

BANARAS HINDU UNIVERSITY
FACULTY OF MEDICINE
INSTITUTE OF MEDICAL SCIENCES
(542/02/ab/CPCSEA 27.12.2012)

No. Dean/2016/ CAEC/ 623

Dated: 21.01.2017

The Head
School of Biomedical Engineering
Indian Institute of Technology
Banaras Hindu University

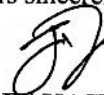
Dear Sir

A meeting of the Central Animal Ethical Committee of the University was held on 21.01.2017 at 3.00 PM in the Chamber of the Dean, Faculty of Medicine, IMS for animal ethical clearance of the proposal submitted by the following.

Name of the Student	Mr. Ajay Kumar Sahi
Title	Engineering corneal epithelial tissue
Ethical Observation	
Remarks	The Synopsis is approved by the Central Animal Ethical Committee of the University

This is for your information and necessary action at your end.

Yours sincerely,



(JAI PRAKASH)

DEAN

&

CHAIRMAN

CENTRAL ANIMAL ETHICAL COMMITTEE OF THE UNIVERSITY

Form B (per rule 8(a))

APPLICATION FOR PERMISSION FOR ANIMAL EXPERIMENTS

Application to be submitted to sent either to the CPCSEA (address in form A above) or Central Animal Ethical Committee of the University (CAECU)

Part A

*1. Name and address of establishment
Not Applicable

*2. Registration number and date of registration
Not Applicable

3. Name, address and registration number of breeder from which animals acquired (or to be acquired) for experiments mentioned in parts B & C


Central Animal House,
Registration Number: **542/AB/CPCSEA**,
Institute of Medical Sciences,
Banaras Hindu University,
Varanasi-221 005

4. Place where the animals are presently kept (or proposed to be kept)
Animal House,
Department of Pharmaceutics,
Indian Institute of Technology,
Banaras Hindu University
Varanasi.


5. Place where the experiment is to be performed
Tissue Engineering and Biomicrofluidics Laboratory,
School of Biomedical Engineering,
Indian Institute of Technology,
Banaras Hindu University
Varanasi.

6. Date on which the experiment is to commence and duration of experiment
10-12-2016; duration: 3year.

7. Type of research involved (Basic Research/Educational/Regulatory)
Basic and Application Research


Name and designation of Investigator: **(Dr. S. K. MAHLA)**
(ASSISTANT PROFESSOR)
SBME, IIT(BHU)

Date: 26/11/2016
Place: Varanasi


(Ajay Kumar Sahi)
Ph. D. Research Scholar

*Applicable only for application to be submitted to CPCSEA

PART B

Protocol form for research proposals to be submitted to the committee/Institutional Animal Ethics Committee, for new experiments or extensions of ongoing experiments using animals other than non-human primates.

1. Dissertation Title:
 “Engineering Corneal Epithelial Tissue”

Principal Investigator / Research Scholar / Research Guide / Advisor:

I. Research Guide:

- a. Name : Dr. Sanjeev Kumar Mahto
b. Designation : Assistant Professor
c. Dept / Div/ Lab : School of Biomedical Engineering
 Tissue Engineering and Biomicrofluidics Lab
d. Telephone No. : +917617052884.
e. Experience :

II. Investigator:

- f. Name : Ajay Kumar Sahi
g. Designation : Ph. D research scholar
h. Dept / Div/ Lab : School of Biomedical Engineering
 Tissue Engineering and Biomicrofluidics Lab
i. Mobile No. : +919435223391
j. Experience : Nil

3. List of names of all individuals authorized to conduct procedures under this proposal
- Dr. Sanjeev Kumar Mahto
 - Ajay Kumar Sahi
 - Ms. Suruchi Poddar
 - Mr. Kiran Yellappa Vajanthri
4. Funding source with complete address (Please attach the proof)
No
5. Duration of the project
- Number of months : 36 Months
 - Date of initiation (Proposed) : 10/12/2016
 - Date of completion (Proposed) : 10/12/2019
6. Detailed study plan may be given (Not more than one page): **Annexure-I**
7. Animals required
- Species/Common name : Charles Foster rats
 - Age / weight / size : 2 month/150-250 gm
 - Gender : Either Sex

- d. Number to be used (Year-wise breakups and total figures needed to be given) : 120
- e. Number of days each animal will be housed : 2-3 months
- f. Proposed source of animals: Central Animal House, IMS, BHU

8. Rationale for animal usage

- a. Why is animal usage necessary for these studies?

Animal experiments are required to be used for the measurement of transparency of adult rat cornea as well as a source for isolation of corneal epithelial cells which are to be incorporated into scaffolds for corneal epithelial tissue engineering applications.

- b. Why are the particular species selected required?

Charles Foster rats are widely used in the studies to isolate cornea and corneal epithelial cells which will be further used for corneal epithelial tissue engineering applications.

- c. Why is the estimated number of animals essential?

Optimization of the corneal cell scaffold construct would require repetition which would give reliable and statistically relevant data.

- d. Are similar experiments conducted in the past? If so, the number of Animals used and results obtained in brief. If yes, why new experiment is required?

Yes, similar experiments have been conducted in the past. Alaminos, Miguel, *et al.* (2006) have reported a successful growth of rabbit corneal epithelial cells and incorporated it into fibrin-agarose scaffold.

- e. Have similar experiments been made by any other organization agency? If so, their results in your knowledge.

No

9. Description of the procedures to be used.

Animals are maintained in an animal room on a 12-h light/12-h dark cycle and at constant temperature (22±2°C). The animals are further subjected to anesthesia of diethyl ether and euthanized by cervical dislocation which ensures no pain to the animal. The animal is further dissected to isolate the eye and the cornea is extracted. These corneal tissue undergo enzymatic cleavage followed by trituration processes to obtain the cells which are used. Few corneas tissue are dipped in bouins's fixative to fix the tissue followed by histological studies.

10. Please provide brief descriptions of similar studies from *in-vitro* / *in-vivo* (from other animal models) on same/similar test component or line of research. If, enough information is available, justify the proposed reasons. Alaminos, Miguel, *et al.* (2006) have reported a successful growth of rabbit corneal epithelial cells and incorporated it into fibrin-agarose scaffold. Tonsomboon *et al.*

(2013) work shows that mechanical properties of compliant hydrogels can be substantially enhanced with electrospun nanofiber reinforcement.

11. Does the protocol prohibit use of anesthetic or analgesic for the conduct of painful procedures (any which cause more pain than that associated with routine injection or blood withdrawal)? If Yes, explanation and justification
No.

12. Will survival surgery be done? No
If Yes, the following to be described.

- a. List and description of all such surgical procedures (including methods of asepsis)
- b. Names, qualifications and experience levels of operators
- c. Description of post-operative care
- d. Justification if major survival surgery is to be performed more than once
- e. On a single individual animal.

13. Methods of disposal post-experimentation
Deep pit burial

14. Animal transportation methods if extra-institutional transport is envisaged
Not Applicable.

15. Use of hazardous agents (use of recombinant DNA-based agents or potential human pathogens require documented approval of the Institutional Biosafety Committee (IBC). For each category, the agents and the biosafety level required, appropriate therapeutic measures and the mode of disposal of contaminated food, animal wastes and carcasses must be identified)
- a. Radionuclides
 - b. Microorganisms / Biological infectious Agents
 - c. Hazardous chemicals or drugs
 - d. Recombinant DNA
 - e. Any other (give name)

If, your project involved use of any of the above, attach copy of the minutes of IBC granting approval.

Not Applicable.

LIST OF PUBLICATIONS

Journals:

1. **Sahi, A. K.,** Varshney, N., Poddar, S., Gundu, S., & Mahto, S. K. (2021). Fabrication and Characterization of Silk Fibroin-Based Nanofibrous Scaffolds Supplemented with Gelatin for Corneal Tissue Engineering. *Cells Tissues Organs*, 1-22.
2. **Sahi, A. K.,** Varshney, N., Poddar, S., & Mahto, S. K. (2020). Comparative behaviour of electrospun nanofibers fabricated from acid and alkaline hydrolysed gelatin: towards corneal tissue engineering. *Journal of Polymer Research*, 27(11), 1-15.

LIST OF PUBLICATIONS

Publications not related to the thesis work:

1. **Sahi, A. K.**, Varshney, N., Poddar, S., Vajanthri, K. Y., & Mahto, S. K. (2019). Optimizing a detection method for estimating polyunsaturated fatty acid in human milk based on colorimetric sensors. *Materials Science for Energy Technologies*, 2(3), 624-628.
2. **Sahi, A. K.**, Verma, P., Varshney, N., Gundu, S., & Mahto, S. K. (2022). Revisiting Methodologies for In Vitro Preparations of Advanced Glycation End Products. *Applied Biochemistry and Biotechnology*, 1-25.
3. Varshney, N., **Sahi, A. K.**, Poddar, S., Vishwakarma, N. K., Kavimandan G., Prakash A., & Mahto, S. K. (2020). Freeze-Thaw Induced Physically Cross-linked Superabsorbent Polyvinyl Alcohol/Soy Protein Isolate Hydrogels for Skin Wound Dressing: In Vitro and In Vivo Characterization. *ACS Applied Materials & Interfaces*. (Accepted).
4. Srivastava, P., **Sahi, A. K.**, Kumar, A., & Mahto, S. K. (2022). Establishing relation between in-vivo and in-vitro Cryospray experiments through thermal characteristics. *International Journal of Thermal Sciences* 176, 107389.
5. Gundu, S., Varshney, N., **Sahi, A. K.**, & Mahto, S. K. (2022). Recent developments of biomaterial scaffolds and regenerative approaches for craniomaxillofacial bone tissue engineering. *Journal of Polymer Research*, 29(3), 1-23.
6. Vishwakarma, N. K., Singh, S., Vishwakarma, S., **Sahi, A. K.**, Patel, V. K., Kant, S., & Mahto, S. K. (2022). Converting CO₂ into heterocyclic compounds under accelerated performance through Fe₃O₄ grafted ionic liquid catalysts. *New Journal of Chemistry*.

7. Poddar, S., Agarwal, P. S., **Sahi, A. K.**, Varshney, N., Vajanthri, K. Y., & Mahto, S. K. (2021) Fabrication and characterization of electrospun psyllium husk-based nanofibers for tissue regeneration. *Journal of Applied Polymer Science*, 50569.
8. Varshney, N., **Sahi, A. K.**, Poddar, S., & Mahto, S. K. (2020). Soy protein isolate supplemented silk fibroin nanofibers for skin tissue regeneration: Fabrication and characterization. *International Journal of Biological Macromolecules*, 160, 112-127.
9. Agarwal, P.S., Poddar, S., Varshney, N., **Sahi, A.K.**, Vajanthri, K.Y., Yadav, K., Parmar, A.S. and Mahto, S.K., (2020). Printability assessment of psyllium husk (isabgol)/gelatin blends using rheological and mechanical properties. *Journal of Biomaterials Applications*, 0885328220979473.
10. Poddar S, Agarwal PS, **Sahi, A. K.**, Vajanthri KY, Pallawi, Singh KN and Mahto SK (2019). Fabrication and Cytocompatibility of Psyllium Husk (Isabgol)/Gelatin Composite Scaffolds. *Applied Biochemistry and Biotechnology*, pages 1-19. DOI : 10.1007/s12010-019-02958-7
11. Varshney N, **Sahi, A. K.**, Vajanthri KY, Poddar S, Balavigneswaran CK, Prabhakar A, Rao V and Mahto SK (2019). Culturing Melanocytes and Fibroblasts within Three-dimensional Macroporous PDMS Scaffolds: Towards Skin Dressing Material. *Cytotechnology*, 71 (1): 287-303.

Patent Applications:

1. Mahto S.K., **Sahi A.K.**, Agarwal P., Agarwal P., Maheshwari S., “A double-chamber rotating bioreactor with axial flow of culture medium for tissue engineering” Date of Filing: February 8, 2018. Indian Patent (IPR/2021-22/002).
2. Saini V, Pradhan S, **Sahi A.K.**, and Mahto S.K. “3D Printed Phantom Simulating Abdominal Structure of Human Body for Brachytherapy Applications.” (Filed).
3. Mahto SK, Varshney N, Sahi AK, “A Non-invasive Polymeric Wound Closure Device”. Application filed to Institute IPR Cell on October 9, 2019 [Application ID: IPR/2019-20/008]
4. Mahto SK, Varshney N, **Sahi AK**, “Soy-based electrospun nanofibrous sheet and the method of electrospinning thereof.” Application filed to Institute IPR Cell on October 9, 2019 [Application ID: IPR/2019-20/007]
5. Mahto S.K., **Sahi A.K.** and Rai. S, “A Syringe with Controlled Volume Dispensing System”. Application No. 201811030441. Date of filling: August 14, 2018. Indian Patent.(Published online)

Book chapters:

1. **Sahi, A. K.**, Verma, Pallawi, Singh, K., & Mahto, S. K. (2019). “Advancements and New Technologies in Drug Delivery System.” In *Biomedical Engineering and its Applications in Healthcare* (pp. 681-700). Springer, Singapore.
2. **Sahi, A. K.**, Varshney, N., Sidu, R. K., Poddar, S., Singh, K., & Mahto, S. K. (2020). “Clinical Implications of Cortisol and Bioanalytical Methods for Their Determination in Various Biological Matrices.” In *Immunodiagnostic Technologies from Laboratory to Point-Of-Care Testing* (pp. 195-221). Springer, Singapore.

International Conferences:

1. **Sahi, A.K.**, Varshney N, Mahto, S.K. (2019) “Fabrication of Silk-Gelatin Based Corneal Analogs Using Electrospinning Technique”, Annual Next-Gen Tissue Engineering and Regenerative Medicine Conference, scheduled on October 14-15, 2019 at **Singapore City, Singapore, 2019.**
2. **Sahi, A.K.**, Mahto, S.K. (2019) “An Affordable Prefilled Dual-Chamber Syringe for Drug Delivery applications”, 2nd National Biomedical Research Competition (NBRC) on 17th of November organized by PGIMER, Chandigarh under the aegis of Society of Young Biomedical Scientists (SYBS), India, 2019. **(Received appreciation award in top 10 best poster)**
3. Varshney N, **Sahi, A. K.**, and Mahto, S.K. (2019) "Optimization and Development of Natural Protein Based Electrospun Composite Scaffolds for Tissue Engineering Applications" International Conference on Biomaterial-Based Therapeutic Engineering and Regenerative Medicine BioMET-2018), IIT Kanpur, India, 2019.
4. Varshney N, **Sahi, A. K.**, and Mahto, S.K. (2019) “Development and characterization of a polymeric scaffold as a wound dressing material”. Annual Next-Gen Tissue Engineering and Regenerative Medicine, from 14-15 October 2019, **Singapore city, Singapore, 2019.**
5. **Sahi, A. K.**, Mahto, S. K. (2018) “Electrospun Silk mat for Corneal Tissue Engineering, 1st IEEE EMBS International Student Conference in Ramaiah Institute of Technology, India, from 19-21 December 2018. **(Awarded 1st prize in oral presentation)**

6. **Sahi, A. K.**, Rai. S., Mahto, S. K. (2018) “A 3D Printed Dual Chamber Syringe for Drug Delivery” 7th Inter IIT Tech Meet 2018 - Engineers' Conclave, IIT Bombay, India. December 2018.
7. Suruchi Poddar, Agarwal, P.S., **Sahi, A.K.**, Vajanthri, K.Y., Mahto, S.K. (2018) “Stabilization and Characterization of Psyllium Husk (Isabgol) Scaffolds for Wound Care Applications” International Conference on BioMaterials, BioEngineering and BioTheranostics (BioMET 2018) in association with SBAOI and STERMI 24-28th July 2018, Vellore Institute of Technology, Tamil Nadu, 2018.
8. **Sahi, A. K.**, Mahto, S. K. (2017) “Histological characterisation of corneal tissue for fabrication of physiologically functional tissue construct” Institute day 2017, Indian Institute of Technology (Banaras Hindu University), Varanasi, India, 2017.
Anjali; Sahi, A. K., Mahto, S. K. (2016) “Development of affordable and portable microfluidic device for estimation of Poly Unsaturated Fatty Acids in human milk” SYSCON 2016- Recent advances in Biomedical Research, AIIMS, New Delhi, India, 2016.
9. Anjali, **Sahi, A. K.**, Mahto, S. K. (2016) “Development of an Affordable Microfluidic based POCD for Estimation of DHA in Human Milk” Institute day 2016, Indian Institute of Technology (Banaras Hindu University), Varanasi, 2016.
10. Vajanthri, K. Y., Saxena, S., Poddar, S., Periwal, A., Agarwal, P., **Sahi, A. K.**, Chaudhary, A., Mahto, S. K. (2016) “Fabricating Functional Skeletal Muscle Tissue Constructs Using Decellularized Matrices” Indian Medical Device Expo, a joint initiative by IIT Bombay, COE Pune and VNIT Nagpur, held at College of Engineering, Pune, India, 2016.

Workshops and Hands-on-trainings:

1. **Training program in generation and maintenance of Human iPS cells**“CiRA-ADBS training program in generation and maintenance of human induced pluripotent stem cells,” organized by the Accelerating the application of Stem cell technology in Human Diseases (ASHD) program and The Center for iPS Cell Research and Application (CiRA), **Kyoto University, Japan**. November 27th-December 3rd, 2019. **(Was among the 8 selected researchers to be awarded with DBT travel grant to attend the Workshop)**
2. Participated in a device design challenge hands-on-workshop organised by Collaborative Medical Device Design Initiative (CO-MEDDI) in collaboration with King George's Medical University, Lucknow, Uttar Pradesh, India. (February 25-27, 2020)
3. Participated in Idea Exposition on “Innovation in Bioengineering & Healthcare” organised by BIRAC Regional Center (BRIC), in collaboration with IKP and IIT (BHU), Varanasi on October 23-24, 2019 at School of Biochemical Engineering, IIT (BHU), Varanasi.
4. Participated in three-day Workshop on “Innovative Design & Manufacturing Education” supported by Pandit Madan Mohan Malaviya National Mission on Teachers and Teaching (PMMMNTT), IIT(BHU), Varanasi, India (7-9th April 2018).
5. Actively Participated in two-day Workshop on “Research Methods and Data Analysis Using SPSS & R” organized by Department of Management Studies, Indian Institute of Information Technology (IIIT) Allahabad, India (13-14 November 2017).

6. Successfully completed the Workshop on ÄKTA, organized by School of Biochemical Engineering, IIT(BHU), Varanasi, India (10-11th august 2017).
7. A GIAN course on “Nano- biotechnology for Healthcare” organised by Indian Institute of Technology Kharagpur (IIT-Kgp) (29th June-8th July, 2017).
8. Participated in three-day Workshop on “Emerging Trends in Drug Design and Molecular Modelling” organized by Design Innovation Centre (DIC), Banaras Hindu University and IIT(BHU), Varanasi, India (19th -21st July, 2017).
9. Volunteered one-day Brainstorming workshop on establishing “Centre for Advanced Biomaterials and Tissue Engineering” Indian Institute of Technology (Banaras Hindu University), Varanasi, India (Mar. 19, 2018)
10. Volunteered one-day Brainstorming workshop on establishing “Centre for Advanced Biomaterials and Tissue Engineering” Indian Institute of Technology (Banaras Hindu University), Varanasi, India (Mar. 19 2018)
11. Volunteered a short-term course on “Tissue Engineering” under the Quality Improvement Programme (QIP) sponsored by AICTE, New Delhi (Jan. 09-14, 2017)