21 CFR Part 1271, and promoted regulations via final rules that encompass registration of tissue establishments and listing of products (2001) [11], donor eligibility requirements (2004) [12], expectations for maintaining Current Good Tissue Practice (2004) [13],
and reporting, inspection, and enforcement regulations. The last of the final rules published in 2004 became effective for cells and tissues recovered from donors after May 25, 2005. The regulations in 21 CFR 1271 describe the scope of the expanded oversight to be applicable to human cells, tissues, and cellular and tissue-based products (HCT/Ps). These are described as conventional tissue (e.g., bone, including demineralized bone, skin, tendons, ligaments, fascia, pericardium, dura mater, cartilage, heart valves, veins/arteries, amniotic membrane), ocular tissue (i.e., corneas, sclera), reproductive tissue (i.e., semen, oocytes, embryos), and hematopoietic stem/progenitor cells (including cells derived from peripheral or cord blood). Federal regulations have matured to include a wide variety of allograft cell/tissue products intended for implantation, transplantation, infusion, or transfer into human recipients, whether sourced from living or deceased donors.

21 CFR 1271 is not applicable to some human-derived therapeutic products, such as solid organs used for transplantation; blood or blood components; minimally manipulated bone marrow; secreted or extracted human products such as milk, collagen, and cell factors; and products derived from or exposed to cells, tissues, or organs from non-human animals. In addition, exemptions include autologous tissue re-implanted within the “same surgical procedure.” A regulatory ladder (tiered, risk-based approach) was created to address nuances if HCT/Ps are combined with other materials or whose use and effect on the body become complicated after further manipulation. In a number of ways, HCT/Ps may be elevated to a higher regulatory scheme that encompasses requirements for medicinal products, specifically those attributable to FDA designation as a “biologic” or a “medical device.”

**Australia**

In the Therapeutic Goods Act 1989 and subsequent amendments, human organs, tissue, and cellular products, as well as tissue- and cell-based derivatives, are regulated by several different routes. The related Australian Code of Good Manufacturing Practice (GMP) [14] is applicable to human tissue for transplantation when procured, stored, and supplied without deliberate alteration to its biological and mechanical properties (e.g., dura mater, heart valves, skin, corneas, and bone). It was published by the Therapeutic Goods Administration (TGA) in 2000 and adopts GMP expectations for tissue establishments. The quality system requirements include quality objectives, organizational structure, monitoring systems, and management review. It incorporates many quality systems elements from the ISO 9000 series of standards and equally applies these principles for the control of blood products. Similar to the regulatory designations for human tissues and cells in Europe and the Americas, these allografts may be regulated as medicines or therapeutic devices, depending on their biological/mechanical properties or their therapeutic purpose. The regulation of viable human and animal
tissues that undergo processing and modification prior to implantation or infusion to patients have yet to be fully addressed in the current legislation. In July 2002, the Australian Health Ministers supported the TGA for a regulatory framework for human tissues and emerging biological therapies. The ensuing Human Cellular and Tissue therapies (HCT) framework was originally planned as a part of the regulatory partnership between Australia and New Zealand, but its postponement in July 2007 meant the HCT framework was delayed. In July 2009 the Australian Government moved forward independently with this framework, which excludes assisted reproductive tissues and solid organs. Hematopoietic progenitor cells are envisaged to be part of the framework, after a public consultation phase with that profession’s stakeholders. Recently, amendments to the Therapeutic Goods Regulations 1990 that create a new regulatory framework for biologicals were passed by Executive Council on March 10, 2011. The biologicals framework commenced on May 31, 2011. After this date, all products within the scope of the framework will need to comply with the requirements made under the new legislation, but a three-year transition period is provided for establishments to come into compliance. Four classes of biologicals have been developed, based upon risk, extent of manipulation applied, and whether use is homologous or not. In this regard, similarities exist with the European and US perspective but a major difference is that the submission of an extensive dossier for products is required for not only Class 3 and 4 biologicals, but also for Class 2. A dossier of information on a product is akin to a “device history file,” so requiring this extensive compilation to characterize a Class 2 biological, which is equivalent to a conventional tissue allograft in the USA, is an onerous task for tissue banks. Guidance documents are being developed to assist stakeholders to better understand the TGAs expectations. It is also interesting to note that these biological products (of any class), once reviewed and approved by the TGA, become officially listed on the Australian Registry of Therapeutic Goods (ARTG) and a reimbursement fee is assigned to each one. This is an agreed minimum fee for supplying the product within Australia and must be honored by insurance companies and healthcare providers.

Within the TGA regulation for process control, the quality assurance of the manufactured product is described and encompasses specific requirements for documentation, materials, procedures, conditions, quality control related to sampling, validation of processes to be supported by data, and product release. The glossary contains definitions helpful for the qualification of equipment and validation of processes, prospective and retrospective; and yet, to date, specific guidance is not offered regarding the means to validate the steps used in the processing/manufacture of human tissues. The TGA regularly reviews technical data submitted by tissue establishments that supports the processing steps they use and their claims.
Canada
Continuing with the development of national regulations, in June 2007, Health Canada published “The Safety of Human Cells, Tissues and Organs for Transplantation Regulations” (CTO regulations) [15], with a further supplement which came into force in June 2008. The draft “Guidance Document for Cell, Tissue and Organ Establishments, Safety of Human Cells, Tissues and Organs for Transplantation” [16] was released at this time and finalized in April 2009. The CTO regulations establish safety requirements relating to the processing and handling of CTO products, resulting in improved protection of the health and safety of recipients. These regulations directly reference sections of the General Standard CAN/CSA Z900.1 [17], entitled “Cells, Tissues, and Organs for Transplantation and Assisted Reproduction: General Requirements,” along with four of the five standards for specific organ and tissue types, thus making them mandatory. Based on the National Standards, the regulations set out basic safety requirements with respect to donor screening, donor testing, collection/retrieval, processing, preservation, packaging, labeling, storage, quarantine, record-keeping, distribution, importation, error, accident and adverse reaction monitoring, reporting, and investigation. Establishments distributing CTOs are required to have a quality assurance system [17] in place that complies with the requirements of the regulations, which enables them to perform their activities effectively. Important components of a quality assurance system are the standard operating procedures (SOPs), which must be kept current and approved by the medical or scientific director.

The prescription of requirements for CTO processing, as defined by regulations in Europe, the USA, and elsewhere, are not clearly elucidated in the federal regulation but are described in the CSA standards for tissues and cells. By definition, the term “processing” (French: “traitement”) means a series of steps, which in other regulations are “manufacturing” phases, such as: donor screening, testing and suitability determination; retrieval; testing/measurements performed on tissue; preparation for use; preservation; quarantine; banking; packaging; and labeling. The CTO regulations/guidance and the CSA standards have not yet defined qualification, validation, or verification, and these terms are attributed to personnel, records, equipment, supplies, labeling, and technical review for tissue release. The section of Process Control in the CSA standards (Z900.1–03) lists requirements for the major components of the operations for a tissue establishment. Updates to these CSA standards are expected during late 2011 or early in the following year.

Brazil
The public health regulations in Brazil are structured under the guidance of Agência Nacional de Vigilância Sanitária (ANVISA) whose mission is to foster health protection by exercising contamination control on the production and
marketing of products and services. Blood, other tissues, cells, and organs are within this oversight and ANVISA works, with state and county systems, to promote sanitation vigilance as a social right/protection. ANVISA was established by law [18] in January 1999 and is designated an autonomous agency operating as an independently administered, financially separate regulatory agency. In the Federal Public Administration, ANVISA has a management contract with the Ministry of Health [19] for their responsibilities to coordinate the National Sanitary Surveillance System (SNVS) [20], the National Program of Blood and Blood Products, and the National Program of Prevention and Control of Hospital Infections.

Specific to tissues, a resolution was enacted in December 2006 and provides for the technical regulations for the functioning of musculoskeletal tissue banks and banking of human skin [21]. It promotes the principles of quality and risk reduction by the adoption of measures outlining critical controls for tissue banking operations and assists with the development of objective guidelines for inspections. The tissue banking profession is fairly young in Brazil but is rapidly developing and, like other tissue establishments located elsewhere, some room for improvement has been identified as a result of inspections by authorities. These regulations are well written, comprehensive, and offer excellent directions for establishing and maintaining quality systems, including requirements for the physical facility, equipment and materials, and operations of the tissue bank. The annex contains sections that are reminiscent of a technical manual and contain good tissue practices garnered from professional standards and international regulations. For instance, some particular requirements relating to processing and packaging include the following:

- It should be performed in a controlled and classified environment.
- Throughout processing the lyophilizer should be maintained in a controlled environment.
- The final package must ensure the moisture content during product life.
- The sterility of tissues should be guaranteed by a complementary validation sterilization process.

By following these and other prescribed mandates, the professionals have a protocol for developing successful systems to provide safe, quality tissues for transplantation. A major reorganization and some revisions to these regulations are planned during 2012.

**Singapore**

In Singapore, the regulatory control for the processing of human tissue rests with the Licensing and Accreditation Branch of the Ministry of Health. This is implemented through site establishment licenses issued under the Private Hospitals and Medical Clinics Act, and establishments are expected
to duly comply with the formalized “Guidelines For Healthcare Institutions Providing Tissue Banking” [22]. In addition, the Health Sciences Authority (HSA) of Singapore will be implementing the regulatory framework for regulating human cells and tissue-based therapies (CTTs) under their current legislation. In line with the regulatory approach undertaken by regulatory agencies in the EU and USA, the HSA intends to adopt a risk-based approach in regulating CTTs. These are viewed as substantially manipulated, for non-homologous use as well as in combination with a drug/biologic/device, and will be subjected to regulatory control similar to pharmaceuticals and biologic medicinal products. These controls include product registration, clinical trial certification, manufacturer’s licensing or GMP certification, and reporting of serious adverse events. In tandem with the regulatory framework applicable to medicinal products, the applicant for product registration for CTTs will be required to submit dossiers, including the product information for chemistry, manufacturing, and control, while the respective manufacturer will be required to conform to the PIC/S GMP guide [23] and its relevant annexes, or equivalent. As a science-based organization recognizing the vibrant development in the field of CTTs, the HSA is committed to closely collaborate with benchmark regulatory agencies and work with stakeholders to fulfill its national responsibility in promoting and advancing public health and safety effectively.

Highlighted above are some brief insights to the history and current status of several national regulations and established systems for ensuring the quality and safety of human tissues and cells. Other countries, for example Korea, India, and Japan, have active work programs for implementing similar controls to improve the quality and safety of human tissues and cells.

**Regulations – tissue and cell processing**

**European Union**

For compliance with Directive 2004/23/EC, national regulators are required to authorize the tissue and cell preparation processes performed in tissue establishments. Annex II of Directive 2006/86/EC assists this assessment activity by defining the requirements for the evaluation of donor selection criteria and procurement procedures, the relevant protocols for each step of the process, the quality management criteria, and the final quantitative and qualitative criteria for cells and tissues. Consequently, regulatory oversight of these control systems (i.e., the evaluation of critical processing procedures) is becoming the norm and it can be approached in several ways. Validation of critical processing steps may be based on studies performed by the establishment itself or on data from published studies or, for well-established processing procedures, by a retrospective evaluation of the clinical results.
of the tissues or cells supplied by the establishment. Consistency as well as effectiveness of the preparation process in the establishment environment is of particular importance. The quality system should be updated for any significant change to the process and/or after revalidation and qualification programs. A formalized review process is expected to periodically verify that they achieve their intended purpose. As the tissues and cells are native to the human body, with established functionality and proven beneficial purpose – in contrast to medicines or medical devices – the tissue establishment has the duty to demonstrate the preparation process did not render the tissues or cells clinically ineffective or harmful to the recipient.

Annex 1 of Regulation No.1394/2007, for advanced therapy medicinal products, includes a list of processes which are not regarded as “substantial manipulation”. The list includes:

- cutting, grinding, shaping, centrifugation, soaking in antibiotic or antimicrobial solutions, sterilization, irradiation, cell separation, concentration or purification,
- filtering, lyophilization, freezing, cryopreservation and vitrification.

The exclusion list, subject to modification as technology progresses, is more commonly associated with the processing operations in tissue establishments. Where, for example, “separation,” “concentration,” and/or the “purification” of human cells or tissues (e.g., for the isolation of pancreatic islet cells) is undertaken, it is achieved by the application of process technologies and selective media to maintain the natural state, biological characteristics and native functions of the cells or tissues. The aforementioned processes are viewed as suitable candidates for the use of the “preparation process dossier” (see later section). The technical requirements for the validation of manufacturing processes of advanced therapy medicinal products are specified in a guideline on human cell-based medicinal products [24] from the European Medicines Agency (EMA).

The processing of tissues and cells shall take place in a controlled environment with specified air quality and cleanliness to minimize the risk of contamination, with appropriate measures to prevent cross-contamination. The four classifications (i.e., Grade A, B, C, and D) of environmental standards, with their defined physical and microbiological parameters, are specified in the European Guide to Guide for Manufacturing Practice for Medicinal Products [25]. Where there is no subsequent microbial inactivation process, an air quality with particle counts and microbial colony counts equivalent to those of Grade A, with a background environment of at least Grade D, is the normal requirement according to the European Directives. In exceptional circumstances, an alternative standard may be applied where it achieves the quality and safety required for the type of tissue and cells, the process itself, and the proposed human application. Illustrative examples include:
• Where a validated terminal sterilization process has been applied.
• If the expected environment might have a detrimental effect on tissue or cell properties.
• Where the mode/route of application to the recipient has a lower risk of infection transmission than its transplantation.
• Where it is not technically possible to carry out the process in the specified environment.

In all cases the environmental standard applied and its rationale is to be specified within the quality system of the tissue establishment so that it can be scientifically demonstrated to achieve the required standards for quality and safety, taking into account the intended purpose, mode of application, and recipient status.

**Advanced therapy medicinal products**

The transitional phase for the EU healthcare program of Regulation (EC) No.1394/2007 [26] for advanced therapy medicinal products (ATMPs) started in January 2009 and includes three types of medicinal products for human use: gene therapy, somatic cell therapy, and tissue engineering (e.g., expanded chondrocytes for the repair of damaged cartilage or cultured fibroblasts/endothelial cells for treating skin burns or ulcers). The prerequisite for an ATMP is to ascertain its product characteristics and intended purpose meet the definition for a medicinal product and secondly to review the applicability of Regulation (EC) No.1394/2007. Where these are met, Article 2 of Directive 2004/23/EC stating that “Where such manufactured products are covered by other Directives then it shall apply only to donation, procurement and testing” becomes relevant. For example, tissue engineering is focused towards viable “engineered” human or animal cells and is defined as a product that “contains or consists of engineered cells or tissues, and is presented as having properties for, or is used in or administered to human beings with a view to regenerating, repairing or replacing a human tissue”. It subsequently follows the cells or tissues are “engineered” where they fulfill at least one of the following conditions:

• The cells or tissues have been subject to substantial manipulation, so that biological characteristics, physiological functions, or structural properties relevant for the intended regeneration, repair or replacement are achieved.
• The cells or tissues are not intended to be used for the same essential function or functions in the recipient as in the donor.

A process such as “cell culture/expansion” is not included in the list of processes in Annex 1 of the Regulation, implying that it is considered to be substantial manipulation and is therefore “engineering” (e.g., the
expansion of chondrocytes, so the same characteristics, functions or properties are maximally optimized for the reformation of new cartilage). This presumptive approach is consistent with the FDA regulations for “351 products,” where cell culturing/expansion is viewed as “more than minimal manipulation.” In the second case above, separated cells or tissues from a donor site are processed and transplanted into a different site in the recipient where it is intended they perform a different essential function from their natural role in the donor site. For example, isolated stem cells from bone marrow subjected to stimuli (i.e., chemical, enzymatic, or electrical) can in vitro differentiate to a cardiac cell lineage and be transplanted for the repair of myocardial tissue. Conversely, human amniotic membrane functioning as a physical barrier can be processed for ophthalmology applications where the essential function is the same and this is generally not viewed as an ATMP (see Case Study 1.3).

The principles of both regulations, for tissues and cells and ATMPs, overlap with each other. As the regulatory programs for tissue establishments are implemented, there is a parallel initiative for the newer regulations on ATMPs. It is beneficial that the common responsibilities (Box 1.1) at the interface of these work programs are coordinated and mutually compatible, to ensure the timely provision of modern alternative treatments to patients. In early 2012 a European Commission meeting of these health sectors reviewed the common interfaces of both legislative frameworks.

**Box 1.1 Common responsibilities at the interface of tissue establishments (TEs) and ATMP manufacturers**

- Training/expertise of the specialist inspectors
- The verification system for donation, procurement, and testing requirements
- Coordination of planning site inspections by related sectors
- Mutual recognition of site certificates between the regulators
- Compatibility of the roles and status of the “responsible person” (defined in the Tissue and Cell Directives) and the “qualified person” (defined in GMP for medicinal product manufacture)
- Systems for the long-term storage of quality and traceability-related records
- Reporting of serious adverse events and reactions linked to tissues and cells used in ATMPs to the TE sector
- Verifying coding systems for tissues and cells from the donor to/from the end recipient
- Traceability systems between the intermediaries of TEs and ATMP companies
- Management and control of TE requirements in the clinical investigations of ATIMPs
- Defining practical start and end points of quality systems and GMP practices in the two sectors
Management of borderline products in Europe

The Tissue and Cell Directives provide a unified platform (see Figure 1.1) for different healthcare sectors and the application of a regulatory definition (e.g., medicinal product, medical device) and knowledge on the mode of action for the stated intended purpose(s) are significant criteria to determine the regulatory status of a product. Classically, those acting principally by pharmacological, immunologic, or metabolic means are regarded as medicinal products, whereas those acting principally by mechanical means are medical devices. Other related considerations include presentational form, mode of application, any statements or claims, and the instructions for use. This case-by-case approach is well established and applied to assign the regulatory sector, or shared responsibility, to lead the conformity assessment procedure. Conformity with the donation, procurement, and testing requirements becomes the prerequisite for the application of other European regulations, such as the Medicinal Products Directive 2001/83/EC (see [26]) and theoretically the Medical Device Directives (MDD) [27]. The latter presently excludes the use of human materials (Article 1,5 (f) of MDD), and the European Commission was invited by the Council of Europe in early 2011 to consider how to address the regulatory gaps with respect to medical devices utilizing nonviable human tissues and cells. Preliminary work was initiated by the European Commission in 2011 to review the existing regulatory practices to ensure the earlier regulatory gaps (e.g. human bone for remodeling purposes or collagen facial implants), which are not covered by the more recent ATMP regulation, are addressed adequately.

Figure 1.1 Interface between the Tissue and Cell Directives and other healthcare sectors.
United States
In the USA, federal regulatory oversight of medical products that contain human tissues and/or cells is divided between two Public Health Service (PHS) Acts, which provide the legal authority to regulate these products to protect the public health. Section 351 authorizes the Secretary of Health and Human Services (HHS) to approve regulations for requirements to address the safety and effectiveness of drugs, biologics, and medical devices. Section 361 authorizes the HHS to set forth requirements with the objective to avoid the transmission of communicable disease.

“361 products”
Conventional and reproductive HCT/Ps do not require premarket approval by FDA, which means there is no formal review of the manufacturing steps or the provision of a license for the establishment or the product, unlike blood products where interstate distribution and licensing of blood establishments occurs. HCT/Ps qualifying within Section 361 of the PHS Act should meet the following four criteria:

• Minimal manipulation (for structural tissue, processing that does not alter the original relevant characteristics of the tissue relating to the tissue’s utility for reconstruction, repair or replacement; and for cells or nonstructural tissues, processing that does not alter the relevant biological characteristics of the cells or tissues).
• Intended for homologous use (repair, reconstruction, replacement, or supplementation of a recipient’s cells or tissues with an HCT/P that performs the same basic function or functions in the recipient as in the donor).
• Not combined with another article (with a few exceptions).
• No systemic effect and not dependent on metabolic activity of living cells except if for autologous use, use in a first-degree or second-degree blood relative or for reproductive use.

“351 products”
When a product containing or derived from an HCT/P falls outside the four criteria, a request for designation can be made to the FDA’s Tissue Reference Group (TRG). Specific attributes are scrutinized and risk assessed accordingly, such as biologic and structural characteristics (after the processing/treatments), and its action or effect on, or function in, the body. Product efficacy and additional safety concerns must also be assessed. The TRG, with members from CBER and CDRH, classifies the product as a biologic or a medical device. Where a product is combined with or is sourced from an HCT/P, the regulations in 21 CFR 1271 apply, and thereafter the regulations for biologics (i.e., Current Good Manufacturing Practice (CGMP)) and/or medical devices (i.e., Quality System Regulations (QSR)) apply. In such cases, a product determined to be a biologic or medical device falls under the FDA’s
authority as described in Section 351 of the PHS Act, or the Food, Drug, and Cosmetic (FD&C) Act. Examples of combination products already designated by FDA include:

- demineralized bone combined with a handling agent/carrier (i.e., a medical device)
- encapsulated pancreatic islet cells (i.e., a biological product)
- cultured cells on a synthetic membrane or combined with collagen (i.e., a biological product or medical device)
- bone–suture–tendon allograft (i.e., a medical device).

To date, HCT/Ps that have been reviewed and determined to qualify as “351 products”, due to either nonhomologous use or manipulation considered to be more than minimal, include:

- autologous chondrocytes expanded in vitro for repair of cartilage defects
- allogeneic hematopoietic stem/progenitor cells and cord blood
- genetically modified cell therapy products [28].

Thus, “351 products” in the USA are broadly very similar to ATMPs in the EU.

In 2002, the FDA issued the “Guidance for Industry: Validation of Procedures for Processing of Human Tissues Intended for Transplantation” [29]. This followed several serious transmissions of bacterial disease that occurred in the USA due to contaminated allografts, involving processing deficiencies related to validation [30]. Although the guidance lacked the instructional detail on how to specifically perform adequate validations for processing, it did set out the FDA’s expectations. A short list of approaches was finalized to aid a tissue establishment to document the effectiveness of procedures to prevent contamination and cross-contamination. These approaches are methodologies to be used to support a process, which is similar to those described later in the EU Directives:

- Verify full and proper implementation of a previously validated procedure such as may be found in a technical manual of another organization, or
- Conduct literature searches to demonstrate that the procedures implemented are known to be effective in preventing the infectious disease contamination (e.g., Environmental Protection Agency-approved chemical sterilants for laboratory surfaces), or
- Conduct off-line or on-line challenges with indicator organisms, as appropriate, or evaluating the capacity of the manufacturing process to prevent contamination during processing.

The AATB has subsequently formed a task force to develop guidance on all aspects of a “microbiological surveillance program,” which addresses process
validation, the validation of culturing methodologies used for various human tissue types, and how to properly establish a sterility assurance level (SAL) based on processing steps.

Title 21 CFR 1271 at Subpart D includes GMP-like requirements and these are termed Current Good Tissue Practice (CGTP). These requirements aim to control the introduction of contamination during the performance of cell or tissue processing steps and so avoid cross-contamination between donor materials. It contains a description of “core CGTPs” that includes expectations for facilities; environmental control and monitoring; equipment; supplies and reagents; processing and process controls; recovery; processing and process controls; labeling controls; storage; receipt, predistribution shipment, and distribution of an HCT/P; and donor eligibility determinations, donor screening, and donor testing. Additionally, it includes other functions that support this control: Establishment and maintenance of a quality program; personnel; records; tracking; complaint files; procedures; process changes; process validation; and exemptions and alternatives. There are differences between the requirements found in CGTP and those in CGMP for products determined to be “biologics” and similar requirements in QSR for products determined to be “medical devices.” These include all donor eligibility requirements; prevention of the introduction, transmission, or spread of communicable diseases; prohibition on pooling; predistribution shipment; HCT/P availability for distribution only after donor eligibility has been established; and record-keeping for 10 years. These differences make sense since human tissues and cells are a very different source material than metals and plastics (synthetic raw materials).

The role of professional standards in the context of regulations

The establishment of standards, and implementation into tissue banking operations via policies and procedures delivers added value to the quality system and processing controls of an establishment. As national regulations and supporting documents have been produced to govern cell and tissue banking, the regulators have relied on the collective expertise of tissue banking professionals and referenced “industry” standards that guided them in developing regulation. This has been most evident in Europe, Canada, and the USA. Professional standards and guidelines were produced as long ago as the mid-1980s (i.e., AATB) but other tissue and cell banking groups have supported their constituencies by providing guidelines and promoting safe practice. From the mid-1990s, the following cell and tissue banking professional associations have published standards: European Association of Tissue Banks (EATB); British Association for Tissue Banking (BATB); Asociación
Española de Bancos de Tejidos (AEBT), the Spanish Association of Tissue Banks (SATB); European Association of Musculoskeletal Tissues (EAMST); European Eye Bank Association (EEBA); Joint Accreditation Committee of the International Society for Cellular Therapy and the European Group for Blood and Marrow Transplantation (JACIE); and the International Atomic Energy Association (IAEA); as well as one of the earliest standards-setting groups, the Eye Bank Association of America (EBAA). The Council of Europe also began to publish guidelines via “Recommendations and Resolutions” in 1994 that led to the first edition of the “Guide to Safety and Quality Assurance for Transplantation of Organs, Tissues, and Cells” [5], which is presently in its 4th edition (2010) and thoroughly describes quality management systems.

Standards that address processing requirements, much like national and regional regulations, require processes be validated but currently do not offer specific step-by-step instructions on how to do this. However, the systems for enhancing the safety of tissues and cells provided by these associations by the publication of standards and guidelines cannot be stressed enough. On a global scale, the continuous expertise, knowledge, and dedication of cell and tissue banking professionals promoted safety and quality long before it was required by regulations.

**Implementation and practice**

Consensus in regulations is often achieved by compromise, and the transposition into national legislation, which may take some time, precedes the roll-out and implementation by national authorities for systems to verify the conformity of tissue establishments. Since April 2006 the European regulators have learned, initiated, and phased in a series of implementation measures (i.e., for management and monitoring), which will continue long into the future. In the sharing of these experiences and systems some highlights of several surveys, each for the activities performed in the preceding year, are provided in this section.

The first survey in 2007 was responded to by 30 European countries, of which 12 (40%), 8 (26%), and 4 (13%) had completed their transposition of the principal and two technical Directives respectively. The survey indicated there were 2360 different tissue establishments operating within the EU. The second survey in 2008 had a similar purpose, to monitor the state of conformity. This time 31 European countries submitted their data on line: 25 (80%), 20 (65%), and 18 (58%) had transposed the principal and two technical Directives respectively. At least five countries had not included reproductive cells in their transposition process and by mid-2008 four were referred to the European Court of Justice, for lack of progress [31].
3075 establishments noted there were 1920 sites “authorized” by the relevant Competent Authorities. The survey also identified the different type and numbers of tissue establishments as: skin (127), muscle (262), ophthalmic (183), cardiac (103), HSCs – bone marrow/peripheral blood (541), HSCs – cord blood (123), cell banks (55), reproductive centers (1055), other banks (67), and multitissue banks (256). Twelve (38%) of the countries had finalized their annual report of site activities at establishments, most of which were publicly accessible. Twenty-three countries (74%) maintained a regulatory list or electronic database of their establishments, while 15 (48%) presented their core data in a public register, at their agency website or the EUROCET Registry [32]. The third survey performed in early 2009 was more comprehensive than the earlier two, and the data was presented at meetings in 2009. There had been significant progress in the finalization of national transpositions, especially for the two technical Directives. The comprehensive information of all three surveys, regarding the implementation and practices by European countries, is on the Commission’s website [33].

Presently the regulatory resources are primarily focused towards performing inspections of unlicensed tissue establishments or their ongoing reinspection programs. There are some indications that certain types of tissue establishments (e.g., bone, heart valves) are moving towards regionalization of services, to share operational systems and professional skills. With new regulations, and the diversity of different tissues and cells, there are inevitably some technical queries and interpretations which give rise to further debate: for example, the “time of donation,” the terms of “distribution” and “import,” and the “standards of environment” expected for processing of reproductive cells. These and other topics need to achieve a consensus in the near future to ease the transfer of tissues and cells across national and international borders.

As a general comparison, there are more than 2000 tissue establishments registered with the FDA's electronic Human Cell and Tissue Establishment Registration System (eHCTERS). These facilities “manufacture” HCT/Ps if they screen or test donors of tissues or cells, or if they recover, process, package, label, store, or distribute tissue or cell products. As of mid-November 2009, there were 1099 tissue establishments performing “processing” for HCT/Ps and 921 of these were located in the USA. Overall, the type and numbers of tissue establishments registered in the eHCTERS in 2007 that listed the tissue types they manufacture is comparable to the European statistics above. They were: dura mater (24), somatic cells (86), other (157), sclera (159), vascular grafts (174), HSCs – cord blood (185), heart valves (186), cornea (190), pericardium (222), skin (254), cartilage (276), ligaments (281), fascia (284), tendons (311), HSCs – bone marrow/peripheral blood (377), embryos (476), oocytes (508), semen (574), and bone (740). Each establishment may well be responsible for more than one type of HCT/P.
Verification of compliance with regulations

The national regulatory authorities have obligations to perform inspections of the establishments under their jurisdiction. Many have prioritized inspection programs by performing a risk assessment on the type of human tissues or cells, the complexity of site operations, and their compliance with existing measures. A two-tier inspection program is typical for the management of applications; a desk-based assessment of documentation with a self-declaration of conformity, or more traditionally, the on-site inspection of systems and practices at the establishment. Where compliance is demonstrated the center is granted site authorization by a certificate for the site’s activities and types of tissues/cells. The inspector’s role is to verify compliance with the regulations so the regulatory body can decide on the authorization of a site. National regulators should have plans with sufficient resources for subsequent inspections to ensure the continuity of the program. On-site inspection covering all areas of activity should be performed at least every 4 years, whereas a thematic inspection may focus on a particular system or process. Other types of inspection may well be performed, depending on circumstances, such as system-oriented, specific tissues or cells, third parties, or processes. Where the processes are complex or innovative they are likely to be assessed independently of the site inspection. Alternatively, where simplified processing is undertaken, they can be audited during the regular inspection of the establishment. Within the EU, a regulatory guideline was formally established for the conditions of inspections and control measures, together with the training and qualification of officials, in the field of human tissues and cells [34].

The recently finalized EUSTITE project developed an outline format (i.e., the preparation process dossier) for a structured approach to the assessment of critical processes, to be evaluated prior or after the inspection. There are recognizable benefits to the off-site assessment of complex, innovative or unique processes, which should ideally involve input from other knowledgeable technical experts. Where significant modifications are planned to be implemented, an updated process dossier should be submitted for review. Supplementary information on the management of evaluating processes can be found at www.eustite.org.

In the fiscal year 2010, the FDA performed 565 inspections of registered tissue establishments. Approximately one fourth of these inspections resulted in an FDA Form 483, which report observations indicating noncompliance with regulations. Specific findings related to processing include failure to maintain facility in good state of repair; failure to maintain documentation of equipment maintenance, cleaning, sanitization and calibration; lack of environmental controls and monitoring to assure consistency and maintain validated state; and lack of or inadequate validation of microbiological test methods.
Case studies

CASE STUDY 1.1

FDA order to cease manufacturing of stem cells

Background

In September 2009, an FDA notification was published on the internet which identified a registered tissue establishment [35] that did not have adequate protections to prevent the transmission of communicable diseases. The company offered services for collection, serological testing, processing, and long-term storage of stem cells derived from umbilical cord blood. A public order had been issued by the FDA to cease manufacturing and the quarantine of existing stock based on evidence that: 9 of 25 donor record files failed to contain test results for HIV and HCV; written procedures were not established or maintained for several significant on-site systems; and contaminants, including insects, were found in the processing and storage areas. The majority of the 27 other privately owned cord blood banks in the USA were found to be compliant. The notification was seen by an EU tissue and cell regulator. With equal interest, and legal responsibilities, the Members States and the European Commission have the duty to ensure the protection of patients and enquiries were initiated. Their purpose was to verify if any of the stored stem cells had been distributed to other countries and used in clinical applications. This was found not to be the case and the EU regulator informed others their follow-up had verified other non-US patients were not affected. Fortuitously the annual meeting of the Member States’ tissue and cell regulators and the European Commission was held shortly after and national representatives were updated on this matter.

Lessons learned

The communication network between international regulators, operating on an informal basis, was effective and maintains confidence in the safety of these services. Regulators, tissue establishments and clinics need to be aware that activities in countries other than their own could in some circumstances have a significant impact on the management of their patients. Monitoring certain public information sources should be part of active surveillance programs to keep relevant parties up to date.
CASE STUDY 1.2

Donor exclusion criteria are not always the same

Background

The European Commission issued a notification in August 2008 to representatives of Member States on the recall of a bone injectable putty manufactured by a commercial company. Earlier, the Belgian authorities had become aware that some lot numbers of these products – derived from eight donors who had presence or previous history of malignant disease – had been distributed to Belgian tissue banks, and possibly to others. Where relevant, the Member States initiated their own national procedures for informing the clinics (e.g., orthopedic and dental) of the voluntary recall. As a parallel activity, the Dutch Health Inspectorate performed an inspection of the import site (i.e., point of entry) situated in the Netherlands, which found systems and practices to be compliant with the Tissue and Cell Directives. In cooperation with the European Commission, the company issued an updated statement to the Member States on the state of play, with assurances their practices met all the requirements of the FDA and AATB. The presence or history of malignant disease is listed in Directive 2006/17/EC as a criterion for the exclusion of a potential donor. However, the Directive states that donors not meeting the general acceptance criteria may be accepted on the basis of a documented risk assessment authorized by the “responsible person” of the tissue establishment. The Directives provide a common framework of minimum requirements, and stricter requirements in the transposition of the national or local laws can be applied. Consequently at least four European countries, including Belgium, do not accept donors with the presence or previous history of malignant disease, under any circumstances. Similarly, some view that the risk assessment should not be interpreted as a permanent waiver to this exclusion criterion for a donor, even where there exists scientific evidence that extensive processing techniques may remove malignant cells or render them ineffective.

Lessons learned

As the regulations are progressively implemented there are technical perspectives which may highlight subtle differences in the specific requirements of several countries. Knowledge and interpretation of the science and its practical application is needed to reach a consensus to ensure the availability of state of the art treatments. One should recognize in the common framework there may well be other regulatory differences at the national or local level; similar events are likely to be experienced in the future.
**CASE STUDY 1.3**

**Status of amniotic membranes**

Currently there are several products for the repair and regeneration of damaged ocular surface tissue that are derived from the processing and cryopreservation of donated placental tissue. The methodology retains the structural properties of the amniotic membrane for the natural biological actions (antiscarring, antiinflammation, antiangiogenesis) of the in vivo tissue. Since the 1990s, the membrane has been used for reconstruction, where the frozen form consists of a non-viable matrix maintaining and preserving its original function and tissue integrity. It is procured and processed to the standards of Good Tissue Practices (GTP) and GMP, with strict protocols for donor history and screening for infectious markers. In 2001, a Request for Designation to the FDA on a commercial amniotic membrane for ocular surface reconstruction concluded it was within the scope of the tissue banking regulations. Within Europe, a similar review of its regulatory status initially considered the second point of Article 2 of Regulation 1394/2007/EC, which is applicable if cells or tissues from a donor site are processed and transplanted into a different site within a recipient, where the biological activity of cells in the recipient performs a different essential function compared to their natural role, as exhibited in the donor site. However, discussions at an EU level noted, in line with the earlier decision of the FDA, that amniotic membrane, where it functions as a physical barrier, is not regarded as an advanced therapy medicinal product by many of the Competent Authorities for the Tissue and Cell Directives, since the intended purpose at the recipient site represents the same essential function (i.e., a physical barrier). In the future, supplementary clarification on the meaning of “same essential function” is expected to achieve a consensus between the sectors. As presently interpreted this primarily relates to the functional properties of the native tissue of the donor, and yet does not exclude any other functions when transplanted to the recipient.

**Conclusions**

The history of the evolution of the professional standards for tissue and cell banking and the subsequent promulgation of regulations is interesting to explore and understand. During the past few decades, human tissues and cells have been shown to have increasingly beneficial clinical applications for needy patients, and this is no different than the revolution in other therapeutic products, namely pharmaceuticals/drugs, medical devices, and blood or blood products. All of these, for human applications, have the ability to improve or save lives but they can also cause morbidity and mortality if contaminated or defective. The processing steps and methodologies are key components which must be controlled to ensure their quality and safety. The professional experience that allowed agreement on best practices and volun-
tary standards provided a basis for the establishment of mandatory requirements by regulatory authorities in many countries. A global prescription for the quality and safety of human tissues and cells is likely to be the next development phase, which moves a step forward towards facilitating their equitable availability.

**KEY LEARNING POINTS**

- The recent implementation of the European regulations and interaction with other countries worldwide have stimulated further focus on the compatibility of the requirements for quality and safety to achieve the equitable availability of human tissues and cells.
- The standards and guidelines for the management and control of preparation processes (“simplified,” “complex,” or “unique”) are predominantly prepared and updated by professionals in the tissues and cells community and the mechanism for their authorization by regulatory authorities is still predominantly in the developmental phase.
- The parallel implementation of the regulatory work programs for tissue establishments and ATMPs are linked by several common responsibilities and achieving mutually beneficial approaches will contribute to maintaining the standards of quality and safety for the end product.
- The professional skills and expertise within the tissue establishment community and the diverse specialized societies are critical to the effective functioning of regulatory frameworks for improving the standards of care to patients.
- Reciprocal communication systems and their interconnectivity between procurement sites, tissue establishments, clinicians, national regulators, and international parties are presently undergoing a revolution, to improve reporting and surveillance systems, the exchange of skills/experience and the transfer of technical developments.
- The rise in the circulation of human tissues and cells on a global scale reflects, to a certain extent, the increasing interchange ability of national requirements/standards and the shortage of certain types of tissues and cells to meet the clinical need of patients.

**References**


32. EUROCET (European Registry for Organs, Tissues and Cells). www.eurocet.org