

Human tissue-engineered products

-

Today's markets and future prospects

A synthesis report prepared by
Anne-Katrin Bock, Dolores Ibarreta, Emilio Rodriguez-Cerezo
(**IPTS-JRC**)

based on studies conducted by
B. Hüsing, B. Bühlren, S. Gaisser (Fraunhofer ISI, Germany)
J. Senker, S. Mahdi (SPRU, UK)
C.J. Kirkpatrick (University of Mainz, Germany)

October 2003



EUROPEAN COMMISSION
JOINT RESEARCH CENTRE

**European
Science and
Technology
Observatory**

European Commission

Joint Research Centre (DG JRC)

Institute for Prospective Technological Studies

<http://www.jrc.es>

Legal notice

Neither the European Commission nor any person acting on behalf of the Commission is responsible for the use which might be made of the following information.

Report EUR 21000 EN

© European Communities, 2003

Reproduction is authorised provided the source is acknowledged.

PREFACE

Tissue engineering is a multidisciplinary, young and emerging biotechnology sector, which promises to change medical practice profoundly, regenerating diseased tissues and organs instead of just repairing them. There is a lot of hope connected to this novel biotechnology development concerning improved treatment possibilities, enhanced quality of life of patients and not least the ability to overcome in the long run the constant shortage of donor organs for transplantation. But expectations are also high regarding the potential markets these products could cover. Estimations are in the range of 4 to 400 billion Euro per year world-wide.

To fulfil these promises several challenges concerning scientific, technological and also societal issues need to be met. Basic research is still required for elucidation of fundamental processes in for example cell differentiation and growth. However, the first tissue-engineered products have already reached the markets, a lot more are in the pipeline, and many companies in the USA as well as in Europe are involved in this area. Yet there has been little information on the situation of tissue engineering in Europe.

This report is the first providing an overview on tissue engineering companies in Europe, commercialised products, research activities, and factors influencing the development of the sector. It is the result of a study that was carried out in the context of development of a specific European legislative framework for human tissues and tissue-engineered products. Tissue-engineered products differ in many ways from medical devices and pharmaceuticals. For that reason they are not appropriately covered by current European legislation. The European Commission is approaching this issue via new European legislation. A directive on standards for quality and safety of human tissues and cells is already in the decision process of the European institutions, a regulation covering human tissue-engineered products is currently being developed.

In the framework of the Lisbon strategy, a European strategy and an action plan for life sciences and biotechnology were developed to exploit the full potential of biotechnology and to strengthen the sector's competitiveness while ensuring environmental and consumer safety and consistency with common values and ethical principles (European Commission, 2002). It gives a mandate to the DG Joint Research Centre's Institute for Prospective Technological Studies (DG JRC-IPTS) for carrying out biotechnology foresight with the objective of identifying newly emerging issues and possible proactive policy measures (Action 29).

Against this background and on request of DG Enterprise from July 2002 DG JRC-IPTS initiated a study on European commercial and research activities in the field of tissue engineering. It was executed by the IPTS and the European Science and Technology Observatory (ESTO) between December 2002 and May 2003. The Fraunhofer Institute for Systems and Innovation Research, Germany, carried out research on tissue-engineered products and companies and cost-effectiveness of tissue engineering treatments. SPRU Science and Technology Policy Research of the University of Sussex, UK, did the review of today's and possible future research activities and legal and socio-economic issues. Professor James Kirkpatrick, University of Mainz, Germany, an external partner, supported the study regarding scientific issues.

This synthesis report is mainly based on the final results of the following four working packages (WP):

- WP1: Analysis of the actual market situation – Mapping of industry and products
- WP2: Comparison of tissue engineering treatment costs with conventional treatment
- WP3: Research activities and future developments of human tissue engineering in Europe and the USA
- WP4: Legal situation and socio-economic impacts of tissue engineering

The complete contents of these working packages are available on the IPTS web page (<http://lifesciences.jrc.es>).

Preface

This report and the co-ordination of the study was done by Anne-Katrin Bock, Dolores Ibarreta and Emilio Rodriguez-Cerezo, DG JRC-IPTS.

Whilst IPTS is grateful for the help and input received from Didier Bouis, DG Enterprise, and the ESTO network, and would like to thank especially the experts who took the time to be interviewed, responsibility for the content of this synthesis report rests solely with IPTS.

CONTENTS

PREFACE	I
1 INTRODUCTION	1
2 HUMAN TISSUE ENGINEERING: PRODUCTS ON THE MARKET	3
2.1 Tissue-engineered skin products	3
2.2 Tissue-engineered cartilage products	5
2.3 Tissue-engineered bone products	7
2.4 Products in the pipeline.....	8
3 SCIENCE AND TECHNOLOGY – RESEARCH AND EUROPEAN ACTORS	13
3.1 Current research areas	13
3.1.1 Biomaterials	13
3.1.2 Cells	15
3.1.3 Biomolecules.....	16
3.1.4 Engineering design and manufacturing aspects	17
3.1.5 Bioinformatics to support tissue engineering.....	19
3.2 Developments expected in the coming ten years	19
3.3 Characterisation of the European tissue engineering research community	20
4 COMMERCIAL TISSUE ENGINEERING IN EUROPE – COMPANIES AND INFLUENCING FACTORS	23
4.1 Characteristics of European tissue engineering companies.....	23
4.2 Factors influencing the development of the commercial tissue engineering sector in Europe.	25
4.2.1 Regulatory framework	25
4.2.2 Cost effectiveness	28
4.2.3 Business models and strategies	30
4.2.4 Research support.....	32
4.2.5 Ethical issues.....	32
4.2.6 Employment and training needs.....	33
5 CONCLUSIONS	35
REFERENCES	39
ANNEX 1: TISSUE ENGINEERING COMPANIES IN EUROPE	45
ANNEX 2: METHODOLOGIES APPLIED	48

List of figures

Figure 3.1: Tissue engineering and the interactions of biomaterials, cells and biomolecules.....	13
Figure 3.2: Tissue engineering publications 1991 - 2002.....	20
Figure 3.3: Domestic public/private co-authorships 1992-2002	21
Figure 4.1: Tissue engineering companies in Europe.....	23
Figure 4.2: Company size of European tissue engineering companies	24

List of tables

Table 2.1: Estimation of sales figures for selected tissue-engineered skin products.....	4
Table 2.2: Maximum market potential for tissue-engineered skin products	5
Table 2.3: Sales figures of autologous chondrocyte transplants (ACT, for knee injuries).....	6
Table 2.4: Market sizes correlated with cartilage defects/cartilage repair	7
Table 4.1: Business models for pharmaceuticals, medical devices and tissue engineering products.....	30
Table A: Tissue engineering companies in Europe	45
Table B: The exchange rates used for conversion to Euro	49

1 INTRODUCTION

Tissue and organ damage and loss are normally treated using allogeneic transplants, the patient's own tissue, medical devices and/or pharmaceuticals. All these treatments have their inherent shortcomings, replacing the diseased organ or tissue only imperfectly or, in the case of transplants, being available only in small numbers compared to the demand and also requiring immunosuppressive treatment.

With the advancement of the concept of "regenerative medicine" and in particular the field of tissue engineering a completely new form of medical treatment can be envisaged with the potential to change medical practice profoundly. Tissue engineering aims at regenerating the diseased tissues (and organs as a future perspective) *in vitro* or through a combination of *in vitro* and *in vivo* processes and implanting the product at the diseased site to achieve full functionality. Improved healing processes and a higher quality of life are expected results, probably leading to lower costs of treatment in the long term.

Tissue engineering is an essentially multidisciplinary field, combining various aspects of medicine, materials science, engineering and biology. There is not yet a specific definition, resulting in differences of what is understood as "tissue engineering". Often cell therapy or tissue regeneration *in vivo* is included. There are several definitions with a broader or more focused scope. The term tissue engineering was first coined in 1988 at a meeting of the US National Science Foundation as "*the application of the principles and methods of engineering and the life sciences towards the fundamental understanding of structure/function relationships in normal and pathological mammalian tissues and the development of biological substitutes to restore, maintain or improve functions*"¹ (Chapekar, 2000). The Scientific Committee on Medicinal Products and Medical Devices (SCMPMD) of the European Commission's DG SANCO, in its opinion from October 2001 used the following, narrower definition: "*Tissue engineering is the regeneration of biological tissue through the use of cells, with the aid of supporting structures and/or biomolecules*" (European Commission, 2001). This definition includes products that combine cells with (degradable) matrices or scaffolds plus, if necessary, biomolecules such as growth factors. In the context of regulatory issues, the SCMPMD anticipates the need for a science-based, stringent definition for tissue engineering to enable its differentiation from medical devices, medicinal products and cell therapy. The SCMPMD definition has been used in the present study, excluding xenogeneic cells and tissues, direct transplantation, pure cell therapy (e.g. injection of bone marrow stem cells to repair heart tissue after infarction), and *in vivo* tissue regeneration (implanting acellular biomaterials with bioactive behaviour to enhance e.g. bone repair). However, boundaries are often blurred.

Tissue-engineered products might act primarily in a structural way (e.g. bones, cartilage), thus resembling medical devices. Also, they might act like pharmaceuticals (e.g. bioartificial liver or pancreas), or in both ways. Their special characteristics and the associated risks concerning the materials involved, production process and mode of action led the SCMPMD to the conclusion that none of the existing European regulatory frameworks (i.e. Directive on Medical Devices 93/42/EEC², Directive on Medicinal Products 2001/83/EC³) in their current form encompass tissue-engineered products appropriately (European Commission, 2001). A specific regulatory framework was deemed necessary for these products. As the first products have already reached the market, some EU Member States, realising the need for regulation, have started to develop national rules.

¹ Langer and Vacanti (1993) defined tissue engineering in 1993 as "*an interdisciplinary field that applies the principles of engineering and life sciences toward the development of biological substitutes that restore, maintain or improve tissue or organ function.*" Williams (1999) defined tissue engineering as the "*persuasion of the body to heal itself, through the delivery to the appropriate sites of molecular signals, cells and supporting structures.*"

² Official Journal L 169 12/07/1993, p. 0001-0043

³ Official Journal L 311 28/11/2001, p. 0067-0128

Introduction

At the EU level initiatives for a complementary regulatory framework have also been started. A proposal for a Directive on tissue banking (with the aim of assuring quality and safety of human tissues and cells for medical purposes) was published in June 2002 (European Commission, 2002a) and is currently discussed by the EU institutions. This directive is supposed to cover only the donation, procurement and testing of tissue in the context of tissue engineering, provided processing, preservation, storage and distribution are covered by other Community legislation. Regarding the need for a separate legislative framework covering the marketing of human tissue-engineered products to safeguard consumer and user protection and enable a common market for these products, a public consultation was carried out between June and September 2002 by the European Commission. A regulation is currently under preparation by Directorate-General Enterprise.

Because of the very recent development of tissue engineering, little information is available for this sector for the European Union and acceding countries. Against this background the Institute for Prospective Technological Studies (IPTS), DG Joint Research Centre, was requested by DG Enterprise in July 2002 to carry out a study on tissue engineering in Europe with the following objectives:

- To provide an overview and analysis of tissue engineering products and companies in the EU and acceding countries
- To provide information on the state of the art and future directions of tissue engineering research
- To identify and analyse factors possibly influencing tissue engineering development.

The study was co-ordinated by DG JRC-IPTS and was executed by the European Science and Technology Observatory (ESTO) between December 2002 and May 2003. This synthesis report is mainly based on the final reports resulting from four different working packages. The complete content of these working packages are available on the IPTS web page (<http://lifesciences.jrc.es>).

2 HUMAN TISSUE ENGINEERING: PRODUCTS ON THE MARKET⁴

Increasing knowledge on cellular and molecular processes governing tissue growth is applied to grow human tissue *in vitro*, aiming to develop better medical treatments and to overcome transplantable organ and tissue shortage. Commercial tissue engineering is still in its initial phase: the first tissue-engineered product (cartilage) was approved for marketing in 1996 in the USA. Since then, progress in tissue engineering has resulted in several commercialised products for skin substitution, knee cartilage repair (autologous chondrocyte transplantation, ACT) and a few bone repair products from several companies in Europe and the USA. Although very special, these products are relatively simple, consisting of few cell types and not needing vascularisation during growth. They yet have to gain broad acceptance in clinical practice. More advanced products are under development, also covering other application areas such as cardiovascular diseases or bioartificial organs (see also Chapter 3 on research activities).

2.1 Tissue-engineered skin products

Human skin is a complex organ with several tasks to fulfil:

- Thermoregulation
- Microbial defence (mechanical and immune)
- Desiccation barrier
- Mechanical defence and wound repair.

During the past 30 years many attempts have been made to develop products that support wound healing and could be used as substitutes for skin in severe cases. These substitutes (permanent or temporary) should ideally fulfil additional requirements to the ones listed above (Schulz et al., 2000):

- Elicit regeneration response from the wound bed without causing inflammation or rejection
- Be durable and elastic to provide normal function and cosmetic appearance
- Have pigmentation and control of contraction to resemble natural skin
- Be available for acute cases and be easy to use.

Indications for skin substitutes are burns, chronic wounds (ulcers), plastic and aesthetic surgery and defects in oral mucosa. Severe burns, with about 150 patients per year in Western Europe estimated to require treatment with tissue-engineered skin substitutes, present a much smaller market than chronic wounds with a significantly higher prevalence (Jones et al., 2002). Chronic wounds (wounds which do not heal within 6 weeks) include pressure ulcers, venous ulcers and diabetic ulcers and can persist for several years, requiring cost-intensive treatments. Underlying diseases, which might result in chronic wounds, such as diabetes or venous diseases, are increasing due to changing life styles and are also age-related. For Germany alone it is estimated that 2 to 3 million people suffer from chronic wounds, with direct and indirect costs of more than 1 billion € (Augustin et al., 1999; Landesbank Baden-Württemberg Equity Research, 2001). Other indications for skin substitutes are for example the pigmentation disorder vitiligo or dental surgery.

Currently there are about two dozen tissue-engineered skin substitutes on the market in Europe and the USA. In general they consist of a sheet-like matrix (made of e.g. collagen, hyaluronic acid, biodegradable synthetic or semisynthetic polymers) and different skin cells (keratinocytes and/or fibroblasts and/or melanocytes). American companies tend to base their products on allogeneic cells, while European companies, probably because of regulatory reasons (see Chapter 4.2), focus on autologous cell-based products. The first products on the market were approved for the treatment of severe, full thickness burns: Epicel (Genzyme Biosurgery, USA), Integra (Integra Life Sciences, USA) and Transcyte (marketed by Smith & Nephew, UK). There

⁴ See also WPI "Analysis of the actual market situation – Mapping of industry and products"

Human tissue engineering: Products on the market

are many more products on the market for the treatment of chronic wounds, for example: Apligraf (developed and manufactured by Organogenesis (USA), marketed by Novartis (CH/USA) until June 2003), Dermagraft (developed by Advanced Tissue Sciences, marketed by Smith & Nephew, UK), Hyalograft™ 3D and Laserskin™ (Fidia Advanced Biopolymers, Italy), BioSeed-S (BioTissueTechnologies, Germany; marketed by Baxter Healthcare), autologous Autoderm and allogeneic CryoCeal (XCELLentis, Belgium), Epidex (production stopped by Modex Therapeutics (CH), product licensed to Autoderm (Germany) in spring 2003), Collatamp (Innocoll GmbH, Germany), Epibase (Laboratoire Genevrier, France), CellActiveSkin (production stopped in late 2002 by IsoTis SA (NL), because the product was not profitable), OrCell (Ortec, USA) and VivoDerm (Convatec, USA).

Due to difficulties with reimbursement by European health insurance schemes of treatments based on tissue-engineered skin products (see Chapter 4.2) companies target increasingly the “self-payer” patients’ segment, such as aesthetic surgery. Products that fall in this category are for example BioSeedM and MelanoSeed, both produced by BioTissueTechnologies (Germany).

Tissue-engineered skin products also have applications as skin models for *in vitro* testing in toxicology, pharmacology and cosmetics. Companies active in this segment are e.g. Biopredic and SkinEthicLaboratories, France.

Actual sales figures for selected tissue-engineered skin products are presented in Table 2.1. Data have been obtained from literature and through estimations on the basis of companies’ annual reports. Although not all products presently on the market are covered, the total estimated annual sales for tissue-engineered skin products world-wide are likely below 20 million €

Table 2.1: Estimation of sales figures for selected tissue-engineered skin products

Trade name	Company	Year	Sales (M €)
Apligraf	Organogenesis Inc (USA), Novartis (USA/CH)	2000	12
Dermagraft	Advanced Tissue Sciences (USA) ¹ , Smith & Nephew (UK)	2002	4.405
CellActiveSkin	IsoTis (NL)	2002	0.545
Epidex	Modex Therapeutics (CH)	2002	0.157
BioSeedS, BioSeedM, MelanoSeed	BioTissueTechnologies (D)	2002	0.450
Epicel	Genzyme Biosurgery (USA)	2001	n.a. 75 patients treated annually worldwide
Total			17.557

1: Advanced Tissue Sciences (USA); had a marketing agreement with Smith & Nephew for Dermagraft and Transcyte; both products were completely taken over by Smith & Nephew in 2002 after Advanced Tissue Sciences had to file for bankruptcy.

Source: Fraunhofer ISI

The sales figures estimated in Table 2.1 are much lower than the estimations for market sizes for tissue-engineered skin products (Table 2.2). These markets range from 300 million € to 800 million € for the year 2001. Chapter 4.2 will discuss some of the factors influencing the market

situation of tissue-engineered products. Smith & Nephew, the market leader in wound care, assumes that the active wound care market, to which tissue-engineered skin belongs, will grow annually by 28%, having a share of the overall wound market in 2002 of about 10% (392 million €). This growth will be based on the replacement of traditional wound management products, resulting in overall reduction of healing costs while increasing the share of material costs and reducing the cost share of nursing time (Dick, 2002).

Table 2.2: Maximum market potential for tissue-engineered skin products

Market	Market Size 2001 (M €)	Region	Source
Global wound management market potential	6,250	world	Landesbank Baden-Württemberg Equity Research, 2001
Maximum market potential for tissue-engineered skin, only applicable to chronic wounds	625	world	Landesbank Baden-Württemberg Equity Research, 2001
Global market for skin replacement products for wound repair	800	world	Russell & Cross, 2001
Market for skin substitutes	300	USA	Russell & Cross, 2001

Source: Fraunhofer ISI

2.2 Tissue-engineered cartilage products

Cartilage can be found in the human body as “unstressed cartilage”, for example in the ear and nose, or as “stressed” cartilage as for example in joints or intervertebral discs. Cartilage is considered to have a limited capacity to regenerate itself, thus injuries do not heal easily.

Tissue engineering applications in the field of unstressed cartilage are reduced to singular cases, engineering the outer ear or the nasal septum, or the more famous case of building a rib cage using cartilage cells grown on a scaffold in 1994 in the USA (Arnst & Carey, 1998). This technology has not yet become part of regular clinical methods.

At present, tissue-engineered cartilage products targeting defects in hyaline cartilage are commercially more important. Since 1994, autologous chondrocyte transplantation (ACT) also termed autologous chondrocyte implantation (ACI) is available for traumatic knee joint injuries. This technique and its several modifications are currently the most important clinical application of cartilage tissue-engineered products. Research is being carried out to extend the applications of tissue-engineered cartilage to joints other than the knee, to treat other knee cartilage defects (e.g. arthritis), to develop combined tissue-engineered cartilage and bone products, and to develop treatments for intervertebral disc damage.

The classic ACT procedure includes a biopsy to obtain healthy chondrocytes from the patient’s knee. These cells are then expanded and cultivated for about three weeks and transferred to the damaged knee in an open knee surgical procedure. The cells are covered with a periosteal flap, which is sutured over the cartilage lesion. After the surgery, 2 to 6 months are needed for full regeneration of the cartilage. Instead of a periosteal flap, an artificial cover made of e.g. collagen or hyaluronic acid can be used. Another new variant uses chondrocytes cultured on a three-dimensional, biodegradable scaffold (e.g. matrix-induced ACT). The scaffold, cut to the required size, is inserted into the lesion and fixed with anchoring stitches. This method opens up the possibility of arthroscopic surgery and does not need the cover, thus simplifying the surgery.

Human tissue engineering: Products on the market

Several companies are active in this field of tissue engineering, the most important being Genzyme Biosurgery (USA), Fidia Advanced Biomaterials (Italy), Verigen (Germany), co.don (Germany), BioTissueTechnologies (Germany) and TETEC AG (Germany). Sales figures for some ACT products are listed in Table 2.3. Data have been collected from expert interviews, literature and companies' annual reports. Currently, total sales world-wide are unlikely to exceed 40 million €per year.

Table 2.3: Sales figures for autologous chondrocyte transplants (ACT, for knee injuries)

Country	ACT/year performed	Calculated sales volume *	Important companies/products	Sales information from important companies
USA	2,000-3,000	16 – 24 M€	Genzyme Biosurgery/ Carticel®	Sales of Carticel ®: Sales 2001: 18.4 M US-\$ Sales 2002: 20.4 M US-\$
Germany	600	3 M€	Verigen/ ACI/MACI/MACI-A co.don/co.don chondrotransplant® BioTissue Technologies/ BioSeedC®	Sales of co.don chondrotransplant®: 2000: 550,000 €(ca. 100 transplants plus 100 without reimbursement), 2001: 1,000,000 €(260 transplants plus 80 without reimbursement) Sales by BioTissueTechnologies 2002: 500.000 € ca. 100 transplants
UK	300-850**	1.5-4.3 M€	Verigen/ ACI/MACI/MACI-A	
Italy	300-400	1.5-2 M€	Fidia Advanced Biomaterials/ HYALOGRAFT® C	
Spain	40	187,000€	IsoTis/CellActive Cart	Sales of IsoTis' CellActive Cart: 187,000 €in 2002
Total	3,240-4,850	22.2-33.3 M€		

*assuming retail prices of €5,000 /autologous chondrocyte transplant in Europe and €8,000/transplant in USA. These costs do not include costs for surgery and rehabilitation.
**Estimates by NICE of the number of potential ACT operations in England and Wales

Source: Fraunhofer ISI

Table 2.4 shows estimated market sizes for treatments associated with cartilage defects. Similar to the situation for skin products, the potential market sizes are significant. They are in the range of several billion €and contrast heavily with the estimations of actual sales. One reason for this discrepancy is the present restriction of ACT treatment to traumatic knee joint cartilage defects. Additionally, the ACT treatment requires co-operation of the patient during the rehabilitation time of 2 to 6 months when the knee cannot be fully used. A full knee prosthesis can bear weight already a few days after the surgery and is thus the preferred option for elderly patients for whom the life span of the prosthesis correlates with their life expectancy. In several European countries ACT treatment is not generally reimbursed by health insurance.

New developments in ACT treatment such as the matrix-induced ACT might open up new application fields for ACT and thus open larger, attractive market segments, which are not accessible for cell suspensions (e.g. osteoarthritis treatment in the knee, treatment of cartilage defects in other joints such as hip or shoulder). Furthermore matrix-induced ACT simplifies the knee surgery and might therefore become more attractive. The annual market for effective new

repair techniques for the USA has been estimated between 300 million € and 1 billion € which would represent a significant increase compared to the present market.

Table 2.4: Market sizes correlated with cartilage defects/cartilage repair

Region	Market size (€)	Year	Remarks	Source
Europe	2 billion	1999	Market value for joint implants (prosthesis costs only)	Biomet Merck
World	1.5 billion	1999	Market value for knee implants (prosthesis costs only)	Datamonitor
USA	5.2 billion	2001	annual spending for total knee replacement; estimation based on incidence (200.000 patients/year) and cost per treatment (26.000 US-\$)	Russell & Cross, 2001
World	6.5 billion	2001	market potential of surgical procedures for cartilage regeneration	Landesbank Baden-Württemberg Equity Research, 2001
World	25 billion	2011	market potential of surgical procedures for cartilage regeneration	Landesbank Baden-Württemberg Equity Research, 2001

Source: Fraunhofer ISI

2.3 Tissue-engineered bone products

Tissue-engineered bone products could be used for treatment of bone fractures, jawbone surgery and periodontal surgery as well as for the treatment of osteoporosis and bone tumours. For small bone fractures standard therapies are available and fulfil clinical needs satisfactorily. Gypsum and plaster as well as nailing, screws and plates are well known materials. For larger defects autologous bone grafts give the best results. If these are not available, allogeneic or xenogeneic grafts or synthetic bone material can be used, but with increased risk of infection or immunogenic rejection. Tissue-engineered bone competes with the established treatments in the case of small bone lesions. The treatment of larger lesions would also fill a treatment gap, but no tissue-engineered bone products are yet available that could be used for this indication. Additionally, most bone defects result from trauma and accident and need acute treatment, so that there would not be enough time to produce an autologous tissue-engineered bone product.

These might be the reasons why only a few tissue-engineered bone products are on the market. Most of them are not cell-biomaterial combination products and thus do not fully comply with the definition of tissue engineering used in this study. Accordingly, only a few companies are active in this field:

BioTissueTechnologies (Germany) produces BioSeed-Oral Bone for applications in jawbone surgery. Annual sales are in the range of 250,000 € Co.don (Germany) produces the autologous product co.don osteotransplant for several applications in bone surgery (sales figures were not available). Osiris Therapeutics (USA) developed Osteocel, a product derived from human mesenchymal stem cells from bone marrow for promoting new bone formation. The product is still in the development phase. CellFactors (UK) have an osteoinductive product called Skeletex, a cell-free product comprising collagen and growth factors. IsoTis SA (CH/NL) has stopped the development of the autologous bone product VivescOs and now focuses on the marketing of the osteoconductive scaffold product OsSatura, which the company considers to be economically much more favourable than the more expensive tissue engineering option. The

additional costs of the tissue engineering option are considered not to be justified by the additional therapeutic benefit. Several other companies are producing growth factors and bone morphogenic proteins, others offer biomaterials and synthetic bone fillers.

The worldwide market for bone replacement and repair is estimated at about 300 million € including autologous, allogeneic, xenogeneic and synthetic bone materials (Concord Corporate Finance Research, 2002; IsoTis Corporate Communications and Investor Relations, 2003). For the time being, due to competing products and not sufficiently advanced technology in bone tissue engineering the application field seems to be focussing on niche markets such as dental or maxillofacial surgery.

2.4 Products in the pipeline

Research on tissue engineering applications is on-going and several developments have been identified leading to more sophisticated products, some of them already in clinical trials. Other areas as those already mentioned, such as cardiovascular diseases or diseases concerning the nervous system are the focus of many research activities, with products still being far from the market.

Skin, cartilage and bone

The next steps in product development for skin substitutes include the integration of growth factors and the construction of a complete skin, consisting of the dermis with its principal cellular components, fibroblasts and blood vessels, and also the epidermis. Currently many skin substitutes concentrate on epidermal replacement with keratinocyte sheets. The possibility to grow epithelial layers would also open other applications such as oesophagus, stomach and windpipe repair.

Tissue-engineered cartilage will most probably be extended to three-dimensional constructs with the aim of replacing intervertebral disc (co.don has already the product Chondrotransplant Disc in clinical trials). The treatment of arthritis is envisaged using a combination of chondrocytes and biomaterials in the form of spheres. Mechanical strength of tissue-engineered cartilage, which is lower than for natural cartilage might be improved by providing necessary mechanical stress during tissue growth in the bioreactor.

Bone repair will profit from improved scaffold design, making it possible to provide accurately tailored site-specific scaffolds. Better vascularisation and the use of bone marrow-derived stem cells with ceramic-based scaffolds are regarded as fruitful for progress in bone repair. Ceramics have the advantage of great similarity with the inorganic phase of bone. Products using osteoblasts or bone marrow cells combined with several biomaterials are currently being tested in clinical trials.

Cardiovascular diseases

Cardiovascular diseases (CVD) pose a significant public health problem because they cause about 50% of mortality (240 to 260 death per 100,000 population). Additionally CVD are the leading cause of disease burden with 21.8% of the overall burden of disease and injury. Some 175,000 heart valve replacements per year are carried out worldwide, with sales worth about 830 million € in 2001 (Di Lullo, 2002). 240,000 to 320,000 coronary and peripheral bypass graft surgeries are carried out in Europe annually. For Germany alone, it is estimated that total cost of CVD amounted to 16 billion € in 1990 (Kohlmeier et al., 1993). Developing tissue engineering alternatives for CVD is well underway for heart valves, vessel grafts and heart muscle tissue, but no products have yet reached the market.

Heart valves

Mechanical heart valves and heart valves from human donors or xenogeneic ones, which are used today have several shortcomings (thrombogenicity, limited durability, shortage of supply)

and they are not able to grow, which poses a problem for patients in childhood. Research is being carried out to develop a custom-made heart valve by tissue engineering that avoids the problems of other products and as it is a living replacement, is able to grow with the patient. The right cell source, the best suited scaffold material and structure, as well as bioreactor conditions are under investigation. First trials with large animals (sheep) have been carried out as a proof of principle (Stock et al., 2002). Several US American and European companies are carrying out research on tissue-engineered heart valves.

Blood vessels

Blood vessels are another focus of product development in tissue engineering. Blood vessels are not only essential for applications in e.g. bypass surgery but also for the construction of larger tissues that need vascularisation. Current vessel replacements use either autologous grafted vessels or stents made of Dacron or expanded polytetrafluoroethylene (ePTFE). Autologous grafts are not always available, depending on the patient's health, and stents tend to clog rather quickly when small diameters below 4 mm are used. Development of tissue-engineered blood vessels aims at constructing blood vessels which are able to replace natural vessels regarding vasoactivity, appropriate mechanical properties and no thrombogenic activity (Nerem & Seliktar, 2001). Clinical trials are carried out with stents (synthetic or natural) seeded with autologous endothelial cells. Long term experience has already been published for synthetic polymer vascular grafts (Deutsch et al., 1999; Meinhart et al., 2001) and seeded small-diameter grafts show enhanced clinical performance (Seifalian et al., 2002). In the EU three German tissue engineering companies are active in this field: Vascular Biotech GmbH with cryopreserved allogeneic vessels lined with autologous endothelial cells, co.don and BioTissueTechnologies with endothelialised synthetic vessel grafts.

Other approaches use collagen-based blood vessels grafts, but better results concerning rupture strength have been achieved with biodegradable synthetic polymers. Human smooth muscle cells, genetically engineered to enhance their proliferative capacity, have been grown on a degradable scaffold to form arteries with a diameter of 3 mm (McKee et al., 2003). Tissue-engineered vessels constructed of layers of different cell types are under investigation. Cells are grown in sheet form, then rolled around a mandrel and the construct is cultivated over several weeks to enable the development of a stable tubular form (Nerem & Seliktar, 2001).

Heart muscle tissue

Heart attacks, due to reduced blood supply, result in damaged heart muscle tissue, which impairs the functioning of the heart. Reversal of tissue damage is usually not possible and tissue engineering research focuses on growing patches of heart muscle tissue or, for the time being, on cell therapy-like approaches such as transplantation of healthy cells into the damaged heart area. Different types of cells are applied: hematopoietic stem cells (Stamm et al., 2003; Strauer et al., 2001; clinical trials), embryonic stem cells (Roell et al., 2002; preclinical experiments with rodents), skeletal muscle cells (clinical trials phase II under way, Menasche, 2002) or primary heart muscle cells (Kessler & Byrne, 1999). Research is in the preclinical and early clinical phase. Several US companies (e.g. Genzyme Biosurgery, Diacrin, BioHeart, Osiris) are active in this field.

Peripheral and central nervous system

Diseases or damage of the central nervous system (CNS) could be in principle a very interesting market for tissue-engineered products as there are only few competing conventional treatments available. Potential targets for tissue-engineered products are neurodegenerative diseases, damage of nerve fibres and spinal cord injury, epilepsy, impaired generation of nerve impulses, stroke, and pain. Neurodegenerative diseases, like Alzheimer and Parkinson, are more prevalent in the elderly population, so it can be expected that these diseases will play an increasingly important role in the overall disease spectrum in the future. Over the last 40 years the proportion of elderly people aged 65 years and over in the population of the OECD countries has increased steadily: on average from 8.9% in 1960 to 13.8% in 1999 (OECD, 2001). It is assumed that by 2020 elderly people will represent about 25% of the total population world-wide. The total

(direct and indirect) costs caused by Alzheimer's disease are estimated in the range from 2,470 € to 32,000 € per patient per year (Bloom et al., 2003). In Parkinson's disease, these estimates average 13,800 € per patient (Hagell et al., 2002). The costs are high and the overall health care costs for these diseases are likely to rise due to potentially increasing number of cases given the expected demographic shifts in all countries. The world market for drugs targeting the CNS is estimated in 2000 at about 41 billion € (Informa Pharmaceuticals, 2000), but drugs against neuro-degenerative disorders are not among the top-sellers. An exception are pharmaceuticals against multiple sclerosis with an estimated market volume in 2001 of 2.15 billion €, expected to increase to 3.7 billion € by 2005 (Frost & Sullivan, 2001).

The development of feasible tissue engineering strategies to target the CNS in order to treat major clinical problems such as Parkinson's and Alzheimer's disease is a challenging field. A major hurdle for delivering therapeutic substances to the brain is the blood-brain barrier. One of the current endeavours is the development of nanoparticles to carry a relevant drug or even genetic material to the brain (Kreuter, 2001). Various polymer systems are being developed to act as biomaterial carriers, such as poly(butylcyanoacrylate) (Alyaudtin et al., 2001) and PEGylated poly(cyanoacrylate) (Calvo et al., 2001).

One approach to promote the regeneration of peripheral nerves is to use acellular nerve grafts colonised by Schwann cells, although in a rodent model this approach was not as effective as the use of autologous nerve grafts (Frerichs et al., 2002). A further biomaterial approach is to design a nerve guide from a biodegradable polymer which is colonised by Schwann cells (Schlosshauer et al., 2003). Biodegradable co-polymers of trimethylene carbonate and vareosilon-caprolactone in the form of flexible porous tubes have been synthesised and have been shown to support the growth of human Schwann cells (Pego et al., 2003).

Several companies from the USA and Europe focus for the time being on cell therapies, mainly for Parkinson's disease, Alzheimer's disease, Huntington, stroke and spinal cord injuries. Most of the products are at the preclinical stage. Human adult neural stem cells, human adult bone-marrow-derived stem cells as well as immortalised human cell lines are used to develop novel therapies. Some of the companies active in this field are NeuroNova AB (Sweden), ReNeuron Holding (UK), Cellfactors (UK), ReInnervate Limited (UK), StemCells Inc. (USA), Neuronix Inc. (USA) and Acorda Therapeutics Inc. (USA). Diacrin Inc. (USA) develops cell transplantations on basis of porcine neural or spinal cord cells. The company already carried out clinical trials, but seems to have stopped these in 2001.

Complete organ engineering

The overall, but still distant goal of tissue engineering is to construct *in vitro* human organs to overcome scarcity of donor organs and improve disease treatment. Research has been carried out on urinary bladder (Oberpenning et al., 1999), kidney (Humes, 1996; Humes, 2000; Woods & Humes, 1997), heart (see above), liver and pancreas. Products are still far from clinical use and the market. Furthermore, several scientific and technical hurdles still need to be overcome (e.g. vascularisation, controlled three-dimensional structure, coordinated action of different cell types).

Pancreas

Because of lifestyle changes and unhealthy nutrition habits the incidence of diabetes is increasing worldwide. Tissue engineering activities aim at relieving the patients from the need for regular injections of insulin as well as prevent associated diseases like CVD, blindness, kidney failure, impaired circulation leading to ulcers and leg amputation. However, many companies have stopped their R&D efforts to construct a bioartificial pancreas after more than 187 million € of private sector funds had been invested (Lysaght & Reyes, 2001). The transplantation of pancreatic islet cells (allogeneic or xenogeneic) seems to be more promising but the use of xenogeneic cells is controversial because of possible infection risks. Positive results have recently been achieved using allogeneic pancreas cells (Ryan et al., 2001; Shapiro

et al., 2000). Several encapsulated products are in clinical trials. The companies involved are Amcyte and Novocell from the USA, and Diatranz from New Zealand.

Bioartificial liver

The bioartificial liver, which uses liver cells in a specific extracorporeal bioreactor, has advanced further than the bioartificial pancreas, but all products are still in the clinical trial phase. In Europe around 1000 people develop acute liver failure each year, which can lead to death within 2 to 10 days. Some patients are able to recover, some need a liver transplant (about 4850 liver transplants were carried out in Europe in 2001, corresponding to total costs of about 414 million Euro per year). An artificial liver could help in bridging the time until a liver transplant becomes available or support the regeneration of the patient's liver. Several scientific and technical problems have to be solved:

- definition of cell type and cell source to be used in the device
- production of the required quantity of cells with the desired functionality
- keeping the cells physiologically active during cultivation in the device
- appropriate bioreactor design
- logistics such as shelf life, availability
- safety and efficacy in clinical application.

Two bioartificial liver devices with human hepatocytes (ELAD[®], Vitagen, USA; MELS[®], Hybrid Organ, Germany) and three devices using porcine cells (HepatAssist[®] 2000 System, Circe Biomedical, USA; BLSS[®], Excorp Medical, USA; LIVERX2000 System, Algenix Inc., USA) are already in the clinical trials phase or the company plans to start phase I trials in the near future.

3 SCIENCE AND TECHNOLOGY – RESEARCH AND EUROPEAN ACTORS⁵

3.1 Current research areas

Tissue engineering is the product of the integration of several lines of research from medicine, materials science, biology and engineering. According to the definition (see Annex 2), it encompasses supporting structures (i.e. scaffolds made of biomaterials), seeded with cells specific for the tissue that is to be replaced, and biomolecules with cell stimulating capacities. These three items present the backbone of tissue engineering and the main research fields and are described below. Figure 3.1 outlines the correlation between these different fields. Also the design of bioreactors and the overall logistics and automation are components of tissue engineering and the respective research areas are discussed below. Unavoidably, there is some overlap between the selected categories.

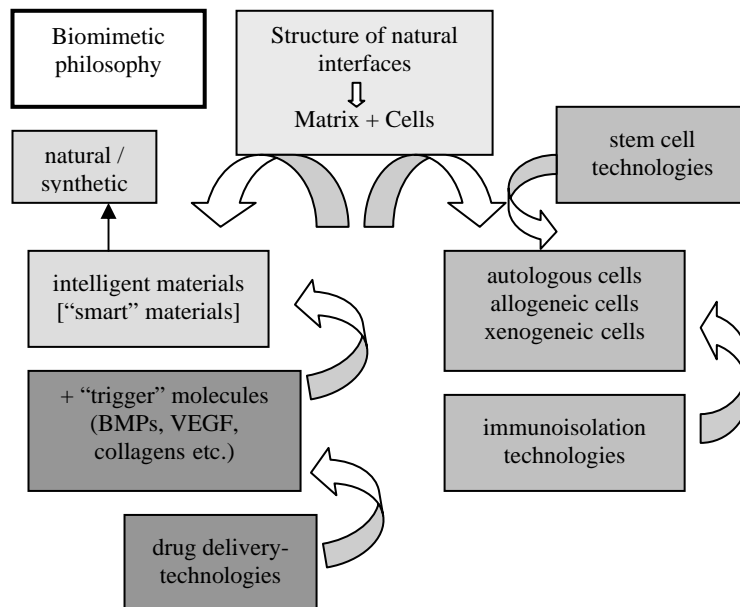


Figure 3.1: Tissue engineering and the interactions of biomaterials, cells and biomolecules
Source: C. J. Kirkpatrick

3.1.1 Biomaterials

Biomaterials in tissue engineering applications are used to support and guide the growth of cells in specific two- or three-dimensional structures. They take over the task of the extracellular matrix (ECM), which naturally provides cells with a supportive framework of structural proteins, carbohydrates and signalling molecules. The ideal scaffold, mimicking the ECM, would be made of a biomaterial that provides all the necessary signals for the cells to grow, differentiate and interact, forming the desired structure while at the same time slowly being degraded into physiological compounds as the new tissue is formed and integrated in the patient’s tissue. Moreover, the biomaterial should be safe and easily available at acceptable costs. The first degradable biomaterials used in tissue engineering were developed for surgery in the first place, building on the experience made regarding non-toxicity and non-immunogenicity of the material and its degradation products. Research now focuses on the development of

⁵ See also WP3 “Research activities and future developments of human tissue engineering in Europe and the US”

biomaterials with new mechanical properties for e.g. weight bearing, enhanced degradability, better processing characteristics and novel cell-interaction supporting properties. Of major interest is the integration of bioactive signal molecules into the biomaterial matrix or scaffold (see Chapter 3.1.3).

Biomaterials can be made of synthetic materials (e.g. lactide, glycolide, ceramics), naturally derived (e.g. collagen, natural polysaccharides) or semisynthetic polymers (e.g. poly-4-hydroxybutyrate). Poly(lactic acid) and poly(glycolic acid) or combinations of these two are already approved for clinical use and widely applied in tissue engineering research and products. Innovative developments in this field include:

- New degradable synthetic polymers that are for example thermoplastic and have a shape-memory capability so their conformation could be altered by changing the temperature (e.g. from room temperature to body temperature) (Lendlein & Langer, 2002).
- Composite materials made from polylactide-co-glycolide and bioactive glass or porous bioactive glass alone are promising candidates especially for bone tissue engineering (Livingston et al., 2002; Lu et al., 2003).
- Synthetic hydrogels that are modified with adhesive peptides to enable cells to colonise the gel (Park et al., 2003). Due to their good permeability for nutrition and gases, biocompatibility and physical characteristics, hydrogels are promising materials for tissue engineering applications.
- Photopolymerisation which enables great flexibility in scaffold formation (Nguyen & West, 2002).
- Biologically derived polymers such as poly-4-hydroxybutyrate, which have a high elasticity and controlled biodegradability and open up new possibilities in scaffold design.
- Synthetic protein-based polymers, that are produced with the help of genetic engineering and present a novel class of biomaterials which are biodegradable and biocompatible (Xue & Greisler, 2003). The integration of artificial amino acids could result in new polypeptide building blocks with distinct characteristics (Prestwich & Matthew, 2002).

Natural polymers such as extracellular matrix and derivatives such as collagen, hyaluronic acid and polysaccharides from plants and seaweed (alginate) are being tailored to be used in tissue engineering. Alginate hydrogels with a high porosity could be used for minimally invasive application techniques because of their ability to change physical status from liquid to gel. Fibrin is being modified with additional ligands. Another source of natural biomaterials is extracting and purifying whole tissues such as bones to be used as matrices for tissue engineering, thus making use of their natural structure and characteristics (e.g. associated growth factors). Chitin, chitosan as well as native or bioengineered variants of silk from silkworm, spiders and insects are natural polymers of animal origin, which are currently investigated for suitability as scaffold material (Altman et al., 2003; Khor & Lim, 2003).

Apart from the material chemistry itself, the structure of the scaffold plays a major role in tissue engineering. Of significance is the porosity with large void volumes resulting in a good surface-area-to-volume ratio to enable three-dimensional growth and supply with nutrients and oxygen. Woven and non-woven fibre-based fabrics are modified and adapted for use in layer structures for skin, bladder and intestine but also for blood vessels and cartilage. The surface characteristics of biomaterials are important for determining the behaviour of cells. Gas-plasma-treatment and other physico-chemical modifications can be used to render scaffold surfaces more receptive for cells (Claase et al., 2003). Nanotopographical methods are used to structure or pattern surfaces (Curtis & Riehle, 2001; Dalby et al., 2003; Dalby et al., 2002). Cell behaviour is also controlled by cell-ligand interactions and research is being carried out to understand this interaction using model systems and the effect of controlled combination of adhesion peptides and growth factors. Patterning technologies with specific distribution of cell anchoring sites could be used to direct and regulate distribution of cells on the scaffold (Xue & Greisler, 2003). Advanced manufacturing approaches based on computer-aided design and computer-aided manufacturing, solid free-form fabrication processes and microelectromechanical systems will lead to improved scaffold designs (Chaikof et al., 2002). The combination of imaging technologies and solid free-form fabrication promises precisely

formed scaffolds with the necessary microstructure (Warren et al., 2002). Organ printing, the computer-aided, jet-based three-dimensional tissue engineering of living human organs, is being developed to build three-dimensional organs or tissues integrating cells during the build-up of the scaffold. This could enable the introduction of a blood vessel system in the tissue (Mironov et al., 2003). Further examples will be given in the relevant sections below (e.g. 3.1.3 and 3.1.4).

A specific topic of major relevance for the future success of tissue engineering concepts is the interaction of cells of the immune system with the implanted biomaterial (Al-Saffar & Revell, 2000; Rhodes et al., 1997; Werthen et al., 2001).

3.1.2 Cells

Cells are the crucial part of any tissue engineering effort and comprehensive understanding of cell biology, extracellular matrix biology, developmental biology and physiology as well as immunology and inflammation are prerequisites to be able to predict cell responses to biomaterials and their contained biomolecules. New analytical tools, being developed in the context of progress in genomics and proteomics, e.g. chip technology, will help to gain more insight in cell culture methodology and regulation of cell response to external stimuli.

A significant issue is cell sourcing. There are different possibilities to derive cells:

- From the patient (autologous cells)
- From a donor not identical with the patient (allogeneic cells)
- From a different species (xenogeneic cells).

A specific cell type, so-called stem or progenitor cells, can be derived from

- The patient or another donor (autologous or allogeneic adult stem cells)
- Embryos. Embryonic stem cells can be extracted from surplus embryos created by *in vitro* fertilisation, which are no longer required by the parents (allogeneic embryonic stem cells), from embryos created by *in vitro* fertilisation for research purposes (allogeneic embryonic stem cells) or potentially from embryos created by somatic cell nuclear transfer (SCNT), a technology which has been applied successfully for several mammals but so far not for humans. SCNT is supposed to enable the production of tissue that perfectly matches immunologically the respective patient by using “quasi-autologous” embryonic stem cells.

Another option to derive stem cells is currently being researched, using unfertilised human oocytes that are coaxed to develop into parthenogenetic human embryos. These embryos do not have the potential to develop further and die within a few days, but it might be possible to derive stem cells from them (Lin et al., 2003; Pagán Westphal, 2003).

All cell sources have different risks and benefits concerning availability, immunogenicity, pathogenicity, and quality. The choice of cells will also influence product development time, the regulatory framework to comply with and marketing strategy (see Chapter 4.2). Only the use of allogeneic or xenogeneic cells will make possible the off-the-shelf availability of tissue-engineered products.

Regarding autologous cells, the availability of cells is limited for some cell types due to difficulties obtaining them through biopsy (e.g. neural tissue). However, the use of autologous cells has the advantage of avoiding immunogenic reactions and a low risk of infection with pathogens. The cells differ in quality and behaviour from patient to patient and it will be difficult to achieve consistent product quality under these circumstances. Autologous cell products will not be available in urgent acute cases.

For allogeneic cells there is a better availability because a larger number of donors can be used. The pooling of cells from different donors can provide constant starting material. On the other hand there is an increased risk of disease transmission and the recipient’s immune system may need to be suppressed to avoid an adverse immune response.

Concerning xenogeneic cell sources, the risk of animal pathogen transmission and immunogenic rejection is an issue. Genetic modification of the source animal and immunoisolation techniques might be able to reduce immunogenicity of the graft but future research might reveal other problems such as mutagenicity and tumour formation in the cultured cells, a problem which also applies to cultured autologous and allogeneic cells. Another possible barrier might be ethical concerns regarding manipulation and use of source animals.

Stem and progenitor cells are of considerable interest and the focus of research of many companies and academic research institutions. The availability and properties of adult stem cells are currently being explored and their potential ability to develop into a wide range of different cell types makes them a promising candidate for tissue engineering. Mesenchymal stem cells derived from bone marrow are studied e.g. for bone and cartilage applications (Noel et al., 2002; Risbud & Sitterling, 2002) and for applications in heart failure (Tran et al., 2003). They are easily accessible and lack immunogenic properties, which opens up the possibility to use them as allogeneic cell sources. Muscle-derived stem cells are considered another promising cell type (Deasy & Huard, 2002). Embryonic stem cells seem to be more versatile and easier to handle to date but the derivation from embryos which leads to the embryo's destruction is associated with severe ethical concerns in many European Member States. Research on the abilities of adult stem cells to differentiate is on-going (Holden & Vogel, 2002; Jiang et al., 2002; Verfaillie, 2002; Vogel, 2002).

Research is being done on the culture conditions for routine proliferation, the controlled and permanent differentiation of stem and progenitor cells into pure populations of specific cell types (lineage differentiation) and to control proliferation and differentiation for therapeutic purposes, especially for avoiding any cancer risk. Recently a specific class of signalling proteins (Wnt proteins) has been identified as inducers of self-renewal of haematopoietic stem cells and as critical for homeostasis, which might be important for tissue engineering application (Reya et al., 2003; Willert et al., 2003). Interesting work is being performed on the use of fatty tissue as a source of adult mesenchymal stem cells and the lineage induction for cardiomyocytes (Rangappa et al., 2003). A further source of progenitor cells is the blood. Thus, Shirota et al. (2003) have demonstrated that the isolation and expansion of endothelial progenitor cells (EPCs) from human blood could be a possible source of cells for seeding vascular grafts. Replacing the epithelial lining of the urogenital tract is also a major problem. *In vitro* expansion of urothelial cells obtained from bladder washings has been shown to yield a cell population which can be expanded in the laboratory and could offer a useful source for tissue engineering of the bladder (Fossum et al., 2003).

3.1.3 Biomolecules

Cell growth and wound healing normally take place embedded in the ECM. This gel-like network provides proteins and carbohydrates with specific functions for cell proliferation, differentiation and interactions. To mimic the ECM is one aim of ongoing research on scaffolds and biomolecules.

Biomolecules include proteins with the function of growth factors, differentiation factors, angiogenic factors and bone morphogenic proteins, which influence cell behaviour and growth. Significant effort is put into elucidation of effects of specific molecules, the appropriate selection for the tissue engineering application and the method to deliver the molecules to the tissue and cells. ECM functions as a major reservoir for many of these biomolecules.

Delivery of biomolecules can be achieved externally in the form of proteins or the respective gene. Research on gene therapy has been a source of much currently available knowledge in transfection of cells to promote protein expression. The efficiency of expression, its regulation and cell- and tissue-specific transfer are key research areas. Another possibility is the stimulation of the cells to produce the selected biomolecules themselves through the choice of the biomaterial or the tissue culture conditions.

Cell adhesion and motility can be influenced by integrating adhesion peptides in the scaffolds. One of the best investigated biosignal molecules is the small amino-acid sequence RGDS, which is a cell-binding site in many matrix proteins. It is hoped that its incorporation into matrices and scaffolds for tissue engineering a rapid and effective binding of the cells of interest can be achieved. One example is its incorporation into synthetic polymer hydrogels to assist osteoblast adhesion and growth for bone regeneration (Behravesch et al., 2003). The right density needs to be determined to hold the delicate balance between adhesion and migration (Hench & Polak, 2002). The optimal concentration, spatial and temporal availability as well as the effects of single biomolecules and combinations of biomolecules are research topics. Griffith & Naughton (2002) describe the possibility of constructing a dual-release scaffold, releasing factors like PDGF (platelet-derived growth factor) and VEGF (vascular endothelial growth factor) in different quantities and at different times, thus mimicking physiological conditions for blood vessel induction and maturation (Perets et al., 2003), which is a prerequisite for the success of most TE implants.

The covalent incorporation of bioactive molecules into the scaffold and the controlled release to achieve physiological concentrations is the next challenge. The use of so-called drug delivery systems integrated in the tissue-engineered construct can be used to provide a combination of essential biological signals such as growth factors and directed at several cell types from blood vessel lining cells (Wissink et al., 2001) to bone cells (Boden et al., 2000). Microfluidic technology is one potential way to achieve targeted delivery (Saltzman & Olbricht, 2002). Encapsulation in degradable microspheres or the direct incorporation in the scaffold and release through diffusion and degradation of the scaffold, present other ways of biomolecule delivery. Supercritical carbon dioxide is being tested to create scaffolds with incorporated growth factors (Whitaker et al., 2001). It will continue to be a major task to ensure that the pharmacokinetics and -dynamics are such that no massive biological stimulus is present which would result in too much cell proliferation (hyperplasia) or even tumour development (neoplasia).

3.1.4 Engineering design and manufacturing aspects

Once feasibility of the tissue engineering approach has been confirmed, several design challenges need to be addressed to enable large-scale product manufacture.

Adaptation of existing bioreactor technology to large-scale expansion of cells and three-dimensional growth of tissues

Bioreactors might need different properties depending on whether autologous cells or allogeneic cells are to be used for just one or several patients, respectively. In the case of autologous cells, fewer cells from a small biopsy will have to be expanded and the tissue will be delivered to one specific patient only. Therefore robust, mobile bioreactors would be needed, or smaller items stationed locally or regionally. Larger bioreactors for growing allogeneic tissue for a larger number of patients might be located at one single site. Designing bioreactors for three-dimensional structures presents a major challenge. In the tissue engineering of blood vessels using biodegradable polymer scaffolds, specially designed rotational culture systems are being developed (Nasseri et al., 2003). Other groups are focussing on the design of perfusion systems (Sodian et al., 2002).

Understanding stimuli for the development of tissue with certain physical properties

Tissues such as blood vessels, bone and cartilage have specific physical properties to fulfil their function in the body. The relationship between the composition and structure of the tissue and its mechanical performance is not yet very well understood. Additionally, for many tissues the specific properties are not yet well defined. For blood vessels and bone a considerable amount of information on *in vivo* stresses and strains is available, but in the case of cartilage information on the *in vivo* mechanical environment is lacking (Guilak, 2001). A major challenge for tissue engineering is to design replacement tissues with the essential mechanical properties. Research focuses on elucidating the molecular and cellular responses to certain external stimuli such as flow of culture medium, shear stress, and pressure to translate this information into the design of

bioreactors. Considerable improvements have already been achieved for blood vessels, cartilage and cardiac muscle (Naughton, 2002). An additional important factor in the design of tissue-engineered vascular structures is the simulation of the mechanical forces exerted in the natural blood vessel as a result of smooth muscle activity. Thus, for example, *in vitro* systems employing cyclic mechanical strain are being developed and tested (Stegemann & Nerem, 2003). Most probably other factors are also involved in the development of tissue properties, such as soluble signalling molecules or the extracellular matrix (Bottaro et al., 2002).

Strategies for vascularisation of tissue after implantation but also during tissue growth

A prerequisite for growing larger tissues is the vascularisation to facilitate oxygen and nutrient supply to every part of the growing tissue. This is one focus of research activities and includes specific structuring of scaffolds and seeding them with endothelial cells to promote the growth of blood vessels (see Chapter 3.1.3). Specifically engineered scaffolds, which release angiogenic growth factors might also be a solution for small tissues which cannot exploit existing vascular beds, although the process of blood vessel growth might still be too slow (Griffith & Naughton, 2002). The use of stem cells or progenitor cells could offer a further advantage with respect to the need for rapid vascularisation as these cells are resistant to low oxygen conditions (Hevehan et al., 2000; Ivanovic et al., 2000).

Techniques for the storage and preservation of cells and tissues

Tissue-engineered products need to be shipped from the production site to the patient. A prerequisite for the regular use of tissue-engineered products is the development of strategies for stable storage of cells and three-dimensional tissues prior to their clinical utilisation. One cost-efficient and effective method could be cryopreservation as it provides a long shelf life and low risk of contamination. However, there are many unanswered questions about the response of different cell types and tissues to the freeze-thaw process and specific protocols need to be developed. Cryoprotectants are used to prevent deleterious ice formation within the cells and tissues but how protection works in detail and how the cryoprotectants can be removed from stored tissues needs to be clarified (Toner & Kocsis, 2002).

Development of specific bioreactors for bioartificial (extra-corporal) organ support devices

Large numbers of cells need to be produced for temporary treatment of severe kidney and liver diseases with extra-corporal bioartificial organ devices. The devices aim at replacing dialysis in case of kidney failure, support liver regeneration or bridge the time until a liver transplant becomes available. Bioreactors need to be adapted to this biologically very complex and specific application (Jasmund & Bader, 2002). Research is being done on the type and source of cells used, providing the appropriate number of cells with optimal physiological functionality, improving the transport between the cells and the patient's blood and minimising the volume of the device.

There is a huge sector of research associated to tissue engineering efforts concerning the development of suitable *in vitro* testing methods to evaluate the quality of tissue engineering concepts and constructs (Kirkpatrick et al., 2002; Pariente et al., 2000). Standards need to be defined for a successful tissue-engineered product and its performance that are based on the structure and function of the native tissue. Methods for the long-term assessment of cell and tissue function also need to be developed (National Institutes of Health Bioengineering Consortium, 2001).

3.1.5 Bioinformatics to support tissue engineering

The field of bioinformatics is considered to be less well developed for tissue engineering than for other areas. New analytical tools such as gene array technology and protein chip technology will produce a huge amount of useful data for tissue engineering, providing insight into relationships between external conditions and cell responses at the genetic level and for functional parameters of cell populations. Imaging technology will help to assess structural parameters as well as observe and control cells and engineered tissues.

All these data need to be collected and processed, stored and analysed. Databases for tissue structure, function and biomaterial response are expected to be generated in the future. Data analysis tools will be developed to analyse functional and structural relationships useful for assessment of efficacy and safety. Tissue modelling systems, standardised manufacturing datasets and automated quality assurance systems will foster development and production.

The storage, integration, visualisation, modelling and distribution of new data will pose a challenge that requires the development of standards for databases and for terminology and classification. Efforts might need to be undertaken to develop a common standard database construction to be able to share information and build on synergistic effects.

3.2 Developments expected in the coming ten years

Tissue engineering is a field which is still in its very early development. Several scientific as well as technical problems need to be solved before products can be commercialised on a large scale. Several experts in the field from the UK, Germany and the USA have been interviewed to provide their view on key breakthroughs and key research areas in tissue engineering in the near future.

- They believe that **adult stem cells** will play a significant role in research in the coming years and key breakthroughs would be to know how to program them, control their differentiation and be able to use them for tissue engineering. It is probable that adult stem cells will be one of the main cell sources for tissue engineering in the medium-term future, with the advantage of avoiding immune rejection. Embryonic stem cells would mainly be used for basic research on cell differentiation and the necessary molecular signals, the results of which could be applied to tissue engineering.
- The structural properties of **scaffolds** will be improved and also include important signal molecules to influence and control cell development and behaviour. As a result of increasing knowledge about molecular signals, scaffolds used in growing tissues will be improved to become “smarter”, providing the cells with the biomolecules and environmental stimuli needed for optimal growth and development. Three-dimensional scaffolds will be needed, with custom-made form and size for the individual patient’s needs. New materials as well as the integration of imaging technologies such as magnetic resonance imaging (MRI) to produce tailor-made scaffolds with the correct shape, will be a major focus of research.
- **Controlling differentiation of cells** is a significant issue in tissue engineering. Cell interactions within the same tissue and with other tissues need to be elucidated to control tissue growth and implantation. The same is true for the transduction of physical signals into intracellular molecular signals and change of cell behaviour. Also the distinction between different cell types will be a main issue in research, ensuring that the correct cell types are isolated for tissue engineering. It is assumed that the knowledge on how to control cell function and differentiation especially with a view to three-dimensional tissue-engineered constructions will be available within the next ten years.

- **The test and control of metabolic activity and functionality** is a prerequisite for further progress in this area. Testing tools for TE products will need to be developed. This will lead to research on new analytical tools but also on improved information processing and networking. In general, to enable cooperation and be able to make use of all the data from cellular genomic and proteomic research, harmonised standards for databases need to be put in place.
- **Bioreactors** for three-dimensional tissues and the supply of the necessary physical stimuli will be developed. Additionally, efforts are necessary to develop bioreactors with automated processes able to standardise the production of tissue-engineered products from individual patient's cells.
- The **growth and control of blood vessels** in engineered tissues is an essential prerequisite for growing three-dimensional tissues and also for their survival once the tissue is implanted in the patient's body. Knowledge about how to foster and control vascularisation in growing tissues will enable the construction of thicker, three-dimensional tissues, a prerequisite for enlarging the application range of tissue-engineered products. With a view to being able to use allogeneic cell sources, the **prevention of rejection** of the engineered tissue by the patient's immune system will also be a field of intensive research.

3.3 Characterisation of the European tissue engineering research community

With the aim of identifying the performers of research an analysis of scientific publications for the period 1991 to 2002 was undertaken. A short description of the bibliometric search and analysis can be found in Annex 2. The following keywords were used:

[human + (tissue engineer*)] or [human + regenerat* + (skin or bone or vascular or valves or cartilage or chondrocyte or osteochondral or endothelial or keratinocytes)].

3080 publications have been identified, nearly all of them authored by researchers from public institutions. This is not surprising as researchers have to “publish or perish”, whereas companies generally do not care to publish. On the contrary patent issues generally delay publication. Figure 3.2 shows the increase in number of publications in this area from 1991 to 2002.

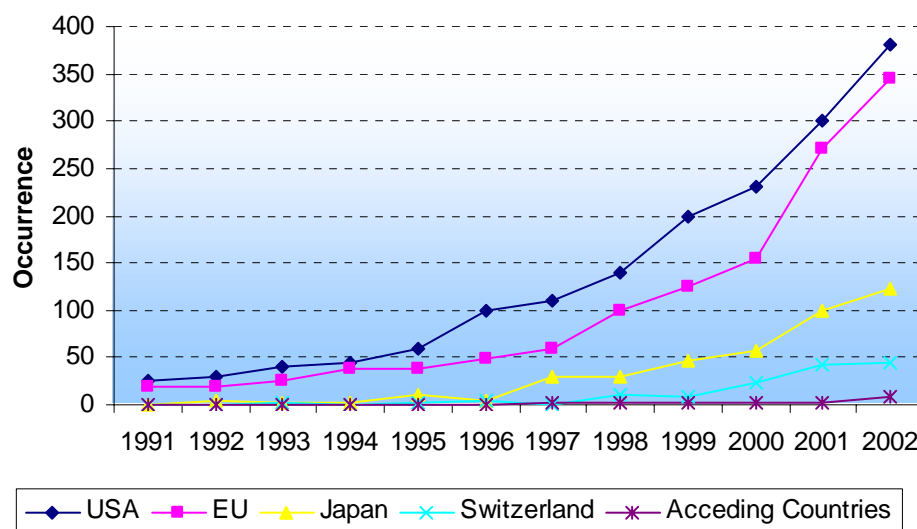


Figure 3.2: Tissue engineering publications 1991 - 2002
Source: SPRU, University of Sussex

US authors have been clearly ahead in the mid-nineties, but the EU has been catching up since 2000. Looking at the public institutions with most authors occurring in any of the publications, the US institutions lead, with Harvard University, Massachusetts Institute of Technology, University of Michigan and University of Pittsburgh ahead of all the others. Their leadership is not surprising since these were the institutions first to introduce the subject (Murray, 2002). 12 institutions from 3 EU Member States are represented in the Top 50: UK (University College London, Imperial College), Germany (Humboldt University, Universities of Munich, Heidelberg, Freiburg, Hamburg, Regensburg, Hannover Medical School, TH Aachen), and The Netherlands (University of Nijmegen, Erasmus University). The Swedish Karolinska Institute is also present in the Top 50 institutions when first authorships only are taken into account. Also Japan is showing an increasing activity in tissue engineering. These findings are supported by a recent report, characterising Japan as the leading country in Asia concerning tissue engineering, with major government investments and the creation of new research centres (The Royal Academy of Engineering, 2003). Relating the number of occurrences shown in Figure 3.2 to the size of the respective populations of the USA, the EU and Japan, it becomes clear that the EU as well as Japan is not performing as well as the USA (data not shown).

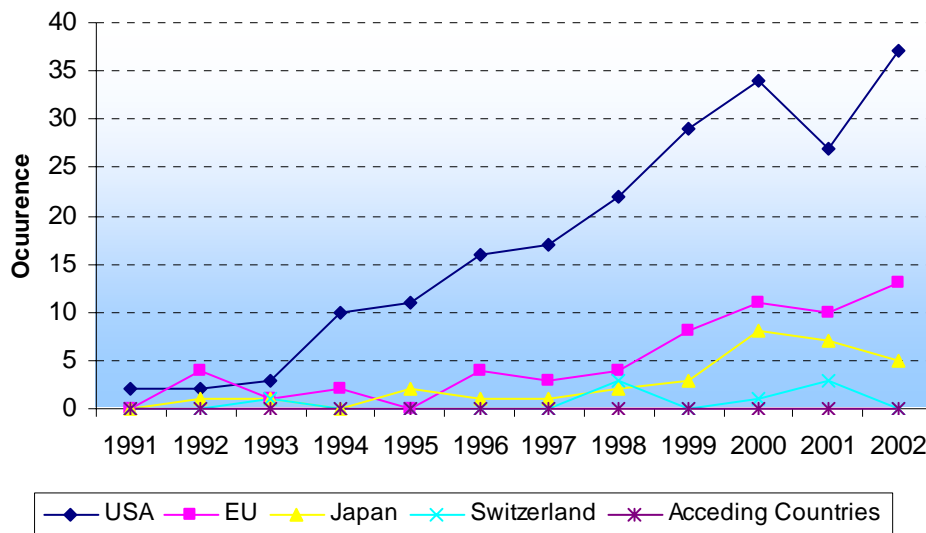


Figure 3.3: Domestic public/private co-authorships 1992-2002
Source: SPRU, University of Sussex

The analysis of public/private co-authorships reveals an interesting detail. In the USA more than a third of all public/private collaborations in the period 1991 – 2002 are domestic (211 out of 551 co-authorships), whereas they account for less than one quarter of EU collaborations (61 out of 264 co-authorships). The reason behind this discrepancy is not clear, perhaps there is a lack of certain expertise in Europe or companies are not aware of existing expertise. The perceived lack of a strong organisation of the tissue engineering community might contribute to this situation. However, Figure 3.3 shows that domestic public/private collaborations are increasing since 1995 in the USA as well as in the EU. This could reflect the recognition that the multidisciplinary needed in advancing tissue engineering might demand increased research collaboration between institutions. National governments as well as the European Commission may have a role in promoting networking in this sector.

4 COMMERCIAL TISSUE ENGINEERING IN EUROPE – COMPANIES AND INFLUENCING FACTORS

4.1 Characteristics of European tissue engineering companies⁶

In this study, about 113 companies have been identified which are active in the field of tissue engineering in Europe. These companies can be divided into core tissue engineering companies⁷, “broader definition” tissue engineering companies⁸ and companies that are active in developing tissue-engineered products for *in vitro* use only, i.e. not for therapeutic purpose.

Out of the 113 companies identified, 54 belong to the core tissue engineering category, 48 are companies characterised as broader definition tissue engineering companies and 11 companies produce tissue-engineered products for *in vitro* use. The distribution of these companies within European countries is shown in Figure 4.1. Most tissue engineering companies are based in Germany, with 39 companies including 19 core tissue engineering companies. Germany is followed by the UK, with 18 tissue engineering companies of which 11 are core tissue engineering companies. In France, despite being the second largest Member State, a comparatively small number of tissue engineering companies was identified (10). In contrast, small countries such as Sweden and The Netherlands are relatively active in tissue engineering (10 and 6 tissue engineering companies, respectively). Only few companies have been identified in Italy (2) and Spain (3), and none in other Mediterranean countries such as Greece or Portugal. The only acceding country with tissue engineering companies identified is the Czech Republic with 3 tissue engineering core companies (list of companies see Annex 1).

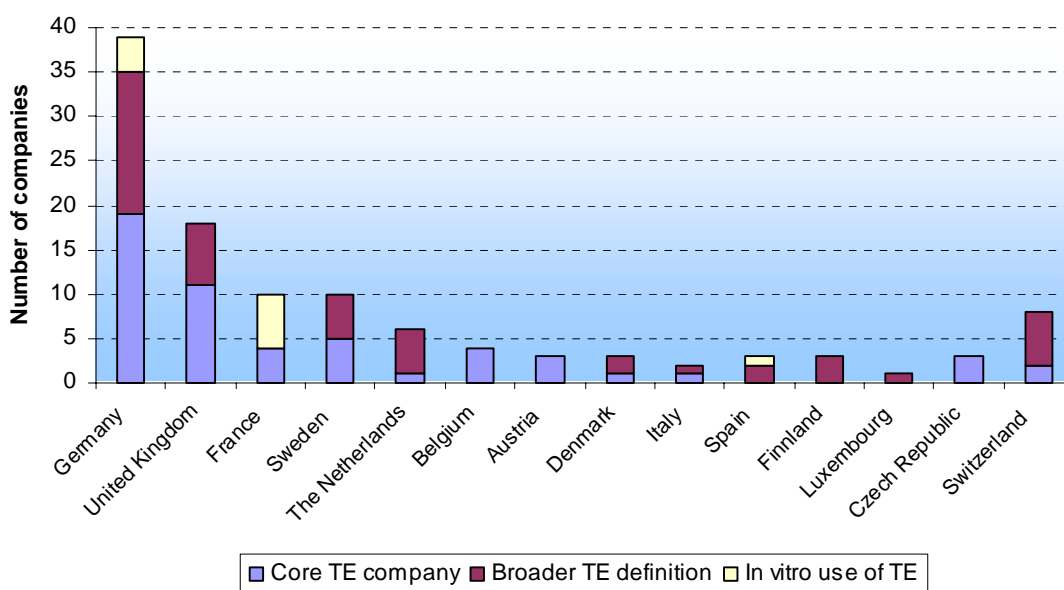


Figure 4.1: Tissue engineering companies in Europe
Source: Fraunhofer ISI

⁶ See also WP1 “Analysis of the actual market situation – Mapping of industry and products”

⁷ The activity of core tissue engineering companies fully complies with the tissue engineering definition selected for this study.

⁸ “Broader definition” tissue engineering companies carry out activities which are directly relevant for tissue engineering, but do not comply fully with the definition, for example the activity concentrates on the construction of bioreactors for tissue engineering. Additionally medical device and pharmaceutical companies which are involved in joint R&D activities in tissue engineering, but this presents only a minor activity in the company, are included in this category

Of the 113 companies identified 80 are biotechnology companies (71%), 24 are medical device companies (21%) and 9 belong to the pharmaceutical sector (8%). Most of the tissue engineering companies identified are small and medium sized companies (91 of 113; 49 of 54 core tissue engineering companies), with less than 500 employees (Figure 4.2). For 44 of these 91 SMEs more information on the number of employees was available. These figures show that about 75% of the SMEs have less than 50 full time equivalent employees. Only one company was identified with more than 100 employees.

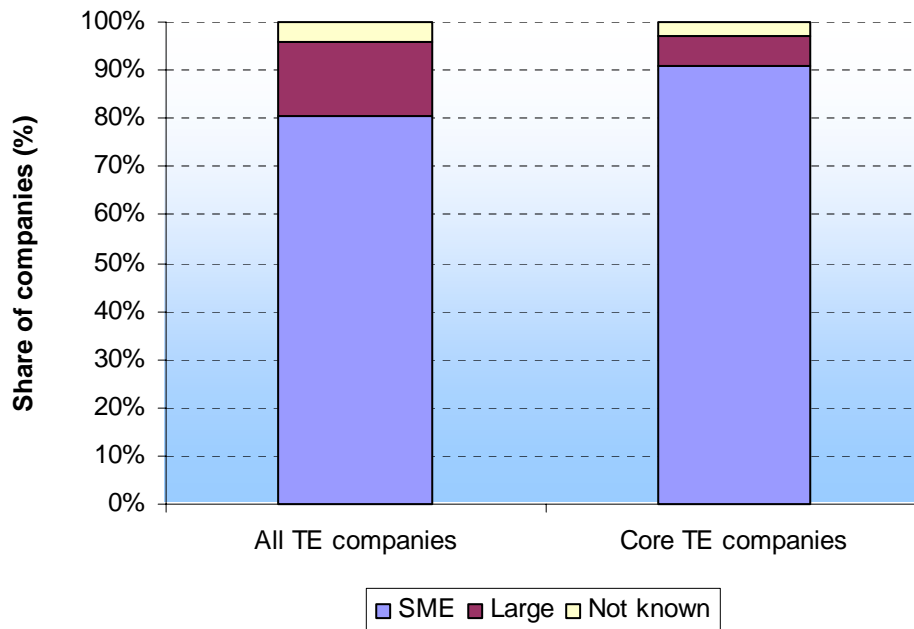


Figure 4.2: Company size of European tissue engineering companies
Source: Fraunhofer ISI

The characteristics of tissue engineering companies in the USA have been described by Lysaght & Reyes (2001). Around 70 start-up companies are active in tissue engineering, 14 of them not from the USA. The definition used for tissue engineering was a bit broader than the one used for this study⁹. Most of the companies are young and small. 40% have less than 16 full time equivalent employees, 40% less than 51 and 20% have more than 51 employees (overall about 3500 full time equivalent employees).

The majority of US companies is active in the area of structural applications (skin, bone, heart valves, arteries, myocardial particles), which is still expanding. The sector for metabolic applications (bioartificial organs, encapsulated cell therapies) has shrunken by 30% since 1998, reflecting the early stage of development and basic scientific problems that need to be solved for further development. The sector concerning cellular applications (cell transplantations, therapeutic cloning) is stagnating since 1998, but the focus is changing with newly emerging companies having activities involving human embryonic and adult stem cells (Lysaght & Reyes, 2001, Lysaght & Hazlehurst, 2003).

Cumulative spending since 1990 exceeded 3.5 billion € increasing at about 16% per year. About two dozen of the companies are listed on stock exchanges (representing 35% of the workforce employed). However, according to the authors no profitable tissue-engineered product seems to be yet on the market. The bankruptcy of Advanced Tissue Sciences and Organogenesis by the end of 2002, the leading companies for tissue-engineered skin products

⁹ The definition for tissue engineering includes devices or processes that combine living cells and biomaterials, utilise living cells as therapeutic, diagnostic reagents or regenerate tissues or organs in-vitro for subsequent implantation, or provide materials or technology to enable such approaches.

on the market, might be the first result of the disproportion between investing in research and development and very low sales numbers and a first sign for consolidation of the sector.

It can be concluded that the tissue engineering sector seems to be quite similar in Europe and the USA. It is characterised by young, small, research-based and technology-oriented companies, a structure which reflects the recent emergence of tissue engineering and the dynamics of this field.

4.2 Factors influencing the development of the commercial tissue engineering sector in Europe¹⁰

4.2.1 Regulatory framework

Regulation in the EU

Currently there is no specific European regulation for human tissue-engineered products. Member States mostly use existing regulation for medical devices or medicinal products and approach the tissue-engineered products on a case-by-case basis (see below). Authorisations cover only the respective national market. The regulatory framework for medical devices is based on the Council Directive 93/42/EEC¹¹. This Directive explicitly does not apply to “transplants or tissues or cells of human origin nor to products incorporating or derived from tissues or cells of human origin”. Directive 2001/83/EC on the Community code relating to medicinal products for human use¹², is considered inappropriate to the specific properties and requirements of human tissue-engineered products. However, the Committee for Proprietary Medicinal Products of the European Agency for the Evaluation of Medicinal Products (EMEA) considered human adult cell therapy products as medicinal products, provided the cells have been subject to a manufacturing process in dedicated facilities encompassing more than minimal manipulation, and the resulting product is definable in terms of qualitative and quantitative composition (EMEA, 2001). This describes very roughly the regulatory grey zone in which tissue-engineered products are situated at the moment. The Scientific Committee on Medicinal Products and Medical Devices (SCMPMD, DG SANCO) stated that current legislation does not encompass all aspects of tissue engineering and concluded that there is a need for specific legislation. It recommended the establishment of a Tissue Engineering Regulatory Body to oversee tissue-engineered products (European Commission, 2001).

In June 2002 the Commission published a proposal for a Directive on setting standards of quality and safety for the donation, procurement, testing, processing, storage, and distribution of human tissues and cells (European Commission, 2002a). The draft Directive aims at harmonising the quality and safety of human cells and tissues used for applications to the human body. Concerning industrially manufactured products derived from tissues and cells, it covers only the first steps of the process, namely donation, procurement and testing of cells and tissues, provided the other steps are regulated by Community legislation. Embryonic stem cells, if their use is authorised in a Member State, would be covered by this directive. Research activities are covered as they concern cell and tissue applications to the human body in clinical trials. In June 2002, the Commission launched a public consultation about the need for a complementary legislative framework for human tissue engineering and human tissue-engineered products, which was open until end of September 2002. Industry and several Member States supported the development of a new specific legislation, as none of the existing regulations can provide sufficient safety for the patient and still allow rapid innovation (European Commission, 2003).

¹⁰ See also WP2 “Comparison of tissue engineering treatment costs with conventional treatment” and WP4 “Legal situation and socio-economic impacts of tissue engineering”

¹¹ Official Journal L 169 12/07/1993, p. 0001-0043

¹² Official Journal L 311 28/11/2001, p. 0067-0128

Meanwhile the regulatory framework applied differs from Member State to Member State, which presents an obstacle to free movement of tissue-engineered products in the EU. Some Member States, such as Austria, Finland and Sweden are preparing new national measures on the basis of regulations or guidelines. Others apply either the medical devices or the pharmaceuticals regulation, on a case-by-case basis. The cases of Germany and the UK are described in more detail below. Germany follows a pharmaceutical approach, and the UK the medical devices approach.

Germany

Germany has not yet developed any specific regulation for human tissue-engineered products. According to Gassner (2001), from a technical point of view tissue-engineered products could be classified as medical devices in line with the Medical Devices Law (Medizinproduktegesetz), but human transplants, tissues and cells and products containing human tissues and cells are not covered by this law, thus excluding human tissue-engineered products. Instead, tissue-engineered products are currently subsumed under the Medical Drug Law (Arzneimittelgesetz). For autologous products or allogeneic products, which are produced for an individual patient, a manufacturing approval by the authority of the respective Land is needed. This has been the case for products such as autologous chondrocyte transplants (ACTs) or autologous skin substitutes. An approval process as required for drugs including clinical trials is not required. However, information on effectiveness and costs, available only through clinical trials, is needed for reimbursement decisions by insurance companies (see below). Allogeneic products produced for a wide variety of patients would need a product approval as for drugs of the federal authority (Bundesinstitut für Arzneimittel und Medizinprodukte BfArM) or from EMEA, which would encompass clinical trials.

In general, Good Manufacturing Practice (GMP) for pharmaceuticals as well as guidelines from the German Medical Association (Maintenance and management of tissue banks, Manufacture of transfusion related products, Collection and application of stem cells¹³), although not being specific for tissue-engineered products, should be taken into account and applied (Dieners et al., 2002).

United Kingdom

The UK recognised the fact that tissue-engineered products fall outside the scope of European regulations for medical devices and medicinal products. These products still fall under the Consumer Protection Act (1987) and the General Product Safety Regulations (1994), but no other regulation is deemed applicable in the UK. In June 2002 a “Code of Practice for the Production of Human-derived Therapeutic Products” was published by the Medical Devices Agency of the Department of Health (since April 2003 merged with the Medicines Control Agency to Medicines and Healthcare Products Regulatory Agency MHRA), oriented at medical device regulation. The Code of Practice complements the Code of Practice for Tissue Banks (March 2001) and the Guidance on the Microbiological Safety of Organs, Tissues and Cells for Transplantation (August 2000). It was developed consulting with professional organisations, commercial producers and hospitals. It has no statutory force and is not legally binding but could be used by producers, certification organisations and regulatory agencies as a basis for assessing product safety and quality. Its aim is to enhance confidence in tissue-engineered products and to protect companies from liability in case of unforeseen adverse effects, provided they followed the Code. The Code covers therapeutic products, which contain human-derived material (viable or non-viable). This includes engineered human skin or cartilage products, but excludes viable animal tissue. It aims at controlling and safeguarding the quality of materials used, the microbiological safety, the production and processing practices and the product performance. The latter will be achieved through pre-market clinical studies and post-market surveillance. When recombinant genetic methods and/or products are used in the development

¹³ Bundesärztekammer (www.bundesaerztekammer.de) Richtlinien zum Führen einer Knochenbank 1998, Richtlinien zum Führen einer Hornhautbank 1997, Richtlinien zur Transplantation peripherer Blutstammzellen 1997, Richtlinien zur Transplantation von Stammzellen aus Nabelschnurblut 1999

of a tissue-engineered product or are present in the final product, the product is covered by regulations for pharmaceutical products.

Regulation in the USA

In the USA the U.S. Food and Drug Administration (FDA) is responsible for cell and tissue products. In 1997 the FDA initiated the Tissue Action Plan (Proposed approach to the regulation of cellular and tissue-based products) with the aim to revise existing regulation and to develop a comprehensive and workable regulatory framework for tissue-engineered products. In 2001 a final rule on “Human cells, tissues, and cellular and tissue based products; Establishment Registration and Listing” was published. It requires the manufacturers of human cells, tissues, and cellular and tissue-based products to register and list their products. Excluded are vascularised human organs for transplantation, whole blood or blood components or blood derivatives, secreted or extracted human products, except semen, minimally manipulated bone marrow for homologous use, and xenogeneic cells or tissues. Furthermore, two proposals have been published in 1999 and 2001, respectively, “Suitability determination for donors of human cellular and tissue-based products” and “Current Good Tissue Practice for manufacturers of human cellular and tissue-based products; Inspection and enforcement”. The first proposal aims at screening and testing of cell and tissue donors for risk factors and clinical evidence for relevant communicable disease agents and diseases. The latter aims at requiring manufacturers to follow certain requirements for methods used, facilities used, controls and a quality system to be put in place. Additionally labelling, reporting, inspections and enforcement will be regulated. Both proposals have not been finalised yet.

Medical products are either regulated as drugs, medical devices, biologics or combination products, the latter covering products that combine several characteristics and which will be categorised according to the primary mode of action. Regulation will be dependent on potential risks associated with the product. More than minimally manipulated tissue products need a market approval according to their categorisation, most probably as medical device or biologics. Product classification disputes can be clarified by an ombudsman, and a Tissue Reference Group has been proposed. For example tissue-engineered skin products are currently classified as medical devices. For medical devices intended for implantation or acting as a life-sustaining or life-supporting device, the FDA requires pre-market approval involving preclinical studies and clinical trials to demonstrate safety and effectiveness. Additionally compliance with good manufacturing practice is necessary. Autologous chondrocyte transplantation products such as Carticel[®] have been classified as biologics. Pre-market approval includes the demonstration of product safety, purity and potency through clinical trials as well as quality control and good manufacturing practice.

Legal situation and reimbursement

The main difference between the USA and the EU concerning regulatory approaches for tissue-engineered products is a uniform approach in the USA contrasted to differing national approaches in the EU at the level of Member States. Once a product receives approval in the USA a large national market can be accessed and the reimbursement of costs by health insurance companies does not pose a major problem. In Europe for the time being each country has its own approval system, which differs from the others. Approval is only valid for the respective (small) national market, and access to other national markets might have different requirements. Tissue-engineered products can be marketed on the basis of pre-clinical data, which results in the advantage of earlier access to the market compared to US companies. On the other hand, insurance companies often ask for additional data for reimbursement, for example on cost-effectiveness and long-term efficacy of the treatment, which can only be provided through specifically designed clinical studies. Furthermore, due to the unharmonised and unclear legislative situation in the EU and many of its Member States, predominantly autologous products have been developed and produced to date. Concerning allogeneic products, the investment for development and clinical trials would probably be too costly for targeting only a small national market.

4.2.2 Cost effectiveness

Tissue-engineered products open up a new way of treating diseases. The hope is that they deliver superior treatments, improving the speed, extent and duration of healing compared to conventional treatments. This needs to be proven for individual products and applications in comparative clinical trials or at least in studies with comparable outcome criteria. The comparison of treatment costs alone is not sufficient, as different treatments can result in different outcomes. The effectiveness of treatments needs to be included in the comparison.

Today only a few tissue-engineered products are already on the market, mainly in the skin and cartilage sector, as shown in Chapter 2. A literature search was carried out for publications on cost-effectiveness of available tissue-engineered products. Only few publications could be identified that present costs of treatments and even fewer that present a cost-effectiveness analysis or a cost-utility analysis of tissue-engineered products compared with conventional treatments. It seems that only few clinical trials have been carried out to prove effectiveness and superiority of tissue-engineered products in terms of improved healing process, increased quality of life and costs of treatments compared to other treatments. It should be noted that neither is this kind of information available for all conventional treatments.

The cost-effectiveness of tissue-engineered products has been investigated on the basis of publicly available data for three case studies: skin substitutes for burns and ulcer treatment, autologous chondrocyte transplantation (ACT) and cell seeded synthetic vascular grafts. For all products the amount of data was rather scarce, the few studies that could be identified (all in all 38 publications) often having methodological shortcomings (small patient groups included, rarely controlled trials, data based on modelling) or not being sufficiently transparent. Furthermore, the studies usually refer to different comparative conventional treatments and include different or not clearly defined cost factors, thus making comparisons difficult. Accordingly, the results presented below need to be taken with care.

Skin substitutes for burns and ulcers

Severe burns

In case of severe burns (full-thickness burns which do not heal by autoregeneration) the quick closure of the wounds is very important. Several conventional as well as tissue-engineered products are available for that purpose. Three publications have been identified that provide information on treatment costs of tissue engineering and conventional treatments. No study was available that includes treatment costs as well as measurement of effectiveness. Product costs alone differ strongly between tissue-engineered and conventional products. Conventional products have costs in the range between 0.37 €/cm² to 8.66 €/cm² whereas tissue-engineered products are more expensive with costs between 9.92 €/cm² and 20.85 €/cm². Treatment costs of severe burns can vary considerably for each individual patient, making comparisons between treatment options difficult.

A study based on model calculations (Parente, 1997) estimates treatment costs between 18,815 € and 94,550 € for tissue engineering treatment and 28,165 € to 156,534 € for conventional treatments, the range depending on the area of body surface injured. In this case tissue engineering treatment results in lower costs, however, the model has not been validated with clinical data and similar product costs were assumed for the tissue-engineered and the conventional product (cadaver allograft), which does not reflect actual price differences of a factor higher than 10. Rue et al. (1993) present average treatment costs per patient of 40,758 €, not specifying the type of costs included, whereas direct hospital costs of 15,853 € for unspecified skin grafts have been calculated by the NHS Centre for Reviews and Dissemination (2003f). The authors of the analysed publications could not find a positive effect of tissue-engineered products on wound closure compared to conventional treatments (e.g. Rue et al., 1993). Because of a lack of strong evidence for superiority of tissue engineering treatments the cost-effectiveness for burn treatment favours the conventional treatment. Tissue-engineered skin replacement options currently are mainly used for critical burn patients.

Chronic wounds (ulcers)

For the treatment of ulcers the data base is slightly better than for burns. Six studies are available that analyse treatment costs as well as effectiveness of tissue-engineered products, some derived data from clinical trials. But also in this case calculated costs differ for the tissue engineering treatment as well as for the conventional ones, mostly due to differences in the cost factors included in the studies, the type of ulcer treated, the time frame observed and other methodological aspects.

Regarding the average product costs per patient, conventional products (different gauze types or hydroactive dressing plus collagenase) are estimated at 10 € to 600 €, whereas tissue-engineered product costs per patient can be expected at 763 € to 2,774 €, depending on the product. Meaume & Gemmen (2002) calculate costs 5 to 7 times higher for the tissue engineering treatment than for the conventional treatment, for a time period of 12 weeks. Allenet et al. (2000) present average treatment costs per ulcer that are slightly lower for the tissue-engineered product due to a better healing rate (8,159 € and 8,641 €). Sibbald et al. (2001) show similar costs for the tissue engineering and the conventional treatment (about 1,200 € to 1,400 €). Compared to treatment with Unna's boot, which is considered to be a rather expensive conventional alternative, tissue engineering treatment is estimated to be 7,000 € cheaper (Augustin et al., 2002; NHS Centre for Reviews and Dissemination, 2001; Schonfeld et al., 2000). Augustin et al. (2002) compared costs for the tissue engineering treatment (4,370 €) with costs for hydrocolloid dressing (7,530 €) and lipid gauze dressing (10,897 €), but the data have not been collected under the same conditions at the same time and cannot be compared directly. Clinical trials are on-going to fill this data gap.

Concerning effectiveness, positive results for tissue-engineered products have been recorded, with more rapid healing and a higher rate of ulcers healed compared to conventional treatments (e.g. Sibbald et al., 2001, model calculations, Meaume & Gemmen, 2002; NHS Centre for Reviews and Dissemination, 2001; Schonfeld et al., 2000). According to the publications identified, one tissue-engineered product seems to have proven cost-effectiveness, especially for severe and recalcitrant ulcers. Other products seem to be cheaper in purchasing and further studies might prove also their cost-effectiveness.

Autologous chondrocyte transplantation (ACT)

Six studies have been analysed for ACT, providing a comparison of costs, cost-effectiveness analysis or even cost-utility analysis. However, the studies have several shortcomings, partly not specifying the conventional treatment they use as a comparator, lack of sound effectiveness data for conventional as well as for the tissue engineering option, results on costs are often based on model computations, experts opinion or literature, small patient groups have been investigated, control groups are missing, or the calculation of the quality adjusted life years (QUALYs)¹⁴ is unclear.

For ACT the total direct medical costs including rehabilitation range from 7,410 € to 27,044 € (Minas & Chiu, 2000; NHS Centre for Reviews and Dissemination, 2003c). Conventional treatments are calculated with 1,250 € to 4,200 €. Also German experts considered the ACT treatment being more costly (ACT: 5,000 € to 7,000 €, conventional: 1,250 € to 4,200 €, not including costs of hospital stay), but the assumed reduced need for revision surgery and reduced need for total knee replacement at young age would balance the cost in favour of ACT treatment (Arbeitsgemeinschaft ACT und Tissue Engineering, 2002). The NHS Centre for Reviews and Dissemination (2003a) state a slightly better effectiveness of 0.8 QUALYs for ACT after 10 years, but at a high price (13,213 €QUALY). Direct comparisons of total knee replacement with ACT calculating QUALYs revealed that the costs for each additional QUALY after two years were 6,207 € for total knee replacement and 6333 € for ACT (Jackson et al., 2001; Minas & Chiu, 2000). Accordingly cost-effectiveness for selected patients is seen positively by the authors. However, ACT is not reimbursed by health insurances in Germany, Switzerland or Canada. In the USA FDA approval is restricted to treatments of specific knee defects.

¹⁴ In cost-utility-analyses utility measures are calculated in form of QUALYs which enables comparison of the cost-effectiveness of different treatments.

Cell seeded synthetic vascular grafts

No tissue-engineered vascular graft product is currently on the market. There have been several long-term clinical trials for synthetic vascular grafts seeded with autologous endothelial cells, which show very promising results concerning similar or improved patency rates (Deutsch et al., 1999; Laube et al., 2000; Meinhart et al., 2001; NHS Centre for Reviews and Dissemination, 2003d). Synthetic grafts are used if autologous vessels are not available for bypass surgery. However, small diameter synthetic grafts are prone to thrombotic events. No data for tissue-engineered product or treatment costs are available. Generally costs for coronary bypass surgery are very high, the costs mainly originating from the surgery, the hospital stay and possible complications post-operative (10,500 € to 18,000 €, NHS Centre for Reviews and Dissemination, 2003b; NHS Centre for Reviews and Dissemination, 2003e). As the conventional graft itself is rather inexpensive, and, based on experiences with the other fields of tissue engineering, the production of seeded grafts might be rather expensive, cost-effectiveness of tissue-engineered grafts might be difficult to achieve at this stage.

The overview on cost and effectiveness analyses clearly shows a need for further and better clinical studies to be able to evaluate cost-effectiveness of tissue-engineered products. The available data, with all necessary reservations due to the small number of studies and several methodological shortcomings, do not provide clear evidence for the superiority of tissue-engineered products. On the other hand, the available data do not exclude potential benefits regarding quality of life, long-term savings or indirect effects of tissue-engineered products. Additionally, the tissue-engineered products currently on the market have to compete with a variety of alternative treatment options, and do not target new and unique treatment strategies or having a life saving function. This might be different for future tissue-engineered products, providing treatments for diseases which could not be treated satisfactorily before (e.g. diseases or damage of the CNS). Furthermore, product costs of tissue-engineered products might go down in the future due to streamlined production technologies and economies of scale.

4.2.3 Business models and strategies

Tissue engineering is considered a hybrid business, being located between pharmaceuticals and medical devices. Accordingly, also the business models differ (Table 4.1). Pharmaceutical companies have to calculate with long development times of more than 8 years and high up-front investments in research and development of several hundred million Euro. This is balanced by large markets of several billion Euro, high gross margins and exclusivity because of patent protection. Medical device companies face short development times of about 1 year, lower investment cost in R&D, but also focused markets with lower gross margins and less exclusive patent protection. Tissue engineering seems to combine long development times and medium investment costs in R&D with low gross margins and small markets – at least for the products being commercialised up to now. Tissue engineering is a very recent development and it is far too early to try and determine or identify a successful business model.

Table 4.1: Business models for pharmaceuticals, medical devices and tissue engineering products

Pharmaceuticals	Medical Devices	Tissue Engineering products
High up-front investment in R&D	Lower up-front investment in R&D	Medium up-front investment in R&D
Long development times	Short development times	Medium to long development times
High gross margins	Low gross margins	Low gross margins
Large markets	Focused markets	Focused markets

Source: Fraunhofer ISI

There are several other unique issues to be faced: The manufacturing of autologous products has a limited scale, but still is a labour intensive process, with intra-patient variability, limited possibilities for planning as products are manufactured on demand and logistics and customer assistance for these novel products present an important cost factor. The production of allogeneic products can eliminate some of these problems and offers more space for cost containment through e.g. automation. The delivery on demand poses the challenge of longer-term storage. Generally, the distribution and storage of tissue-engineered products has not been solved yet and probably can be compared to the handling of whole organs. The logistics need to be coordinated with the specific needs (e.g. surgery procedure schedules) of the users.

The marketing of tissue-engineered products could be done directly or via licensing. Partners might be helpful, but it needs to be decided if they should come from the pharmaceutical or the medical device industry and if large or small organisations (product specialist) would be preferable. Tissue-engineered products are very complex and unfamiliar for many physicians and surgeons. Thus highly educated marketing staff is required as well as other marketing strategies e.g. workshops, practical trainings.

Tissue engineering companies today seem to have a rather narrow but not unique scientific-technological basis. However, a broad knowledge base seems to be necessary reflecting the multidisciplinary character of tissue engineering, comprising cell biology, developmental biology, material sciences, engineering, medicine, chemistry etc. Clinical expertise is essential also for the development of tissue-engineered products. Intellectual property rights and know-how seems to be fragmented between different companies and need to be creatively combined (Petit-Zeman, 2001). A limited patent search in the database of the US Patent and Trademark Office (USPTO) for the time period 1988 to 2001 (for details see Annex 2) identified about 470 patents, mostly from companies (88%). US companies hold most of the patents (355), followed by companies from the European Union (30), Switzerland (15) and Japan (10). Almost no patents could be found in the database of the European Patent Office using the same keywords although it records several tissue engineering patent applications. However, US patents received from countries outside the USA are a clear indication for the high market-relevance these patents have. Additionally, the overall number of patents identified was rather low, probably partly due to the combination of keywords used for the search. Industry representatives pointed out that many of the applications had been developed and published by physicians in academia long before commercialisation, making patenting impossible. Moreover, patenting in the cell culture area seems to be difficult because a process needs to be patented which might be difficult. A patent on a process might also be difficult to enforce, thus these developments possibly will be handled rather as a business secret.

Many tissue engineering companies today compete with alternative conventional products and with each other having similar product portfolios (e.g. skin sector). Considering the high investment costs and the relatively small sales in limited markets (much smaller than for pharmaceuticals), products must have an excellent clinical performance. Especially the products currently available, which face strong competition and are often based on an improvement of the quality of life rather than survival of the patient, must provide added value in disease treatment. Longer-term clinical trials, carried out according to scientific standards might help to convince medical doctors to apply tissue-engineered products and might also provide the necessary data for reimbursement.

As pointed out by several interviewed experts, often a clear focus on the market is missing. The product development starts at the scientific level and considers the scientific and technological feasibility. The demand side and the application of the products need to be taken into account and the respective markets need to be critically assessed. Already during product development manufacturing and quality control standards, which are not regulated yet on a European level, need to be respected.

4.2.4 Research support

Experts as well as literature (WTEC Panel, 2002) coincide with the view that generally there is an equally high level of research in the EU and in the USA. According to a brief survey carried out in the context of this study and covering research funding organisations in the EU and acceding countries, research funding focuses mainly on the sectors biomaterials, cells and biomolecules. Engineering design aspects and automation are relatively neglected areas. Some countries do not prioritise funding support (Czech Republic, Belgium, Hungary, Ireland). The US National Institutes of Health indicated that they support research in all tissue engineering areas, which could be a reason for the perceived lead of the USA in sub-fields of engineering design aspects and automation (WTEC Panel, 2002). Furthermore, there are more border-line approaches in research in the USA. France, Germany, Sweden and UK are considered to make the greatest contribution to research in the EU through funding of large scale research programmes and research centres, which is partly reflected in the ranking of publication authorship occurrence (see above). The relative lack of authorship of France and Sweden might be connected to the fact that tissue engineering has become a funding priority only recently.

The perception of experts is that support for tissue engineering research is less intensive and focused in the EU than in the USA. Europe also has less networking between the public and private tissue engineering research communities. This situation could improve with the commencement of the European 6th Framework Programme and the implementation of two new instruments: the Integrated Project and the Network of Excellence. Both instruments aim at creating large scale research cooperations within the EU in the context of realising the European Research Area. Tissue engineering research will be funded within the Thematic Priority 1 (Genomics and Biotechnology for Health, 2,255 million €) and Thematic Priority 3 (Nanotechnology and nanosciences, knowledge-based multifunctional materials and new production processes and devices, 1,300 million €). From the Expressions of Interest submitted in June 2002, tissue engineering projects concerning connective tissue, organ repair and biobanks as well as applications of stem cells in new cell-based therapies and for tissue regeneration have been evaluated as highly relevant and the topics might be funded in the coming years.

4.2.5 Ethical issues

The use of human tissue raises questions about the conditions of donation, ownership and financial interest, which have to be clear so as not to inhibit development of therapies involving cells and tissues. The proposed Directive on tissue banking (European Commission, 2002a) states that donors of parts of the human body for cell and tissue procurement should not benefit financially from the donation. It also refers to the European Convention on Human Rights and Biomedicine from 1997 and its additional protocol from 2002, stating the need for free, informed and specific consent from the donor and prohibiting financial gains from donation. Ownership of donated tissue is not discussed. A court decision in the USA in 1990 denied ownership to patients of cells harvested during the course of medical treatment (WTEC Panel, 2002). On the other hand, the making of profits with donated tissues by companies without compensating the donors, raises concerns. Privacy and anonymity against the background of traceability of tissue donors and recipients for safety purposes is an issue.

Another topic that raises ethical concerns in the context of tissue engineering is embryonic stem cell research. The question about whether such research should be allowed, involving as it does the destruction of an embryo to gain embryonic stem cells, is connected to issues about the dignity and moral status of the embryo. This problem is dealt with differently in the European Member States, ranging from prohibition (e.g. Germany, Austria) to the possibility of embryo creation for research purposes under certain conditions (UK). The European Commission recently published an overview on the situation of human embryonic stem cell research in Europe (European Commission, 2003a). The possibility to fund embryonic stem cell derivation for research via the 6th Framework Programme is currently under discussion, but a moratorium

is in place until the end of 2003. In July 2003 the European Commission submitted to the Council and the Parliament a proposal for establishing detailed guidelines for EU funding of research involving human embryos and human embryonic stem cells, which should be implemented by end of 2003 (European Commission, 2003b). The European Group on Ethics in Science and New Technologies (EGE), an advisory body reporting directly to the President of the European Commission, has issued several opinions with relevance to tissue engineering¹⁵. Regarding research and use of embryonic stem cells, the EGE calls for strict public control by a centralised authority and the consideration of basic ethical principles, notably respect for human dignity, principles of consent, justice and beneficence, freedom of research and proportionality between the research methods used and the aims pursued. European pluralism concerning philosophical, moral and legal approaches need to be respected. Embryonic stem cells and processes involving embryonic stem cells are considered patentable as long as the stem cells have been modified by *in vitro* treatments or genetically modified to fulfil the legal requirements for patentability.

Another type of cells with potential use in tissue engineering are embryonic germ cells retrieved from fetal tissue. The acceptability is closely related to concerns about abortion (Nuffield Council on Bioethics, 2000). Xenogeneic cells are viewed critically as a potential source of cells because of their pathogenic potential. However, the rights of animals, their welfare and the importance of maintaining respect for nature are ethical questions associated to the genetic modification of donor animals.

The potentially high costs of growing and storing tissues and organs might become an issue in the future, due to potential financial restrictions for health care systems. Another issue concerns equal access to tissue engineering treatment. Will those who can afford the treatment perhaps live longer and gain political power? Also the conception of oneself and of being “human” might change if the body consists of replacement parts and is considered “renewable” (Satava, 2002).

4.2.6 Employment and training needs

The effects of tissue engineering on employment are difficult to predict as tissue engineering is in its infant to adolescent phase. Currently it seems that the effects are small, with employees in the order of 3300 for 73 companies in USA and Europe (Lysaght & Reyes, 2001). There is certainly a training need for people involved in the whole process from research and production to authorisation, selling and application. Especially sales personnel need to be well trained to have the expertise to explain the products and to convince the users. On the other hand physicians need to be trained to be able to use the novel products. Towards this aim specific centres for excellence are being developed by companies in cooperation with specific hospitals to assure the high quality of surgery involving their products.

¹⁵ Ethical Aspects of Human Tissue Banking dated 21 July 1998; Ethical Aspects of Research Involving the Use of Human Embryo in the context of the 5th Framework Programme, dated 23 November 1998; and Ethical Aspects of Human Stem Cell Research and Use dated 14 November 2000, Ethical aspects of patenting inventions involving human stem cells, dated 7 May 2002.

5 CONCLUSIONS

Tissue engineering is a comparatively young area of interdisciplinary research which led to the first product approval 1996 in the USA (ACT product Carticel). Research in this area presents a priority funding field in many Member States and European research activities can be considered equal to the USA, with Germany and UK leading. Funding initiatives cover mainly the central tissue engineering areas of biomaterials, cells and biomolecules. Technical engineering aspects as well as bioinformatics, both significant supporting sectors, seem to be less emphasised. The overall aim of on-going research is to improve the performance of tissue-engineered products and to enlarge application areas, which for the time being concentrate on comparatively simple tissues such as skin, cartilage and bone. In this innovative field human stem cells play a crucial role. Research on human embryonic stem cells is seen as essential for gaining insight into fundamental processes during cell development, differentiation, interaction with biomolecules, cells and biomaterials. At the moment, because of overriding ethical concerns in several Member States, but also because of probable immunogenic reactions to allogeneic cells, the use of adult stem cells is given more attention for tissue engineering applications in Europe.

Several research results have been transferred to the commercial sector. A total of 113 companies was identified as being active in tissue engineering, with 54 companies engaging in tissue engineering as defined. Comparing the structure of the sector in the EU and the USA, it seems that the sector is developed to a similar extent regarding the number and size (number of employees) of companies. The results of this study indicate that the European market is characterised by young, small, research-based and technology-oriented companies, most of them SMEs with less than 50 employees. A survey in the USA in 2001 came to comparable results. The medical devices and pharmaceutical companies represent only about 30% of the companies. There seems to be more public/private collaborations in research in the USA compared to the EU, which indicates a certain lack of networking of the tissue engineering sector in Europe.

Market estimates for tissue-engineered products have been very promising, ranging from 80 billion € for the USA alone (MedTech Insight, 2000) to 400 billion € worldwide (Langer & Vacanti, 1993). More moderate estimates still calculated a global market of 3.9 billion € by 2007 (Business Communication Company, 1998) or of 270 million € by 2007 for skin products alone (MedMarket Diligence, 2002). The reality provides much lower figures with world-wide sales of tissue-engineered products probably not surpassing 60 million € in 2002. There are several factors that influence the development of the sector:

- To date tissue-engineered products are available only for specific applications in the wound sector (skin substitutes with sales of about 20 million €), in knee cartilage repair (autologous chondrocyte transplantation ACT with sales of about 40 million €) and bone repair (two tissue-engineered products commercialised). Scientific (e.g. issue of vascularisation, interaction of different cell types, control of cell behaviour and proliferation) and technical problems (e.g. design of bioreactors, transportation and storage) need to be solved for the development of larger and more complex tissues.
- Tissue engineering treatments compete with alternative conventional treatments, several of the latter being much simpler and also cheaper. Those tissue-engineered products which are currently on the market do not have unique life saving function or outstanding comparative advantages regarding effectiveness or treatment costs, but rather improve the quality of life through potentially faster and better healing of wounds and less need for repeated surgery. This situation might change in future if more sophisticated and novel tissue-engineered products (e.g. tissue-engineered intervertebral discs, larger bone substitutes, tissue-engineered heart valves) become available which show clear comparative effectiveness or which allow the treatment of diseases for which no other treatment exists. Also product costs might be lowered in the future due to advanced production techniques and increased scale of production.

Conclusions

- As yet there are only few products commercialised and even fewer products have been subject to scientific studies to prove the cost-effectiveness of the treatment compared to conventional alternatives. Additionally, most of the tissue-engineered products are still in an early stage of their development and are developed and marketed by small biotech companies, which do not have the resources for large, long-term clinical trials to provide this kind of data.
- Lack of cost-effectiveness data on the other hand is the main reason for insurance companies not reimbursing treatment with tissue-engineered products. It must be added that these data are also not available for all conventional treatments. No reimbursement results in difficulties in accessing the market, apart from niche markets where patients are prepared to pay by themselves (e.g. aesthetic surgery).
- Another challenge for tissue engineering companies is the current European legal situation concerning tissue-engineered products. At the moment there is no specific regulatory framework for this special kind of product, for which neither the directives on medical devices nor on pharmaceuticals seem fully appropriate or applicable. Thus Member States proceed on a case-by-case basis, applying different rules and enabling access to the respective national markets. This is one reason why European companies focus on autologous products, for which authorisation procedures are simpler. For products, which have high development costs and would need clinical trials as in the case of allogeneic products, national markets might be too small.
- Tissue engineering companies still have to develop a functioning business model, combining medium to high up-front investment costs with - at least for the currently commercialised product types - rather focused markets and small gross margins, thus combining characteristics of the pharmaceutical and medical devices sectors. Current sales are too small to cover the operating costs. The markets need to be developed and presently there seems to be redundancy and overcapacity, with companies competing with each other and well established alternative conventional products. Two major US tissue-engineering companies, producing skin substitutes, went into bankruptcy at the end of 2002. This might be a first sign of an early consolidation of the tissue engineering sector.

The tissue engineering sector is too young to enable the definition of a successful business model and strategy. Many perceive it as being in its infant phase. Several of the companies involved seem to have a narrow, but not unique scientific-technological basis, which may need to be enlarged and combined in an intelligent way in this interdisciplinary field. Progress in the development of a European legal framework will certainly create a more favourable environment for further development of the sector. A workable solution needs to be found for small biotechnology companies to enable them to carry out studies to prove cost-effectiveness of their products, also with a long-term perspective. On the other hand these additional requirements might discourage the development of new tissue-engineered products, which often are expensive and for which cost-effectiveness might not be easy to show. A convincing performance of tissue-engineered products will certainly help to persuade physicians and surgeons to apply these novel products. As Seifalian et al. (2002) put it for grafted vessels: “the only way fully biological tissue-engineered coronary or biological grafts will ever get to replace native vessels will be to outperform all currently available grafts in clinical trials.” However, this would imply the necessity of cost-effectiveness information for the traditional treatment options, which is also lacking in many cases.

Initiatives like LIFE (Living Implants from Engineering, USA) in 1999 promised to be able to tissue engineer a human heart within ten years. The time scale for a lab-grown heart has subsequently been extended to about 25 years by the founder of LIFE (Zandonella, 2003), which may still be significantly over-optimistic. Painting too bright a picture of the potential of tissue engineering will probably not help the development and the creation of confidence in this innovative technology. Several significant scientific and technological challenges still have to be met, of which fundamental understanding of cell and tissue growth and behaviour, constant quality in large-scale production and storage and control of quality and functionality are only a few examples. However, in the medium to long term, the manifold research activities surely will

result in the development of novel products, for example in the cardiovascular area or concerning diseases of the central nervous system, for which no treatment is available today.

REFERENCES

- Allenet, B., Parea, F., Lebrun, T., Carr, L., Posnett, J., Martini, J. & Yvon, C. (2000). Cost-effectiveness modeling of Dermagraft for the treatment of diabetic foot ulcers in the French context. *Diabetes & Metabolism*, **26**, 125-132.
- Al-Saffar, N. & Revell, P. (2000). Differential expression of transforming growth factor-alpha and macrophage colony-stimulating factor/colony-stimulating factor-1R (c-fms) by multinucleated giant cells involved in pathological bone resorption at the site of orthopaedic implants. *J Orthop Res*, **18**, 800-807.
- Altman, G., Diaz, F., Jakuba, C., Calabro, T., Horan, R., Chen, J., Lu, H., Richmond, J. & Kaplan, D. (2003). Silk-based biomaterials. *Biomaterials*, **24**(3), 401-406.
- Alyaudtin, R. N., Reichel, A., Lobenberg, R., Ramge, P., Kreuter, J. & Begley, D. J. (2001). Interaction of poly(butylcyanoacrylate) nanoparticles with the blood-brain barrier in vivo and in vitro. *J Drug Target*, **9**(3), 209-21.
- Arbeitsgemeinschaft ACT und Tissue Engineering. (2002). Beantwortung des Fragenkatalogs zur Autologen Chondrozytenimplantation (ACI). Stellungnahme zur ACT an den Bundesausschuss Krankenhaus "Neue Untersuchungs- und Behandlungsverfahren". Arbeitsgemeinschaft ACT und Tissue Engineering unter Schirmherrschaft der Deutschen Gesellschaft für Unfallchirurgie (DGU) und der Deutschen Gesellschaft für Orthopädie und Orthopädische Chirurgie (DGOOC) pp. 77
- Arnst, C. & Carey, J. (1998). Biotech Bodies. Business Week 27 July 1998
- Augustin, M., Ermuth, T., Johnsen, S., Zschocke, I., Schöpf, E. & Vanscheidt, W. (2002). Kosteneffekte der autologen Keratinozyten-Transplantation bei schweren therapieresistenten chronischen Wunden. *ellipse*, **18**, 1-10.
- Augustin, M., Siegel, A. & Heuser, A. (1999). Chronic leg ulcers: cost evaluation of two treatment strategies. *Journal of Dermatological Treatment*, **10**(Supplement 1), S21-S25.
- Behraves, E., Zygourakis, K. & Mikos, A. (2003). Adhesion and migration of marrow-derived osteoblasts on injectable in situ crosslinkable poly(propylene fumarate-co-ethylene glycol)-based hydrogels with a covalently linked RGDS peptide. *J Biomed Mater*, **65A**, 260-270.
- Bloom, B. S., de Pourville, N. & Straus, W. L. (2003). Cost of illness of Alzheimer's disease: how useful are current estimates? *Gerontologist*, **43**(2), 158-64.
- Boden, S. D., Zdeblick, T. A., Sandhu, H. S. & Heim, S. E. (2000). The use of rhBMP-2 in interbody fusion cages. Definitive evidence of osteoinduction in humans: a preliminary report. *Spine*, **25**(3), 376-81.
- Bottaro, D., Liebmann-Vinson, A. & Heidaran, M. (2002). Molecular signaling in bioengineered tissue microenvironments. *Ann N Y Acad Sci*, **961**, 143-153.
- Business Communication Company. (1998). Cell therapy and tissue engineering: Emerging products. Business Communication Company pp.
- Calvo, P., Gouritin, B., Chacun, H., Desmaele, D., D'Angelo, J., Noel, J., Georjgin, D., Fattal, E., Andreux, J. & Couvreur, P. (2001). Long-circulating PEGylated polycyanoacrylate nanoparticles as new drug carrier for brain delivery. *Pharm res*, **18**, 1157-1166.
- Chaikof, E., Matthew, H., Kohn, J., Mikos, A., Prestwich, G. & Yip, C. (2002). Biomaterials and scaffolds in reparative medicine. *Annals of the New York Academy of Sciences*, **961**, 96-105.
- Chapekar, M. (2000). Tissue engineering: Challenges and opportunities. *J Biomed Mater Res (Appl Biomater)*, **53**, 617-620.
- Claase, M., Grijpma, D., Mendes, S., De Bruijn, J. & Feijen, J. (2003). Porous PEOT/PBT scaffolds for bone tissue engineering: Preparation, characterization, and in vitro bone marrow cell culturing. *J Biomed Mater Res*, **64A**, 291-300.
- Concord Corporate Finance Research. (2002). Biomaterialien und Tissue Engineering am Neuen Markt. Concord Effekten AG pp. 91
- Curtis, A. & Riehle, M. (2001). Tissue engineering: the biophysical background. *Phys Med Biol*, **46**, R47-65.
- Dalby, M., Riehle, M., Yarwood, S., Wilkinson, C. & Curtis, A. (2003). Nucleus alignment and cell signaling in fibroblasts: response to a micro-grooved topography. *Exp Cell Res*, **284**, 274-282.

References

- Dalby, M., Yarwood, S., Riehle, M., Johnstone, H., Affrossman, S. & Curtis, A. (2002). Increasing fibroblast response to materials using nanotopography: morphological and genetic measurements of cell response to 13-nm-high polymer demixed islands. *Exp Cell Res*, **276**, 1-9.
- Deasy, B. & Huard, J. (2002). Gene therapy and tissue engineering based on muscle-derived stem cells. *Current Opinion in Molecular Therapeutics*, **4**(4), 382-389.
- Deutsch, M., Meinhart, J., Fischlein, T., Preiss, P. & Zilla, P. (1999). Clinical autologous in vitro endothelialization of infrainguinal ePTFE grafts in 100 patients: a 9-year experience. *Surgery*, **126**(5), 847-55.
- Di Lullo, S. (2002). Heart valve therapy. Presentation at the Investor Conference, November 7, 2002 pp. www.edwards.com/AboutUs/InvestorInformation/Events/webcast.aspx.
- Dick, J. (2002). Advanced Wound Management. Smith & Nephew interim result presentation 1 August 2002 pp. www.smith-nephew.com/investors/interim02/ns-index.html.
- Dieners, P., Sonnenschein, E. & Köhler, M. (2002). Tissue Engineering - Rechtliche Grundlagen und neue Entwicklungen. *PharmaR*, **9**, 325-333.
- EMA. (2001). Points to consider in the manufacture and quality control of human somatic cell therapy medicinal products. Committee for Proprietary Medicinal Products CPMP pp.
- European Commission. (2001). Opinion on The State of the Art Concerning Tissue Engineering, The Scientific Committee on Medicinal Products and Medical Devices, SANCO/SCMPMD/2001/0006 Final.
- European Commission. (2002). Commission Communication: Life Sciences and Biotechnology - A Strategy for Europe (COM/2002/27), OJ, Vol. C55, pp. 3-32.
- European Commission. (2002a). Proposal for a Directive of the European Parliament and of the Council on setting standards of quality and safety for the donation, procurement, testing, processing, storage, and distribution of human tissues and cells (COM/2002/319).
- European Commission. (2003). Human tissue and cell engineering products. DG Enterprise pp. http://europa.eu.int/comm/enterprise/medical_devices/consult_tissue_engineer.htm.
- European Commission. (2003a). Commission staff working paper: Report on human embryonic stem cell research, SEC (2003) 441.
- European Commission. (2003b). Proposal for a Council Decision amending decision 2002/834/EC on the specific programme for research, technological development and demonstration: "Integrating and strengthening the European research area" (2002-2006) (COM/2003/390).
- Fossum, M., Gustafson, C., Nordenskjold, A. & Kratz, G. (2003). Isolation and in vitro cultivation of human urothelial cells from bladder washings of adult patients and children. *Scand J Plast Reconstr Surg Hand Surg*, **37**, 41-45.
- Frerichs, O., Fansa, H., Schicht, C., Wolf, G., Schneider, W. & Keilhoff, G. (2002). Reconstruction of peripheral nerves using acellular nerve grafts with implanted cultured Schwann cells. *Microsurgery*, **22**, 311-315.
- Frost & Sullivan. (2001). Multiple sclerosis therapies: poised for take-off pp. [wysiwyg://110/http://www.inpharm.com/intelligence/frost011101.html](http://www.inpharm.com/intelligence/frost011101.html).
- Gassner, U. (2001). Tissue Engineering im Normenschungel. *MedR*, **11**, 553-561.
- Griffith, L. & Naughton, G. (2002). Tissue engineering - current challenges and expanding opportunities. *Science*, **295**, 1009-1014.
- Guilak, F. (2001). Functional tissue engineering of articular cartilage: The role of biomechanics. *Proceedings of the WTEC workshop on tissue engineering research in the United States*, 175 - 179.
- Hagell, P., Nordling, S., Reimer, J., Grabowski, M. & Persson, U. (2002). Resource use and costs in a Swedish cohort of patients with Parkinson's disease. *Mov Disord*, **17**(6), 1213-20.
- Hench, L. & Polak, J. (2002). Third-generation biomedical materials. *Science*, **295**, 1014-1017.
- Hevehan, D. L., Wenning, L. A., Miller, W. M. & Papoutsakis, E. T. (2000). Dynamic model of ex vivo granulocytic kinetics to examine the effects of oxygen tension, pH, and interleukin-3. *Exp Hematol*, **28**(9), 1016-28.
- Holden, C. & Vogel, G. (2002). Plasticity: time for a reappraisal? *Science*, **296**(5576), 2126-9.

- Humes, H. D. (1996). Application of gene and cell therapies in the tissue engineering of a bioartificial kidney. *Int J Artif Organs*, **19**(4), 215-7.
- Humes, H. D. (2000). Bioartificial kidney for full renal replacement therapy. *Semin Nephrol*, **20**(1), 71-82.
- Informa Pharmaceuticals. (2000). CNS-markets and therapies pp. [wysiwyg://138/http://www.inpharm.com/intelligence/inpharma011000.html](http://www.inpharm.com/intelligence/inpharma011000.html).
- IsoTis Corporate Communications and Investor Relations. (2003). Tissue-engineered cartilage pp. www.isotis.com.
- Ivanovic, Z., Bartolozzi, B., Bernabei, P. A., Cipolleschi, M. G., Rovida, E., Milenkovic, P., Praloran, V. & Dello Sbarba, P. (2000). Incubation of murine bone marrow cells in hypoxia ensures the maintenance of marrow-repopulating ability together with the expansion of committed progenitors. *Br J Haematol*, **108**(2), 424-9.
- Jackson, D. W., Simon, T. M. & Aberman, H. M. (2001). Symptomatic articular cartilage degeneration: the impact in the new millennium. *Clin Orthop*, **391**, S14-S25.
- Jasmund, I. & Bader, A. (2002). Bioreactor developments for tissue engineering applications by the example of the bioartificial liver. *Adv Biochem Eng Biotechnol*, **74**, 99-109.
- Jiang, Y., Jahagirdar, B. N., Reinhardt, R. L., Schwartz, R. E., Keene, C. D., Ortiz-Gonzalez, X. R., Reyes, M., Lenvik, T., Lund, T., Blackstad, M., Du, J., Aldrich, S., Lisberg, A., Low, W. C., Largaespada, D. A. & Verfaillie, C. M. (2002). Pluripotency of mesenchymal stem cells derived from adult marrow. *Nature*, **418**(6893), 41-9.
- Jones, I., Currie, L. & Martin, R. (2002). A guide to biological skin substitutes. *Br J Plast Surg*, **55**(3), 185-93.
- Kessler, P. D. & Byrne, B. J. (1999). Myoblast cell grafting into heart muscle: cellular biology and potential applications. *Annu Rev Physiol*, **61**, 219-42.
- Khor, E. & Lim, L. (2003). Implantable applications of chitin and chitosan. *Biomaterials*, **24**(13), 2339-2349.
- Kirkpatrick, C., Krump-Konvalinkova, V., Unger, R., Bittinger, F., Otto, M. & Peters, K. (2002). Tissue response and biomaterial integration: the efficacy of in vitro methods. *Biomol Eng*, **19**, 211-217.
- Kohlmeier, L., Kroke, A., Pöttsch, J., Kohlmeier, M. & Martin, K. (1993). *Ernährungsabhängige Krankheiten und ihre Kosten*. Nomos-Verlagsgesellschaft mbH und Co KG, Baden-Baden.
- Kreuter, J. (2001). Nanoparticulate systems for brain delivery of drugs. *Adv Drug Deliv Rev*, **47**, 65-81.
- Landesbank Baden-Württemberg Equity Research. (2001). Tissue Engineering. Landesbank Baden-Württemberg Equity Research pp. 62
- Langer, R. & Vacanti, J. P. (1993). Tissue engineering. *Science*, **260**(5110), 920-6.
- Laube, H. R., Duwe, J., Rutsch, W. & Konertz, W. (2000). Clinical experience with autologous endothelial cell-seeded polytetrafluoroethylene coronary artery bypass grafts. *The Journal of Thoracic and Cardiovascular Surgery*, **120**, 134-141.
- Lendlein, A. & Langer, R. (2002). Biodegradable, elastic shape-memory polymers for potential biomedical applications. *Science*, **296**, 1673-1676.
- Lin, H., Lei, J., Wininger, D., Nguyen, M., Khanna, R., Hartmann, C., Yan, W. & Huang, S. (2003). Multilineage potential of homozygous stem cells derived from metaphase II oocytes. *Stem Cells*, **21**(2), 152-161.
- Livingston, T., Ducheyne, P. & Garino, J. (2002). In vivo evaluation of a bioactive scaffold for bone tissue engineering. *Journal of Biomedical Materials Research*, **62**(1), 1-13.
- Lu, H., El-Amin, S., Scott, K. & Laurencin, C. (2003). Three-dimensional, bioactive, biodegradable, polymer-bioactive glass composite scaffolds with improved mechanical properties support collagen synthesis and mineralization of human osteoblast-like cells in vitro. *Journal of Biomedical Materials Research*, **64A**(3), 465-474.
- Lysaght, M. J. & Reyes, J. (2001). The growth of tissue engineering. *Tissue Eng*, **7**(5), 485-93.
- Lysaght, M. J. & Hazlehurst, A. L. (2003). Private sector development of stem cell technology and therapeutic cloning. *Tissue Eng*, **9**(3), 555-561
- McKee, J., Banik, S., Boyer, M., Hamad, N., Lawson, J., Niklason, L. & Counter, C. (2003). Human arteries engineered in vitro. *EMBO Rep*, **4**(6), 633-638.

References

- Meaume, S. & Gemmen, E. (2002). Cost-effectiveness of wound management in France: pressure ulcers and venous leg ulcers. *Journal of Wound Care*, **11**, 219-224.
- MedMarket Diligence. (2002). Cell-based tissue engineering. MedMarket Diligence LLC pp. 247
- MedTech Insight. (2000). Cost-benefit - new approach to EU regulation of human tissue products. Medtech Insight pp. 1-5
- Meinhart, J., Deutsch, M., Fischlein, T., Howanietz, N., Froschl, A. & Zilla, P. (2001). Clinical autologous in vitro endothelialization of 153 infrainguinal ePTFE grafts. *Ann Thorac Surg*, **71**(5 Suppl), S327-S331.
- Menasche, P. (2002). Cell therapy of heart failure. *C R Biol*, **325**(6), 731-738.
- Minas, T. & Chiu, R. (2000). Autologous chondrocyte implantation. *Am. J. Knee Surg*, **13**, 41-50.
- Mironov, V., Boland, T., Trusk, T., Forgacs, G. & Markwald, R. (2003). Organ printing: computer-aided jet-based 3D tissue engineering. *Trends in biotechnology*, **21**(4), 157-161.
- Murray, F. (2002). Innovation as co-evolution of scientific and technological networks: exploring Tissue Engineering. *Research Policy*, **31**, 1389-1403.
- Nasser, B., Pomerantseva, I., Kaazempur-Mofrad, M., Sutherland, F., Perry, T., Ochoa, E., Thompson, C., Mayer, J. J., Oesterle, S. & Vacanti, J. (2003). Dynamic rotational seeding and cell culture system for vascular tube formation. *Tissue Eng*, **9**, 291-299.
- National Institutes of Health Bioengineering Consortium. (2001). Reparative medicine: Growing tissues and organs. National Institutes of Health pp. 55
- Naughton, G. (2002). From lab bench to market. Critical issues in tissue engineering. *Ann. N.Y. Academy of Sciences*, **961**, 372-385.
- Nerem, R. & Seliktar, D. (2001). Vascular tissue engineering. *Annu Rev Biomed Eng*, **3**, 225-243.
- Nguyen, K. & West, J. (2002). Photopolymerizable hydrogels for tissue engineering applications. *Biomaterials*, **23**, 4307-4314.
- NHS Centre for Reviews and Dissemination. (2001). An economic assessment of Apligraf (R) (Graftskin) for the treatment of hard-to-heal venous leg ulcers. NHS Economic Evaluation Database (NHS EED), York: University of York.
- NHS Centre for Reviews and Dissemination. (2003a). Autologous chondrocyte transplantation for cartilage defects in the knee joint. The Cochrane Library, Issue 1. Oxford: Update Software.
- NHS Centre for Reviews and Dissemination. (2003b). The clinical and financial impact of portaccess coronary revascularization (Structured abstract). The Cochrane Library, Issue 1. Oxford: Update Software.
- NHS Centre for Reviews and Dissemination. (2003c). Effectiveness of autologous chondrocyte transplantation for hyaline cartilage defects in knees: a rapid and systematic review. The Cochrane Library, Issue 1. Oxford: Update Software.
- NHS Centre for Reviews and Dissemination. (2003d). Impact of an aggressive stenting strategy on initial and one-year follow-up costs in patients undergoing coronary angioplasty (Structured abstract). The Cochrane Library, Issue 1. Oxford: Update Software.
- NHS Centre for Reviews and Dissemination. (2003e). Morbidity, cost, and six-month outcome of minimally invasive direct coronary artery bypass grafting (structured abstract). The Cochrane Library, Issue 1. Oxford: Update Software.
- NHS Centre for Reviews and Dissemination. (2003f). Outcome and cost analysis for outpatient skin grafting. The Cochrane Library, Issue 1. Oxford: Update Software.
- Noel, D., Djouad, F. & Jorgensen, C. (2002). Regenerative medicine through mesenchymal stem cells for bone and cartilage repair. *Current opinion in investigational drugs*, **3**(7), 1000-1004.
- Nuffield Council on Bioethics. (2000). Stem cell therapy: The ethical issues. Nuffield Council on Bioethics pp.
- Oberpenning, F., Meng, J., Yoo, J. J. & Atala, A. (1999). De novo reconstitution of a functional mammalian urinary bladder by tissue engineering. *Nat Biotechnol*, **17**(2), 149-55.
- OECD. (2001). *Health at a glance*. OECD.

- Pagán Westphal, S. (2003). Virgin birth method could found stem cell dynasties. *NewScientist*, **26 April 2003**, 17.
- Parente, S. (1997). Estimating the economic cost offset of using dermagraft-TC as an alternative to cadaver allograft in the treatment of graftable burns. *Journal of Burn Care & Rehabilitation*, **18**, 18-24.
- Pariente, J., Bordenave, L., Bareille, R., Baquey, C. & Le Guillou, M. (2000). Cultured differentiated human urothelial cells in the biomaterials field. *Biomaterials*, **21**, 835-839.
- Park, Y., Tirelli, N. & Hubbell, J. (2003). Photopolymerized hyaluronic acid-based hydrogels and interpenetrating networks. *Biomaterials*, **24**, 893-900.
- Pego, A., Poot, A., Grijpma, D. & Feijen, J. (2003). Biodegradable elastomeric scaffolds for soft tissue engineering. *J Control Release*, **87**, 69-79.
- Perets, A., Baruch, Y., Weisbuch, F., Shoshany, G., Neufeld, G. & Cohen, S. (2003). Enhancing the vascularization of three-dimensional porous alginate scaffolds by incorporating controlled release basic fibroblast growth factor microspheres. *J Biomed Mater Res*, **65A**, 489-497.
- Petit-Zeman, S. (2001). Regenerative medicine. *Nature Biotechnology*, **19**, 201-206.
- Prestwich, G. & Matthew, H. (2002). Hybrid, composite, and complex biomaterials. *Annals of the New York Academy of Sciences*, **961**, 106-108.
- Rangappa, S., Fen, C., Lee, E., Bongso, A. & Wei, E. (2003). Transformation of adult mesenchymal stem cells isolated from the fatty tissue into cardiomyocytes. *Ann Thorac Surg*, **75**, 775-779.
- Reya, T., Duncan, A. W., Ailles, L., Domen, J., Scherer, D. C., Willert, K., Hintz, L., Nusse, R. & Weissman, I. L. (2003). A role for Wnt signalling in self-renewal of haematopoietic stem cells. *Nature*, **423**(6938), 409-14.
- Rhodes, N., Hunt, J. & Williams, D. (1997). Macrophage subpopulation differentiation by stimulation with biomaterials. *J Biomed Mater Res*, **37**, 481-488.
- Risbud, M. & Sittering, M. (2002). Tissue engineering: advances in in vitro cartilage regeneration. *Trends in Biotechnology*, **20**(8), 351-356.
- Roell, W., Lu, Z. J., Bloch, W., Siedner, S., Tiemann, K., Xia, Y., Stoecker, E., Fleischmann, M., Bohlen, H., Stehle, R., Kolossov, E., Brem, G., Addicks, K., Pfitzer, G., Welz, A., Hescheler, J. & Fleischmann, B. K. (2002). Cellular cardiomyoplasty improves survival after myocardial injury. *Circulation*, **105**(20), 2435-41.
- Rue, L. W., 3rd, Cioffi, W. G., McManus, W. F. & Pruitt, B. A., Jr. (1993). Wound closure and outcome in extensively burned patients treated with cultured autologous keratinocytes. *J Trauma*, **34**(5), 662-667; discussion 667-668.
- Russell, J. & Cross, S. (2001). Commercial prospects for tissue engineering. Business Intelligence Program-SRI Consulting pp. 15
- Ryan, E. A., Lakey, J. R., Rajotte, R. V., Korbitt, G. S., Kin, T., Imes, S., Rabinovitch, A., Elliott, J. F., Bigam, D., Kneteman, N. M., Warnock, G. L., Larsen, I. & Shapiro, A. M. (2001). Clinical outcomes and insulin secretion after islet transplantation with the Edmonton protocol. *Diabetes*, **50**(4), 710-719.
- Saltzman, W. & Olbricht, W. (2002). Building drug delivery into tissue engineering. *Nature Reviews Drug Discovery*, **1**, 177-186.
- Satava, R. (2002). Disruptive visions: moral and ethical challenges from advanced technology and issues for the new generation of surgeons. *Surgical Endoscopy*, **16**(10), 1403-1408.
- Schlosshauer, B., Muller, E., Schroder, B., Planck, H. & Muller, H. (2003). Rat Schwann cells in bioresorbable nerve guides to promote and accelerate axonal regeneration. *Brain Res*, **963**, 321-326.
- Schonfeld, W. H., Villa, K. F., Fastenau, J. M., Mazonson, P. D. & Falanga, V. (2000). An economic assessment of Apligraf (Graftskin) for the treatment of hard-to-heal venous leg ulcers. *Wound Repair Regen*, **8**(4), 251-257.
- Schulz, J., Tompkins, R. & Burke, J. (2000). Artificial skin. *Annu. Rev. Med.*, **51**, 231-238.
- Seifalian, A. M., Tiwari, A., Hamilton, G. & Salacinski, H. J. (2002). Improving the clinical patency of prosthetic vascular and coronary bypass grafts: the role of seeding and tissue engineering. *Artif Organs*, **26**(4), 307-20.

References

- Shapiro, A. M., Lakey, J. R., Ryan, E. A., Korbitt, G. S., Toth, E., Warnock, G. L., Kneteman, N. M. & Rajotte, R. V. (2000). Islet transplantation in seven patients with type 1 diabetes mellitus using a glucocorticoid-free immunosuppressive regimen. *N Engl J Med*, **343**(4), 230-8.
- Shirota, T., He, H., Yasui, H. & Matsuda, T. (2003). Human endothelial progenitor cell-seeded hybrid graft: proliferative and antithrombogenic potentials in vitro and fabrication processing. *Tissue Eng*, **9**, 127-136.
- Sibbald, R. G., Torrance, G. W., Walker, V., Attard, C. & MacNeil, P. (2001). Cost-effectiveness of Apligraf in the treatment of venous leg ulcers. *Ostomy Wound Manage*, **47**(8), 36-46.
- Sodian, R., Lemke, T., Fritsche, C., Hoerstrup, S., Fu, P., Potapov, E., Hausmann, H. & Hetzer, R. (2002). Tissue-engineering bioreactors: a new combined cell-seeding and perfusion system for vascular tissue engineering. *Tissue Eng*, **8**, 863-870.
- Stamm, C., Westphal, B., Kleine, H. D., Petzsch, M., Kittner, C., Klinge, H., Schumichen, C., Nienaber, C. A., Freund, M. & Steinhoff, G. (2003). Autologous bone-marrow stem-cell transplantation for myocardial regeneration. *Lancet*, **361**(9351), 45-46.
- Stegemann, J. & Nerem, R. (2003). Phenotype modulation in vascular tissue engineering using biochemical and mechanical stimulation. *Ann Biomed Eng*, **31**, 391-402.
- Stock, U., Vacanti, J., Mayer Jr, J. & Wahlers, T. (2002). Tissue engineering of heart valves - current aspects. *Thorac Cardiovasc Surg*, **50**, 184-193.
- Strauer, B. E., Brehm, M., Zeus, T., Gattermann, N., Hernandez, A., Sorg, R. V., Kogler, G. & Wernet, P. (2001). [Intracoronary, human autologous stem cell transplantation for myocardial regeneration following myocardial infarction]. *Dtsch Med Wochenschr*, **126**(34-35), 932-8.
- The Royal Academy of Engineering. (2003). *The Japanese Approach to Tissue Engineering*. London, pp. 59.
- Toner, M. & Kocsis, J. (2002). Storage and translational issues in reparative medicine. *Ann N Y Acad Sci*, **961**, 258-62.
- Tran, N., Li, Y., Bertrand, S., Bangratz, S., Carteaux, J., Stoltz, J. & Villemot, J. (2003). Autologous cell transplantation and cardiac tissue engineering: potential applications in heart failure. *Biorheology*, **40**, 411-415.
- Verfaillie, C. (2002). Adult stem cells: assessing the case for pluripotency. *Trends in Cell Biology*, **12**(11), 502-508.
- Vogel, G. (2002). Studies cast doubt on plasticity of adult cells. *Science*, **295**, 1989-1991.
- Warren, S., Hedrick, M., Sylvester, K., Longaker, M. & Chen, C. (2002). New directions in bioabsorbable technology. *J Neurosurg (Spine 4)*, **97**, 481-489.
- Werthen, M., Sellborn, A., Kalltorp, M., Elwing, H. & Thomsen, P. (2001). In vitro study of monocyte viability during the initial adhesion to albumin- and fibrinogen-coated surfaces. *Biomaterials*, **22**, 827-832.
- Whitaker, M., Quirk, R., Howdle, S. & Shakesheff, K. (2001). Growth factor release from tissue engineering scaffolds. *Journal of Pharmacy and Pharmacology*, **53**, 1427-1437.
- Willert, K., Brown, J. D., Danenberg, E., Duncan, A. W., Weissman, I. L., Reya, T., Yates, J. R. & Nusse, R. (2003). Wnt proteins are lipid-modified and can act as stem cell growth factors. *Nature*, **423**(6938), 448-52.
- Williams, D. (1999). *The Williams Dictionary of Biomaterials*. Liverpool University Press, Liverpool.
- Wissink, M., Beernink, R., Pieper, J., Poot, A., Engbers, G., Beugeling, T., van Aken, W. & Feijen, J. (2001). Binding and release of basic fibroblast growth factor from heparinized collagen matrices. *Biomaterials*, **22**, 2291-2299.
- Woods, J. D. & Humes, H. D. (1997). Prospects for a bioartificial kidney. *Semin Nephrol*, **17**(4), 381-386.
- WTEC Panel. (2002). Panel report on tissue engineering research. International Technology Research Institute pp. 208
- Xue, L. & Greisler, H. (2003). Biomaterials in the development and future of vascular grafts. *Journal of Vascular Surgery*, **37**(2), 472-480.
- Zandonella, C. (2003). The beat goes on. *Nature*, **421**, 884-886.

ANNEX 1: TISSUE ENGINEERING COMPANIES IN EUROPE

Table A: Tissue engineering companies in Europe (as of April 2003)

Source: Fraunhofer ISI

Country	Company	Company Type	Company Size	TE Relevance
Austria	Educell Zellkultivierung F&E GmbH	Biotech	SME	Core
Austria	InnovaCell	Biotech	SME	Core
Austria	igor – Institut für Gewebe- und Organrekonstruktion	Biotech	SME	Core
Belgium	Genzyme Europe	Biotech	large	Core
Belgium	TIGenix NV	Biotech	SME	Core
Belgium	beta-cell	Biotech	SME	Core
Belgium	XCELLentis	Biotech	SME	Core
Czech Republic	Altius Co.Ltd.	Biotech	SME	Core
Czech Republic	Educell	Biotech	SME	Core
Czech Republic	CPN, Ltd.	Biotech	unknown	Core
Denmark	Interface Biotech SA	Biotech	SME	Core
Denmark	Nordic Bioscience A/S	Biotech	SME	Broader def.
Denmark	Coloplast A/S	Medical Device	large	Broader def.
Finnland	Ark Therapeutics Oy	Biotech	SME	Broader def.
Finnland	Cellomeda Oy	Biotech	SME	Broader def.
Finnland	Fibrogen Europe Oy	Biotech	SME	Broader def.
France	Imedex Biomateriaux	Pharma	SME	Core
France	Laboratoires Genevrier	Pharma	SME	Core
France	Myosix SA	Biotech	SME	Core
France	Neurotech SA	Biotech	SME	Core
France	Biopredic International	Biotech	unknown	<i>in vitro</i> use
France	Coletica	Biotech	SME	<i>in vitro</i> use
France	Galderma R & D	Pharma	large	<i>in vitro</i> use
France	Groupe DermScan	Biotech	SME	<i>in vitro</i> use
France	L'Oreal Recherche	Medical Device	Large	<i>in vitro</i> use
France	SkinEthic Laboratories	Biotech	SME	<i>in vitro</i> use
Germany	Ars Arthro AG	Biotech	SME	Core
Germany	ARTISS GmbH	Biotech	SME	Core
Germany	BioTissue Technologies AG	Biotech	SME	Core
Germany	Cell Lining GmbH	Biotech	SME	Core
Germany	CO.DON AG	Biotech	SME	Core
Germany	MeGa Tec GmbH	Biotech	SME	Core
Germany	Switch Biotech AG	Biotech	SME	Core
Germany	TETEC Tissue Engineering GmbH Technologies GmbH	Biotech	SME	Core
Germany	Vascular Biotech GmbH	Biotech	SME	Core
Germany	Verigen Transplantation Service International AG	Biotech	SME	Core
Germany	IBFB GmbH	Biotech	SME	Core
Germany	Innocoll GmbH	Medical Device	SME	Core
Germany	CellMed AG	Biotech	SME	Core
Germany	Cytonet AG	Biotech	SME	Core
Germany	DeveloGen	Biotech	SME	Core
Germany	Trans Tissue Technologies GmbH	Biotech	SME	Core

Annex 1

Country	Company	Company Type	Company Size	TE Relevance
Germany	Kourion Therapeutics GmbH	Biotech	SME	Core
Germany	CellTec GmbH	Biotech	SME	Core
Germany	Hybrid Organs	Biotech	SME	Core
Germany	CellSystems Biotechnologie Vertrieb GmbH	Biotech	SME	in vitro use
Germany	ACM-Biotech GmbH	Biotech	SME	in vitro use
Germany	EDI GmbH	Biotech	SME	in vitro use
Germany	In Vitro Biotec GmbH	Biotech	SME	in vitro use
Germany	ALVITO Biotechnologie GmbH	Biotech	SME	Broader def.
Germany	B.Braun Melsungen	Medical Device	Large	Broader def.
Germany	Biomet Merck Biomaterials GmbH	Medical Device	SME-Large	Broader def.
Germany	Aventis Behring GmbH	Pharma	Large	Broader def.
Germany	Beiersdorf AG	Pharma	Large	Broader def.
Germany	Biovision GmbH Biomaterial	Biotech	SME	Broader def.
Germany	Cardion AG	Biotech	SME	Broader def.
Germany	Curasan	Biotech	SME	Broader def.
Germany	Dr. Suwelack Skin & Health Care AG	Pharma	SME	Broader def.
Germany	Teraklin AG	Medical Device	SME	Broader def.
Germany	Aesculap AG & Co.	Medical Device	Large	Broader def.
Germany	In Vitro Systems & Services GmbH	Biotech	SME	Broader def.
Germany	Innovent Technologieentwicklung e.V.	Biotech	SME	Broader def.
Germany	Osartis GmbH & Co. KG	Biotech	SME	Broader def.
Germany	ProBioGen AG	Biotech	SME	Broader def.
Germany	Minucells and Minutissue Vertriebs GmbH	Biotech	SME	Broader def.
Italy	Fidia Advanced Biopolymers srl	Pharma	large	Core
Italy	Novamont SpA	Medical Device	unknown	Broader def.
Luxembourg	Cellon S.A.	Biotech	SME	Broader def.
Spain	Advancell	Biotech	SME	in vitro use
Spain	Grupo Ferrer Internacional S.A.	Pharma	Large	Broader def.
Spain	Genetrix SL	Biotech	SME	Broader def.
Sweden	Cell Therapeutics Scandinavia	Biotech	SME	Core
Sweden	Cellfactory	Biotech	SME	Core
Sweden	Karocell Tissue Engineering AB	Biotech	SME	Core
Sweden	Neuronova	Biotech	SME	Core
Sweden	Vitrolife AB	Medical Device	SME	Core
Sweden	AnaMar Medical	Biotech	SME	Broader def.
Sweden	Biora AB	Medical Device	SME	Broader def.
Sweden	Medicarb	Biotech	SME	Broader def.
Sweden	Q-Med	Medical Device	SME	Broader def.
Sweden	Angio genetics AB	Biotech	SME	Broader def.
Switzerland	Kuros	Biotech	SME	Core
Switzerland/The Netherlands	IsoTis SA	Biotech	SME	Core

Country	Company	Company Type	Company Size	TE Relevance
Switzerland	Novartis	Pharma	Large	Broader def.
Switzerland	Centerpulse AG	Medical Device	large	Broader def.
Switzerland	Degradable Solutions AG	Medical Device	SME	Broader def.
Switzerland	Nisco Engineering Inc.	Medical Device	SME	Broader def.
Switzerland	Synthes stratec AG	Medical Device	Large	Broader def.
Switzerland	BD Biosciences	Biotech	Large	Broader def.
The Netherlands	Matrix Medical BV	Biotech	SME	Core
The Netherlands	Biomat BV	Medical Device	SME	Broader def.
The Netherlands	Bioscan BV	Medical Device	SME	Broader def.
The Netherlands	Leadd	Biotech	SME	Broader def.
The Netherlands	Pharming Group NV	Biotech	SME	Broader def.
The Netherlands	Polyganics BV	Medical Device	SME	Broader def.
United Kingdom	Axordia	Biotech	SME	Core
United Kingdom	Cell Factors	Biotech	SME	Core
United Kingdom	Cerestem	Biotech	SME	Core
United Kingdom	Intercytex Limited	Biotech	SME	Core
United Kingdom	Multicell	Biotech	unknown	Core
United Kingdom	Odontis	Biotech	SME	Core
United Kingdom	Regentec	Biotech	SME	Core
United Kingdom	ReInnervate	Biotech	SME	Core
United Kingdom	Reneuron	Biotech	SME	Core
United Kingdom	Renovo Ltd	Biotech	SME	Core
United Kingdom	Smith & Nephew Ltd	Medical Device	large	Core
United Kingdom	Advanced Medical Solutions	Medical Device	SME	Broader def.
United Kingdom	Apatech	Medical Device	SME	Broader def.
United Kingdom	Enact Pharma	Biotech	unknown	Broader def.
United Kingdom	Johnson & Johnson Advanced Wound Care	Medical Device	large	Broader def.
United Kingdom	PPL Therapeutics plc	Biotech	SME	Broader def.
United Kingdom	Tissuemed Ltd	Medical Device	SME	Broader def.
United Kingdom	TissueScience Laboratories	Medical Device	SME	Broader def.

SME: <500 employees, large: >500 employees

ANNEX 2: METHODOLOGIES APPLIED

In this study tissue engineering is defined as *the regeneration of biological tissue through the use of cells, with the aid of supporting structures and/or biomolecules* (European Commission, 2001). The study focuses on tissue engineering for therapeutic purposes. The definition includes autologous and allogeneic cells and tissues and excludes xenogeneic materials, apart from genetically humanised xenogeneic cells. Gene therapy as well as direct transplantation are also excluded. Tissue-engineered products are combined tissue/non-tissue type products that do not exert their effect primarily through metabolic, pharmacological or immunological means. The cells and tissues used have been substantially modified in the production process, i.e. they are not directly transplanted.

The study covers the European Union as well as the ten acceding countries that will join the EU in May 2004.

The part on current research activities is based on the WTEC Panel report on tissue engineering research published in 2002 (WTEC Panel, 2002). The information was complemented with other recent scientific publications based on literature database search and advice from Prof. Kirkpatrick. Expert interviews (8 experts from Germany, UK and USA) are the basis for information on future trends in tissue engineering research.

The overview on tissue-engineered products and markets is based on market studies, company reports and internet home pages, scientific literature searches, health statistics and questionnaire-guided interviews with experts from industry and academia (14 experts from the Netherlands, France, Germany, Italy, Portugal, Belgium).

A bibliometric analysis was the approach to characterise the European tissue engineering research community compared to the USA. For the period 1991 to 2002, the Science Citation Index in the ISI Web of Science database was searched for the keywords [Human + (tissue engineer*)] or [human + regenerat* + (skin or bone or vascular or valves or cartilage) or chondrocyte or osteochondral or endothelial or keratinocytes].

The list of keywords and the search combination represents a compromise considering limited resources and time. As a result publications not containing the key word “tissue engineering”, but being relevant to that field such as publications on basic research in the area of cell-biomaterial interactions, might not have been captured by the search. After these procedures, the relevant publications were analysed according to the country of the author, his/her affiliation to the private or public sector and for joint authorship between public and private sectors. For this publication analysis, the multiple counting system was used to calculate multi-authored publications. Consequently, the statistics show the number of authorship occurrences within the total number of publications instead of the number of publications produced by particular authors.

A similar methodology was used for a patent search in the USPTO database for the same time period. In addition to keywords, patents produced by companies that are assumed to be active in tissue engineering were also searched.

Tissue engineering companies have been identified and characterised on the basis of international and national biotechnology directories, internet and scientific literature searches, market studies, companies reports and internet home pages as well as direct contacts.

Two small surveys were carried out to cover research funding activities in Europe and regulatory issues in the Member States. 45 research funding organisations in 13 EU Member States, 8 acceding countries and the USA were approached to answer a short questionnaire on their funding priorities concerning tissue engineering research. 10 Member States, 5 acceding countries and two organisations from the USA answered the questionnaire. Another brief

questionnaire was sent to regulatory bodies in 13 Member States and Slovenia for information on applied or planned regulation concerning human tissue-engineered products.

The analysis of cost-effectiveness of human tissue-engineered products was based on available publications and focused on three case studies, selected for relevance for present and future tissue engineering applications and availability of data. The literature search was done using MEDLINE database¹⁶, the Cochrane Library¹⁷, the NHS Centre for Reviews and Dissemination Database¹⁸, and the German Agency for Health Technology Assessment¹⁹. 38 articles were included in the cost-effectiveness analysis. The cost data that were presented in the various studies in different currencies were converted to €. The exchange reference rates used for this purpose are shown below. The cost information was taken to serve as a landmark. This means they were taken "as is" from the publications without any discounting, i.e. without taking into account that costs and effects can be realised at different points in time.

Table B: The exchange rates used for conversion to Euro

AUD	(Australian dollar)	1.7852	GBP	(Pound sterling)	0.68110
BEF	(Belgian Francs)	40.3399	SEK	(Swedish krona)	9.2527
CAD	(Canadian dollar)	1.5711	USD	(US dollar)	1.0723
FRF	(Francs Français)	6.55957			

Source: European Central Bank, <http://www.ecb.int/stats/eurofxref/eurofxref-xml.html>, retrieved 27 March 2003, and for the conversion rates of the EURO-Member Countries: <http://www.ecb.int/change/conversion.htm>

Additional information about the applied methodologies can be found in the respective reports of the WPs on the IPTS web page (www.jrc.es).

¹⁶ <http://www.ncbi.nih.gov/entrez/queri.fcgi>

¹⁷ via the German Network on Evidence-Based Medicine

¹⁸ <http://agatha.yorck.ac.uk/welcome.htm>

¹⁹ http://www.dimdi.de/en/hta/hta_dahta_dimdi/index.html