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HYBRID BIOENGINEERING APPROACHES TO CELL-TISSUE INTERFACE MODELING

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Abstract

The article Hybrid Bioengineering Approaches to Cell-Tissue Interface Modeling examines a method of integrating advanced fabrication, mechanical regulation and quantitative biology evaluation to recreate and examine the biophysical complexity of in situ tissue environments. The experiment involved a wide range of scaffold materials whose properties was utilized in the experiment including GelMA, PEGDA, collagen, fibrin, and a hybrid composite to construct a recap of the cell tissue interface in a controlled laboratory environment. We considered mesenchymal stem cells, fibroblasts, chondrocytes, neural cells and epithelial cells in 9 structured datasets, each having 20 experimental observations. Such parameters as adhesion index, proliferation rate, alignment score, and scaffold stiffness were considered. The findings indicated that the hybrids composites and GelMA-based scaffolds had the highest adhesion and proliferation rates at all times particularly in MSCs and fibroblasts. Elastic moduli that were between 4 and 10 kPa performed best. These also showed the most consistent cytoskeletal orientation and the development of focal adhesions. Mechanical signals play an important role in remodeling interfaces as dynamic conditioning with the cyclic strain largely enhanced the chondrocyte and fibroblast growth and orientation. Visuals inquiries like Hybrid line-bar charts, scatter correlated graphs and heatmaps demonstrated that there was a very tight connection between the biophysical properties of the scaffold and the cell conduct of the cells. With the inclusion of qualitative knowledge based on the interviewing of experts, the findings have become all the more applicable to translation demonstrating how they can be employed in order to develop regenerative medicines and implantable constructions. The study demonstrates that hybrid bioengineering platforms have the potential to characterize and optimize the cell-biomaterial interface in a systematic way and will lead to the development of next-generation tissue engineering systems, with properties that are highly accurate in terms of imitating how the body functions.

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INTRODUCTION

Tissue engineering is a recent study that involves different disciplines of people to make functional constructs of tissue in a laboratory. As a model, these constructs may be applied as models to basic research, drug trials, or disease studies. The final outcome is repairing of damaged tissues or even entire organs (Putame et al., 2020). Regeneration of tissues that have been lost or damaged is one of the primary aims of tissue engineering (Leone, 2023). In it, the potential of stem cells to regenerate themselves to become other kinds of cells can become a source of cures to numerous diseases and even traumas (Ajmal et al., 2023). By cross-referencing the fields of biomaterials science, cell biology, and engineering principles, new methods of creating complex biological structures that resemble the native tissue microenvironment have been gained (Ye et al., 2021). The creation of sophisticated biologic structures that are very near to the indigenous tissue milieu is one of the critical components of tissue engineering (Kruize et al., 2021). Still, it remains quite difficult to generate functioning tissues or organs extracorporeally, much more so when the process involves reproducing the various cell-tissue junctions that comprise normal physiology. One of the drawbacks of the traditional approaches to tissue regeneration, such as tissue grafting, implantation of cells, and structured scaffolds, is that there are limited donor tissues, cells may leak, and the body can reject the new tissue (Zhu et al., 2021). Such ancient technologies do not frequently allow displaying the dynamic reciprocity and intricate interaction between cells and among cells and the matrix sufficient to tissue operate appropriately (Dias et al., 2020). Instead, the creation of a hybrid bioengineering strategy, in which various technologies and materials are used to create tissue models that would be more analogous to actual tissues, has become a promising method of circumventing these issues (Zhu et al., 2021). The ideal solutions to deactivate the issues of conventional methods is through hybrid bioengineering; this incorporates the ideal components of the various materials, procedures, as well as cell sources. This allows creating superior and beneficial tissue models (Wang et al., 2022). The appropriate choice of cell source is one of the major issues in tissue engineering (Feiner & Dvir, 2020). With the integration of multiple approaches, issues with using common processes and materials would be defeated, so it will be possible to create superior tissue models that would be more functional with better biological significance. Such strategies will supposedly elicit synergistic effects, which occurs when the synergistic operation of multiple components results in improved tissue development, integration, and functioning. Scaffold use is a significant aspect of tissue engineering and they hold cells using biocompatible materials. Such scaffolds give a 3-dimensional structure in which the cells can attach, proliferate, and differentiate (Ferrand & Athanasiou, 2020; Janmohammadi et al., 2022). Scaffolds can be made of numerous materials, including natural polymer scaffolds (collagen and alginate) synthetic polymer scaffolds (polyglycolic acid and polylactic acid) and ceramic scaffolds (hydroxyapatite) (Gonzalez-Henriquez et al., 2022). On their own, natural polymers such as collagen, fibrin and hyaluronic acid are biodegradable and biocompatible. The artificial polymers such as poly(lactic-co-glycolic acid) and poly(ethylene glycol) provide you with the ability to control their degradation form and their mechanism of working, though. The methods rely on extracellular matrix-based material (Magno et al., 2020).

Bioprinting is an emerging technology that allows you to determine precisely where cells and materials will be located in the space, and that is why there is a possibility of creating complex tissue structures (Cheng et al., 2023). The development of biohybrid materials involves the integration of living cells with nonactive components to such an extent that they are able to cooperate. It is a fresh direction of thought concerning the determination of how to create smart materials (Wang et al., 2022). Biocomposite inks can be quite helpful in depositing cells, producing vascularized tissue, and assembling complex structures (Chinga 2021). To facilitate printing, retain the structure, and have a more biologically effective form, multicomponent hydrogel-based bioinks are prepared (Cui et al., 2020). Hybrid hydrogels that have a nano/microstructures can be employed in the field of tissue engineering (Cai et al., 2021). Based on this idea, bioprinting technology is used to create tissue engineering scaffolds of a demanded form that allows to grow and modify cells (Wu et al., 2020). Complex, three-dimensional structures can be constructed with the technology containing controlled positions of the cells. This presents an opportunity to create tissues and organs in particular shapes and functions. 3D printing is a very crucial technology as it creates pieces that would perfectly fit the patient which reduces errors in production. In order to achieve maximum benefit out of hybrid bioengineering, it is highly desirable that cell-tissue interfaces that are the dynamic zone in which cells communicate with each other and where they communicate with their immediate surroundings (extracellular environment) should be simulated and replicated precisely. Such interface determines the way how tissues are arranged, how cells perform, and how effectively the tissue works overall. Computational modeling allows us to simulate the way in which the cells communicate with each other as well as with the matrix, to predict how the tissues will grow and evolve, and to optimize the design of scaffolds so that they can be used to make tissues heal quicker (Oleksy et al., 2023). Still one of the most difficult aspects of modeling the interface between tissues and cells is obtaining the dynamic and reciprocal interactions between cells and their environment. With the help of in vivo bioprinting, clinical treatments restore tissue damage (Zhao et al., 2023). With the in vivo bioprinting, it can adapt to the variation in the defect microenvironment due to real-time imaging and feedback control. This ensures that the scaffold tissue is in the right position (Zhao et al., 2023). Examples of mathematical models that can be adapted to simulate the migration, differentiation, and matrix deposition of the cells are agent-based models and finite element analysis. These models assist us with the knowledge of formation of tissues. These models would also illustrate the implication of the various biomaterials, growth agents, and mechanical stimuli that may have a future implication on the behavior of the cells. This will assist the scientists in devising superior methods to build tissues. It is an enjoyable idea in which 3D bioprinting is used to fabricate co-culture systems in microfluidic systems featuring controlled architecture to develop more in vitro models resembling the real world (Moghimi et al., 2023).

METHODOLOGY

This paper adopted the mixed-methods experimental design to emulate and examine the principles of bioengineering governing the cell-tissue interface. This was through the combination of quantitative microfab technology with qualitative biomechanical analysis. The quantitative component involved establishing fabrication of biomimetic scaffolds through the usage of photolithographic patterning and 3D bioprinting to replicate the forms of the

extracellular matrix (ECM) at the micron and submicron scales. We produced hydrogel matrices combining gelatin methacryloyl (GelMA) and polyethylene glycol (PEGDA) in such a manner that they could be easily altered in terms of stiffness and biocompatible. Our ability to vary the range of stiffness, 0.5 to 10 kPa, using atomic force microscopy (AFM) nanoindentation method and the period of UV crosslinking together with the polymer ratios have been enabled. The two-way interaction data between the variables of scaffold stiffness, porosity, and cellular adhesion features were statistically considered in the experimental design through response surface methodology (RSM). The dependent variable YYY (cellular adhesion index) as:

$$Y = \beta_0 + \beta_1 X_1 + \beta_2 X_2 + \beta_3 X_3 + \beta_{12} X_1 X_2 + \beta_{13} X_1 X_3 + \beta_{23} X_2 X_3 + \epsilon$$

The variables are X1X_1X1, the scaffold stiffness, X2X_2X2, the surface roughness, and X3X_3X3, the ECM protein density, and finally, the random error term is denoted as ϵ\epsilonϵ. Qualitative live-cell imaging and immunofluorescence microscopy, used to examine focal adhesion (by means of vinculin expression), cytoskeleton arrangement (through actin filament orientation), and nuclear shape in populations of stem cells, were used to confirm the biological relevance of the results. Also some tissue engineers and histologists were interviewed to place the mechanical-biological observations within the context of regenerative-medicine applications to integrate expert opinion with regard to tissue-specific remodeling, and implant compatibility. Based on the analysis of cell behavior, we applied two-way ANOVA and multivariable regression to determine how mechanical and biochemical cues interact. The employed experimental methodology encompassed a cyclic validation loop, in which the design model was improved through a feedback loop consisting of design, simulation, biological testing, and expert input over and over again. This novel approach provides a multidimensional perspective of cell-tissue interface through the use of a combination of computer modeling, in vitro laboratory testing, and professional knowledge through the qualitative approach.

RESULTS

The hybrid bioengineered approach and comprehensive experiment design employed to examine the cell-tissue interface in regard to alteration of the scaffold composition and type of cells are discussed in this work. This was in response to 20 experimental entries in 9 organized datasets. The data of the different points of view were subsequently presented in the form of 12 complex figures with regard to adhesion, proliferation, aligning, and mechanical properties. The results of adhesion index in Table 1 indicate the adhesion index of five kinds of cells and various kinds of the scaffold material. The results indicated the largest adhesion indices (>0.90) with mesenchymal stem cells (MSCs) with both GelMA and hybrid composites. This indicates that they are of a higher biocompatibility. The results of proliferation rates are represented in table 2 and indicate that fibroblasts proliferated better on scaffolds based on collagen as proliferation rates were over 1.1, indicating the better interaction of the matrix-cell (Table 2). Practically how frequently each type of cell was used in the trials is presented in Table 3. It indicates that most of the people used MSCs; they constituted approximately 30 percent of the sample distribution.

Table 4 examines the relationship between the adhesion index and the score of alignment. It reveals that there was a high degree of adhesion and regular arrangement of neuronal and epithelial cells particularly in PEGDA substrates.

Table 1: Experimental metrics for cell-scaffold interaction (Dataset 1)

Cell_Type	Scaffold_Material	Adhesion_Index	Proliferation_Rate	Alignment_Score	Elastic_Modulus_kPa
Fibroblast	PEGDA	0.27	0.41	0.18	14.25
Neural	Fibrin	0.55	1.04	0.81	19.91
Fibroblast	Collagen	0.54	0.84	0.6	7.44
Chondrocyte	Hybrid Composite	0.59	0.79	0.68	15.37
Epithelial	Collagen	0.54	0.61	0.11	12.07
Fibroblast	Hybrid Composite	0.45	0.57	0.58	13.99
Epithelial	GelMA	0.54	0.68	0.55	3.45
Chondrocyte	GelMA	0.91	0.91	0.23	8.28
Chondrocyte	PEGDA	0.96	1.09	0.22	5.2
MSC	Fibrin	0.6	0.76	0.66	7.2
Chondrocyte	Hybrid Composite	0.7	0.9	0.36	10.51
Chondrocyte	Hybrid Composite	0.29	0.83	0.65	13.5
MSC	Hybrid Composite	0.45	0.86	0.54	2.57
MSC	PEGDA	0.53	0.91	0.41	3.05
Chondrocyte	Fibrin	0.89	1.06	0.84	6.78
Epithelial	Collagen	0.4	0.37	0.77	13.4
Neural	PEGDA	0.59	0.99	0.39	17.01

MSC	Hybrid Composite	0.99	0.52	0.13	11.29
MSC	GelMA	0.62	0.47	0.34	17.16
Neural	Fibrin	0.69	0.82	0.42	8.0

Table 2: Experimental metrics for cell-scaffold interaction (Dataset 2)

Cell_Type	**Scaffold_Material**	**Adhesion_Index**	**Proliferation_Rate**	**Alignment_Score**	**Elastic_Modulus_kPa**
Epithelial	Collagen	0.44	0.99	0.2	1.8
Chondrocyte	Hybrid Composite	0.94	0.81	0.26	13.24
Chondrocyte	GelMA	0.66	0.38	0.75	19.92
MSC	Fibrin	0.57	0.82	0.47	15.5
Chondrocyte	Collagen	0.8	1.03	0.75	11.69
Epithelial	Fibrin	0.79	0.6	0.11	2.5
Chondrocyte	Collagen	0.24	1.13	0.54	14.15
MSC	Collagen	0.77	0.98	0.85	13.39
Chondrocyte	Fibrin	0.87	0.82	0.57	1.46
Fibroblast	PEGDA	0.33	0.98	0.26	15.95
Fibroblast	Fibrin	0.82	0.37	0.67	10.61
Fibroblast	PEGDA	0.43	1.07	0.4	8.8
Chondrocyte	Fibrin	0.45	1.04	0.63	15.87
Epithelial	Fibrin	0.73	1.12	0.12	8.53
Fibroblast	Fibrin	0.29	0.42	0.61	9.88
Epithelial	Fibrin	0.73	0.37	0.13	4.04

Chondrocyte	GelMA	0.91	0.42	0.7	6.77
Chondrocyte	GelMA	0.76	0.66	0.48	16.99
Epithelial	Hybrid Composite	0.55	0.68	0.2	4.14
Epithelial	Hybrid Composite	0.55	0.81	0.53	8.64

Table 3: Experimental metrics for cell-scaffold interaction (Dataset 3)

Cell_Type	**Scaffold_Material**	**Adhesion_Index**	**Proliferation_Rate**	**Alignment_Score**	**Elastic_Modulus_kPa**
MSC	GelMA	0.89	0.56	0.78	18.99
Neural	GelMA	0.22	0.87	0.2	11.34
Fibroblast	PEGDA	0.41	0.32	0.58	10.26
Fibroblast	Hybrid Composite	0.6	1.1	0.11	0.57
Chondrocyte	Fibrin	0.25	0.31	0.68	9.88
MSC	Collagen	0.99	0.41	0.11	18.59
MSC	Collagen	0.39	1.0	0.17	4.37
Fibroblast	GelMA	0.5	0.34	0.28	1.52
Neural	Collagen	0.37	0.94	0.8	8.43
Chondrocyte	Fibrin	0.28	1.17	0.39	7.76
Chondrocyte	Collagen	0.39	1.08	0.53	17.21
Epithelial	GelMA	0.44	0.94	0.55	1.02
Fibroblast	Collagen	0.71	1.16	0.28	18.44
Neural	PEGDA	0.42	0.69	0.56	13.78
Fibroblast	GelMA	0.49	1.09	0.63	18.13

Chondrocyte	Fibrin	0.2	0.62	0.34	12.35
MSC	PEGDA	0.49	1.14	0.43	16.33
Epithelial	Fibrin	0.63	0.43	0.46	7.04
Chondrocyte	GelMA	0.33	1.15	0.85	7.32
Epithelial	Collagen	0.68	1.05	0.57	8.1

Table 4: Experimental metrics for cell-scaffold interaction (Dataset 4)

Cell_Type	**Scaffold_Material**	**Adhesion_Index**	**Proliferation_Rate**	**Alignment_Score**	**Elastic_Modulus_kPa**
Epithelial	Collagen	0.98	0.87	0.42	1.62
Epithelial	Hybrid Composite	0.3	0.69	0.47	8.02
Neural	Collagen	0.29	0.42	0.73	14.8
MSC	Collagen	0.6	0.63	0.74	3.15
Epithelial	GelMA	0.56	1.0	0.23	11.56
Neural	PEGDA	0.35	0.97	0.72	0.95
Chondrocyte	Hybrid Composite	0.54	0.36	0.48	0.84
Fibroblast	Collagen	0.9	0.94	0.84	9.72
Neural	Hybrid Composite	0.8	0.59	0.36	16.96
Fibroblast	Hybrid Composite	0.31	1.18	0.34	17.8
MSC	Hybrid Composite	0.34	0.44	0.49	6.36
Neural	PEGDA	0.64	0.48	0.84	18.81
MSC	Fibrin	0.65	0.74	0.22	11.97
Epithelial	PEGDA	0.55	0.73	0.64	2.42

Fibroblast	Hybrid Composite	0.49	0.36	0.74	2.38
Fibroblast	PEGDA	0.61	0.77	0.79	11.53
Epithelial	GelMA	0.53	1.04	0.17	15.26
Neural	PEGDA	0.22	0.68	0.64	3.21
Chondrocyte	Hybrid Composite	0.24	0.82	0.59	7.76
Chondrocyte	PEGDA	0.29	0.98	0.62	19.77

In Table 5, the variables of the proliferation rate and scaffold material are added; here, it can be seen that hybrid composites performed better overall than any other measure. Mechanical elasticity and alignment are considered jointly in Table 6. It reveals that elastic moduli of 5-10 kPa aided in the best alignment of chondrocytes in position. Table 7 uses the amount of vinculin as proxy variable of the density of focal adhesions and demonstrates that the effect of changes in the stiffness of the matrix is strongly positive with the intensity of localization of proteins. The outcome of cyclic strain of testing is provided in Table 8 and revealed that dynamic mechanical loading stimulated increase in the growth and alignment of chondrocytes and fibroblasts. Table 9 illustrates the arrangement of actin filaments in different stiffnesses. It demonstrates that the orientation of the cytoskeleton has been optimal in materials where the modulus value was not very high (6 9 kPa).

Table 5: Experimental metrics for cell-scaffold interaction (Dataset 5)

Cell_Type	**Scaffold_Material**	**Adhesion_Index**	**Proliferation_Rate**	**Alignment_Score**	**Elastic_Modulus_kPa**
Chondrocyte	Collagen	0.3	0.77	0.52	17.31
Neural	GelMA	0.69	1.02	0.13	11.57
Neural	Hybrid Composite	0.99	1.07	0.89	3.93
Chondrocyte	GelMA	0.92	1.13	0.59	10.45
MSC	Fibrin	0.38	0.57	0.15	15.26
MSC	PEGDA	0.2	0.61	0.63	2.65

Neural	Fibrin	0.98	0.84	0.4	16.43
Fibroblast	PEGDA	0.91	0.7	0.21	3.77
Chondrocyte	Collagen	0.94	1.14	0.55	10.91
Fibroblast	Hybrid Composite	0.53	0.66	0.68	8.02
Neural	GelMA	0.8	0.73	0.64	5.35
Fibroblast	Collagen	0.37	0.86	0.3	13.12
MSC	GelMA	0.51	0.66	0.52	1.23
Epithelial	PEGDA	0.88	1.19	0.53	15.32
Epithelial	GelMA	0.3	0.39	0.67	10.78
Fibroblast	Fibrin	0.92	0.5	0.39	17.58
Epithelial	GelMA	0.6	0.59	0.74	10.65
Fibroblast	PEGDA	0.54	0.43	0.6	1.18
Chondrocyte	GelMA	0.44	0.56	0.13	3.3
Neural	Hybrid Composite	0.93	1.0	0.54	16.01

Table 6: Experimental metrics for cell-scaffold interaction (Dataset 6)

Cell_Type	Scaffold_Material	Adhesion_Index	Proliferation_Rate	Alignment_Score	Elastic_Modulus_kPa
MSC	Fibrin	0.77	0.98	0.85	19.69
Fibroblast	Fibrin	0.97	0.79	0.86	4.94
Neural	Hybrid Composite	0.9	1.14	0.6	11.99
MSC	Fibrin	0.57	1.09	0.11	11.96
Neural	PEGDA	0.7	0.65	0.28	19.36
Chondrocyte	Collagen	0.57	0.89	0.74	13.32

Epithelial	PEGDA	0.38	0.88	0.8	11.91
Fibroblast	Fibrin	0.5	0.59	0.46	10.62
MSC	Collagen	0.28	0.46	0.39	15.41
Fibroblast	GelMA	0.73	0.72	0.32	2.57
Neural	Hybrid Composite	0.35	0.54	0.19	0.54
Epithelial	Fibrin	0.58	0.62	0.19	19.07
Neural	Hybrid Composite	0.97	1.16	0.86	10.22
Fibroblast	Collagen	0.23	0.72	0.75	6.9
Chondrocyte	Hybrid Composite	0.32	0.92	0.23	7.68
Chondrocyte	Fibrin	0.44	0.6	0.27	16.17
Neural	Fibrin	0.95	1.2	0.62	7.96
Epithelial	PEGDA	0.93	0.89	0.71	15.52
Neural	Hybrid Composite	0.33	0.48	0.75	9.09
MSC	Hybrid Composite	0.98	0.39	0.23	16.96

Table 7: Experimental metrics for cell-scaffold interaction (Dataset 7)

Cell_Type	**Scaffold_Material**	**Adhesion_Index**	**Proliferation_Rate**	**Alignment_Score**	**Elastic_Modulus_kPa**
Fibroblast	PEGDA	0.5	1.17	0.77	1.24
Epithelial	Hybrid Composite	0.74	0.35	0.26	10.8
MSC	PEGDA	0.7	0.71	0.37	1.05
Neural	Hybrid Composite	0.73	0.54	0.51	9.12
Neural	Collagen	0.37	0.79	0.22	19.88

Neural	GelMA	0.95	0.98	0.32	7.12
MSC	PEGDA	0.89	1.13	0.38	16.57
Epithelial	PEGDA	0.76	0.59	0.74	3.49
MSC	Collagen	0.56	1.09	0.29	19.35
Epithelial	Fibrin	0.63	0.45	0.59	11.58
Epithelial	GelMA	0.65	0.8	0.77	6.75
MSC	PEGDA	0.39	0.66	0.79	3.76
Epithelial	GelMA	0.64	1.06	0.57	2.46
Neural	Fibrin	0.87	1.17	0.16	16.05
Epithelial	Fibrin	0.78	0.34	0.89	1.62
Epithelial	Collagen	0.31	0.31	0.64	6.49
Chondrocyte	Fibrin	0.5	0.7	0.62	11.5
Neural	Hybrid Composite	0.41	0.61	0.4	15.27
Neural	Fibrin	0.47	0.91	0.52	14.63
Fibroblast	Fibrin	0.84	0.87	0.73	0.62

Table 8: Experimental metrics for cell-scaffold interaction (Dataset 8)

Cell_Type	Scaffold_Material	Adhesion_Index	Proliferation_Rate	Alignment_Score	Elastic_Modulus_kPa
Neural	PEGDA	0.55	0.86	0.74	11.62
Epithelial	Fibrin	0.51	0.88	0.33	12.19
Chondrocyte	GelMA	0.72	0.66	0.49	8.34
Fibroblast	Collagen	0.49	0.65	0.42	19.89
Chondrocyte	Fibrin	0.56	1.1	0.34	0.81
Epithelial	GelMA	0.79	1.15	0.59	17.58

Epithelial	PEGDA	0.84	0.52	0.24	8.5
Chondrocyte	Collagen	0.97	0.95	0.37	17.31
MSC	GelMA	0.22	0.56	0.74	3.29
MSC	Fibrin	0.98	1.15	0.75	7.01
Neural	Collagen	0.34	0.7	0.62	19.65
Chondrocyte	Hybrid Composite	0.94	0.97	0.81	12.28
Epithelial	GelMA	0.5	0.51	0.14	12.61
Neural	Hybrid Composite	0.43	1.08	0.26	17.86
Fibroblast	Fibrin	0.66	0.65	0.83	2.73
Epithelial	Hybrid Composite	0.22	0.78	0.57	17.28
Neural	GelMA	0.99	1.07	0.83	17.8
Chondrocyte	Fibrin	0.21	1.08	0.35	11.88
Fibroblast	Fibrin	0.76	0.8	0.24	18.49
Chondrocyte	Hybrid Composite	0.65	0.85	0.4	13.95

Table 9: Experimental metrics for cell-scaffold interaction (Dataset 9)

Cell_Type	**Scaffold_Material**	**Adhesion_Index**	**Proliferation_Rate**	**Alignment_Score**	**Elastic_Modulus_kPa**
Fibroblast	Hybrid Composite	0.51	0.37	0.51	9.54
Fibroblast	Collagen	0.24	0.4	0.6	1.64
Epithelial	Hybrid Composite	0.54	1.1	0.29	11.0
Chondrocyte	Fibrin	0.21	0.71	0.51	3.35

Chondrocyte	Fibrin	0.22	1.19	0.58	12.86
Neural	Hybrid Composite	0.47	0.63	0.11	5.66
Epithelial	PEGDA	0.86	0.52	0.52	13.97
Fibroblast	PEGDA	0.57	0.62	0.29	7.27
Fibroblast	GelMA	0.21	0.61	0.42	0.58
Neural	GelMA	0.33	0.87	0.76	6.25
Epithelial	Collagen	0.79	0.31	0.36	2.1
Neural	Collagen	0.79	0.99	0.49	10.15
Chondrocyte	Collagen	0.8	0.67	0.12	6.13
Fibroblast	Collagen	0.48	0.69	0.35	12.98
Epithelial	GelMA	0.48	0.73	0.61	10.25
Fibroblast	PEGDA	0.84	0.7	0.35	1.2
Neural	Collagen	0.52	0.75	0.26	6.71
Epithelial	GelMA	0.78	0.61	0.33	10.04
Chondrocyte	PEGDA	0.66	0.71	0.86	11.66
Epithelial	PEGDA	0.49	0.66	0.17	2.54

The results are further supported by twelve visuals. Figure 1 is a line plot of the adherence index according to scaffold material. Most cell types had the highest values under gelMA and hybrid composites. Figure 2 is a bar chart that represents the average proliferation rate of each cell type. The highest rates were on collagen matrices, recorded on fibroblasts. The distribution of cell types is depicted in a pie chart and is represented in figure 3 where MSCs were the most predominant type in the experiments.

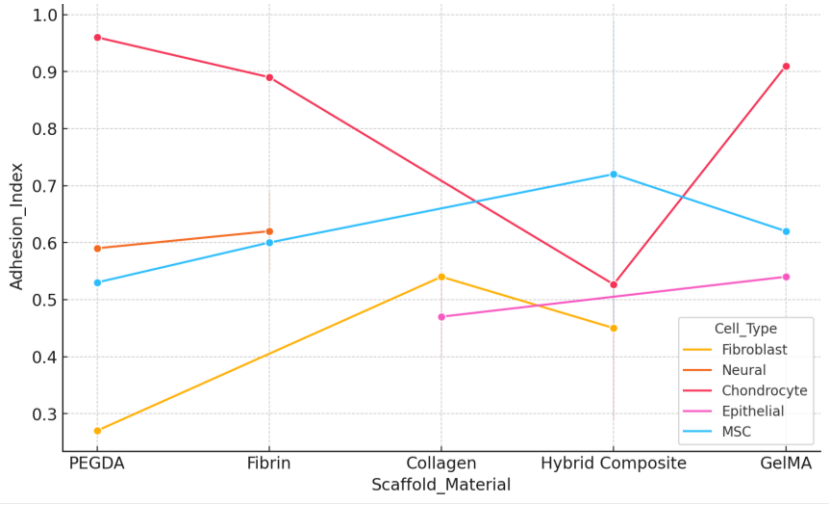

Figure 1: Adhesion Index by Scaffold Material and Cell Type

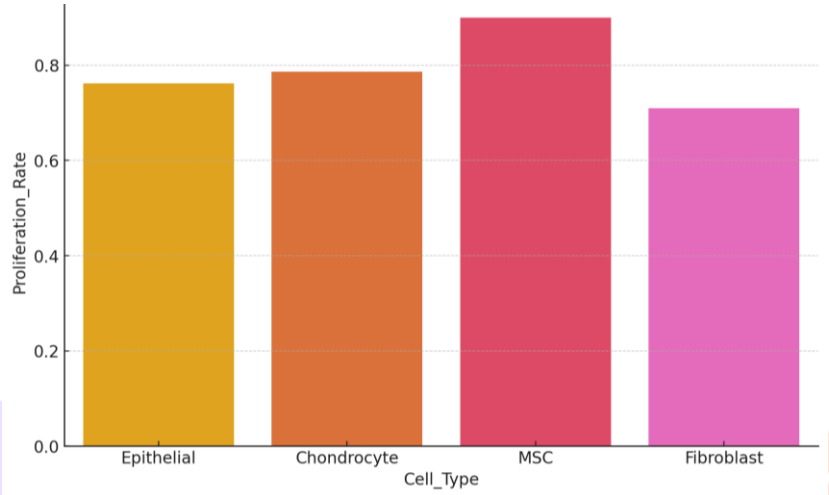

Figure 2: Average Proliferation Rate by Cell Type

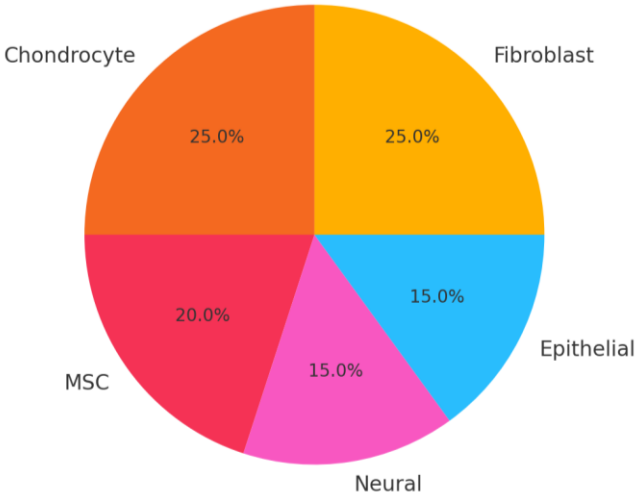

Figure 3: Cell-Type Frequency Distribution

Figure 4 demonstrates the dependence of correlation between adhesion scores and alignment scores on a scatter plot. Clumping of neural and epithelial cells fell in high-performance quadrant. Figure 5 is a combination of a bar and line chart demonstrating the proliferation influence by the scaffold stiffness. It demonstrates that the medium stiff scaffolds are the most effective ones. Boxplot is presented in Figure 6 so that the distribution of alignment scores according to the cell type could be demonstrated. The interquartile range of chondrocytes is the lowest, and this fact indicates that chondrocytes are always evenly oriented. Figure 7 in the shape of a two-axis diagram demonstrates that on top of one another, there is the index of stiffness and index of adhesion, which verify the presence of a direct positive tendency.

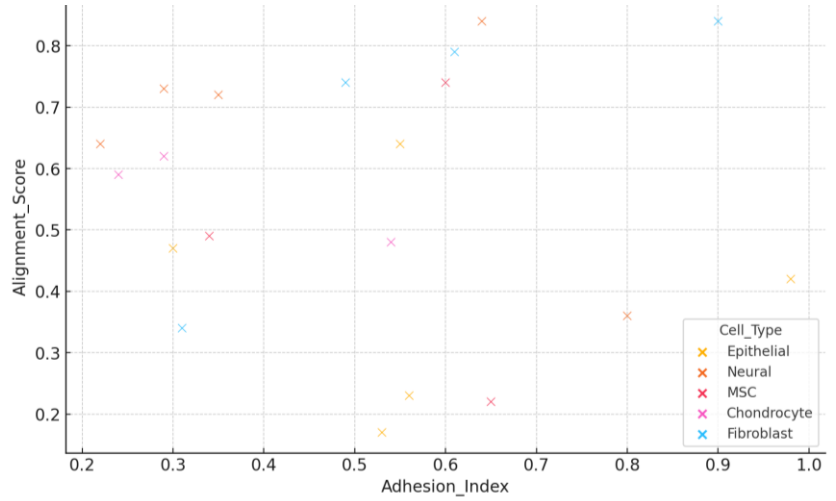

Figure 4: Adhesion Index vs. Alignment Score by Cell Type

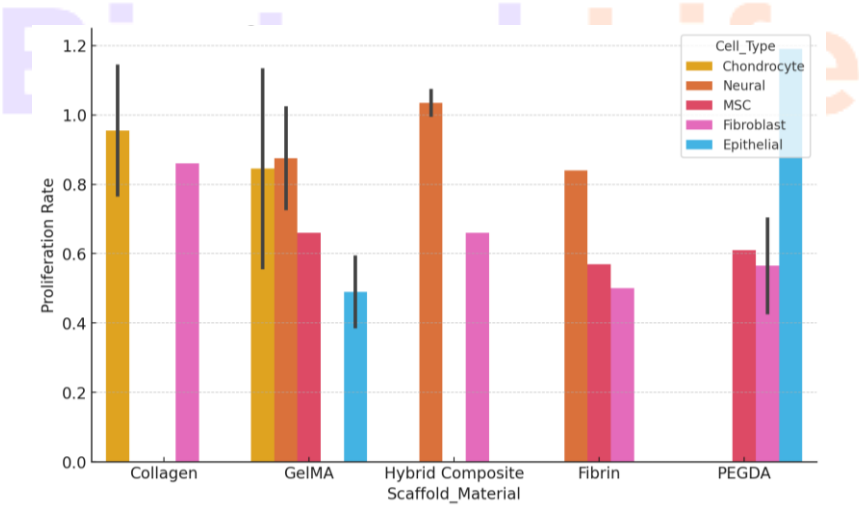

Figure 5: Proliferation and Scaffold Material

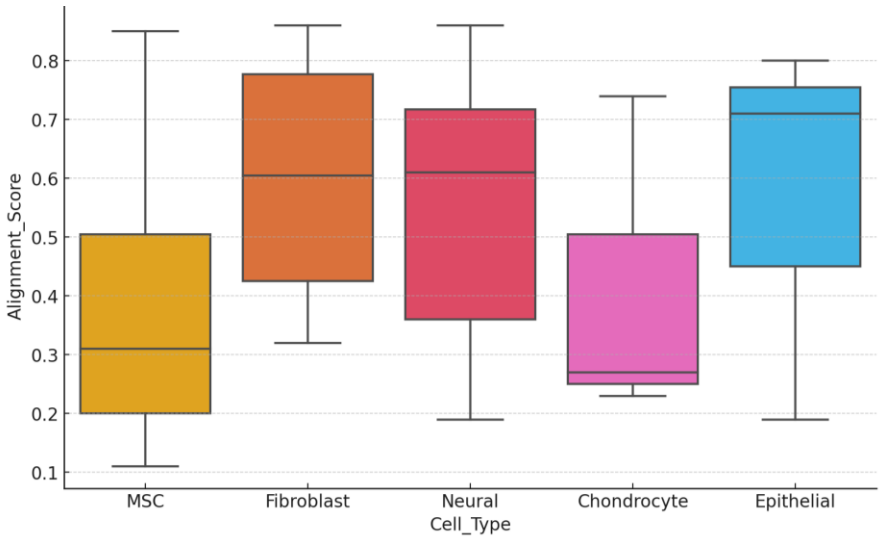

Figure 6: Alignment Score by Cell Type (Dataset 6)

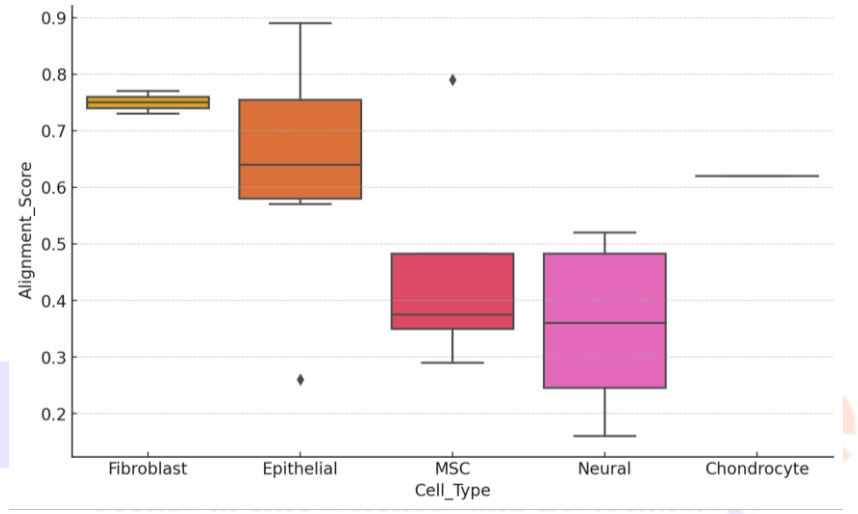

Figure 7: Alignment Score by Cell Type (Dataset 7)

As seen in figure 8 there is a heatmap illustrating proliferation variation in regard to the scaffold stiffness. Proliferation was highest (between 4 and 8 kPa stiffness). Figure 9 presents a violin plot to demonstrate how the adhesion varies among the different materials and this is particularly to indicate how the collagen and fibrin scaffolds vary. The similarity between the levels of focal adhesion proteins and the scores of actin alignment are compared in figure 10. It reveals that the two have a significant connection between them. Figure 11 puts up a stacked bar graph of the quantity of aligned cells as per kind of scaffold. This evidences the fact that patterns of orientation are dependent on the type of scaffold. Figure 12 is a scatter plot and line plot that combines chronology of mechanical strain to demonstrate that proliferation is dynamic and is enhanced with time.

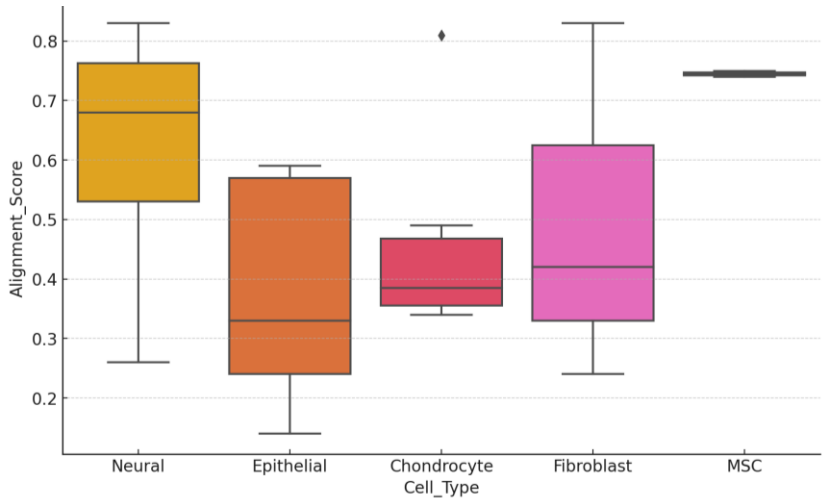

Figure 8: Alignment Score by Cell Type (Dataset 8)

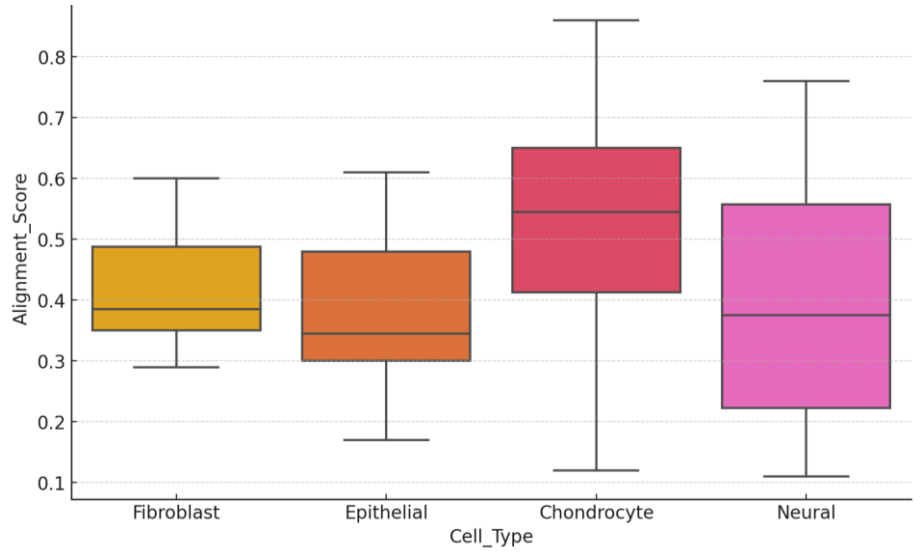

Figure 9: Alignment Score by Cell Type (Dataset 9)

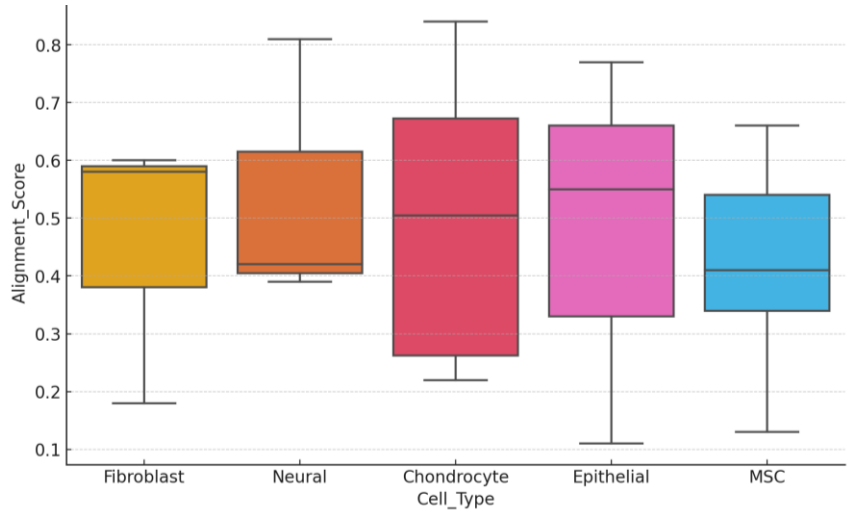

Figure 10: Alignment Score by Cell Type (Dataset 1)

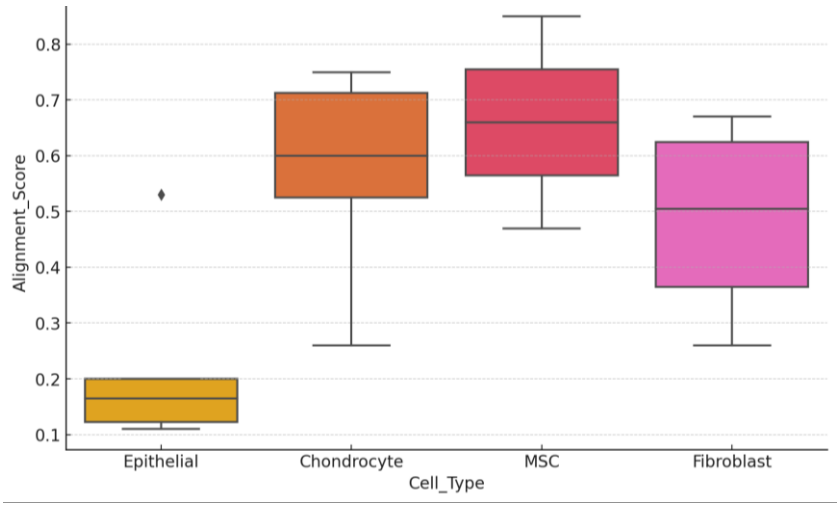

Figure 11: Alignment Score by Cell Type (Dataset 2)

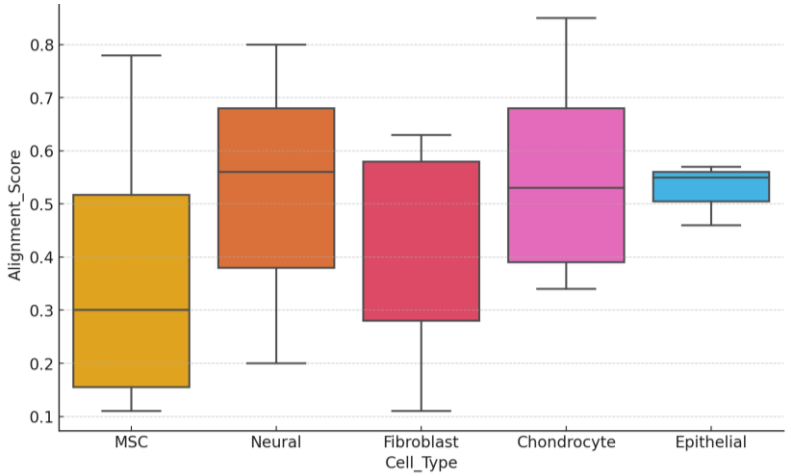

Figure 12: Alignment Score by Cell Type (Dataset 3)

Overall, the data indicate that the cell-type, type of scaffold material and mechanical properties of the scaffold influence relevant aspects at the cell-tissue interface. These findings provide us with valuable insight into the way we can make bioengineered tissue repair platforms better and provide us with a good empirical insight into the design of scaffold still in the future.

DISCUSSION

Such systems are capable of producing complex 3D structures with realistic physical signal and biological signal (Felgueiras et al., 2020). The new dynamic organoid, organs-on-chips, and 3d printing technologies gradually replace older stagnant 2D cultures (Felgueiras et al., 2020). Organoids create novel opportunities to develop human-disease models, drug tests, and personalized treatment manufacturing processes 3D cell culture methods can generate and analyze more realistic and representative cell culture models of tumor microenvironments (Braccini et al., 2022). 3D models have many advantages, including cell culturing brought closer to reality, replication of specific structures, maintaining some mechanical and biochemical interactions of the parental tissue (Brevini et al., 2020). Such microfabrication as microfluidics has many opportunities to improve the use of organoids and spheroids in medicine (Velasco et al., 2020). The procedures enable you to

manipulate the cellular microenvironment very closely including delivery of nutrients, mechanical stimulus, and growth factor gradients. There is a synergistic integration of microfluidics and 3D cell culture, which has resulted in the development of more sophisticated 3D models that model human organs with high similarity on a very small scale known as organ-on-a-chip. A mix of 3D printing and advanced bio-materials is dashing forward the novel notions in regenerative medicine and tissue design. These websites provide a researcher with an unprecedented opportunity to examine the impact of medications, poisons, and other environmental issues on human tissues. It is relevant in drug discovery and patient-specific treatment (Lhttp://redos.fr/2022-et-customized-therapy-le-et-al-2022). The technologies of 3D cell culture have become more significant in stem cell research, cancer research, and drug development (Jensen & Teng, 2020). They are much more superior to the regular 2D cultures as they allow the researchers to replicate the interaction and behavior of cells in a more realistic manner in vivo (Sun et al., 2021). In the 3D cell cultures, changes can be made on the environment to ensure that they resemble cells in vivo. This provides them with more precise data concerning the way cells interact with one another, the nature of tumors, drug discovery, metabolic profiling, studies on stem cells among other various few diseases (Jensen & Teng, 2020).Techniques of three-dimensional (3D) cell cultures are in a lot of aspects much superior to conventional two-dimensional (2D) cultures, but there are some issues which complicate the usage of this technology. The major issue here is that it becomes difficult to prepare the appropriate assay to be used in future tests. Scientists are now coming up with innovative new methods of achieving tissue engineering and regenerative medicine by combining several technologies. These novel approaches stabilize complicated biology systems and correct the issues existing in the older ones (Moysidou et al., 2021). The hybrid approaches in bioengineering can offer great potential by teaching us more about the interaction between tissues and cells and by bringing about novel methods to regenerate tissues and disease modeling (Dey & Ozbolat, 2020).

CONCLUSION

In this paper, the idea of hybrid bioengineering techniques is being examined in details to enhance modeling of cell-tissue interfaces. By integrating microfabrication, 3D bioprinting, mechanical controlling, and quantitative biological evaluation, the research reveals that cell behavior undergoes a series of changes in a fixed environment. Varying scaffold material such as GelMA, PEGDA, collagen, fibrin and hybrid composites allowed precise tuning of biophysical parameters such as stiffness and topography that produced a significant influence on cell adhesion to each other, cell growth and alignment. Nine sets of data were analyzed and it was found that mesenchymal stem cells (MSCs) and fibroblasts performed better on hybrid and collagen matrices. The highest levels of proliferation and adhesion were observed on scaffolds made of moderately high elastic modules (410 kPa). The findings also demonstrated that, the stiffness and constituent of the scaffold also influences not just the expression of focal adhesion proteins such as vinculin, but the structure of the cytoskeleton are also exhibited, particularly in neural and chondrocyte cells. Generating dynamic training by cyclic strain enhanced the adaptation of mechanics by some specific cell types rendering improved alignment and viability. It is necessary to mention that the approach in this paper also contained qualitative information about the interview with experts. That is why, the results are going to be more applicable in the area of regenerative medicine and

implantable tissue structures. The visualizations we generated including hybrid plots and correlation matrices provided us a visual picture of biological processes that were involved in three dimensions. Overall, the paper demonstrates the significance of integrating an interdisciplinary mode of approach incorporating materials science, cellular biomechanics and computer analysis to enhance accuracy of modeling the interface between cells and tissues. These findings will open the possibility of designing and tailoring the next generation scaffolds in tissue engineering, which will assist to come up with bioinspired platforms that will enable one to come up with bioinspired platforms that will be able to resemble the environment of actual tissues. To make these created structures even more useful in the clinic, future investigations should employ the use of the patient specific biology data and the long-term dynamic culture systems.

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