

Poster Presentation

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Functional improvement of hemostatic scaffold by addition of recombinant batroxobin

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ncontrolled hemorrhaging is the leading preventable cause of infectious complications in accidents, surgical procedures and battle fields. It indicates that excessive blood bleeding could ultimately be correlated with life. Therefore, topical hemostats and sealants are required that they can assist the effective treating arrest bleeding and stabilize the casualty. In many types of hemostatic agents, collagen and chitosan play an important role for clot formation and platelet activation and aggregation in blood coagulation. Additionally, chitosan adheres to red blood cell in blood coagulation. However, some studies have been reported natural substances were not adequately quick to hemostasis within relevant time. Thus, this study focused on recombinant batroxobin (rBat), from cDNA expressed in Pichia pastoris. In the hemostat dressing, rBat functions as fibrinogen to convert enzymes, plasminogen activators, prothrombin activators, factor X activators, or hemorrhagins such as thrombin. rBat interacts with proteins in the blood coagulation cascade and acts specifically on the fibrinolytic pathway. In contrast to thrombin, batroxobin splits off only the fibrinopeptide α -chain, leaving the β -chain of

fibrinogen unaffected. In addition, the rBat, thrombin-like enzyme components, does not influence other hemostatic factors or cells. Therefore, in this study, we fabricated a novel collagen and chitosan hemostatic scaffold containing rBat using freeze-dry method. Because each of the materials used in the scaffold involve different hemostatic mechanisms within the coagulation cascade, the scaffold was predicted to be effective at controlling bleeding. Also, incorporation of rBat was predicted to provide a synergetic effect to natural substances for induce rapid hemostasis. Experiments performed here in vitro studies to evaluate respective hemostatic mechanism confirm the efficacy of this novel hemostatic scaffold. For animal experiments, we used a Sprague-Dawley (SD) rat initial hemorrhage model. In conclusion, the scaffold dressing should be a definite improve control of excessive hemorrhage.

Speaker Biography

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Homogeneous distribution of hMSC in 3D PCL scaffold by electrical stimulation

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"issue engineering has shown tremendous promise in creating biological alternatives for harvested tissues, implants, and prostheses. An ideal scaffold for tissue regeneration should possess large porosity and pore size for good infiltration of cells, high pore interconnectivity for tissue ingrowth. Cell culture in 3-D porous scaffolds is often impaired by the difficulty of achieving a homogeneous cell seeding and by the diffusion constraints within the cell-scaffold constructs. In the present study, we investigated the effect of electric stimulation on the migration of hMSCs and infiltration of hMSCs into 3D PCL scaffold by electric stimulation. To fabricate the porous scaffold, we simulated the current distribution in designed scaffold using comsol physics computer program. We designed lattice structure for difficult infiltration without any stimulation. Then, 3D PCL scaffolds was fabricated by 3D printing. During electrotaxis on 2D, hMSCs moved toward the anode or cathode

under direct current electric fields. Cell seeded into PCL scaffold, incubated for 1day, and then treated 1000 µA electric for 3h using a customized agar-salt electrotaxis chamber. After electric current treatments, cell distribution on PCL scaffold were visualized by immunofluorescence staining. As a result, if there was no electric treatment, cell stayed near the surface of scaffold. However, electric stimulation enhanced the infiltration of cells into scaffold and hMSC distributed and proliferated on 3D scaffolds for 28d, homogenously. In conclusion, the infiltration of hMSCs into scaffold was enhanced by the control of migration using physical stimulations as electrical stimulation.

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ROS-induced cell sheet delivery method based on photofunctional polymer film

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ell transplantation is one of the promising technologies for the repair and regeneration of damaged tissues. Previous approaches have been applied the direct injections of cell suspensions and the implantation of biodegradable threedimensional scaffolds seeded cells. However, these treatments have limits due to poor localization of the injected cells and insufficient delivery of oxygen and nutrients to cells, host inflammatory reactions by scaffolds. Cell sheet engineering was recently proposed as a new approach transplanting cell sheets to the lesion without scaffolds. To harvest cultured cell sheets, most studies has focused on culture surface property variations (e.g. wettability, pH, electricity, magnetism). Photodynamic action causes production of reactive oxygen species (ROS) by interaction of light with photosensitive agents and oxygen. This study investigated ROS-induced cell detachment system to transfer cell sheet directly at target area without harvesting procedure. To produce exogenous ROS, a photosensitizerimmobilized polymer film was prepared by spin coating method. Treatment of green light to activate photosensitizer contained in the film generates ROS, which in turn causes cell sheet detachment from the film. We have found optimum conditions to detach an intact cell sheet with extracellular matrix (ECM) protein. This method is capable of efficiently transplanting cells in a simple process, by contacting the cultured film with the lesion where the cell is to be delivered and irradiating light. Also, the study demonstrated in vivo efficacy of applying this approach to subcutaneous tissues of nude mice. Therefore, the study indicates that ROS-induced method for cell sheet transplantation has potential application in tissue engineering.

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Different aspect of electro taxis in long term culture cell related to senescence

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Ctem cells may differentiate into various cell lineages. Thus, Jthese cells are promising tools for tissue engineering and cell therapies. But stem cells have limited lifespan in vitro, so the use of stem cells are restricted. It has been reported that longterm culture of stem cells causes the cells undergo senescence with cell morphology, alteration in differential potential and proliferating ability. In this study, we confirmed the possibility of applying direct current electric field to cells, known as electro taxis, to be a useful way to distinct cell. We used 3 types of stem cells (TMSC, hMSC, ADSC) and 3 passages (P5, P10, P15) for each stem cells in every experiment. We performed MTT assay to evaluate proliferative ability of 3 types of stem cells. The proliferation rate of stem cells cultured for long term was significantly decreased compared with the early phase passage 5. One of well-known senescence marker, senescenceassociated ß-galactosidase was stained, and its activity was detected. The number of ß-galactosidase stained cells were increased in long term culture compared with the early phase.

The activity of β -galactosidase shown similar pattern compared with β -galactosidase staining. In electric induced cell migration, we applied cells in charge of 1000µA for 3 hours in customized agar-salt electro taxis incubator and chamber system. Early cultured cells move toward anode with directedness direction. But in long-term culture cells directedness has been decreased. In conclusion, our result showed that long term culture of stem cells undergoes senescence which can be known by proliferation assay and senescence-associated β -galactosidase assay. Also, in electro taxis result, different result about passage increasing could be influenced by stem cell senescence which is similar aspect shown in senescence assay. Therefore, applying direct current electric field to cells can be another method to confirm senescence cells.

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Accepted Abstracts

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Deciphering spiral ganglion neurons heterogeneity by single-neuron transcriptome profiling

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Type I spiral ganglion neurons (SGNIs) are myelinated and carry all the auditory information from organ of Corti inner hair cells to the auditory brainstem. SGNIs are morphologically and physiologically diverse; they consist of low and high threshold subtypes. Moreover, they differ in their susceptibility to noise insult. These differences between types and subtypes are likely driven by distinct gene expression profiles. We used a transgenic mouse with the unique feature of fluorescently labeled SGNI and single cell gene expression profiling to distinguish different SGNI subtypes. We employed quantitative single cell RT-PCR for a preselected group of 192 candidate genes aimed to specifically distinguish among subtypes of SGNIs. These studies will contribute to elucidate significant questions in the field such as molecular profiles that define the different SGNI subtypes.

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Correction of the contracted noses

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Datients seeking dorsal augmentation often have features of a shortened and associated small nose. The small nose can be defined as a nose with a low dorsum and a short length. This is particularly common among the people of South China and South East Asia where many patients have a short and overly rotated nose. Objective markers of the short nose include a nasolabial angle of more than 100-105 degrees and when the middle third of the face (from brow to base of nose) to total facial height ratio is less than 1:3. The causes of a severely shortened nose, particularly in South East Asians include congenital causes and post-operative contraction. Post-operative causes occur due to immunologic reaction to graft material. Repeated surgery and post-operative infection are other known causes. The correction of this deformity requires dorsal augmentation and nasal lengthening. Poorly performed surgery can results in worsening of the contracture and shortening. The mainstay of surgical treatment is the extended spreader graft. In congenital cases, skin elasticity is good as no prior surgery has been performed. Adequate lengthening can be achieved and maintained with autologous septal and auricular cartilage. However, in cases of post-operative contraction, the rigid and scarred skin is often resistant to lengthening. A stronger, more rigid graft may be required. The choice of graft material in this case is often the autologous rib cartilage graft. Although bilateral extended spreader grafts give good lengthening and symmetry, the increased pressure on the dorsal septum can cause cartilage necrosis and perforation, to avoid this, the width of the spreader grafts should not exceed 5mm. The author uses a unilateral extended spreader graft to avoid this complication. When there is severe and asymmetric contraction. Adequate correction may require the use of several types of local flaps. These include unilobular and bilobed flaps, alar rotation/ advancement flaps and the subnasale flap.

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Evaluation of the hearing results after mastoidotympanoplasty operation with or without Ossiculoplasty - a percentage (%) change versus absolute change; A different methodology

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Our article provides a comprehensive review of parameters of percentage change of the results in terms of the documented hearing results (not based on any particular classification) in ear surgeries in chronic suppurative otitis media (CSOM) cases. Materials and methods: Our series is a prospective cohort observational study in 230 cases of CSOM from 2009-2016 wherein we have evaluated the hearing improvement with a parameter of percentage (%) change of hearing improvement between pre-operative and post-operative period in various types of ear surgeries. A statistically significant difference was seen in 4 out of the 14 surgical groups namely in mastoidotympanoplasty, modified

radical mastoidectomy (MRM), MRM with tympanoplasty and in tympanoplasty. The rest of the 10 surgical groups had less than 10 cases and therefore the p significance could not be determined however these groups still showed hearing improvement. The superiority of the surgical techniques like mastoidotympanoplasty, MRM, MRM with tympanoplasty and in tympanoplasty as demonstrated in our study aims to find an ideal surgical procedure in CSOM which gives the best chance to improve or preserve the hearing with complete eradication of the disease.

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Obesity and Otorhinolaryngology

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Introduction: Overweight and obesity are defined as abnormal or excessive fat accumulation that may impair health. In 2014, 39% of adults aged 18 years and over (38% of men and 40% of women) were overweight. The worldwide prevalence of obesity more than doubled between 1980 and 2014. Obesity can influence whole the body including Ear, Nose and Throat.

Overview of the course: The course will discuss diagnosis and classification of obesity. The causes, Pathophysiology, and systemic health hazards of obesity. The course will reveal the influence of obesity on the diseases of the ear, nose and throat as well as the diseases that obesity play an important role in its etiology. The attendees will learn the importance of the obesity in our daily practice. Systemic hazards of obesity on the human. Obesity as a risk factor in the etiology of some ENT diseases. How obesity and overweight influence the diseases of ENT. Counselling the obese patients before General anesthesia. At the end of the course the attendees will be able to: diagnose and classify overweight and obese patients. Know systemic and ENT hazards of obesity. Risks of General anesthesia on obese patients.

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HPV and Laryngeal cancer

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Introduction: Laryngeal cancer is the most common type of cancer in the head and neck. Human papilloma virus (HPV) represents a group of more than 150 related viruses. Infection with certain types of HPV can also cause some types of cancer. HPV now causes most Oropharyngeal Cancer; Common sites base of the tongue and tonsil. HPV as a cause of Cancer larynx is still unclear.

Overview of the course: The course will reveal all risk factors of laryngeal carcinoma, role of HPV in oropharyngeal cancer, global prevalence of HPV in cancer larynx, epidemiology of HPV in laryngeal cancer, clinical implication and prognosis of HPV

positive cases with cancer larynx, comparison between HPV positive cases with laryngeal and oropharyngeal cancer.

Learning outcome: At the end of the course attendees will lean, risk factors of cancer larynx including HPV, epidemiology of HPV in laryngeal cancer, pathophysiology of laryngeal carcinoma when infected by HPV, comparison between Oropharyngeal and laryngeal carcinoma that infected by HPV. Prognosis of HPV infected laryngeal carcinoma and biological difference between HPV positive and negative laryngeal carcinoma.

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Congenital deformed nose

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Congenital deformed noses are usually combined serious deformities which is nostril asymmetry, hypoplasia, defect of some part, deviation etc. The deformities are not only the nose but also lip, philtrum and other face too. Types of congenital deformed noses are Cleft nasal deformities, Scleroderma, Binder syndrome, Hypoplastic nose, etc. The most common congenital nasal deformity is a Cleft nose. There are two types cleft nose which is unilateral and bilateral one. The characteristics of Binder Syndrome is a very flat nose and hypoplasia of mid face. Usually there is underdeveloped nasal septum. Scleroderma has a kind of fissure which is across whole face including nose. Many of them is needed reconstructive rhinoplasty. Hypoplastic nose means underdeveloped some part of nose or whole nose. The author wants to report the experience how they have treated the congenital deformed nose.

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Cochlear implant surgery by transcanal VERIA technique: Experience from 3000 cases

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The Veria technique for Cochlear implantation is a nonmastoidectomy technique which is done through the end aural route for the cochleostomy with a transcanal tunnel drilled in the posterior canal wall. This technique has been used for implanting in more than 3000 cases. This technique uses a specially designed perforator to make the tunnel in the posterior canal wall. Though the conventional technique has been successful it is more time consuming and is prone to various complications especially in children with small facial recess, cochlear malformations and cochlear rotation. This technique is simple, helps in faster healing and earlier fitting of the processor, is precise thereby minimizing trauma to the facial nerve. The surgery can be performed in infants who have not yet developed the mastoid completely. This technique can be applied in difficult cases of common cavity, ossified cochlea, cochlear hypoplasia, otosclerosis, high jugular bulb, rotated cochlea with great ease and minimal difficulty.

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Intestinal stem cell differentiation after massive small bowel resection is regulated by Notch signaling in a rat model

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Objective: Notch signaling promotes differentiation to the absorptive cell lineage rather than to the secretory cell lineage. The objective of this study was to determine the role of Notch signaling in intestinal stem cell differentiation in a rat model of short bowel syndrome (SBS).

Methods: Male Sprague-Dawley rats were randomly assigned to one of two experimental groups of 8 rats each: Sham rats underwent bowel transection and re-anastomosis, SBS- rats underwent 75% mall bowel resection. Rats were sacrificed on day 14. Illumina's Digital Gene Expression (DGE) analysis as used to determine Notch signaling gene expression profiling. Notchrelated gene and protein expression were determined using Real Time PCR, Western blotting and immunohistochemistry.

Results: From 7 investigated Notch-related (by DGE analysis) genes 6 genes were up-regulated in SBS vs control animals

with a relative change in gene expression level of 20% or more. A significant up-regulation of Notch signaling related genes in resected animals was accompanied by a significant increase in Notch-1 protein levels (Western Blot) and a significant increase in NOTCH-1 and Hes -1 (target gene) positive cells (immunohistochemistry) compared to sham animals. Evaluation of cell differentiation has shown a strong increase in total number of absorptive cells (unchanged secretory cells) compared to control rats.

Conclusions: Two weeks after bowel resection in rats, stimulated Notch signaling directs crypt cells population toward absorptive progenitors.

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Retinal stem cells reprogramming

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A new therapeutic method of Regenerative Opthalmic Medicine. A Homotoxicological Pharmaceutical blend therapy through retrobulbar injections. The author reports his experience derived, 10 years ago, from his first Autologous Stem Cells live explantation from "Mallow Bone" (through the Hip) and re-implantation into the retrobulbar fat of the Eye. To define a new concept of "Re-Informational Therapy" for "Regenerative Opthalmic Medicine" using a system for "Reformatting" - Pathological Retina through the therapeutical "Re-regulation" of affected Tissues of the inner Eye no more capable to spontaneously regulate its normal biological functions.

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