Fraunhofer

AUTOMATED TISSUE ENGINEERING ON DEMAND – THE TISSUE FACTORY

MAKING VISIONS HAPPEN

SKIN FROM THE FACTORY

Automated Tissue Engineering on Demand

Today´s rising demand for more reliable, effective, humane and affordable alternatives to animal testing has lead to a growing market for in vitro test systems. These test systems employ human tissue equivalents for testing the safety of new substances or products which are supposed to be applied to the human body. However, the state-of-the-art processes for producing these so called tissue models are still very expensive, difficult and time-consuming, due to the complex and personnel-intensive manual manufacturing protocol.

For the purpose of a more cost-effective and quicker production of high-quality human tissue models, we from Fraunhofer-Gesellschaft are the first institution worldwide having realized the automation of this production process.

With a dedicated team of cell biology and engineering professionals, we have entirely reengineered the manual manufacturing protocol into an automatable and industrially feasible manufacturing process. Commercialization of the human tissue models resulting from this innovative production process is planned in 2013.

Want to learn more? Let's take a closer look at how we put the Tissue Factory into action!

THE TISSUE FACTORY

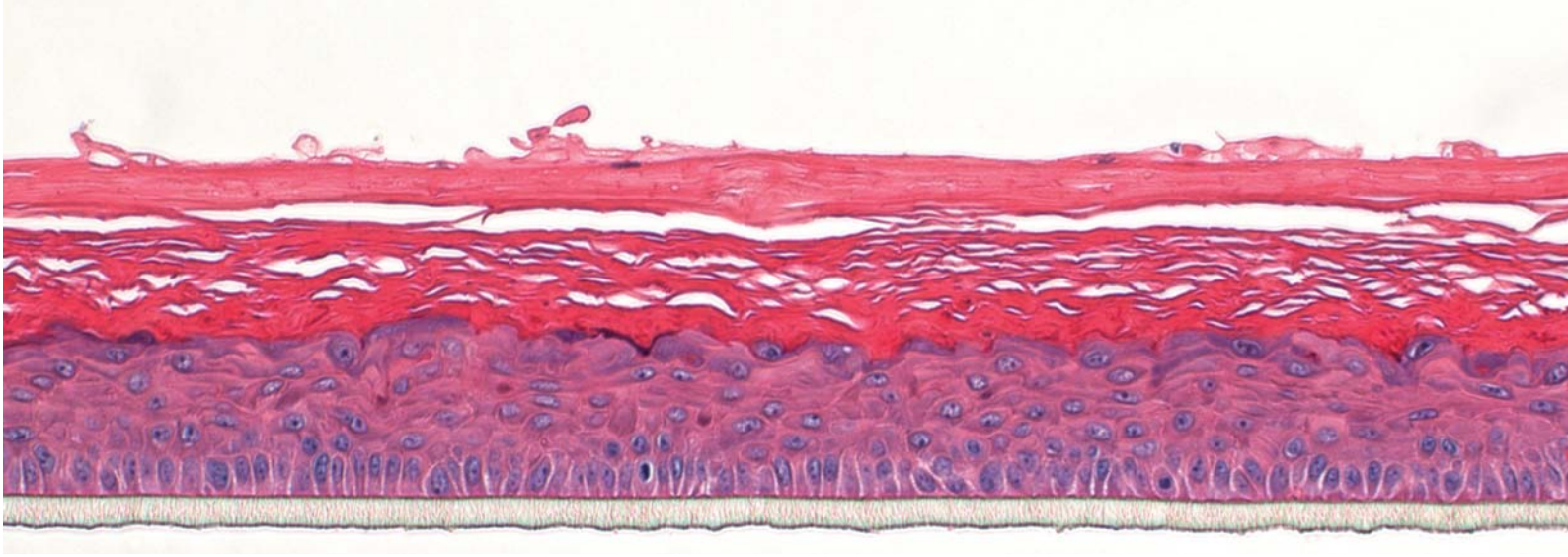
Automated production of human tissue equivalents

An interdisciplinary group of Fraunhofer scientists has implemented a continuous process chain for tissue engineering into a single production plant - starting with the extraction of cells out of human biopsy material up to the fully automated tissue culture of human skin equivalatents. The tissue factory provides a powerful platform for novel product, process and material technologies for the automated manufacturing of various kinds of human tissues.

The production system of the Tissue Factory is subdivided into three main modules: cell extraction module, cell expansion module and tissue cultivation module.

Specifications

- Automated manufacture of more than 5,000 human skin equivalents per month
- Portfolio of diverse human tissue products in significant quantities in a near future
- 100% quality control via non-invasive methods
- Widely GMP-compliant
- Modular plant assembly for flexible use of individual modules as stand-alone solutions



THE SKIN MODELS

Open Source Epidermal Model

The usage of reconstructed epidermal tissues for testing applications such as toxicity and efficacy is gradually expanding all around the world. Various different skin models have been already developed and have also passed successfully scientific validation at ECVAM (European Centre for the Validation of Alternative Methods to Animal Testing). However, all these validated skin models are either protected by patents and/ or are being commercialized using proprietary tissue culture procedures by only a few private companies. This makes their availability dependent on the corporate strategies of these companies.

Regarding this situation, the Tissue Factory now endorses the open source concept for the reconstruction of human epidermis. The Open Source Reconstructed Epidermis was originally developed and published by Professor Yves Poumay (Arch. Dermatol. Res. 296, 203-211, 2004) and further developed by Henkel to assure the continues availability of skin models to anyone – independent of use or party whatsoever.

The Tissue Factory is going to support the development of the OS-REp model and the setting of a new standard in epidermal tissue reconstruction, since this is going to allow the production of reproducible epidermal skin equivalents of constant high quality in independent laboratories worldwide.

Fraunhofer Fullthickness Human Skin Equivalent

Our ip-protected fullthickness skin model is constructed using human keratinocytes and fibroblasts embedded in a specific collagen scaffold. Keratinocytes are cultivated in special media and differentiate to a multi-layered epidermis with a well stratified horny layer (stratum corneum) holding an important barrier function for the penetration of substances through the skin. This distinct double-layer construction of the skin equivalent affords the opportunity to investigate different sorts of interaction between epidermal and cutaneous cells. This opens up various application areas in testing the toxic effects of new substances such as corrosion and irritation of the skin or photoxicity. Besides, the effects of active ingredients in cosmetic or pharmaceutical products can be tested in vitro using our fullthickness human skin equivalent.

Our fullthickness human skin equivalent has been granted an international patent (WO/2001/092477) and is officially accredited for in vitro biocompatibility testing of medical devices (DIN ISO 10993-5).

Take a closer look at the production process of the fullthickness model in the following pages!

MAIN MODULES OF THE PRODUCTION SYSTEM

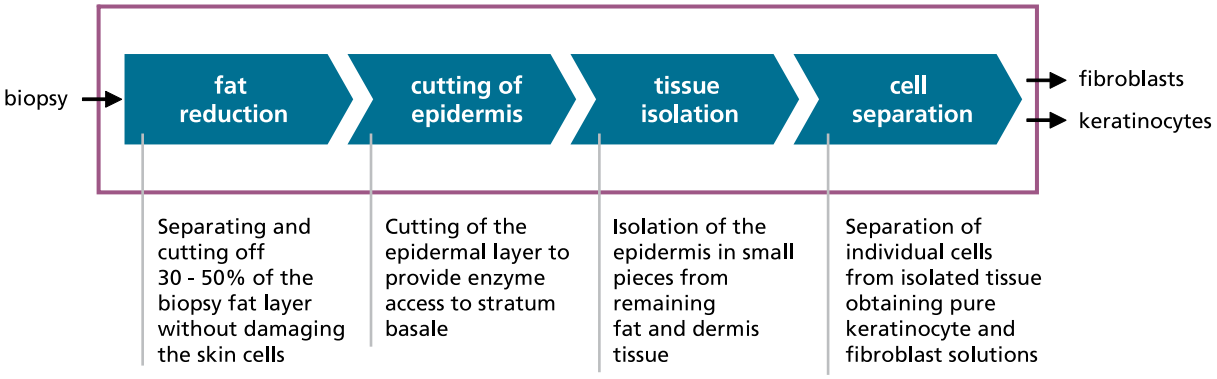
Example process for the production of our fullthickness human skin model

CELL EXTRACTION MODULE

Extraction of primary fibroblasts and keratinocytes from human skin biopsies

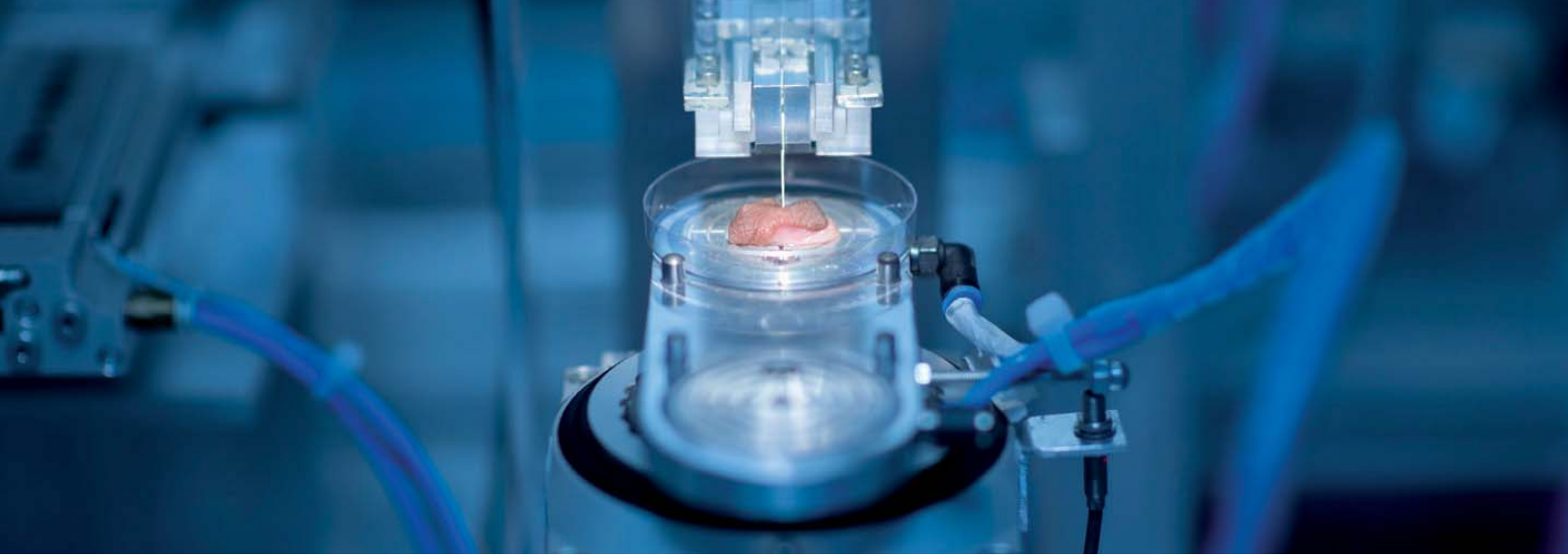
In order to provide the required cell material for cell proliferation and for the subsequent tissue structure build-up, skin cells have to be extracted and isolated from human biopsies. As the quality and amount of producible skin equivalents is directly dependent on the amount and vitality of primary keratinocytes and fibroblasts, efficient downstream processing requires maximized proliferation rates as well as large quantities of vital cells in pure solutions. While these requirements can only be fulfilled by an automated process, the wide range of sizes and lack of structural definition make human biopsies a challenging material for automated processing.

We have developed a novel process chain comprising a combination of specific mechanical and enzymatic applications, focusing on high throughput and maximized efficiency while at the same time maintaining high cell vitality rates.



Specifications

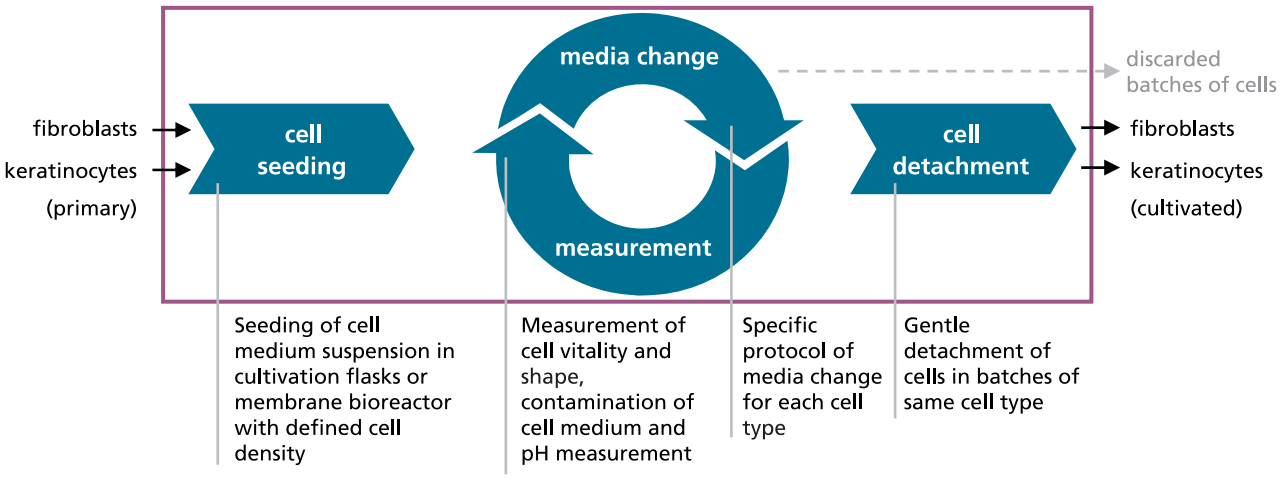
- Fully automated cell extraction from skin tissue
- Wide range of processable biopsy sizes and shapes
- Increased cell output and vitality compared to conventional manual lab process
- High throughput of up to 15 biopsies per day



CELL EXPANSION MODULE

Proliferation of primary fibroblasts and keratinocytes

The next step in the production process is the cultivation of the extracted primary keratinocytes and fibroblasts in closed cell culture flasks. A newly developed, highly flexible membrane bioreactor in standard MTP format facilitates droplet seeding, steady media flow, online proliferation density and contamination measurements as well as enzyme-free cell detachment. With a capacity of 500 flasks/bioreactors, the cells are treated according to a cultivation protocol that was specifically adapted for the fully automated process. The effect of the inherent donor dependency of the batches on cell proliferation is reduced significantly by the 14-days cultivation protocol. Source material contamination is detected by regular, multi-parametric measurements before and after media changes. In the case of positive result, the concerned flask (batch) is discarded and a fully sterile setup is restored by multi-level cleaning protocols. In consequence, the cell expansion module acts as a filter for batch homogeneity and additional quality assurance with regard to the subsequent tissue build-up.



Specifications

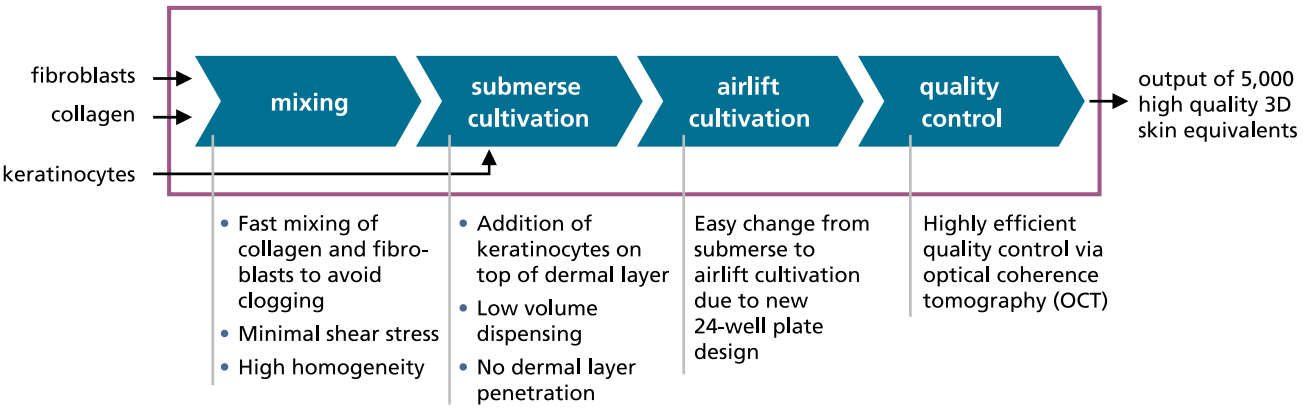
- Total capacity of 500 cultivation flasks in MTP format
- Monthly throughput of 375 million fibroblasts and 750 million keratinocytes
- Non-invasive cell vitality and contamination measurements as well as multi-level cleaning protocols
- Membrane bioreactor in standard MTP format

TISSUE CULTIVATION MODULE

Build-up and cultivation of 3D skin equivalents

The build-up and cultivation of fully functional 3D skin equivalents is the final production step after cell extraction and expansion. Carrying out this process without any human intervention is a challenging task with respect to process control and automation technology. It requires reliable handling and mixing of dispensed cells and other liquids with heterogeneous time and process dependent properties. Reproducible application of these liquids into tissue cultivation inserts requires precise positioning and dynamic control of active and passive dosing systems.

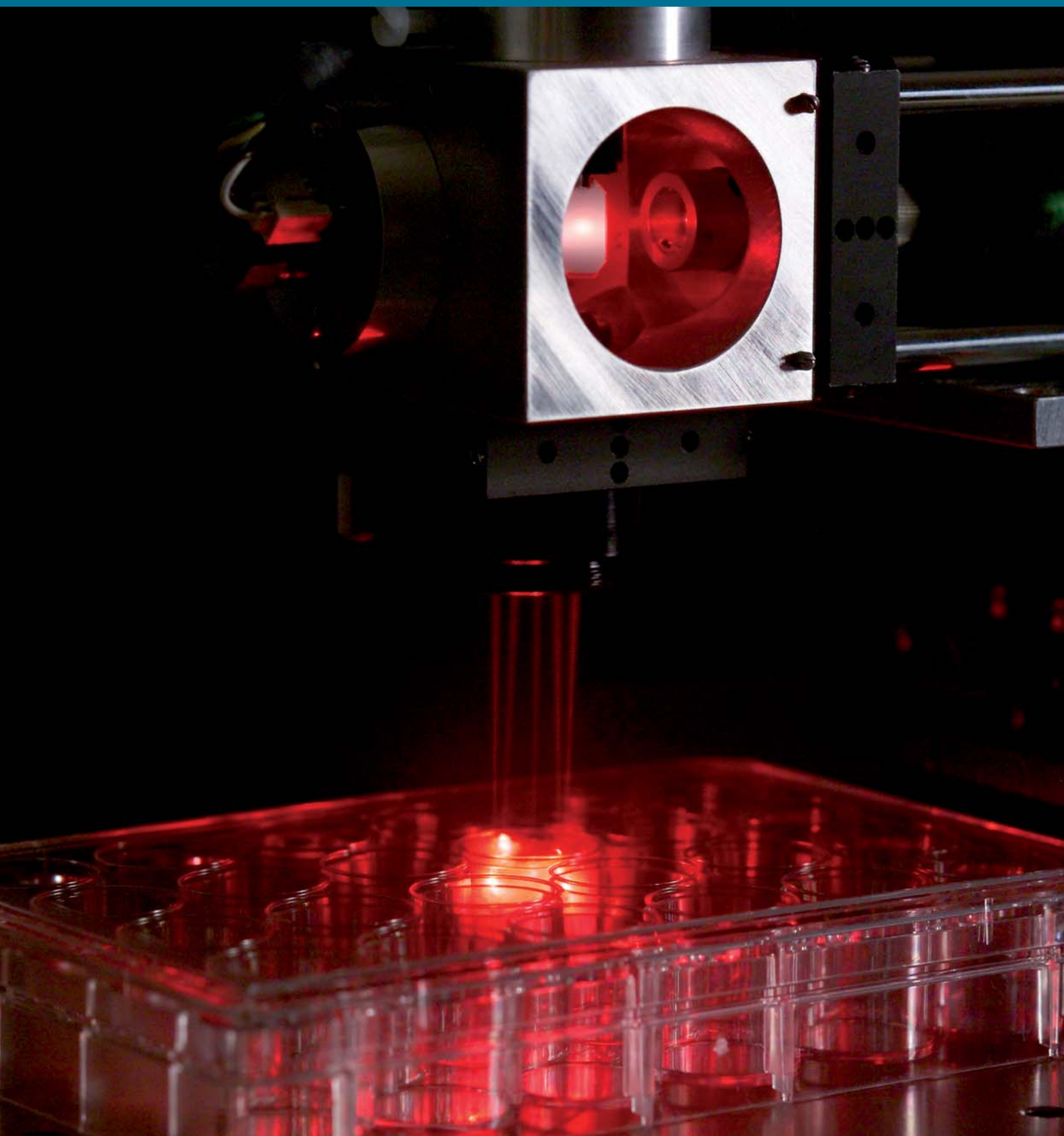
To ease process automation we have developed a new 24-well plate concept with integrated cultivation inserts, leading to a reduction of the total number of handling steps compared to the manual process. Additionally, distinct liquid handling procedures are eased considerably, thus enhancing overall process reliability. Thanks to the novel 24-well plate design, initial growth medium application as well as consecutive media changes every other day can be carried out without any interaction between dosing devices and tissue cultivation inserts.



Specifications

- Total capacity for up to 240 full loaded 24-well plates
- Maximum throughput of 5,000 fully functional skin equiva-lents per month
- Each skin equivalent is treated exactly the same way
- Minimized contamination risk due to fully automated process
- 100 % quality control before shipment of skin equivalents

QUALITY CONTROL



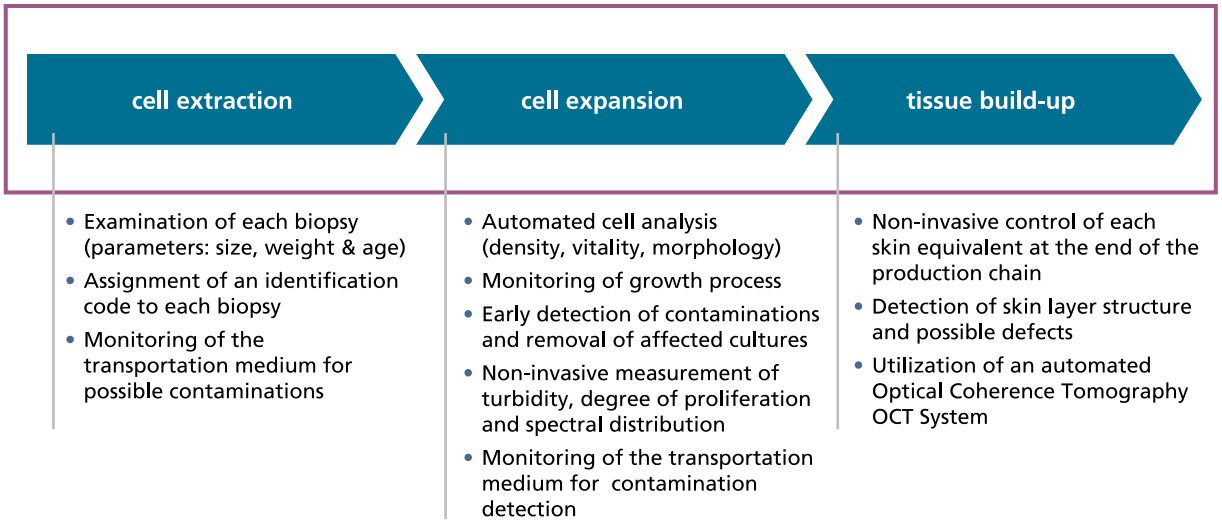
SMART SENSOR SOLUTIONS

Monitoring of the production process

With the aim to guarantee a high quality product, we have developed a broad spectrum of smart sensors to monitor the production process and perform effective quality controls for each individual skin equivalent before leaving the tissue factory. With this sensor network, growth parameters of cells and tissue are captured during the cultivation processes and emerging contaminations of cultivation media are detected in an early stage. With the implementation of quality management systems to the concept, layout and performance of our tissue factory, a high level of process reliability is achieved.

This includes a concept for sterility, a widely GMP-compliant system design, a strategy to minimize error rates and reliable information management.

Monitoring and controlling the automated production of skin equivalents requires the integration of a wide spectrum of metrological instruments into the production process. In order to guarantee superior quality of the end product, every single ready-to-ship skin equivalent finally has to be tested for defects and inhomogeneities such as the concentration of air bubbles and layer geometry.



Specifications

- Early detection of contaminations by analysis of pH, optical density (OD), dissolved oxygen (DO), chirality and spectral distribution of the cell culture media for individual bioreactors / cell culture flasks
- Non-invasive determination of proliferation progress for optimum cell throughput
- Monitoring of cell quantity and vitality for optimization of the production processes
- Non-invasive quality inspection based on optical coherence tomography for each skin equivalent

INTERESTED?

BUSINESS OPPORTUNITIES

We are looking for partners!

Our objective to develop and demonstrate a novel production system which for the first time worldwide enables a large-scale supply of human tissue engineered products has generated considerable interest throughout media and industry. Up to now, our project team has fostered several new product and process technologies for which we have filed comprehensive patent protection. Currently, a total number of nine international patents are pending.

If you are interested in

- initiating R&D cooperations,
- obtaining product or production licenses,
- performing substance tests with skin equivalents,
- automation solutions customized to meet your specific requirements,

or if you have other opportunities in mind for leveraging the Tissue Factory, feel free to contact us in order to discuss future perspectives.

CONTACT INFORMATION

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THE FRAUNHOFER-GESELLSCHAFT

Transferring research results to industry

Research of practical utility lies at the heart of all activities pursued by the Fraunhofer-Gesellschaft. Founded in 1949, the research organization undertakes applied research that drives economic development and serves the wider benefit of society. Its services are solicited by customers and contractual partners in industry, the service sector and public administration.

At present, the Fraunhofer-Gesellschaft maintains more than 80 research units in Germany, including 60 Fraunhofer Institutes. The majority of more than 20,000 staff are qualified scientists and engineers, who work with an annual research budget of $2.3 billion (€1.8 billion). Of this sum, more than $1.9 billion (€1.5 billion) is generated through contract research. Two thirds of the Fraunhofer-Gesellschaft's contract research revenue is derived from contracts with industry and from publicly financed research projects.

The Fraunhofer-Gesellschaft is a recognized non-profit organization that takes its name from Joseph von Fraunhofer, the renowned Munich researcher, inventor and entrepreneur.

PARTNER INSTITUTES

The interdisciplinary project team consists of over 25 scientists from the fields of biology, chemistry, physics, mechanical engineering and computer science. Biological aspects are covered by the two Fraunhofer Institutes IGB and IZI, whereas the engineering part is covered by the two Fraunhofer Institutes IPA and IPT.

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