



Sanquin

Sanquin Cellular Therapy Services

Your experienced partner in translational research



Blood and Beyond

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Our laboratory for stem cell transplantation started its activities in 1979 and is now part of Sanquin Blood Supply Foundation. The laboratory is positioned within the Research division of Sanquin to benefit optimally from the scientific environment. All research within this division is related to the many aspects of blood and blood transfusion, the division employs around 300 scientists. The stem cell laboratory serves both academic and non-academic hospitals and is actively involved in the development of new (stem) cell products for clinical trials. Since we broadened our services into the field of cellular therapies, we currently offer our services under the name Sanquin Cellular Therapy Services.



Sanquin Cellular Therapy Services:

- State-of-the art preparation of cellular products, including stem cell products, for patient care or diagnostics
- Highly qualified and well-trained staff
- Extensive experience with handling cellular products
- Participation in translational research
- Tailor-made solutions for the generation of cellular products for clinical trials
- Registration & documentation, including CCMO approval
- Analysis of production processes to guarantee the quality and safety of products
- Clean room facilities meeting the latest GMP-requirements

Cellular therapies currently available

Stem Cell Therapies

Hematopoietic stem cell (HSC) transplantation is the procedure to restore stem cells that have been destroyed by high doses of chemotherapy and/or radiation therapy. The stem cells can be derived from bone marrow, peripheral blood or umbilical cord blood. Within the Laboratory for Stem Cell Transplantation, blood and bone marrow is processed for patients who are in need of stem cell transplantation. Besides hematological recovery, HSCs are also used for other applications, such as for treatment of autoimmune diseases (Systemic lupus erythematosus, Crohn's disease) or acute myocardial infarction or chronic peripheral artery disease.

Many novel cellular products for individual therapy have been developed over the past few years, which are now being translated into the clinic. These new cellular therapies can be subdivided into regenerative therapy or immunotherapy. Here we describe which cell types are presently used in our facility and how they are applied in therapy.

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Cellular therapies in development

Expanded hematopoietic stem cells

Previous research in a mouse model has shown that transplantation of *ex vivo* expanded cord blood (CB) stem cells results in a significantly accelerated platelet recovery without adverse effects on long term engraftment. Moreover, in this mouse model, double cord transplantation using one un-manipulated CB and one expanded CB resulted in engraftment of both cords. The experimental CB expansion model is now being translated into a clinical protocol. The goal is to use expanded CB cells in a phase II study performing double cord transplantation in high risk adult patients lacking suitable stem cell donors.

Dendritic cells

Dendritic cells (DCs) are the best antigen presenting cells in the human body and are involved in activation of the adaptive immune response. DCs can be cultured *in vitro* from monocytes and subsequently used as immunotherapy, to modulate the immune response. Immuno-activatory DCs have the potential to treat cancer, for example melanoma, prostate cancer or colon cancer. Recently we have developed a method to generate clinical grade immuno-activatory DCs. Starting in 2011, these DCs will be studied in a phase I clinical trial with patients suffering from oesophageal adenocarcinoma. Besides immuno-activatory DCs, tolerogenic DCs may also have a clinical application in the near future. In situations where the immune response is non-beneficial or unwanted, such as transplantation rejections and auto-immunity, as for example in diabetes, these tolerogenic DCs can be used to suppress the immune reaction.

Mesenchymal stromal cells

Bone-marrow derived mesenchymal stromal cells (BM-MSC) are multi-potent cells that provide support for hematopoietic progenitor cells. They have the ability to migrate to sites of inflammation and injury and have immunomodulatory properties. Clinical applications of MSC include treatment of therapy-resistant acute graft versus host disease (GvHD) and treatment of rejection after hematopoietic stem cell or solid organ transplantation. A clinical-grade MSC expansion protocol is being developed to generate a standardized MSC product from third-party donors.

T cells

Patients that suffer from a relapse after stem cell transplantation can be treated with a donor lymphocyte infusion (DLI). With this procedure the lymphocytes from the original donor are infused after the transplantation to augment the anti-tumour response or to ensure that the donor stem cells remain engrafted.

Sanquin's scientific expertise in regenerative & immunotherapy*

Hematopoietic Stem Cells:

Sanquin Research Amsterdam, Department of Experimental Immunohematology
prof. dr. Ellen van der Schoot

Hematopoiesis/Mesenchymal Stromal Cells:

Sanquin Research Amsterdam, Department of Hematopoiesis
dr. Marieke von Lindern, dr. Carlijn Voermans, dr. Daphne Thijssen-Timmer

Dendritic Cells:

Sanquin Research Amsterdam, Department of Immunopathology
dr. Marieke van Ham, dr. Anja ten Brinke

Expanded Cord Blood Stem Cells:

Sanquin Research Leiden, Department of Clinical Transfusion Medicine
prof. dr. Anneke Brand, dr. Yvette van Hensbergen

Sanquin's collaborations in regenerative & immunotherapy

Radboud University Nijmegen Medical Centre
Netherlands Cancer Institute – Antoni van Leeuwenhoek Hospital
Academic Medical Center, Amsterdam
VU University Medical Center, Amsterdam
Leiden University Medical Center

* For key publications see page 7.

Sanquin's facilities

Regulatory Requirements & Accreditation

Manufacturing of cellular products or so-called Advanced Therapy Medicinal Products (ATMPs) has to meet the most stringent requirements with regard to the production process, to ensure the highest safety of the end product. The European Community has defined the standards in directive EC/1394/2007 with respect to the ATMPs and directive 2001/83/EC and guideline 2001/83/EC on all aspects, including GMP of human medicinal products. For the production of ATMPs in The Netherlands, approval by the CCMO or the Inspection of Health is obligatory.

The Laboratory for Stem Cell Transplantation is ISO-9001, ISO-13485 and ISO-14971 certified since 2004 and is acknowledged by the Dutch Ministry of Health as a tissue bank according to EU directive 2004/23/EC. In April 2007 the laboratory obtained the JACIE accreditation which was prolonged for 4 years in June 2010.



Techniques

- Cell isolation and expansion in closed systems
- Magnetic cell sorting using CliniMACS
- Loading of dendritic cells with RNA
- Cryopreservation with Planer® freezers
- Storage of cellular products in liquid nitrogen vapour storage freezers
- Thawing of cellular products for clinical use
- Progenitor cell assays
- Quality control assurance

Equipment

- Cobe 2991 cell processor (Gambro-BCT)
- CliniMACS®
- Planer® freezers

Sanquin's key publications

Van Hensbergen Y, Schipper LF, Brand A, Slot MC, Welling M, Nauta AJ, Fibbe WE. Ex vivo culture of human CD34+ cord blood cells with thrombopoietin (TPO) accelerates platelet engraftment in an NOD/SCID mouse model.

Exp Hematol 2006; 4:943-50.

Tijssen MR, van Hennik PB, di Summa F, Zwaginga JJ, van der Schoot CE, Voermans C. Transplantation of human peripheral blood CD34-positive cells in combination with ex vivo generated megakaryocytes results in fast platelet formation in NOD/SCID mice.

Leukemia 2008; 22:203-8.

Ten Brinke A, van Schijndel G, Visser R, de Gruijl TD, Zwaginga JJ, van Ham SM. Monophosphoryl lipid A plus IFN γ maturation of dendritic cells induces antigen-specific CD8(+) cytotoxic T cells with high cytolytic potential.

Cancer Immunol Immunother 2010; Epub ahead of print.

Ten Brinke A, Karsten ML, Dieker MC, Zwaginga JJ, Vrieling H, van Ham SM. Generation of dendritic cells for immunotherapy is minimally impaired by granulocytes in the monocyte product.

Immunobiology 2006; 211(6-8):633-40.

Ten Brinke A, Karsten ML, Dieker MC, Zwaginga JJ, van Ham SM. The clinical grade maturation cocktail monophosphoryl lipid A plus IFN γ generates monocyte derived dendritic cells with the capacity to migrate and induce Th1 polarization.

Vaccine 2007; 25(41): 7145-52.

Milano F, Rygiel AM, Buttar N, Bergman JJGHM, Sondermeijer C, van Baal JWPM, ten Brinke A, Kapsenberg M, van Ham SM, Peppelenbosch MP, Krishnadath KK. An ex-vivo read out for evaluation of Dendritic Cell induced autologous CTL responses against esophageal cancer.

Cancer Immunology and Immunotherapy 2007; 56(12):1967-77.

Milano F, van Baal JWPM, Rygiel AM, Bergman JJGHM, Van Deventer SJH, Kapsenberg ML, Peppelenbosch MP, Krishnadath KK. An improved protocol for generation of immunopotent dendritic cells through direct electro-poration of CD14+ monocytes.

J Immunol Methods 2007; 321(1-2):94-106.

Tijssen MR, Woelders H, Vries-van Rossen A et al. Improved postthaw viability and in vitro functionality of peripheral blood hematopoietic progenitor cells after cryopreservation with a theoretically optimized freezing curve.

Transfusion 2008; 48:893-901.

Van Beem RT, Hirsch A, Lommerse IM, Zwaginga JJ, Noort WA, Biemond BJ, Piek JJ, van der Schoot CE, Voermans C. Recovery and functional activity of mononuclear bone marrow and peripheral blood cells after different cell isolation protocols used in clinical trials for cell therapy after acute myocardial infarction.

EuroIntervention 2008 May; 4(1):133-8.

Van Hemert FJ, Voermans C, Van Eck-Smit BL, Bennink RJ. Labeling monocytes for imaging chronic inflammation.

QJ Nucl Med Mol Imaging 2009; 53(1):78-88.

Bennink RJ, Thurlings RM, van Hemert FJ, Voermans C, Dohmen SE, van Eck-Smit BL, Tak PP, Busemann-Sokole E. Biodistribution and radiation dosimetry of 99mTc-HM-PAO-labeled monocytes in patients with rheumatoid arthritis.

J Nucl Med 2008; 49(8):1380-5.

Tijssen MR, Woelders H, de Vries-van Rossen A, van der Schoot CE, Voermans C, Lagerberg JW. Improved post-thaw viability and in vitro functionality of peripheral blood hematopoietic progenitor cells after cryopreservation with a theoretically optimized freezing curve.

Transfusion 2008; 48(5):893-901.

Braakman E, Schuurhuis GJ, Preijers FW, Voermans C, Theunissen K, van Riet I, Fibbe WE, Slaper-Cortenbach I. Evaluation of 'out-of-specification' CliniMACS CD34-selection procedures of hematopoietic progenitor cell-apheresis products.

Cytotherapy 2008; 10(1):83-9.

Sanquin Cellular Therapy Services

Daphne Thijssen-Timmer, Manager R&D Cellular Therapies

Carlijn Voermans, Head of Laboratory

Plesmanlaan 125

1066 CX Amsterdam

The Netherlands

stamcellab@sanquin.nl

www.cellulartherapies.nl