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3D CANCER TISSUE MODEL ORGANOTYPIC CULTURE AND IT'S IMPACT ON DRUG SCREENING

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ABSTRACT

The ability to accurately forecast the effects of possible cancer chemotherapeutic medications in human systems using in vitro simulations is essential for their successful translation to the clinic. The necessity of improving efficient in vitro drug screening is driven by the high cost and extended duration of preclinical and clinical research. In many cases, 3D models are used instead of conventional cultures to simulate cell morphology and cell-interactions are applicable physiologically for forecasting treatment results for complicated varying malignancies. The development of tissue mimetic models and complex simulations of physiology are made possible by bio printing and microfluidic technology, respectively, providing more accurate prediction of drug interactions. Realistic tissue models are specifically organotypic tissue structures built using organ-on-a-chip or cell-laden matrices. The advancement in the screening of in vitro cancer medication led by bio printing and microfluidic chips, as well as associated difficulties, are highlighted in this review's projection of the progress made toward the development of biomimetic tissue models.

Keywords- 3D bioprinting, organotypic models, cancer tissue model, drug screening, microfluidic chips, tumour micro-environment.

I. INTRODUCTION

In vitro and in vivo investigations have historically been used to clarify the pathophysiology, processes, and drug screening of human disease. Drug toxicity and efficacy testing in preclinical animal studies raises ethical questions and does not always accurately forecast the impact on human clinical trials. Approximately 8% of translations from animal models to clinical cancer trials are successful on average[1]. Animal models are only partially capable of simulating complicated processes including physiology, inter tumoral heterogeneity in malignancies, and human carcinogenesis and progression. In order to create more accurate organotypic tissue model in vitro that are alike to the *in vivo* physiological condition and produce more foreseeable interaction between host and drug, the current situation justifies their development. Organotypic cancer tissue models include components that are similar to the matrix to imitate the intricate exchanges found in tissues and often include two or more primary cell types[2]. These in vitro models saw a rapid transition from standard 2D cultures to co-cultures of varying cells and 3D spheroids constructs with materials that resemble biomimetic micro physiological systems and represent the extracellular matrix (ECM)[3]. A multidisciplinary integrated technique called 3D bio printing makes it easier to recreate organ or tissue systems. The intricate anisotropic tissue structure is mimicked by the cell-filled bio ink that is dispersed in a robotically controlled manner. There are further benefits to printing microfluidic devices by skill fully manipulating chip geometries and multiphase fluid flow[4]. Any biological sample on a chip drug screening that is high throughput, dependable, and sensitive is made possible by microfluidic chips. When evaluating modified T cells for T-cell associated therapy and combination therapy, microfluidic 3D models replicate the tumour micro-environment. The development of organotypic 3D models using subject-specific culture holds potential as a viable method to lessen the reliance on *in vivo* testing. With a focus on the advancement and difficulties in 3-D bio printing and micro fluid chip technology, this paper examines the strategies that combine the precision of source of cells selected, materials used, and innovative technological solutions like High Throughput Screening(HTS) for creating organotypic tissue models in vitro[5],[6],[7],[8].

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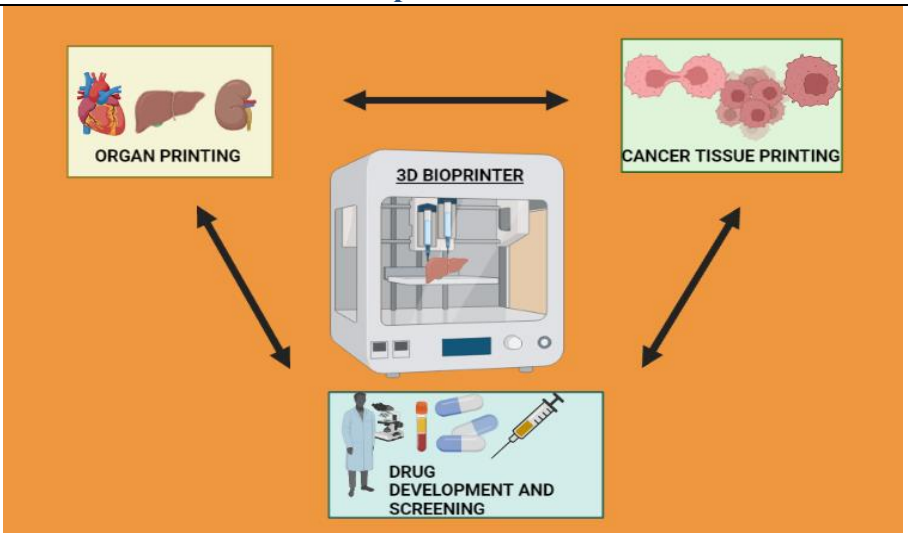

Figure.1 Role Of 3d Bioprinter In Development Of Cancer Model And Drugs

II. METHODS FOR OBTAINING CELLS FOR CULTURE

A complicated micro-environment with variety of cells, extra cellular matrix, and different types of cell interactions is what distinguishes the proliferation and spreading of cancer. Standard laboratorial tests on cell reactions to treated subjects are being conducted for more than a century using adherent cells of continuous cell lines and monolayer cultures[9]. Even though the widely- established two dimensional cultures are well recognised, and provided knowledge about how drugs work, there are still certain restrictions. The time-consuming, expensive, and labour-intensive screening process is ineffective in accurately predicting how drugs or candidate compounds would interact with cells. The association between 2D cultures and the *in vivo* environment is constrained by the missing of complexity of the structure, micro-environmental tissue structure, and cell and matrix interactions[10]. Protein expression, genetic molecular expression, cellular signalling, migration of cells, morphological features, proliferation, cell viability, their arrangement, and metabolism of drug are all different in response to the anti-tumour medications in 2-D and 3-D cultures. Different 3D models based on different sources of cells and cell-matrix combinations have been created to appropriate the efficacy of drugs in order to get around the drawbacks of monolayer cultures. By enabling intra-cellular interactions for preserving functionality of cells and homeostasis of the tissue, the 3D environment simulates *in vivo* physiology. It is possible to use existing secondary cell lines, primary cells, or more modern, tailored patient-tissue-derived cells when evaluating cancer drugs[11-12]. The development of sources from which the cells are obtained, components of culture, and technical improvements for in vitro model development is schematically depicted in

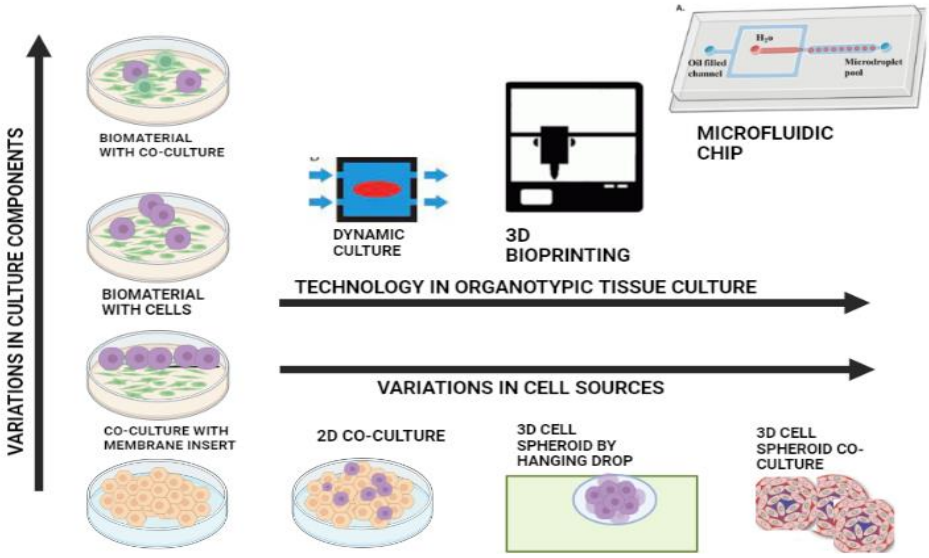

Figure.2 Advancement In Technology Toward The Development Of Organotypic Models.

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The simplest 3D system is done using hanging drop technique called cell spheroids; other techniques involve organic or inorganic matrix, substrates with minimal adherence coating, scaffolds supported with paper layer and cell levitation using magnet. These techniques are useful for maintaining differentiated cell states, anticipating drug penetration, and determining how cellular interactions affect drug fate[12]. For subsequent biochemical or cell investigation, homogeneous size, comparable gradient diffusion, and physiology of cells in micro-well plates are ensured by standardizing spheroid formation via high-throughput technologies. Contrary to the cumbersome traditional techniques of spheroid models, Ivascu and Kubbies created a quick approach for producing analogous spheroids from a single cell group within 24 hours[16]. These spheroids obtained from suspension culture from the breast cancer cell lines and other cell lines are possibly used in the screening of drug cytotoxicity and investigating expression of genes from oncological models. The use of automated HTS eliminates the problems with traditional cultures for evaluating drug ADMET(absorption, distribution, metabolism, excretion and toxicology) by enabling repeatability, accuracy, and sensitivity and quick readouts of tests[13-15]].

Individual cell cultures There has been a noticeable advancement in the fabrication of greatly yielding platforms for the manufacturing of analogous spheroids. The analysis demonstrated differences between 3D and 2D tumour, in identification of distinct components that impact the cell viability of breast and colorectal cancer cell lines, and in determining the impact of collagen producing cells on tumours[17,18]. Shuford et al. have detailed methodical and subsequent clinical evaluation of cell spheroids-tissue obtained from ovarian cancer patients for medication screening prior to treatment[19]. The successful forecasting of subject's physiological reaction in this study exhibits strong personalized prognosticative and remedial advice to combat ovarian cancer. In complicated malignancies with diverse cells and ECM, single cell microtissues are insufficient as models for drug evaluation[18].

Multicellular cultures or cell co-cultures Because it replicates the vascularized tumour micro-environment, cells that are co-cultured has become more common in drug screening against cancer. Ivanov et al. created biologically realistic cancer spheroids by using human medulloblastoma cell line that are co-cultured and cells of brain tissue obtained from human fetus[9]. They also examined the neurotoxicity and effects of several chemotherapy medicines. The tumor-host contact affects the physiological, chemical and molecular characteristics of the tumour, and thus paracrine(co-cultured) successfully imitated the interaction between healthy and malignant tissue. [14]Anil-Inevi et al. used stem cell line of the bone marrow and MDA-MB-231 cells to create a magnet induced levitation device for 3D cells to assemble on their own in situ in micro-gravity stimulation. For biomedical applications, the cluster size changeable 3D spheroids can be a suitable model. By sub-culturing tumour cells of pancreas thrice, connective tissues and endothelial cells, which are aggregated to form a central focus that is fibroblastic in nature with endothelial cells equally distributed throughout the spheroid[9,19], Lazzari et al. created a multi-celled tumour spheroid structure for pancreatic carcinoma. Because molecular cross-talk is so important to disease progression and therapies, the complicative micro-environment deduced the chemical sensitivity that represents subjects aversion to treatment and underscores the significance of heterogeneous physiological systems. Compellingly, Movia et al. created three dimensional multi-layer sub-cultures. MRC-5 cell line and A549 cells at the interface between liquid and air as the initial artificial instrument for evaluating the effectiveness of respiratory anti-cancer drugs that are inhaled[13]. The analysis effectively compared ALI multilayered co-cultures to monocultures to assess simultaneous resistance to these drugs docetaxel, cytarabine, vinblastine, and methotrexate (multidrug resistance).[20]

ORGANOIDS GENERATED FROM CELLS

The inability to grow large numbers of primary cells and keep them viable in vitro limits their usefulness in translational medical research. Over primary cells, stem cells have a number of benefits including infinite cultures in vitro, ability to be pluripotent, intercellular signalling pathways that are simulated, and generational inheritance of traits. In order to create precursor for pancreatic organoids with specific mutation in pancreal cells, Huang et al. employed human stem cells that are pluripotent. The cellular epigene-targeting drugs were evaluated on these organoid models, which kept their differentiated status and demonstrated biological changes and responses similar to real tumours[22]. This served as a precursor to find treatment approaches for pancreatic exocrine cancer. Colon-like organoids were created using Human induced Pluripotent Stem Cells (iPSCs) by Crespo et al. iPSCs were created using cells extracted from patients with familial adenomatus

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polyposis in association with mutation in the germline, and rapamycin, XAV939, and geneticin were tested on these cells. It was reported that the organoid system has the potential to be translated into precision medicine along with medication screening for colorectal cancer[21-22].

TECHNIQUES IN CULTURE ELEMENTS

To more accurately reproduce natural physiology and forecast therapeutic performance, 3D culture platforms are becoming increasingly complicated. In contrast to 2D culture hypersensitivity to chemotherapeutic medicines, recent techniques that aim to imitate 3D *in vivo* complexity incorporate microwells inserted with membrane for categorization and ECM like biomaterial components. A membrane implant for culture In order to sustain 3-D micro-tumours in separate microwells for drug screening both single and sequential,[24] Mosaad et al. used a mesh made of nylon in the microwell. With the newly designed HTS, 150 micro-tumors could be cultured in each well of 48-well plates, and the anticancer medication response could be measured. Two cancer cell lines androgen-associated prostrate cancer cells and prostrate stromal myofibroblast cells were combined to form micro-tumours. Two medications that are frequently prescribed for advanced prostate cancer were assessed for their effects on the micro-tumor response: therapy targeted on androgen using enzaltumide or abiraterone acetate; and taxane chemotherapy of either single or sequential dosage using docetaxel. [23]The three dimensional organisation of a tumour and its multi-cell complex nature have been recreated by Vorsmann et al. using an organotypic cultured complete skin analogous that contains predetrmined sizes of spheroids derived from melanoma tumour. Metastatic melanoma 451-LU cells were cultured using a hanging drop method, and melanoma spheroids with certain dimensions and cell counts were produced. In order to create the dermal compartment, ten 451-LU(a subline of WM164) spheroids were combined with primary fibroblast culture, initiated into collagen I in a cell insert, grown, and co-generated during differentiation of epidermis.[24] Significant differences in the therapeutic success between complicated *in-vivo*-like 3D models and 2D cell sensitization of melanoma towards TNF-related apoptosis-inducing ligand by simultaneous administration of sub-lethal dosages of UVB or cisplatin were observed. TRAIL + cisplatin treatment effectively killed skin-equivalent-embedded melanoma spheroids, whereas TRAIL + UVB treatment had essentially no effect, offering a possibility for the exploration of substantially relevant cytotoxic medicines and customized therapies. 3D structures made of biomaterials Any organotypic system's size is extremely important since it defines the facet: focal ratios that affect drug efficiency assessments. Spheroid models generate a three-dimensional environment for cell proliferation, but the mechanical and physical ECM component is absent[25]. As a result, biomaterials that have cells embedded in them in 3D structure have been created, providing an aligned and physiological micro-environment for a further accurate evaluation of cell response to medications[26-28]. Cancer designs that simulate the three dimensional micro-environment have been created employing cancer cells embedded in artificial scaffolds such Matrigel1, hyaluronic acid, and decellularized matrices. The successful creation of a PEG hydrogel micro-well array that produces homogenous tumour spheroids that are multicelled was described by[27] Lee et al. These structurally distinct, sturdy multicellular HeLa/ovarian cancer and HeLa/HUVECs 3D sphere constructs determined the efficiency of therapy with graphene oxide-wrapped gold nanoparticles and were an excellent tool for drug screening. Table 1 lists various 3D organotypic tissue structures made from biomaterials and cells, as well as the in vitro screening potential examined [1][71-78],87.

Table 1. Investigation 3d Tissue Mimics For Diverse Drug Evaluation And Biological Activity Assessment

CANCER MODEL MADE THROUGH ORGANOTYPIC CULTURE	MATERIAL SOURCE OBTAINED FROM BIOLOGICAL AND SYNTHETICAL SOURCES	CELL SOURCE	OBSERVED DATA	REFERENCES
Breast carcinoma	Rat's tail collagen 1	Myoepithelial cells surround the co-unit of a dual cell that contains MCF-7 cells.	It was investigated if MMP or HGF c-met signaling may be involved in the disruption of cellular structure caused by	[71]

			inhibitors.	
Cancer of the prostate and breast	Antherae mylitta's silk fibroin	LnCaP prostatic carcinoma cells and MDA MB-231cells from breast cancer	The anticancer medications MMPs analyze the effects of growth regulators and signaling sequences on matrices, cleaving the ECM-mimicking tumour.	[72]
Throat and head cancer	Matrices of collagen	Hypopharynx squamous carcinoma cells (FaDu) and fibroblasts from patients were co-cultured.	Radiation therapy had little to no impact, according to the Tissue Roll for the Assessment of Cell's Exposure and Response (TRACER) framework, which examined stromal interactions and tumor cell reaction to treatment.	[73]
Hepatocellular carcinoma(HCC)	polyglucuronic acids with phenol grafts	hepatoma cells from humans and fibroblasts from mice, human umbilical vein endothelial cells, hepatic stellate cells	Multi-cellular tumor spheroids (MCTS) showed tumor heterogeneity and clinically important pathophysiology variables for assessing cancer therapy.	[78,79],[87]
cancer of oesophagus	rendered free of biomaterial	minimal mesenchymal stem cells co-cultured with oesophagus cancerous cells from a tumour sample	Cancer was anticipated in cancer cells specific to the patient combined with mesenchymal stem cells to stimulate proliferation and chemotherapeutic dosage resistance in the 3-D designs, which allowed for the precise forecasting of chemosensitivity against models that are specific to the patients.	[77,78]
osteosarcoma	Alginate, gelatin, as well as carboxymethyl cellulose, among	osteosarcoma cells from human	To assess a new treatment and carcinoma, they were found to increase cell proliferation, biological	[87]

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	others.		compatibility, and collagen synthesis. Replicates tumour micro-environment	
Cancer of ovarian cells	Three-dimensional printing using collagen peptides-methacryloyl.	Ovarian follicular cell grown on spheroids	Offers conditions necessary for ovarian follicle formation in the scaffold, it might be utilized as a substitute method for follicular growth. Effective prediction of reactions specific to the patients for first-line chemotherapy	[76]
Vestibular schwannomas	Collagen- IV protein	NF1 PN and Schwann cells	In 2D and 3D cultures, the ability of the MEK inhibitor(selumetinib), the IGF-1R inhibitor picropodophyllin, and the BMP2 inhibitor LDN-193189 to limit the proliferation of cancer cells was examined.	[74]

ORGANOTYPIC CULTURE TECHNOLOGY

Organotypic tumour model bioprinting Regenerative medicine, cancer research, drug development, toxicology, and fundamental science all use 3D-bioprinted tissue models. The method has many benefits, including the capacity to create precise, dependable, and repeatable cell models for HTS and the ability to place biomaterials with exact spatio-temporal placement to obtain layer-on-layer printing[1,2,29]. In order to replicate both healthy and pathological tissue, the approach requires cell printing using biomaterials, extra cellular matrix elements, and various biochemical components. It operates on three different extrusion principles a)micro-extrusion, b)inkjet printing, and c)laser assistance. Inkjet printing uses printers that are drop on demand to print bioink on a predetermined appropriate substrate while creating digital information[10]. To produce the droplets from the ejector, a vapor bubble is created by electrically heating the nozzle with a piezoelectric actuator. A nozzle-free technique known as "aim and shoot" is used in laser-assisted printing to get over the drawback of bioink blockage. A better knowledge of disease causation, progression of disease, screening of drugs, assessment of drug ADMET, and the development of advanced cancer therapeutics is made possible by 3Dprinted models in vitro that replicate the morphological and biological features of actual tumours[30].

Model of bone metastases in breast cancer Zhu et al. designed a bone matrix which is 3D bioprinted using stereolithograph technology, HA(hyaluronic acid) nanoparticles, and PEG composites to simulate the tumor microenvironment. The MDA-MB-231 and MCF7 cells' cellular structure and migration on bioprinted models were similar, however, the 3D model with hydroxyapatite nanocomposites demonstrated greater migratory potential[11,10]. When cells were treated with 5-fluorouracil on three dimensional matrices, cell toxicity was reduced than in 2-D. Human cord endothelial cells, mammary fibroblasts, and an ER+ve MCF-7 cells were used in the bioprinting of breast tumour with stromal cells and core in order to study breast cancer invasion.[12]

BEZ235, a PI3K/mTOR inhibitor, was used for 3D bioprinting human breast cells (HC1143). This treatment decreased the S6 ribosomal protein phosphorylation across the stromal cells and cancer tissues. While 2D

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designs demonstrated a 50% reduction in Ki67-positive cancer cells, three dimensional designs did not demonstrate any form of discernible reduction in these cells[31].

Ovarian cancer Cervical tumour models in vitro have been created by 3-D bioprinting cervical cancerous cells using hydrogels. These hydrogels use fibrous proteins like fibronectin, collagen, or laminin to simulate the extra cellular matrix of the tumour micro-environment[87]. Protein expression of printed cells on tumour designs was unfold by proliferation of cells, morphological imaging of cells, and analytics, which exhibited a degree of resemblance to the natural cervical carcinoma environment. The three dimension and two dimension models showed elevated MMP-2 and matrix metalloproteinase 9(MMP-9) expression, similar to a native cervical carcinoma. Both models of chemotherapy with paclitaxel had uneven and loose morphology in the cell structures' cytoskeleton. The difference in chemotherapeutic impact between the two models was concluded by seeing the cells in two dimension culture floating away from the substrate's receptor while the 3D cell structure steroid remained stable and intact[31] [26].

Ductal pancreas adenosarcoma A stromal combination of human primary pancreatic stellate cells and HUVECs was printed with a xenograft of pancreatic cancer cell line derived from patient. Alpha-smooth muscle actin, which is a specific hallmark for tumour activation, was expressed by bio-printed cells. The tumour micro-environment's intrinsic and extrinsic signals were investigated using a 3D-printed model. In the examination of tumour physiology, micro-environment of tumour, and associated mechanical research, this model appears to be more pertinent[31].

Brain cancer The fatality of brain tumors or glioblastomas is increased by the widespread metastasis of brain tumor cells, higher recurrancy, and resistance to cancer medications. A 3D bioprinting of human glioblastoma SU3 cells were designed in order to better understand the glioma genesis, susceptibility to anti-cancer drug, and resistance to treatment. Temozolomide was used to study chemoresistance, and the results were compared to 2D models. A 3D bioprinting based on coaxial -extrusion has recently been used to recreate the in vitro glioma tumor micro-environment[32]. To explore the interactions between the stroma and the tumor, the bioprinted model created heterogeneous self-assembled microfibers in a multicellular heterogeneous milieu. In this study, mesenchymal stem cells and glioma stem cells were used to coaxially 3D bioprint alginate and gelatin[33]. Table 2 lists enlist the 3D models biprinted using organotypic tissue mimic for cancer treatment. [1,26,32,55,79-81,87]

Table 2. 3d Bio-Printed Cancer Models For Toxicity Testing And Medication Screening

BIO PRINTED DESIGNS	MATERIALS OBTAINED USING SYNTHETIC POLYMERS	CELL SOURCE	OBSERVED DATA	REFERENCES
Cerebral cancer	Gellan gum modified with RGD polypeptide.	Glioma cord cells and primary cortical neurons	Greater temozolamide resistance 400–1500 mgml1 compared to 2-D model. Neurodegenerative ailments were also evaluated.	[32]
Tumour of cervix	bio-inks made of hydroxyethyl cellulose with varying concentrations of sodium alginate	Hela cells	Extrusion printing with 3-D printers had no negative effects on cell survival. Cell growth, MMP protein expression, and drug resistance to chemotherapy were evaluated.	[26], [87]

Malignant tumour of breast	boronic acid-functionalized laminarin and alginate combined	MCF-7 cancer cells and other malignant breast cancer cells	Cellular spheroids are used for drug testing and replicate the structure of the tumour micro-environment. Up to 14 days after printing, cell viability could be maintained at more than 90%.	[79], [87]
Metastatic breast tumour model	Combining nanohydroxyapatite with gelatin methacrylate	Combined culture of breast cancer and stromal cells	The course of post-metastatic breast cancer is investigated using the interactive action of cells in an artificial bone micro-environment.	[55,80,87]
Hepatic carcinoma	Bio-ink made of collagen I and hyaluronic acid	The hepatic stellate cells and human primary liver cells	The metabolism of 7-ethoxy-trifluoromethyl coumarin, drug resistance against acetaminophen, and radiation resistance was investigated.	[81,87]
Skin	Gelatin, diethylaminoethyl cellulose, and alginate	Fibroblasts and keratinocytes	Drug toxicity and its effect on cancer. The 3D constructions showed improved collagen expression, higher cell viability, alongside additional skin-specific indicators.	[81]
Osteoblast precursor	Laminarin functionalized with boronic acid	Bone cells and bone marrow	cell viability could be maintained at more than 90% and developed precursors were able to replicate the micro-environment of tumour cells	[87]

Organ-on-a-chip with microfluidics Microfluidic-chips has revolutionized drug development by making drug discovery easier, more convenient, and quicker. Micro-reaction chambers for drug manufacturing finely regulate reactions in terms of both space and time, leading to larger surface-to-volume ratio, more hood flow, and an increases thermal transfer gradient[5]. On a single chip, several medication concentrations can be examined, and dual sides of a singular cell can be used to expose one side of the drug. Microtissues-on-a-chip include cells, extra cellular matrix, vascularization, and biomimics cell behaviour and native biological environment[7]. The main goal of an organ-one-chip is to create a system that functions as a single unit of the target organ and models a particular aspect of human physiology[37]. Figure 2 illustrates the various applications of microfluidic chips in disease diagnostics and drug screening.

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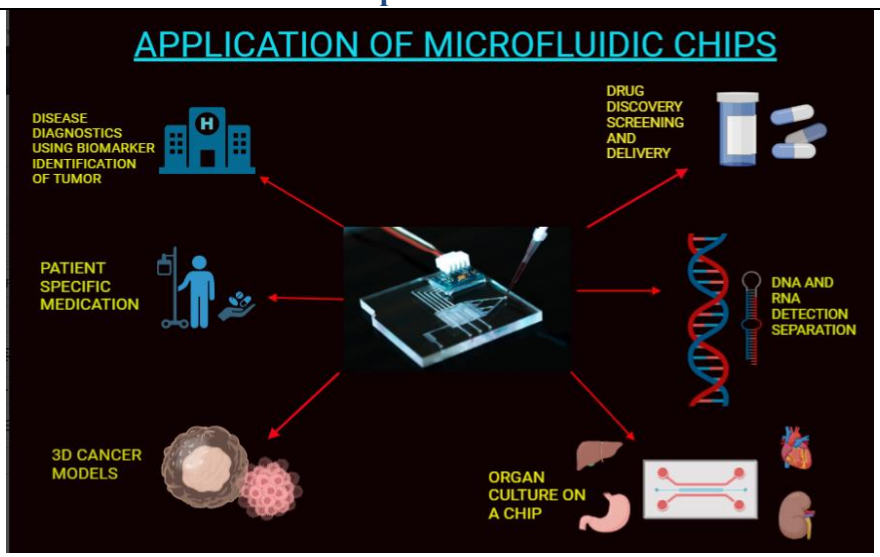

Figure 3. Various Applications Of Microfluidic Chips In Disease Diagnostics And Drug Screening

Chip model of intestine

The chip can be used to replicate the absorption and delivery of medications taken orally into the bloodstream. A minimal functional intestinal unit should produce a motion similar to peristalsis and a steady coating of mucosal intestine, which is crucial for drug absorption by the intestinal epithelium[38,39]. Low oxygen concentration and anaerobic microbiota that goes along with it are key aspects of the intestine. Human intestinal-microvascular endothelial cells and colorectal adenocarcinoma cells (Caco-2) were successfully cultivated on a chip that Jalili-Firoozinezhad et al. created. Additionally, a strain of intestinal bacteria called Bacteroides fragilis was added to the side of the epithelial lining of the intestine and also co-cultured for a number of days. These instruments aid in the development of drugs that incorporate the therapeutic role of the microbiota[41].

Chip model of liver

The functional unit of liver, sinusoid, is made up of various cell types. Its hepatic vessels and hepatic bile duct cells carry out a variety of tasks, including the elimination of bile, harmful substances, and oxygen[41,42]. By sub-culturing cells in a substratum supportive of cellular development, such as extra cellular matrix of decellularized liver, several ways have been developed to imitate these activities in a single chip. It is crucial to include a bile duct in a Liver chip model since this aids in the elimination of waste materials and bile acid. The production of albumin and urea has been greatly increased by the chip, and the IC50 data for medications such acetaminophen (APAP), tacrine, and chlorpromazine have been reached or nearly reached in vivo[43].

Heart-on-a-chip

Blood is pumped by the heart and serves as the vehicle for drugs. Physical aspects including blood flow rate, heart valve contraction should be taken into account when developing heart on a chip(HOAC) technology. Hydrogels and micropillar arrays can be used to create 3D matrices utilizing microfluidics technology[44-46]. Such layouts make it easier for cells to avoid direct contact with blood flow and receive vital nutrients by diffusion technique. Homogenous 3D scaffolds that co-culture endothelial and cardiac cells produced from iPSCs show patient-specific modeling. On a related HOAC, Sidorov et al. Assessed the contractility, mechanism potential, and 3D microtissue elasticity. These tools can be used to characterize a cardiac microtissue's electrical and mechanical properties[46]. A cardiac-like flow profile can now be produced by varying the closing timing of various pneumatic valves on the chip. Improved alignment of cell and on HOAC with increased aSMA intensity are the cardiac cells' responses to these simulated flow patterns. Because the heart is heavily vascularized, vascularization is another crucial characteristic that must be replicated in vitro. Through the use of microfluidic 3D stamping,[47-48] Zhang et al. created an angio-chip that included capillary-like channels and promoted the proliferation of endothelial cells. Cardiomyocytes combined with such designs will prove to be one of the best models for more accurately simulating in vivo conditions and predicting drug responses. The aforementioned microfluidic models will enable more accurate prediction of the cardiotoxicity of cancer chemotherapy medicines. [52]

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Lung-on-a-chip

The alveolar epithelium of the lung is a special type of tissue which is on one side is exposed to air while having a surface that is vascularised highly on the other. Additionally, the pleural parenchyma is constantly being mechanically stretched[50]. Expansion of the pleural parenchyma is crucial for preserving in vivo healthy cell activity. In the breathing microfluidic environment, human metastatic adenocarcinoma cells develop resistance to tyrosine kinase inhibitors in third-generation, but not in static cells. Numerous subjects with late-stage lung cancer showed a similar outcome[49-51]. In a model of on-chip IPF(Idiopathic Pulmonary Fibrosis), demonstration has been done for breathing behaviour to hinder the wound healing. Various designs have analysed the impact of medication using lung as route of administration and evaluated toxic levels by nanoparticles addition. Analogous models of other tissues of organs, including kidney, ovaries and components of reproductive system, pancreatic cells, arteries, have also been created using chip technology[48,51-54]. Tumour model on a chip Thriving growth of tumour cells with their capacity for progression and medication toxic level is necessary for tumour modeling on a chip. A vascular mimicing which delivers nutrition and transports drug to the cancer tissues and research metastatic mechanisms, is another crucial component. Microfluidics can produce dose-dependent cell killing by fabricating a concentration gradient production device[55,56]. Diverse techniques, such as the radial injection of a medication to the core of the malignant tumour made up of cancer cells of colorectal, have been researched to imitate in vivo. Cultures made of micro-3D with multicellular efficiently investigated stromal activation and tumor growth. It accurately depicted the variations in an abnormal tissue's ECM composition, architecture, and transport properties.[57] Gioiella et al. noted ECM remodeling in stromal tumour tissue that was similar to physiological condition in vivo by overexpressing hyaluronic acid and fibronectin. The capability of tumor cells to cause elevated fluid leakage in the presence of mononuclear cells(neutrophils) was revealed using tumor-on-a-chip (TOAC)[58,59]. When compared to traditional 2D cultures and spheroids of 3D produced from the ovarian cancer cell line SKOV3, these models provided more accurate predictions of anticancer treatment efficacy. Micro-dissected tumours from patients were loaded onto chips to enable person-specific designing and direct screening of drug. The tumors included four xenografts of mouse made from cancer cell lines of human, one from a subject with prostatic hyperplasia which is non-malignant and four from patients with prostate and ovarian carcinoma[60]. Several organs on a chip Drugs are given specifically for one organ interact with other organs by travelling via the circulation and reaching different tissues. Some illnesses affect multiple organs, such as lung cancer, and the simulation of the drug ADMET is possible on the same platform[63-66]. These platforms make it possible to simulate the single drug toxicity on multi-organs or the mimicking of specific disease-related organs. Lung cancer cell trans-endothelial migration via the blood-brain barrier can be effortlessly simulated on a chip device. In one analysis, media were sequentially passed tissue-tissue on the surface of a chip to simulate drug's mechanism and transportation of metabolites in vivo[67,68].

The toxicity values and the outcomes from the clinical trials agree. Organ cross-talk and media flow by on-chip programmable electromagnetic micro-actuators are among the sophisticated devices that runs without any need of exterior pumping mechanisms promised by recent advancements in microfluidics[69,70].

III. CONCLUSION AND FUTURE PROSPECTS

Models of cancer tissue made through organotypic technology are useful alternatives to animal models for managing the costs associated with drug discovery borne by the pharmaceutical sector. By building diverse 3D cancer mimicking in vivo structures, the inadequate prediction of a traditional chemotharapy treatment reaction has been significantly conquered. These biomimetic organotypic three dimensional models have drastically improved the way medicines are depicted and given more depth to basic research. With the development of current technologies like bioprinting and microfluidic devices, trials to closely recreate the tumour micro-environment appear to be feasible. Multidisciplinary bioprinting technology challenges call for knowledge from the fields of engineering, materials science, and biological science. In contrast, scaling up for HTS in microfluidics requires sophisticated technical advancements. In vitro tissue models have improved as a result of developments in high-throughput technologies and the creation of bioengineered regenerative constructs. However, there is still room for improvement in our knowledge about 3D tissue models because of their complex nature making simple duplication difficult and allows for the subsequent evaluation of additive therapy. Although it will be difficult, integrating organotypic constructions with physiological simulation in

microfluid-device has the potential to produce the most useful in vitro models. Future strategies should seek to identify patient-specific pathogenic pathways and forecast their response to chemotherapy in the oncoming era of customized medicine. For clinical derivation of 3D cancer tissue models to achieve predicted results of malignant tumour treatments, levelling up and cutting the rate of the procedure would be additional obstacles. These methods should become effective individualized diagnostics and prognostics for treating cancer.

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