Review Article

Regenerative medical therapy from the viewpoint of biomaterials

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Regenerative medical therapy has been expected as the third therapy following the reconstructive surgery and organ transplantation to compensate for the technological and methodological disadvantages. There are three therapeutic objectives. The first is to create a new therapeutic strategy. The second is to enlarge the clinical applications of the conventional therapy. The third is to suppress the progressive deterioration of diseases. For every objective of regenerative therapy, basic idea is to induce and accelerate the regeneration and repairing of defective and damaged tissues based on the natural healing potential of patients themselves. For the successful tissue regeneration and repairing, it is indispensable to provide cells with a local environment which enables them to efficiently proliferate and differentiate, resulting in cell-induced tissue regeneration. Biomaterials play an important role in the creation of this regeneration environment in terms of the cells scaffold of artificial extracellular matrix and the delivery technology of bio-signaling molecules to enhance the cells potential for tissue regeneration. In addition, biomaterials give cells culture conditions suitable for their *in vitro* proliferation and differentiation to obtain a large number of cells with a high quality for cell transplantation therapy. Cells can be genetically engineered to activate the biological functions by using the non-viral carrier of biomaterials. Several examples of *in vivo* tissue regeneration and basic researches for stem cells with the technology of cell scaffold and drug delivery system (DDS) of growth factors and genes are introduced to emphasize significance of biomaterials in the full realization of regenerative medical therapy.

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Significance of Biomaterials in Regenerative Medical Therapy

Advanced surgical therapies currently available consist of reconstruction surgery and organ transplantation. Although there

is no doubt that these therapies have saved and improved countless lives, they have several therapeutic and methodological limitations. In the case of reconstruction surgery, biomedical devices cannot completely substitute the biological functions even for a

single tissue or organ, and consequently cannot prevent the progressive deterioration of tissue and organ injured or damaged. One of the biggest issues for organ transplantation is the shortage of donor tissues or organs. Additionally, the continuous and permanent use of immunosuppressive agents to prevent immunological rejection responses often causes side effects, such as the high possibility of bacterial infection, carcinogenesis, and virus infection. To resolve these issues in the two advanced therapies, a new therapeutic solution that is clinically mild to patients, is required.

In this clinical situation, a new therapeutic trial, in which disease healing can be achieved based on the natural healing potential of patients, has been explored. This trial is termed regenerative medical therapy where the regeneration of tissues and organs is naturally induced to therapeutically treat diseases by artificially promoting the proliferation and differentiation of cells. To realize this cell-induced regeneration therapy, there are two approaches. One is cell transplantation where cells with a high potential of proliferation and differentiation are transplanted to induce tissue regeneration based on their potentials. The other is the therapeutic approach with biomaterials. In the latter approach, an *in vivo* local environment which enables cells to promote their proliferation and differentiation is created by making use of biomaterials. If the environment manipulates efficiently cells inherently present in the body to enhance the biological functions, cell-induced natural healing of tissues and organs will be achieved without cell transplantation. This approach is called tissue engineering. This basic concept of biomaterials-based tissue engineering was originally introduced by R. Langer and J. Vacanti¹⁾. Several technologies and methodologies with biomaterials have been demonstrated to create the cell environment for tissue regeneration²⁻⁹⁾.

This new regenerative therapy cannot always therapeutically substitute the reconstructive surgery and organ transplantation clinically available, and has advantages and disadvantages in therapy. However, it is clinically expected as the third therapeutic choice. If regenerative medical therapy is realized, it will enable us to create new therapeutic strategies as well as increase the therapeutic choice of clinicians, which consequently brings about large therapeutic benefits for patients who have not been able to receive clinically effective therapies. There are three objectives of regenerative therapy. The first objective is to create a new therapeutic strategy which is well known generally. The second is to enlarge the clinical application of therapies conventionally available. The conventional therapy is not always effective in treating patients who are aged or suffer from other

diseases, such as diabetes and hyperlipemia, nor clinically applicable because the number of key cells is small and their potential for proliferation and differentiation is low. In this case, it is practically possible that combination with the technology and methodology to promote cell-induced self-healing potentials improves the therapeutic efficacy. The third is to suppress the progressive deterioration of diseases. The deterioration and progress of disease conditions are artificially suppressed by promoting the cell potentials to induce tissue regeneration. For example, in chronic fibrosis diseases, the fibrous tissue of excessive collagen fibers and fibroblasts causes the physical impairment of natural healing process at the disease site. If the fibrosis can be loosened and digested, and additionally the natural healing potential of the surrounding healthy tissue can be augmented, it is highly expected that the disease deterioration is physiologically suppressed. One of the therapeutic advantages is the ability to accelerate the natural healing of body injury through promoted angiogenesis or the infiltration and recruitment of key cells at the injured site. This will enable patients to shorten the healing period and suppress the deterioration process of disease even under inflammation and infection conditions. A disadvantage of this therapy is that, generally, at least a few days are required to induce and activate cell-based tissue regeneration. Consequently, it cannot be expected that regenerative therapy alone will achieve the rapid healing of wounds or diseases. Depending on the clinical situation, it is necessary for better medical treatment to combine the conventional therapies with the regenerative medical strategy.

Fundamental Technology and Methodology for Biomaterials-Based Regenerative Therapy

Fundamentally, the body tissue comprises of three factors; cells, the extracellular matrix (ECM) for cell proliferation and differentiation (natural scaffold), and bio-signaling molecules. There are some cases where tissue regeneration is achieved by the single or combinational use of the factors in an appropriate way. However, since better tissue regeneration cannot always be expected only by their simple combination, it is necessary to biomedically contrive the way how to combine. To this end, proper and positive assistance of biomaterials will be practically promising. There are five fundamental technologies or methodologies that are necessary for biomaterials-based regenerative medical therapy.

The first key technology is the preparation of cells scaffold for their proliferation and differentiation for *in vivo* tissue regeneration (Fig.1A). ECM is not only a physical support for the

cells, but also provides a natural environment for cell proliferation and differentiation or morphogenesis, which contributes to tissue regeneration and organogenesis. Generally, it is difficult to naturally regenerate and repair a large-size tissue defect only by supplying cells to the defective site, because cells and the ECM are both lost. Therefore, to induce tissue regeneration at the defective site, one way is to artificially build a local environment for cells which is a three-dimensional scaffold of artificial ECM to initially assist their attachment and the subsequent proliferation and differentiation, inducing cell-based tissue regeneration. It is expected that cells residing around the scaffold implanted infiltrate into the scaffold and consequently proliferate and differentiate therein if the artificial ECM is biologically compatible. Biomaterials play an important role in the preparation of cell scaffold. The scaffold should be porous and biodegradable. The pore structure is necessary for the infiltration of cells into the scaffold and the supply of oxygen and nutrients to cells proliferated therein. Once the cell-induced tissue regeneration is naturally initiated, cells eventually produce the ECM of natural scaffold. Thus, a long-term remaining of cell scaffold often causes physical hindrance against the natural process of tissue regeneration. It is key for successful tissue regeneration to control the time profile of scaffold biodegradation at the defect as well as the three-dimensional structure. The mechanical property of biomaterials scaffold is also important. If the mechanical strength is low, the scaffold readily deforms in the body. As the result, the regeneration of bulky tissue cannot be always expected. An appropriate mechanical design is highly required.

The second key technology is to protect cells transplanted from immunological attack and fibroblasts infiltration naturally occurring and provide the space for tissue regeneration (Fig. 1B). When a body defect is generated, the defect space is generally occupied rapidly with the fibrous tissue produced by fibroblasts, which are ubiquitously present in the body and can proliferate rapidly. This is one of the typical wound healing processes to temporarily fill and emergently repair the body defect. However, once this in-growth of fibrous tissue into the space to be regenerated takes place, the regeneration and repairing of a target tissue at the space can no longer be expected. To prevent the tissue in-growth, a barrier membrane to provide a space for tissue regeneration is required. In addition to the membrane-induced tissue regeneration, there is another approach where cells with an activity to secrete physiologically important substances, such as pancreatic islets and hepatocytes, are used to substitute the biological functions of damaged organs. For this cell-based organ substitution, functional cells are encapsulated in a hydrogel mem-

brane to prevent them from immunological attack by antibodies and host cells.

When the tissue around a defect does not have the inherent potential to regenerate, tissue regeneration cannot always be expected only if the scaffold or the space providing membrane is supplied. The cell scaffold and membrane of biomaterials should be used in combination with cells or/and signaling molecules (e.g. growth factors, cytokines, and chemokines) that have a potential to accelerate cell-induced tissue regeneration. Cells with a high potential for proliferation and differentiation are being prepared and applied to a tissue defect to induce tissue regeneration thereat. Although there are some cases where a growth factor is required to promote tissue regeneration, the direct injection of growth factor in solution into the site to be regenerated is not generally effective. This is because the growth factor rapidly diffuses from the injected site and is enzymatically digested or deactivated. To enable the growth factor to efficiently exert its biological function, a new technology is required. This comes in the form of the third key technology of tissue engineering — drug delivery system (DDS) (Fig. 1C). Although every DDS technology is available for tissue engineering, the release technology of growth factors and genes has been mainly applied to induce tissue regeneration. For example, the controlled release of growth factor at the site of action over an extended period of time is achieved by incorporating the factor into an appropriate carrier of biomaterial. It is also possible that when incorporated in the release carrier, the growth factor is protected against proteolysis to prolong the activity retention *in vivo*. The release carrier should be degraded in the body, because it becomes useless after the release function finishes. In addition to the controlled release, the improvement of signaling molecules stabilization, the prolongation of the *in vivo* half-life, and the targeting to the site of action will enhance the factor-induced tissue regeneration.

Over time, DDS has been investigated and developed as only a technology or methodology to enhance the *in vivo* efficacy of therapeutic drugs. Based on the fixed idea and historical background, it has been thought that DDS cannot scientifically and technologically be applied to regenerative medical therapy. Little has been investigated on DDS-based tissue regeneration. Considering that drugs applicable for regenerative therapy include proteins and genes effective in promoting the proliferation and differentiation of cells to induce tissue and organ regeneration, generally, they are unstable *in vivo*. Therefore, upon *in vivo* administering the signaling molecules, it is necessary for enhanced *in vivo* biological activity to make use of DDS technology and methodology. Many types of biomaterials have been used for

Fig. 1 Important role of biomaterials and key technologies to realize regenerative medical therapy

the DDS applications. Cells with high proliferation and differentiation potentials, so-called stem cells, are important to induce tissue regeneration. For example, in addition to hematopoietic stem cells, mesenchymal stem cells (MSC) are present in the adult bone marrow. It has been elucidated that the MSC of adult stem cells have an inherent potential to differentiate into osteogenic, chondrogenic, adipogenic, and myocardial cell lineages. Presently, human MSC are isolated and are commercially available¹⁰. Several researches of regenerative medical therapy with MSC have been reported to demonstrate their therapeutic feasibility in tissue regeneration^{8,11-14} while the clinical experiments have been begun^{14,15}. If it is possible to clinically use a differentiated type of a patient's own MSC, immunological rejection will be no longer be a question. In addition, neural stem cells¹⁶⁻¹⁸ and stem cells isolatable from fat tissue^{19,20} have been extensively being investigated. However, one of the problems is the shortage of cells clinically available. Therefore, it is necessary to develop a technology or methodology for the preparation of a large number of stem cells with a high quality. The fourth technology is for the efficient isolation and proliferation of cells (Fig.1D), which are achieved by providing a cell culture substrate of biomaterials as the artificial ECM. The cell scaffold for *in vivo* tissue regeneration mentioned previously can be also used

as the culture substrate. The three-dimensional substrate can be designed and prepared from biomaterials of cytocompatibility to prepare cells for transplantation therapy. From the viewpoint of the nutrients and oxygen supply to cells, the research and development of cell culture methods and bioreactors are required. Cell sheet engineering is one of the promising cell culture technologies to realize tissue regenerative therapy²¹. The fifth technology is for the genetic engineering of cells (Fig.1E). There are some cases where cells transplanted do not function well to induce cell-based tissue regeneration. As one trial to tackle this issue, cells are genetically engineered with biomaterials to activate the biological functions. It is necessary for genetic engineering of cells to develop the carrier of gene transfection and the cell culture system for efficient gene expression. Non-viral gene carrier of biomaterials is needed to develop from the clinical viewpoint of cell therapy because it is practically tough to use viral vectors of gene transfection clinically. This technology of gene transfection is also applicable for the basic research of stem cell biology and medicine which gives important knowledge and results for cell therapy. Non-viral carriers for gene transfection and cell culture technologies with biomaterials are needed to induce iPS cells at higher efficiency for their clinical applications.

This combination of cell scaffold, space providing, and DDS

technologies is practically promising to create an environment which enables cells to promote the proliferation and differentiation for tissue regeneration. The culturing and genetic engineering of cells are both key technologies to prepare cells clinically available for cell therapy. Every biomaterials-based technology is important not only to develop the basic research of stem cells biology and medicine, but also to realize the cell-based tissue regenerative therapy.

Frontier of Tissue Regeneration Based on Biomaterials Technologies

Tissue engineering for clinical regenerative medicine can be classified as either *in vitro* or *in vivo* depending on the site where tissue regeneration or organ substitution is performed. The *in vitro* tissue engineering involves tissue reconstruction by cell culture methods and organ substitution with functional cells — termed bioartificial hybrid organ. If a tissue can be reconstructed *in vitro* in factories or laboratories on a large scale, it can be supplied to patients when required. However, it is difficult to reproduce the *in vivo* event completely *in vitro* only by using the basic knowledge of biology and medicine or cell culture technologies currently available. At present, it is practically difficult

to realize *in vitro* tissue engineering because artificial arrangement of a biological environment for cell-based tissue reconstruction is impossible. In addition, limited oxygen nutrients supply to cells cultured three-dimensionally should be improved. Human dermal fibroblasts cultured on a biodegradable polymer sheet are applied clinically for the treatment of skin burn. A cell sheet is prepared from the corneal epithelial cells with a temperature-responsive polymer dish and clinically applied for the regenerative therapy of cornea²²⁾. Another application of *in vitro* tissue engineering is the functional substitution of liver and pancreas by combination of allo- or xeno-geneic cells with an immuno-isolation membrane of biomaterials although it is still an experimental trial.

Biomaterials have been investigated to design and create the biological environment which can regulate the proliferation and differentiation of cells. It has been demonstrated that the direction of cells differentiation can be modified by the softness of cell substrates²³⁾ and the surface immobilization of bioadhesion molecules²⁴⁾.

Distinct from the *in vitro* tissue engineering, the *in vivo* tissue engineering is advantageous from the viewpoint of the environment to induce tissue regeneration. It is likely that most of bio-

Fig. 2 Concrete examples of tissue regeneration therapy based on the release technology of growth factors with biodegradable gelatin hydrogels: Connective tissue growth factor (CTGF) and vascular endothelial growth factor (VEGF)

* $p < 0.05$, significant against the value of 20 μg of VEGF solution. ** $p < 0.05$, significant against the value of VEGF-free gelatin hydrogels.

logical components necessary for tissue regeneration, such as growth factors and cytokines for tissue regeneration, are naturally supplied in the body. Based on this advantage, almost all the approaches of tissue engineering have been performed *in vivo* with or without biodegradable cell scaffolds. There are several examples where *in vivo* tissue regeneration is achieved by use of the scaffold with or without cells²⁵). As described previously, if patients are young and healthy, and the tissue to be repaired has a high potential to induce regeneration, active and immature cells infiltrate into the biomaterial scaffold implanted from the surrounding healthy tissue, resulting in the formation of new tissue. However, additional means are required if patients are aged and/or suffer from other diseases, such as diabetes and hyperlipemia, or if the regeneration potential of tissue is low as a result of, for example, a low concentration of cells and growth factors. The simplest method is to supply a growth factor to the site of regeneration for cell differentiation and proliferation in a controllable fashion. Recent researches of tissue regeneration using a growth factor of *in vivo* instability indicate that the DDS technology is necessary to enable the growth factor to exert its biological activity for *in vivo* tissue regeneration. However, DDS research of growth factor for tissue regeneration has not been studied extensively. This is mainly because growth factor is costly and has not been able to use at a large amount.

One of the largest problems in the release technology of growth factor protein is the loss of biological activity of protein released from a protein-carrier formulation. It has been demonstrated that this activity loss mainly results from denaturation and deactivation of protein during the preparation process of the formulation. Therefore, a method to prepare the formulation of protein release with biomaterials should be exploited to minimize protein denaturation. From this viewpoint, polymer hydrogel may be a preferable candidate as a protein release carrier because of its biosafety and its high inertness toward protein drugs. However, it will be impossible to achieve the controlled release of protein

over a long period of time from hydrogels since the protein release is generally diffusion-controlled through the water pathway present in hydrogels. Thus, a possible approach is to allow the growth factor to immobilize in a biodegradable hydrogel. The immobilized factor is not released by simple diffusion, but only by the solubilization of factor in water as a result of hydrogel biodegradation. In such a release system, the time profile of growth factor release is governed and can be changed only by that of *in vivo* hydrogel degradation. The final goal of regenerative therapy is to treat patients, but not to develop the basic science of biology and medicine. Based on this, gelatin was selected to prepare the biodegradable hydrogel of protein release carrier, because it has been clinically used in medical and pharmaceutical applications and proven to be biocompatible. By the gelatin hydrogel, the controlled release of bioactive growth factors over the time range of 5 days to 3 months could be achieved. We have succeeded in the controlled release of various growth factors to induce the regeneration of various tissues (Fig.2). Basic fibroblast growth factor (bFGF) is reported to have a variety of biological activities²⁶) and be effective in enhancing wound healing through induction of angiogenesis and inducing the regeneration of bone, cartilage, skin, nerve, periodontal, and fat tissues or the elongation of hair²⁷). When human recombinant bFGF (Fibrast® spray, Kaken Pharmaceutical Co., Tokyo; <http://www.kaken.co.jp>) was incorporated into a gelatin hydrogel and subcutaneously implanted into the mouse back, significant angiogenic effect was observed around the implanted site, in marked contrast to the injection of bFGF solution even at higher doses²⁸).

There are two important objectives of angiogenesis in tissue engineering; the therapy of ischemic disease and 'in advance angiogenesis' for cell transplantation. As the first example, when injected into the ischemic site of myocardial infarction²⁹) or leg ischemia³⁰), gelatin microspheres incorporating bFGF induced angiogenesis to a significantly greater extent than the bFGF solution. This angiogenic therapy for leg ischemia has been clini-

Table 1 On-going clinical experiments of regenerative therapy with the release technology of growth factors

Disease or operation	Growth factor	Therapeutic effect	Number of facilities
Vascular graft surgery for heart	bFGF	Angiogenesis	1
Arteriosclerosis obliterans (ASO) and Buerger diseases	bFGF	Angiogenesis	4
Diabetes skin ulcer	bFGF	Angiogenesis, Dermis regeneration	3
Periodonal disease	bFGF	Regeneration of alveolar bone	3
Hardness of hearing	IGF1	Suppression of nerve degeneration	2
Meniscus injury	PRP	Chondrogenesis	1
Facial plastic surgery	bFGF	Chondrogenesis, soft tissue regeneration	1
Chest plastic surgery	bFGF	Regeneration of sternum bone, angiogenesis	1
Plastic surgery of soft tissue	bFGF	Adipogenesis	1
Facial nerve paralysis	bFGF	Nerve function recovery	1

cally started to demonstrate good therapeutic results³¹). In addition, the bFGF-induced angiogenic therapy was also effective in accelerating the regeneration healing of the diabetic foot ulcer and periodontal tissue defect. Several DDS-based regenerative therapies of sternum, fat, and meniscus are being proceeded clinically to demonstrate good therapeutic efficacy (Table 1).

The sufficient supply of nutrients and oxygen to cells transplanted is indispensable for their survival and maintenance of biological functions *in vivo*. For successful cell therapy, it is beneficial to induce angiogenesis in advance or around the site where cells are transplanted, by using the bFGF release system. This technology of 'in advance angiogenesis' efficiently improved the biological functions of pancreatic islets³², hepatocytes³³, cardiomyocytes³⁴ and kidney cells³⁵, as well as the engrafting of a bioartificial dermis-epidermis skin-tissue construct³⁶.

The release system enabled transforming growth factor (TGF)- β 1, and bone morphogenetic protein-2 (BMP-2) to enhance the biological activity of bone regeneration and bone healing, as well as to synergistically promote MSC-induced bone regeneration²⁷. It is well known that insulin-like growth factor (IGF)-1 suppresses the apoptosis of nerve cells. Controlled release of IGF-1 from the gelatin hydrogel inhibited the aging of acoustic nerve, resulting in suppressed deterioration of hearing hardness³⁷. This IGF-1 treatment has been started clinically. In addition, the hydrogel system can release not only one type of growth factor, but also two or more types in different concentration and release profiles. Upon applying a hydrogel incorporating a low dose of either basic fibroblast growth factor (bFGF) or transforming growth factor β 1 (TGF- β 1) to a bone defect of rabbit skulls, no bone tissue was regenerated at the defect. However, a synergistic effect on bone regeneration was observed by the simultaneous release of two factors (Tabata et al. unpublished data). The synergistic angiogenesis of bFGF and hepatocyte growth factor (HGF) was observed³⁸. The platelet contains a cocktail of autologous growth factors. The hydrogel enabled the controlled release of the growth factor cocktail to induce the regeneration of tissues, in remarked contrast to the use of cocktail alone³⁹⁻⁴¹.

Further Applications of Biomaterials to Regenerative Therapy

In addition to growth factors, gene has been used for tissue regenerative therapy. There are two future directions in the use of gene therapy. The first direction is the conventional gene therapy where a plasmid DNA and adenovirus are injected directly. It is practically difficult to use viruses for clinical therapy although the transfection efficiency is high. On the other hand, to enhance

the *in vivo* gene transfection efficiency of plasmid DNA, assistance of DDS technologies is required. Several carriers of biomaterials have been investigated to enhance the level of gene expression. It has been found that the controlled release of plasmid DNA from a biodegradable hydrogel of cationized gelatin enhanced the level of gene expression as well as prolonged the time period of expression^{42,43}. The second direction is to genetically activate cells by gene transfection for cell therapy. There are some cases where the transplantation of stem cells does not always induce a therapeutic effect clinically acceptable. A promising and practical way to break through this problem is to genetically engineer stem cells by gene transfection for activation of their biological functions. So far, such cell genetic activation has been of great success. However, the good results cannot be clinically applied because of the virus use. Therefore, it is necessary to develop a biomaterial-based DDS technology or methodology for non-viral system with gene transfection efficiency as high as that of viral system⁴⁴. The controlled release of plasmid DNA inside cells enhanced the level of gene expression and prolonged the expression period⁴⁵. Reverse gene transfection was effective in enhancing the gene expression of stem cells to genetically engineer the biological function⁴⁶. In addition, MSC genetically engineered showed the efficacy of cell therapy superior to the original cells⁴⁷. The gene engineering technology with biomaterials is available for the basic research of stem cells biology and medicine. Small interfering RNA (siRNA) can silence the genetic activity in a RNA-sequence specific manner. The biomaterial combination of siRNA enhanced the silencing effect, resulting in genetic modification of cell functions. The controlled release of siRNA significantly enhanced the *in vivo* biological activity^{48,49}.

At present, there is no effective therapy for chronic fibrosis diseases, such as lung fibrosis, cirrhosis, dilated cardiomyopathy, and chronic nephritis. The disease site is normally occupied with fibrous tissue of excessive collagen fibers and fibroblasts. It is possible that this tissue occupation causes impairment of the natural healing process at the disease site. Therefore, if the fibrosis can be loosened and digested to disappear by the drug treatment of internal medicine, it is highly expected that the disease site will be regenerated and repaired based on the natural healing potential of the surrounding healthy tissue. This new strategy is defined as physical regenerative therapy of internal medicine (Fig.3). The physical regeneration therapy is conceptually identical with the surgical one in terms of active utilization of natural healing potential. We have demonstrated that the controlled release of a MMP-1 plasmid DNA at the medulla of

Fig. 3 Conceptual illustration of surgical and physical therapies of tissue regeneration

The basic idea of both the therapies is similar in terms of active utilization of the natural healing potential of body itself.

chronic renal sclerosis induced the histological regeneration of kidney structure, in contrast to the plasmid DNA solution⁵⁰). The intraperitoneal injection of gelatin microspheres incorporating hepatocyte growth factor (HGF) allowed to histologically cure rat liver fibrosis in remarked contrast to the injection of HGF solution⁵¹). Combination with biomaterials enabled siRNA to enhance the *in vivo* biological activity and consequently suppress tissue fibrosis^{52,53}).

Closing Remarks

The therapy of regenerative medicine — a new therapy to achieve the tissue regeneration based on the cell-induced natural healing potential of body — is the third therapy following the reconstructive surgery and organ transplantation. There are two approaches of cell transplantation and tissue engineering to clinically realize the regenerative therapy. Even if superior stem cells can be practically obtained with recently developed basic researches of biology and medicine, it is impossible to therapeutically treat patients only by transplanting the cells, unless a local environment of cells suitable to promote the proliferation and differentiation is created and provided properly. To create the environment, it is no doubt that biomaterials-based tissue engineering technology and methodology are needed. However, one of the large problems to create the regeneration environment at present is the absolute shortage of biomaterial researchers who

investigate the cell scaffold, the DDS, the protective barrier, and cell culture, aiming at tissue regeneration and the biological substitution of organ functions. Such researchers must have knowledge in medicine, dentistry, biology and pharmacology, in addition to material sciences. It is indispensable to educate the researchers of an interdisciplinary field who have an engineering background and can also understand basic biology, medicine and clinical medicine. The latest knowledge and information of stem cells and the related biology and medicine must be combined and integrated with biomaterials science. Toward the design and creation of regeneration environment, substantial collaborative research between material, pharmaceutical, biological, and clinical scientists is needed.

Biomaterial-based regenerative therapy can be not only applied surgically at the tissue defect, but also physically done for a new therapy of chronic fibrosis diseases by the drug treatment of internal medicine. Tissue engineering is still in its infancy, although some growth factor-induced tissue regeneration therapies have been clinically started. It is no doubt that the increasing significance of biomaterials in cell scaffold and DDS in future will help progress in the clinical regeneration therapy as well as the basic research of stem cells biology and medicine necessary for future regenerative therapy. We will be happy if this review stimulates readers' interest in the idea and research field of tissue engineering to assist understanding how important biomaterials

are to realize tissue engineering-based regenerative therapy.

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