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Research Article

Development of Organ-on-Chip for the Study of Placental Pathologies: A Ten-Year Study of Literature Published

Ravindra Kalode[1]*, Pranoti R. Kalode[2]

[1]MBBS, MD. (Radio Diagnosis) from IGMC, Nagpur. Assistant Professor at Dr. Panjabrao Deshmukh Memorial Medical College, Amravati, India
[2]MBBS, DGO. from Dr. Panjabrao Deshmukh Memorial Medical College, Amravati, Consultant at Kalode Hospital, Amravati, India

*Corresponding Author: Ravindra Kalode, MBBS, MD. (Radio Diagnosis) from IGMC, Nagpur, Assistant Professor at Dr. Panjabrao Deshmukh Memorial Medical College, Amravati, India. Email: drravindraniar@gmail.com

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Abstract

Context: The placenta performs a crucial function in nutrient exchange, but studying this tissue poses a number of challenges. Utilizing microfluidic and microfabrication technologies, a 3D placenta-on-a-chip model provides a biomimetic alternative for studying placental diseases and treatments.

Objectives: Aim: To review and analyze the currently available placenta-on-chip data to study placental pathologies in patients.

Methods: By systematically searching the PubMed, Scopus, and Science Direct databases, research papers that employed 3D printing techniques for the development of organoids and Organ-on-Chip (OoC) systems for in vitro experiments were gathered and scrutinized.

Results: When exposed to glucose transfer, placenta-on-a-chip mimics the features of an in vivo human placenta. Microchips have the potential to become a platform for diagnostic purposes for placental diseases and a model for duplicating the important features of these diseases.

Conclusions: The microfluidic placenta-on-a-chip platform holds promise as an affordable solution with versatile applications. However, research is essential to develop a comprehensive in vitro pregnancy model in the future to expand our understanding of feto-maternal communication.

Keywords: Placenta; Organ-on-chip; Engineering Technologies

1. Context

The placenta is essential for successful reproduction (1) and a vital organ that sustains the development of embryos. This organ provides essential nutrients and oxygen to the fetus while eliminating waste products such as xenobiotics, bacteria, viruses, parasites, and CO2 from the fetus (2). Inadequate placental growth can impact these functions, potentially leading to mortality and morbidity in both the fetus and the mother. Research has demonstrated that inadequate oxygen and nutrient transport through the placental barrier can result in undesirable outcomes for the fetus (3). Throughout pregnancy, the placenta and fetal membrane act as a defensive shield, comprising trophoblasts, connective tissue, basal lamina, and fetal endothelium. These unique anatomical features of the placenta are designed to regulate substrate exchange and prevent direct contact between maternal and fetal blood (4).

Inspecting the biological aspects of the human placenta poses a major experimental challenge. Various models, including in vivo, ex vivo, and in vitro, have been developed for studying the placenta (5). In vivo investigations on mice models are widely used as prototype systems, but even these systems harbor certain challenges. Because of the variations in placental anatomy, the placenta stands out as one of the most organ-specific structures among different species (6, 7). In order to advance basic and clinical research in the field of obstetrics, it is vital to understand the feto-maternal interface during pregnancy and parturition (8). In ex vivo models, the main challenges are those related to the setup of placental perfusion, obtaining consent from female participants for taking part in research, and resolving ethical concerns (9).

To overcome these challenges, in vitro models mimicking in vivo conditions, such as Organ-on-Chip (OOC) platforms, came into light. These platforms are developed using bioengineering and microfluidics-based approaches,

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providing a better understanding of in vivo functions and responses of the placenta, which is expected to move this field forward significantly (8). Using Placenta-on-a-Chip micro devices, human trophoblasts (JEG-3) and human umbilical vein endothelial cells (HUVECs) can be co-cultured on a thin extracellular matrix (ECM) membrane to simulate a physiological placental barrier in vitro (1). Two layers of human cells are embedded in a flash-drive-sized device that replicates the interface between the mother and the fetus. These microfluidic channels on either side of the interface allow researchers to study how molecules are transported through or are blocked by these routes.

2. Objectives

In this study, we sought to analyze the currently available data on the applications of the placenta-on-chip technology for the study of placental pathologies.

3. Methods

3.1. Literature Search

We systematically and thoroughly collected all research studies from the PubMed, Scopus, and Science Direct databases, which employed 3D printing techniques to create organoids and Organ-on-Chip platforms for in vitro investigations. This was achieved by using a combination of the following keywords, as well as the Boolean operators of AND and OR to combine them: "bio-printing," "rapid prototyping," "3D printing," "organ-on-a-chip," and "placenta on the chip."

3.2. Eligibility Criteria

For study selection, we defined certain eligibility (i.e., inclusion and exclusion) criteria. Specifically, the inclusion criteria revolved around the following factors: (1) Articles that were published between 2010 and 2020; (2) research investigations utilizing 3D printing and/or bio-printing methods in the context of Organ-on-a-Chip devices; and (3) articles that their full-text was available. Meanwhile, articles of the following types were excluded: (1) Abstracts, (2) non-English literature, (3) review articles, systematic reviews, or meta-analyses, (4) book chapters, (5) brief communications, (6) conference proceedings, (7) patents, and (8) case reports.

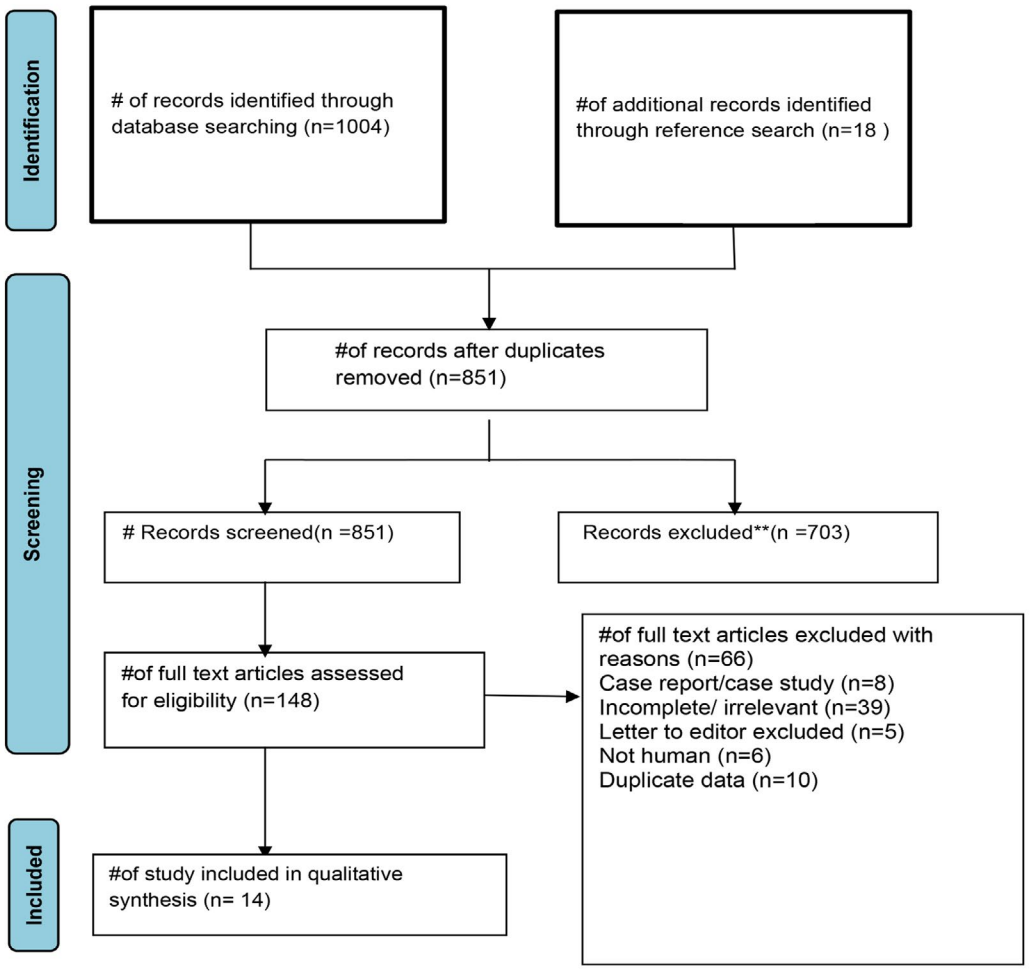

Figure 1. The PRISMA flow diagram for systematic reviews. Our search was conducted in the mentioned databases and registries

4. Results

The initial search strategy yielded 1004 articles (Figure 1). Subsequently, manual screening uncovered an additional 18 articles. We eliminated 153 duplicate articles. Following the screening of 851 articles based on their titles and abstracts, 703 articles were excluded. This left us with 148 articles that underwent a full-text assessment to determine their eligibility. Out of these, 137 were excluded for various reasons, as detailed in Fig. 1. Ultimately, 14 studies spanning the past three decades met our criteria for full-text review, and all 14 were subsequently included for further analysis. Statistical analyses were conducted using Review Manager Version 5.3 (The Nordic Cochrane Centre, Copenhagen, Denmark).

We here reviewed the literature published over a ten-year period on the progress of organ-on-chip platforms for studying placental pathologies. The results reveal that this technology holds huge promise for providing accurate and reproducible models for studying the complex interactions between the placenta and the maternal-fetal environment. The use of organ-on-chip platforms has resulted in notable progress in our comprehension of placental physiology and pathophysiology, as well as conditions such as preeclampsia, intrauterine growth restriction, and gestational diabetes. These models have the potential to be used for screening responses to treatment and expanding personalized medicine in the future, ultimately improving the health of pregnant women and their fetuses. However, further research and validation are needed before these models can be fully integrated into clinical practice.

4.1. Studies' Characteristics

The parameters tested using the placenta on-chip technique vary in different studies, including drug transport across the placenta, the rate of glucose transport, health risks associated with nanoparticles, caffeine transport, IgG antibody transport, the effects of environmental toxins, preterm birth, and amnion membrane cells transition. Out of 16 studies included, 3 were conducted in the USA, and one study was from each of Switzerland and England. The rest of the studies included did not mention the region where the study was conducted. Table 1 summarizes these studies' characteristics.

Table 1. Placental Parameters Investigated Using the Placenta-on-chip Technology

First Author	Year	Country	Parameters Tested	Results
Antoine Malek et al. (10)	2003	Switzerland	Transport of immunoglobulin G and its subclasses	The human placenta establishes a dedicated transport system for IgG in both the fetal and maternal directions.
Villano K et al. (11)	2007	NA	Maternal transport of interleukin-6	An ex vivo perfusion model demonstrated minimal transfer of interleukin-6 from the maternal to fetal side.
Jisoo Lee et al. (1)	2015	NA	Glucose transport	Regarding glucose transport, placenta-on-a-chip mimics the features of a natural human placenta. The microchip had the potential to become a platform for disease-related diagnostics and for duplicating the important components of placental diseases.
Cassidy Blundell et al. (12)	2016	USA	Drug transport	Glyburide, as a representative medication, was capable of replicating the inherent function of efflux transporters and imitating the restricted transfer of a drug administered to the mother using a placenta-on-a-chip model.
Juan Gnecco et al. (13)	2017	England	Preterm birth (PTB) and premature rupture of membrane (PROM)	Subclinical infections such as chorioamnionitis were the factors responsible for PTB and PROM.
Zhu Y et al. (14)	2018	NA	Bacterial infections	Bacterial infections triggered the activation of inflammatory cytokines in maternal channels, promoting the stimulation of maternal macrophages.
Rajeendra L Pemathilaka et al. (15)	2019	USA	Caffeine transport	Caffeine concentration on the fetal side increased until it reached a steady-state condition.

Fangchao Yin et al. (16)	2019	NA	Impact of TiO2 nanoparticles	Production of reactive oxygen species was observed.
BabakMosavati t al. (2)	2020	NA	Rate of glucose transport	Glucose diffusion rate increased with membrane porosity and decreased with the elevation of the flow rate.
Patrick Schuller et al. (17)	2020	NA	Nanomaterial risk assessment	Particles with diameters ranging from 100 to 200 nm could be absorbed by the placental barrier via endocytosis, potentially leading to inflammation in both the placenta and fetus, fetal growth restriction (FGR syndrome), and fetal infections.
Marta Cherubini et al. (18)	2021	NA	Placental dysfunction, infections, and maternal-fetal toxicology	Placental function and material exchange at the maternal-fetal barrier were assessed.
Sungjin Kim et al. (19)	2021	USA	The effects of an environmental toxin (cadmium)	Cadmium exposure led to maternal cell death and some degrees of fetal cell death, triggering inflammation in both maternal and fetal cells.
Lauren Richardson et al. (8)	2019	NA	Interactions between cells and paracrine communication between maternal and fetal cells throughout pregnancy and the process of childbirth.	Membrane permeability, oxidative stress, toxin-induced senescence, and cytokine production were analyzed.
Lauren Richardson et al. (20)	2019	US	Amnion membrane cells' transition and migratory properties under interactive environmental conditions	Cells from the amnion membrane had the capability of transitioning and moving through microchannels filled with type IV collagen.

5. Discussion

The placenta is the organ that functions as a biological barrier during human fetal development. The human placenta is regarded as a crucial organ that sustains fetal development throughout the gestational period (21). A healthy and perfectly functional fetal placenta facilitates the effective circulation of oxygen and nutrients, waste removal, metabolite production, and immunity against diseases, infections, and xenobiotics (18). Recently, in vitro models of the human placenta have substantially helped understand the mechanism of the placental transport of materials. For re-creating the physical microenvironment of an organ, the tissue-tissue interface, and blood perfusion, as well as studying human biology, organ chips offer a powerful new tool (22).

The in vivo placental barrier is a complex structure comprising layers of trophoblastic and fetal endothelial cells, with a basement membrane separating them. As pregnancy advances, this villous membrane gradually becomes thinner, reaching an average thickness of around 4.53 μm at full term (23). Placental barrier function is critically dependent on appropriate intercellular junctions in the intervillous space and fetal capillaries to transmit substances from maternal blood to the fetus.

The "Placenta-on-a-Chip" device carries viable human cells that replicate the essential fetal-maternal interface found within the villous tree of the human placenta. Trophoblasts and Human Umbilical Vein Endothelial Cells (HUVECs) are co-cultivated within a compartmentalized three-dimensional PDMS microsystem. This system comprises upper and lower cell culture chambers divided by a thin vitrified collagen membrane. This setup enables the replication of the crucial features of the placental barrier (1).

In a study conducted by Cassidy Blundell et al., they utilized the BeWo trophoblastic cell line and demonstrated that the transport of glyburide across the placental barrier could be analyzed using the placenta-on-a-chip technology. Placenta-on-a-chip was capable of reproducing native efflux transporters and mimicking inadequate placental transport of maternally administered drugs (12).

BabakMosavati et al. designed a microfluidic-based placenta-on-a-chip platform that included a polycarbonate membrane and two polydimethylsiloxane (PDMS) microchannels to assess glucose transport across the membrane. The findings indicated that in the co-cultured cell model, the rate of glucose diffusion was lower in comparison to both the monoculture and the cell-free microdevice. Numerical simulations were conducted to explore the impact of other factors, such as the flow rate and membrane porosity, on glucose transport through the placental barrier model. The findings revealed that

as membrane porosity increased, the rate of glucose diffusion also increased, while higher flow rates predicted a lower rate of glucose diffusion (2). These results were in accordance with a study conducted by Shah et al., who analyzed the transport of glucose and the presence of glucose transporters in the human choriocarcinoma cell line (24). Antoine Malek et al. explored the active transport and passive diffusion mechanisms of small molecules, as well as the transport of immunoglobulin G and its subclasses using a dual in vitro perfusion approach, and the transfer of substances across the human placenta in an isolated cotyledon. The results demonstrated that all 4 immunoglobulin G subclasses could be transported across the human placenta from the maternal to the fetal side. (10) A similar technique was used by Schneider et al., who studied the transfer of amino acids across the placenta (25).

Pemathilaka et al. were the first to report caffeine transport across the placenta using a placenta-on-a-chip model. They studied the rate of caffeine transfer across a micro-engineered placental barrier. The results revealed that on the fetal side, caffeine concentration increased until reaching a steady-state condition (15). In a recent study, Zhu et al. created a micro-engineered model simulating the human placental barrier on a microchip and examined the inflammatory reactions linked to bacterial infections. Their findings revealed that bacterial infections could trigger inflammatory cytokine release in the maternal channels, which in turn promoted the activation of maternal macrophages (14).

Schuller et al. developed a lab-on-chip system for nanoparticle risk assessment in placental BeWo trophoblastic cells and concluded that nanoparticles with diameters ranging from 100 to 200 nm could be absorbed by the placental barrier through endocytosis, which might lead to placental and fetal inflammation, fetal growth restriction (FGR) syndrome, and fetal infections (17). A similar lab-on-chip model was used by Yin et al. to study the effect of nanoparticles on placental barrier function (16).

Kim et al. investigated the effects of an environmental toxin (cadmium) and found that cadmium induced death in maternal cells and some fetal cells, accompanied by the inflammatory activation of these cells (19). Lee et al. analyzed glucose transport across the placenta using a placenta-on-chip model, and their results showed that with regard to glucose transportation, placenta-on-a-chip mimicked the features of the natural human placenta (1). Microchips have the potential to become a platform for disease-related diagnostics and a model for duplicating the important components of placental diseases. Yin et al. examined the impact of TiO2 nanoparticles on the placenta, demonstrating the production of reactive oxygen species (ROS) (16), which can be associated with the progression of numerous inflammatory diseases, as stated by Mittal et al. (26).

Richardson et al. assessed an organ-on-chip model of the feto-maternal interface by analyzing cell-cell interactions between maternal and fetal cells and the exchange of signaling molecules during pregnancy and childbirth. They evaluated membrane permeability, oxidative stress, toxin-induced senescence, and cytokine production (8). Gnecco et al. assessed preterm birth (PTB) and premature rupture of membrane (PROM) and showed that subclinical infections such as chorioamnionitis were associated with these complications (13). In another study conducted by Richardson et al. using a placenta-on-chip model, they showed that amnion membrane cells had the capacity to undergo transitions and migrate within microchannels filled with type IV collagen (20). Overall, this systematic review revealed that placenta-on-chip models are advantageous for studying the placental transport of various substances, which can aid in studying placental pathologies.

5.1. Conclusions

The microfluidic placenta-on-a-chip platform exhibits the potential to function as an economical platform with a broad spectrum of applications in various scenarios, offering fresh opportunities for studying the transport of substances across the placenta. This microdevice can serve as an in vitro model for research on nutrients' transportation across the maternal-fetal interface, thereby contributing to the comprehension and management of diseases. Although organ-on-chip technologies have been increasingly applied to the field of obstetrics in recent years, substantial research is essential in the future to establish a comprehensive in vitro pregnancy model to enhance our understanding of feto-maternal communication, the mechanisms underlying term and preterm labor induction and the factors governing the permeability of drugs and toxicants across these critical interfaces.

Authors' Contribution:

Dr. Ravindra Kalode: Conceptualization, formal analysis, resources, manuscript writing and reviewing, data collection and interpretation, methodology. Dr. Pranoti Kalode: Writing and reviewing the manuscript.

Conflict of Interests:

No competing interest was reported.

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Data was made available by the corresponding author.

Ethical Approval:

Not applicable

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