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Organ on a Chip in Advanced Clinical Study: A Review

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Atul Anil Phalke*[1], Shital Pharate[2], Unmesh Wankhede[3]

L.S.D.P Collage Of Pharmacy Mandavgan Pharata, Dist-Pune, Tal-Shirur, Pin code-412211. India.

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ABSTRACT

Organ on a chip (OOAC) is an advanced clinical technology in recent years at various stages of the drug discovery and development process. Animal experiments are too unusual for preclinical screening in the drug development process, various complications like ethical considerations and species differences remain. To solve this problem, the assay in which the human-derived cells are actively pursued. Although it remains difficult to accurately predict drug discovery, toxicity, efficacy, and drug interactions, because cultivated cells do not recognize their original organ functions and morphology in vitro culture system so, the technology organ on chip based on the microfluidic device have been recently used in vitro organ model. It is a multi-channel 3D microfluidic cell culture chip that stimulates the activities, mechanics, and physiological response of entire organs and organ systems The chips are lined with human cells and their tiny fluidic channels. various organs like the liver, heart, kidney, gut, etc. are have been reproduced as in vitro models. We herein provide that the organ on chip is an advanced clinical method for drug discovery and their challenges in the future.

INTRODUCTION:

An organ-on-chip is a multi-channel 3D microfluidic cell culture, an Integrated circuit that stimulates the activities, mechanics, and physiological response of entire organs and organ system, a type of artificial organ. It constitutes the subject matter of significant biomedical engineering research, more precisely in bio-MEME. The convergence of lab on chip (LOC_s) cell biology has permitted the study of human physiology in an organ of a specific context, introducing a novel model of in vitro multicellular human organisms. Organs that have been stimulated by microfluidic devices include the brain, lung, heart kidney, liver, prostate, vessels(artery), skin, bone, cartilage, and more.

In 2010, Harvard's Wyss Institute led by Donald Ingber, produced the first successful hip, a lung model. Two years later Ingber's lab was included in a public-private collaboration tasked with creating 10 different Human organ-on-chips.[22]

Fig 1: Seeding of cells in the chip

Liver on chip:

It is important to detect the metabolic ability and toxicity during the drug discovery process, so the liver is the main organ related to drug metabolism.[1] The liver constitutes a series of complex hepatic lobules that confer multicellular functional communication, so the hepatic system is the major site of drug metabolism. Maintaining the physiology of hepatocytes is challenging.[2]Three-dimensional scaffolds were combined with a cell-retaining filter and structural support in a cell culture medium across the top of the array and through the 3D cell aggregates in each channel. A cell culture chamber was designed so that flow rates meet

estimated values of cellular oxygen consumption while providing fluid shear stress within the physiological range. The scientist demonstrated that this device enables the formation of hepatocellular aggregates reminiscent of structures seen in hepatic acini, and should maintain their structure and viability for up to 2 weeks using this device.[1]

Kidney on chip:

The kidney is a more important organ responsible for metabolism and excretion in vivo. During drug discovery, drug candidate efficacy and toxicity in the kidney are evaluated by animal testing because there are not suitable in vitro models.[1]The initial design of the kidney on-chip has two parts. A top channel increases the urinary lumen and has fluid flow, whereas the bottom chamber mimics interstitial space and is filled with media. Kidney cells are under much lower shear stress than lung or endothelial cells. This device used rat distal tubular cells or MDCK cells, and its shear stress was ˷1dyn/cm^2.[3]

The kidney contains more than 10 cells, types which are highly organized in the 3D structure surrounded by extracellular matrix and complex vasculature. The renal tubular system is key in the development of DIKI and drug interaction including cellular influx, efflux, and intracellular metabolism. Clinically relevant DIKI biomarkers monitored after drug exposure of kidney-on-a-chip devices need to be established to reflect in vivo situations.[4]

Intestine on chip:

The small intestine is the major site for digestion, drug and nutrient absorption, interaction with the commensal microbiome, and development of mucosal immunity, as well as a primary site of diseases.

The Intestine Chip contains normal human intestinal epithelial cells derived from endoscopic biopsies or tissue reactions of living human intestine and intestinal tissue-specific microvascular endothelial cells.[5]

Microfluidic devices containing hollow microchannels less than 1 mm in width support laminar fluid flow and control of nanoliter to microliter scale fluid volumes, and thus, they are amenable to use for the culture of living cells. By using a syringe or a peristaltic pump, a culture medium may be perfused at desired flow rates through each microchannel, which can mimic the dynamic ranges of fluid flows and associated shear stresses on the cell surface that are observed in the human intestine lumen and the blood capillaries. This fluidic control also

enables the delivery of growth factors, nutrients, and drug compounds to the intestinal epithelium grown on the microfluidic channels in a highly regulated spatiotemporal manner.[6]

Lung on chip:

The human lungs serve the vital purpose of gas exchange in the respiratory system. The lung-on-a-chip is a complex, three-dimensional model of a living, breathing human lung on a microchip. The device is made using human lung and blood vessel cells and it can predict absorption of airborne nanoparticles and mimic the inflammatory response triggered by microbial pathogens. It can be used to test the effects of environmental toxins, absorption of aerosolized therapeutics, and the safety and efficacy of new drugs. It is expected to become an alternative to animal testing.

The lung-on-a-chip places two layers of living tissues-the lining of the lung's air sacs and the blood vessels that surround them across a porous, flexible boundary. Air is delivered to the lung lining cells, a rich culture medium flows in the capillary channel to mimic blood, and cyclic mechanical stretching is generated by a vacuum applied to the chambers adjacent to the cell culture channels to mimic breathing.

The research findings for lung-on-a-chip were published in the June 25, 2010, issue of Science,[7] the academic journal of the American Association for the Advancement of Science. The research was funded by the National Institutes of Health, the American Heart Association, and the Wyss Institute for Biologically Inspired Engineering at Harvard University.

Inventors:

The technology was developed by Donald E. Ingber, M.D., Ph.D., an American cell biologist who is the Founding Director of the Wyss Institute for Biologically Inspired Engineering at Harvard University, and Dan Dongeun Huh, Ph.D., who was a Technology Development Fellow at the Wyss Institute and is now Wilf Family Term Chair Assistant Professor in Bioengineering at the University of Pennsylvania. The device was created using a microfabrication strategy known as soft lithography that was pioneered by George M. Whitesides, an American chemist, who is a professor of chemistry at Harvard, as well as a Wyss Institute core faculty member.

Testing:

The response of the lung-on-a-chip to inhaled living pathogens was tested by introducing E. Coli bacteria into the air channel on the lung air sac side of the device while flowing white blood cells through the channel on the blood vessel side. The lung cells detected the bacteria and, through the porous membrane, activated the blood vessel cells, which in turn triggered an immune response that ultimately caused the white blood cells to move to the air chamber and destroy the bacteria.

Researchers also introduced a variety of nanoscale particles (such as those found in commercial products, and air and water pollution) into the air channel. Several types of these nanoparticles entered the lung cells and caused the cells to overproduce free radicals and to induce inflammation. Many of the particles passed through the model lung into the blood channel, and mechanical breathing was found to greatly enhance nanoparticle absorption from the air sac into the blood.

The Wyss Institute team is working to build other organ models, such as a gut-on-a-chip, as well as bone marrow and even cancer models. They are exploring the potential for combining organ systems, such as linking a breathing lung-on-a-chip to a beating heart-on-a-chip. The engineered organ combination could be used to test inhaled drugs and to identify new and more effective therapeutics that lack adverse cardiac side effects.

Gut-on-a-chip:

The human gut-on-a-chip contains two microchannels that are separated by the flexible porous Extracellular Matrix (ECM) coated membrane lined by the gut epithelial cells: Caco-2, which has been used extensively as the intestinal barrier.Caco-2 cells are cultured under spontaneous differentiation of their parental cell, a human colon adenocarcinoma, that represents the model of protective and absorptive properties of the gut.[8] The microchannels are fabricated from polydimethylsiloxane (PDMS) polymer.[9] To mimic the gut microenvironment, peristalsis-like fluid flow is designed. By inducing suction in the vacuum chambers along both sides of the main cell channel bilayer, the cyclic mechanical strain of stretching and relaxing is developed to mimic the gut behaviors.[9]Furthermore, cells undergo spontaneous villus morphogenesis and differentiation, which generalizes characteristics of intestinal cells.[10]Under the three-dimensional villi scaffold, cells not only proliferate but metabolic activities are also enhanced.[11]

They showed that cells cultured using the device are reported 4/6 undergo spontaneous 3D villus morphogenesis and small intestinal cell differentiation under these physiological conditions. To replicate more accurate physiological conditions, this device was used to co-culture multiple commensal microbes in contact with intestinal epithelial cells, and to analyze resulting physiological phenomena. Oral administration is one of the most common methods for drug administration. It allows patients, especially out-patients, to self-serve the drugs with minimal possibility of experiencing acute drug reactions and in most cases: pain-free. However, the drug's action in the body can be largely influenced by the first-pass effect. The gut, which plays an important role in the human digestive system, determines the effectiveness of a drug by absorbing its chemical and biological properties selectively.[12] While it is costly and time-consuming to develop new drugs, the fact that the gut-on-a-chip technology attains a high level of throughput has significantly decreased research and development costs and time for new drugs.[13]

Modeling inflammatory bowel disease. (IBD):

Even though the cause for inflammatory bowel disease (IBD) is elusive, its pathophysiology involves the gut microbiota.[14] Current methods of inducing IBD are using inflammatory cues to activate Caco-2. It was found that the intestinal epithelium experienced a reduction in barrier function and increased cytokine concentrations, [13] The gut-on-a-chip allowed for the assessment of drug transport, absorption, and toxicity as well as potential developments in studying pathogenesis and interactions in the microenvironment overall. [15]

Modeling radiation-induced cell injury:

The chip was used to model human radiation-induced injury to the intestine in vitro as it recapitulated the injuries at both cellular and tissue levels. Injuries include but are not limited to the inhabitation of mucus production, promotion of villus blunting, and distortion of microvilli.[16]

Pancreas on chip:

Pancreas-on-a-chip (PoC), which refers mainly to the study of the endocrine part of the pancreas on a microfluidic chip, may be used as a standardized and real-time assessment platform for evaluating islet potency and quality. The design of the chip depends largely on the function of the organ. Therefore, each organ type needs an individualized system to mimic its micro-environment. Microfluidic platforms for islets may have trapping sites in

which islets are immobilized and cultured under medium flow. Trapping of islets is mostly done by micro-wells or by constrictions that are fabricated in micro-channels under continuous flow. Using such microfluidic platforms, islet viability and composition, as well as hormone and metabolite secretions by islets, could be tested "on-chip". Perfusion is essential to maintain the viability of trapped islets, removal of metabolic products, and stimulation of islets with chemicals, hormones, and nutrients such as glucose. Precise fluid control is crucial to minimize any damage to the trapped islets and to analyze hormone kinetics in relation to biochemical and metabolic stimulation in the islets. Jun et al. have developed a microfluidic perfusion system providing an osmosis-driven low-speed flow (1.54-5.04 μm/s) comparable to in vivo interstitial flow levels. This allows culturing reaggregated islets in "concave" microwells for a month with a low flow rate that could prevent shear force damage to cells and provide continuous oxygen and nutrient supplies for islets. This model aids in studying islet function post-isolation and has the potential to be used as an in vitro model for diabetic drug testing.[17] Physical protection of islets from the flow within a chip platform by placing islets in a micromesh sheet has also been reported as a strategy to avoid damaging islets by shear force while allowing perfusion of oxygen and nutrient to the islet in the chip. Size heterogeneity of isolated islets (50-400 μm) reduces the viability of islets in microfluidic chips due to the fact that large islets could damage to a greater extent by the flow rate. Therefore, having islets of different sizes in a microfluidic platform could create challenges in the amount of oxygen and nutrient that islets need. It has been shown that smaller islets perform better than large islets both in clinical settings and in culture. Standardization of islet size by engineering size-controlled cell clusters is one strategy to overcome the variation in oxygen and nutrient demand by islets in the microfluidic platform and also flow rate damage in the system.[18]

Ovaries on chip:

Conventional 2D or even 3Din vitro culture models for human reproductive organs cannot properly recapitulate the bidirectional endocrine crosstalk between the uterine endometrium and the ovary. This crosstalk is essential for maintaining the various physiological features and functions of each tissue. Moreover, most in vitro models for the female reproductive tract also fail to mimic its multicellular structure. We, therefore, developed a novel 'dual reproductive organ on a chip' that reflects the bidirectional endocrine cross-talk and the complex multicellular structures by integrating various cellular components of both the human uterine endometrium and the ovary with several biodegradable natural polymers.

Indeed, the bidirectional endocrine crosstalk between these two tissues is achieved through media sharing between channels, and it can markedly improve the viability of loaded cells within each chamber of the chip platform. In addition, we also identified a reliable reproductive toxicity marker, SERPINB2, which is significantly increased in response to various toxic exposures in both endometrial and ovarian follicular cells. Based on these findings, we next established a SERPINB2 luciferase reporter system that was specifically designed for detecting and quantifying the toxicity of certain substances. By introducing this SERPINB2 luciferase reporter system into the loaded cells within the chip platform, we ultimately developed an effective 'dual reproductive organ-on-chip that was successfully used to predict the reproductive toxicity of various hazardous materials.[19]

Over the past decade, organs-on-a-chip and microphysiological systems have emerged as disruptive in vitro technology for biopharmaceutical applications. By enabling new capabilities to engineer physiological living tissues and organ units in the precisely controlled environment of microfabricated devices, these systems offer great promise to advance the frontiers of basic and translational research in biomedical sciences. Here, we review an emerging body of interdisciplinary work directed towards harnessing the power of organ-on-a-chip technology for reproductive biology and medicine. The focus of this topical review is to provide an overview of recent progress in the development of micro-engineered female reproductive organ models with relevance to drug delivery and discovery.[19] We introduce the engineering design of these advanced in vitro systems and examine their applications in the study of pregnancy, infertility, and reproductive diseases. We also present two case studies that use organ-on-a-chip design principles to model placental drug transport and hormonally regulated crosstalk between multiple female reproductive organs. Finally, we discuss challenges and opportunities for the advancement of reproductive organ-on-a-chip technology.[20]

Testing on chip:

Multi-organ-chip cocultures of human liver and testis equivalents were maintained at a steady state for at least 1 week and the co-cultures reproduced specific natural and drug-induced liver-testis systemic interactions.

Current benchtop nephrotoxicity models typically do not include. hepatic metabolism and interactions of the liver-testis axis. However, these are important to study the biotransformation of substances.

Testicular organoids derived from primary adult testicular cells and liver spheroids consisting of cultured HepaRG cells and hepatic stellate cells were loaded into separate culture compartments of each multi-organ-chip circuit for co-culture in liver spheroid-specific medium, testicular organoid-specific medium, or a combined medium over a week. Additional multi-organ chips (single) and well plates (static) were loaded only with testicular organoids or liver spheroids for comparison. Subsequently, the selected type of medium was supplemented with cyclophosphamide, an alkylating anti-neoplastic prodrug that has demonstrated germ cell toxicity after its bioactivation in the liver, and added to chip-based co-cultures to replicate a human liver-testis systemic interaction in vitro. Single chip-based testicular organoids were used as a control.[21]

Bone Marrow on chip:

Bone marrow is a complex organ that is responsible for producing all of the Bone, blood cell types of our body, and immune system cells in a process known as hematopoiesis, erythrocytes, granulocytes, monocytes, lymphocytes, and platelets. Bone marrow-on-a-chip could also generate blood cells, which could circulate in an artificial circulatory system to supply a network of other organs-on-chips Bone. Bone marrow is located in trabecular cavities of bones and comprises several cell types, such as hematopoietic stem cells (HSCs), mesenchymal stem cells (MSCs), osteoblasts, and other niche cells. The microvascular system of Bone marrow.[23]

Brain on chip:

The brain is the most complex organ in the human body, comprises the central nervous system (CNS) along with the spinal cord.[24] Many organ-on-a-chip methods are in process, researchers have tried to mimic the physiology of human tissue on an engineered platform. In the case of brain tissue, structural connections and cell-cell interactions are important factors for brain function. The brain comprises multiple layers in which numerous neuronal, glial, and immunological cells interact and are functionally protected by the skull from mechanical stress and by the tight Blood-brain barrier from the toxicants. The biochemical member structure is specific to the brain. Due to the neuronal plasticity and cellular interactions, modeling the complexity of the human brain is a huge challenge. The brain consists of endothelial cells, with tight junctions, which separate blood and extracellular fluid of the CNS., the brain potentiality of a 3D-microfluidic model of the BBB based on human cells to include blood flow movement by adding shear stress. Brain chip implants are part of modern

culture. In 1929 the device called EEG (Electro Encephalography) was invented by HANS BERGER in the field of human brain research which helped to record the human brain signals. Brain chip technology involves communication based on the neural activity of the brain.[25]

The skin on chip:

The skin is the largest organ of the body. The main functions of the skin are sensory, thermoregulatory, metabolic, and protective. As the physical barrier against the environment, it controls the passage of molecules and ions while protecting against microorganisms, ultraviolet radiation, and toxic or mechanical agents.

The skin also acquires a thermoregulatory role, keeping body temperature constant. Skin cells synthesize vitamin D.It decreases the risk of developing diseases such as osteoporosis, arthritis, fractures, muscle weakness, and cancers. Human skin is composed of three structural layers: the epidermis, the dermis, and the hypodermis.

The cellular skin substitutes include epidermal cell monolayers and dermo-epidermal bilayers (holding keratinocytes and fibroblasts) skin substitutes. Microfluidic devices with micrometer-sized chambers that allow the dynamic culture of cells inside, to model or mimic the physiology of a tissue or organ.[26]

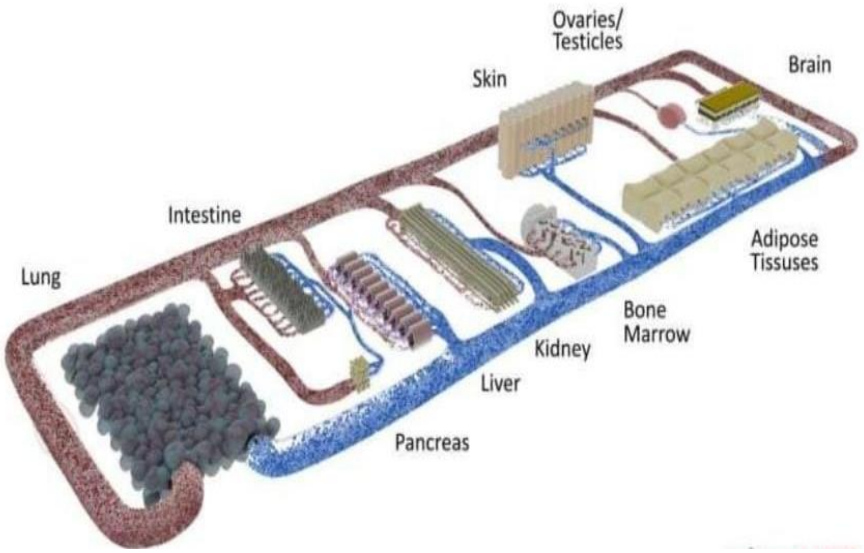

Fig 2: Different organ on a chip

ABBREVIATIONS:

OOC: Organ-On-Chip,3D: Three-dimensional,2D: Two-dimensional, LOC: Lab on-chip, MDCK: Madin-Darby Canine Kidney, ECM: Extracellular matrix, PDMs: polydimethylsiloxane, CNS: Central nervous system, IBD: Inflammatory Bowel Disease, POC: Pancreas on-chip, HSCS: Hematopoietic stem cell, MSCs: Mesenchymal stem cell, EEG: Electro Encephalography, BBB: Blood-brain barrier.

CONCLUSION:

In this review, we outlined the organ-on-chip model is used in the drug discovery process in vitro study. This technology is most useful in the different research fields. We hope this paper will help to improve and understand such collaborative work.

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Image Author -1 	***Atul Anil Phalke*** *Loknete Shri Dadapatil Pharate College of Pharmacy ,Shirur ,Pune ,Maharashtra,412211.* *At Post Pimpalsuti ,Tal-Shirur,Dist-Pune,412210.*
Image Author – 2 	***Shital Pharate*** *Loknete Shri Dadapatil Pharate College of Pharmacy ,Shirur ,Pune ,Maharashtra,412211* *At post Mandavgan Pharata,412211*
Image Author -3 	***Unmesh Wankhede*** *Loknete Shri Dadapatil Pharate College of Pharmacy, Shirur, Pune, Maharashtra,412211.* *At post Mandavgan Pharata, Shirur, Pune,412211.*