ORGAN EXTRACT- AND CELL THERAPY
FOR REJUVENATION AND TREATMENT OF CHRONIC–DEGENERATIVE DISEASES

INTRODUCTION

The living organism, as a part of the universe, is embedded in the magnitude of the latter's dimensions. Within the wide boundaries of the electron mass of 10^{-31} to the cosmic dimension of 10^{17}, life takes up only a small span of 10^{-5} to 10^{1}, thus only 6 out of 48 dimensions. Life begins at the organizational stage of the single cell and ends in the domain of the multi-cellular organism. Life is characterized by the capability of the cells to transform the continuous energy and material losses of lifeless nature into new energies and structures. A cellular state deriving from these principles stands in reciprocal harmony with its lifeless environment and is described as healthy. Loss of utilization or deficiency of material of lifeless nature leads to defective functioning of the cellular state, to disease.

The paramount objective of any medical treatment should be the restoration of all capabilities of the functional unit of life, the cell. Only healthy functional cells will enable their associates, the tissues and organs to fulfil their task ensuring development and survival of the organism. Medicine today orientates itself towards relieve of symptoms that can be registered or are deducible by means of scientific and technical aids (microscope, electron microscope, biochemical data, electronic recordings). It is thereby neglecting all dimensional areas below and above the so-called objective detection methods, and in doing so, it defines its limits. Thought levels below the visible correlations with nature, such as in homeopathy, or above them, such as in the embedment of life in earthly and cosmic relations in anthroposophy lead a reluctant and marginal existence in the medical concept of today.

It may be that the brilliant idea Niehans put to practice 70 years ago of making young cells available to diseased or ageing organs was erroneous. The idea was, nevertheless, rewarded with practical success. We know today that the implanted cells are decomposed in order of magnitude under microscopic observation, but it is precisely by these easily transportable and incorporable particles that important building substances for the repair of cellular and sub-cellular defects are supplied to the diseased organism. Moreover, the repair of cellular defects opens the possibility of a new materialization of the elementary functions of life, the utilization of the materials and energies of the environment. Where upon, not only are symptoms eliminated, but the opportunity created of producing afresh the fundamental principle of life, and with it health.

The evolution of life embraces a semi-circle consisting of maturing, maturity and ageing. Disabilities, disorders and diseases increase with the distance from the middle of this semi-circle in the direction to the beginning and end of the biological existence. The main field for a therapy aimed at repairing the biological potential therefore lies inevitably in the first and last decades of life. In the course of practical and clinical experiences with organ extract- and cell therapy the following areas of indications have crystallized:

I. Congenital and infantile development disturbances
    1. Metabolic disorders
    2. Chromosome aberrations
    3. Insufficiencies and depressions in the blood-forming system
    4. Immunologic deficiencies

5. Infantile disturbances of the central nervous system

II. Degenerative changes caused by old age
    1. General degeneration and loss of vitality
    2. Degenerative manifestations
        a. Cardiovascular system
        b. Central nervous system
        c. Connective tissue
        d. Digestive tract
        e. Skin

III. Defective functioning of organs or organ systems arising from constitutional causes or disease
    1. Chronic organic diseases of the heart, circulatory system, liver, joints
    2. Chronic organic diseases and malfunctioning of the endocrine and immune system
    3. Hereditary degenerative diseases of the central nervous systems

IV. Concomitant tumour therapy

Within these indications the documented experimental values of organ extract- and cell therapy in respect of the strength of testimony range between single observations of uncommon diseases and statistical substantiations of up to thousand-fold observations.

The implantation treatment with foetal or young cell suspensions which has taken its place in medical history under the term 'Cellular Therapy' operates by way of the following therapeutic factors:

- The rapidly growing intrinsic content of the foetal and young tissue of biochemical substrates and enzymes
- The foetal tissue's own composition of minerals and trace elements
- The foetal tissue's own biological development power which leads to rapid tissue growth

Whilst biochemical substances and elements are analyzed in great detail, the biological development power is not easily measurable with scientific parameters. We know that roots, street pavements and stone work can lift, but we are not in a position to interpret and to measure this power. In our therapeutic concept, this force however plays a big role since it alone creates the possibility and precondition for the application of the elements and the utilization of enzymes and substrates for new structures.

Away from the indicated connections with microcosm (elements, trace elements, elementary particles) and macrocosm (solar energy, cosmic radiation), cell therapy should always be practiced in the framework of holistic medicine. This means that necessary measures in the conduct of life, nutrition, physiotherapy, psychotherapy and medical treatment must be incorporated insofar as they are required in the individual patient's situation. No form of therapy is a one and only redeeming religion. The 'mono-symptom - mono-substance' claim of modern pharmacotherapy is one of the most disastrous dogmatizing efforts of our time.

Organ extract- and cell therapy provides a body, under suitable application, with the opportunity of transforming the elementary function of life, the utilization of environmental energies and materials into new energies and structures. This step in a new dimension in medicine leads, in the longer aspect from a 'medicine for disease' to a 'medicine for health'. The therapeutic efforts are not focused on the elimination of single symptoms of disease but serve in the restoration of the vital elementary functions of an organism [1].

BASIC PRINCIPAL OF ORGAN EXTRACT- AND CELL THERAPY

In organ extract- and cellular therapy, cells or extracts of various foetal animal tissues are administered via intramuscular injection into the human body for therapeutic purposes. These xenograft implants are broken down into their basic elements (enzymes, polypeptides, deoxyribonucleic acids, ribonucleic acids and other basic organic substances) and reused by the cells, tissues and organs of the person treated. The fundamental theory behind organ extract – and cell therapy is the principle 'Similia Similibus' or 'Like Cures Like', as stated by Paracelsus, a Swiss physician and philosopher of the 16th century. Paracelsus and many other early physicians believed that the best way to rebuild or revitalise ill organs or ageing tissue was to use healthy living cells of the same tissue type. Modern organ extract- and cell therapy refers to treatment by injection with cellular elements and whole cells from healthy unborn or foetal sheep or calves specially bred for medical purposes under a controlled environment. Organ extract- and cell therapy actually "wakes up" dormant cells within the human body, thereby stimulating the growth and function of existing tissue and repairing or regenerating old and malfunctioning cells. Organ extract- and cell therapy offers something that vitamins, minerals and other conventional or natural treatments cannot. It can provide the exact components necessary for injured or diseased tissue to heal and regenerate. While most pharmaceutical drugs work by suppressing certain symptoms over a short period of time and only as long as they are taken, organ extract- and cell therapy are stimulating the body's own healing and revitalizing powers and exert a long term effect.

THE THERAPEUTIC TECHNIQUE Preparation of Organ Extracts- and Cells for Therapy

The method of injection-implantation of xenograft organ extracts and cellular preparations for therapeutic purposes as defined by Niehans had its beginnings in the surgical transplantation of glands. Organ extract- and cellular therapy is a modified and expanded form of this transplantation technique. Instead of the surgical transplantation of the entire gland, an organ extract or a suspension of single cells and minute units of cell clusters are injected intramuscularly (implantation per injectionem). The method was actually first reported in 1927 by Küttner, a surgeon in Breslau, and was soon forgotten [2]. In 1931, Niehans re-discovered this method and developed it further over the next decades into a comprehensive therapeutic procedure. The novelty of Niehans work was not only the technical variation of transplantation per injection. Of far greater importance was that he extended the injection-implantation to parenchymal organs and other types of tissue, as well as the fact that he preferred the use of foetal cells as source material. Foetal animal tissues are much easier to obtain under sterile conditions, and they possess only very low-grade anaphylactic properties, hence they are much better tolerated by the recipient. However, in most animal foetuses, the pituitary gland, the testes, the ovary, the adrenal gland and the parathyroid glands are either too small or not sufficiently activated for the use as source material for organ extract- and cell therapy. These tissues are therefore often obtained from very young animals after birth, mostly calves and sheep. As a matter of principle, only differentiated foetal tissues and not embryonic cells should be used for organ extract- and cell therapy, since embryonic (undifferentiated) cells could increase the risk of uncontrolled growth (cancerous growth) after implantation. Sheep are the preferred donor animals because of their sturdy health and strong resistance to disease, and also for the ease they can be kept in isolated flocks. In addition, allergic reactions are rare after exposure to sheep protein (sheep are rarely used for the production of sera, hence prior exposure to sheep protein with antibody development unlikely).

The resection or harvest of the organs must be performed with strict surgical asepsis in order to avoid contamination. The operating veterinarian and all assistants must wear protective surgical gowns and sterile face masks. The operating room must provide a fully aseptic atmosphere. The

use of bacteriostatic solutions or antibiotic drugs to control possible contamination during surgical harvest of the foetal organs is discouraged, since it is not known whether these drugs will change the biochemical properties of the tissues and thus influence their therapeutic efficacy. Sterility tests are done for all organ extracts- and cell therapy preparations that are preserved by either deep-freezing or lyophilization. Unfortunately, there is no time for such testing when the fresh cell method is used for therapy, all the more important is here absolute sterility during harvest and cell preparation.

When preparing organ extract- and cell therapy preparations, speed and aseptic conditions are of outmost importance. In order to prevent autolysis of the tissues and organs harvested, the preparation time must be kept to a minimum until the suspended cells are either injected fresh (fresh cell therapy) or deep frozen (fresh frozen cell therapy) or freeze-dried (lyophilization).

Preparation of Fresh Organ Extract and Cells

It is believed that a prerequisite for best therapeutic efficacy of organ extract- and cell therapy preparations is the biochemical freshness of the source material, hence the importance that the transplanted cells remain in their un-denatured state. Denaturation of cellular material (autolysis) sets in shortly after the donor animal has been sacrificed. In organs with high concentrations of enzymes, such as pancreas, liver and adrenal gland, autolysis might start as early as 15 minutes after the death of the animal. Therefore, it is advisable that the organ harvest is performed by a well trained and skilled veterinarian and that the harvested organs are used right away for the preparation of the fresh cell therapy suspension. Niehans used this preparation method in the early days of cell therapy. This method has several disadvantages. In particular, it does not fulfil today's safety requirements for biological injectables since sterility can be compromised during the harvest of the cells and there is no time for microbiology testing of the material injected. This form of cell therapy also bears a higher risk for allergic reaction and more frequently induces local inflammation at the injection site. For these reasons, fresh cell therapy is rarely practiced today.

Preparation of Fresh Frozen Organ Extracts and Cells

This method uses fresh organ extracts or cells from sheep or calf foetus that are shock frozen according to a proprietary cryopreservation technique in liquid nitrogen at -196°C and afterwards stored at or below -80°C until injection. The advantage of this method is the preservation of physiological integrity and biological activity of the extracted substances and cellular material for a long period of time in the frozen state. Ultrastructural studies showed no significant tissue damage via the technique of cryopreservation: cellular membranes were intact, the mitochondria showed only discrete swelling and vacuoles were found in the endoplasmic reticulum. After thawing the fresh frozen cell preparations for injection their biological activity is fully maintained. Since the preparation of the fresh frozen cells involves several steps of cell washing to reduce blood contamination and thereby antigenic potential due to the ABO antigen system on the erythrocytes or other blood cell antigens, there is only a very low risk for allergic reaction and rarely local inflammation at the injection site. Therapy with fresh frozen organ extracts or cells is an improved method compared with the fresh cell therapy. The fresh frozen cell preparations are all tested for sterility and antigens; they can be stored at or below -80°C for years and shipped that way without problems to all corners of the world. Once thawed the contents of the injection vial has to be used immediately and should not be re-frozen.

Preparation of Lyophilized (Freeze-dried) Organ Extracts and Cells

Lyophilised organ extracts and cells are prepared by the process of freeze-drying. Organ extracts and cellular material will be frozen and dehydrated in vacuum at the same time, thereby maintaining the biological integrity of the biochemical substances in the organ extract and also maintaining cellular components. Lyophilization must not be confused with mere desiccation. At present, there is no better procedure known for preservation of tissue. With other methods of desiccation, denaturation of protein may result, whereas lyophilization, i.e. sublimation of the tissue fluid from the frozen material, will preserve specific biochemical compounds as well as microscopic and sub-

microscopic cell structures so well that micro-organisms, yeast cells, bacteria and some viruses can be stored for decades and still maintain their reproductive ability after re-hydration. On the other hand, the freeze drying process destroys a protein substance called intracellular cementum which holds cells together, but also triggers allergic reactions. Removing this substance by lyophilization thereby reduces the allergic potential of the organ extracts or cell preparations. Prior to administration, the freeze-dried organ extract- and cell preparations are reconstituted with normal saline or Ringer's solution. Therapy with lyophilised organ extracts or cells is a more improved method over the fresh cell method. The preparations can be tested for sterility and antigens; they can be stored easily and shipped without problems to all corners of the world.

Suspension Media

Most cellular therapists use Ringer's solution as the suspension medium. This solution contains NaCl 0.8%; KCL 0.02%; Ca CL_2 0.02%, and Na HCO_3 0.1%. Ringer's solution is however not the best suspension medium, and for that reason, we recommend either a specially modified Tyrode solution or Compton solution as a more suitable solution.

Implantation Technique

Organ extracts and cell suspensions are implanted by intramuscular injection, usually into the outer, upper quadrant of the M. gluteus maximus. In patients likely to develop decubitus ulcers, the injection can also be done into the M. quadriceps. Subcutaneous injections should be avoided, because the slowed resorption from fatty tissue and the risk of sterile abscess formation. Since organ extracts and cell suspensions contain large particles, large bore needles (outer diameter at least 1.5 mm) should to be used for injection. Care should be taken that such large bore needles do not punch out a piece of epidermis which could be carried into the deeper layers of the subcutaneous fat or muscle and cause sterile abscess formation. Intravascular injection must be avoided; otherwise embolism or shock could develop. The deposit of organ extract- and cell suspension should not be too large; otherwise resorption may be rendered more difficult. Deposits of more than 10 ml into one area should be avoided. If larger amounts are to be used therapeutically, then the injection needle should be re-directed after each 5-10 ml deposit to reach other layers of the muscle.

Dosage

The dosage of fresh organ extract- and fresh cell suspensions cannot be determined exactly. Due to the method of preparation neither the volume and weight nor the composition can be standardized. If the technique of tissue cutting and preparing the suspension is mastered, then a 20 ml suspension is approximately equal to 0.7 to 1.0 g of fresh tissue. With lyophilized preparations the circumstances are more favourable. Here, the exact weight of the desiccated cell material can be determined, knowing that present lyophilization leaves rest moisture of about 1%. Nevertheless, even when knowing the exact weight of cellular material after lyophilization, proper dosing is still difficult, since the efficacy of the biological material varies. Therapists with experience in organ extract- and cell therapy have realized that there is a direct relationship between the severity of organic dysfunction and the amount of suspended cells necessary for proper treatment of the respective condition. This corresponds clearly to the so called HalstedPrinciple, which states that autologous or isologues transplants from endocrine glands produce fewer reactions in the recipient if the corresponding gland is in a hypo-functioning state, thus in need of the transplant. Following the Halsted-Principle the dosage of organ extract- and cell therapy could be adjusted according to the severity of the disease for which the transplant is administered.

Prophylaxis of Zoonoses

Infectious diseases which are transmitted from animals to humans are called zoonoses or anthropozoonoses. If the animal used for harvest of organs and tissues should be diseased, the infectious agents could be transmitted to the receiving patient, even though complete aseptic conditions were observed throughout the resection of the organs or tissues and during preparation

of the organ extract or cell suspension. Customary clinical and pathological – anatomical examination of the donor animal are considered not sufficient to exclude, with reasonable certainty, the presence of disease. Therefore, additional safety steps must be taken to ensure that only healthy animals are used for organ and tissue harvest. These steps include the close supervision of the animal herd by a certified veterinarian, the clinical examination of the living animal prior to sacrifice (including body temperature daily for one week, erythrocyte sedimentation rate, complete blood count, tuberculin test, interpalpebral test for Q-fever, intradermal test for listeriosis, etc.) the autopsy of the donor animal, as well as serological and bacteriological tests. Even before introduction of these safety steps for the preparation of xenogeneic organ extract- and cell therapy there was no conclusive reports about transmission of zoonoses after treatment, despite the popularity of this treatment method and millions of transplants being performed over the years. Only one publication reports about a case of brucellosis which was believed to be transmitted by a fresh cell implant [3]. Today, as a rule, source animals should be selected and handled according to the guidelines of the European Agency for the Evaluation of Medicinal Products (London, 17 December 2003; Document: CPMP /1199/02: "Points to consider on xenogeneic cell therapy medical products"). Only when all examinations and test results (see above) are certifying the health of the donor animal and the preparation of the organ extract and/or cells for therapy has been carried out as is customary for manufacture of vaccines, sera or blood components, transmission of zoonoses by cellular therapy can, in all probability, be prevented.

IMMUNOLOGICAL ASPECTS OF ORGAN EXTRACT- AND CELL THERAPY

Xenogeneic Transplantation and Rejection

According to the laws of transplantation, one would expect organ extracts and cells from a xenogeneic source (for example sheep and calf) to be rejected immediately by the recipient patient. We know four immunological mechanisms by which xenotransplants are usually rejected: First, ***hyperacute rejection***, which is caused by xenoreactive natural antibodies and complement of the recipient acting against endothelial cells of the source animal organ. Second, ***acute vascular rejection*** caused by the combined effect of elicited xenoreactive antibodies and activated host natural killer cells and monocytes. In combination these stimuli (the anti-graft antibodies and the activated host cells) result in activation of the endothelial cells of the source organ. Endothelial cell activation leads to general inflammation with resultant thrombosis (platelet aggregation and activation of the coagulation cascade) resulting in organ rejection. Third, the xenograft counterpart of ***classical T cell mediated rejection*** of allografts (transplantation between individuals of the same species), which is directed against the major histocompatibility antigens of the donor cells, will almost certainly occur. Finally, xenografts may also be subject to ***chronic rejection*** in a manner analogous to allografts.

Organ Extract- and Cell Therapy without Immune System Suppression

More than 70 years of clinical practice and experience in the therapy with organ extracts and suspended cells mostly from sheep and calf foetuses, however, have shown that xenotransplantation of these materials does not invoke clinically appreciable rejection and is generally well tolerated. There are several reasons for this obvious discrepancy between theoretical anticipation and practical experience: First, hyper-acute and acute rejections are less likely with xenograft cells and tissues than with whole xenograft organs, because there is no immediate spatial contact of the transplant with the corresponding host organ to be treated, and the suspended cells and tissue fragments do not require connection to an intact vasculature of the host for survival. In comparison with the conventional implantation techniques of solid tissue fragments (endocrine glands or whole organs), injection-implantation has the advantage that the suspended cells or cell groups are readily taken up by bodily fluids of the host and transported from the site of injection. By virtue of their being suspended in a fluid medium, the cells and cell groups are readily able to come into close contact with the metabolic processes of the host. Blood transfusions and bone marrow

implants as performed in clinical practice can be quoted as examples. Second, the xenograft organ extract and suspended cells are not from adult animals with fully expressed major histocompatibility antigens on their cell surfaces, but they originate, as a rule, from foetal or immature juvenile tissue, which is known to have much less antigenic expression. Foetal cells have four basic properties that make them clinically useful for grafting or transplantation applications: their intrinsic plasticity, their ability to grow and proliferate, their ability to produce growth factors, and their reduced antigenicity compared to adult tissue. Murine and early human foetal and embryonic stem (ES) cells, for example, do not express HLA class I and II antigens, and demonstrate reduced surface expression of co-stimulatory molecules important for T cell activation. Transplantation of murine ES cells demonstrates long-term graft survival despite the fact that these cells do acquire HLA class II antigen expression after in vivo differentiation. Since they are able to accomplish long-term engraftment without the need for immune system suppression, their inability to induce an immune response is not likely to be the result of escaping immune surveillance, but rather due to their ability to colonize the recipient thymus and induce intra-thymic deletion of allo-reactive recipient T cells. Therefore, with the use of foetal organ extract- and cell therapy preparation a much higher tolerance of the recipient can be expected, allowing therapy without immune system suppression of the host.

Immune Reactions and Side Effects in Organ Extract- and Cell Therapy

Despite the low antigenicity and clinically excellent tolerability of foetal organ extract- and cell therapy preparations, there is no doubt that the human organism reacts to the implantation according to the laws of allergology. In principle, the immune mechanisms involved after injection of xenogeneic organ extract- and cell therapy preparations will depend on:

- the qualitative antigenic properties of the injected material
- the injection method with regard to site of injection
- the amount of implanted material and difference of the cells as to their organ and species-specificity
- the interval between the injections
- the nature and mode of reaction of the antibodies formed by the recipient

Low antigenicity of foetal xenograft cells due to low or lacking expression of major histocompatibility complex antigens has been explained above. The following discussion will concentrate on other factors that could invoke immune responses and untoward side effects with organ extract- and cellular therapy.

First of all, the tissues and cells used for implantation are contaminated with the foetal blood cells. These blood cells have clear antigenic potential. In sheep, for example, seven blood group systems have been identified and termed A, B, C, D, M, R and X. In cattle the number of blood such groups is eleven. Similar to cattle, the B system in sheep is highly polymorphic. The R system is similar to the J system in cattle, in that the antigen is soluble. The M-L system is involved in active red cell potassium transport and polymorphisms in this system result in breeds of sheep with varying erythrocyte potassium content. All blood group antigens could potentially increase antigenicity of the cell preparation. Therefore, the cells are thoroughly washed in Ringer's or Tyrode solution several times to reduce blood contamination and thereby antigenic potential. After these antigens have been removed by the repeated washing steps, the cells are either shock frozen with liquid nitrogen at -196°C according to a proprietary cryopreservation protocol or centrifuged with the resulting cell-pulp being used for lyophilisation.

Looking at dissolved protein as potential antigenic material in cell preparations, we must understand that following findings: Even after thorough washing of the cells to remove all blood contamination, xenogeneic protein may be found after suspension of these cells in nitrogen-free isotonic solution. This protein will be set free by the washed and suspended cells. Fresh cell-pulp and lyophilized cells re-suspended and ready for injection will also contain this soluble antigenic material in addition to the cellular tissue elements. According to Knuchel, dissolved proteins from lyophilized cells

correspond electrophoretically to the serum protein of the respective animal species. Therefore, with each cell injection, a certain amount of antigenic protein in solution is simultaneously injected [4]. The question arises how much antigen this is when calculated in protein weight units.

Dahmen observed a total of 3.25 mg of nitrogen was dissolved by 5.0 mg of N-free liquid (0.65mg Nitrogen / ml) from 100 mg of dry substance within two minutes at 37°C [5]. Instead of viewing all the nitrogen in this solution as antigen, approximately 77% (in this case 2.5 mg of nitrogen) should be considered antigen. Calculating this into protein (1 g nitrogen = 6.2 g protein), one obtains 15.5 mg of protein, which corresponds to 100 mg of lyophilized cells.

If one extracts an amount of fresh cells corresponding to 100 mg of lyophilized cells for one hour with 5 ml Ringer's solution at 37°C, the solution contains for example:

Origin	Weight	Solution Contents
Liver	514 mg	6.5 mg Nitrogen / 5 ml
Placenta	584 mg	5.65 mg Nitrogen / 5 ml

If only 77% of this total nitrogen is assumed to be antigen, then approximately 1 g of fresh tissue cell suspension contains 58 mg of soluble protein. If this is compared with ordinary tetanus prophylaxis with 3000 IU in 2.0 ml of a 1500-fold serum having a protein content of 10% or 200 mg, it becomes clear that in order to inject an equal amount of protein with fresh cells, approximately 3.45 g are required. With lyophilized cells this equals 13 vials of 100 mg each, containing a total of 200 mg of soluble protein. This comparison shows that the quantity of dissolved antigen-protein is smaller in cellular therapy than with prophylactic and therapeutic serum injections.

However, the injection of such small amounts of soluble antigen can induce the formation of antibodies by plasma cells in accordance with the laws of immunology and allergy. The formation of such antibodies can be demonstrated in the blood. If large amounts of such antibodies are produced, symptoms of generalized serum sickness and local reactions such as the Arthus phenomenon may result. As a rule, clinical symptoms by these antibodies will only be caused if the quantity of the antigen injected is relatively large, if cells of various animal species are applied simultaneously, if the interval between the repeated applications is relatively short, or most particularly, if the material is injected intravenously. All of the above factors could favour the occurrence of anaphylactic reactions and serum sickness. Despite of these facts, anaphylactic reactions due to antigen-antibody reactions with cellular therapy are extremely rare. Most likely, the antigenic potency of the foetal cells is very low that a complete immune response cannot be mounted and the structural protein of the injected cell depot is absorbed so slowly and in such small quantities that is has a desensitizing effect.

What can be done to further reduce the already low risk of allergic reaction in cellular therapy?

Firstly, one should consider the possibility of an allergy of the recipient to the non-specific tissue. A preliminary sensitivity test consisting of intra-cutaneous injection with 0.01 ml of normal serum protein from the corresponding animal species could help in the regard and is recommended by some cell therapists. A reaction occurring within minutes after injection would indicate a dangerous allergy potential for the respective patient.

There are, however, patients who tolerate the first cell therapy injection without developing any reaction, but soon afterwards, as a result of circulating antibodies, acquire a hypersensitivity which makes a second injection of cells from the same animal species after a short period of time (one week to four months) somewhat dangerous. Unfortunately, it is uncertain whether the intracutaneous skin test can be relied upon in every case in order to predict dangerous hypersensitivity. Nevertheless, the test should not be ignored if there is anything in the case history that indicates the possibility that hypersensitivity might already have been acquired.

The antigenic substance injected in cellular therapy leads to the formation of circulating antibodyglobulins, as well as to the formation of cells capable of causing a reaction upon renewed contact with the injected antigenic substances. If this reaction is artificially produced intra-cutaneously; it can be of the delayed skin reaction type. These cells may cause a reaction at the site of the cellular therapeutic injection, clinically resembling a delayed Arthus phenomenon running a relatively mild course. Thus it would not – especially with repeated cell injections – correspond to the rapidly occurring Arthus phenomenon caused by circulating anaphylactic blood antibodies; its occurrence would remain clinically unnoticed besides this phenomenon. There are apparently two types of cells which cause these reactions: in animals lymphocytic cells identified by Chase and in human beings the granulocytic leukocytes identified by Lawrence [6-7]. The latter are also capable of transferring a reaction when in extract form. This is due to specific reagents which even humans with agammaglobulinemia are capable of building [7]. The nature of this transferfactor is still being investigated so that specific reactions that are entirely dependent on cellular properties can be better understood.

Summarizing the possible side effects from organ extract- and cellular therapy we can safely state that the number of cases of immediate anaphylactic shock symptoms reported in literature is strikingly small in proportion to the wide use of this treatment method. The injection implantation of xenograft cells, non-specific to the human being, is clinically very well tolerated. The expected anaphylactic and allergic reactions fail to appear or are so few that there is no reason to give up the practice of cellular therapy [8]. More than 5 Million treatments have been recorded with only a handful of patients experiencing serious side effects. This makes this therapy actually safer than prescribing aspirin.

THE HISTORY AND THE SCIENCE OF ORGAN EXTRACT- AND CELL THERAPY

Cell therapy dates back thousands of years. Written in 1600 B.C., the Eber Papyrus of medicine recommended injection of animal organs to improve human vitality. In the Middle Ages, Parcelsus observed, for the first time, that the organisational unit of all life; the cell, was the element in ‘like heals like’.

In the late 19th century, French Nobel laureate Dr. Alexis Carrel discovered the potentially immortal nature of cells by keeping alive fragments of a chicken heart 25 years after the fowl had died. This accomplishment was performed by combining cellular material from different hearts into one cell culture.

At the end of the 19th century, Paris physiologist C.E. Brown-Sequard recognised the potency of cellular therapy by injecting himself with an extract made from the testicles of a young bull. His virility was subjectively increased due to the testosterone in the extract.

In the 1920’s, ophthalmologist Vladimir Filatov initiated the application of foetal cellular and aloe plant extract therapies for non-specific rejuvenation of chronically ill patients. His earliest claimed successes were in reversing retinitis pigmentosa and involutional retinal macular degeneration.

In the 1930’s, surgeon Niehans became increasingly interested in endocrinology while serving as head of staff at one of the renowned hospitals in Switzerland. He studied the work of colleagues who were experimenting with the implantation of animal glands into patients whose organs were malfunctioning. One of Niehans’ first discoveries was that cells derived from the organs of foetal sheep could be injected into the human body without triggering the natural defence mechanism that acts to reject foreign protein.

In 1931, Swiss physician Paul Niehans was summoned to an emergency operation where he was requested to perform a transplant for an elderly woman whose parathyroid glands were removed during a thyroid surgery by accident. The patient was in critical condition and in a race against time, Niehans sought instead to inject the woman with a steer's parathyroid cells suspended in a saline solution, crudely prepared at the scene. The woman's condition quickly stabilized and continued to improve as she went on to live another 30 years.

In the forty years following his first successful experiments, Niehans applied his discoveries in organ extract- and cellular therapy over 50,000 times. Among Niehans' patients were celebrities like Charles Chaplin, Robert Cummings, Joan Crawford, Charles de Gaulle, Dwight and Marnie Eisenhower, Winston Churchill, Charles Boyer, Bernard Baruch, the Duke and Duchess of Windsor, Joseph Kennedy, and many others.

In 1953, Niehans was called to the bedside of ailing Pope Pius XII. In gratitude for successful results of his own cell therapy, Pope Pius XII invited Niehans to become a member of the Papal Academy of Sciences. Due to this success, physicians slowly began to accept Niehans' work with cell therapy. Organ extract- and cellular therapy was en route to becoming an accepted regenerative technique in Europe.

Niehans continued his research and work into the 1960's, publishing extensively, only interrupted by World War II. His major opus on theory and practice of cellular therapy was published in German in 1954. Niehans later collaborated with Bauer of Clarens Clinic in Switzerland in studying the therapeutic effects of fresh and preserved cells. Niehans also conducted research into the cancer resisting properties of foetal mesenchyme cells within a well-regulated connective tissue matrix. He later developed the freeze-drying process of fresh cells termed lyophilization. He used cells from the frontal brain to treat mongolism. He used skin and eye cellular extracts to treat albinism, injected liver cells to treat cirrhosis, and utilised testicle cells to treat impotence. Swiss publishing company Thoune released the English version and update of Niehans' original work which also included papers by researchers from Germany, Austria, Greece and Spain.

Over one thousand scientific studies have demonstrated the effectiveness of cell therapy. By radioactively marking tissue extract, researchers have been able to ascertain what exactly happens with the organ extract and cellular material after it is injected. Within forty eight hours of administration, 90% of cell extract of the liver, for example, is attracted to its respective organ in the recipient, making its way to this destination. Various research studies conducted by Kment and Schmid conducted at the University of Vienna, the University of Heidelberg and other leading European universities have demonstrated this by means of radioactive marking of undifferentiated embryonic sheep cells. These always found their way to their respective human organs to catalyse regeneration and repair [1, 9].

Dr. Paul Weiss of the Rockefeller Institute in New York City conducted research in an attempt to explain the regenerative effect of exogenous cells. He concluded that cells have the inherent ability to 'auto organise'. This mechanism of cellular homing and recognition relates to the unique vibration frequencies emitted by DNA coils and membrane fibre optics of all cells. In the 1920's, Tesla and Lakhovsky described that 'like seeks and congregates with like' due to vibration signals, not chemical or mechanical binding sites.

A scholar and President of the International Society of Cellular Therapy Research, Kment experimented with old rats and their ability to learn, perceive, and heal after tissue laceration. Kment concluded that older animals injected with cells showed improved mental acuity and a faster ability to heal. Kment's extensive work in Austria in the 1970's indicated that cellular therapy improved cognitive abilities, connective tissue elasticity and tissue respiration. Furthermore, ageing animals injected with foetal cells from other species 'drew more significantly closer to the state of the young control animals' [9].

By the late 1970's, a large number of experiments had been conducted on cell therapy yielding empirical results. A wide range of diagnoses, including degenerative diseases and genetic malfunction in infancy were found to improve with cellular therapy. Despite positive results in these areas, researchers were unable to explain exactly how cellular transplants of foetal glands and other tissues functioned. Genetic information of foreign cells is transferred to somatic cells, yet it is not incorporated into the genome for transfer to offspring.

While organ extract- and cell therapy specialists were working in Europe along the lines suggested by the late Paul Niehans of Switzerland, the U.S. allopathic community in the 1980's had begun to move into the arena through various alternate routes. Many of these physicians and scientists were seemingly oblivious to the fact that they were simply corroborating the theories and postulates of the 'unorthodox' European cellular therapy and solid-state biophysics pioneers of decades earlier.

The 1980's was a decade in which allopathic medicine began to legitimise cell therapy (to find some way to fit it into the allopathic paradigm and alter semantics while doing so) began with Dr. Michael Osband whose work was published in the New England Journal of Medicine in 1981. He showed that 10 of 17 children treated for the immunosuppressive condition called Histiocytosis X underwent complete remission after being treated with daily intramuscular injections of thymus extract from five day old calves [10]. This was the first reported use of a non-human organ extract- and cell therapy in the U.S.

In 1983, the American Paralysis Association convention was told that cells taken from human aborted foeti and injected into animals had provided evidence of being useful in repair of spinal cord accidents and degenerative diseases.

In January 1988, The *Los Angeles Times* reported on the work of Dr. Kevin Lafferty from the University of Colorado Medical Centre, who saw 'good results' in 6 of 17 diabetic patients treated with implanted cells from foetal pancreases [11]. The *Times* also reported that about 200 patients worldwide had received foetal liver cells, primarily to restore bone marrow loss as a result of cancer therapy.

Allopathic medicine regards Parkinson's disease as an incurable condition. In the late 1980's, a wheelchair bound victim of Parkinson's was the first American to undergo what the American medical establishment decided to call 'human foetal cell transplantation therapy'. This new designation was hoped to be distanced from the line of medical research of cellular therapy; the European animal embryo based treatments the allopathic industrial cartel in the United States had long written off as medium tech quackery. Ongoing orthodox research into the Parkinson's case had shown that since the 1989 injection of brain tissue from an aborted human foetus, the patient's Parkinson's symptoms had lessened by 50%. Unfortunately for the pharmaceutical industry, nothing about cell therapy is patentable, or highly profitable, nor does it consign a patient to endless expensive courses of therapy.

Dr. Mitchell Golbus of the University of California in San Francisco had unsuccessfully attempted to transplant adult tissue into human foeti to cure genetic conditions [12]. The fact that an American university would try to inject adult cells indicated just how provincial the U.S. healthcare system could be. For classical European organ extract- and cell therapy practitioners who utilise animal embryonic, foetal or placental tissue injected subcutaneously, the interest of the Americans into human cell therapy borders on the laughable. Why would they utilise adult human tissue? And why would they directly graft into the brain or any other organ, which is risky and potentially dangerous?

In October 1991 and as a follow up in February 1992, American researchers were also reporting early success with foetus to foetus cell therapy with a severe genetic abnormality known as Hurler's Syndrome. Buckley et al. reported that transplanted human foetal tissue had 'taken hold' in an infant born a year before, with the child's blood making cells of the transplanted tissue [13]. The

developing foetus had been injected with foetal cells from an aborted human foetus in a controversial application of therapy. The parents of this child had also lost two children prior to this to mucopolysaccharidosis syndrome, which causes crippling skeletal problems, blindness and severe mental retardation. The case brought to the public eye the controversy of using human foetal tissue in experimental therapy.

Over the last two decades, xenotransplantation is progressing into areas of treatment previously not treated by transplantation, such as degenerative brain conditions and diabetes. Phase I clinical trails have been conducted with foetal islet of Langerhans cells for treating diabetes and foetal brain cells for Parkinson's disease and Huntington's chorea [14-20]. It was found that foetal xenotransplants of brain cells would continue to secrete neurotrophic factors and trophic substances in the recipient patient thereby improving their own survival and promoting regeneration of nearby damaged tissue. Release of nerve growth factors by foetal neuroblasts can assist in neural tissue regeneration and healing of the degenerative brain processes [17-20].

Xenotransplantation of foetal animal cells is also a novel experimental strategy to treat heart disease, such as myocardial infarction and heart failure. Its beneficial effects may include active contribution of transplanted cells to contractile function, passive improvement of the mechanics of the heart, induction of neoangiogenesis (angiogenic factors secreted from foetal tissue can promote blood vessel formation) or other indirect influences on the biology of the heart. Several cell types have been used for cardiac cell transplantation including cardiac cells from foetal or newborn animals and cardiac muscle cell lines, skeletal myoblasts and skeletal muscle cell lines, smooth muscle cells, and a variety of stem cells, either adult or embryonic. With many of these cells, encouraging results in experimental ischemic and non-ischemic heart disease have been obtained including successful cell survival after transplantation, integration into the host myocardium, and improvement of the function of diseased hearts. It has to be noted, however, that there are many issues that need to be addressed before this strategy will add to the therapeutic options for patients with heart disease [21].

The first trial of restoring brain functions with cell grafting was performed in 1979 using a rat model of Parkinson's disease. Foetal nigral tissue was demonstrated to survive grafted tissue and to repair motor dysfunction in the model rat. The encouraging results indicate that cell transplantation may be useful to restore brain functions in neurodegenerative disorders in which a certain type of neuronal populations is specifically damaged. The observation that xenogeneic foetal neurons are able to survive and function when transplanted into the adult brain fostered the development of cellular therapy as a promising approach to achieve neuronal replacement for treatment of diseases of the adult central nervous system. This approach has been demonstrated to be efficacious in patients with Parkinson's disease after transplantation of human foetal neurons. The use of human foetal tissue is limited by ethical, infectious, regulatory, and practical concerns. Other mammalian foetal neural tissues have served as an alternative cell source. Cell xenotransplantation has been used as an experimental therapy for Parkinson's disease and other movement disorders. Several open-label research trials have shown clinically meaningful improvement in Parkinson’s Disease signs and symptoms after striatal transplantation of allogeneic foetal ventral mesencephalic tissue [22-23].

Summarising the scientific advancements in organ extract- and cellular therapy, it can be stated that the ability to cross species lines has dramatically expanded the number of patients and the scope of human diseases that can be treated successfully with xenotransplantation. In addition to whole organs, the transplantation of cells and tissues with specific differentiated functions represents an important conceptual and medical advance. In the USA alone, over 15 million patients suffer from diabetes, over 7 million patients suffer from neurodegenerative diseases, and millions more suffer from liver failure, AIDS, haemophilia and other disorders caused by tissue loss or dysfunction. Clinical trials using animal cells to treat many of these diseases are already under way, and it seems likely that this list will continue to grow as researchers identify new bioactive

molecules and expand their understanding of the role different cells play in the human disease process [24-27].

Administered via intramuscular injection, cell therapy proves to be advantageous over conventional procedures of surgical transplantation for the following reasons:

- Implantation of foetal xenogeneic cells by injection brings about rapid dispersion and only mild degrees of rejection and antibody formation by the host
- Xenogeneic cells are less likely injured due to lack of blood supply as commonly seen in whole organ transplants
- Injected in the form of a suspension, xenogeneic cells are rapidly incorporated into the body helping to optimise its metabolic processes
- Organs that are impossible or difficult to transplant (for example brain or some of the endocrine glands) can be implanted in the form of xenogeneic cells very easily

In addition to general body revitalization, health maintenance, and life extension, cell therapy will effectively stimulate the cells of the specific organ systems concerned, and thereby help in the treatment of a multitude of human diseases and ailments.

Other indications

To support treatment of chronic fatigue syndrome, immune deficiency, infertility, frozen shoulder, shoulder stiffness, migraine, joint pain, depression, melasma, ageing skin, acne, hay fever, premenstrual syndrome, uterine fibrosis, endometriosis, insomnia, liver dysfunction, fungal infection, digestive problems, nerve related discomfort, extreme coldness, anaemia, low libido, constipation, gastritis, intestinal intoxication and hormonal imbalances

Dosage Instruction and Type of Application

Organ extract and cellular therapy are non-standardized treatments designed for each individual patient. Recommendations can therefore only be generalised to a limited extent. Selection of tissue type and dosage for treatment depend on the goal of the treatment (general rejuvenation or treatment of a specific degenerative disease), the duration and severity of the aliment or disease, the age of the patient, the observed healing tendency and other factors.

Most often treatment is administered for general rejuvenation. In such cases a set of organ extract or cell therapy preparations of the following tissue types are often combined in one treatment session: Thymus, spleen, liver, kidney, heart, adrenal gland, pituitary gland, ovary or testis.

Before any organ- or cell therapy, it is of utmost importance to gain as much information as possible in regards to the patient's medical history. Whenever possible, the root cause of a physical ailment or symptoms should be determined in order to adopt a specific and individualised cell therapy. The absence of such information would direct therapeutic efforts toward alleviation of symptoms only, and therefore, the desired therapeutic goal may not be attained consistently and with lasting effect.

The injection is to be administered intramuscularly, into the upper outside quadrant of the major gluteus muscle. Opened ampoules are to be injected immediately, as sterility can be compromised. Once thawed, any unused vial should not be re-frozen, but rather discarded.

Safety and Effectiveness of Cell Therapy

Since organ extract preparations have very low antigenicity; they are not readily recognized as foreign by the recipient's immune system, and therefore, they will not cause significant clinical side effects. Donor animals and all preparations are thoroughly tested to rule out transmission of

zoonoses and infections; there are no known cases with the use of organ extract preparations. Acute allergic reactions with fresh frozen or freeze-dried organ extracts and cell preparations are exceedingly rare. If such reactions should occur, they will be easily manageable with antihistamines, steroids, and intravenous volume fluid expansion. Organ extract and cell preparations of foetal xenogeneic cells will imprint their vigour upon old, tired, and degenerating cells, stimulating them to function with renewed efficiency, thereby improving organ function and vigour and vitality of the entire organism.

REFERENCES AND ADDITIONAL INFORMATION

[1] Schmid F (1983) *Cell Therapy: A new dimension of medicine.* Switzerland: Ott Publishers Thoune
[2] *Küttner;* Zbl Chir 12, **39**, 390-397 *(1912)*
[3] *Bennhold H;* Deutsche Medizinische Wochenschrift 79, **17**, 704-711 *(1954)*
[4] *Knuchel F et al.;* Medizinische, **16**, 578-593 *(1955)*
[5] *Dahmen H;* Deutsch. Schlacht- und Viehhof-Ztg., **1**, *(1956)*
[6] *Chase MW;* Proc Am Soc Exp Biol Med, **134**, 156-169 *(1945)*
[7] *Lawrence H et al.;* Journal of Immunology, **68**, 159 *(1952)*
[8] *Carstens CE;* Hippokrates, Stuttgart, **2**, 40 *(1957)*
[9] *Kment A et al.;* Therapiewoche 11, **9**, 489 *(1961)*
[10]*Osband ME et al.;* New England Journal of Medicine, **304**, 146-153 *(1981)*
[11](http://www.uchsc.edu/misc/diabetes/oxessa.html)
[12]*Golbus M et al.;* New England Journal of Medicine, **307**, 1651-1653 *(1982)*
[13]*Buckley RH et al.;* New England Journal of Medicine, **341**, 291-292 *(1999)* [14] *Heneine W et al.;* Lancet, **352**, 695-699 *(1998)*
[15]*Groth CG et al.;* Lancet, **344**, 1402-1404 *(1994)*
[16]*Lanza RP et al.;* Mol Med Today 4, **1**, 39-45 *(1998)* [17] *Deacon T et al;* Natur Med, **3**, 350-353 *(1997)*
[18]*Fink JS et al.;* Cell Transplant 9, **2**, 273-278 *(2000)*
[19]*Brevig T et al.;* Transplantation 72, **2**, 190-196 *(2001)*
[20]*Kakishita K et al.;* Nippon Rinsho 61, 3, 457-462 *(2003)*
[21]*Muller-Ehmsen J et al.;* Congest Heart Fail 8, **4**, 220-227 *(2002)*
[22]*Lindvall O et al.;* Clin Chem Lab Med 39, **4**, 356-361 *(2001)*
[23]*Subramanian T;* Semin Neurol 21, **1**, 103-115 *(2001)*
[24]*Metzger R et al.;* Pediatr Surg Int 18, **7**, 640-647 *(2002)*
[25]*Till H et al.;* Eur J Pediatr Surg 8, **4**, 240-243 *(1998)*
[26]*Danilovs et al.;* Tissue Antigens 21, **4**, 296-308 *(1983)*
[27]*Edge AS et al.;* Cell Transplant 7, **6**, 525-539 *(1998)*
[28]Schmid F & Stein J (Eds.). (1967) *Cell Research and Cellular Therapy.* Switzerland: Ott Publishers Thoune
[29]Stephan, P (1971) *The Secret of Eternal Youth: Rejuvenation Through Dr. Niehans' Cell Therapy:* New York: Arco Publishing Company, Inc
[30]Cellular Therapy (http://www.karlloren.com/biopsy/p68.htm)
[31]*Chapman LE et al.;* New England Journal of Medicine, **333**, 1498-1501 *(1995)*
[32](http://www.cells.de/cellseng/1medienarchiv/Zellstruktur/index.jsp#Cell%20structure)
[33]*Weiss RA*; British Medical Journal, **317**, 931-932*(1998)*

- **The International Federation for Cell Therapy Professionals Europe & U.S.A**