

# ORGANOID AS A NOVEL TECHNOLOGY FOR DISEASE MODELING

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**Received:** 06.01.2021; **Accepted:** 08.02.2021; **Available Online Date:** 04.03.2021

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**Cite this article as:** Sever T, Cevik I, Calibasi-Kocal G, Basbinar Y, Ellidokuz EB. Organoid as a novel technology for disease modeling. J Basic Clin Health Sci 2021; 1: 94-101.

## ABSTRACT

The organoid technology is capable to create more real-like in vitro models in terms of structure and function of the origin of the tissue. Since the three-dimensional model is able to illustrate disease pathology, cell differentiation, and recapitulation of self-renewal, lead organoid technology as a promising disease model to fill the gap between conventional two-dimensional, and in vivo disease models. The review describes the recent development of organoid disease modeling approaches.

**Keywords:** Organoid, Disease Modeling, Stem Cell

## INTRODUCTION

Disease modeling via animal and cell culture techniques have been providing great contribution in explaining the mechanism of diseases and identifying potential curative approaches. However, there is still a significant lack of models that could show more physiologically and predictive relevant results. Two-dimensions (2D) cell cultures have been routinely and ardently used worldwide for the past decades. However, the 2D cell cultures are tenable ancient and do not reflect the physiology or anatomy of the regarding tissues for enlightening studies (1). Moreover, in vivo cancer models have some limitations such as consumption in time and resources, expensiveness, and interspecies differentiation (2).

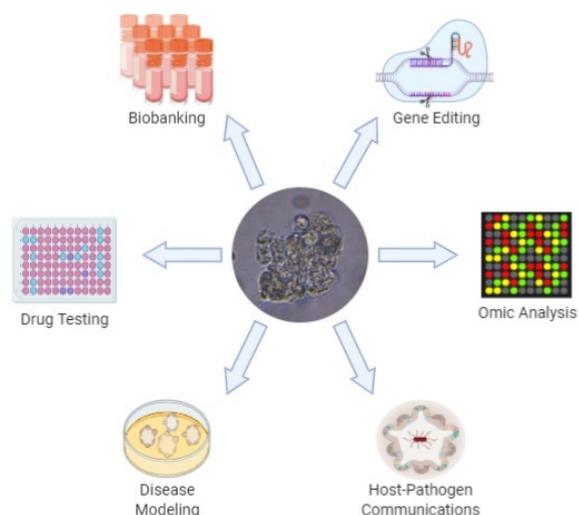
These limitations have prompted scientists to search for new models that better mimic the parental tissue.

Designing a three-dimension (3D) cell culture model requires multidisciplinary expertise and approach (3). Several 3D cell culture models have been studied up to now such as the spheroid, ex-vivo, and organoid. Organoids, as a novel and promising in vitro culture model, were generated via the self-organizing capacity of stem cells that could recapitulate the function, architecture, genetic signatures of the parental tissue (4). Despite the organoid technology has numerous advantages, the technology has several limitations. Development of vascularization, as one of the obstacles, is crucial for cell-cell and stromal-cell interactions did not occur except several studies in the xenograft model generation process (5). Other limitations of the technology are the lack of microbiota, for the gut model, and the immune system that has slightly solved via co-culture systems (6,7). Another obvious drawback of the technology is the

absence of interorgan communication since they mimic a specific part of the human body, not the entire. Thus, they could not recapitulate the micro-physiology of specific tissue or organ. Several studies have been already focused on the limitation. In a study, various organoids have been connected to investigate the interaction between the pancreas, liver, and gastrointestinal tract (8).

Organoids have been used to search for cancer, genetic disorders, and infectious diseases via directly patient-derived or gene editing of human stem cells. (9) Recently, human patient-derived organoids have been generated that provide a more accurate disease model. A large body of studies shows the great potential of organoid technology in drug screening, drug optimization, regenerative medicine, and personalized medicine (10).

Taken together these pieces of information, in this study, we reviewed the applications and advantages/disadvantages of organoids in several disease models.



**Figure 1.** The figure summarizes the application areas of organoid technology and created by using BioRender (<https://biorender.com/>).

### Organoid Modeling for Cancer

Up to now, vigorous efforts have been done in the cancer research area and important development has been reached in the treatment and diagnosis (11,12). Still, cancer indicates a major health concern globally due to the low life quality and survival of patients with cancer. Since poor mimicking of the parental tumor by traditional cancer models, the progress of efficient

therapy is one of the major obstacles which are working on these cancer models lastly fails in clinical practices. The lacks of conventional cancer models are microenvironment, stromal compartments, organ-specific function, immune system, genetic heterogeneity (2,13,14). The promising organoid technology bridges the gap between traditional in vitro and in vivo cancer models (15). Tumoroids have been generated from tissue biopsies, surgical resections, ascitic fluid, and circulating tumor cells (16–19). Tumoroids could be used for various downstream implementations since they could be propagated and passaged indefinitely (20). Tumoroids could be an important branch of organoid technology, such as genetic carcinoma, infection-cancer development, and mutation-tumorigenesis processes (21–25). Tumoroids have been generated and bio-banked from various types of primary and metastatic cancers as colorectal, pancreatic, prostate, lung, liver, ovaries, kidney, bladder, brain, cancers, etc. (26).

Nowadays, drug response analysis of patients and their matched tumoroids showed that drug responses are very parallel. In case of drugs did not affect the tumoroids, the matched patient was not affected, and drugs that displayed efficiency in tumoroids were matched by the patient response with 90% of cases. In several studies, this initial study has been found corroborated (27,28). An accelerated number of studies on patient-derived tumoroids, their molecular profiling, and usage in xenograft formation, may provide more reliable in vitro screening platforms for personalized medicine (29). Analyzed cancer types and sample sizes are limited. Thus, more stringent researches are necessary for routinely adopting patient-derived tumoroids as in vitro patients 'avatars'.

### Organoid Models for Neurodegenerative Diseases

Recently, human brain organoids, generally generated from PSCs, have been used in neuroscience to evaluate mainly neurodevelopmental processes and related disorders. In a study, PSCs-derived brain organoids have been utilized to illustrate transcriptional dysregulation and developmental malformations which take place in schizophrenia. Since the organoid technology has 3D brain conformations, the research group could have evaluated the disruptive organoid region of

**Table.1** Applications of organoid technology in disease modeling. The table includes studies about organoid modeling of cancer and genetic, neurodegenerative, and infectious diseases.

Disease Model	Organ	References
<b>Cancer</b>	Bladder	Mullenders et al., 2019(49)
	Colorectal	Yao et al., 2020(50); Schnalzger et al., 2020(51); Fujii et al., 2016(20); Drost et al., 2015(52)
	Breast	Yang et al., 2020(53); Griscelli et al., 2017(54); Sachs et al., 2018(55)
	Kidney	Hwang et al., 2019(56); Wang et al., 2017(57); Batchelder et al., 2015(58)
	Ovary	Nanki et al., 2020(59); Kopper et al., 2019(60); Hill et al., 2018(19)
	Lung	Shi et al., 2020(61); Kim et al., 2019(62); Dijkstra et al., 2018(63)
	Liver	Broutier et al., 2017(64)
	Brain	Bhaduri et al., 2020(65); Ballabio et al., 2020(66); Jacob et al., 2020(67)
	Prostate	Beshiri et al., 2018(68); Gao et al., 2014(18)
<b>Neuroscience</b>	Malformation	Dunnack et al., 2017(69)
	Microencephaly	Gabriel et al., 2020(70); Kelava et al., 2016(71)
	Autism/macrocephaly	Chan et al., 2020(72); Hohmann et al., 2020(73); Mariani et al., 2015(74)
	Alzheimer's disease	Papaspyropoulos et al., 2020(75)
	Parkinson's disease	Kim et al., 2021(76); Monzel et al., 2017(77)
<b>Genetic Diseases</b>	Familial adenomatous polyposis	Sommer et al., 2018(78); Crespo et al., 2017(79)
	Cystic fibrosis	Berkers et al., 2019(80); Dekkers et al., 2013(81)
	Alagille syndrome	Guan et al., 2017(82)
	Polycystic kidney disease	Freedman et al., 2015(83)
	Miller–Dieker lissencephaly syndrome	Bershteyn et al., 2017(84); Iefremova et al., 2017(85)
	Rett syndrome	Gomes et al., 2020(86); Feldman et al., 2016(87);
	Timothy syndrome	Sloan et al., 2018(88)
<b>Infectious Diseases</b>	Hereditary multiple intestinal atresia	Bigorgne et al., 2014(89)
	Brain	Watanabe et al., 2017(90); Gabriel et al., 2017(28)
	Intestinal	Lamers et al., 2020(91)
	Stomach	Bartfeld et al., 2015(92)

schizophrenic patients. The study suggests that organoid technology could be used for such disease models (30). The prenatal hypoxic injury cause early life neurological defects. Since the human corticogenesis evaluation model is lack, all consequences of hypoxia are still not known. The prenatal hypoxic injury cause early life neurological defects. Since the human corticogenesis evaluation model is lack, all consequences of hypoxia are still not known. Daviaud et al. reported the brain organoid model as a starting point to analyze new approaches in therapeutic to regenerate and protect affected cell populations in the neurodevelopmental process (31). Neurodegenerative disease modeling is also applicable as, Parkinson's disease, Fronto-Temporal

Dementia, Alzheimer's disease and Amyotrophic Lateral Sclerosis (32). On the other hand, brain organoids could reflect various sites of the brain such as the midbrain and neocortex. In order to evaluate more complex biological mechanisms in the human brain, Chen et al. generate an organoid model so-called assembloid which pre-patterned into a specific cite of the brain (33). As a result, brain organoids supply an interesting overview of various ways that could be utilized for neuroscience.

#### Organoid Models for Genetic Diseases

Recently revealed CRISPR–Cas9 endonuclease technology provides a genetic engineering method, that readily available to researchers (34). A large

body of studies performed the gene-editing system on human pluripotent stem cells (hPSCs) in order to generate specific mutated isogenic cell lines. Since different cell lines have strong phenotypical variations, these cell lines have been served as a crucial control for genetic analysis (35). CRISPR-Cas9 genome editing combined organoid technology enlarges the organoid applications in various ways. Genetic diseases modeled with organoid technology are cystic fibrosis, hereditary multiple intestinal atresias for intestine, familial adenomatous polyposis for colon, alagille syndrome for liver, polycystic kidney disease for kidney, microlissencephaly, miller–dieker lissencephaly syndrome, rett syndrome, timothy syndrome for the brain, enhanced s-cone syndrome, retinitis pigmentosa for the retina, and leber congenital amaurosis (36). As an example, in the Caucasian population, the leading lethal genetic disorder is cystic fibrosis (CF) that is a multiorgan disease including, pancreas, lung, reproductive tract, intestine, and liver (37). In a study of cystic fibrosis, the F508del in cystic fibrosis transmembrane conductance regulator (CFTR) gene that encodes CFTR protein, leading to misfolded CFTR channel protein, mutant human intestinal organoids that cause to fast degradation were corrected via CRISPR–Cas9. The edited CFTR amino acid sequence displayed a normal channel activity in vitro (38). The study clearly showed that targeted gene therapy could be another application of the organoid technology that has the potential for more accurate targeted gene therapy approaches. Taken together, organoids could be combined with other technologies to create better disease models.

### Organoid Models for Infectious Diseases

The organoid has been accepted as a promising technology for disease modeling that fulfills this expectation more day by day. One of the most important advantages of organoid technology is eliminating the interspecies differences when applied for disease modeling. Additionally, organoids include various cell types present in the parental tissue that could be altered by changing the medium compositions to have interested cell lineages (39). These advantages make the organoid technology amenable to study infectious diseases. Brain organoids are derived from human pluripotent stem cells that self-organize and recapitulate the in vivo fetal brain tissue better than 2D cell cultures in terms

of architecture and composition such as progenitor, glial, and neuronal cell types (40,41).

A myriad study has been focused on the Zika virus (ZIKV), a member of the Flaviviridae family, that transmit to the human being from mosquitos. It has been associated with microcephaly in newborns and possessed a crucial risk for pregnancy (42). The organoid technology has been applied to reveal the mechanism of the infectious since it can reflect the malformation of the brain driven by the ZIKV. In a study, ZIKV-exposed brain organoids show that favored infection of neuronal progenitor cells, which inhibits the proliferation and induces a decrease of cell viability that leads to reduced organoid size (43). Recent studies have been used brain organoid technology in drug screening to preventing and curing ZIKV infection (44,45).

Nowadays the coronavirus disease 2019 (COVID-19) has been affecting a vigorous number of humans. Despite COVID-19 mostly affects the lung, clinical data indicate a growth in both chronic and acute neurological symptoms such as meningitis/encephalitis and persistent fatigue (46,47). In a recent study, COVID-19 infected brain organoids illustrate that the virus damages the choroid plexus epithelium leads to leakage in the blood-brain barrier that prevents immune cells, pathogens, and cytokines into the brain and cerebrospinal fluid (48). Thus, organoid technology is crucial to study host-pathogen interactions via modeling infectious diseases.

### CONCLUSION

Already myriad studies have been declared highly efficient generation of various human diseases modeled with organoid technology. Despite several limitations of this promising and novel 3D technology, the application of the model for understanding the mechanism of diseases and evaluation of more precise therapy options are possible. In order to bring cumulative information produced in the lab to clinical practice fast, the technology is pointed out as a milestone. Still vigorous developmental progress is necessary for the model.

**Conflict of Interest:** No conflict of interest was declared by the authors

**Financial Disclosure:** This study was supported by Dokuz Eylul University, Scientific Research Projects (2018.KB.SAG.039).

**Peer-review:** Externally peer-reviewed.

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