THERAPEUTIC AND ECONOMIC IMPLICATIONS OF REGULATING STEM CELL THERAPY AND BLOOD COMPONENTS

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Abstract

The objectives of the study were to 1) Identify European collaborators in the area of stem cell therapy and blood components 2) Establish a network of collaborators to investigate the setting up of a stem cell therapy unit in Malta 3) Address requirements for setting up a stem cell therapy unit and consider economic implications 4) Identify regulatory sciences norms for regulators of stem cell therapy facilities and blood establishments and 5) Compile a glossary of terminology for use in the field of stem cell therapy and blood components.

The methodology included building a structured collaboration involving the identification of key stakeholders and key indicators. Business models for developing a stem cell therapy unit were analysed through the network. Economical implications for set-up, maintenance and treatment were determined and evaluated. A quality manual for regulators inspecting stem cell therapy facilities and blood establishments was developed as part of the regulatory sciences norms. The terms selected for the glossary were identified through collaborations and discussions with experts in the field of stem cell therapy and blood components.

Five site studies were carried out at King's College in London (academic), Holostem Terapie Avanzate in Modena (industry), CTP System in Florence (technical specialists) and the Health Products Regulatory Authority in Ireland (regulator). A public-private partnership was identified as a business model for setting up a stem cell therapy unit to include the skills and assets of the public and the financial acumen of the private investors. Identified facilities for the Malta stem cell project were a research laboratory ($36m^2$) at the University of Malta (UOM) and a Good Manufacturing Practice (GMP) laboratory ($500m^2$) as the manufacturing facility at the Malta Life Sciences Park

(MLSP). Economical implications involved MLSP rent (€100/m^2/year), setting up a Class B processing laboratory (€5,000/m^2), equipment (c€300,000) and personnel (c€600,000/year). Therapy charges range from c€3,000/treatment for hepatocyte transplantation to c€50,000/patient for adipose stem cells for aesthetic medicines. The quality manual for regulators included an 'Aide Memoire' as a practical guidance and a glossary of one hundred and sixty terms was compiled and validated.

Collaborative partnerships served to up-skill knowledge and competency in stem cell therapy and blood components. The results point to the need of a feasibility study which in itself costs between €15,000 and €25,000.

Keywords

Blood components - Collaboration – Public-Private Partnerships - Regulatory science - Stem cell therapy

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List of Abbreviations

AAV	Adeno-associated virus
AIDS	Acquired Immune Deficiency Syndrome
ASC(s)	Adult stem cells
ATMP(s)	Advanced Therapy Medicinal Products
BMR(s)	Batch Manufacturing Record
CAT	Committee for Advanced Therapies
CCMI	Centre for Cell Manufacturing Ireland
CFU-F	Fibroblastic Colony Forming Unit
CHMP	Committee for Medicinal Products for Human Use
CJD	Creutzfeldt Jakob disease
CLI	Critical Limb Ischaemia
CMV	Cytomegalovirus
COMP	Committee for Orphan Medicinal Products
CRF	Clinical Research Facility
CVD	Cardiovascular disease
EEA	European Economic Area
EC	European Commission
EDQM	European Directorate for the Quality of Medicines
EMA	European Medicines Agency
EU	European Union
FACS	Fluorescence activated cell sorting
FBC	Full blood count
FMEA	Failure Mode and Effects Analysis
GBTE	Galway Blood and Tissue Establishment
GM-CSF	Granulocyte macrophage colony-stimulating factor
GMP	Good Manufacturing Practice
GvHD	Graft versus host disease
Hb	Haemoglobin
HBsAg	Hepatitis B surface antigen
HBV	Hepatitis B virus
HCV	Hepatitis C virus

HEPA	High Efficiency Particulate Air
HIV	Human immunodeficiency virus
HLA	Human leukocyte antigen
HLQ	Health lifestyle questionnaire
HMA	Heads of Medicines Agencies
hMSC	human Mesenchymal Stem Cells
HPRA	Health Products Regulatory Authority
HSV-TK	Herpes simplex I virus thymidine kinase
HTA	Human Tissue Authority
HTLV	Human T-lymphotropic virus
HVAC	Heat and Ventilation Air Condition
IBTS	Irish Blood Transfusion Service
iPS	Induced pluripotent stem cells
IMP(s)	Investigational Medicinal Product
IMPD	Investigational Medicinal Product Dossier
ISO	International Standards Organisation
LPL	Lipoprotein lipase
MDH	Mater Dei Hospital
ME	Malta Enterprise
MHRA	Medicines and Healthcare Products Regulatory Agency
MLN	Malta Laboratories Network
MLSP	Malta Life Sciences Park
MMA	Malta Medicines Authority
MOU	Memorandum of Understanding
MSCs	Mesenchymal stem cells
NAT	Nucleic Acid Testing
NBTS	National Blood Transfusion Service
NHIRL	National Histocompatibility and Immunogenetics Reference Laboratory
PDL	Population doubling levels
PI	Principal Investigator
PIC/S	Pharmaceutical Inspection Co-operation Scheme
QA	Quality Assurance
QC	Quality control

QMS	Quality Management System
QP	Qualified Person
RCC	Red cell concentrate
ROI	Return of Investment
RP	Responsible Person
SAE	Serious Adverse Event
SAR	Serious Adverse Reaction
SAWP	Scientific Advice Working Party
SME	Small and medium sized enterprise
SOP	Standard Operating Procedure
SPH	Superintendent of Public Health
TSE	Transmissible spongiform encephalopathy
UCB	Umbilical cord blood
UK	United Kingdom
UOM	University of Malta
vCJD	Variant Creutzfeldt-Jakob disease
WHO	World Health Organisation

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CHAPTER 1

INTRODUCTION

Malta is considering a scenario for the regulation of stem cell therapy and blood components, analogous to the rules applied for medicinal products on the basis of quality, safety and efficacy. This chapter gives an overview of the state-of-the-art advancements in the field of stem cell therapy and blood components. Stem cells as medicinal products are classified as advanced therapy medicinal products (ATMPs).

1.1 Development of advanced therapy medicinal products

Recent years have witnessed rapid development of several cutting edge technologies, including stem cell therapy, gene therapy and tissue engineering, which have the potential to transform the way conditions are treated (Zhang et al, 2015). The Bioindustry Association in the United Kingdom (UK) highlighted that 80% of healthcare costs are spent on treating the late stages of illness, such as heart failure, which in the future could possibly be managed better using stem cell therapies.[1] Stem cell therapies have been developed for different therapeutic indications that address areas of unmet medical need, including cardiovascular diseases, diabetes mellitus, eye disorders, Crohn's disease, graft versus host disease (GvHD), bone fractures, cartilage defects and neurological conditions, such as Parkinson's disease and multiple sclerosis (Figure 1.1).

[1] Department for Business Innovation and Skills. Taking Stock of Regenerative Medicine in the United Kingdom[Online]. London: Department of Health; 2011 [cited 2017 May 28]. Available from: URL: www.bis.gov.uk / assets / biscore / innovation/ docs / t / 11 - 1056 - taking - stock - of - regenerative - medicine

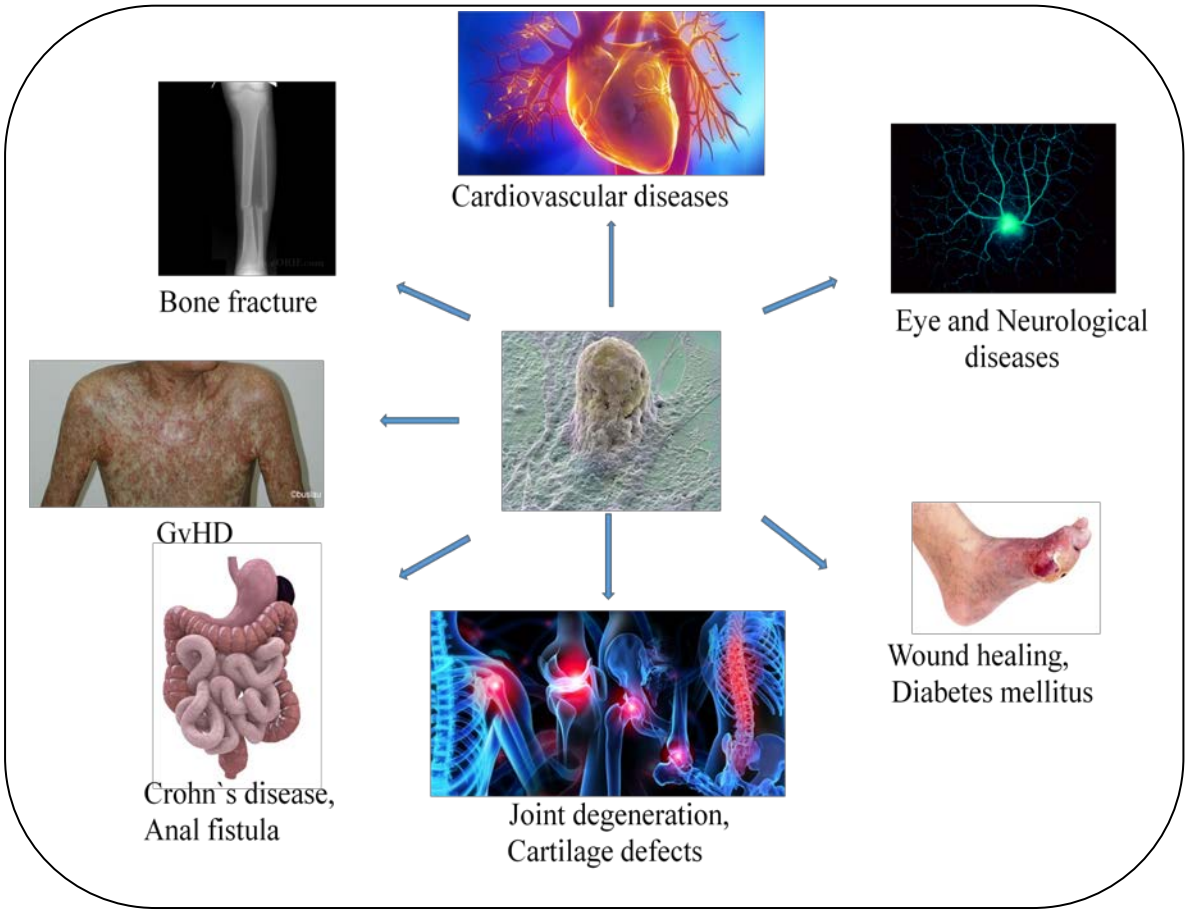


Figure 1.1 Stem cell therapies developed for different therapeutic indications

Reproduced from a presentation by the Chair of the Committee for Advanced Therapies (CAT), Martina Schüßler-Lenz, during the 2017 Heads of Medicines Agency meeting, Malta

Research and development of ATMPs is almost exclusively carried out by academia, hospitals and small and medium-sized enterprises (SMEs), with 80% of ATMPs currently under development are carried out by academia (Hartmann-Fritsch et al, 2016). A study by Hanna et al (2016) identified 939 ATMP clinical trials between 1999 and 2015 (Figure 1.2).

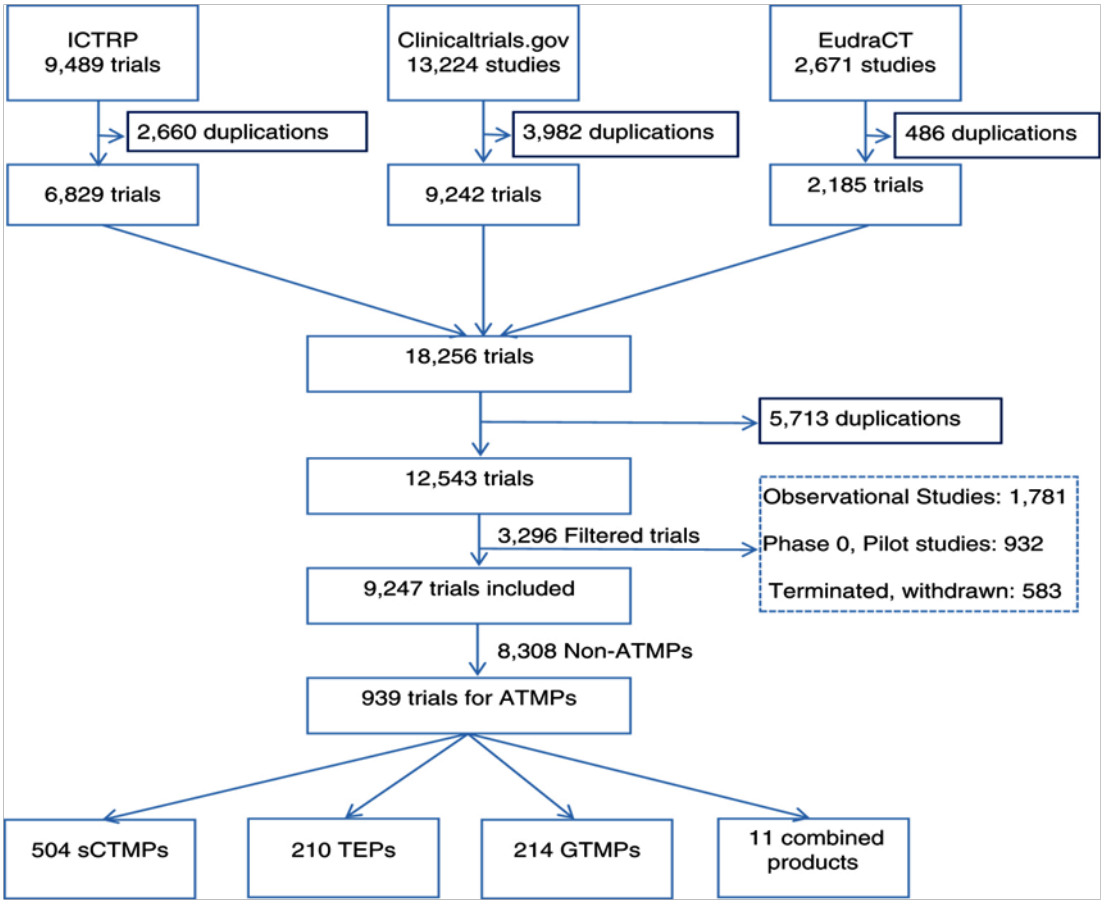


Figure 1.2 Flow chart of clinical trials identification

Abbreviations ATMPs: advanced therapy medicinal products; sCTMPs: somatic cell therapy products; TEPs: tissue-engineered products; GTMPs: gene therapy medicinal products; ICTRP: International Clinical Trials Registry Platform

Reproduced from Hanna E, Rémuzat C, Auquier P, Toumi M. Advanced therapy medicinal products: Current and future perspectives. Journal of Market Access and Health Policy 2016; 4(1): 31036.

Results from a research study carried out by Hanna et al (2016) shows that oncology is the dominant therapeutic area for ATMP trials (24.8%), while cardiovascular disease (CVD) is the second largest therapeutic area where the majority of ATMP clinical trials are in the early stages of development (Table 1.1).

Table 1.1 Classification of advanced therapy medicinal products trials by disease area and phase of development

Therapeutic area	Phase 1 and 1/II	Phase II and II/III	Phase III	N/A	Total
Cancer	146 (24.2%)	69 (26.4%)	18 (27.8%)		233 (24.8%)
Cardiovascular disease	104 (17.2%)	67 (25.7%)	11 (16.7%)		182 (19.4%)
Immune system /inflammation	68 (11.3%)	29 (11.1%)	9 (13.9%)	2 (22.2%)	108 (11.5%)
Musculoskeletal system	59 (9.8%)	25 (9.6%)	9 (13.9%)	6 (66.7%)	99 (10.5%)
Neurology	61 (10.1%)	23 (8.8%)	1 (1.5%)		85 (9.1%)
Gastrointestinal disease and diabetes mellitus	25 (4.1%)	15 (5.7%)	8 (12.3%)	1 (11.1%)	49 (5.2%)
Ophthalmology	34 (5.6%)	7 (2.7%)	3 (4.6%)		44 (4.7%)
Pulmonology	25 (4.1%)	6 (2.3%)	1 (1.5%)		32 (3.4%)
Dermatology	19 (3.1%)	7 (2.7%)	3 (4.6%)		29 (3.1%)
Haematology	16 (2.6%)	4 (1.5%)	0		20 (2.1%)
Others	47 (7.8%)	9 (3.4%)	2 (3.1%)		58 (6.2%)
Total	604 (64.3%)	261 (27.9%)	65 (6.9%)	9 (0.9%)	939 (100%)

Reproduced from Hanna E, Rémuzat C, Auquier P, Toumi M. Advanced therapy medicinal products: Current and future perspectives. Journal of Market Access and Health Policy 2016; 4(1): 31036.

1.2 Regulatory framework for the development of advanced therapy medicinal products

All medicinal products, whether of chemical or biological origin, are scientifically evaluated for quality, safety and efficacy before being granted a marketing authorisation. European Commission (EC) Regulation 1394/2007[2] provides the overall legal framework for ATMPs which mandates that ATMPs are subject to a centralised procedure. Marketing authorisation applications for ATMPs are subject to a 210-day assessment procedure by the European Medicines Agency (EMA) to ensure a high level of scientific evaluation and to facilitate market access (Van Wilder, 2012). The complexity of ATMPs led to establishment of the EMA Committee for Advanced Therapies (CAT) to address challenges associated with ATMPs and to support the marketing authorisation procedure through ATMP classification, certification and provision of regulatory scientific advice (Figure 1.3).[2,3]

[2] European Commission. Regulation (EC) 1394/2007 of the European Parliament and of the Council of 13 November 2007 on advanced therapy medicinal products and amending Directive 2001/83/EC and Regulation (EC) 726/2004 [Online]. Official Journal of the European Union 2007; L324:121-137 [cited 2017 May 28]. Available from: URL: http://eur-lex.europa.eu/legal content/EN/TXT/PDF/?uri=CELEX:32007R1394&from=EN

[3] European Commission. Commission Regulation (EC) No 668/2009 of 24 July 2009 implementing Regulation (EC) No 1394/2007 of the European Parliament and of the Council with regard to the evaluation and certification of quality and non-clinical data relating to advanced therapy medicinal products developed by micro, small and medium-sized enterprises [Online]. Official Journal of the European Union 2009; 194:7–10. [cited 2017 May 28]. Available from: URL: http://eur-lex.europa.eu/LexUriServ/LexUriServ.do?uri=OJ:L:2009:194:0007:0010:EN:PDF

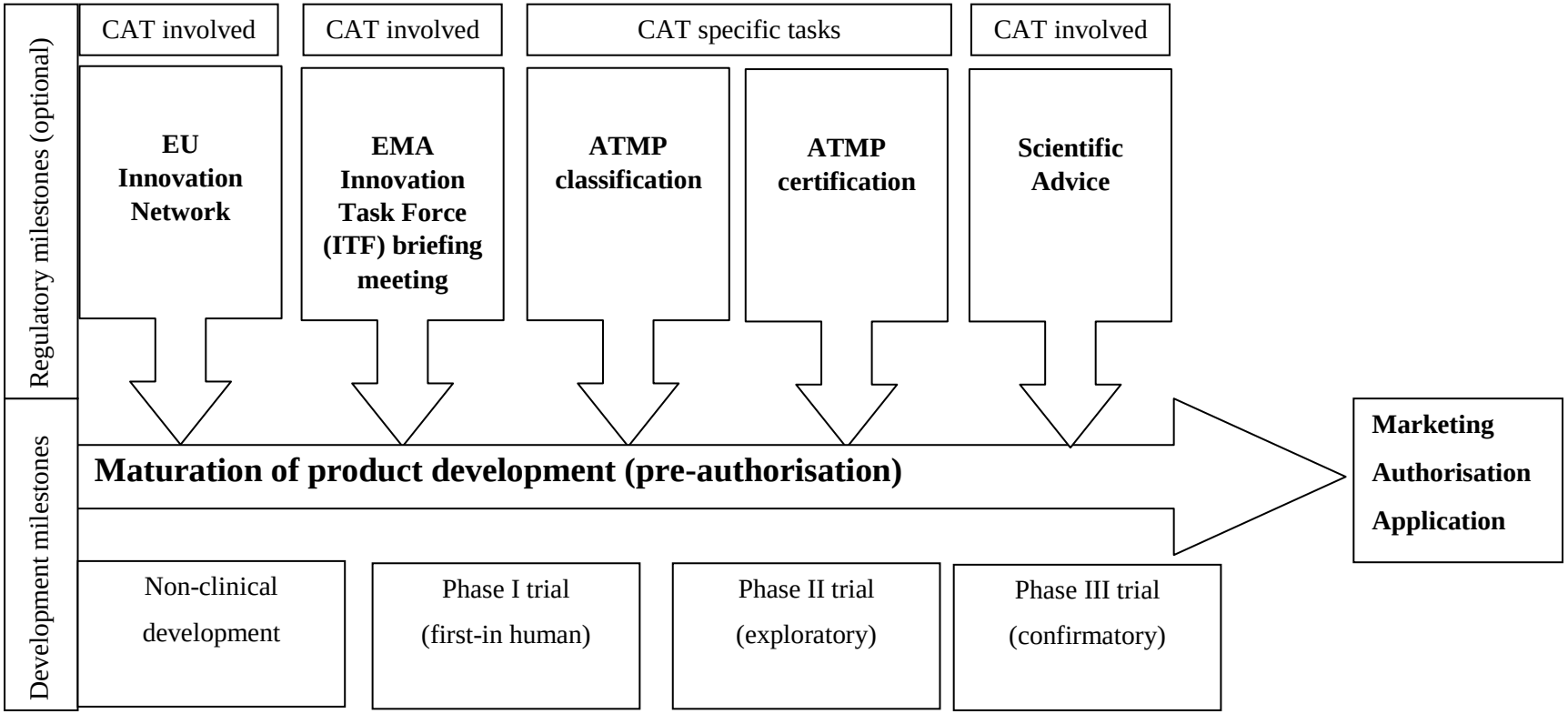


Figure 1.3 Regulatory pathways for advanced therapy medicinal products in Europe

Adapted from Maciulaitis R, D'apote L, Buchanan A, Pioppo L, Schneider CK. Clinical development of advanced therapy medicinal products in Europe: Evidence that regulators must be proactive. Molecular Therapy 2012; 20(3):479-82.

Clinical trials using ATMPs as investigational medicinal products (IMPs) are currently regulated under the Clinical Trials Directive 2001/20/EC.[4] The same approach as for other medicinal products applies, where both the opinion of the Ethics Committee and approval of the national competent authority are required prior to commencing a new clinical trial. EU Regulation 536/2014 on clinical trials, which comes into effect in 2018 and repeals Directive 2001/20/EC, includes a streamlined application process through the EU portal, a harmonised review, increased transparency with regard to clinical trial data and simplified rules for safety reporting[5] (Hurley et al, 2017).

"Quality plays a major role in the safety and efficacy profile of ATMPs".[6] From the beginning of the process, cells and tissues for clinical use must be in accordance with Directive 2004/23/EC[7], commonly referred to as the 'European Tissues and Cells Directive'. This directive covers the quality and safety standards for the entire chain of activities, from donation, to procurement, testing, processing, preservation, storage and

[4] European Commission. Directive 2001/20/EC of the European Parliament and of the Council of 4 April 2001 on the approximation of the laws, regulations and administrative provisions of the Member States relating to the implementation of good clinical practice in the conduct of clinical trials on medicinal products for human use [Online]. Official Journal of the European Union. 2001; 21: 34–44 [cited 2017 May 28]. Available from: URL: http://eur-lex.europa.eu/LexUriServ/LexUriServ.do?uri=OJ:L:2001:121:0034:0044:en:PDF

[5] European Commission. Regulation (EU) No 536/2014 of the European Parliament and of the Council of 16 April 2014 on clinical trials on medicinal products for human use, and repealing Directive 2001/20/EC [Online]. Official Journal of the European Union 2014; 158:1–75 [cited 2017 May 28]. Available from: URL: http://ec.europa.eu/health//sites/health/files/files/eudralex/vol-1/reg_2014_536/reg_2014_536_en.pdf

[6] European Commission. Consultation Document – Draft Guidelines on Good Manufacturing Practice for Advanced Therapy Medicinal Products[Online]. Brussels: EC; 2016 [cited 2017 May 28]. Available from: URL: http://ec.europa.eu/health//sites/health/files/files/advtherapies/2016_06_pc/2016_06_draft_guideline.pdf

[7] European Commission. Directive 2004/23/EC of the European Parliament and of the Council on setting standards of quality and safety for the donation, procurement, testing, processing, preservation, storage and distribution of human tissues and cells [Online]. Official Journal of the European Union 2004; L102:48-58 [cited 2017 May 28]. Available from: URL: http://eur-lex.europa.eu/Lex-UriServ/LexUriServ.do?uri=OJ:L:2004:102:0048:0058:en:PDF

distribution of human tissues and cells, as well as technical implementing directives (Table 1.2).

Table 1.2 Technical implementing tissue and cells directives

Directive	Purpose
2006/17/EC[8]	Technical requirements for donation, procurement and testing of human tissues and cells
2006/86/EC[9]	Standards for traceability, notification of serious adverse reactions and events, and requirements for coding processing, preservation, storage and distribution of human tissues and cells
2015/565[10]	Technical requirements for the coding of human tissues and cells
2015/566[11]	Procedures for verifying the equivalent standards of quality and safety of imported tissues and cells

Technical implementing Directive 2006/86/EC[9] addresses the specific requirements for traceability of the individual product from its starting raw material through to final use. The Single European coding system was developed to facilitate traceability and information on the main characteristics and properties of tissues and cells. This system

[8] European Commission. Commission Directive 2006/17/EC of 8 February 2006 implementing Directive 2004/23/EC of the European Parliament and of the Council as regards certain technical requirements for the donation, procurement and testing of human tissues and cells [Online]. Official Journal of the European Union 2006; 38:40–52 [cited 2017 May 28]. Available from: URL: http://eur-lex.europa.eu/LexUriServ/LexUriServ.do?uri=OJ:L:2006:038:0040:0052:EN:PDF

[9] European Commission. Commission Directive 2006/86/EC of 24 October 2006 implementing Directive 2004/23/EC of the European Parliament and of the Council as regards traceability requirements, notification of serious adverse reactions and events and certain technical requirements for the coding, processing, preservation, storage and distribution of human tissues and cells [Online]. Official Journal of the European Union 2006; 294:32–50 [cited 2017 May 28]. Available from: URL: http://eur-lex.europa.eu/LexUriServ/LexUriServ.do?uri=OJ:L:2006:294:0032:0050:EN:PDF

[10] European Commission. Commission Directive (EU) 2015/565 of 8 April 2015 amending Directive 2006/86/EC as regards certain technical requirements for the coding of human tissues and cells [Online]. Official Journal of the European Union 2015; 93:43-55 [cited 2017 May 28]. Available from: URL: http://eur-lex.europa.eu/legal-content/EN/TXT/PDF/?uri=CELEX:32015L0565&from=EN

[11] European Commission. Commission Directive (EU) 2015/566 of 8 April 2015 implementing Directive 2004/23/EC as regards the procedures for verifying the equivalent standards of quality and safety of imported tissues and cells [Online]. Official Journal of the European Union 2015; 93:56-68 [cited 2017 May 28]. Available from: URL: http://eur-lex.europa.eu/legal-content/EN/TXT/PDF/?uri=CELEX:32015L0566&from=EN

is supported by an online database, the 'EU Coding Platform', and enables identification of the donor, tissue establishment, recipient and manufacturing facility processing the cells, as well as identification of any products and materials coming into contact with the tissues or cells.[12]

1.3 Advanced therapy medicinal products granted a marketing authorisation

Notwithstanding the success of ATMPs in *in vitro* models and animal studies, few have reached more advanced regulatory milestones, such as ATMP certification from CAT or a regulatory scientific advice from the EMA (Maciulaitis et al, 2012). Between 2009 and 2016, eight ATMPs were granted a marketing authorisation (Table 1.3), out of which only five are currently authorised, namely Glybera 3×10^{12} genome copies/ml solution for injection, Holoclar 79,000 - 316,000 cells/cm^2 living tissue equivalent, Imlygic 10^6 and10^8 plaque forming units (PFU)/mL solution for injection, Strimvelis 1-10 million cells/ml dispersion for infusion and Zalmoxis 5-20 x 10^6 cells/mL dispersion for infusion.[13]

[12] European Commission. Commission decision of 3.7.2015 establishing a model for agreements between the Commission and relevant organisations on the provision of product codes for use in the Single European Code [Online]. Brussels: EC; 2015 [cited 2017 May 28]. Available from: URL: http://ec.europa.eu/health//sites/health/files/blood_tissues_organs/docs/c_2015_4460_decision_en.pdf

[13] European Medicines Agency. EMA/388480/2016. Regulation of advanced therapy medicines[Online]. London: EMA; 2016 [cited 2017 May 28]. Available from: URL: http://www.ema.europa.eu/docs/en_GB/document_library/Press_release/2016/06/WC500208078.pdf

Table 1.3 Advanced therapy medicinal products granted a marketing authorisation between 2009 and 2016

Name of ATMP	Authorisation date	ATMP classification	Authorisation state	Description of ATMP and Indication
ChondroCelect[14]	2009	Tissue Engineered Product	Withdrawn in 2016 for commercial reasons	*Ex-vivo* expanded autologous chondrocytes for cartilage repair of single symptomatic defects of the femoral condyle of the knee (International Cartilage Repair Society grade III or IV) in adults
Glybera[15]	2012	Gene Therapy Medicinal Product	Authorised Orphan designation Exceptional authorisation	Adeno-associated virus serotype 1 (AAV1) vector transferring lipoprotein lipase (LPL) gene for adult patients diagnosed with familial LPL deficiency and suffering from severe or multiple pancreatitis attacks despite dietary fat restrictions
MACI[16]	2013	Tissue engineered product	Suspended in 2014 Manufacturing site in Denmark closed	Autologous cultured chondrocytes indicated to repair cartilage defects of the bones of the knee joint

[14] European Medicines Agency. EMA/421074/2014. ChondroCelect: EPAR summary for the public [Online]. London: EMA; 2014 [cited 2017 May 28]. Available from: URL: http://www.ema.europa.eu/docs/en_GB/document_library/EPAR_Summary_for_the_public/human/000878/WC500026033.pdf

[15] European Medicines Agency. EMA/470328/2012. Positive opinion on the marketing authorisation of Glybera (alipogene tiparvovec) [Online]. London: EMA; 2012 [cited 2017 May 28]. Available from: URL: http://www.ema.europa.eu/docs/en_GB/document_library/Medicine_QA/2012/07/WC500130153.pdf

[16] European Medicines Agency. EMA/733307/2014. Closure of EU manufacturing site for MACI [Online]. London: EMA; 2014 [cited 2017 May 28]. Available from: URL: http://www.ema.europa.eu/docs/en_GB/document_library/Referrals_document/Maci_20/WC500173680.pdf

Name of ATMP	Authorisation date	ATMP classification	Authorisation state	Description of ATMP and Indication
Provenge[17]	2013	Cell Therapy Medicinal Product	Withdrawn in 2015 for commercial reasons	Autologous peripheral-blood mononuclear cells activated with prostatic acid phosphatase granulocyte-macrophage colony-stimulating factor. Indicated for metastatic castrate-resistant prostate cancer
Holoclar[18]	2015	Tissue Engineered Product	Conditional authorisation Orphan designation	*Ex-vivo* expanded autologous corneal epithelial cells containing stem cells for adult patients with moderate to severe limbal stem-cell deficiency caused by burns
Imlygic[19]	2015	Gene therapy medicinal product	Authorised	Oncolytic attenuated Herpes Simplex Virus-1 transferring human granulocyte macrophage colony-stimulating factor (GM-CSF) gene for treatment of adults with unresectable melanoma regionally or distantly metastatic with no bone, brain, lung or other visceral disease

[17] European Medicines Agency. EMA/303072/2015. Provenge: Witdrawal of the marketing authorisation in the European Union [Online]. London: EMA; 2015 [cited 2017 May 28]. Available from: URL: http://www.ema.europa.eu/docs/en_GB/document_library/Public_statement/2015/05/WC500186950.pdf

[18] European Medicines Agency. EMA/786996/2014. First stem-cell therapy recommended for approval in EU [Online]. London: EMA; 2014 [cited 2017 May 28]. Available from: URL: http://www.ema.europa.eu/docs/en_GB/document_library/Press_release/2014/12/WC500179333.pdf

[19] European Medicines Agency. First oncolytic immunotherapy recommended for approval. [Online]. London: EMA; 2015 [cited 2017 May 28]. Available from: URL: http://www.ema.europa.eu/ema/index.jsp?curl=pages/news_and_events/news/2015/10/news_detail_002421.jsp&mid=WC0b01ac058004d5c1

Name of ATMP	Authorisation date	ATMP classification	Authorisation state	Description of ATMP and Indication
Strimvelis[20]	2016	Gene therapy medicinal products	Authorised Orphan designation	Autologous CD34+ cells transduced with retroviral vector encoding the human adenosine deaminase cDNA sequence. Indication for patients with severe combined immunodeficiency disorder due to adenosine deaminase deficiency, whom no suitable HLA-matched related stem cell donor is available. Administration only in specialised transplant centre
Zalmoxis[21]	2016	Cell therapy medicinal product	Authorised	Allogenic T cells genetically modified with a retroviral vector encoding for a truncated form of the human low affinity nerve growth factor receptor (ΔLNGFR) and the herpes simplex I virus thymidine kinase (HSV-TK Mut2) for adjunctive treatment in haploidentical haematopoietic stem cell transplant of adult patients with high-risk of haematological malignancies.

[20] European Medicines Agency. EMA/CHMP/230486/2016 New gene therapy for the treatment of children with ultra-rare immune disorder recommended for approval [Online]. London: EMA; 2016[cited 2017 May 28]. Available from: URL: http://www.ema.europa.eu/docs/en_GB/document_library/Press_release/2016/04/WC500204145.pdf

[21] European Medicines Agency. EMA/CHMP/429337/2016 New cell-based therapy to support stem cell transplantation in patients with high-risk blood cancer [Online]. London: EMA; 2016 [cited 2017 May 28]. Available from: URL: http://www.ema.europa.eu/docs/en_GB/document_library/Press_release/2016/06/WC500209256.pdf

1.4 Applying a risk-based approach in the development of advanced therapy medicinal products

ATMPs must comply with Good Manufacturing Practice (GMP) for medicinal products under Directive 2003/94/EC.[22] In view that ATMPs are mainly developed by hospitals, academia or SMEs rather than by pharmaceutical companies, compliance with GMP standards is a major challenge for ATMP manufacturers that may lead to "a translational gap" of product development. The challenge to establish GMP-compliant manufacturing processes could be due to limited financial resources, regulatory expertise or limited knowledge about the investigational product, such as potency or how the manufacturing process will evolve during the development phases of the product (Maciulaitis et al, 2012; Milmo, 2015). These manufacturing issues could be a reason for having only eight ATMPs with a marketing authorisation in the EU.

The risks involved in the manufacturing process of ATMPs differ according to the type of product, characteristics of the starting materials and complexity of the manipulation steps. The finished product may entail some degree of variability, where the ATMP manufacturer may need to implement flexibility in the application of GMP requirements depending on the specific characteristics of the manufacturing process and product. The EC developed a draft GMP guideline for ATMPs[6] addressing the need of a risk-based approach in the development of ATMPs while ensuring the quality, safety and efficacy

[6] European Commission. Consultation Document – Draft Guidelines on Good Manufacturing Practice for Advanced Therapy Medicinal Products[Online]. Brussels: EC; 2016 [cited 2017 May 28]. Available from: URL: http://ec.europa.eu/health//sites/health/files/files/advtherapies/2016_06_pc/2016_06_draft_guideline.pdf

[22] European Commission. Commission Directive 2003/94/EC of 8 October 2003 laying down the principles and guidelines of good manufacturing practice in respect of medicinal products for human use and investigational medicinal products for human use [Online]. Official Journal of the European Union 2003; L262:22-26 [cited 2017 May 28]. Available from: URL: http://ec.europa.eu/health/sites/health/files/files/eudralex/vol-1/dir_2003_94/dir_2003_94_en.pdf

of the product. The guideline was drafted following consultations with the EMA and EU national competent authorities which was followed by a targeted stakeholder consultation launched on 28 June 2016.[23]

The proposed guideline for ATMPs was supported by the majority of respondents, particularly academia and SMEs, however further amendments were suggested including 1) Reducing the stringent requirements for dedicated production areas where additional clarification was requested when biological safety cabinets and closed systems are used, 2) Adaptation of the aseptic process validation and requirements for on-going stability program according to the specific characteristics of ATMPs, 3) Additional flexibilities on the role of the Qualified Person (QP) considering that for many ATMPs each unit is considered as a separate batch. A number of respondents submitted their feedback to the EC that the relationship between the draft guideline and the GMP guidelines in Volume 4 was unclear.[24] There were different suggestions that the proposed guideline should be either a stand-alone document or annex to the GMP guidelines in Eudralex Volume 4.[25]

[23] Taylor NP. European Regulatory Roundup: EC Opens Draft Consultation on Advanced Therapy GMP Guidelines [Online]. Regulatory Affairs Professional Society, 2016 [cited 2017 May 28]. Available from: URL: http://www.raps.org/Regulatory-Focus/News/2016/06/30/25245/European-Regulatory-Roundup-EC-Opens-Draft-Consultation-on-Advanced-Therapy-GMP-Guidelines-30-June-2016/

[24] European Commission. Summary of the responses to the targeted stakeholder consultation on the development of Good Manufacturing Practice for Advanced Therapy Medicinal Products pursuant to Article 5 of Regulation 1394/2007 [Online]. Brussels: EC; 2016 [cited 2017 May 28]. Available from: URL: https://ec.europa.eu/health/sites/health/files/files/advtherapies/2016_06_pc/responses/2016_11_09_outcome_public_consulation.pdf

[25] European Commission. EudraLex Volume 4 EU Guidelines to Good Manufacturing Practice Medicinal Products for Human and Veterinary Use: Annex 1 Manufacture of Sterile Medicinal Products [Online]. Brussels; 2008 [cited 2017 May 28]. Available from: URL: http://ec.europa.eu/health//sites/health/files/files/eudralex/vol-4/2008_11_25_gmp-an1_en.pdf

The Pharmaceutical Inspection Co-Operation Scheme (PIC/S) committee and the GMP/GDP Inspectorate Working Group submitted their concerns to the EC that the proposed GMP guideline for ATMPs is establishing lower standards and risking de-harmonisation that could lead to serious consequences to the current global regulatory framework. The PIC/s committee highlighted risks relating to patient safety in the proposed GMP guideline for ATMPs including insufficient requirements to assure aseptic production, process validation, control of starting materials, address product release process, integrate Quality Risk Management concepts in the production of ATMP and to adopt a well-organised pharmaceutical quality system.[26] The critical points highlighted by the PIC/S committee are challenges on the manufacturing aspect of ATMPs which need to be addressed to ensure that patients receive high quality medicinal products.

1.5 Challenges in the development of Advanced Therapy Medicinal Products

Given the unique biological nature of ATMPs and uncertainties in their clinical use, ATMPs present new challenges to both research and regulatory scientists that may not have been foreseen within the existing regulatory framework (Yuan and Wang, 2014). Product characterisation is a critical quality control requirement that needs to be designed to address concerns regarding risks of cross-cell contamination, transmitting infectious or genetic diseases, and ensuring that the ATMP administered to the patient is the one that has been approved. The incomplete understanding of stem cell science leads to lack of established standard operating procedures relating to product

[26] Pharmaceutical Inspection Co-Operation Scheme. PS/L 11/2017 - Letter to the European Commission on Guidelines on Good Manufacturing Practice for Advanced Therapy Medicinal Products [Online]. Geneva: PIC/S; 2017 [cited 2017 May 28]. Available from: URL: https://picscheme.org/layout/document.php?id=1043

characterisation during ATMP development that affects the reliability of clinical trial results[6] (Yuan and Wang, 2014). Establishing the link between the functional capability of the product and the intended clinical use may be a challenge where it may take several years to measure long-term clinical outcomes. Conducting lengthy clinical trials is a financial burden especially on small companies and may be problematic to maintain patient follow-ups. EMA has tackled this challenge with the possibility of accepting part of the clinical efficacy results as a post-marketing obligation as long as a positive-risk balance has been demonstrated at the time of the marketing authorisation application (Schneider et al, 2010).

Non-clinical studies in animal species can be powerful tools to further characterise the product and for toxicity assessment, however, finding a relevant animal model may be challenging. It is difficult to find an animal model for testing the product that resembles humans in terms of cellular and molecular interactions. Schneider et al (2010) gave an example where adult human stem cells for tissue repair, grow exponentially and can differentiate into various phenotypes, for example mesenchymal stem cells to an osteogenic or mycogenic phenotype, which is driven by the respective micro-environment. Cells react in a species-specific manner, where nothing may happen when injecting the cells into animals due to lack of interaction with animal tissue or results in artificial immunotoxic effects as the immune system of the animal attacks the foreign human cells.

[6] European Commission. Consultation Document – Draft Guidelines on Good Manufacturing Practice for Advanced Therapy Medicinal Products[Online]. Brussels: EC; 2016 [cited 2017 May 28]. Available from: URL: http://ec.europa.eu/health//sites/health/files/files/advtherapies/2016_06_pc/2016_06_draft_guideline.pdf

Quick testing methods for assessing safety and efficacy of ATMPs are extremely needed where products, such as autologous stem cell therapy, have very short shelf life, limiting the time of controls before release (Pellegrini et al, 2016). Sterility, endotoxin and mycoplasma testing are classical examples of when fast and efficient testing methods are required during the manufacturing and release process of ATMPs. While the ATMP may be released on the basis of rapid testing methods, post-release confirmation would still be expected and a plan in case of positive results should be in place (Wilson and Cockroft, 2013).

Lack of effective criteria for product release is a challenge in the development of ATMPs, where unlike conventional medicines that are manufactured to a high degree of homogeneity, the manufacture of ATMPs is characterised by biological variability which at times could be highly significant, such as in the case of allogenic stem cell therapies. The biological variability of ATMPs presents other challenges for developing regulatory tools, including establishment of standards. Development of standards, including standard testing methods and specifications, relies on risk assessment of the manufacturing process and clinical procedures. Common reference standards are required to assess minimal acceptable changes during cell culture and to facilitate comparisons among different studies. Standardisation efforts should include development of uniform standards relating to donor eligibility, consent, procurement, manufacturing, storage, delivery and selection of patients to receive the ATMP (Yuan and Wang, 2014). Hurley et al (2017) discuss another challenge concerning the reimbursement of ATMPs, where the current health eco-systems are not appropriately set-up to fund ATMPs, limiting access to few or no patients. Payers still need to

implement cost/benefit models for ATMPs which are often indicated for orphan use with uncertainties on clinical outcome measures.[27]

The examples highlighted in this section show that these challenges apply to both the developers and regulators of ATMPs. The interaction between regulators, developers and academic groups to exchange scientific views and ensure compliance with the regulatory framework for ATMPs is essential to ensure the quality, safety and effectiveness of the medicinal product. Unregulated advanced therapies have caused clinical problems, where in March 2017 it was reported that three women with non-neovascular age-related macular degeneration experienced profound visual loss after receiving intravitreal injection of autologous adipose tissue-derived stem cell therapy at a clinic in Florida (Kuriyan et al, 2017). Similar cases raise concerns over the quality, safety and effectiveness of these products. It is generally agreed that before applying advanced therapies, such as stem-cell based medicinal products, comprehensive studies to understand stem cell behaviour and the mechanism of interaction upon transplantation are needed (Yuan and Wang, 2013).

1.6 Stem cell-based medicinal products

Potential use of stem cells was first revealed in the late 1950s, when Dr. Donnall Thomas from the Fred Hutchinson Cancer Research Centre in the United States of America performed the first ever haematopoietic stem cell transplant, also referred to as bone marrow transplant (Shihadeh, 2015). While haematopoietic stem cell transplantation has become standard health care to treat leukaemia, this discovery raised

[27] McArdle P. ATMP development challenges: From scientific advice to market authorisation. EMA-Europa Bio Information Day [Online]. London; 2015 [cited 2017 May 28]. Available from: URL: http://www.ema.europa.eu/docs/en_GB/document_library/Presentation/2015/11/WC500196332.pdf

hope in the development of advanced therapies for treating many serious diseases and conditions that are currently untreatable (Yuan and Wang, 2014).

There are thousands of international ongoing clinical trials using stem cells which are moving rapidly into clinical application (Heslop et al, 2015). Stem cells are characterised by two properties which differentiate them from all other cells in the body, namely self-renewing capacity and multi-lineage differentiation capacity.[28] The promise of stem cells as innovative tools for various therapeutic applications resides in these two properties, and in the ability to functionally re-populate a damaged tissue when transplanted and to generate differentiated progeny *in-vivo*, even in the absence of tissue damage[29] (Mosaad, 2014).

Embryonic stem cells, adult or somatic stem cells and induced pluripotent stem cells can be used as starting materials for medicinal products. The final stem cell-based medicinal product may consist of terminally-differentiated cells derived from stem cells, undifferentiated stem cells or a mixture of cells with varying differentiation profile.[27] Embryonic stem cells were the first type of pluripotent stem cells isolated from human embryos in 1998 (Thomson et al, 1998). Embryonic stem cells are pluripotent which suggest possible widespread use of these cells in therapeutic areas such as diabetes

[27] McArdle P. ATMP development challenges: From scientific advice to market authorisation. EMA-Europa Bio Information Day [Online]. London; 2015 [cited 2017 May 28]. Available from: URL: http://www.ema.europa.eu/docs/en_GB/document_library/Presentation/2015/11/WC500196332.pdf

[28] European Medicines Agency (EMA). Reflection paper on stem cell-based medicinal products EMA/CAT/571134/2009 [Online]. London: EMA; 2011 [cited 2017 May 28]. Available from: URL: www.ema.europa.eu/docs/en_GB/document_library/Scientific_guideline/2011/02/WC500101692.pdf

[29] Barfoot J, Kemp E, Doherty K, Blackburn C, Sengoku S, van Servelle, A et al. A Stem Cell Research: Trends and Perspectives on the Evolving International Landscape [Online]. EuroStemCell 2013; [cited 2017 May 28]. Available from: URL: https://www.elsevier.com/research-intelligence/resource-library/stem-cell-research-trends-and-perspectives-on-the-evolving-international-landscape

mellitus, CVD, Parkinson's disease or spinal cord injury.[30] There are three main sources of human embryonic stem cells namely, already existing embryonic stem cell lines, from in vitro fertilisation procedures and nuclear transfer technique (Hug, 2005). The ethical and political uncertainties regarding embryonic stem cells has led to more research on other types of stem cells such as reprogramming of somatic cells to produce induced pluripotent stem cells (Lo and Parham, 2009). Induced pluripotent stem cells (iPS cells) were first reported in 2006 using mouse cells, and a year later the procedure was replicated in human cells (Takahashi et al, 2007; Yu et al, 2007). IPS cells share similar features with embryonic stem cells namely, self-renewing capacity, pluripotent and form teratomas.[27] Human IPS cells have been used for disease modelling, drug discovery and stem cell therapy development (Shi et al, 2016).

Somatic or adult stem cells (ASCs) have more limited expansion and differentiation capacity than pluripotent stem cells. ASCs include various cell types, such as mesenchymal and hematopoietic stem cells, and various types of progenitor or precursor cells existing in foetal, adult or birth-associated tissue, including Wharton's Jelly of umbilical cord and the placenta.[27] Mesenchymal stem cells (MSCs) are the most frequently used cell types in stem cell clinical trials. This is largely due to substantial amount of evidence supporting the safety of administration of human MSCs in a variety of disease states. MSCs are capable of differentiating into different cell types such as bone, cartilage and muscle and express specific surface antigens. In addition, MSC

[27] McArdle P. ATMP development challenges: From scientific advice to market authorisation. EMA-Europa Bio Information Day [Online]. London; 2015 [cited 2017 May 28]. Available from: URL: http://www.ema.europa.eu/docs/en_GB/document_library/Presentation/2015/11/WC500196332.pdf

[30] National Institutes of Health. Stem Cell Information: Embryonic stem cells [Online]. U.S: NIH; 2016 [cited 2017 May 28]. Available from: URL: https://stemcells.nih.gov/info/Regenerative_Medicine/2006Chapter1.html

transplantation may exert its therapeutic effects through the secretion of paracrine factors that may have anti-inflammatory and immunomodulatory effects, increasing the success of engraftment with reduced risk of GvHD. MSCs can be used for both autologous and allogenic cell transplantation (Liew and O'Brien, 2012). MSCs were originally isolated from bone marrow, however limitations with this source include a painful isolation procedure, low frequency of cells in the bone marrow and decline in MSC characteristics with donor age (Batsali et al, 2013). The Wharton's Jelly of umbilical cords was identified as an alternative source for MSCs, and therefore cord blood banking increases the availability of MSCs for clinical application (Chatzistamatiou et al, 2014).

1.7 Stem cells banking

Umbilical cord blood (UCB) is an important source of stem cells for the treatment of both haematological and non-haematological diseases (Meissner-Roloff et al, 2012). Worldwide, over 600,000 UCB units have been stored for transplantation and more than 30,000 UCB transplantations have been performed (Sun et al, 2016). UCB banking may be private, public, or hybrid private-public banks. Private banks preserve UCB stem cells for autologous or familial usage, while public banks store cells for autologous use (Roura et al, 2015).

The scientific quality of the cell source is crucial in stem cell research. Researchers are responsible for assuring the characteristics of the cells used. Stem cell banks are established to preserve characteristics of the cells and to facilitate stem cell research. In addition, it is essential for a stem cell bank to assure the supply of standardised and quality-controlled stem cell lines, with high quality control systems in place to ensure

the reliability, safety and reproducibility of test results (Antoniadou and David, 2015; Stacey, 2017).

The procedure for stem cell banking involves collection of cells isolated from various sources, proliferation, storage and preservation for future use. The set-up of a stem cell bank is similar to a stem cell laboratory, where GMP principles should be applied from the first stage of cell banking. Major functional areas commonly required in a stem cell bank include preparation areas for cell isolation and culture, quality control laboratories to test the cell characteristics and ensure that cultured cells meet the required GMP standards, storage areas including freezer rooms, and information areas to archive donor information, consent forms and data files where file management should conform to GMP requirements (Figure 1.4).

When a cell line is used over different manufacturing cycles a two tiered cell banking system is recommended namely master cell bank and a working cell bank (Figure 1.5). Tissue and cells arriving at a stem cell banking facility should be quarantined for donor screening and testing. Donor screening tests include human immunodeficiency virus (HIV) type 1 and 2, hepatitis B virus (HBV), hepatitis C virus (HCV) treponema pallidum (syphilis), Creutzfeldt Jakob disease (CJD), human T-lymphotropic virus (HTLV) types 1 and 2 and cytomegalovirus (CMV). Other quality control tests carried out at the master cell bank for characterisation of cells include, but are not limited to, identity, purity, viability, stability, tumourigenicity potency, sterility, mycoplasma and endotoxin testing. The working cell bank can be prepared from fresh cultures or frozen vial from the master cell bank. Stem cells from the working cell bank may be distributed to researchers. Establishment of a master and a working cell bank enables a

supply of quality controlled cells over a long period of time that may be used for research purposes and product development for stem cell therapies (Sun et al, 2016).

Figure 1.4 Major functional areas in a stem cell bank

Reproduced from Sun C, Yue J, He N, Liu Y, Zhang X, Zhang Y. Fundamental principles of stem cell banking. Advances in Experimental Medicine and Biology 2016; 951: 41–58.

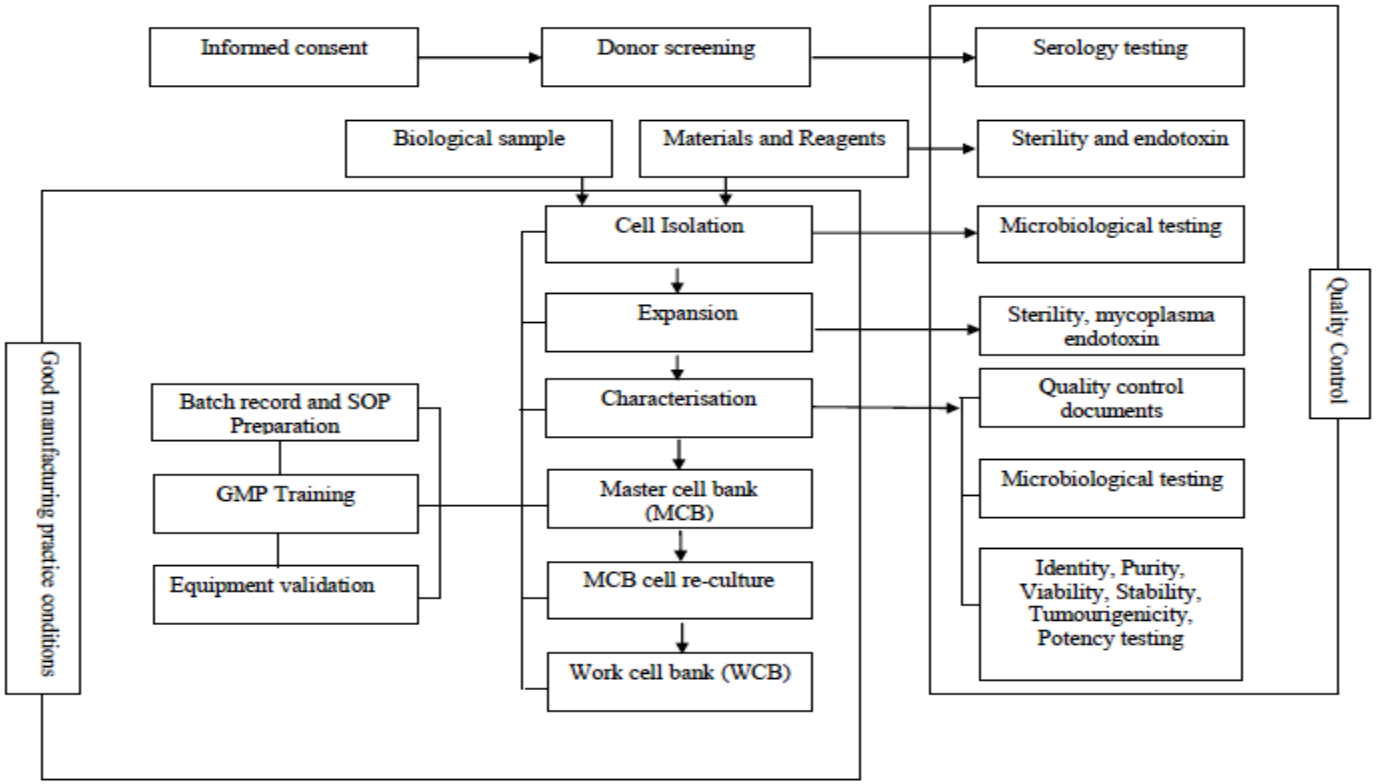


Figure 1.5 Workflow for stem cell banking under GMP conditions

Adapted from Sun C, Yue J, He N, Liu Y, Zhang X, Zhang Y. Fundamental principles of stem cell banking. Advances in Experimental Medicine and Biology 2016; 951: 41 – 58.

1.8 Ensuring quality and safety of blood components

Blood component therapy represents a form of cellular therapy, however, regulatory, technical and operational differences exist between the well-established processes used for the preparation of blood components and the innovative technologies used in stem cell therapies (Rebulla and Giordano, 2012). Blood components used for transfusion therapy include platelet concentrates, red cell concentrates and plasma (Ramirez-Arcos et al, 2016). Ensuring that blood components are of high quality, safe and deliver the intended benefits to patients throughout the shelf-life is a complex mission (Acker et al, 2016).

In the wake of the Acquired Immune Deficiency Syndrome (AIDS) tragedy in the haemophilia population between 1982 and 1984, blood safety became a major public concern (Evatt, 2006; Epstein, 2012). Other tragedies and scandals threatening blood supply have been reported internationally, including the outbreak of hepatitis C in Ireland in 1977 associated with the administration of anti-D immunoglobulin contaminated with the hepatitis C virus that infected more than 1600 people.[31] Increasingly recognised threats from hepatitis viruses and new emerging diseases including vCJD and Zika virus intensify concerns on the safety of blood supply (Epstein, 2012; Lanteri, 2016). The approach to blood safety depends on donor education, screening, testing and discarding blood components in inventory if donor exposure or illness is reported post-donation (Epstein, 2012).

[31] O Malley K. Considering the policy of indefinite deferral imposed on MSM blood donors in Ireland. UCD Student Medical Journal [Online] 2017; [cited 2017 May 18]. Available from: URL: http://www.ucdsmj.com/2-4/

The inherent risks of blood and the complexity of collecting, testing, processing, storing and distributing good quality and safe blood and blood components in an adequate timely and equitable access requires set-up of an organised national blood establishment (Epstein, 2012). A specialised directive regulating blood transfusion was enacted in 2002.[32] EU competent blood regulatory authorities organise inspections and control measures to ensure that the blood establishment complies with the requirements listed in Directive 2002/98/EC.[32]

1.9 Regulatory sciences for stem cell-based medicinal products and blood components

A regulatory science approach is applied by developing and validating new standards and tools to evaluate and assess the benefit–risk of medicinal products and blood components, facilitating sound and transparent regulatory decision-making. Regulatory science also contributes to the evaluation of regulatory systems to ensure patient safety, enhancing public health and stimulating the development of medicines for unmet medical needs (Leufkens and Eichler, 2011; Yuan and Wang, 2014).

Yuan and Wang (2014) address the need of promoting stem cell regulatory sciences, where all stakeholders including researchers, clinicians, regulators, manufacturers, investors, public and the media must understand the importance of effective regulation. Networking between different stakeholders promotes an effective regulatory environment that is critical to build a culture of continuous improvement of all

[32] European Commission. Directive 2002/98/EC of the European Parliament and of the Council of 27 January 2003 setting standards of quality and safety for the collection, testing, processing, storage and distribution of human blood and blood components and amending Directive 2001/83/EC [Online]. Official Journal of the European Union 2003; L33:30-40 [cited 2017 May 28]. Available from: URL: http://eur-lex.europa.eu/LexUriServ/LexUriServ.do?uri=OJ:L:2003:033:0030:0040:EN:PDF

regulatory sciences perspectives, including benefit-risk assessment, research and incentives to support regulatory science and expertise. Risk assessment for each product and change in the manufacturing process must be performed to determine the impact on the finished product (Yuan and Wang, 2014). Figure 1.6 summarises actions that should be taken in the context of establishing regulatory sciences in stem cell therapy to facilitate translating innovative stem cell science into promising products, a model that could be adapted to any other process or product such as the preparation of blood components.

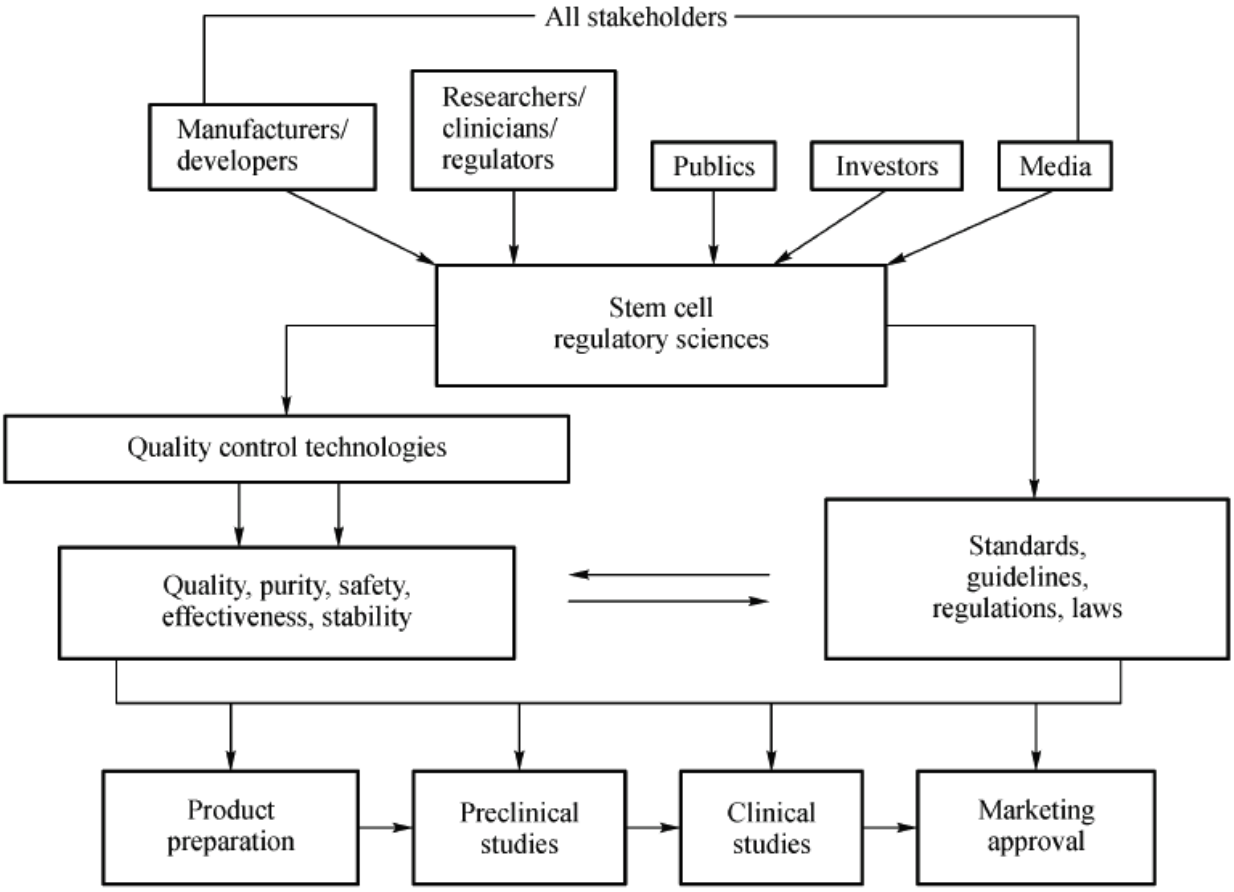


Figure 1.6 Promoting stem cell regulatory sciences

Reproduced from Yuan BZ and Wang J. The regulatory sciences for stem cell-based medicinal products. Frontiers of Medicine 2014; 8(2): 190-200.

1.10 Applying regulatory sciences in this research study

The promise of stem cell research in making regenerative medicine accessible, has found great public interest, and has further attracted clinicians, scientists and entrepreneurs to this field of research (Inamdar et al, 2012).

Malta is relatively new to stem cell research and it is important to get involved in these innovative therapies since it forms a vital therapeutic armamentarium for many clinical chronic conditions. Life sciences are a priority area in the economic plan for Malta in line with EU 2020 targets to have a smart, sustainable and inclusive economy.[33] The Maltese Government is investing to create an optimal setting for companies operating within the life sciences area to improve the healthcare sector. There is a unique opportunity for Malta to get involved in stem cell therapy which is expected to have a positive impact to achieve health and economic goals, where advanced therapies are an important income-generating component of the biotechnology sector. The recently developed Malta Life Sciences Park (MLSP) by Malta Enterprise, in collaboration with the University of Malta (UOM) and Mater Dei Hospital (MDH), provides state-of-the-art facilities for research.[34] The Malta Medicines Authority (MMA) was relocated to the MLSP to offer opportunities for open dialogue and collaboration between the international scientific, regulatory and medical sectors encouraging exchange of knowledge and ideas in innovative areas.

[33] The Malta Council for Science and Technology (MCST). Malta's national strategic plan for research and innovation: A vision for knowledge-driven growth 2011-2020 [Online]. Malta: MCST; 2011 [cited 2017 May 28]. Available from: URL: http://mcst.gov.mt/files/uploaded/National%20Strategy%20DRAFT.pdf

[34] Malta Enterprise. Life Sciences [Online]. Malta: ME; 2014 [cited 2017 May 28]. Available from: URL: www.maltaenterprise.com/en/media/news/2014/life-sciences

In the last two years, there was an increase in national requests by potential investors for regulatory advice on how the MMA regulates stem cell therapies. In response to this interest, the MMA is proactively exploring new ways to nurture a regulatory environment that supports development of innovative medicines and to establish scientific and technical competence to regulate stem cell therapy and blood components. The rationale of this research study was to leverage European scientific excellence through networking, to support the setting up of a stem cell therapy unit in Malta and to up-skill regulators' competence to effectively regulate stem cell therapy facilities and blood establishments.

1.11 Aim and objectives

The aim of this research was to study the therapeutic and economic implications of regulating stem cell therapy and blood components in Malta.

The objectives were to:

1. Identify European collaborators in the area of stem cell therapy and blood components
2. Establish a network of collaborators to investigate the setting up of a stem cell therapy unit in Malta
3. Address requirements for setting up a stem cell therapy unit in Malta and consider economic implications
4. Identify regulatory sciences norms through development of a quality manual for regulators of stem cell therapy facilities and blood establishments
5. Compile a glossary of terminology for use in the field of stem cell therapy and blood components.

CHAPTER 2

METHODOLOGY

The methodology included building a structured collaboration with key stakeholders in Europe, namely academia, industry, technical specialists and regulators. The accumulation of knowledge in the area of stem cell therapy and blood components was analysed through scientific review case studies identified during site studies. A network of collaborators of European and local public institutions was established to set up a stem cell therapy unit in Malta. The requirements and economic implications to set up a stem cell therapy unit in Malta were identified and evaluated through the network. A quality manual for regulators was developed through identified regulatory sciences norms and a glossary was compiled and validated to define terms used in the field of stem cell therapy and blood components.

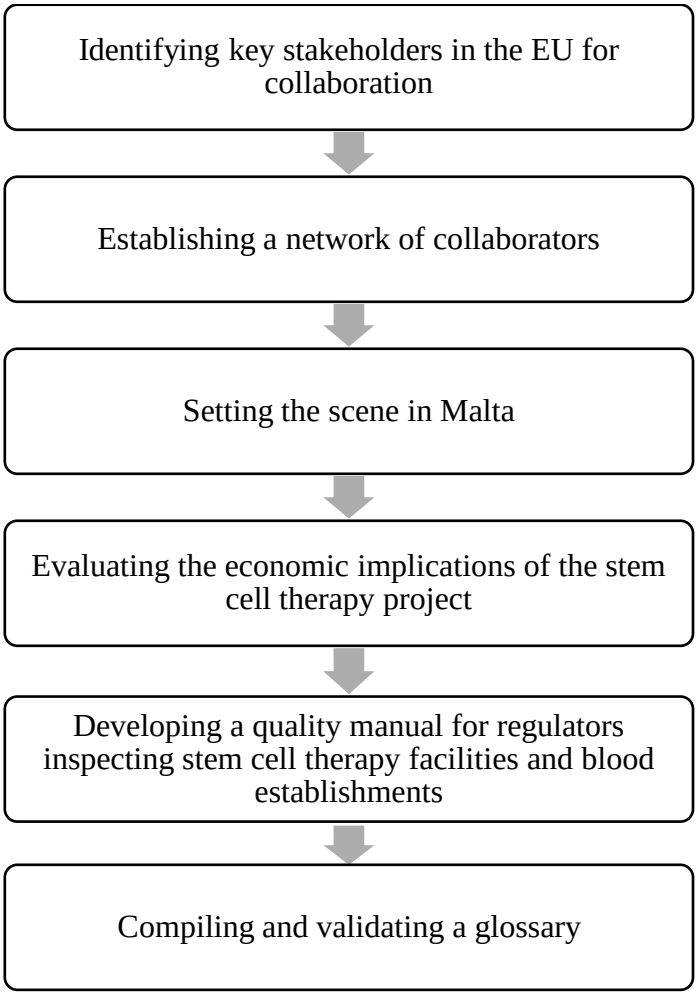


Figure 2.1 Development of a stem cell therapy and blood components research

2.1 Identifying key stakeholders in the EU for collaboration

Knowledge and experience in manufacturing of stem cell therapy products is important for the development of successful stem cell therapy facilities (Pearce et al, 2014). Site studies to established stem cell therapy facilities and blood establishments can provide valuable insights from user experiences, and collaborations with such facilities can be beneficial for sharing of resources, experiences, ideas and expertise (Inamdar et al, 2012).

Academia, industry, technical specialists in the field of stem cell therapies and the European medicines regulatory network were identified as potential key stakeholders for collaboration.

2.1.1 Collaborating with academia

Research plays an important role in academic and scientific circles, where academic institutions are considered as major stakeholders in the development of ATMPs (Hanna et al, 2016). A site study in an academic setting was selected to 1) Understand the set up of a stem cell therapy unit, including evaluation of the environmental monitoring system, equipment required and the implemented quality management system and 2) Evaluate their experience in the clinical translation of stem cell research. The site study was selected through internet search of European universities carrying out research in stem cell therapy. The search was narrowed to universities in the UK to avoid language barriers during the site study.

2.1.2 Collaborating with industry

Products initially developed by non-commercial sponsors may be licensed to the industry for commercialisation of the product, therefore translating the research into viable clinical therapies to obtain a marketing authorisation (Burningham, 2013). The pharmaceutical industry was identified as a key stakeholder in this research to understand the challenges experienced by the industry to obtain a marketing authorisation of stem cell-based medicinal product. The site study was selected following systematic review of stem cell-based medicinal products that have a valid centralised marketing authorisation using the European Medicines Agency website[35]. A site study in the pharmaceutical industry was selected to evaluate the 1) Set up of a GMP stem cell therapy facility including specifications of cleanroom facilities and equipment, 2) Clinical and regulatory aspects of stem cell therapy and 3) Manufacturing process of stem cell therapy.

2.1.3 Collaborating with technical specialists

The rationale for including technical specialists as key collaborators in the research was to avail of their knowledge and experience in this innovative field to support the setting up of a stem cell therapy unit in Malta. A site study at a company offering technical specialist services was carried out to identify 1) Potential areas of collaboration, 2) The appropriate business model for the Malta stem cell project, 3) Experience in the clinical application of stem cells and 4) Possibility to facilitate the conduct of a feasibility study for the setting up of a stem cell therapy unit in Malta.

[35] European Medicines Agency. Home [Online]. London: EMA; 2017 [cited 2017 May 28]. Available from:URL: http://www.ema.europa.eu/ema/index.jsp?curl=pages/home/Home_Page.jsp&mid=

2.1.4 Collaborating with the European medicines regulatory network

Potential collaborations with European regulators were explored through the European medicines regulatory network. The Malta Medicines Authority (MMA) participates actively within the European medicines regulatory network through meetings, such as Heads of Medicines Agencies meetings and bilateral meetings with national competent authorities, where contacts and collaborations with other EU authorities are established. Site studies through participation of regulatory inspections with an identified European regulator were planned to determine regulatory sciences norms in the field of stem cell therapy and blood components. Observing inspections of a stem cell therapy facility and a blood establishment allows the research to evaluate the regulators approach in licensing and regulating such facilities to ensure the good quality and safety of stem cell therapies and blood components. Language barrier was taken into consideration when selecting the national competent authorities for collaboration in the European medicines regulatory network. While carrying out an inspection, the inspector should facilitate exchange of information and it is important to be able to understand and communicate clearly and effectively with all persons involved during the inspection in order to achieve the objectives of the inspection.[36]

Through a Memorandum of Understanding (MOU) (Appendix 1) between the MMA and the Health Regulation Department, which is the local competent authority responsible to regulate the use of human blood and blood components organs, tissues

[36] European Commission. Eudralex – Volume 10 Chapter IV Recommendation on Inspections [Online]. Brussels: EC; 2016 [cited 2017 May 28]. Available from: URL: http://ec.europa.eu/health//sites/health/files/files/eudralex/vol-10/v10_chap4_en.pdf

and cells,[37] the need to collaborate and share technical expertise in inspecting the local blood establishment was identified.

2.1.5 Presenting case studies through site studies

During site studies with academia, industry and technical specialists, discussions with experts in the field of stem cell therapy were conducted to learn from their knowledge and experiences. During these discussions, the researcher asked questions related to their experiences in the clinical and regulatory aspect of stem cell therapy, and requirements for the setting up of a stem cell therapy unit in Malta. Questions asked related to special considerations to be taken up during design of a stem cell therapy unit, equipment required to set up a stem cell laboratory, and experiences in the clinical application of stem cell therapy. The questions were adapted according to the activities undertaken by the site study to identify practices in different institutions and to evaluate different case studies related to stem cell therapy, thereby serving as a platform for this research study.

Sites studies as a result of collaboration with the European medicines regulatory network were conducted through observed inspections with a national competent authority. The information gathered through observed inspections was compiled and case studies were presented to highlight regulatory sciences norms in stem cell therapy facilities and blood establishments.

[37] Ministry for Health. Health Care Standard [Online]. Malta; 2017[cited 2017 May 28]. Available from: URL: https://health.gov.mt/en/hcs/Pages/hcs.aspx

2.2 Establishing a network of collaborators

Local public institutions were identified as key stakeholders in the establishment of a Malta stem cell project, both in research and development of the stem cell therapy, and in translation of the product from bench to bedside (Table 2.1). Discussions with local institutions started from the conceptualisation of the project, to interact openly with all stakeholders and to identify the resources available, including the skills and assets of the public. A network of collaborators was established through multi-site studies with different European institutions and the networking of these European institutions with local public institutions. Representatives from the identified local public institutions were invited to participate in the site studies at European institutions to network and together identify the roadmap for the setting up of a stem cell therapy unit in Malta.

The costs for setting up a stem cell therapy facility and costs of research and development of stem cell-based products are high (Pellegrini et al, 2016). Developers of stem cell therapies require support in funding the projects and would benefit from access to capital investment (Giebel, 2005). Potential investors for the Malta stem cell project were identified through stakeholder meetings with the MMA. Identified stakeholders approached the MMA for regulatory advice and to discuss their interest to invest in stem cell therapy. Potential investors for the Malta stem cell project were assessed for financial capacity, experience of working in the pharmaceutical sector and appreciation of regulatory sciences.

Networking meetings were organised to bring together the investors, academia, technical specialists, regulator and local public institutions to interact openly and to identify ways how the project can be developed to set up a stem cell therapy unit in

Malta. This early interaction strengthens the network to meet the interests of all stakeholders (Pellegrini et al, 2016).

Table 2.1 Identified local public institutions to collaborate in the Malta stem cell project

Local public institution	Rationale for collaboration
Malta Medicines Authority (MMA)	MMA took a proactive role in this project, where the involvement of the regulator ensures that the appropriate regulatory scientific approach is applied from the early stages of the stem cell project.
Malta Laboratories Network (MLN)	Establishment of a strategic partnership through signing of a MOU on collaboration between the MLN and all parties involved in the Malta stem cell project facilitates exchange of knowledge, expertise and resource allocation to support the setting up of a stem cell therapy unit in Malta.
University of Malta (UOM)	Meetings with the Dean of the Faculty of Medicine and Surgery at UOM and Head of the Department of Physiology and Biochemistry were coordinated to explore the possibility of utilising the new laboratories in the Biomedical Sciences Building, University of Malta.
Malta Enterprise (ME)	Meetings between the Chief Executive Office of ME and other potential collaborators interested in the stem cell project were organised to identify measures and incentives that can be offered to support start-up companies interested in research and development of innovative products.
Malta Life Sciences Park (MLSP)	The strategic location and state-of-the art laboratory spaces at MLSP could be ideal to set up a stem cell therapy unit. Meetings and site visits with the Chief Executive Officer of MLSP were organised to identify potential laboratory spaces suitable for the project and to discuss laboratory features and required adjustments for the facility to be GMP compliant.
Mater Dei Hospital (MDH)	MDH was identified as a potential clinical setting to conduct clinical trials with stem cell therapy. Although clinical translation of stem cell therapy would come at a later stage of the project, involvement of MDH was considered as an asset in the planning phase.
National Blood Transfusion Service (NBTS)	Banking and manipulation of cells needs to be carried out according to GMP regulations, however the long experience of blood establishments in managing aseptic procedures and their active involvement in haemovigilance programs, including an established system for the traceability of the products, cannot be undermined (Rebulla and Giordano, 2012). NBTS may represent a convenient setting for development of stem cell therapy, and meetings with the Director at NBTS were organised.

2.3 Setting the scene in Malta

The next step in the research study was to consolidate the information and experiences gained through collaboration with European institutions, along with realisation of the level of support available locally through the establishment of the network. The methodological framework to set up a stem cell therapy unit in Malta was outlined (Figure 2.2).

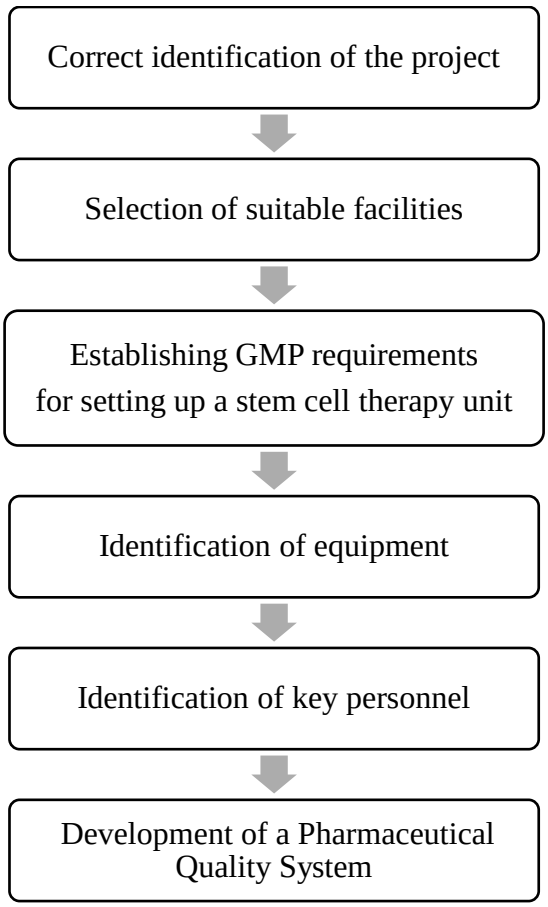


Figure 2.2 Methodological framework to set up a stem cell therapy unit in Malta

2.3.1 Correct identification of the project

Each project has a specific product and process, regulatory positioning and customisation which depend on the interests and specific requirements of the investors. Different business models for developing a stem cell therapy unit were identified and analysed through the network. The plan for implementing the stem cell project was established through sharing of experiences of collaborators in the network.

2.3.2 Selection of suitable facilities

A review of available infrastructure and facilities to set up the stem cell therapy project in Malta was carried out. Factors taken into consideration by the researcher included:

- Laboratory space available to accommodate the process flow, including product receipt area, material storage, production areas, quality control laboratory, microbiological laboratory, administration and changing rooms
- Laboratory finishes such as walls, flooring, ceilings, air supply, environmental monitoring system and air quality
- Access control
- Risks of contamination from the surrounding environment and control measures from sources of contamination
- Possibility to upgrade and maintain laboratory to comply with GMP standards
- Services such as supply of liquefied gases, ventilation, electricity and water

2.3.3 Establishing GMP requirements for setting up a stem cell therapy unit

Key considerations in the planning and design of a GMP stem cell therapy unit include:

- Major functional areas
- Number of cell processing laboratories
- Classification of different rooms (Grade A, B, C and D)
- Air handling system specifications including air exchange per hour, temperature and humidity
- Pressure gradient between different classified rooms
- Material flow
- Equipment flow
- Interior finishes

These requirements were evaluated and verified during site studies and collaborations with EU institutions.

2.3.4 Identification of equipment

Equipment required for the cell processing laboratories was identified through a scientific literature search on equipment used in stem cell laboratories (Wesselschmidt et al, 2011; Leemhuis et al, 2014) and through the site studies and collaborations with EU institutions.

2.3.5 Identification of key personnel

GMP personnel are crucial for a pharmaceutical manufacturing industry (Montemurro et al, 2015). Key personnel required to set up a stem cell therapy unit were identified

through the site studies and as outlined in the GMP guidelines.[38] The level of commitment of the identified personnel to work in the stem cell therapy unit was evaluated. The importance of having personnel with the necessary skills to work in highly technical operations, such as aseptic techniques, cannot be underestimated. The required competences for personnel working in cell processing laboratories were evaluated.

2.3.6 Development of a pharmaceutical quality system

The pharmaceutical quality system should be designed to facilitate innovation and continuous improvement, which can be applied throughout the lifecycle stages from the development stage of the product up to manufacturing, and finally product discontinuation.[39]

Throughout the research study, the importance of documentation and structuring specific standard operating procedures (SOPs) was emphasised. A list of policies and SOPs was compiled to give a foundation of core procedures that should be captured in a good pharmaceutical quality system.

[38] European Commission. EudraLex – Volume 4 EU Guidelines for Good Manufacturing Practice for Medicinal Products for Human and Veterinary Use – Part 1 Chapter 2: Personnel [Online]. Brussels: ECl 2013[cited 2017 May 28]. Available from: URL: https://ec.europa.eu/health/sites/health/files/files/eudralex/vol-4/2014-03_chapter_2.pdf

[39] International Conference on Harmonisation. Q10 Pharmaceutical Quality System [online] 2008; [cited 2017 May 28]. Available from: URL: http://www.ich.org/products/guidelines/quality/quality-single/article/pharmaceutical-quality-system.html

2.4 Evaluating the economic implications of the stem cell therapy project

The economic implications of setting up a stem cell therapy facility were identified through the network including the required investment for the project, estimated timelines to set up the facility and anticipated time to reach the market and make an adequate return of investment (ROI) to retire the investment.

A preliminary costing exercise was performed to determine capital and operating expenses for the stem cell therapy unit including: 1) Rent of premises, 2) Costs for setting up a class B processing laboratory, 3) Costs of equipment and 4) Yearly salaries of key personnel (Table 2.2).

Table 2.2 Method to source costing information on expenditure

Expenditure	Source to obtain information on costs
Rent of premises	MLSP
Set up of a Class B processing laboratory	Technical specialists
Equipment	Potential investors
Salaries of key personnel	Pharmaceutical industry

Estimate treatment costs of stem cell-based medicinal products were identified through collaborations during the site studies.

2.5 Developing a quality manual for regulators inspecting stem cell therapy facilities and blood establishments

A quality manual for regulators of blood and stem cell therapy facilities was developed following collaborations through the European medicines regulatory network, and taking into account the following references:

- European Blood Inspection System Manual (Siefried and Seidl, 2010)
- CATIE Manual (Seidl et al, 2012)
- EuSTITE Operational Manual[40]
- Compilation of Community Procedures on Inspections and Exchange of Information[41]

The aim of the quality manual is to support the MMA to set up and organise a system for inspection of stem cell therapy facilities and blood establishments. Four regulatory affairs experts were invited to form part of a panel to validate the quality manual. Suggestions received from the panel were used to update the quality manual.

2.6 Compiling and validating a glossary

A need for a glossary which defines regulatory terms used within the field of stem cell therapy and blood components was identified during the course of the study. Terms and definitions were aligned with existing regulations, guidelines or standards. The glossary was laid out into three columns; term, definition and author-date citation. References for each term were placed at the end of the glossary. The terms selected for the glossary were identified through collaborations and discussion with experts in the field of stem cell therapy and blood components.

[40] European Commission. Inspection of tissue and cell procurement and tissue establishments – Operational Ma+nual for Competent Authorities. EC: Brussels, 2015 [cited 2017 May 28]. Available from: URL: http://ec.europa.eu/health//sites/health/files/blood_tissues_organs/docs/manual_11_en.pdf

[41] European Medicines Agency. EMA/572454/2014 Rev 17. Compilation of Community Procedures on Inspections and Exchange of Information. [Online]. London: EMA; 2014 [cited 2017 May 28]. Available from: URL: http://www.ema.europa.eu/docs/en_GB/document_library/Regulatory_and_procedural_guideline/2009/10/WC500004706.pdf

The glossary was subjected to a pilot validation, where two pharmacists and one layperson were recruited to assess the glossary. The feedback received from each member was used to update the glossary.

CHAPTER 3

RESULTS

The results cover 1) The identified European collaborators as key stakeholders in academia, industry, technical specialists and the European medicines regulatory network, 2) Case studies from the five site studies carried out through collaborations with European centres of excellence in stem cells and blood regulatory sciences namely, Cell Therapy Unit at King's College London, Holostem Terapie Avanzate, CTP System, Centre for Cell Manufacturing Ireland and Irish Blood Transfusion Service, 3) Establishment of a network of collaborators to investigate the setting up of a stem cell therapy unit in Malta, 4) Requirements to set up the GMP stem cell therapy unit, 5) Economical implications of setting up a stem cell therapy unit, 6) Establishment of a quality manual for the inspection of stem cell therapy facilities and blood establishments, 7) Compiled glossary of terms related to stem cell therapy and blood components.

3.1 Identified European collaborators

The collaborations established through the carried out five site studies (Figure 3.1) were:

1. King's College London site study which represents collaboration with academia (Appendix 2)
2. Holostem Terapie Avanzate site study in Modena which represents collaboration with industry (Appendix 3).
3. CTP system site study in Siena which represents collaboration with technical specialists in the field of advanced therapies (Appendix 4).
4. Health Products Regulatory Authority (HPRA) site studies in Ireland which represent collaboration with the European medicines regulatory network. Two inspections with HPRA, blood establishment inspection at Irish Blood

Transfusion Service (IBTS) in Dublin and a stem cell therapy facility inspection at the Centre for Cell Manufacturing Ireland (CCMI) in Galway were carried out (Appendix 5 and 6).

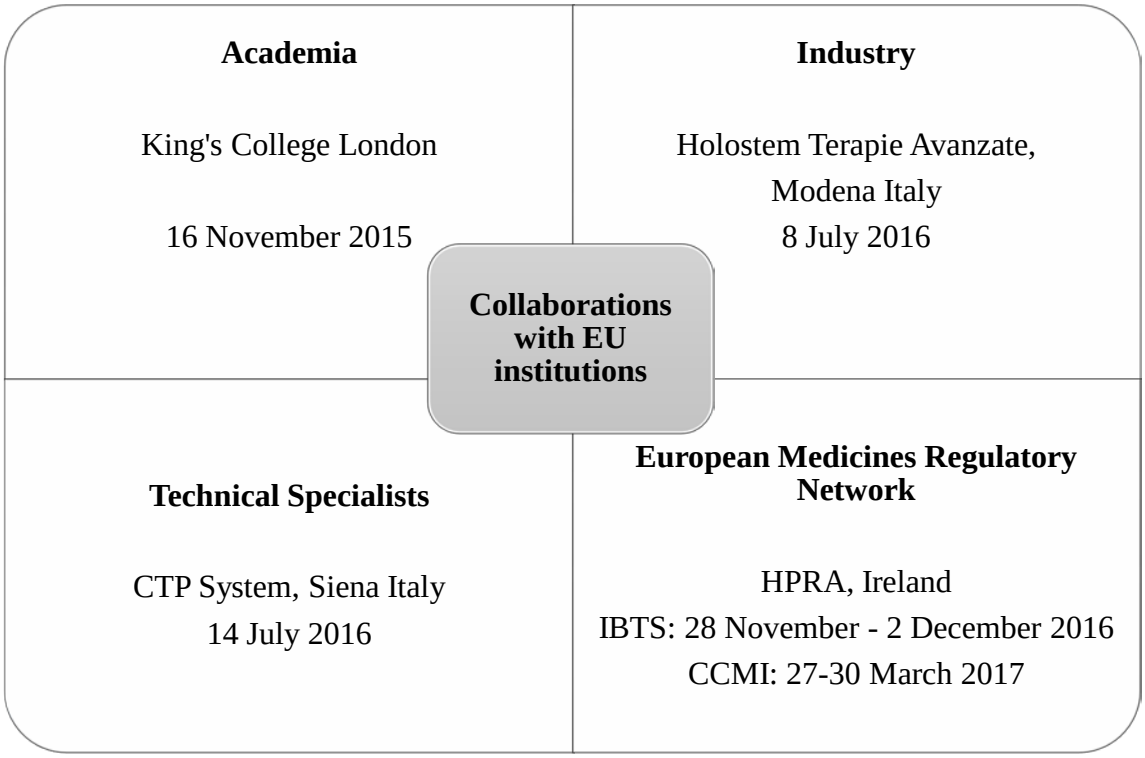


Figure 3.1 Collaborations with EU institutions

3.1.1 Site Study 1: Identified collaborator with academia

King's College London has one of the largest state-of-the-art GMP cell therapy units in the academic setting in Europe, with extensive research in hepatocyte, islet and bone marrow transplantation. Experts at King's College London have published numerous scientific papers on stem cell research-based innovations involving islet, liver and bone marrow transplants,[42] King's College London is renowned for the successful clinical application of hepatocyte transplantation. A press release 'World first liver treatment at

[42]King's College London. Clinical research Facility launched [Online]. 2014 [cited 2017 May 28]. Available from: URL: http://www.kcl.ac.uk/ioppn/news/records/2014/May/Clinical-Research-Facility-

King's'[43] published in November 2011 highlights how a case of a seven month-old boy with liver failure due to a herpes-simplex virus infection was treated successfully for the first time with human hepatocyte transplantation. Scientific research (Dhawan et al, 2006; 2010; 2015) published by the Hepatocyte Biology and Transplantation Group at King's College supported the technique used to transplant liver cells in order to restore the metabolic functions of the failing liver. King's college is one of the major centres in Europe for the treatment of liver disease and carries out over 200 liver transplants yearly.[42] Dr Ragai Mitry, Head of Liver Cell Production at King's College London was contacted to arrange a site study.

3.1.2 Site Study 2: Identified collaborator with industry

Holostem Terapie Avanzate in Modena, Italy was selected as a collaborator from industry due to its successful achievements with Holoclar, the first stem cell therapy medicinal product approved in the EU (Milazzo et al, 2016). Holostem is a bioengineering company in Modena, founded in 2008 by internationally renowned researchers Professor Michele de Luca and Professor Graziella Pellegrini supported by the University of Modena and Reggio Emilia, and Chiesi Farmaceutici S.p.A, a leading Italian pharmaceutical company in Parma.[44] Various scientific papers on the work carried out at Holostem were published (Pellegrini et al, 1997; De Luca et al, 2006; Pellegrini and Luca, 2014; Milazzo et al, 2016), including a scientific review published by Pellegrini et al, (2016), which focus on recent developments in stem cell-based

[43] King's College Hospital NHS Foundation Trust. World first liver treatment at King's [Online]. 2011[cited 2017 May 28]. Available from: URL: https://www.kch.nhs.uk/news/media/press-releases/view/9460

[44] Holostem Terapie Avanzate. About us [Online]. 2017 [cited 2017 May 28]. Available from: URL: http://www.holostem.com/en/Homepage.html

corneal therapy. Professor Tor Paaske Utheim, ophthalmologist at the Oslo University Hospital in Norway stated that approval of this promising technology "is a very important step in the right direction for other cell therapies" (Dolgin, 2015).

On the 25 January 2016, Professor Anthony Serracino Inglott, Chairman of the MMA and Professor John Joseph Borg, Post-Licensing Director at MMA and CHMP member, received an invitation letter (Appendix 3) from Dr Paolo Chiesi, President and Dr Andrea Chiesi, Former Chief Executive Officer of Holostem, to visit the manufacturing facility in Modena and gain first-hand personal experience of the work and efforts involved in the manufacturing process of stem cells. Exploring potential collaboration on regulatory sciences with this pharmaceutical company of significant standing, with experience in the marketing of a stem cell product provide valuable contribution in development of a stem cell therapy unit in Malta.

3.1.3 Site Study 3: Identified collaborator with technical specialists

CTP System is a leader in Italy in the provision of GMP services in the field of life sciences. The organisation engages around 180 competent and professional scientific experts including engineers, computer scientists and biologists. Standard services range from design and project management, to qualification and validation, support for building quality systems and validation advice on IT systems, preventive maintenance and regulatory support for pharmaceutical companies, as well as microbiological and chemical/physical analysis through certified laboratories.[45]

[45] CTP System. CTP System [Online] 2017 [cited 2017 May 28]. Available from: http://www.ctpsystem.com/index.php/en/system-components/ctp-tecnologie-di-processo

The University of Malta and the MMA has collaborated with CTP System through various seminars organised for students undertaking courses in the area of pharmaceutical industry. Seminars are open to stakeholders in the pharmaceutical industry to improve skills and competences related to pharmaceutical processes and pharmaceutical regulatory requirements. Discussions with CTP on the setting up of a stem cell therapy unit started in March 2016 during one of the aforementioned seminars. CTP System invited the MMA to visit their headquarters in Siena, Italy to discuss ATMP projects carried out by CTP and how technical specialists in the advanced therapy field can support the Malta project such as in conducting a feasibility study.

3.1.4 Site Study 4 and 5: Identified collaborator in the European Medicines Regulatory Network

Previous successful collaborations between the MMA and the Medicines and Healthcare products Regulatory Agency (MHRA) and HPRA led to initiation of discussions with the two authorities to explore possible collaborations on training related to inspections of stem cell therapy facilities and blood establishments. MHRA and HPRA have vast experience in regulating and licensing of stem cell therapy facilities and blood establishments. Training opportunities were discussed and consolidated during visits to the MMA by the Chief Executive of MHRA, Dr Ian Hudson, and the Chief Executive of HPRA, Dr Lorraine Nolan, in 2016. HPRA invited MMA to participate in two inspections in Ireland, IBTS (blood establishment) and CCMI (stem cell therapy facility).

3.2 Case studies through collaborations with EU Institutions

The case studies presented in this section exemplify the potential of collaboration in the process of knowledge generation and provide valuable insights from experts in the field of stem cell therapy and preparation of blood components. The experiences from the five site studies identified regulatory sciences norms that support the setting up of a stem cell therapy unit in Malta and competence building for regulators inspecting stem cell therapy facilities and blood establishments.

3.2.1 Academia case study: Clinical application of hepatocyte transplantation

Hepatocyte transplantation is an alternative to orthotopic liver transplantation in patients with acute liver failure or metabolic liver disease, where it supports the native liver to regenerate or helps in restoring liver enzymatic function (Dhawan et al, 2006). Two major advantages of hepatocyte transplantation are that multiple recipients can benefit from one donor liver and it is less invasive than orthotopic liver transplant. The Institute of Liver Studies and the Paediatric Liver Centre at King's College London carried out extensive research on the clinical application of hepatocyte transplantation (Mitry et al, 2002; Dhawan et al, 2005; Dhawan et al, 2006; Fitzpatrick et al, 2009; Mitry, 2009; Dhawan et al, 2010; Dhawan, 2015).

Hepatocytes are isolated from unused or rejected liver for transplantation, where cellular activity and viability of the cells depends on the quality of the original tissue. The hepatocyte transplantation occurs via intraportal injection accessed by percutaneous transhepatic puncture of the portal vein or inferior mesenteric vein catheterisation. Dr

Ragai Mitry and his team developed an innovative technique, licensed by the MHRA, to purify and encapsulate hepatocytes for clinical application. GMP grade hepatocyte microbeads were developed to protect cells from host immune attack while still maintaining cell viability and function. Cryopreservation techniques have been developed to store hepatocytes for a long period and can be thawed for immediate use and microencapsulation technique protects hepatocytes from cryoinjury. The case illustrates an example of clinical translation of stem cell research from 'bench to bedside' where patients have been successfully treated with this stem cell therapy.

3.2.2 Industry case study: Challenges for the marketing authorisation of stem cell-based medicinal products – Holoclar case

Clinical studies for Holoclar where conducted before implementation of the ATMP regulation EC 1394/2007[2] in 2008. The implications of the new regulation were that Holoclar:

- Was classified as a tissue engineered product
- Falls under Directive 2001/83/EC[46] which regulates medicinal products in the EU
- Required demonstration of quality, safety and efficacy of the product to obtain a marketing authorisation

[2] European Commission. Regulation (EC) 1394/2007 of the European Parliament and of the Council of 13 November 2007 on advanced therapy medicinal products and amending Directive 2001/83/EC and Regulation (EC) 726/2004 [Online]. Official Journal of the European Union 2007; L324:121-137 [cited 2017 May 28]. Available from: URL: http://eur-lex.europa.eu/legal content/EN/TXT/PDF/?uri=CELEX:32007R1394&from=EN

[46] European Commission. Directive 2001/83/EC of the European Parliament and of the Council of 6 November 2001 on the community code relating to medicinal products for human use [Online]. Official Journal of the European Union 2001b; L311:67-128 [cited 2017 May 28]. Available from: URL: www.ema.europa.eu/docs/en_GB/document_library/Regulatory_and_procedural_guideline/2009/10/WC500004481.pdf

- Manufacture must be compliant with EU GMP standards

Challenges to obtain approval for the marketing authorisation of Holoclar included:

- Translation of clinical experience into ICH-Good Clinical Practice (GCP) format. Holoclar was approved on the basis of two retrospective, multicentre, case series based, non-randomised and uncontrolled observational trials, HLSTM01 and HLSTM02 (Table 3.1), and long term follow-up. By 2007, 219 patients were treated in 21 different centres in Italy and 82 patients were not included in the two retrospective studies because centres declined to release patient data. Registered clinical data, availability of photographic records of patient progress and follow-up analysis made it possible to apply ICH- GCP principles to retrospective data for regulatory review. The EMA Scientific Advice Working Party (SAWP) supported in generating a prospective protocol to capture and evaluate all retrospective data.

Table 3.1 Holoclar in retrospective Good Clinical Practice studies

Trial Name	Number of Patients	Study Features
HLSTM01	106	▪ Two surgical centres ▪ Homogeneous eligibility criteria ▪ Common surgical protocol, post-transplantation treatment and follow-up ▪ Efficacy evaluation 1 year post-implant ▪ Availability of eye pictures ▪ High data consistency
HLSTM02	29	▪ Seven surgical centres ▪ Common surgical protocol ▪ Heterogeneous eligibility criteria, post-transplantation treatment and follow up, efficacy assessment ▪ No eye pictures ▪ Appropriate data integrity

Adapted from European Medicines Agency. EMA/25273/2015 Holoclar Assessment report [Online]. London: EMA; 2015 [cited 2017 May 16]. Available from: URL: http://www.ema.europa.eu/docs/en_GB/document_library/EPAR_-_Public_assessment_report/human/002450/WC500183405.pdf

- Comparability between the old and new manufacturing process in line with GMP requirements stipulated by the ATMP regulation[2], such as transmissible Spongiform Encephalopathies (TSE) and European Pharmacopoeia compliant raw material, setting in-process controls and validating analytical methods.

[2] European Commission. Regulation (EC) 1394/2007 of the European Parliament and of the Council of 13 November 2007 on advanced therapy medicinal products and amending Directive 2001/83/EC and Regulation (EC) 726/2004 [Online]. Official Journal of the European Union 2007; L324:121-137 [cited 2017 May 28]. Available from: URL: http://eur-lex.europa.eu/legal content/EN/TXT/PDF/?uri=CELEX:32007R1394&from=EN

- Defining therapeutic indications and clinical end points. The primary endpoint was the presence of a stable corneal epithelium without significant recurrence of corneal neovascularisation at 12 months after Holoclar transplantation. Secondary endpoints included ocular symptoms, inflammation, visual acuity improvement and keratoplasty success. The slit lamp examination and impression cytology were tools used to determine the clinical endpoint.
- Demonstrating comparability and quality consistency of pre- and post-change products. p63 transcription factor is used as a potency marker, where the release specifications of Holoclar require a specified amount of p63 transcription factor for clinical success.
- Demonstrating that changes do not have an adverse effect on the quality, safety and efficacy of the product.
- Retention of testing samples from the starting material or from manufacturing intermediates is limited due to limited number of cells isolated from the small biopsy.

The joint collaboration between the University of Modena and Reggio Emilia, which had a scientifically and clinically proven stem cell therapy, and the industrial partner Chiesi Farmaceutici, with a European commercial infrastructure and the regulatory know-how to provide adequate level of GMP standards, facilitated the approval of Holoclar. The marketing authorisation of Holoclar was granted via the conditional approval procedure, where it was requested to conduct a multinational, multicentre, prospective, open-label, uncontrolled interventional study to assess the efficacy and safety of the stem cell therapy. The prospective trial as part of the conditional marketing

authorisation is another challenge since it takes years to collect clinical data from patients with a rare disease such as limbal stem cell deficiency.

3.2.3 Case studies with technical specialists

Technical specialists in the field of advanced therapies have experience in addressing scientific, organisational and technical challenges in the development of stem cell therapy projects. The following two case studies address potential areas of collaboration with technical specialists from the engineering phase of the project to identifying specialising areas in the clinical application of stem cells.

3.2.3.1 Potential areas of collaboration

A multidisciplinary team is essential when setting up a stem cell therapy unit. Collaborations with technical specialists in the area of engineering, validation, quality, product development and IT compliance facilitate realisation of the project in Malta (Figure 3.2). Through collaborations with CTP System, it was identified that conducting a feasibility study is the first step to support potential entrepreneurs taking an investment decision for the project. The feasibility study incorporates the design and layout of the facility, equipment required for the project, preliminary process analysis, personnel needs, estimation of the costs involved and estimated time to finish the project.

Engineering	Validation	Process and Quality	IT Compliance
• Feasibility studies • Project management • GMP review and risk assessment • Design of the facility • Building supervision • Equipment and critical system supplier selection	• Validation master plan preparation • Design, installation, operational and performance qualification of facility, systems and equipment • Metrology and calibration	• Support in regulatory sciences to obtain GMP license of the facility and marketing authorisation of the product • Design and implementation of the pharmaceutical quality system including drafting of SOPs • Quality assurance and quality control support • Personnel training • Stem cell therapy process development and validation • Technology transfer	• GMP IT assessment and review • Feasibility for IT projects • Support for IT infrastructure management and IT networks qualification • Provision of quality integrated IT systems • Software and supplier selection • Computer system validation • Audits to systems supplier • Training

Figure 3.2 Potential areas of collaboration with technical specialists[47]

[47] The case of CTP System

3.2.3.2 Clinical application of stem cells in aesthetic therapy

Adipose-derived stem cells are attractive candidates for regenerative therapies. Stem cells are now used in cosmetic procedures such as skin anti-aging therapies (McArdle et al, 2014). CTP System suggested that Malta is an excellent destination for medical tourism and investing in dermatological and aesthetic stem cell therapies is an attractive business investment, since these therapies takes a short time to market, there is fast return of investment, good market potential, lack of competition and strong potential partnership with aesthetic and plastic surgery clinics (Figure 3.3).

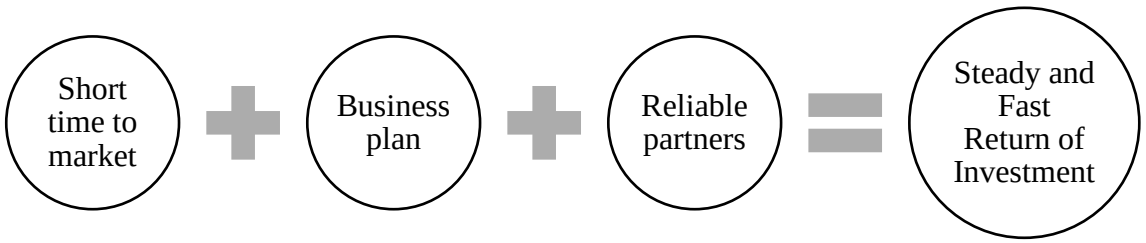


Figure 3.3 Investment in aesthetic stem cell therapy

3.2.4 Case studies through the European medicines regulatory network

The critical scientific reviews of the following case studies were identified and assessed during the inspections with HPRA in Ireland at a stem cell therapy facility (CCMI) and a blood establishment (IBTS). The case studies highlight the regulatory scientific approach by the facilities and the regulators to ensure the quality, safety and efficacy of stem cell-based medicinal products and blood components. Case studies reviewed include manufacturing process of stem cell therapies for clinical trials, challenges in the quality control testing of stem cell therapies, aseptic process validation, supply chain

control, handling of emerging infections to ensure safe blood supply, and management of the quality management system such as handling of deviations, complaints, and recalls.

3.2.4.1 Clinical application of stem cells in critical limb ischaemia

Manufacturing sites involved in the preparation of medicinal products for use as investigational medicinal products (IMPs) are inspected by national competent authorities to monitor compliance with GMP requirements provided for in the clinical trial authorisation.[5,6] In 2014, HPRA approved the conduct of a Phase Ib, open label, uncontrolled, non-randomised dose-escalation Critical Limb Ischaemia (CLI) Trial. The aim of the clinical trial is to examine the safety of intramuscular autologous transplantation of mesenchymal stem cells ($2x10^6$ cells/mL) into patients with no option critical limb ischaemia. Manufacturing of the stem cell therapy is carried out at CCMI and entails the isolation, expansion and cryopreservation of hMSC (Figure 3.4).

[5] European Commission. Regulation (EU) No 536/2014 of the European Parliament and of the Council of 16 April 2014 on clinical trials on medicinal products for human use, and repealing Directive 2001/20/EC [Online]. Official Journal of the European Union 2014; 158:1–75 [cited 2017 May 28]. Available from: URL: http://ec.europa.eu/health//sites/health/files/files/eudralex/vol-1/reg_2014_536/reg_2014_536_en.pdf

[6] European Commission. Consultation Document – Draft Guidelines on Good Manufacturing Practice for Advanced Therapy Medicinal Products[Online]. Brussels: EC; 2016 [cited 2017 May 28]. Available from: URL: http://ec.europa.eu/health//sites/health/files/files/advtherapies/2016_06_pc/2016_06_draft_guideline.pdf

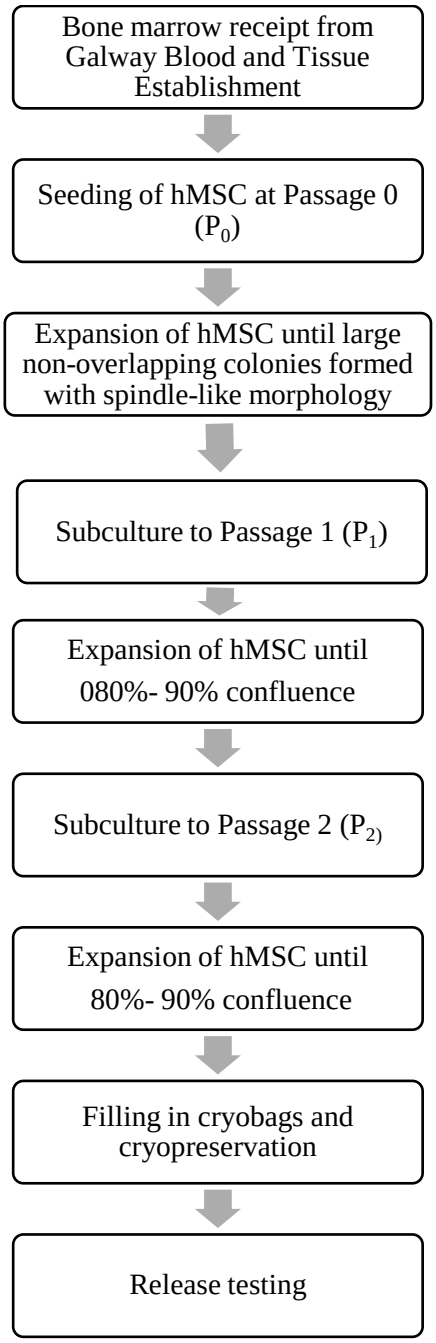


Figure 3.4 Manufacturing process for the Critical Limb Ischaemia Trial

Quality control tests including bone marrow testing, in-process controls testing and release testing for the manufacturing of hMSCs in CLI clinical trial are outlined in Table 3.2.

Table 3.2 Quality control testing for the manufacturing of hMSC

Quality control measures	Tests	Acceptance criteria
Bone marrow testing	HIV-1 antibody	Negative
	HIV-2 antibody	
	Hepatitis A antibody	
	Hepatitis B surface antigen	
	Hepatitis B core antibodies	
	Hepatitis C core antigen antibody	
	Anti-cytomegalovirus antibody IgG	
	Syphilis	
	Human T-lymphotropic virus 1	
	Sterility	
In-process controls	Colony size	Process stopped if colonies are not formed by Day 21 of P_0
	Cell count	Variable
	Fibroblastic Colony Forming Unit (CFU-F)	Process stopped if sufficient confluence is not observed by Day 7 of P_1
	Viability	≥70%
	Sterility	Negative
	Mycoplasma	Negative
	Endotoxin	≤ 5 EU/mL
Release testing	Sterility	Negative
	Mycoplasma – none detected	Negative
	Endotoxin	≤ 5 EU/mL
	Karyology	No chromosomal abnormality detected
	Viability	≥70%
	Immunophenotyping positive	≥90% positive and ≤5%negative markers

The results of the CFU-F assay are used to calculate the number of potential hMSC capable of being cultured in the bone marrow aspirate and this allows accurate assessment of the number of population doubling time of the cells. Population doublings, doubling time and percentage viability are calculated for each batch at every passage.

3.2.4.2 Clinical application of stem cells in osteoarthritis

ADIPOA-2 clinical trial is a phase IIb, multicentre prospective, randomised, double-blind study funded by the EU Horizon 2020 research funding program. The purpose of this clinical trial is to assess the safety and efficacy of a single injection of either $2x10^6$ or $10x10^6$ autologous adipose derived mesenchymal stromal cells in the treatment of mild to moderate osteoarthritis of the knee, active and unresponsive to conservative therapy for at least 12 months. This randomised clinical trial is open in 10 medical centres across Europe involving 150 patients. CCMI manufactures cell batches for centres in the UK and Ireland.[48, 49]

The defined roles of the tissue establishment, the clinical research facility and the manufacturing facility are reviewed through a protocol during an inspection. In the case of the ADIPOA-2 trial, lipo-aspirate active substance is manufactured from the patient adipose tissue. The adipose aspirate is managed by the Galway Blood and Tissue Establishment (GBTE) located at Galway University Hospital. The protocol involves the roles and responsibilities of the Clinical Research Facility (CRF), GBTE and CCMI.

[48] NUI Galway. Centre for Cell Manufacturing Ireland – Clinical Trials [Online]. Galway; 2017 [cited 2017 May 28]. Available from: URL: https://www.nuigalway.ie/stem-cells/clinical-trials/

[49] Regenerative Medicine Institute. ADIPOA-2 [Online]. Ireland: Galway; 2016 [cited 2017 May 28]. Available from: URL: http://www.remedi.ie/news/adipoa-2

The Responsible Person (RP) for GBTE has the overall responsibility for the procurement of lipo-aspirate, as well as the responsibility to report serious adverse reactions or serious adverse events to the relevant authorities and inform CCMI and CRF accordingly.

The manufacturing process of adipose-derived stem cells takes between 12 to 15 days. Cryopreservation of the product is not necessary since the shelf-life of the product is 24 hours. CCMI informs GBTE and CRF of culture and expansion progression, and provides a date closer to the time of availability of the release IMP for administration to the patient. CRF reserves the appropriate hospital space for administering the product to the patient (Table 3.3).

Table 3.3 Manufacturing process for ADIPOA-2 clinical trial

Timeline	**Manufacturing Process**
Day -1	Adipose tissue collection
Day 0	▪ Receipt control ▪ Specimen is usually processed immediately but can be stored between +2 and +8°C and stored for <24hours from time of aspiration ▪ Adipose tissue digestion, filtration and centrifugation ▪ Culture set up
Day 1	Washing and medium exchange
Day 4 ± 1	Medium exchange
Day 6 ± 1	Medium exchange
Day 8 ± 1	Cell harvesting, quality control and re-seeding
Day 11 ± 1	Medium exchange

Timeline	Manufacturing Process
Day 13 ± 1	Medium exchange
Day 12 – 15	Cell harvesting and quality control
Day 12 – 15	Cell packaging ⟶ Shipping to operating room

The IMP is QP certified and released in line with Clinical Trial Directive 2001/20/EC,[4] and transported to the designated research pharmacist at the clinical research facility in validated cell safe box or similar to ensure cold chain storage. At this point, the product would be under the responsibility of the Principal Investigator (PI). A dispensing prescription is issued to the pharmacist by the physician. The IMP is dispensed to the patient who is waiting at the theatre. The IMP is administered by the consultant radiologist to maintain blinding of the PI and sub-investigators. Participants will be followed up the by the PI for 2 years with assessments conducted according to protocol.

3.2.4.3 Challenges in the quality control testing of stem cell-based medicinal products

The autologous adipose-derived stem cell-based medicinal product in the ADIPOA-2 trial has a shelf-life of 24 hours that limits the time of quality control testing before release from CCMI (Table 3.4). The sterility testing, mycoplasma and endotoxin tests will be available after release of the product, hence the, product is released based on the negative results at Day 8. Additional batch test results are available after release of the product (Table 3.5).

[4] European Commission. Directive 2001/20/EC of the European Parliament and of the Council of 4 April 2001 on the approximation of the laws, regulations and administrative provisions of the Member States relating to the implementation of good clinical practice in the conduct of clinical trials on medicinal products for human use [Online]. Official Journal of the European Union. 2001; 21: 34–44 [cited 2017 May 28]. Available from: URL: http://eur-lex.europa.eu/LexUriServ/LexUriServ.do?uri=OJ:L:2001:121:0034:0044:en:PDF

Table 3.4 Tests specified in the Certificate of Analysis for the adipose stem cells finished product

Test	Time points	Acceptance criteria
Cell Count	Day 12-15	Dose $2x10^6$ Dose $10x10^6$
Viability[a]	Day 12-15	≥90%
Immunophenotype (using Flow cytometry)	Day 12-15	CD73+, CD90+, CD105+ ≥90% CD45+ <5% CD14+ <5% CD34+ ≤10%
Microbial sterility	Day 0 Day 8 ± 1	Negative
Mycoplasma	Day 0 Day 8 ± 1	Negative
Endotoxin	Day 8 ± 1	≤ 1.0EU/ml
Visual Inspection	Day 12 - 15	Volume of filling; Integrity of the primary container; Inspection against lighted white and black background for evidence of visible particulates or other foreign matter; Presence of combi-stopper; No damage of the syringe; Correct labelling of product

[a] A sample is stained with trypan blue, a blue dye that is excluded from viable cells and absorbed by non-viable cells allowing their enumeration. Percentage viability is calculated

During the review of the report on the three validation batches for the ADIPOA-2 clinical trial, one of the validation batches had viability values out of specification. Viability of the cells is reduced with high confluency. High confluence results in cells detaching from the surface of the culture chamber, hence losing their viability. This is more commonly seen in clinical trials involving healthy individuals and which must be

taken into consideration in trials involving diseased patients. The viability issue was resolved by daily confluency testing and harvesting the cells earlier than day 15, hence allowing harvesting between day 12 and 15 of the procedure depending on confluency testing results. Population doubling, doubling time and percentage viability is calculated for each batch at every passage.

Table 3.5 Additional batch test results (available after release)

Test	Time Point	Acceptance criteria
Microbial sterility	Day 12- 15	Negative
CFU-F		Variable
Mycoplasma		Negative
Endotoxin		≤ 1.0EU/mL
Human telomerase reverse transcriptase (hTERT)		Negative
Maximum population double		≤16
Maximum doubling time		≤4 days

3.2.4.4 Challenges with batch sizes and variability of the starting material

CCMI received a complaint from the clinical trial facility that a batch as part of the CLI trial had low volume of medicinal product to be reconstituted and administered to the patient. The clinical trial protocol states that the final product volume should be 12mL and the patient should be treated with 10mL of 2 x10^6 cells/mL bone marrow derived mesenchymal stem cells. The complainant reported that recovery on reconstitution did not meet the requirements in the protocol, and 6mL of product was filled for administration to the patient. The root cause of the low cell yield was attributed to donor variability. A deviation was carried out to maximise the fill volume of the batch. The

batch manufacturing record (BMR) includes the ‘total viable cell number’ to be used when calculating the required volume of freezing media, in the case of this batch the ‘total cell number’ was used. The planned deviation was captured through a change control. A variation was submitted to HPRA for approval to update the clinical trial protocol, detailing that a volume of up to 100mL bone marrow aspirate as starting material is required to ensure that sufficient volume is available for QC sampling and for administration to the patient.

3.2.4.5 Deviation on the use of incorrect batch manufacturing record

During review of batch records, CCMI QP noted that the incorrect version for recording ‘Isolation, Counting and Seeding of bone marrow’ was used. QC results showed that the seeding density per flask was within the acceptable range at the time of processing. The root cause for the deviation was due to internet connectivity problems inside the cleanrooms and the latest version of the BMR is saved on a memory stick, which is transferred to the production areas in a zip sterile pocket. The BMR is printed in the production area from a printer in a Grade B area. The deviation required a risk-assessment to ensure that the latest version of BMRs is saved on the memory stick, which should be controlled by the quality unit.

3.2.4.6 Aseptic gowning within classified environments

Aseptic gowning is essential to contamination control in sterile manufacturing facilities. Correct aseptic gowning technique may be assessed as part of the environmental monitoring programme through microbial sampling using contact plates.[50] The gowning procedure observed at the CCMI inspection indicates areas that may require particular attention during the training process, including:

- Aseptic procedure for placing the hood over the bouffant cap using the inner thumb loops and fastening the poppers at the front and back of the hood.
- Procedure for placing the sterile mask over the mouth and nose, ensuring the glossy side faces outwards. Straps are to be tied behind the head and neck ensuring no part of the skin is exposed.
- Procedure to remove the suit from the packaging. The suit should be held high up from the collar and unzipped away from the body, ensuring that it does not touch the floor, walls or bench and worn with minimal contact on the outside of the suit. Poppers should be fastened at neck, cuffs and legs.
- Importance of checking in the mirror to ensure that no skin or hair is exposed, bouffant cap is not protruding from under the hood, and the hood and suit are properly fastened.

[50] Cleanroom Technology. Managing aseptic gowning within classified environments; 2015 [cited 2017 May 28]. Available from: URL: https://www.cleanroomtechnology.com/news/article_page/Managing_aseptic_gowning_within_classified_environments/107339

3.2.4.7 Supply chain control

CCMI uses heparin as an anticoagulant in the media exchange procedure. The sourcing of heparin was investigated during the inspection. Heparin is supplied by the University of Galway Hospital, however no agreements between CCMI and the hospital to ensure supply chain compliance and integrity are in place. Agreements with the suppliers of these materials should be in place to ensure that if materials, such as heparin, are recalled, CCMI would be notified.

3.2.4.8 Performance qualification of sterility testing equipment

BD BACTEC™ FX 40 sterility testing equipment was recently installed in the microbiological laboratory at CCMI. Performance qualification of the BACTEC™ system was carried out in accordance with the 7th Edition of the European Pharmacopoeia (EDQM, 2010), where reference strains are specified by the pharmacopoeial method. The European Pharmacopoeia states that there may be the need "to modify the list of micro-organisms depending on the origin of the cells and any micro-organisms previously found or associated with the particular type of cells" (EDQM, 2010).

Fangoldia magma is an organism found in the hospital where stem cells are sourced, and CCMI are carrying out a risk assessment on whether to include the sterility test for this organism in the protocol. The sterility testing protocol should include the decision-making process and appropriate justifications for the inclusion or exclusion of microorganisms to be detected by the methodology of the test.

3.2.4.9 Environmental Monitoring

Environmental monitoring should be performed post-breach, 'in operation' and 'at rest'.[6] The environmental monitoring program at CCMI was reviewed during the inspection. The program does not define the frequency of environmental monitoring 'at rest' where it was recommended that the sampling plan for monitoring should be risk-based. The approach to environmental monitoring should be one of proactively identifying ways to reduce contamination, rather than identifying ways how to detect contamination. Environmental monitoring was recommended to be carried out every two weeks to evaluate the actual process and to understand microbial contamination risks. Frequent environmental monitoring program results in more confidence in the manufacturing process and ensures that risk mitigations to help prevent contaminations are in place.

3.2.4.10 Media fill test

Media fill test, also referred to as process simulation, is part of aseptic processing validation. The test is carried out using a sterile microbial growth medium to test whether the aseptic procedure is adequate to prevent contamination during production.[6] The media fill is used to evaluate the aseptic assembly and operation of the critical equipment, qualify the operators, assess their technique and demonstrate that the environmental controls are adequate to meet the basic requirements to produce sterile hMSCs by aseptic processing.[51]

[6] European Commission. Consultation Document – Draft Guidelines on Good Manufacturing Practice for Advanced Therapy Medicinal Products[Online]. Brussels: EC; 2016 [cited 2017 May 28]. Available from: URL: http://ec.europa.eu/health//sites/health/files/files/advtherapies/2016_06_pc/2016_06_draft_guideline.pdf

[51] European Commission. EudraLex Volume 4 EU Guidelines to Good Manufacturing Practice Medicinal Products for Human and Veterinary Use: Annex 1 Manufacture of Sterile Medicinal Products [Online].

At CCMI process simulations are carried out twice a year per process and all personnel authorised to enter the aseptic processing area during manufacture should participate in the media fills, at least every 6 months. The negative control in the test is an unopened bottle of media incubated in the CO_2 incubator from Day 1 until the end of the process. Process simulation is successful if all cryobags are free from turbidity, as compared to the negative control. The negative control must be free from contamination and this is supported by submitting the negative control to the contract testing laboratory and growth promoted against certain bacteria to show that the media is capable of supporting growth of bacteria.

The media fill process test is time-consuming (around 10 days) and deplete materials and resources. A risk-based approach when conducting this test was recommended, by assessing the critical "worst-case" steps in the manufacturing process that are at greater risk of microbial contamination. The media fill test method for a particular product should be designed according to the risk analysis and testing may be carried out on the critical time points of the process.

3.2.4.11 Failure to detect donors with low haemoglobin levels

In July 2014, the Irish Blood Transfusion Service (IBTS) changed the point-of-care testing from HaemoCue to Haemospect device to determine pre-donation haemoglobin levels. Donors are deferred if the haemoglobin level is <13.5g/dL for males and <12.5g/dL for females. The point-of-care device Haemospect allows non-invasive measurement of blood haemoglobin levels. On the 1st October 2014, a donor donated blood and fifteen days later the patient was hospitalised and received a blood

Brussels; 2008 [cited 2017 May 28]. Available from: URL: http://ec.europa.eu/health//sites/health/files/files/eudralex/vol-4/2008_11_25_gmp-an1_en.pdf

transfusion. The donor had severe iron deficiency anaemia with a haemoglobin level of 7.1g/dl. The Haemospect reading at donation was 13.9g/dl. The red cell concentrate (RCC) had been discarded as it was underweight (<231mL). It was reported that the donor felt faint at the clinic. Three other similar donor incidents were reported following this case.

These cases prompted a series of actions by the blood establishment to resolve the issue (Appendix 7). The serious adverse events were reported to HPRA, where a rapid alert was issued and a dedicated inspection was performed in November 2015. Hundreds of donors were followed up as a result of this incident. Donor information sources were:

- Review of incident reports, product complaints, service complaints, product non-conformances that may be linked to problems with the Haemospect device
- Full blood count results taken from donors as a result of implemented actions
- Donors or relatives who have contacted IBTS

Not all donor information is relevant or required medical follow-up. A database was created by the Medical Department to capture all relevant donors requiring follow-up. A total of 244 donor files were created and donors with evidence of serious outcomes (transfused as a result of anaemia) were entered in the donor vigilance database by completing the incident form. The data formed part of the annual donor vigilance submission to the HPRA.

The procedure at the time of inspection was that donors whose capillary haemoglobin is outside the threshold venous haemoglobin range will have to undergo venous haemoglobin testing (Table 3.6).

Table 3.6 Threshold venous haemoglobin values used to assess acceptability or deferral of donor

Male donor venous haemoglobin level (g/dl)	Female donor venous haemoglobin level (g/dl)	Action
≥18.0	≥16.5	Defer donor and refer to doctor
≥13.5 - ≤17.9	≥12.5 - ≤ 16.4	Eligible
≥13.0 - ≤13.4	≥12.0 - ≤ 12.4	Defer donor for 3 months
≥10.0 - ≤12.9	≥11.5 - ≤11.9	Defer donor for 6 months
≤9.9		Defer donor and refer to GP

The Haemospect case identified areas to be monitored and assessed during blood establishment inspections (Figure 3.5).

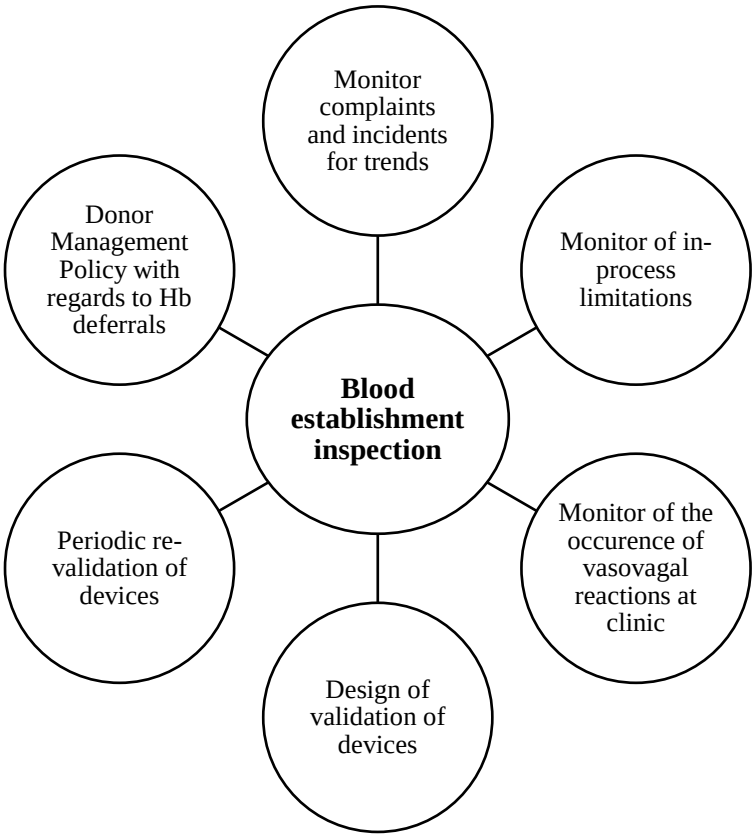


Figure 3.5 Areas to monitor and assess during a blood establishment inspection following Haemospect case

3.2.4.12 Risk mitigation procedures following incident report

During blood donation, four blood samples are taken for virology testing, nucleic acid testing, donor grouping and for archiving purposes. The blood group of repeat donors is checked and verified with records from previous donations and a ‘confirmed group’ label is affixed on the blood pack. The ‘confirmed group’ label is a risk reduction measure that does not detect wrong blood group in bag.

IBTS received correspondence from hospitals querying the reason for only receiving red cell units with ‘confirmed group’ status on labels. This is unusual as hospitals would expect to receive a number of units (up to 10-15%) without this status on the label. The correspondence from hospitals prompted a check of units in the inventory at IBTS which revealed that all units contained ‘confirmed group’ status on labels. Selected units were checked on eProgresa (IT system). First time donations that contained ‘confirmed group’ status on labels were identified. An incident report was raised and the IT department discovered that the parameter controlling the printing of the label with ‘confirmed group’ was incorrect. The feared clinical outcome was that the blood in the unit would be of a different group to the blood in the pilot tube, leading to an ABO incompatible transfusion. It is critical that the unit of blood donated by a donor and the four blood samples in pilot tubes taken from the sample pouch on needle insertion are from the same donor and are labelled with identical donation numbers to prevent a situation where there is an incorrect ABO group label being applied to the red cell unit. During the inspection, the procedure in place to ensure that the donated unit and donor samples are from the same donor was reviewed.

IBTS has a policy to check donor grouping of all new donors to Rh phenotype and Kell type (C, c, E, e and K). Confirming the group of a new donor on the second donation would not always detect a 'wrong donor in tube' since quite a large proportion of donors tested and confirmed would be group 'O RhD positive'. However, knowing the Rh phenotype from the first typing would ensure that on the second typing, rare Rh phenotypes would be selectively taken out and confirmed. This gives a high level of confidence in the system.

The virology laboratory at IBTS tests all blood donations for the presence of HIV-1/2 antigen/antibodies, Hepatitis C antibody, HTLV-I/II antibody, Hepatitis B core antibodies, Hepatitis B antigen and antibody to Treponema Pallidum (syphilis). A sample is taken from the segment line of the whole blood or plasma pack of repeat reactive samples for testing in duplicate to ensure that no discrepancy exists between the donor tube and the donor blood pack. An investigation is instigated if a discrepancy is detected such as samples tests positive for a virology marker and negative from the sample pack.

To mitigate against the possibility of such an occurrence, the following clinical procedures are documented in SOPs:

- **Handling of donation numbers:** Donors are issued with donation numbers at registration and ensured that each label is accounted for by the end of the donation, including unused or spare donation numbers which should be securely affixed on the Health and Lifestyle Questionnaire (HLQ) if the donor is deferred or samples are taken without a donation.

- **Clearing of the workstation** before each donor is brought to the donation bed and after the donor is escorted from the donation bed. Staff member must ensure that HLQ, blood pack and sample tubes including all donation numbers of the previous donor, have been removed from the donation bedside.
- **Identifying the donor:** Checking that donor's name and date of birth are the same as on the HLQ and asking donor if the signature on the HLQ is his/her signature.
- **Labelling of donations and sample tubes:** Staff member only moves to the next donor when all labelling is completed according to the procedure.
- **Verification of correct labelling** of the HLQ, sample pilot tubes and the main blood pack is carried out before the donor leaves the donation bed. The HLQ, donation and pilot tubes are placed in a transit tray.
- **Blood tray check:** Placing of labelled packs in cooler tray and pilot tubes in numerical order in the pilot tube holders. Reconciliation at the end of the clinic to ensure the correct number of pilot tubes is present and initials the relevant form confirming that s/he has done so.

To date, there have been no incidents of mismatching between blood in the pilot tubes and blood in the packs. The National Haemovigilance Office has not received any reports of acute haemolytic transfusion reactions.

3.2.4.13 Selection of red cell components for cross-match

IBTS received a complaint were an adult red cell was issued to an infant <12 months. According to the transfusion protocol the red cell unit should be C-/K-/CMV- irradiated. The result was that the unit met all neonatal criteria. The electronic system will stop non-CMV negative and non-K negative units from being issued to a patient < 12 months.

It was recommended to update the 'IBTS Guideline for the selection of suitable red cell components for cross-match for adults, neonates and infants' to include:

- Requirements for neonates and infants
- Different red cell products available and their uses
- List of conditions where patients become transfusion dependent and the requirements to phenotype

The changes will differentiate and clarify transfusion requirements for the different categories of patients.

3.2.4.14 Personnel training

The Medical Officer at the blood establishment received a complaint from a donor who developed shingles a few days post-donation, where similar complaints trigger a recall. The Quality Control Environmental Monitoring Unit responsible for handling blood recalls were not informed about the recall until the next day. An incident report was carried out as a result of delay in recall. The red cell unit had already been transfused to the patient.

Preventive and corrective action included retraining of the concerned personnel on the complaint procedure. A temporary change control was carried out to revise the process for product recalls, where all product recalls are handled at the National Blood Centre. Recalls are reported to the Quality Control Environmental Monitoring unit, Monday to Friday between 07:00 and 19:00 or the Component Department Monday to Friday between 19:00 and 07:00, weekends and public holidays. It was recommended to close the temporary change control, train staff on the new times and streamline the recall system according to the change control.

During the inspection, the organisation chart and evidence of various job descriptions were requested to ensure that individual responsibilities are clearly recorded and defined within the organisation. Training records were provided as evidence that personnel receive training and that training needs are identified and followed-up, including evidence of training as a corrective action and preventive action (CAPA) following an incident report which was due to non-conformance with a procedure.

3.2.4.15 Trend analysis

While reviewing the index list of incident reports, it was observed that blood tube heat-sealing error non-conformance had repeatedly occurred in 2016. Further investigations were carried out and a trend analysis report was requested. The target for venepuncture technical unusable is 2%, while the target for non-venepuncture technical unusable is 0.5%. Two IBTS blood collection sites were exceeding the limits. Analysis in terms of equipment used, staffing levels, donor flow, technique used by personnel and training was carried out. It was observed that this non-conformance occurred mostly in February and June 2016 during specific weeks which correlates with an increase in donor flow.

Contributing factors to this error could be an increase in donor flow, times of significant staff shortage and level of competency to cover the whole blood and apheresis program. The quality management system internal audit program was requested with the aim to review the audit related to donation collection activities. This observation was left pending to the next inspection of the questioned collection sites.

3.2.4.16 Donor interview

In 2016, IBTS prepared a change control to amend the requirements for extended donor interview from one who has donated in the last two years to one who has donated in the last five years. The risk assessment for this change was reviewed and the blood establishment can benefit by:

- Increasing pool of donors possibly suitable for donating blood for neonatal use
- Decreasing the burden on nursing staff and use resources to manage donor clinic more effectively

3.2.4.17 Emerging infections – Zika virus case

Zika virus can be transmitted to patients through blood transfusion, however no transmission cases have been reported. Blood establishments and regulatory authorities should be vigilant of the risk of donor-derived Zika virus transmission, especially since it is known that Zika virus infection remains asymptomatic in 80% of cases. Blood

establishments should update donor information materials to include information on Zika virus infection and ways how the infection can be transmitted.[52]

The IBTS HLQ (Appendix 8) included the following questions related to Zika virus transmission:

1. In the past four weeks have you:
    a. Had sex with anyone who has ever had Zika Virus Infection – with or without protection?
    b. Been in contact with an infectious disease?
2. Have you been outside Ireland or the UK in the past 12 months for any reason?

In Ireland, donors who have visited possible Zika-infected regions are deferred for three months and donors diagnosed with the disease are deferred for six months. A major deficiency was observed in the HLQ, where donors who had sexual contact with persons who travelled or lived in Zika-affected areas, during the three months prior to donation were not captured. This information should be included in the HLQ in line with the scientific advice published by the European Centre for Disease Prevention and Control, 2016.[52]

[52] European Centre for Disease Prevention and Control. Scientific advice on Zika virus and safety of substances of human origin: A guide for preparedness activities in Europe. [Online]. Stockholm: ECDC; 2016 [cited 2017 May 28]. Available from: URL: http://ecdc.europa.eu/en/publications/Publications/Zika-virus-safety-of-substances-of-human-origin.pdf

[52] European Centre for Disease Prevention and Control. Scientific advice on Zika virus and safety of substances of human origin: A guide for preparedness activities in Europe. [Online]. Stockholm: ECDC; 2016 [cited 2017 May 28]. Available from: URL: http://ecdc.europa.eu/en/publications/Publications/Zika-virus-safety-of-substances-of-human-origin.pdf

3.3 The network of collaborators for the Malta Stem Cell Project

The multisite collaborations with different European institutions offered several compelling advantages for this research project for sharing experiences and best practices, and cooperating and networking with local public institutions (Appendix 9) and potential investors for the establishment of a stem cell therapy unit in Malta. King's College London and CTP System came to Malta to discuss the project with potential investors and public institutions including the University of Malta (UOM), Malta Enterprise (ME), Malta Life Sciences Park (MLSP), National Blood Transfusion Service (NBTS), Malta Medicines Authority (MMA) and the Malta Laboratories Network (MLN). Networking meetings (Table 3.7) were coordinated by the MMA and MLN to establish a local stem cell therapy unit. These meetings were followed by teleconferences and email correspondence for realisation of the regulatory science collaboration network for the stem cell project in Malta (Figure 3.6).

Table 3.7 Networking meetings with local institutions

Date	Participants	Outcome
21/12/2015	Dr Ragai Mitry (King's College) Dr Alex Aquilina (NBTS) Alison Attard (MMA/researcher)	Ministry of Health through NBTS are seeking EU Funding for the provision of a new Innovative Centre of Excellence for Cells, Blood and Tissues. This centre will include the blood establishment, banking and manipulation of cells and tissue banking. King's College and NBTS discussed how the technical expertise at King's College can support this national project which is a requisite for the approval of EU funding.
22/12/2015	Professor Godfrey LaFerla (UOM) Professor Richard Muscat (UOM) Dr Ragai Mitry (King's College) Professor Anthony Serracino Inglott (UOM/MMA/MLN) Alison Attard (MMA/researcher) Potential Private Investor	The UOM offered the new laboratory facilities at the Biomedical Sciences Building for the research part of this project, while King's College proposed to collaborate in the training of students by establishing post-graduate training programmes on the technical aspects of working in the field of stem cells and tissue banking.
20/01/2016	Dr Ragai Mitry (King's College) Professor Christian Scerri (UOM) Alison Attard (MMA/researcher)	Review of the available laboratories at the Biomedical Sciences Building at UOM to identify gaps in the infrastructure and equipment to establish a stem cell therapy facility and whether these facilities should be used only for stem cell research or if GMP standards for the production of cell therapy could be met.

Date	Participants	Outcome
15/03/2016	Giulia Dini (CTP) Riccardo Salvagnini (CTP) Peter Meli (ME) John Buttigieg (MLSP) Potential Private Investor Professor Anthony Serracino Inglott (UOM/MMA/MLN) Alison Attard (MMA/researcher)	During this preliminary meeting, CTP discussed how they can collaborate in this project by offering their technical services and support in terms of setting up of a stem cell therapy facility in Malta. A visit to CTP's headquarters in Tuscany was planned to discuss this collaboration with their technical experts. ME and LSP discussed how the state-of-the-art facilities at the LSP were purposely built to accommodate such life sciences projects.

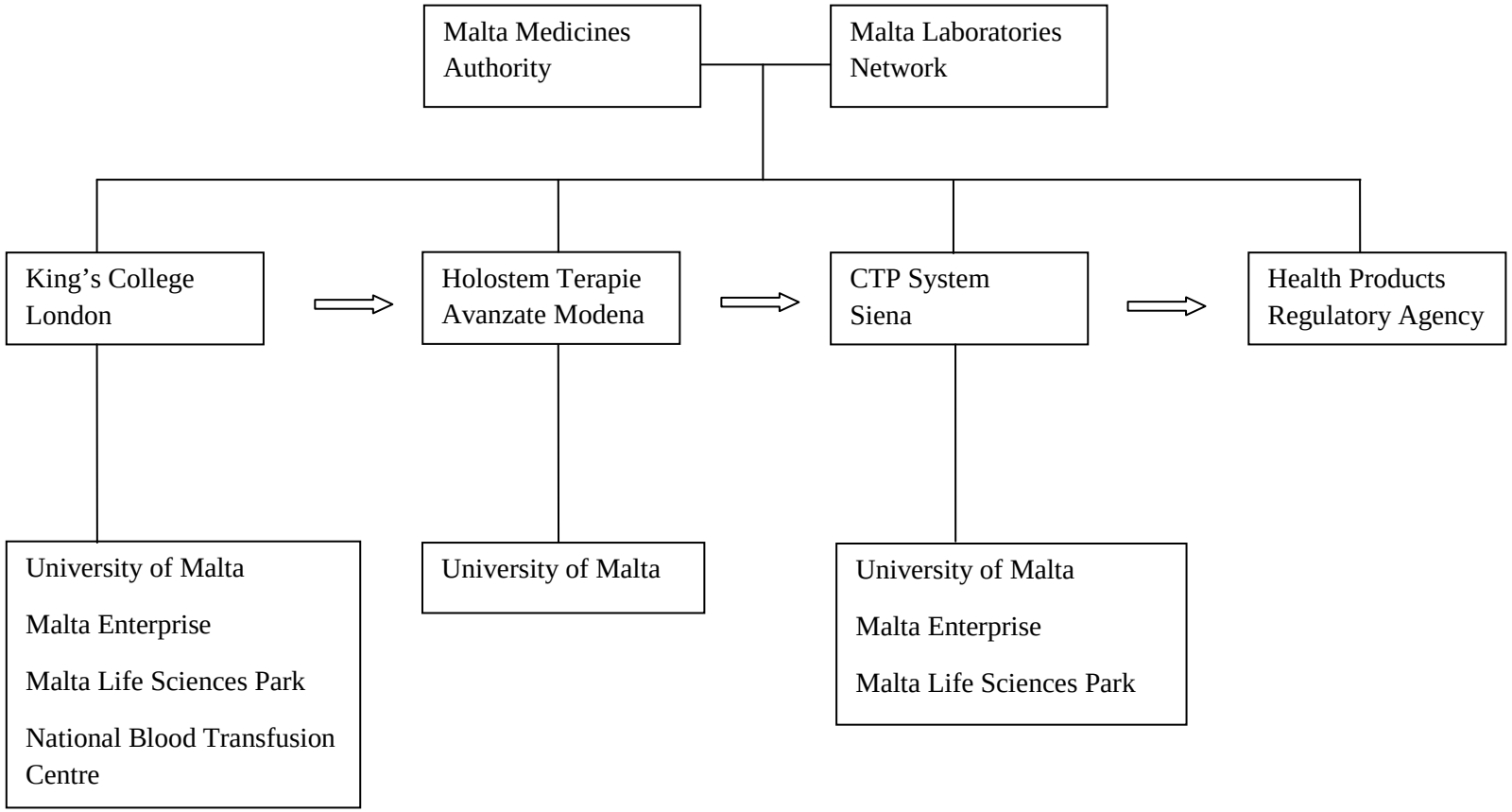


Figure 3.6 Structure of the regulatory science collaboration network for the stem cell project in Malta

3.4 Potential investors

Between 2015 and 2016, two companies approached the MMA with interest to invest in the promising field of stem cell therapy. The first potential investor has a well-established company in Malta not related to the pharmaceutical industry. The company wanted to expand their activities in innovative areas that can be tangibly utilised in the health sector for the benefit of patients. The stem cell therapy project met the desire of the local investor who was willing to invest one million Euro. The potential investor had several meetings with experts from King's College and other local public entities to discuss product development, targeted markets, time market entry and the required investment for this project taking into consideration realistic costs for the early development, GMP requirements and regulatory procedures.

In December 2016, a local pharmaceutical distributor, Cherubino Limited, approached the MMA expressing interest to embark on a project to set up a stem cell therapy unit in Malta. The project was being proposed in collaboration with Spanish foreign investors with established state-of-the-art clinical analysis laboratories and a stem cell bank in Europe and internationally, including Cerba Internacional, APA Laboratoris Clinics, Analiza, Biovault and BIOCORD stem cell bank.

The foreign investors visited Malta on the 27 February 2017 and high level meetings were organised according to the agenda (Appendix 10) to interact and explore on how to collaborate to establish a stem cell therapy unit in Malta.

3.5 Identification of the project

The importance of correctly identifying the project from the very early stages was discussed with the technical specialists. Projects related to stem cell therapy need to be customised according to the type of client and specific requirements, including the specialisation area and site where the product is intended to be developed and marketed. Different business models which could be adapted to the Malta Stem Cell Project were identified (Table 3.8).

In the business model there are two important project actors; the investor, who can accommodate the stem cell therapy unit, and the know-partner or source, who has the knowledge of the production process. The investor and the source can interact in three different ways:

1. Investor obtains the process technology and the know-how including the licenses from the source
2. Investor and source create business in franchising
3. Investor and source create a new joint company, where the source transfers technology of products to the new company, including the licenses

Institutional projects have scientific value since they are more focused to meet unmet medical needs. Advantages of institutional models include the participation in multi-centred clinical trials, publication of scientific papers and provision of therapies under compassionate use programmes.

Table 3.8 Business models for stem cell therapy projects

Type of model	Business	Institutional	Hybrid
Project Actors	Private investor/s and 'know-how' partner	Public entity or foundation	Joint venture public/private
Interests	Forecast return of investment	Focused on clinical results	Setting a research centre to focus on product development
Examples of ojects	Cord blood and placenta stem cell banking Adipose-derived stem cell banking Dermatological aesthetic therapies	Pancreatic islet manipulation and stem cell transplantation Cell therapy for bone reconstruction Cell therapy for multiple sclerosis	Autologous transplantation of limbal corneal epithelium
Priority	Business Time-market entry Budget priority	Clinical	Research

Different business models suitable for the Malta project were discussed between the investors, technical specialists, academia, regulators and other local entities during the networking meetings. During the meetings, it was highlighted that in stem cell research, the quality of the source of cells as the raw material is crucial. The umbilical cord has a good source of multi-potent stem cells which can be banked and would be available for use when the need arises. Cord blood banks are currently not established in Malta. Malta Enterprise remarked that there were other private investors interested in establishing private family banks, where the umbilical cord blood (UCB) is stored for the benefit of the donor and family members, however, the feasibility studies showed that such projects may not be sustainable, mainly due to the small population of Malta.

The experience of the foreign investors with the public-private partnership between Biovault, one of the largest tissue banks in the UK, and the National Health Service (NHS) in UK, was identified as the appropriate business model for the Malta stem cell project. The implementation plan of the Malta project includes:

1. Setting up a centre for GMP cell manufacturing and UCB banking (Figure 3.7).
2. Establishing the procedure for cord blood banking, where the support of the Government is essential for the project to be cost-effective. The cord blood unit can be split into three sections, one part available for possible future use by the donor or family members, another part available for public donation and another part available for research purposes. It is proposed that the costs of collection are assumed by the Government. Costs can be recouped from the public share of the bank that stores the donated UCB from consenting parents. A searchable inventory needs to be maintained to facilitate the supply of stem cells to other hospitals, healthcare organisations or Universities which can be used for medical applications or for research purposes.
3. Initiating stem cell research in developing a stem cell medicinal product.

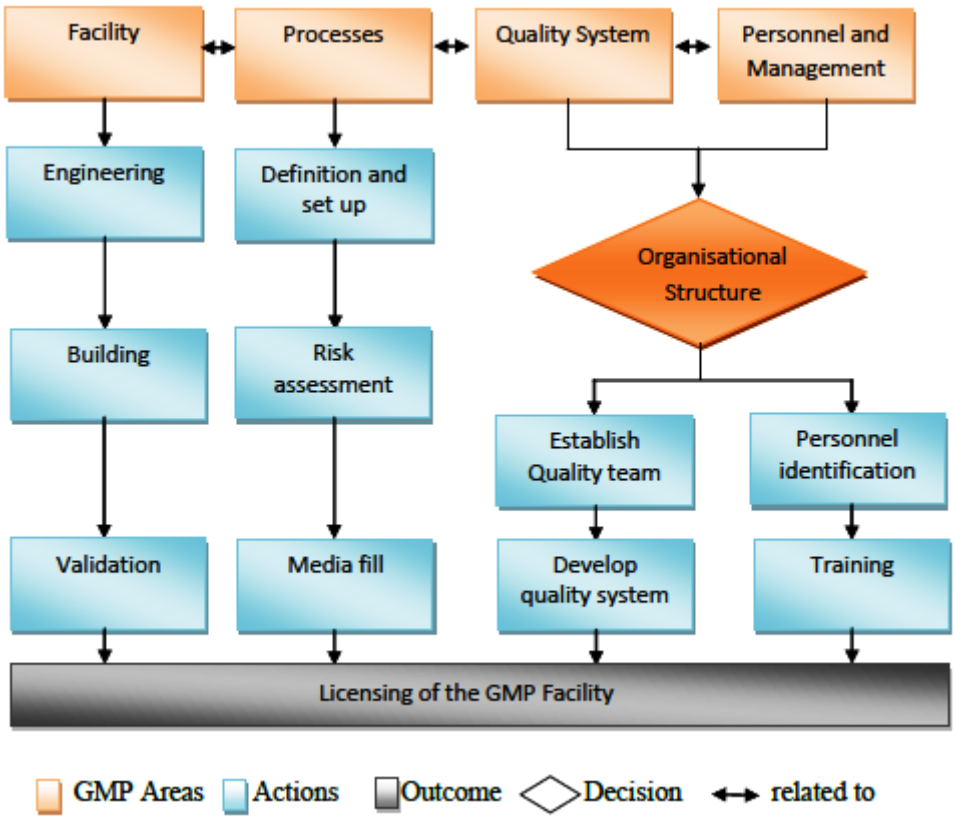


Figure 3.7 Proposed model for the GMP project

3.6 Facilities identified for the Malta stem cell therapy unit

Two facilities were considered for the setting up of a stem cell therapy unit:

1. UOM Biomedical Science Laboratories
2. Malta Life Sciences Park

There are two cell culture rooms available at the UOM Biomedical Sciences building, where the laboratory area has been designed to have positive differential pressure inside the processing area to ensure that air flows from the cleaner to the less clean space. Each room is equipped with stainless steel laboratory furniture, including two laminar flow cabinets as working stations in a clean environment. A sink and water supply for hand washing and cleaning are available in the laboratories.

For the purpose of the project, one cell culture room needs to be allocated for the stem cell research and another room for the quality control testing[53] (Appendix 11). In a separate area from the cell culture laboratories, the UOM Biomedical Sciences facility has a room containing liquid nitrogen tanks for storing the cryopreserved cells and is equipped with oxygen sensors that detect low oxygen levels.

Whilst the laboratories at the UOM Biomedical Sciences building can be used for pre-clinical studies, a purposely designed GMP cell manufacturing facility is required for cell isolation and preparation of clinical grade cells.[54] The cell culture laboratories at the UOM were not found to be fully compatible with GMP requirements for aseptic production due to:

1. Limited laboratory space to develop a defined manufacturing pathway, including product receipt area, quarantine area, storage area, quality control and other testing laboratories and aseptic area for cell manipulation and processing
2. Air supply is not filtered through HEPA filters
3. Lack of access control since the facilities are shared with other research groups that increases product safety risks such as cross contamination or product mix ups

Available laboratory spaces at the MLSP for the proposed project of establishing a cord blood bank and a stem cell therapy unit were identified (Appendix 11). The facilities at the MLSP are rentable. The laboratory space identified for this project are in the LS2 block which has two floors and 27 units ranging from $50m^2$ to $395m^2$ and the internal partioning between the units has been designed to allow flexibility according to the

[53] As recommended by King's College London

[54] As recommended by King's College London and University of Malta

needs of the tenants. The laboratory space required at the MLSP is around 500m^2 which does not need to be located on the same floor, where the liquid nitrogen tanks may be located at ground floor while the GMP processing areas can be set up on the first floor.[55] The laboratory spaces are finished as follows:

- Walls plastered with first paint coat
- High loading screed flooring ready for vinyl surfacing
- HVAC ducting complete with fire dampers and HEPA filters
- Fire rated doors leading in from corridors
- Fire detection units
- Water, electricity and chemical pipe drainage
- Data service points near the entrance to the unit
- External apertures are double-glazed and cannot be opened

The access control system at the MLSP using an electronic card-key system allows only authorised persons to have access to the facility. The secure basement of the MLSP can accommodate storage of gases including carbon dioxide and liquid nitrogen cylinders which can be piped directly to the laboratory and cryobank. MLSP has a generator for backup in case of failed power supply which is essential for the cord blood bank.

3.7 Requirements to develop the GMP stem cell therapy unit

The laboratory spaces at the MLSP were identified as adequate facilities for this project. Development of the cleanrooms, including environmental specifications (Table 3.9) was identified as one of the most important steps for a stem cell manufacturing facility.

[55] As recommended by the potential investors

Table 3.9 Specifications of the cleanroom facility[56]

Parameter	Cleanroom specification
Air exchange per hour	B class >60, C class >30, D class > 20
Temperature	18 - 22°C
Humidity	50 + 10 HR %
Material flow	Over-pressurised pass boxes
Equipment flow	Through an airlock
Control and Monitoring system	Centralised system with broadcast of critical alarms by means of a 24-hour alarm system phone availability. Local indicators for warning alarms to the operators.

The clean rooms and clean air devices are to be classified in accordance with ISO 14644-1 and Eudralex Volume 4 Annex 1 of GMP Manufacture of Sterile Products, both at rest and in operation conditions.[51] The Heating Ventilation and Air Conditioning (HVAC) system is a critical technology to control environmental microbial contamination, where the air supply is filtered through HEPA filters in line with the maximum permitted number of airborne particle concentration depending on the room classification (Table 3.10).

[51] European Commission. EudraLex Volume 4 EU Guidelines to Good Manufacturing Practice Medicinal Products for Human and Veterinary Use: Annex 1 Manufacture of Sterile Medicinal Products [Online]. Brussels; 2008 [cited 2017 May 28]. Available from: URL: http://ec.europa.eu/health//sites/health/files/files/eudralex/vol-4/2008_11_25_gmp-an1_en.pdf

[56] As identified at Holostem Terapie Avanzate

Table 3.10 Maximum permitted number of particles per m^3 equal to or greater than the tabulate size

Grade	Particle size			
	At rest		In operation	
	0.5µm	5.0µm	0.5µm	5.0µm
A	3 520	20	3 520	20
B	3 520	29	352 000	2 900
C	352 000	2 900	3 520 000	29 000
D	3 520 000	29 000	Not defined	Not defined

The GMP facility shall be designed to have two processing laboratories with Class A biosafety cabinets for aseptic preparation and filling of aseptically prepared products in a Class B background. The processing laboratories shall be accessed through two different sections with separate changing rooms to allow different procedures to be performed at the same time without risking cross-contamination.[57]

The aseptic gowning rooms and corridors of access shall be in increasing value of air cleanliness from Grade D to B. The area for preparing material to be passed to the production areas will be classified as Grade C, while the ventilated hatches should be of Grade B. Clean areas should be accessed through air lock with electronically-controlled interlocked doors to ensure that both doors are not opened simultaneously. The air supply in the laboratory spaces at the MLSP are maintained at a negative pressure relative to the outside, which needs to be modified to have a unidirectional flow maintained at a positive pressure. The positive pressure cascade will permit air to flow from areas of higher cleanliness (processing areas) to adjacent less clean areas with a pressure differential of 10-15Pa between different classified rooms (Figure 3.8). The

[57] As recommended by CTP system

HVAC system and equipment for storage of materials, such as liquid nitrogen tanks should be equipped with 24-hour alarm systems.

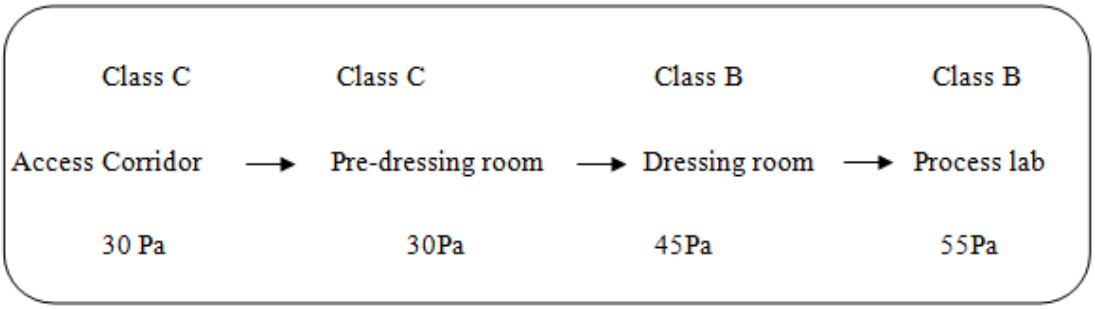


Figure 3.8 Pressure gradient in a cell culture room[58]

Other areas to be considered in the design of the facility include storage room facility, quarantine area, quality control/microbiological laboratory and administrative office for the quality unit. The interiors finishes of the facility shall include vinyl flooring, nonporous and washable ceiling and walls and stainless steel bench-tops.[59]

3.8 Identified equipment

A list of equipment for the setting up of cell processing laboratories, quality control laboratory and the cell bank was identified (Table 3.11). The two processing laboratories shall contain the same equipment.

[58] As recommended by Holostem Terapie Avanzate and CTP system

[59] As recommended by potential investors

Table 3.11 Identified equipment to set up a stem cell therapy unit[60]

Room	Equipment
Cell Processing laboratory	Class II biosafety cabinet
	CO_2 humidified incubator
	Refrigerated centrifuge (bench-top)
	Inverted microscope with camera
	Refrigerated microfuge
	4°C refrigerator
	-20°C freezer
	37°C water bath
	Vortex Stirrer
	Plate shaker
	Sterile tubing welder
	Tube sealer
	Peristaltic pump
	Micropipettes (P10/P20/P200)
	Pipette aid
	Microanalytical balance
Quality control laboratory	Fluorescence-activated cell sorting (FACS) – flow cytometry
	Fluorescent inverted microscope
	Microplate reader that could read fluorescence and luminescence
	Haematocytometre with cover slip
	Refrigerated microfuge
	4°C refrigerator
	-20°C freezer
Cell bank	Controlled-rate freezer
	Liquid nitrogen dewer (for use with controlled-rate freezer)
	Liquid nitrogen tank
	Dry-shipper (small size)
	-80°C freezer
	Astrapac bag sealer

[60] As identified through King's College London and CCMI

3.9 Identified key personnel

Key personnel identified for the establishment and operation of a stem cell therapy unit were: General Manager, Medical Director, Scientific Director, Qualified Person (QP), Quality Manager, Production Manager, stem cell technologists and maintenance technician (Figure 3.9).[61] The Medical Director shall be one of the consultant physicians in cord blood collection or in the clinical transplant of stem cells. The Scientific Director is responsible for the research in the therapeutic application of stem cells. The Quality Manager and Production Manager should be employed on a full-time basis from the start of the project due to their involvement in duties relating to the operations of the stem cell therapy unit, including development and implementation of the pharmaceutical quality system, training of personnel, process validation, establishing in-process controls and finished product specifications, evaluation and trending of environmental monitoring, approving batch manufacturing records and ensuring that activities conform to the required GMP and GLP standards. The QP shall be contracted, starting with 15 hours per week due to the limited batch certification activities.

Two stem cell technologists with a Bachelor degree in a science relevant field will be recruited to support GMP production, formulation and banking of stem cell products in compliance with validated SOPs and regulatory requirements, processes and equipment validation, maintenance of the quality system and documentation systems.

[61] The organisational structure at CCMI was identified as a model for the start-up program in Malta

The rationale of recruiting two stem cell technologists was for:

- Verification of process steps to minimise human errors
- Business continuity in absences due to sick leave, vacation leave or termination of employment
- Management of planned or unplanned increase in workload

The stem cell technologist will report to the Production Manager and preferably have knowledge and experience of:

- Laboratory-based discipline
- Cell culture techniques
- Tissue banking
- Aseptic manufacturing in controlled clean room environment
- Knowledge of the EU regulatory requirements
- Development and implementation of validation programs
- SOPs development
- Reporting on deviation to SOPs
- Maintaining document systems and batch records

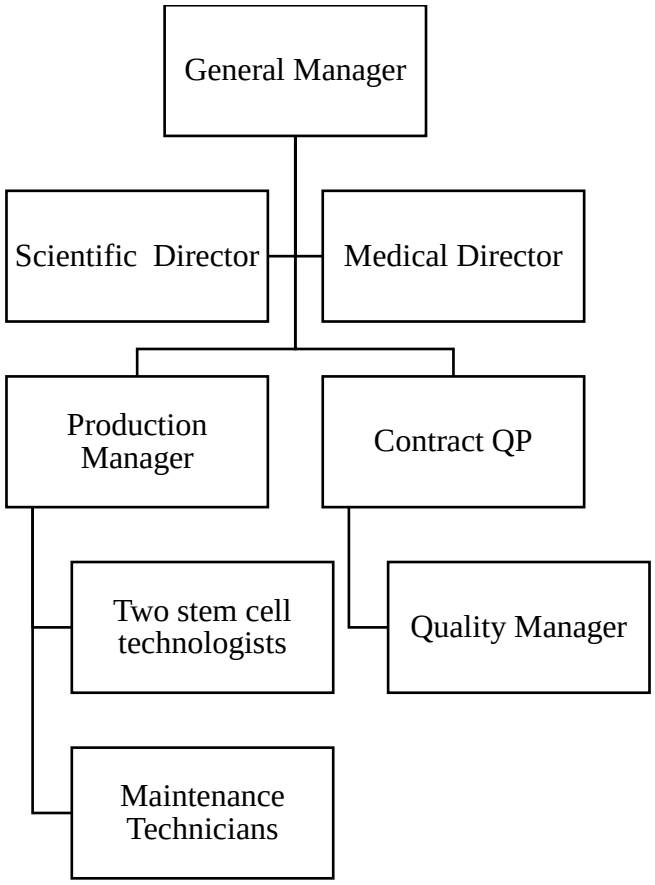


Figure 3.9 Organisational structure at the stem cell therapy unit in Malta

3.10 Documentation of Policies and Procedures

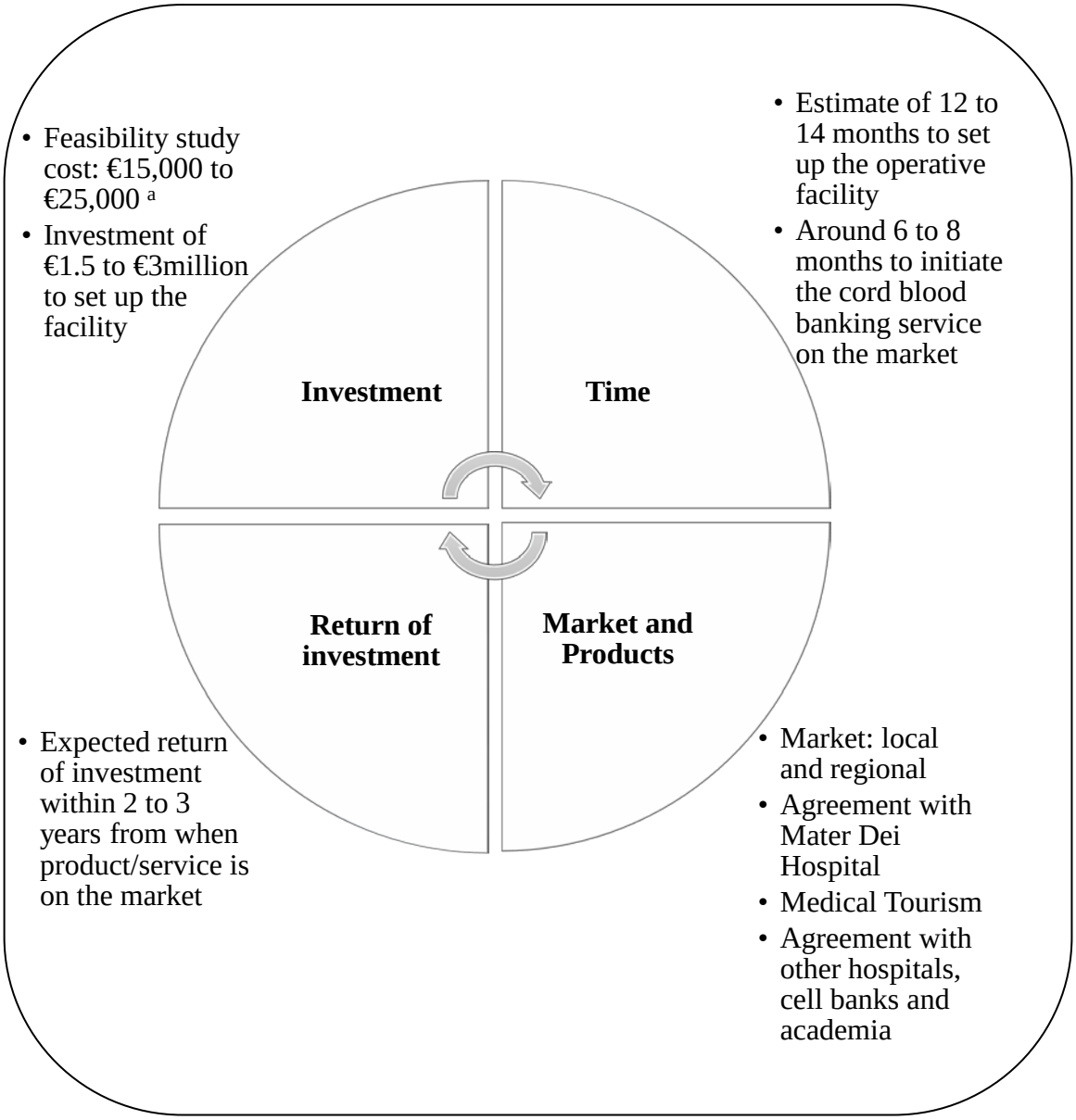
Good documentation is the fundamental basis of GMP and forms an essential part of the quality system.[6] A list of 36 policies and SOPs (Appendix 12) to be developed and implemented in the pharmaceutical quality system of the stem cell therapy unit were identified. The list of policies and SOPs is not comprehensive; however it gives an indication of core policies and procedures to be incorporated in the quality system.

[6] European Commission. Consultation Document – Draft Guidelines on Good Manufacturing Practice for Advanced Therapy Medicinal Products[Online]. Brussels: EC; 2016 [cited 2017 May 28]. Available from: URL: http://ec.europa.eu/health//sites/health/files/files/advtherapies/2016_06_pc/2016_06_draft_guideline.pdf

3.11 Economical implications

The private investors will provide significant funding for this project. During the networking meetings, the economic implications of setting up a stem cell therapy facility were identified, including preliminary projections of the typical investment required to set up the unit, estimated timelines and revenue generating strategies to retire the investment (Figure 3.10).

Technical specialists in the field of stem cell therapy could provide valuable assistance when conducting a feasibility study to support investors take a decision on whether the Malta stem cell therapy project is viable or not. Capital and operational expenses (Table 3.12) were estimated through the network collaboration, including rent of the facilities at MLSP, cost of setting up a Class B processing facility, cost of the identified equipment and yearly salaries of key personnel required for setting up a stem cell therapy unit.



[a] estimated by CTP System

Figure 3.10 Planning the business aspect of the project

Table 3.12 Capital and Operational Expenditures

Capital Expenditure		
Expenditure	**Type of Expenditure**	**Estimated Cost (€)**
Facility[62]	Class B Processing Laboratory (without equipment)	5,000.00 per square metre
Equipment[63]	Class II Biosafety cabinet	5,301.00
	CO_2 humidified incubator	5,793.50
	Refrigerated centrifuge (bench-top)	12,500.00
	Inverted microscope with camera	7,154.23
	Refrigerated microfuge	3,200.00
	4°C refrigerator	4,500.00
	-20°C freezer	790.00
	Digital water bath and stainless steel lid (6L)	1,113.80
	Vortex Stirrer and accessory pack for use	614.93
	Plate shaker, microtitre	379.00
	Sterile tubing welder	7,339.80
	Tube sealer	3,632.50
	Peristaltic pump	3,019.65
	Fluorescence-activated cell sorting (FACS) – flow cytometry	Against consumption
	Fluorescent microscope (preferably inverted microscope)	Against consumption
	Fluorescence microplate reader	18,373.17 – 25,872.94[a]
	Controlled-rate freezer	21,370.00
	Liquid nitrogen dewer	2,950.00
	Liquid nitrogen tank (capacity to store around 8,300 cord blood samples)	73,668.00
	A dry-shipper	4,000.00
	-80°C freezer	12,845.00

[62] Estimated through CTP System

[63] Estimated through foreign investors

	Micropipettes: Pipetman P10 Pipetman P20 Pipetman P200)	 228.77 201.40 201.40
	Pipette aid	200.00
	Haematocytometre with cover slip	20,000.00
	Astrapac bag sealer	3,000.00
	Microanalytical balance	1,810.00
Operational Expenditure		
Expenditure	**Type of Expenditure**	**Estimate Cost (€) /annum**
Rent[64]	Premises	85.00 per square metre
	HVAC system	15.00 per square metre
Personnel salaries[65]	General Manager	69,800 – 94,070[66]
	Medical Director	50,000 – 70,000
	Scientific Director	50,000 – 70,000
	Contract Qualified Person	54,600 (€70/ hour)
	Quality Manager	35,000 – 60,000
	Production Manager	35,000 – 60,000
	Stem cell technologist	37,000[b]

[a] various equipment models were quoted

[b] stem cell technologist salary is the same as that remunerated at CCMI

On the income side, the collaboration with European centres of excellence in stem cell therapy and technical experts in this field facilitated obtaining information on the estimate charges for stem cell therapy medicinal products in various therapeutic

[64] Identified through MLSP

[65] Estimated through industry

[66] MISCO. MISCO Salaries and Benefits Report 2015- 2016 [Online]. Malta; 2015 [cited 2017 May 28]. Available from: URL: http://www.miscomalta.com/details.aspx?id=276789

applications (Table 3.13). The cost for storing cord blood banking is €2,000 per cord blood unit.

Table 3.13 Therapy costs for stem cell-based medicinal products in different therapeutic applications

Stem cell therapy	Estimated cost per treatment (€)
Hepatocyte stem cell transplantation (King's College London)	3,000
Use of adipose stem cells for aesthetic medicine (CTP system)	50,000
Autologous bone marrow derived mesenchymal stromal cells for critical limb ischaemia (CCMI)	22,000

3.12 Regulatory sciences norms for regulators

The initiative taken by the MMA to collaborate in the Malta stem cell project was important to realise the full potential of the project and support sound regulatory decisions. The collaboration between the MMA and HPRA was fundamental to prepare local regulators for conducting inspections of stem cell therapy facilities and blood establishments. A quality manual (Appendix 13) was developed to establish a framework for conducting inspections of blood establishments and stem cell therapy facilities. The quality manual was divided into six sections and three appendices (Table 3.14).

Table 3.14 Structure of the quality manual

Section/Appendix	Content
Section 1	Scope of the quality manual
Section 2	References to EU legislation on blood and blood components, tissues and cells, ATMPs, European guidance documents and local legislation
Section 3	Terms and Definitions used in the quality manual
Section 4	Context of the organisation relating to the role and objectives of the MMA, stakeholders needs and expectations, the quality management system of the MMA
Section 5	Inspections process from scheduling to post-inspection activities
Section 6	Recruitment and training of inspectors
Appendix 1	Quality Risk Management tool
Appendix 2	Scoring of intrinsic factors
Appendix 3	Aide memoire that serves as a guide for inspection team conducting inspections in blood establishments and stem cell therapy facilities

The quality manual included suggestions arising from the pilot validation. The changes implemented in the quality manual following pilot validation were: addition of process maps to make the manual more user-friendly, addition of a glossary of common terms used in the manual and adjoining the checklists used during inspections of blood establishments and stem cell therapy facilities into one Aide-Memoire.

3.13 Stem cell therapy and blood components glossary

A total of 160 terms related to the field of stem cell therapy and blood components to be used by academia, industry and regulators were identified (Appendix 14). The

validation exercise served to identify eight terms to be incorporated in the glossary, including:

1. Comparability
2. Confirmed group label
3. Confluence
4. Donor deferral
5. Haemolytic transfusion reactions
6. Master cell bank
7. Sterility
8. Working cell bank

3.14 Dissemination of results

A poster titled ‘Therapeutic and economic implications of regulating stem cell therapy and blood components’ was accepted for poster presentation at the 77th International Pharmaceutical Federation (FIP) World Congress of Pharmacy and Pharmaceutical Sciences 2017, to be held in Seoul, South Korea from 10-14 September 2017 (Appendix 15).

CHAPTER 4

DISCUSSION

4.1 Advancing regulatory science

Regulators must keep pace with the rapid advances in innovative regulatory sciences. The Malta Medicines Authority identified investing in competences of its scientific personnel to be knowledgeable, up-to-date and proficient in scientific developments and capable of assessing the quality, safety and efficacy of innovative therapies, to accomplish its mission to protect and enhance public health. In line with this strategic plan, the Authority prioritised capacity building in two regulatory sciences areas namely, stem-cell based medicinal products and blood components.

Stem cell-based medicinal products are complex therapeutic agents and their effective regulation depends on continuous improvements of all perspectives of the regulatory sciences, including benefit-risk assessment, research on regulatory processes and incentives to support regulatory science and expertise (Yuan and Wang, 2014). Lack of effective regulation may reduce the fund support for basic research, stop or delay clinical applications and subsequently lead to low confidence among investors in the field of stem cell therapy. The regulatory aspect of the Malta stem cell project may seem bureaucratic, however both the scientific research development and the production with state-of-the-art GMP principles will not reach the ultimate aim of registering the product unless the concept of regulatory sciences is incorporated from the start of the project. It is difficult for scientists to accept new standards and GMP requirements which are essential to obtain a marketing authorisation (Pellegrini et al, 2016). The innovative contribution of this research study in establishing the network for setting up a stem cell therapy facility in Malta was that the researcher could put forward regulatory input and promote regulatory sciences from the start of the project.

Cord blood banking and stem cell therapy is a niche area to Malta, and it is difficult to establish a stem cell therapy unit in Malta without the contribution and expertise of European collaborators. The collaborations with academia (King's College London), industry (Holostem Terapie Avanzate S.r.l), technical specialists in the area of advanced therapies (CTP System) and the European medicines regulatory network (HPRA) were all science-driven and were valuable tools to achieve the mission of advancing regulatory science.

Planning of the site studies was essential to ensure a robust design that collected a significant amount of information and experience in a short amount of time. Participants from different institutions contributed in the site studies to achieve the maximum possible benefit from each site study, for the following reasons:

1. Opportunity for networking between different collaborators in the project, where Malta Enterprise participated in the site study at King's College and University of Malta participated in the Holostem site study.
2. Participants had different experiences which facilitated obtaining more information through various questions asked during the site study.
3. Additional perspectives of the sites studied, facilitated reduction in bias which could be due to incorrect understanding and misinterpretation of the information and qualitative data from each site study. When there were two or more participants during site studies, participants were able to reflect with each other on what they were learning as well as support data collection, where different field notes were available after the completion of the visits.

4. Increasing the competences of scientific personnel from the Malta Medicines Authority, where Ms Roberta Agius, Head of Quality, International and EU Affairs participated in the King's College visit, Holostem visit and the inspection at IBTS.

Malta Enterprise and the potential investor were planning to participate in the site study at CTP System, however due to difficulties in the coordinates of the dates for the visit, the site study was carried only by the researcher. CTP System visited Malta following the site study for further collaboration with all interested parties in setting up a stem cell therapy unit in Malta. The GMP inspection of CCMI in Galway was conducted by one inspector who preferred to have one trainee observing the inspection.

The sequence of these site studies was also carefully planned through teleconferences and exchange of electronic mail with the respective centres and institutions to ensure that the scope of these visits was met. The site studies to established centres of excellence in stem cell therapy, King's College London and Holostem, provided valuable insights of the requirements to set up a stem cell therapy unit and challenges to obtain a marketing authorisation of stem cell-based medicinal products. The history of both King's College London and Holostem in stem cell therapy started from small laboratories carrying out basic stem cell research and only following years of hard work and perseverance was their mission achieved to translate research work from bench to bedside. Following these two experiences, the site study at CTP offered an opportunity to discuss different business models with technical experts that could be adopted to set up a stem cell therapy unit in Malta. The inspections with HPRA were conducted following the other three site studies for two reasons 1) Discussions with HPRA started

at a later stage after gaining knowledge and experience in the advanced therapy field and 2) Inspections on ATMP facilities and blood establishments are not as common as other GMP pharmaceutical inspections due to the limited number of such facilities in Ireland. Since IBTS is the national organisation responsible for collecting, processing, storing and distribution of all blood components in Ireland similar to the National Blood Transfusion Service in Malta, and CCMI is one of the first ATMP facilities in Ireland licensed to manufacture stem cells for use in human clinical trials, the two facilities where identified as suitable site studies to evaluate regulatory sciences norms in the area of stem cell therapy and blood components.

Although the processing and regulatory pathways of blood components and stem cell-based products are different, there were common topics which were observed during the inspections at IBTS and CCMI, including donor management, traceability systems, microbiology and quality control, risks of cross-contamination, requirements for testing laboratories, analytical instrumentation, environmental monitoring, assessment of processes, information technology system such as data processing, integrity and security, evaluation and follow-up notification of serious adverse events and reactions, and risk of transmissible diseases.

One of the areas assessed during the blood establishment inspection were measures that the organisation was taking to reduce the risk of transfusion-transmissible infection. Mitigation measures which were reviewed to improve the safety of blood supply to patients include the testing for Hepatitis E, measures to reduce risk of donor-derived Zika virus transmission and the ongoing project of pathogen reduction for platelets. There is no single method of pathogen inactivation that removes all viruses, bacteria,

spores, protozoa and prions (Klein and Bryant, 2009). Of particular concern are the threats of new emerging agents that are not yet recognised as threats to blood supply. Blood donor screening, pathogen testing and deferrals are ways to drastically reduce the incidence of transfusion-transmitted diseases (Klein and Bryant, 2009). In January 2017, IBTS changed their donor deferral policy from lifelong deferral to one year for men who have sex with men. This decision was taken following review of data presented from other countries that had changed their deferral criteria, where it was concluded that a one year deferral is as effective as lifetime deferral. In light of the past history of IBTS and the contamination scandals described as "the worst... in the history of state", revision of policy to one year deferral surprised many as it was expected to reduce the policy to a five-year deferral period.[31] Malta is also addressing the lifetime ban enforced by the National Blood Transfusion Service on men who have sex with men,[67] IBTS offered to support the technical committee in Malta that will be compiling scientific evidence to support the Maltese Government in reaching a decision on whether to change the national deferral policy.

The major strength of this study was the opportunity to collaborate with different key experts in the stem cell therapy field and gain different perspectives on areas which were being debated in the EU such as the consultation document proposed by the

[6] European Commission. Consultation Document – Draft Guidelines on Good Manufacturing Practice for Advanced Therapy Medicinal Products [Online]. Brussels: EC; 2016 [cited 2017 May 28]. Available from: URL: http://ec.europa.eu/health//sites/health/files/files/advtherapies/2016_06_pc/2016_06_draft_guideline.pdf

[31] O Malley K. Considering the policy of indefinite deferral imposed on MSM blood donors in Ireland. UCD Student Medical Journal [Online] 2017; [cited 2017 May 18]. Available from: URL: http://www.ucdsmj.com/2-4/

[67] Times of Malta. Ban on gay men's blood donation may be relaxed [Online]. Malta; 2016 [cited 2017 May 28]. Available from: URL: https://www.timesofmalta.com/articles/view/20160503/local/ban-on-gay-mens-blood-donation-may-be-relaxed.610813

European Commission regarding GMP guidelines for ATMPs.[6] The consultation document was discussed in-depth with the lead GMP inspector who conducted the inspection at CCMI. The inspector expressed his concerns that the proposed guideline is a diluted form of the GMP guidelines described within EudraLex Volume 4[25] which could be detrimental to patient safety. The risk-based approach described in the guideline could lead to a reduction in the quality and lack of rigour in GMP manufacturing of ATMPs, which are considered as high-risk medicines since they are sterile products susceptible to contamination, and due to their mode of action where they are retained and proliferate in the patient. The draft GMP guideline for ATMPs was also discussed with the technical experts from Holostem to obtain the view points of the industry on this draft guideline. Discussions with Professor Graziella Pellegrini and her technical team showed that the proposed GMP guideline for developers of ATMPs was welcomed. In recent years ATMPs have been regulated by pharmaceutical rules tailored for manufacturing and control of chemical molecules where the intrinsic characteristics of ATMPs introduced bottlenecks in the translation of these complex products from bench to bedside (Pellegrini et al, 2016). The risk-based approach outlined in the proposed guideline provides flexibility in the application of GMP requirements when manufacturing ATMPs. Holostem highlighted that any flexibility applied must be in line with the need to ensure the quality, safety and efficacy of the product and a balanced approach facilitates appropriate scientific development. CCMI expressed a very similar opinion to Holostem. While the industry and the regulators are not in agreement on the approach described in the consultation document, both parties are still uncertain whether this guideline will be an annex to the existing guidance in

[25] European Commission. EudraLex Volume 4 EU Guidelines to Good Manufacturing Practice Medicinal Products for Human and Veterinary Use: Annex 1 Manufacture of Sterile Medicinal Products [Online]. Brussels; 2008 [cited 2017 May 28]. Available from: URL: http://ec.europa.eu/health//sites/health/files/files/eudralex/vol-4/2008_11_25_gmp-an1_en.pdf

Eudralex Volume 4 or a standalone document independent from other guidance. The feedback was that there are still concerns that this guideline contradicts existing GMP requirements and there should be guidance on the meaning of a risk-based approach and which GMP activities cannot be subject to flexibility such as personnel, hygiene and training.

During the discussions with Holostem, Professor Pellegrini commented how the implementation of the ATMP Regulation EC 1394/2007[2] introduced new challenges in the development of Holoclar. One of the challenges faced by Holostem during the development process of Holoclar was the lack of communication between the regulators and scientists. The ATMP regulation was imposing new standards and requirements, while the scientists lacked the regulatory know-how and could not understand what was expected from them considering that they already had the clinical data and the technology to develop Holoclar. The beauty of collaboration is exemplified in the case of Holostem, where the laboratory at the University of Modena and Reggio Emilia was affiliated with Chiesi Farmaceutici, a pharmaceutical company with the regulatory know-how that could provide adequate level of quality manufacturing and GMP compliance required by the new legislation. The public-private partnership facilitated dialogue between the regulators and academia and supported overcoming challenges in obtaining funding to meet the post-approval commitments and deliver the product to patients. The cell therapy unit at King's College London and the Centre for Cell Manufacturing Ireland are also the outcome of successful collaborations.

[2] European Commission. Regulation (EC) 1394/2007 of the European Parliament and of the Council of 13 November 2007 on advanced therapy medicinal products and amending Directive 2001/83/EC and Regulation (EC) 726/2004 [Online]. Official Journal of the European Union 2007; L324:121-137 [cited 2017 May 28]. Available from: URL: http://eur-lex.europa.eu/legal content/EN/TXT/PDF/?uri=CELEX:32007R1394&from=EN

The site studies showed that academic efforts alone are not sufficient. The experiences shared and recommendations made by the technical experts in stem cell therapy during the research study showed that there is the need for a public-private partnership in order to set up a stem cell therapy unit in Malta. A public-private partnership through specific agreements can ensure effective and efficient use of resources and allow sharing of risks and rewards (Pellegrini et al, 2016). This business model is proposed for the project in Malta that aims to work through a network of collaborators in regulatory sciences offering new services currently unavailable in Malta.

Stem cell research in diabetes mellitus was identified as an area of interest to Malta. According to the International Diabetes Federation,[68] over 10% of the population in Malta above 18 years have diabetes mellitus where in 2015 there were 44,100 cases of diabetes mellitus. The incidence is expected to continue rising, making this condition a growing health problem. Diabetes mellitus has a significant impact on the quality of life of patients especially when complications such as peripheral arterial disease arise. It is for this reason that diabetes mellitus is a national public health priority for 2015-2020.[69] Critical limb ischaemia is still an unmet medical need where amputation is often inevitable (Liew and O'Brien, 2012). According to the Diabetes Foot Research Group, approximately 65 major amputations (loss of the lower limb) and over 350 minor foot amputations (loss of toes or part of the foot below the ankle) were carried out in Malta

[68] International Diabetes Federation. Malta [Online]. 2017 [cited 2017 May 28]. Available from: URL: http://www.idf.org/membership/eur/malta

[69] Ministry for Energy and Health. Consultation Document – Diabetes: A national public health priority 2015-2020. [Online]. Malta; 2014 [cited 2017 May 28]. Available from: URL: https://socialdialogue.gov.mt/en/Public_Consultations/MEHHEALTH/Pages/Consultations/DiabetesStrategy.aspx

in 2015 which "translates to a minimum of one amputation every day of the year." [70] The high prevalence of diabetes mellitus in Malta and the experience of studying the research work developed at CCMI on a stem cell therapy product indicated for critical limb ischaemia were two reasons for selecting diabetes mellitus as an area of specialisation during the research phase of the Malta stem cell project. The proposal was discussed and agreed with the potential investors.

Regulatory science is relevant to all collaborators in the Malta stem cell project, including the investors, where the experience of the investors in the pharmaceutical field is considered an important asset. It is more difficult for an investor with no knowledge of the medicines regulatory framework to understand the importance of maintaining high standards to ensure the quality, safety and efficacy of medicines. An advantage to the Malta stem cell project is that the foreign private partners have the knowledge and experience in storage and process of stem cells derived from umbilical cord, peripheral blood and bone marrow. Previous experiences of the investors in setting up cord blood banks can be used to establish a similar facility in Malta. The two facilities identified for setting up this project were the MLSP and the laboratories at the UOM Biomedical Sciences Building. UOM offered the use of the laboratories at the Biomedical Sciences Building for the research and development of the stem cell product, however it was acknowledged that the academic setting lacked the regulatory expertise and resources to establish and operate GMP-compliant manufacturing processes. This arrangement could cause difficulties at a later stage where GMP standards are necessity in the manufacturing of advanced therapies. Two logistical options were identified in this project, 1) Setting up a cord blood bank at MLSP and

[70] Think Magazine. Attacking the silent epidemic of diabetes [Online]. Malta: University of Malta; 2016 [cited 2017 May 28]. Available from: URL: http://www.um.edu.mt/think/attacking-the-silent-epidemic-of-diabetes/

carrying out stem cell research at the University and 2) Setting up one facility at MLSP for both parts of the project. The gap between basic and clinical research is a weak point in a stem cell therapy projects (Arjmand et al, 2012). The investors were more inclined to set up the cord blood bank, stem cell research and manufacturing process of the stem cell therapy at the MLSP. The facility options will be analysed in the proposed feasibility study.

Whether retrofitting an existing laboratory or designing a new laboratory from shell space, the basic GMP requirements for setting up a stem cell therapy unit is the same (Wessleschmidt and Schwartz, 2011). The information and experiences gained during the site studies were used to address the requirements for setting up the new facility layout. The controlled environment of a carefully designed, constructed, validated and maintained cleanroom minimises the risks of environmental contamination during aseptic processing and decreases the risks of cross-contamination. The risk of stem cell product contamination increases with the duration and type of manufacturing process, where open systems require more rigorous in-process controls than closed systems. Sterility testing is one of the controls used to detect the presence of microbial contamination both for in-process test samples and finished product. There is no single screening test that detects all types of contamination, and the testing for strains of microorganisms identified by the European Pharmacopoeia requires 14 days before all results are finalised (Arjmand et al, 2012). Stem cell therapy products have a relatively short shelf-life, at times even as short as 24 hours and the products are released at specific time points provided that sterility testing is negative-to-date. Microbial sterility testing is continued after release of the product. The same bacterial screening programme is also used in blood establishments to identify platelet packs contaminated

with bacteria. An issue with the use of bacterial screening is that contamination could be at very low levels to be detected.[71] The ideal bacterial screening test equipment should have high diagnostic sensitivity, a short test time and a high clinical efficiency (Störmer and Vollmer, 2014).

The design and implementation of an environmental monitoring program is an essential parameter in cleanroom quality control to minimise the risk of contamination. Environmental monitoring is a living process and needs to adapt to changes in the environment and in the manufacturing process.[72] The objective of the environmental monitoring program is to maintain a consistent and controlled environment to eliminate exogenous contaminants (Ottria et al, 2010). The importance of environmental monitoring in accordance with GMP principles including temperature, relative humidity, air particle concentration, air flow, integrity of HEPA filters, differential pressure and microbiological controls, were reiterated during the five site studies.

Regardless of the environmental monitoring systems available in stem cell facilities, general laboratory practice is the heart of laboratory operations. Concerns in the operation of good cleanroom facilities include cleaning, maintenance, appropriate cleanroom garment selection according to the grade of the working area and gowning qualification. Personnel should be appropriately trained and certified competent in their respective areas of experience (Indamar et al, 2015). The Production Manager is responsible to ensure that all personnel in cleanroom facilities are working in areas in

[71] Advisory Committee on the Safety of Blood Tissue and Organs. Pathogen inactivation of platelets: Report of the SABTO Working Group [Online]. UK; 2014 [cited 2017 May 28]. Available from: URL: https://www.gov.uk/government/publications/platelet-transfusion-infection-risk-review

[72] Lee C. Environmental Monitoring: Large academic facility experience. International Society for Cellular Therapy [Online]. 2009 [cited 2017 May 28]. Available from: URL: http://c.ymcdn.com/sites/www.celltherapysociety.org/resource/resmgr/files/PDF/Meetings/ISCT_2009/Presentations/Tuesday/BallroomA/Lee.pdf

which they have been trained and certified competent. Transferring experience of personnel with knowledge in laboratory-based discipline, aseptic and cell culture techniques, GMP sterile manufacturing, clinical application of tissue transplantation, regulatory sciences, development and execution of validation programs and quality system development in stem cell therapy facilities is beneficial (Arjmand et al, 2012; Pellegrini et al, 2016). Blood and tissue establishments have personnel with experience in managing aseptic procedure, including collection, testing, labeling and distribution of blood components or tissues and their active involvement in pharmacovigilance programs and traceability systems from donor to recipient. Personnel with knowledge in Information Technology are also an asset when setting up a stem cell therapy unit as they can support building a comprehensive electronic quality system and aid in implementing the Single European Code labeling with ISBT 128 (Rebulla and Giordano, 2011; Leemhuis et al, 2014). Key personnel to recruit in the stem cell therapy unit were considered early in the project. The expertise required in a stem cell therapy unit is available locally through academia, national blood establishment, tissue establishments, hospitals and industry. The next step would be the time-wise availability of the identified key personnel. Sharing of human resources with other facilities that have the required expertise available may be considered as a cost-effective model to start the project. The stem cell therapy unit needs to have a close relationship with the clinical transplant team where often one of the transplant physicians serves as the Medical Director for the stem cell therapy unit. The close collaboration between the clinical transplant team and the stem cell therapy unit facilitates communication on the patient's therapy, including availability of the released product for administration to the patient which is more important in newly established facilities where "documentation system may be less well-tested and cryopreservation potentially less reliable"

(Leemhuis et al, 2014). Clinical grade stem cell manufacturing cannot be achieved by simply transferring the methodology into a cleanroom facility. Development of a robust pharmaceutical quality system, including validation of procedures for the entire process, is a key principle in GMP (Arjmand et al, 2012).

It is far too easy to decide to set up a stem cell therapy unit on the perception that every country should have one. The amount of funding needed for the setting up and development of ATMPs is impressive (Pellegrini et al, 2016).The financial planning is crucial in setting up a stem cell therapy unit and was identified as a core consideration from the outset of the project. Financial aspects remain critical once the unit has been established in terms of revenue generation, as well as maintenance and processing costs. The economic implications involved in setting up and running a stem cell therapy unit will ultimately impact the cost of the stem cell therapy, where sustainability is needed to provide a therapy to patients on a long term basis (Leemhuis et al, 2014). Scientists and regulators should continue to seek a sustainable balance between optimisation of the manufacturing process to assure the quality, safety and efficacy of products and optimisation of the regulatory framework ensuring that stem cell therapies are accessible to patients.

Blood, tissues and cells for human application are subject to continuous innovation.[73] Each of these innovations brings changes in the operational set-up of the processes along the chain from donor to recipient that may lead to changes in the clinical

[73] European Commission. Commission Implementing Decision of 1.3.2016 concerning the work programme for 2016 in the framework of the third Programme of the Union's action in the field of health (2014-2020) and the EU financial contribution to the WHO Framework Convention on Tobacco Control , serving as a financing decision. Brussels: EC; 2016 [cited 2017 May 28]. Available from: URL: http://ec.europa.eu/research/participants/data/ref/other_eu_prog/hp/wp/hp-workplan_2016_en.pdf

outcomes of patients who benefit from transfusion, transplantation or assisted reproduction. There is a clear legislative requirement for competent authorities to indicate which activities may be undertaken by authorised blood and tissue establishments. The priority launched under the 3rd Health Programme Workplan 2016[72] addresses the need for authorisation of preparation processes which require access to specific technical knowledge and expertise, as well as clear and robust procedures at the competent authority level. The facilitatinG the Authorisation of Preparation Process for blood, tissues and cells (GAPP) Joint Action is a three year programme starting in 2018 aimed at facilitating development of a common and optimal approach to assess and authorise the preparation process in blood and tissue establishments. The joint action will contribute to the implementation of legislation in the area of blood, tissue and cells and will provide tools and training to increase harmonisation of those member state activities that regulate the areas of blood transfusion, transplantation of tissues and cells and assisted reproduction. The GAPP activities will also contribute to greater assurance of receiving safe and effective treatments in these fields where highly specific technical knowledge and clinical expertise is requested for assessing and authorising innovative processes in blood and tissue establishments.[74]

Malta as a small member state would benefit from this joint action through strengthening authorisation procedures and through collaboration with experts from more advanced member states. This is an opportunity to build local capacity for blood, tissues and cells in line with industry and public health requirements. The MMA

[72] Lee C. Environmental Monitoring: Large academic facility experience. International Society for Cellular Therapy [Online]. 2009 [cited 2017 May 28]. Available from: URL: http://c.ymcdn.com/sites/www.celltherapysociety.org/resource/resmgr/files/PDF/Meetings/ISCT_2009/Presentations/Tuesday/BallroomA/Lee.pdf

[74] European Commission. Joint Action Grants (HP-JA) 3rd EU Health Programme. Proposal to a Joint Action facilitatinG the Authorisation of Preparation Processes for blood, tissues and cells (GAPP). Brussels, 2016.

together with the Health Regulation Department and the National Blood Transfusion Service, agreed to collaborate in this EU-funded project and a proposal was submitted in November 2016 where Malta will be one of the associated partners in this joint action. A meeting of nominees for Joint Action on Preparation Process Authorisation was held on the 4 May 2017 at DG Sante in Brussels. A total of 30 nominations from 21 EU countries will participate in this joint action, led jointly by the Italian authorities namely the Italian National Transplant Centre and the Italian National Blood Centre. The joint action proposes 6 defined core work-packages and Malta will be actively involved in knowledge sharing of authorisations of preparation processes between EU competent authorities. Through the work of joint actions and other EU funded projects, member states are able to collaborate as co-partners to address common concerns, share experience and knowledge in complex areas, build on existing activities and expertise, identify commonalities and learn from differences. The quality manual for regulators inspecting blood establishments and stem cell therapy facilities developed in this research study takes into consideration manuals as outcomes of such EU initiatives. The approach taken in the quality manual was to integrate the regulatory responsibilities at the interface between blood, tissues and cells and medicinal sectors where stem cell therapy facilities are GMP sites.

4.2 Limitations

Some of the findings reported in this study may have been influenced by selection bias. The five site studies were carried out in state-of-the-art facilities that have high-level experts in the area of blood, tissues and cells. These sites were selected in order to have valuable insights from the ‘best of the best’ facilities and to collaborate with leading experts in Europe. On the other hand, sites that do not have such a positive experience

in this novel field could still have contributed to give another perspective to the research study. Site studies were established in the four areas determined as key stakeholders to collaborate for this project namely academia (King's College London), industry (Holostem Terapie Avanzate), technical specialists (CTP System) and the European medicines regulatory network (HPRA). Due to limited time and resources, collaborations were established with only one stakeholder from each area, which was a limitation due to the lack of a longitudinal perspective. Another limitation was that the duration of the site studies at King's College and Holostem were short and there were no ongoing operational activities. This limitation restricted the amount of information that was collected from these site studies. The inspection with HPRA at CCMI took place over three full days and was an opportunity to reflect on the real scenario. During the site studies field notes were taken where at times it was difficult to capture key dialogue while understanding the concept of the discussions. This limitation was less significant when others participated in the site studies. The economic implications presented in this research provide an indication of the estimated costs involved to set up a stem cell therapy unit. The investors in this project need to clarify the remaining gaps for the project including confirming the facilities, the laboratory space required and the specialised area for stem cell research. Stem cell transplantation for the treatment of the diabetic foot ulcers was identified as a local potential area for stem cell research. Establishing the manufacturing process for the stem cell therapy provides a clear picture of the specific requirements to develop the product. The business proposal facilitates the conduct of a feasibility study tailored to the specific needs of the project including laboratory finishes, specific equipment, supplies and reagents. A completed feasibility study would support the investors to make a better informed decision on the viability of this project.

4.3 Recommendations for further work

The project for setting up of a stem cell therapy unit is still in its infancy. The next steps of the project are:

1. Drafting a business proposal and presentation to Malta Enterprise with a clear description of the proposed business venture, potential revenue streams and operational costs. Malta Enterprise would then be in a better position to discuss and offer support measures that can be tailored for this project.
2. Signing of a MOU between the public and private sectors through the MLN to improve the likelihood of a positive outcome from the public-private partnership. The MOU will include a definition of the objectives and areas of cooperation, detailed specifications of the responsibilities, established points of contact, confidentiality clause and financial arrangements.
3. Drafting a memo to cabinet to seek Government support to finance the operations of the cord blood bank.
4. Conducting a feasibility study of the project by potentially utilising the services offered by technical specialists in the area of the ATMPs. CTP System estimated that the feasibility study would cost between €15,000 and €25,000.
5. Establishing new networking collaborations with leading scientific experts to involve in local stem cell research for the purpose of developing and commercialisation of the stem cell therapy product.
6. Establishing collaborations with other European national competent authorities to continue building the competences of the regulator. The MHRA invited the MMA to collaborate and visit the agency in London to receive training at the licensing division for ATMPs, clinical trials unit and to visit the National Institute for Biological Standards and Control.

4.4 Conclusions

Identified European collaborators in the area of stem cell therapy and blood components are King's College London (academia), Holostem Terapie Avanzate (industry), CTP System (technical specialists) and Health Products Regulatory Agency (European Medicines Regulatory). These European institutions, together with the potential investors and local public institutions, namely the Malta Medicines Authority, Malta Laboratories Network, University of Malta, Malta Enterprise, and Malta Life Sciences Park, formed part of the established network of collaborators to investigate the setting up of a stem cell therapy unit in Malta. The collaborations contributed to 1) Up-skill knowledge and competence in stem cell therapy and blood components, 2) The exchange of experiences and best practices from professionals with extensive knowledge in the field of ATMPs and preparation of blood components, 3) Identify public-private partnership as the business model to be adopted for the project in Malta, 4) Reach a decision that the project should be divided into two phases: (a) setting up of a cord blood bank subject that the Government contributes towards sustainability of this project and (b) development of a stem cell therapy facility where the cord blood bank supports availability of the raw material, 5) Establish that the facility may undertake stem cell research in the area of diabetes mellitus, in line with national health priorities, 6) Identify how different local public institutions can support the setting up of the stem cell therapy unit in Malta through optimal resource allocation, 7) Identify the requirements to set up a stem cell therapy unit, 8) Identify preliminary projections of the typical investment required to set up a stem cell therapy facility, revenue generating strategies and estimate charges for stem cell therapy products in various therapeutic applications, 9) Prepare the Malta Medicines Authority for inspecting stem cell therapy

facilities and blood establishments and to 10) Develop a quality manual for regulators inspecting stem cell therapy facilities and blood establishments.

The requirements to set up a stem cell therapy unit are 1) Facilities to carry out stem cell research, cord blood banking and GMP activities designed to supply preparations for routine clinical use, where the facility should be designed to minimise the risk of contamination with attention to ergonomics, air quality graded cleanrooms for different kinds of activity, interior finishes and controlled access and areas to be considered in the design of the facility are quarantine, production, storage, ancillary areas, quality control and cell banking areas, 2) Facility services including the availability of generators for backups when power supply fails, HVAC systems and adequate spaces to store liquefied gases cylinders including liquid nitrogen and carbon dioxide, 3) Controlled environmental monitoring system for temperature, humidity, air particle concentration, HEPA filtered air providing pressure cascades with the highest positive pressure in areas where processing contributes to greater risk of contamination to cell cultures and microbiological control, 4) Equipment for the manufacturing process where the equipment identified and listed in this study will continue to develop as the project evolves and more specialised equipment may be required depending on the manufacturing process of the stem cell therapy, 5) Key personnel with the necessary competences to work in sterile manufacturing facilities and 6) A pharmaceutical quality system including SOPs, training records, batch manufacturing records, technical requirements, contracts, traceable records and reports.

The economical implications involved in setting up a stem cell therapy unit require an investment of between €1.5 and 3 million euro. Expenditures involved setting up and

running a stem cell therapy unit include rent of facilities (€100/m^2/year), costs for setting up a Class B processing laboratory (€5,000/m^2), equipment (c€300,000) and salaries for personnel (c€600,000/year). The expected return of investment was estimated to take between 2 to 3 years from when service is on the market where the storage fee for cord blood banking is €2000/cord blood unit and it is envisaged that the therapy charge for stem cell transplantation for the treatment of diabetic foot ulcers would cost around €22,000.

A quality manual was developed as a tool for regulators inspecting stem cell therapy facilities and blood establishments in line with regulatory sciences norms, with the aim to amalgamate existing guidance documents to set a framework for the conduct of inspections. A glossary of 160 terms for use in the field of stem cell therapy and blood components was compiled.

The research experience exemplified the value of collaboration, where the public and private sector, together with the positive interaction of the regulators, can work seamlessly together in a highly complex area, placing Malta at the forefront of stem cell science.

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