HYBRIDA

D5.1: Operational guidelines regarding organoids and organoid-related technologies

Project title: Embedding a comprehensive ethical dimension in organoid-based research and related technologies
Project acronym: HYBRIDA

Hervé Chneiweiss, Ioana Andreescu, Bernard Baertschi, Anne Dubart-Kupperschmitt, Jean-Luc Galzi, Jacques Haiech, Corinne Sebastiani

Grant Agreement no.: 101006012
Lead contractor for this deliverable

Coordination and Support Action
H2020-SwafS-28-2020

HYBRIDA

Deliverables factsheet:

Project Title:	HYBRIDA
Title of Deliverable:	Operational guidelines regarding organoids and organoid-related technologies
Work Package:	WP 5
Due date according to contract:	M 40 (final version)
Authors:	Hervé Chneiweiss, Ioana Andreescu, Bernard Baertschi, Anne Dubart-Kupperschmitt, Jean-Luc Galzi, Jacques Haiech, Corinne Sebastiani
Contributors:	Heidi Beate Bentzen, Mylene Botbol, Emma Capulli, Céline Cougoule, Margherita Daverio, Christine Dosquet, Mette Lindhart Falkenberg, Vincent Flacher, Maxence Gaillard, Xavier Gidrol, Alexei Grinbaum, Soren Holm, Théo Karapiperis, Meriem Karkar, Panagiotis Kavouras, Stefan Krauss, Cécile Legallais, Jonathan Lewis, Tenneille Ludwig, Jeantine Lunshof, Maxime Mahé, Ester Maland, Christine Mummery, Charles Pence, Mario Picozzi, Laurent Poulain, Tine Ravn, Daniel Reumann, Mads Sørensen, Eleni Spyrakou, Henrik Vogt
Keyword List:	Organoids, ethics, reproducibility, trust, transparency, ethics-by-design, conceptual uncertainties, biobanks, adult stem cells, ESCs (embryonic stem cells), iPSCs (induced pluripotent stem cells), protocols, Good Manufacturing Practices (GMP), evaluation and assessment, informed consent, Donor's Tissue Research Under Secure Transparent Ethical Donation (TRUSTED)

Consortium:

	ROLE	NAME	Short Name	Country
1.	Coordinator	University of Oslo	UiO	Norway
2.	Partner	National Technical University of Athens	NTUA	Greece
3.	Partner	University of Manchester	MAN	United Kingdom
4.	Partner	Université Catholique de Louvain	UCL	Belgium
5.	Partner	Aarhus University	AU	Denmark
6.	Partner	Leiden University Medical Centre	LUMC	The Netherlands
7.	Partner	French National Institute for Health and Medical Research	INSERM	France

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

8.	Partner	University of Insubria	UNINS	Italy

Revision history:

VERSION	DATE	Revised by	Reason
1.0	15.10.2022	WP5	Preliminary
2.0	20.04.2023	HYBRIDA Consortium	Work in progress
3.0	30.06.2023	WP5	Work in progress
7.0	30.07.2023	HYBRIDA Consortium	Work in progress
8.5	15.04.2024	WP5	Final Draft
8.8	10.07.2024	WP5	Final version

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

1 INTRODUCTION 5
1.1 Preamble 5
1.2 Executive summary of HYBRIDA's operational guidelines 7
1.3 The HYBRIDA Project 16
1.4 Aims and Scope of HYBRIDA Operational Guidelines 17
2 METHODOLOGY 18
2.1 Concepts and Values 18
2.2 World Health Organization: Choosing ethical values and principles 19
Informing how decisions are made 19
Informing what decisions are made 20
2.3 A matrix to design our operational guidelines for responsible research on organoids and related technologies for researchers and evaluators 21
2.4 Ethics by design and RAD approaches: from ethical principles to practical solutions 22
3 ORGANOIDS SPECIFIC ETHICAL ISSUES 24
3.1 Organoids: an evolving concept 24
3.2 Use of organoids: the identification of four categories 25
For research 25
For bioproduction 25
For preclinical use 25
For clinical use 26
3.3 Organoids and specific issues needing ethical review 28
4 EVALUATION CHECKLIST FOR SCIENTIFIC AND ETHICAL STUDIES USING ORGANOIDS 30
4.1 MINIMUM INFORMATION ABOUT ORGANOIDS AND THEIR USE (MIAOU) FOR RESEARCHERS 31
4.2 Evaluation Checklist for Organoid Experimental Studies (EChOES) 32
5 RICOCheck for International Review Boards (Research Ethics Committee, Research Integrity Committee) 35
6 ORGANOIDS AND INFORMED CONSENT* 37
6.1 Legal capacity to consent 38
6.2 In what contexts does the collection of biological samples take place? 40
6.3 Providing information beforehand and what to consider during the consent process 42
6.4 Construction and use of organoids 43
6.5 Typologies of consent 46
6.6 Anticipating donor's issues: the TRUSTED questionnaire for Tissue Research Under Secure Transparent Ethical Donation 51
6.7 Withdrawal of consent 53
6.8 Need for a taskforce to further analyse the new forms of consent required for research on organoids and related fields 54
7 OPEN ETHICAL QUESTIONS 55
7.1 Embryonic models ethical questioning 55

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

7.2 Nervous system functions: organoids and assembloids and their relationship with sentience, pain, consciousness 60
7.3 Naming of organoids 64
7.4 Ethical and Legal Ownership of Organoids: A complex Landscape 66
8 CONCLUDING REMARKS AND RECOMMENDATIONS 70
9 GLOSSARY 74
10 REFERENCES 88
11 ANNEXES 92
11.1 Annex 1: INSERM team 92
11.2 Annex 2: Ten conceptual uncertainties pertaining to the ontological status of organoids as hybrids 94
11.3 Annex 3: What is behind the "ethics by design" requirement? 95
11.4 Annex 4: SIENNA's Methodological Steps for Ethics by design 105
11.5 Annex 5: How these operational guidelines were drawn up within the framework of the HYBRIDA project 106
11.6 Annex 6: Working groups for the design of MIAOU and EChOES 109
11.7 Annex 7: MIAOU Full questionnaire 111
11.8 Annex 8: EChOES full questionnaire 118
11.9 Annex 9: Working group for RicOCheck design 125
11.10 Annex 10: RICOCheck full questionnaire 126
11.11 Annex 11: Winsconsin-Madison supporting organisation, WiCell, policy for use of cell lines 133
11.12 Annex 12: Organ-on-Chip versus Organoids-on-Chip 135

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

1 INTRODUCTION

1.1 Preamble

Biomedical research is a collective effort. It depends on the contributions of numerous individuals including basic scientists, clinicians and patients who work in a variety of institutions including the academic sector, industry and government agencies, within a framework of national boundaries with different social and cultural beliefs and regulatory systems. Ethical principles and operational guidelines help to secure the foundations for this collective effort. These are essential to building trust among the different actors. The scientific knowledge thus obtained can then be developed using reliable data. Public institutions and private firms can invest in research programmes generating trustworthy preliminary results, knowing that public and institutional support will be forthcoming for the foreseeable future. Patients can enrol in clinical research in the knowledge that these studies are well justified and the risks and burdens are reasonable in terms of their potential benefits.

The present document is proposed within the framework of the European-funded project HYBRIDA, in order to contribute to developing trust among the various actors in research on organoids and related fields. It is accompanied by the European Code of Conduct regarding research integrity in this field. Under the Horizon2020 programme, the HYBRIDA project seeks to establish ethical guidelines for organoid-focused research. As the project involves philosophical and regulatory uncertainties, it is formulating a framework to address these challenges and build trust among stakeholders. The guidelines have been laid down to ensure that all research with organoids is conducted in an ethical and scientifically responsible manner.

In recent years, research on organoids and related fields has emerged as an important area of biomedical science. As for any emerging technology and emerging novel object of research, a precise and consensual definition of organoids still needs to be elaborated. As a minimum, an organoid is an organised cluster of cells generated *in vitro* from different types of stem/progenitor cells (either pluripotent (embryonic or induced), or derived from adult tissue) through the use of 3D tissue culturing methods. Being composed at least in part of organ-specific cell types, such entities might serve as "three-dimensional culture models" that mimic the structural and functional properties of different organs – both human and non-human – such as the retina, heart, brain, intestine, kidney, pancreas, liver, inner ear or skin. We have chosen here to define organoids as

> "Stem or progenitor cell-derived 3D structures that at much smaller scales can re-create important aspects of the 3D anatomy and multicellular repertoire of their physiological counterparts and that can recapitulate at least some basic tissue-level functions."[1]

Organoids have potential applications in various areas of biomedicine, including 1/ development: finding conditions that enable observation of the initiation and growth of a given organ *in vitro*; 2/ physiology/physiopathology: mimicking a given function of an organ in order

[1] Rossi G, Manfrin A, Lutolf MP. Progress and potential in organoid research, *Nature Reviews Genetics*, 2018, 19(11):671–687. Accessible at: 10.1038/s41576-018-0051-9.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

to understand its physiology/physiopathology and permitting pharmacological and toxicological studies; 3/ production: mimicking a given function of an organ to produce a molecule of therapeutic interest; 4/ therapy: producing a living structure that can rescue an organ deficiency in the context of regenerative medicine. Architecture and function(s) are therefore essential features of an organoid as a research and/or clinical counterpart of its related organ. Like all other medical innovations, basic and translational research in the field not only requires a sound scientific rationale, but also needs to take into consideration both ethical, legal and social norms.

To build the framework for operational guidelines, we used several sets of converging basic principles, the first arising from the ALLEA Code of Conduct for research integrity: • Reliability in ensuring the quality of research, reflected in design, methodology, analysis and the use of resources. • Honesty in developing, undertaking, reviewing, reporting and communicating research in a transparent, fair, full and unbiased way. • Respect for colleagues, research participants, society, ecosystems, cultural heritage and the environment. • Accountability for the research from idea to publication, for its management and organisation, for training, supervision and mentoring, and for its wider impacts.

The second set of principles followed the 2021 recommendations of the WHO expert group on the governance of human genome editing: To inform how decisions are made - openness, transparency, honesty and accountability, responsible regulatory stewardship, responsible stewardship of science, responsible stewardship of research resources - and to inform what decisions are made – inclusiveness, caution, fairness, social justice, non-discrimination, equal moral worth, respect for persons, solidarity and global justice.

The third support document used is the guideline from the International Society for Stem Cell research (ISSCR), a non-profit organization which has been working on diverse aspects of human material manipulations that allow organoid progenitors to be obtained. These guidelines serve as premisses of the work carried in this document and were used to define ethically relevant categories of organoids

The HYBRIDA Operational Guidelines aim to help researchers and their recommendations are designed to ensure reliable research, development and production work on organoids and related technologies, as well as advice on assessment and evaluation. Such a document is intended to build trust between scientists through 1- Assurance of the quality of all materials and methods and of their use and application, in order to maintain the integrity, validity and reproducibility of any work conducted; 2- Documentation of the information necessary to track the materials and methods used, to enable reproducibility and ensure that the target audience will understand and evaluate the work; 3- Establishment and maintenance of adequate measures to protect individuals and the environment from any potential hazards; 4- Provision of relevant and adequate education and training for all personnel so as to promote high quality work and safety. Furthermore, the guidelines will support the work of research ethics committees and associated integrity bodies, and address the concerns and challenges experienced by participants who are involved in organoid research studies.

On-going progress of knowledge on organoids will require periodic updates of this document, so that the current Operational Guidelines will support efforts made to develop an appropriate nomenclature for organoids and related technologies, with the aim of “facilitating progress and

This project has received funding from the European Union’s HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

improving communication with the scientific community and the public" [2]. It is therefore anticipated that the Guidelines will probably be updated every 3 to 5 years at the most.

It is important to note that the Operational Guidelines, which mainly serve as a quality management process for the production, characterization, and use of organoids within the scientific community, are distinct from a Code of conduct. The latter fosters trust between scientists and society by promoting responsible research that takes into account the potential social impacts and implications of research objectives and technological advancements. Continuous dialogue between the code of conduct and the operational guidelines is essential to achieve a comprehensive approach to ethical issues. Clearly, this requires a multidisciplinary approach and the engagement of all stakeholders.

The compilation of these Operational Guidelines was a collaborative work involving numerous actors representing the full landscape of organoid research and its potential regulation (see Annex 5 for details of the steps involved in compilation and the various experts who contributed). This resulted in a bottom-up process with multiple discussions among HYBRIDA's partners designed to stabilise the present document and its developments through web-available questionnaires that will remain in a European-funded repository after completion of the HYBRIDA project.

1.2 Executive summary of HYBRIDA's operational guidelines

1. What are organoids?

Organoids are products of biotechnology that emerged in the early 2000s, building on decades of research on the potential of human cells to proliferate and renew themselves, even outside the body. Intuitively, "organoid" appears to describe an entity with similarities to an organ in terms of its cellular composition and/or has a similar architecture that reproduces at least some of the features and functions of an organ. However, the term is more generally used by scientists to refer to a family of entities involving various types of natural or engineered stem cells of healthy or pathological origin. Organoids differ from conventional cell cultures in that the cells can self-organize into three-dimensional complex structure which share some of the anatomical and functional properties of developing organs. Organoid research has been accelerated by (i) overall advances in stem cell research and (ii) innovations in culture medium reagents and devices that make three-dimensional expansion possible, consistent and reproducible between laboratories, whilst capturing physiological tissue functions.
Here are some examples of entities (among many) currently referred to as "organoids" or related to organoids:

- Intestinal organoid: intestinal biopsies collected from patients can be cultured *in vitro* such that small three-dimensional structures composed of stem cells adopt the shape of an intestinal crypt. They can be expanded indefinitely and used to study normal gut physiology or to better understand intestinal diseases in patients.

[2] For further details, go to the article by Pașca, S.P., Arlotta, P., Bateup, H.S. et al., A nomenclature consensus for nervous system organoids and assembloids, *Nature*, 2022. Accessible at: https://doi.org/10.1038/s41586-022-05219-6.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

- Neural organoid: skin cells – or other cells of the body – are reprogrammed to induce pluripotent stem cells and then differentiated into neural tissue that can resemble different parts of the immature nervous system. How the tissue self-organizes, or the different forms of neural cells, can provide insights into brain development or neurodevelopmental pathologies.
- Tumoroid: cancer cells taken as a biopsy from tumours in patients can be grown *in vitro* such that they are a near replica of the original tumour – the (personalised) tumoroid can then be used to test drugs and enable predictions on whether a drug will be effective in treating the patient or not.
- Embryonic model: pluripotent stem cells can differentiate *in vitro* such that they replicate some aspects of early embryonic development rather than a specific organ. This type of laboratory model may help to understand why miscarriages occur, or improve *in vitro* fertilisation procedures, etc.
- Organoid-on-chip: stem cells or organoids can be introduced into engineered microfluidic devices made of plastic or silicon (the 'chip') that supply them with nutrients, oxygen or drugs under conditions of fluidic flow, or with flexible substrates that resemble real tissue.
- Assembloid: organoids from different cell types or tissues are grown together (or assembled). For example, a brain organoid might be connected to a muscle organoid to mimic the innervation of muscle tissue, or gut and stomach organoids are coupled to mimic the gastrointestinal tract. These models do not focus on a specific organ only but can provide insights into global, physiological or pathological processes in the body.
- Chimera: namely a mixture of cells originating from two different species. In the case of organoids and related fields, after being grown *in vitro*, human organoids can be transplanted into animals to enable further development and physiological integration including vascularization. This has mostly been reported for neural and kidney organoids. Transplantation aims to mimic normal physiological conditions in tissues resulting in an improved maturation of the human organoids. A main ethical issue is to know if such transplant confers human properties to the engrafted tissue. In example [3], neural organoids

  Engrafted into injured rat visual cortex in rats survived for up to 3 months, formed afferent and efferent connections with the host visual network, and responded to visual stimulation.

2. What are organoids used for?

Potential applications of organoids range from fundamental biology (understanding developmental mechanisms) and disease modelling (understanding how diseases develop) to their use in drug discovery and therapeutic protocols for (personalised) clinical application in medicine.

More specifically, organoids are, or will be, used in:

- *Basic research*. Organoids as models of development offer windows into the physiology and pathology of an organ. They are useful for basic research in developmental biology and to understanding the mechanisms underlying diseases.

[3] Dennis Jgamadze et al., Structural and functional integration of human forebrain organoids with the injured adult rat visual system, *Cell Stem Cell*, 2023. Accessible at: https://doi.org/10.1016/j.stem.2023.01.004.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

- *Preclinical research on new therapies*. Preclinical research tests potential therapies for their effectiveness prior to clinical trials in patients. Organoids can be used to determine whether disease phenotypes are reversed, to verify toxicity and whether the treatment affects metabolism, etc. Models based on human stem cells could eventually replace some animal models in assessing effectiveness and toxicity.
- *Clinical use*. Organoids are increasingly being used as tools for the selection of personalised treatments in the clinic, but could also offer a source of biomaterial for regenerative medicine, for example for transplantation as Advanced Therapy Medicinal Products (ATMPs).
- *Bioproduction*. Organoids could be engineered and used for the production of biomaterials; for example, the production of viruses for vaccines and for gene therapy that is already of clinical utility.

With regard to the interests of patients, the most important distinction is between research (fundamental or biomedical) and immediate clinical applications (such as personalised medicine, where a single use model is created for each patient) or bioproduction to enable access to expensive drugs by lowering production costs. The vast majority of organoids being developed in laboratories today are used for research. Their clinical applications are still under development, although they are being trialled in some advanced research settings.

Basic research	Developmental biology Disease modelling
Preclinical research	Drug development and controls
Clinical research	Personalised treatment screening Material for regenerative medicine
Bioproduction	Production of molecules, proteins, viruses for treatments and vaccines, etc.

3. Why do organoids warrant special attention from the public, bioethicists and regulators?

Beyond the variety of entities that fall under the umbrella term “organoids” and their many applications, there remains uncertainty regarding a precise definition of the term: what does it actually mean “being similar” to something? Does an organoid become an organ or does it stop short of this? Current organoids are not “mini-organs” or off-the-shelf spare parts for failed organs, as it is sometimes postulated in the popular media. Researchers generally refer to them as ‘models’, because they are essentially biotechnological avatars built to study specific tissue features of interest. They resemble the phenomenon or object of interest to some extent, but cannot be conflated with it. If the model is not an organ, what is its precise nature and how should we relate to it? Organoids are typically hybrid entities: both biological, alive and made from living material, *and* artificial and technologically human-made.

As products of an emerging technology of an uncertain, hybrid nature, organoids could fall within regulatory gaps if, for example, practical, moral or legal issues are not satisfactorily covered, addressed, anticipated, answered or overcome by existing legal instruments or legally binding definitions. Organoids and related technologies might, however, be subject to over-regulation, for instance if current regulations are such that there is uncertainty as to which laws

This project has received funding from the European Union’s HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

apply, if applicable laws give rise to conflicting legal requirements or if there is a lack of appropriate regulatory harmonisation across EU Member States.
A series of workshops within HYBRIDA identified several issues raised by the public that will need to be addressed at some point: whilst there was general support for biomedical research and innovative technologies to find new treatments and improve public health, informed consent and responsible governance were highlighted as concerns throughout the deliberations. Addressing these is essential to safeguard ethical and acceptable use. Proper guidelines for organoid research are required to ensure that any economic interests do not prevail over patient safety and consent and prevent exploitation. The commercialisation of organoids is essential to enabling their implementation in biomedical and preclinical research, but it is a sensitive area, with participants expressing concerns regarding use, ownership and remuneration. These points need clarification in donor consent forms (see below for a discussion of the impact of uncertainty on donation and consent) and consent practices, while bearing in mind that contemporary biomedical research on organoids should not increase existing healthcare inequalities. Potential misuse and a breach of the privacy of personal data in connection with data storage and donation is also another concern.

4. How to promote responsible research with organoids and related technologies?

To ensure integrity in science and respect for the interests of all stakeholders, researchers need to comply with general quality standards regarding scientific integrity and related codes of conduct within their institution and country of origin. Many procedures are already in place within the biomedical research framework to ensure the ethical conduct of research, especially when this involves human material, including human stem cells. Most researchers are familiar with these prerequisites, which also apply to organoid research
Some research on specific organoids introduces new uncertainties: this may relate to sensitive cell types such as germ cells and gametes being formed, or, as noted above, brain-like structures. Guidelines are one way of coping with uncertainty, and especially regulatory uncertainty. The proposed guidelines aim to build trust among the various stakeholders involved in organoid research by ensuring that scientific knowledge is based upon reliable data and simple tools are available to assess the different aspects of research proposals. Guidelines will encourage researchers to report data and metadata in a standardized format that will clarify the methods and purposes of the organoid research to be carried out. These guidelines will also assist funding bodies or other evaluating committees, as well as ethics committees, in ensuring that the research is reported in a commensurable way. Guidelines cannot replace law and will leave some ethical issues open (see section 6) as they are generally subject to a certain degree of interpretation and provide flexibility when considering unforeseen and rapid developments.

4.1. A series of requirements to ensure the quality of reporting on research involving organoids

To enable a real assessment of the quality of research reporting, each batch of organoids should be associated with standard information (metadata). This includes a description of the tissue/cell sources, procurement protocols, the validation and conservation of raw materials, the protocols and databases used, as well as the culture and differentiation protocols and quality control criteria adopted by each level of organization such as biobanks. In this way, researchers receiving organoids can rely on the data that describe and characterise the organoid (structural data: omics; morphological data: imaging and functional data). Further, the metadata should

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

include all regulatory aspects that have been complied within the relevant jurisdiction, noting that the recipient researcher may be subject to different regulations, in order to avoid ethics dumping: material transfer agreements with provisions for the use of the organoids, a prior verification of patient consents, authorisation from the regulatory agencies for organoids created from patient cells, declarations of collection and of material transfers if applicable.
We call this standard set of requirements the **Minimum Information about Organoids and their Use for Researchers** (**MIAOU**). This focuses on the following: the origin of biological material (including informed consent from cell donors), efficacy/reproducibility, the quality of results (size, morphogenesis, cell composition), reliability, genetic integrity, the minimisation of communication errors (an accurate and documented description of the materials and methods), compliance with safety, security and research integrity rules, the prevention of research misconduct and miscommunication with the lay public. A template questionnaire is provided and will be implemented as a user-friendly webpage.
A second checklist mirrors MIAOU: this is intended for the scientific bodies in charge of assessing research proposals on organoids and related technologies. Its goal is to facilitate the work of scientific committees charged with evaluating proposals concerned with building, characterising and using organoids. This **Evaluator checklist for organoid experimental studies** (**EChOES**) describes how to evaluate the quality of organoid descriptions in a grant application in terms of the reproducibility, replicability and rationality of the proposed research. To assess the quality of an application, some elements are mandatory for scientific evaluation, while the others are contextual (for example, depending on the call requirements or the application domain). It is up to the evaluators to judge whether the responses are acceptable regarding a given project.

4.2. A practical guide for Ethics Committees

The Research Integrity Committee Organoid checklist (**RICOCheck**) intends to provide a tool for Research Ethics Committees (RECs) and Research Integrity Offices (RIOs) that will ensure transparency and anticipate ethical issues. Several principles need to be considered by RECs and RIOs, such as data confidentiality, the societal impact of the research project and its anticipated results, the approval of patient associations and the fair and responsible behaviour of Ethics Committees involved in evaluating projects that use organoids.
The research should be based on privacy-by-design, incorporating privacy safeguards at all steps of organoid research. Donors and/or the general public should be substantially involved in the RECs and RIOs, and societal benefits and any potential harm to donors, patients and society should be anticipated.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

5. How to act responsibly when applying organoids and related technologies in the clinic

The clinical applications of organoids are still emerging at the research stage. As such, potential applications are still being explored as a prelude to examining clinical efficacy. In this regard, particular efforts should be made to collect properly controlled evidence, to avoid publication bias that could create hype, and after any successful proof-of-concept studies to move to registered clinical trials. All clinical research outcomes, even those that are negative, should be documented, in line with the FAIR principles. Particular attention should be paid to how best to collect clinical evidence: how does information on a patient-derived organoid relate to the clinical phenotypes manifest in the patient? What are the relevant clinical outcomes? In the case of n-of-1 trials (e.g. when research is conducted in a single patient because of a rare genetic mutation) how should these outcomes be documented?

General efforts should also be made to ensure standardization. As in basic and preclinical research, standardization includes the reproducibility and replicability of experiments, different organoids from the same donor, organoids with the same disease from different donors, the same experiments carried out in different labs and how and if the technology works. Without well-documented procedures on how to grow and use organoids, and how to report their use, etc., it will not be possible to generalize findings or to build robust drug discovery pipelines. Failure to standardize procedures associated with organoid production and characterisation has ethical consequences. The value of patients' donations will be undermined if the results cannot be generalized in an honest, transparent and responsible way, and this will hamper trust between scientists, clinicians and the general public. Furthermore, standardization will be essential to future clinical use: clinicians will require scalable, safe and good manufacturing practice (GMP) products, available at a reasonable cost, in order to enable general access to innovative therapies.

As the clinical applications of organoids are only just emerging, researchers should be wary of creating high expectations among patients and other stakeholders, sharing only a general perspective of what might be possible in the years to come once certain scientific hurdles have been overcome and treatments have been fully validated by evidence. It is of the utmost importance for researchers to refrain from exaggerating prospects regarding the clinical use of organoids; that is, creating 'hype' or an ideology of promise. Precision in communication is essential: clinical prospects should be properly delineated and placed within a reasonable timeframe. Hype is not only relevant in communicating with the general public but is also of importance in reviews written for the scientific community. Obstacles to progress are not always explicit or are even obscured. For instance, practical difficulties in growing organoids (success rates, time, labour, etc.) constitute obstacles to all applications. Not reporting these difficulties might persuade the reader – even a researcher (scientist or clinician) or a funder – to expect outcomes of the technology to be closer to clinical application than justified. A first step towards more responsible communication is to be explicit regarding all known limitations and hurdles, and particularly feasibility, and to remember that clinical standards might be higher than those sufficient for proof-of-concept in research.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

6. Are some organoids more problematic than others?

So far, we have used two distinct strategies to classify organoids. One classification is based on its intended use (organoid destination: basic research / preclinical research / clinical application / bioproduction) while the other is based on organoid type (i.e., the type of organ or physiological process to be modelled). In this regard, some organoids and some potential applications raise more serious concerns than others and they may call for different levels of reflection and regulation. A piece of tissue extracted from a tumour and developed into a tumoroid will be unlikely to raise the same ethical issues as induced pluripotent stem cells turned into entities that model early human embryos. During the past 3 to 5 years, collective discussions have emerged regarding the status of the following organoid-related technologies:

- Embryonic models: advances in three-dimensional stem cell culture, as in organoid technology, have provided new opportunities to study the development of embryo-like structures *in vitro*. Scientists can produce models of human embryos made from embryonic or induced pluripotent stem cells to study the early stages of development. Depending on how this is done, these entities may share some features of natural embryos. Although embryonic research is highly regulated, the status of these embryonic models is unclear in most jurisdictions. These models have been inappropriately called 'synthetic embryos' by some authors, but they are neither synthetic, since they are made of stem cells, taking benefit from their spontaneous auto-organisation properties in given conditions, nor embryos, because such entities do not develop as an embryo. Furthermore, for human models of embryos it is widely considered as unethical to even try to implant them in a uterus. However, it is important to consider future developments of embryonic models. Even if current models are not equivalent or even close to viable embryos, the technology may advance such that they become increasingly closer to their natural counterpart; it is therefore clear that these entities will undoubtedly deserve further moral consideration.
- Organoids of the nervous system, and assembloids composed of various neural organoids: the nervous system supports cognitive function, and although the molecular and cellular bases of cognition are unknown, it is possible that cerebral organoids or similar entities could develop a form of sentience or consciousness, although no consensus yet exists regarding the definition of these two terms. Given the current state of the art, there is instead broad consensus among biologists and neuroscientists that the possibility of consciousness in organoids is negligible at present. However, in the longer term, this may change as organoids and assembloids become increasingly complex, or they develop as chimeras (as cerebroids transplanted into the brains of animals). The tools necessary to assess this possibility still need to be developed by cognitive neuroscience; the normative status of these artificial and sentient entities would need to be discussed, alongside the criteria applied to detect sentience and consciousness.
- Naming: An organoid does not have the same properties and functions as an organ. It is therefore incorrect to refer to organoids as mini-organs, and communicating incorrect information constitutes misconduct. Misnaming is both prejudicial for the advancement of science and for the trust of the public. The HYBRIDA project recognizes the effort of the scientific community to raise a consensus for an adequate nomenclature, for

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

example for nervous system organoids and assembloids[4], proposes a simple set of guidelines for naming that is rooted in developmental neuroanatomy. We anticipate that this effort will facilitate communication and scientific advancement, promote collaborations, and initiate the development of quality control measures and benchmarking in the field.

For the reasons mentioned above, these specific organoids call for wide-ranging debate on their status and regulation. Depending on scientific advances, *in vitro* models of embryos and the nervous system might fall into a regulatory gap in years to come. As models start to better simulate their natural counterparts, general design principles and values will not be sufficient to ensure ethical behaviour. Collective discussions on the status of these artificial entities will be essential. Such discussions are ongoing in bioethics regarding embryo models and brain organoids, but models of the reproductive organs (gonads) and germ cells also need to be taken into account.

7. Do organoids change anything about informed consent?

Organoids are relatively new entities that can be derived routinely in most laboratories from donor tissue. The HYBRIDA consortium reviewed different models of informed consent. Proper informed consent regarding the deposition and use of human biomaterials faces two specific challenges when it comes to organoid research. First, given the unknown future direction of research, it is impossible to anticipate all the potential information and uses that might derive from a given biological sample. Second, integration of the material collected in biotechnological constructs that are shared among laboratories makes it difficult to envision how donors would be able to withdraw their consent, even though it is one of their fundamental rights as stipulated in the Declaration of Helsinki.

In order to address these two specific challenges, one solution might be to use dynamic consent, which provides for continuous information to pass between the donor and users on new projects, allowing the donor to agree for the reuse of their biological samples. This solution involves obtaining initial consent from the donor for the collection and storage of their biological samples in a biobank, and continuous information which requires intense logistics regarding implementation. Because the issue of re-contacting donors poses practical and ethical problems, an alternative might be to adopt the consent for governance model where consent and agreements for the reuse of biological samples would be entrusted to an independent third party representing the donor.

In addition, the donor should be further informed in anticipation of the potential use of their donation so that they can allow or decline specific uses. HYBRIDA is thus proposing a **Donor's Tissue Research Under Secure Transparent Ethical Donation** (**TRUSTED**) that anticipates the conditions of use of biological samples according to the donor's preferences. Donors should complete a questionnaire at the time of donation as part of the consent procedure (see procedure page 41) which also specifies the restrictions they would like to impose on any future use of their tissues, trusting the initial researchers and the secondary users and if necessary with the support of a third party to ensure that these wishes are respected. The TRUSTED would facilitate implementation of the use of biological material, while respecting both the donor's participation and the researcher's investment. We believe that future European

[4] *Ibid*., p. 7.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

regulations regarding informed consent lie beyond the scope of HYBRIDA but we nonetheless call for European action based on our analysis in order to compile appropriate and updated regulations.

8. Recommendations

The HYBRIDA Operational guidelines conclude on 7 groups of recommendations that emphasize the need for a holistic and anticipatory approach to the ethical challenges of organoid research, integrating ethical considerations throughout the research process, and engaging with a broad range of stakeholders to ensure that organoid technologies develop in a way that is socially responsible, inclusive, and aligned with human values.

Anticipate and Address Ethical Concerns Proactively: Stakeholders in the field of organoids are called to employ an ED/RAD framework to proactively anticipate ethical concerns, including the social implications of organoid research. MIAOU/Echoes should be used for these purposes.

Incorporate Responsible Research and Innovation (RRI) Practices: RICOCHECK should be used

Ensure Continuous Ethical Engagement: Maintain ongoing ethical dialogue among researchers, stakeholders, and society, from the initial stages of research to all conceivable applications.

Implement Ethical Reflection: Combine RAD methodology with ED to allow for swift adaptations as ethical considerations and scientific knowledge evolve.

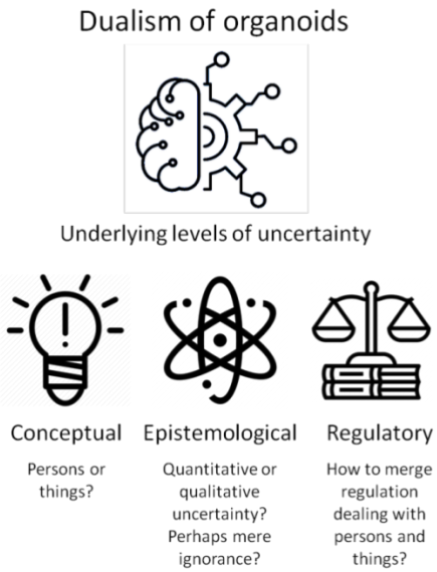
Foster Public deliberation and transparent regulation over Organoid Development: Advocate for transparent oversight and regulation over the development and application of organoid technologies including a Public Advisory Committee for Organoid Research inside an existing agency.

Facilitate Ethical Literacy and Education in the field of organoids: Develop educational resources and training programs to enhance ethical literacy among organoid researchers, stakeholders, and the broader public. This includes fostering an understanding of the ethical dimensions of organoid research and the importance of ethical design principles.

Commit to respecting the informed consent process: including the "TRUSTED" questionnaire, enabling donors to explicitly authorize or prohibit specific potential uses and reuses of their biological material and data. Entrust the Public Advisory Committee for Organoid Research, mentioned above, to assess various consent forms and define the most appropriate consent options, as well as the modalities and consequences of a possible withdrawal of consent for all parties.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

1.3 The HYBRIDA Project



HYBRIDA is a 3-year project funded by the Horizon2020 framework programme. The main aim is to build a comprehensive ethical dimension for organoid-based research and resulting technologies[5]. It aims to address three types of uncertainties regarding organoids:

First, ***conceptual uncertainty*** **(*ontological uncertainty*)**: How should entities be perceived that cannot be categorised as either persons or things? What *are* they? How do we *know* the characteristics of these entities called organoids?

Second, ***epistemological and methodological uncertainty*****:** How can we address forms of uncertainty that cannot be evaluated using statistical methods, i.e. risk assessment? This is particularly pertinent when organoids are intended for personalised or precision medicine, where the number of research subjects with a certain characteristic is too small to enable randomised controlled trials or other statistically based experiments. As precision medicine and new technologies emerge, evidence-based medicine is being challenged to determine new foundations. There are two types of epistemological uncertainty which can be categorised as qualitative, or strict, uncertainty and ignorance or non-knowledge. Qualitative, or strict, uncertainty is where possible positive and negative outcomes can be identified in advance but, unlike risk assessments, the statistical magnitude of each possible outcome cannot be estimated. By contrast, ignorance or non-knowledge is forms of uncertainty where neither possible outcomes nor the statistical magnitude of each can be identified in advance. In order to develop ethically and socially robust ways of assessing the effects of organoid research and related technologies, it is necessary to include these additional forms of uncertainty in Health Technology Assessment (HTA).

Third, ***regulatory uncertainty***: This uncertainty emerges because parts of the regulatory frameworks concerning the rights and duties of persons have been merged with elements of regulation dealing with the stewardship of objects or things. These forms of uncertainty are of particular importance.

HYBRIDA is focusing on how these three types of uncertainties are applicable in organoid research and is developing a conceptual and regulatory framework that can overcome this dualism between persons and things. From this follows the need to communicate the potential and possible pitfalls of organoid research in ways that convey realistic, rather than hyped, scenarios.

[5] The description of HYBRIDA in this section is taken from the project description (HYBRIDA Consortium, 2020, p. 2).

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

1.4 Aims and Scope of HYBRIDA Operational Guidelines

Operational Guidelines regarding organoids and organoid-related technologies are designed to streamline certain working procedures according to best practices[6]. These recommendations should be open to a certain degree of interpretation and provide for flexibility in the event of unforeseen circumstances and for the rapid development of this blooming field.

Further, the HYBRIDA Code of Responsible Conduct for Researchers provides ethical standards of good practice to guide institutions and researchers in the field of organoids and organoid-related technologies, in compliance with the principles of the European Code of Conduct (ECoC): Accountability, Honesty, Reliability and Respect. Both these documents are intended to enhance the existing ethical and normative frameworks: they represent normative foundations for the field of organoids and organoid-related technologies, should reflect HYBRIDA's objectives and should reflect the degree of risk and the forms of uncertainty that society is willing to accept.

[6] In view of the complexity of this field, related technologies (such as chimeras, cloning, organ-on-chips and organoid-on-chips, etc.) will be addressed in future versions of this document.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

2 METHODOLOGY

To address ethical issues in a scientific field – in our case organoids and related technologies – it is useful to define a number of concepts. The elements presented below result from interactions among HYBRIDA partners and with our colleagues. The procedures that were followed are presented in the annex section for the sake of clarity (see Methodology in chapter 11).

2.1 Concepts and Values

Our discussions, and the drafting of the present Operational Guidelines and the future Code of Responsible Conduct for Researchers, stem from three statements formulated according to the work developed by the French philosopher Paul Ricœur[7] and his famous consideration of ethical goals "Une vie bonne, avec et pour autrui, dans des institutions justes" (literally: A good life, with and for others, in fair institutions). We have translated this into three statements relative to scientific integrity, research ethics and professional conduct, respectively:

1) (The desire to) conduct honest and reliable research
2) Loyalty to oneself and to others
3) Fair institutions (which develop governance that promotes honest, upright, fair and accountable research).

We therefore have three compasses to help define our behaviour as actors in a scientific field (in this case, organoids and related technologies) in a given context (cultural, social, individual). These compasses are associated with a set of values that we can or want to render operational in the form of decision-making procedures (principles) or instructions to be followed (standards).

We thus addressed the following issues:

1) How to allow of researchers in the field of organoids and related technologies to be a "good researcher and a good person" in order to build a "good society"? This last question constitutes the foundations for the ethics of virtues[8], a reflection that can be traced back to Aristotle. Very schematically, collective and individual reflections are distinguished in order to guide our behaviour and thus build a good society. All of these questions come under the heading of research on ethics (the field of meta-ethics).
2) How can the used values be made operational?

a. According to a top-down approach that minimises the contextualization of our behaviours by establishing universal norms based on values, and/or principles,

b. According to a bottom-up approach that maximises the context and maintains the values so that they constitute the safeguards of decision-making procedures or by transforming them into principles that will be the heuristics of our behaviour.

We can thus distinguish two axes of ethical reflection:

[7] Paul Ricœur, *Soi-même comme un autre* **[***Self as another***]**, Éditions Seuil, Paris, 1990.
[8] Louisa Yousfi, L'éthique des vertus selon Aristote, Nicolas Journet éd., *La Morale. Éthique et sciences humaines*. Éditions Sciences Humaines, 2012, pp. 96-99. Accessible at: https://www.cairn.info/la-morale--9782361060312-page-96.htm.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

✓ Deontological ethics and teleological (or consequentialist)[9] ethics
✓ Top-down ethics (normative) and bottom-up ethics (ethics of practices[10], ethics of care).

2.2 World Health Organization: Choosing ethical values and principles

Considering that ALLEA values are at the core of the European code of conduct and the document ORF-CoC companion of the present guidelines, only values developed at the level of WHO will be further presented here. In the framework for its *Expert Advisory Committee on Developing Global Standards for Governance and Oversight of Human Genome Editing*, the World Health Organization recommends the implementation of two ongoing processes when selecting values and principles applicable to emerging technologies in life sciences and health. Both processes are related to decision-making: social values and principles take the lead on *how* decisions are made and *what* decisions are made[11]. Inspired by this identification of universal values for genome editing, we attempted to apply them more closely to the field of organoids and related technologies.

Informing how decisions are made

1. Openness, transparency, honesty and accountability

A commitment to openness that encourages collaborative ambition and work, as well as a commitment to use transparent, honest and accountable processes in order to generate and share evidence-informed, accessible and timely information on: (i) the best available data (including information on sources of funding, access and outcomes); (ii) guiding ethical values and principles; and (iii) actionable policy options for organoids and related technologies.

2. Responsible regulatory stewardship

A commitment to support and promote legitimate, evidence-informed: (i) laws and regulations; (ii) programme management and measurement; (iii) data collection, storage, processing, distribution and destruction in accordance with established privacy constraints; (iv) research training and capacity-building; and (v) public awareness regarding the potential benefits, harms and limitations of organoids and related technologies in ways that counterbalance competing influences and demands.

3. Responsible stewardship of science

A commitment to: (i) pursuing rigorous, evidence-informed basic and applied research with appropriate caution with respect to uncertainty and risk; (ii) following established ethical practices for research involving humans, with particular attention to issues of integrity and conflicts of interest; (iii) maximising the potential benefits of research while minimising the potential harms; and (iv) respecting research ethics guidelines and applicable legislation. More

[9] Éthique téléologique, *Philosophie*, 10 October 2020, Accessible at: https://delphipages.live/fr/divers/teleological-ethics. For further information regarding the differences in values, principles and norms, please refer to Annex 6.
[10] *Ethical Place*. Ile de France Region. [Espace éthique. Région Île-de-France]. Accessible at: https://www.espace-ethique.org/ressources/article/de-lethique-vers-la-pratique.
[11] World Health Organization. *WHO Expert Advisory Committee on Developing Global Standards for Governance and Oversight of Human Genome Editing*, p 27-28. Accessible at:https://www.who.int/teams/health-ethics-governance/emerging-technologies/expert-advisory-committee-on-developing-global-standards-for-governance-and-oversight-of-human-genome-editing.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

particularly, a commitment to align the processes and outcomes of organoid and related technologies research with the values, needs and expectations of society, as identified through participatory approaches involving various publics.

4. Responsible stewardship of research resources

A commitment to use finite research resources responsibly when choosing between research options for organoids and related technologies. This requires careful attention to scientific value and validity, as well as social value and validity. Finite research resources include: (i) biological materials; (ii) research skills; and (iii) research funding.

Informing what decisions are made

1. Inclusiveness

A commitment to carefully consider knowledge and perspectives on organoids and related technologies informed by different social, cultural and religious beliefs and moral values, as well as different skill sets. Furthermore, a commitment to ensure that organoid research (basic and applied) and clinical care are representative of global human diversity and are globally accessible.

2. Caution

A commitment to exercise appropriate caution given existing uncertainty and risk. This uncertainty and the balance of potential harms and benefits will be substantial in trials on organoids and related technologies[12].

3. Fairness

A commitment to fair dealings in the pursuit of research on organoids and related technologies and clinical care with individuals, organizations, nations and publics, in support of collective well-being and the common good. A special commitment to benefit sharing that includes giving back to participants and communities whose samples and data are used for research, such as co-research opportunities, the sharing of skills and research capacity and priority access to the benefits of research.

4. Social justice

A commitment to develop organoids and related technologies in ways that: (i) promote human health, collective well-being and the common good; (ii) look after the needs of communities experiencing greater health burdens; (iii) reduce socioeconomic inequality; and (iv) avoid discrimination. In consultation with relevant communities, efforts should be made to ensure access to adequate resources, skills training and capacity-building for researchers, clinicians, policymakers, counsellors and others, as needed.

5. Non-discrimination

A commitment to celebrate and promote diversity by rejecting concepts of eugenics and patterns of discrimination based on personal or group characteristics including race, ethnicity, colour, religion, sex, gender, sexual orientation, age, and mental or physical ability.

6. Equal moral worth

A commitment to recognize and treat all people as having equal moral worth and their interests as deserving of equal moral consideration, with a particular need to recognize and protect the interests of persons with disabilities and of future generations.

[12] For further conceptual distinctions with respect to organoids, please refer to Annex 4.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

7. Respect for persons

A commitment to respect the concerns of competent individuals regarding the most intimate aspects of their lives, including their health and their reproductive options. In addition, a commitment to promote the best interests of individuals who are not competent to make decisions for themselves.

8. Solidarity

A commitment to live and work in harmony, grounded in recognition of the interdependence of humans. In addition, a commitment to share the benefits and burdens of research and clinical care among all people, to minimise the risks of exploitation and to promote the common good.

9. Global health justice

A commitment to equitable access to opportunities and potentially beneficial outcomes from organoids and related technologies for all people, particularly those living in low- and middle-income countries. This includes equitable access to support for health research and the development of health interventions that are appropriate and affordable for the widest possible range of populations with a view to reducing socioeconomic inequality. This also includes equitable protection from potential coercion, exploitation and other harms.

2.3 A matrix to design our operational guidelines for responsible research on organoids and related technologies for researchers and evaluators

To position ourselves in the ethical landscape in order to achieve dialogue and construct consensual guidelines that can be used at least in all European countries, we used the following matrix and put these questions under debate; this will still need to be developed to ensure that this debate remains alive.

	Why be ethical?	**How to be ethical?**	**How do we make communities behave ethically?**
Doing reliable research	Building certified knowledge (through the peer review process)	Three minimal principles of honest research: 1) Reproducibility 2) Replicability 3) Rationality	Establish professional standards and access to information to enable honest and reliable research
Doing fair research	Build laboratory leadership and mentorship that promotes scientific integrity and responsible research	The four principles of fair and responsible research: Accountability, honesty, reliability and respect	Establish a code of conduct for responsible research on organoids
Doing responsible research	Building trust within society/societies	Define the principles that enable the development of these trusting relationships	To set up a Code of Professional Conduct that takes account of the consolidation of trust between scientists and citizens (to deliver certified knowledge, not opinions or beliefs)

To answer such questions in a practical way and build trust among the various partners in research, a certain number of texts that now constitute references for research practice have been written and endorsed by most European institutions and research organisations; these include the Singapore Declaration (2010), TRUST (2018) and the ALLEA Code of Conduct (2023).

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

2.4 Ethics by design and RAD approaches: from ethical principles to practical solutions

Ethics by design is an approach that implies the need to effectively anticipate and reflect upon the ethical issues that might arise with new technologies. According to the artificial intelligence (AI) European Project, the "aim of Ethics by design is to incorporate ethical principles into the development process allowing that ethical issues are addressed as early as possible and followed up closely during research activities"[13]. Furthermore, the EU SIENNA deliverable defines ethics by design as the "systematic inclusion of ethical values, principles, requirements and procedures in design and development processes."[14]
According to this methodology (see Annex 2), developers of new technologies must take ethical challenges into consideration during their design process and thus embed societal values in the project idea, then within the prototype and pilot and finally during the scaling-up process for the emerging technology.

If initially the main field of application of ethics by design was artificial intelligence[15], currently this approach refers to a broader perspective on the ethics of new technologies that could be applied in numerous fields, including biotechnologies and hence the field of organoids and related technologies.

Incorporating the RAD (Reflexivity, Anticipation, and Deliberation) process alongside the established Ethics by Design approach offers a more comprehensive framework, particularly suited to the complex ethical landscape of organoid research and related technologies. While Ethics by Design emphasizes the proactive integration of ethical considerations throughout the technological development process, the RAD process extends this foundational approach by adding layers of ongoing reflection, forward-looking anticipation, and inclusive deliberation, which are crucial for navigating the intricate ethical dimensions specific to organoids.
The advantage of combining Ethics by Design with the RAD approach lies in the dynamic and iterative nature of RAD, which complements the systematic ethical incorporation of Ethics by Design. **Reflexivity** encourages constant self-examination and adjustment to ethical principles in response to new insights or challenges that emerge during the research and development phases. This is particularly relevant for organoid technology, where ethical considerations can evolve rapidly as the technology advances and as its applications broaden. **Anticipation** goes beyond initial ethical assessments to continuously forecast and prepare for future ethical dilemmas and societal impacts, a crucial step given the potential of organoids to revolutionize medicine and pose unique ethical challenges. **Deliberation** ensures that a wide range of stakeholders, including ethicists, biotechnologists, patients, and policy-makers, are involved in discussions about ethical issues, facilitating a more inclusive and holistic approach to ethical decision-making.

[13] Ethics and Research Integrity Sector, DG R&I, European Commission, *Ethics By Design and Ethics of Use Approaches for Artificial Intelligence*. 25 November 2021. Accessible at : https://ec.europa.eu/info/funding-tenders/opportunities/docs/2021-2027/horizon/guidance/ethics-by-design-and-ethics-of-use-approaches-for-artificial-intelligence_he_en.pdf
[14] Brey, Philip, Brandt Dainow, Yasemin J. Erden, Amal Matar, Philip Jansen, Rowena Rodrigues, Nicole Santiago, et al. 2021. *SIENNA D6.3: Methods for Translating Ethical Analysis into Instruments for the Ethical Development and Deployment of Emerging Technologies*, p.53. Accessible at: https://doi.org/10.5281/zenodo.5541539.
[15] See the other document for a longer review. This is particularly the case of the SwafS projects SIENNA and SHERPA.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

This combination not only enhances the ethical robustness of organoid technologies by ensuring that ethical considerations are deeply integrated from the outset but also maintains a flexible and responsive ethical stance throughout the lifecycle of the technology. By leveraging both approaches, developers and researchers can ensure that organoid technologies not only adhere to current ethical standards but are also adaptable to future ethical, societal, and regulatory developments. This dual approach fosters a culture of ethical responsibility and trust, which is essential for the successful integration of organoid technologies into society and for their acceptance by both the scientific community and the public at large.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

3 ORGANOIDS SPECIFIC ETHICAL ISSUES

Building on the values and principles described above, this section focuses on specific issues related to organoids and associated technologies. This section summarizes the general needs for ethical reviews of research conducted on organoids. Further details of our analysis applied to some specific types of research with organoids are developed in chapter 7.

3.1 Organoids: an evolving concept

Because of their novelty and hybridity, organoids received several definitions, sometime misleading. Considering their characteristics, organoids are at least cell-derived 3D structures that self-organise spontaneously, resulting in an architecture that mimics some aspects of a given organ and that perform certain functions of specific organs: "Stem or progenitor cell-derived 3D structures that, on much smaller scales, re-create important aspects of the 3D anatomy and multicellular repertoire of their physiological counterparts and that can recapitulate basic tissue-level functions."[16], [17]However, the rapid development of the field allows to anticipate that refined definitions will be required to describe, among others, patient explant-derived organoids, guided assemblies of complex organoids performing sophisticated natural or artificial functions, chimeras as well as hybridisation between organoids and non-biological devices.

Among the most important ethical issues is referring to an organoid as a "mini-organ" because it can be misleading and oversimplify the complexity of organoids. While organoids share some similarities with organs, such as having multiple cell types and performing specific functions, they are not exact replicas of complete organs. Thus, here are a few reasons why labelling organoids as "mini-organs" may be problematic:

1. Lack of full organ functionality: Organoids often lack the full functionality and complexity of real organs. While they may exhibit some organ-like characteristics, they are typically simpler and may not fully replicate the intricate structures and functions of natural organs. This also includes limited size and scale affecting their ability to fully replicate the physiological processes and interactions that occur within larger organs.

2. Lack of vascularisation, innervation, immune system.

3. Lack of interactions with the system of organs they belong to

Instead of wrongly labelling organoids as "mini-organs," it is more accurate to describe them as in vitro models. Emphasizing their unique properties, capabilities, and limitations can help researchers and the public better understand their potential applications and contributions to biomedical research**.** It is therefore a commitment to behave this way as part of research integrity. Furthermore, we strongly recommend that experts of a given field organize a consensus conference to propose a generic and common way to describe organoids from their specific field.

[16] G. Rossi & al., "Progress and Potential in Organoid Research", *Nature Reviews*, 2018, vol. 19, p. 671. This definition is used in the *Organoids Research: What are the ethical issues?* Memorandum.

[17] Bernard Baertschi, Henri Atlan, Mylène Botbol-Baum, Bertrand Bed'hom, Hélène Combrisson, et al., *Organoids Research: What are the ethical issues?, Note Inserm Ethics Committee*, 2020. Accessible at : inserm-03117706.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

3.2 Use of organoids: the identification of four categories

Depending on the complexity of the organoids and their possible uses, the constraints will be more or less stringent. As previously said, different objectives for organoid use are considered here: 1) for research, 2) for bioproduction, 3) for preclinical use, and 4) for clinical use.

For research

At present, most of the production and use of organoids and related technologies pertains to the research field. This fundamental research field focuses on determining how organoids can be produced and deliver information on the development of a related organ. The acquisition of functions contributes to our understanding of the organ's physiology and pathologies.

Good laboratory practices have been established so that scientists receiving an organoid can rely on the data which describe and characterise it (structural data: omics; morphological data: imaging and functional data). Researchers must also comply with all regulatory aspects depending on their national legislation before receiving the organoid, i.e. MTAs (Material Transfer Agreements) including provisions for the use of the organoid, the prior verification of signed patient consent forms, an authorisation from regulatory agencies for organoids constituted from patient cells, and a declaration of collection and of material transfer if applicable.

What is the process for the reliable and reproducible production of an organoid that a researcher can confidently share with other scientists? In this context, is it necessary to associate a minimum of information (metadata) with each batch of organoids? To answer these questions, the researcher requires access to a description of the sources, procurement protocols, validation and conservation of raw material protocols and the database, as well as culture protocols and quality control criteria for each level of organisation, as well as for biobanking modalities and differentiation procedures. A detailed description can be found in the section on MIAOU (see Section 4).

For bioproduction

In this section, we refer to an organoid specifically engineered for production purposes as a "factoroid." The engineering process for organoids involves making various improvements to optimise them for efficient and precise production through directed evolution. The ultimate goal is to establish a streamlined production line using these factoroids.

Work must be conducted in compliance with Good Manufacturing Practice (GMP) standards, and clinical-grade production processes should be employed when necessary. These quality control measures include:

✓ Ensuring the quality of raw materials, starting products, reagents and other components, up to the final product.
✓ Performing batch analysis in accordance with end-use requirements, such as conducting germ-free tests, functionality tests, impurity testing and environmental controls.
✓ Validating the batch in compliance with to GMP procedures, as necessary.

For preclinical use

The preclinical stage focuses on the research and development of therapies prior to clinical phases I, II, III, and IV, making a distinction between care and research. This stage involves

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

understanding the mode of action of Advanced Therapy Medicinal Products (ATMPs), as well as testing their efficacy and toxicity before administering them to humans.

There are several objectives:

1. Using organoids as a tool for therapy development, including classical chemistry, biologics, and innovative treatments or therapeutic devices (ITDs).
2. Developing organoids as innovative drugs (ATMPs).
3. Personalising treatments within the context of personalised medicine and, more broadly, in order to establish care protocols.
4. Utilising organoids as medical devices, such as theranostics that combine diagnostics and therapy.

A comprehensive description of prediction levels is necessary, and of the components of a toolbox to design and interpret trials, as well as toxicology and pharmacology studies. More broadly, this encompasses efficacy studies, pharmacovigilance (toxicity studies), pharmacodynamics (studies of active substance-target interactions), and pharmacokinetics (the fate of substances administered to a living organism).

For clinical use

At the European Union level[18], since 2007 the status of cell and gene therapy preparations has changed to becoming advanced therapy medicinal products (ATMP) when the cells being used undergo substantial modifications or are directed towards a functional purpose different from their original functions. As 3D structures derived from self-organising cells, organoids fall within the classification of ATMP, which are therapies composed of cells, tissues or genes. This was confirmed by interviews conducted within the framework of HYBRIDA's WP3 deliverable[19].

Multiple clinical uses are being envisaged at present:

1) The integration of organoid technology into clinical applications significantly advances personalized medicine, offering a more nuanced approach to treatment. By cultivating organoids from a patient's own cells, clinicians can test the efficacy of multiple treatments in a controlled environment before administering them to the patient. This method not only increases the success rate of treatments but also minimizes the risks of adverse effects, heralding a new era in healthcare where treatments are tailored to the individual's genetic makeup and disease profile, thereby enhancing patient care and treatment outcomes. At the end of March 2024, 174 studies were collated on the Clinicaltrial.gov website using the keywords "organoid" and "Cancer".

An example of such clinical application can be found in the publication by Tan and colleagues: The article highlighted the use of PDTOs in metastatic colorectal cancer to predict responses to standard-of-care therapies, showcasing organoids as a powerful tool for tailoring treatment plans to individual patients.[20] This approach aligns with the clinical applications of organoids mentioned, including drug efficacy testing and supplementing lost functions. It further

[18] Regulation (EC) No 1394/2007 of the European Parliament and of the Council of 13 November 2007 on advanced therapy medicinal products and amending Directive 2001/83/EC and Regulation (EC) No 726/2004.
[19] https://hybrida-project.eu/deliverables/.
[20] Tao Tan et al., Unified framework for patient-derived, tumor-organoid-based predictive testing of standard-of-care therapies in metastatic colorectal cancer, *Cell Reports Medicine*, 2023. Accessible at: https://doi.org/10.1016/j.xcrm.2023.101335

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

underscores the significance of organoids in advancing personalized medicine, through their application in clinical trials for various cancers and their role in the development of more effective, individualized treatments.

2) Organoids can be administered/grafted to humans in order to replace or supplement lost functions, to promote repair mechanisms or even to stimulate the immune system against viruses or tumour cells.

In July 2020, the American Food and Drug Administration released a guidance document entitled "Regulatory Considerations for Human Cells, Tissues, and Cellular and Tissue-Based Products (HCT/Ps): Minimal Manipulation and Homologous Use"[21] to assist in the classification of tissue therapies. HCT/Ps contain or consist of human cells or tissues intended for implantation, transplantation, infusion or transfer into a human recipient, but not organs, blood or blood products, body secretions or animal tissue. Homologous use means that the tissue performs the same basic function in the recipient as in the donor, but it may be in a different location in the body. Under this regime, some tissue therapies do not require a clinical trial, are not regulated or documented. They are categorized as innovative 'medical procedures', rather than medicinal products requiring marketing approval. This designation also means they are not formally considered to be research, meaning they are not subject to oversight from research ethics committees or required to report results unless clinicians are proactive in these regards. As a result, the rates and types of adverse events and patient benefits arising from these treatments are not known. However, even though according to this guidance, tissue collection, stem cell isolation and implantation could be classified as minimal manipulation, the FDA has classified these 'many steps' as more than minimal manipulation, particularly cell purification and expansion which involves culturing in media, and transportation between sites. Organoids cannot therefore be considered under HCT/Ps and thus should be subject to oversight from research ethics committees.

At the end of March 2024, 221 studies were collated on the Clinicaltrial.gov website using the keyword "organoid", most of them evaluating if testing drugs on organoids may predict the efficacy of chemotherapies for cancers such as lung, pancreas, kidney, haematological, oesophageal, colorectal, etc. Below are a few examples of ongoing clinical trials:

- Novel 3D Hematological Malignancy Organoid to Study Disease Biology and Chemosensitivity (Organoid) ClinicalTrials.gov ID NCT03890614 to compare chemosensitivity between chemotherapy combinations in bone marrow aspirates using 3D organoid models.
- Development of a Prediction Platform for Adjuvant Treatment and Prognosis in Resected Pancreatic Cancer Using Organoid ClinicalTrials.gov ID NCT04736043. The investigators are creating organoids from pancreatic cancer tissues obtained via EUS-FNA and EUS-FNB within the pancreatic cancer diagnostic process and obtained after surgery as part of the treatment process. Reactivity to anti-cancer drugs is also being verified by the use of cell viability assays.

[21] Regulatory Considerations for Human Cells, Tissues, and Cellular and Tissue-Based Products: Minimal Manipulation and Homologous Use, Guidance for Industry and Food and Drug Administration Staff, 2020. Accessible at: https://www.fda.gov/regulatory-information/search-fda-guidance-documents/regulatory-considerations-human-cells-tissues-and-cellular-and-tissue-based-products-minimal.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

The clinical use of organoids, as of all ATMP, requires regulatory supervision. This implies that:

• The preclinical stages (efficacy, toxicology, pharmacovigilance and pharmacokinetic studies) are validated and documented in the regulatory files held by the national regulatory agency.

• The GMP production of organoid medicinal products is assured by an establishment officially qualified as a pharmaceutical establishment and that the production protocols and quality controls are transmitted to the national regulatory agency.

• The filing of a clinical trial authorisation application with the national regulatory agency, including establishment of the various regulatory files which specify the identity and responsibilities of the sponsor and investigator, a description of the methodology and design of the clinical trial together with the possible risks of adverse effects and patient management measures depending on the level of severity, and insurance coverage is ensured by the sponsor and also concerns the post-clinical trial follow-up of patients in pharmacovigilance studies.

Because clinical trials are closely supervised as described above, the ethical problems posed by the transplantation of organoids with respect to the recipient are taken into account (health safety, possible side effects, etc.). It will be incumbent on RECs to ensure that the information process, mandatory for any inclusion into a clinical trial clearly lists all uncertainties, such as the long-term survival and stability of grafted organoids but also the surveillance and assessment methods developed to monitor such uncertainties and guarantee the highest possible degree of safety.

3.3 Organoids and specific issues needing ethical review

This section summarizes the needs for ethical reviews of research conducted on organoids. Further details of our analysis applied to some specific types of research are developed in chapter 7. This section was inspired by the ethics of research review categories proposed by the 2021 ISSCR Guidelines for Stem Cell Research and Clinical Translation.

Applied to the specific field of organoids and related researches, two main areas are considered as priorities for a specific ethical review as they may be subject to measures related to the precautionary principle: i) organoids associated with the dissemination of genetic material in the offspring, and ii) organoids in which higher order brain functions such as consciousness or suffering may emerge.

Four ethical categories result from the classifications above:

Cat. 1a: there is no need for a specific ethical review: a "simple" approach to organoids (kidney, liver, etc.). However, it is clear that all legal and ethical reviews associated with human cell collection (particularly concerning information for and consent of the donor, as described in Chapter 6) should be respected.

Cat. 1b: specific ethical consideration is recommended to the researcher and certain declarations must be made to the authorities: "complex" organoids such as cerebroids (not connected to sensory systems), sexual reproduction organoids, “simple” assembloids (interconnected organoids not reaching high order brain functions) and gastruloids.

This project has received funding from the European Union’s HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

Cat. 2: cases where specific approval by an Ethics Committee is required: blastoids, complex assembloids. A complex assembloid implies cerebroids connected to sensory and possibly motor systems. In such systems, nociceptive processing by some brain circuits may start to suffer or experience a degree of pain, and different degrees of consciousness may emerge from complex neural networks. However, there is currently no clear consensus on how to demonstrate consciousness or suffering in neural organoids mimicking the immature cerebral cortex (see chapter 7).

Cat. 3: a prohibited organoid because there is a lack of compelling scientific rationale and/or ethical standpoint concerning: gestating human stem cell-based embryo models, or the transfer of human-animal chimeric embryos to a human or non-human primate uterus; for instance, any research at present that would cross the gap between germen and soma.

Gestating human stem cell-based embryo models or the transfer of human-animal chimeric embryos to a human or non-human primate uterus are banned because such interventions lack a compelling scientific rationale and/or raise ethical concerns, and remain a subject of debate. Of note, this ban faces criticism due to a lack of clear definitions of terms such as "gestation" (in the context of cell lines or stem cell-based tissues) and "human stem cell-based embryo models." In addition, discussion continues regarding integrated versus non-integrated embryos, with the possibility that the latter could become obsolete if extra-embryonic tissues are replaced by alternative entities. Furthermore, this ban might be seen as problematic because, in the long run, the vascularization of gastruloids could pave the way towards developing organs that address significant clinical needs. Consequently, it is essential to clarify the meaning of these terms in the context of stem cell technology. In our view, germline refers to the genetic material passed down from one generation to another through reproductive cells, while soma refers to the non-reproductive cells that make up the body. It is also worth noting that stem cell-based fertility research, focused on the germline, is a highly active and rapidly evolving international field. Therefore, in view of the present uncertainties, we recommend this ban but open discussions should be pursued in order to envisage for which purpose and under which process of scientific and ethical evaluation and survey some aspects of such research might one day be permitted.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

4 EVALUATION CHECKLIST FOR SCIENTIFIC AND ETHICAL STUDIES USING ORGANOIDS

This section of the Operational Guidelines aims to **build trust** among researchers, among scientific evaluators and between researchers and scientific evaluators. It thus comprises two parts:

a) A description of reliable, honest and transparent organoid design, fabrication, characterization, functionality and applications under the title: **MIAOU** (Minimum Information About Organoids and their Use)
b) A list of criteria to ensure the fair, transparent, respectful and responsible evaluation of studies involving organoids under the title: **EChOES** (Evaluator Checklist for Organoid experimental Studies) for scientific evaluation.

This work is aligned with the broader definition issued by the European Medicines Agency (EMA) on guidelines, which should "reflect a harmonized approach of the EU Member States and the Agency on interpreting and applying the requirements for demonstrating quality, safety, and efficacy set out in the Community directives."[22]

These two documents are built upon a set of shared values discussed in chapters 2.1 and 2.2, among, and between, the stakeholders concerned, namely researchers and evaluators (specific values are shown in the red square in the Table below).

	Researcher	Evaluator	institution	Citizen/patient
Researcher / Physician	Reliability Honesty Transparency	Honesty Reliability Transparency Respect Responsibility	Honesty Reliability Transparency Respect responsibility	Physical, moral and Social well-being Respect for privacy Honesty
Evaluator	Equity (fairness) Transparency Respect Responsibility	**shared principles for evaluation** Benevolence Non-malevolence Autonomy Justice	Transparence Responsibility respect	Honesty
Institution	**Commitment of the institution** Transparency honesty Responsibility	Respect Responsibility Transpareny	Openness	Physical, moral and social well-being Honesty
Citizen & Patients	Consent	Consent	Consent	Consent Honesty

[22] European Medicines Agency, *Quality guidelines*, Accessible at: https://www.ema.europa.eu/en/human-regulatory/research-development/scientific-guidelines/quality-guidelines.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

They serve as guides to compile a document that Research Integrity Committees (RIC) and Research Ethics Committees (REC) can use to i) evaluate research projects while paying constant attention to data confidentiality, ii) assess the societal impact of the research project and its anticipated results, iii) ensure the commitment of patient associations, and iv) ensure the fair and responsible behaviour of Ethics Committees involved in evaluating projects that involve the use of organoids. This part is developed in the next chapter under the title "**RICOCheck**" (Research Integrity Committee check-list), and in the European Code of Conduct for Research Integrity on Organoids and Related Technologies, deliverable D5.2.

Within the framework of HYBRIDA's outcomes, an experimental website and system based on open-source protocols checked the pertinence and ease of use of the files. For example, if the answer to a given question is "No" at the first line, the system jumps to the next section. All our questionnaires will be now available at EBISC and hPSCreg.

4.1 MINIMUM INFORMATION ABOUT ORGANOIDS AND THEIR USE (MIAOU) FOR RESEARCHERS

Documents developed thereafter **are intended to contribute to a reliable and reproducible production of an organoid that a researcher can confidently share with other scientists.** This involves creating a minimum information checklist (metadata) for each batch of organoids, detailing the following aspects:

✓ To describe the sources, procurement protocols, validation and conservation of raw material protocols
✓ To describe culture protocols and quality control criteria for each level of organisation, as well as biobanking modalities
✓ To describe differentiation procedures

The Minimum Information About Organoids and their Use (MIAOU) questionnaire[23] presented here is based on a bottom-up approach, using input from a network of scientists working in the field of organoid design, characterisation and usage to enhance the reproducibility, replicability and rationality of research within and between laboratories and across organoids. The questionnaire encompasses four areas of application: basic, preclinical and clinical research and bioproduction.

The MIAOU questionnaire is designed as an online survey to facilitate user feedback, thus simplifying the process and achieving the aforementioned goals (i.e., building trust among and between scientists and evaluators).

The MIAOU questionnaire consists of five sections:

1) Project identification
   The section aims to clearly identify the project, including its applications and their limits. Particular attention should be paid to not using wording that might induce fears or unrealistic promises. See the discussion on erroneous naming in chapter 7; an example is a consensus paper on the naming of nervous system organoids which has already been published[24].

[23] Check Annex 7.
[24] *Ibid.*, p. 7.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

2) Source materials
Critical elements in this section concern: 1) stem cell metadata based on the ATCC model (batch, structural, morphological and functional data, maintenance and preservation protocol, the presence of informed consent signed by the donor, 2) the declaration of collection (declaration or authorisation of activities for the conservation and preparation for scientific purposes of human body elements) which is mandatory for human samples, 3) the monitoring of possible drifts of starting materials, 4) regulatory documents and medical ethics if relevant (restrictions on use according to donor consent).

3) Organoid manufacturing
Critical elements in this section concern: 1) differentiation protocols and organoid generation (table of differentiation factors, timelines, culture protocols, purification protocols and, if necessary, maintenance and preservation protocols) 2) design and development of master organoid bank and working organoid banks, 3) monitoring of possible drifts of organoids (genetic or protein post translational modifications, metabolism, other biomarkers).

4) Organoid characterisation
Detailed characterisation is project-dependent and should be ensured in line with the proposed use of the organoid (research, bioproduction, preclinical and clinical uses); however, some standards have emerged (omics for structural characterisation, imaging for morphology, and specific functional readouts depending of the foreseen use of the organoid).

5) Organoid use
The critical element in this section is the robustness of the preparation and characterisation of the organoid and anticipation of the future use of organoids, from basic use to the development of innovative applications (for instance, the implementation of good laboratory practices will facilitate the transition from basic to preclinical research).

The full questionnaire is presented in the Annex, chapter 11, and a web version is available at French GDR Organoids and will be implemented at EBISC and hPSCreg.

4.2 Evaluation Checklist for Organoid Experimental Studies (EChOES)

The MIAOU (as mentioned above) is employed to identify the information provided (using Yes/No answers) and to assess the quality of descriptions concerning reproducibility, replicability, and rationality in organoid research.

The Evaluator Checklist for Organoid Experimental Studies (EChOES) derives from the MIAOU list of criteria in order to ensure the fair, transparent, respectful and responsible scientific evaluation of studies involving organoids by scientific committees. To evaluate the

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

quality of a submission, the elements highlighted in red are mandatory for the scientific assessment process, while those in black are contextual (based on the call requirements, project deployment, domain of organoid usage, etc.) and may be beneficial to enabling a comprehensive evaluation of the project. It is up to the evaluators to determine whether a Yes or No response is acceptable for a specific project.

The EChOES questionnaire consists of five sections:

1) Project identification (four critical questions):
   This section aims to clearly identify the project, including its applications and their limits. Particular attention should be paid to not using wording that might induce fears or unrealistic promises.

2) Source materials (13 critical questions)
   Critical elements in this section include: 1) stem cell metadata based on the ATCC model (batch, structural, morphological and functional data, maintenance and preservation protocol, the presence of informed consent signed by the donor*), 2) declaration of collection (declaration or authorisation of activities for the conservation and preparation for scientific purposes of human body elements), mandatory for human samples, 3) monitoring of possible drifts of starting material, 4) regulatory documents and medical ethics if any (restrictions on use according to donor consent).*

3) Organoid manufacturing (seven critical questions)
   Critical elements in this section cover: 1) differentiation protocol and organoid generation (table of differentiation factors, timelines, culture protocols, purification protocols and, if necessary, maintenance and preservation protocols) 2) design and development of master organoid bank and working organoid bank, 3) monitoring of possible drifts of organoids (genetic or protein post translational modifications, metabolism, other biomarkers).

4) Organoid characterisation (four critical questions)
   Detailed characterisation is project-dependent and should be ensured in line with the proposed use of the organoid (research, bioproduction, preclinical and clinical uses); however, some standards have emerged (omics for structural characterisation, imaging for morphology and specific functional readouts depending of the foreseen use of the organoid).

5) Organoid use (five critical questions)
   The critical element in this section is the robustness of the preparation and characterisation of the organoid and anticipation of the future use of organoids, from basic use to the development of innovative applications (for instance, the implementation of good laboratory practices will facilitate the transition from basic to preclinical research).

The full questionnaire is presented in the Annex, Chapter 11, and a web version is under construction.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

A supplementary document, RICOCHECK, is necessary for a complete evaluation by international review boards, such as the Research Integrity Committee (RIC) or Research Ethics Committee (REC). This document is presented in Chapter 5.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

5 RICOCheck for International Review Boards (Research Ethics Committee, Research Integrity Committee)

Several principles need to be taken into account when performing evaluation by RIC and RECs in the field of organoids and related technologies, such as i) constant attention to data confidentiality, ii) the societal impact of the research project and its anticipated results, iii) the commitment of patient associations, iv) the fair and responsible behaviour of Ethics Committees involved in evaluating projects using organoids.
RICOCheck is a European online survey proposed by the HYBRIDA, focused on the ethical issues raised by research programmes involving organoids.
Research Integrity Committees (RICs) and Research Ethics Committees (RECs) evaluate and subsequently approve research projects that correctly address the following issues: a i) personal data confidentiality, ii) societal impact of the research project and its anticipated results, iii) volunteer commitment of patients and donors. In turn, Ethics Committees involved in evaluating projects using organoids are committed to adopting fair and responsible behaviour toward the promoters of the research. The planned use of the organoids produced during the programme is of prime importance and should emphasise societal benefits and consider potential harms to donors, patients and society.
The RicOCheck document was developed using the model in *How to Complete your Ethics Self-Assessment.*[25] Sections 7 (environmental health and safety) & 8 (artificial intelligence) of the European self-assessment document are not pertinent in the field of organoids.

RicOCheck therefore contains eight 8 sections and the questions have been adapted to the organoid field, when necessary:

Section 1: Human embryonic stem cells (hESCs) and human embryos (hEs) (HE, DEP, EU4H and EDF)
Research Integrity and Ethics Committees are particularly attentive to the existence of informed consent for the use of donor biological samples, associated personal data, and to the good adequacy between the consent, the proposed research programme and its applications.
Research on human stem cells, both adult and embryonic, may be financed depending both on the contents of the scientific proposal and the legal framework of the Member States involved. According to the Official Journal of the European Union, no funding shall be provided within or outside the Union for research activities that are prohibited in all Member States; no funding shall be provided in a Member State for a research activity which is forbidden in that Member State[26].

[25] Horizon 2020 Programme, *Guidance 2021: How to complete your ethics self-assessment*, Accessible at: https://ec.europa.eu/info/funding-tenders/opportunities/docs/2021-2027/common/guidance/how-to-complete-your-ethics-self-assessment_en.pdf
[26] Regulation (Eu) 2021/695 of The European Parliament and of The Council of 28 April 2021, Article 18, Chapter 1. Accessible at: https://eur-lex.europa.eu/legal-content/EN/TXT/PDF/?uri=CELEX:32021R0695.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

Section 2: Humans

This section deals with physical, moral and social well-being, together with notions of respect and honesty. Research Integrity and Ethics Committees will be particularly attentive to the existence of informed consent and to the good adequacy between the consent and the TRUSTED, the proposed research programme and its applications. Denomination of the organoids generated should not be misleading nor carry undue hopes. The possibility to use samples taken from cadaveric donors differs from country to country. Projects leaders aiming at using such material will have to refer to the legislation in force where opt-in and opt-out options exist for tissue sampling and usage categories.

Section 3: Humans cells/tissues/organoids

*This section deals with the following values: transparency, honesty and responsibility. Human body elements should be traceable and used in accordance with the conditions of the research and in accordance with the donor's informed consent and responses to the potential future use questionnaire. Denomination of the organoids generated should not be misleading nor carry undue hopes***.**

Section 4: Personal data

This section deals with the notion of privacy-by-design, i.e. data confidentiality. Personal data acquisition should be limited to the strict minimum, anonymised or pseudonymised and their use should be limited to the proposed research programme.

Section 5: Animals/chimeras

This section relates to the 3R rule (reduce, replace, refine use of animals in experimentation). The use of animals instead, or in vitro *or* ex vivo *model systems requires justification.*

Section 6: Non-EU countries

Regulations differ significantly between countries. The promoter of the study should ensure that the export of material to other countries is permitted, and that the study intended will follow the national regulations in force (request for authorisation from the competent authorities).

Section 7: Potential misuse of results

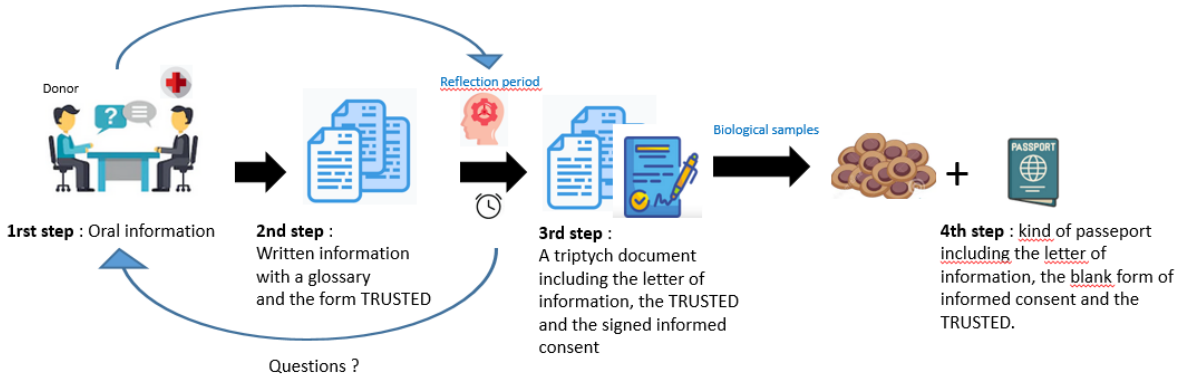
Attention must be paid to the secondary uses of donor samples. The promoter of the research should ensure that all foreseen uses are included in the informed consent and allowed in the donor "TRUSTED list" (restrictions of use), or should obtain a new informed consent.
This section also deals with values such as benevolence and non-malevolence. Potential military uses should be considered here.

Section 8: Other ethical issues

Please specify any other ethical concerns that may arise from the proposed project.
The full questionnaire is presented in the Annex, chapter 11, and a web version is under construction.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

6 ORGANOIDS AND INFORMED CONSENT*



N° 1 - Informed consent procedure

Informed consent is a procedure that includes both oral and written information, with the donor's signature attesting that the information process has been completed and that they freely consent. Once signed, the consent form becomes a formal document that cannot be modified. Any changes, whether to the scientific aspects or to the secondary re-use of samples and/or associated data, will require a new consent form to be drawn up. Considering the unforeseen reuse of cells or tissue, we propose anticipating this by adding the TRUSTED questionnaire for more clarification on application fields.

Informed consent is a basic and fundamental prerequisite of any kind of research involving human subjects, including through the collection of tissues or cells. Consequently, and as stated in section 3.3, specific ethical considerations should be foreseen before starting a research project that involves the generation of organoids with human material. An essential point to consider is the information provided to the donors and their consent for the use of their cells or tissues. Such considerations are rooted in the long history of informed consent. The Nuremberg Code underscores consent and is at the origin of the concept that participation in research is a voluntary activity. The Declaration of Helsinki also enshrines consent as a main guarantee for the promotion and safeguard of "the health, well-being and rights of patients, including those who are involved in medical research"[27]. Article 6 of the Universal Declaration on Bioethics and Human Rights (UDBHR) provides that any preventive, diagnostic and therapeutic medical intervention, as well as scientific research, should only be carried out "with the prior, free, expressed and informed consent of the person concerned"[28]. According to the mini-deliberative workshop recently conducted by the HYBRIDA Project (D4.3) in Denmark, participants were

* This chapter was primarily drafted in collaboration with Dr Christine Dosquet, President of the Inserm Ethics Review Committee, and reviewed by several researchers and experts in the field of informed consent such as Prof. Margherita Daverio (LUMSA, Roma, Italy) one of the collaborators in the European i-Consent project: "Guidelines for tailoring informed consent in the framework of clinical studies" grant EU 741856.

[27] WMA Declaration of Helsinki, *Ethical Principles for Medical Research Involving Human Subjects*. General Principles. Article 4. Accessible at: https://www.wma.net/policies-post/wma-declaration-of-helsinki-ethical-principles-for-medical-research-involving-human-subjects/.

[28] UNESCO, *Universal Declaration on Bioethics and Human Rights*, Article 6 – Consent, 2005. Accessible at: http://portal.unesco.org/en/ev.php-URL_ID=31058&URL_DO=DO_TOPIC&URL_SECTION=201.html.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

particularly sensitive to the information given beforehand, the consent form and follow-up: "informed consent and information to patients and citizens should be clear, and procedures for informing patients and obtaining consent should be simple and understandable for the patients. The participants suggest allocating funds for this specific purpose, e.g. to ensure that there is sufficient time and resources for information"[29]. This is also a main recommendation of the European-granted I-Consent programme. Finally, informed consent implies informing donors about the progress/evolution of the research involving their donation and about their right to exercise autonomy by agreeing or declining to participate. This latter possibility includes the very difficult question of consent withdrawal[30], considering the amount of work involved in making and maintaining organoids (see the specific section dedicated to withdrawal).

Here, we extend the ISSCR Guidelines' list of informed consent considerations for the use of stem cells and adapt these considerations to the organoid field, encompassing the utilization of organoids and related technologies in research or clinical therapy. Specifically, our focus includes aspects such as genetic modifications, tissue and data storage, determining who is entitled to use organoids, donor confidentiality, the right to know the purpose of organoid usage, the therapeutic or commercial use of organoids or their research outcomes, preventing use for reproductive purposes, and ensuring that donation or refusal of donation does not affect clinical care. Since many patients often assume they own their cells by default[31], disclosure regarding potential commercial use should be particularly explicit (refer to Figure n°3 for a description of the informed consent procedure). Furthermore, in the specific context of organoids, some developments might be viewed as undesirable by the donor, such as the production of embryonic models or neural organoids. Therefore, we propose an innovative way to collect donor's views on potential use of their cells or tissues at the best level of information at the time of consent: the inclusion of a Tissue Research Under_Secure Transparent Ethical Donation (TRUSTED) list in the consent form, allowing donors to explicitly permit or prohibit specific potential uses of their biological material and data.

6.1 Legal capacity to consent

Because informed consent is mandatory, when a person is not capable of giving consent to produce organoids using their biological material permission must be obtained from their legal representative, in accordance with applicable law. The legal representative must consider the best interests of the person concerned[32].

Three different types of participants are commonly identified as not having full capacity to consent: mentally incompetent adults, adults in emergency care and children. For the collection of samples from minors, the informed consent of both parents or a legal representative should be obtained, following the minor's assent when possible. The information provided must be

[29] Please check the Deliverable 4.3, *Public attitudes, understandings and perspectives on organoid research*, HYBRIDA Project.

[30] According to the WP6 HYBRIDA findings, "from a legal perspective, donor withdrawals include only donated tissues and cells, and it remains unclear whether withdrawals extend to derived organoids." Lewis, J., and Holm, S. 2022. *Organoid biobanking, autonomy and the limits of consent*, Bioethics, 36, p.14. Accessible at: https://doi.org/10.1111/bioe.13047.

[31] Chirba M, Noble A. *Our bodies, our cells: FDA regulation of autologous adult stem cell therapies*, Bill of Health Boston College Law School Faculty Papers; 2013.

[32] UNESCO, *International Declaration on Human Genetic Data*, 2003. Accessible at: https://unesdoc.unesco.org/ark:/48223/pf0000136112.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

adapted to the age and degree of maturity of the minor, including appropriate vocabulary and visual explanations. In the event that minor children reach the age of majority during the planned studies or for the secondary use or option of their biological samples, it is recommended that they be offered the opportunity to provide consent or withdraw their consent as adults.

Moreover, in the specific case of adults with reading or writing disabilities, informed consent should be obtained with the assistance of an expert patient. Finally, in the specific case of using embryonic stem cells from an IVF embryo, oral and written information must be provided and informed consent will be sought from both parents or, where applicable, from the legal representative of both parents.

Protection of data associated with the sample and personal data: pseudonymisation or anonymisation process. Whatever the context of the collection of biological human samples and associated data, the rights and freedom of donors must be respected. The development and maintenance of organoids must also obey such rules. The General Data Protection Regulation (GDPR) defines personal data as "any information relating to an identified or identifiable natural person" (art. 4.1). In addition, the same document implies that giving consent "means any freely given, specific, informed and unambiguous indication of the data subjects' wishes by which they, by a statement or by a clear affirmative action, signify agreement to the processing of personal data relating to them."[33]

The GPDR (General Data Protection Regulation) requires the pseudonymisation of data for scientific research. Pseudonymisation is presented as an inherent safeguard for the processing of personal data for scientific research purposes as long as it is compatible with the purpose of the processing (art. 89). Pseudonymisation is the processing of personal data in such a way that it is no longer possible to attribute the data to a natural person without additional information[34]. In practice, pseudonymisation consists in replacing directly identifying data (surname, first name, etc.) in a dataset with indirectly identifying data (alias, sequential number, encryption key or code etc.). The GPDR encourages researchers to apply this measure because it offers a good compromise between the needs of the research and safeguarding the interests of the participants. Furthermore, pseudonymisation enables restoration to the donor of any information concerning a life-threatening condition that is discovered or if new research protocols require additional information.

By contrast, the anonymisation process aims to eliminate any possibility of re-identification, so the future use of the data is limited to certain types of use. For example, uses initially anticipated in the informed consent form signed by donors will be possible even if the donors withdraw their consent whilst their cells have already been engineered, cultivated or modified by the researchers, efforts which have already involved a lot of both work and funds so that it seems difficult to oblige them to stop. However, it should be noted that anonymisation means it will not be possible to link together separate sets of data concerning the same individual.

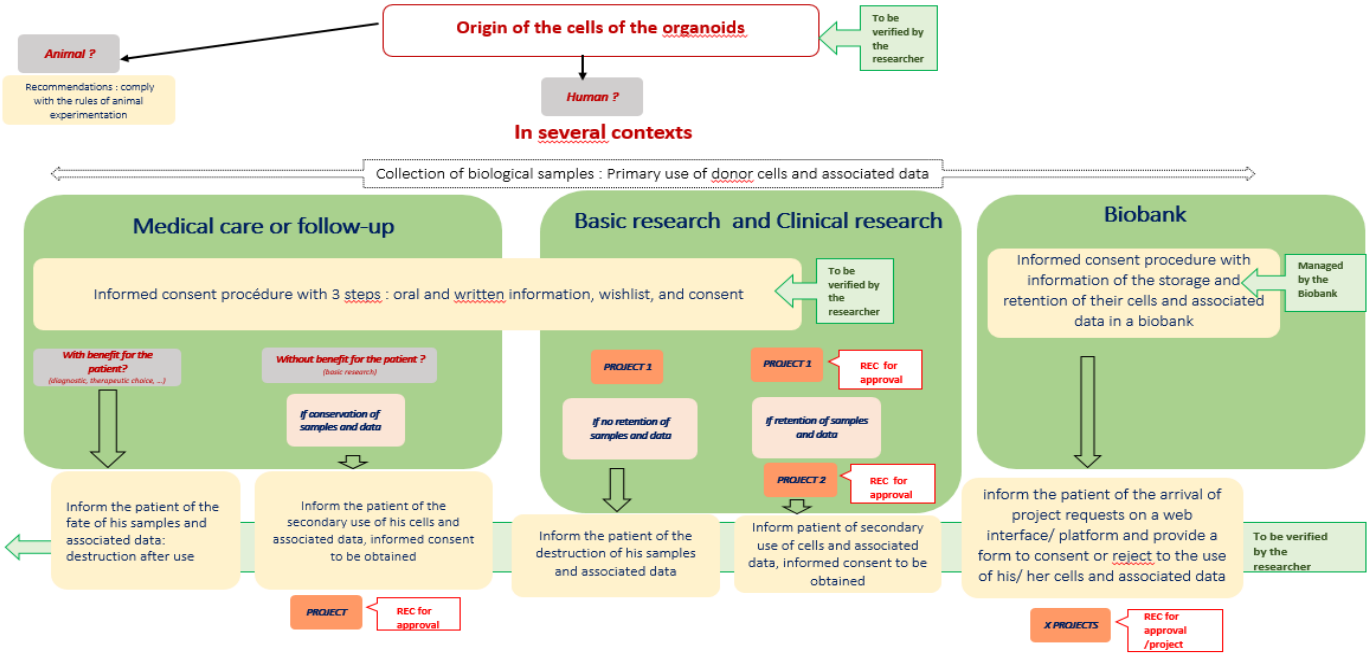
The constraints described above must be taken into account at the start of a project. The GDPR does not include a general obligation of anonymisation. It offers one solution, among others, for the use of personal data in terms of donor rights and freedoms. The choice between anonymisation and pseudonymisation, which must be carefully considered by researchers before a project, is conditional on the need to re-use and exploit personal data without

[33] General Data Protection Regulation (GDPR). Article 4 (11). Accessible at: https://gdpr-info.eu/art-4-gdpr/.
[34] *Ibid.*, p.39.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

infringing the privacy of individuals. It also makes it possible to store data beyond their retention period.

6.2 In what contexts does the collection of biological samples take place?



N° 2 – Tree describing the different contexts

For researchers initiating a project to build an organoid made of human cells, the supply of these cells necessarily involves a closely controlled process for the collection of biological samples and the data associated with the sample. Cells can be supplied from different sources (see Figure n°2 describing the different contexts):

1) from patients as part of their medical care or follow-up or from a volunteer in the context of clinical or basic research

2) from a biobank (institutional or commercial)

6.2.1 Collection of samples in the context of medical care (treatment, follow-up, consultation) or clinical research

When the collection of biological samples and associated data is established in the context of medical care, such as a diagnosis or treatment, and it is further used for research, the patient must receive oral and written information on the purpose of the research and the reasons for storage, its duration and the fate of the materials and associated data at the end of the project. In the particular case of the transfer of biological samples and associated data to a foreign country (or a separate jurisdiction), patients must receive transparent information on the

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

reasons for this transfer, the future of their biological samples and associated data, and the identity of the receiving research team.

If the patients agree, they must give written consent for the use/re-use of the biological samples and associated data according to their Tissue Research Under Secure Transparent Ethical Donation (TRUSTED) list (please see section 6.7) and for how the biological samples and associated data will be secured. The researcher who will be using these samples must ensure that this entire procedure has been rigorously followed and that the patients have consented to the storage and re-use of their biological samples for research purposes. The researcher must also obtain REC approval before the research project starts.

If collection is performed in the context of a donation of biological samples and associated data for research, it is essential that donors are provided with both oral and written information about the research project, as well as the conditions and duration of storage of their donation, prior to the collection of consent.

In the post-mortem context, a specific inquiry should be made for each particular jurisdiction according to the relevant national laws. For example, in France, a collection can be considered if the donor has not refused organ or tissue harvesting, as indicated in the French National Refusal Register[35]. If no refusal has been registered, the next of kin will be asked whether the donor has not expressed opposition in writing or orally during their lifetime. In the case of an oral statement, the medical team will ask the next of kin to specify the circumstances and to sign the written transcript.

If biological samples and associated data are ***retained for secondary use***, the patients must be given additional oral and written information, before the collection of consent, regarding the new research project that involves re-use of their biological samples and their derivatives, and associated data. This could be implemented through the research project website in a dedicated donor area. In addition, donors must be informed of the possible non-commercial use or, on the contrary, the commercialisation of their samples or derived biological material. It is the responsibility of the researcher to ensure that appropriate consent is in place and to submit the research project to a REC for approval before the research project starts.

If a researcher decides to redirect the initial research project to achieve another purpose, the patients must be informed and give their consent for a different use of their samples and associated data.

In all cases, a withdrawal procedure must offer donors the possibility of withdrawing their consent without prejudice to them (see below for details).

[35] https://www.registrenationaldesrefus.fr.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

6.2.2 Collection from biobanks

Both suppliers and scientists must be fully aware of the necessary shift from unethical towards ethical behaviours.

Indeed, when purchasing cells from a commercial supplier or even from institutional biobanks, scientists are often given too little, if any, information on the origins of the cells and the process implemented to obtain initial informed consent. This is a real and frequently encountered problem that raises major concerns because it challenges basic ethical principles. This important hurdle is of importance to organoid researchers as they question their practices regarding the use of such cells at present. It should be noted that no regulations can be retroactive and the present guidelines are designed to offer a framework for best practices in the future. Whereas advances have been made in providing researchers with information on the consent procedure, all commercial suppliers and all types of biobanks need to implement procedures that will ensure a high level of respect for donors, as we recommend below.

When a researcher wishes to obtain biological samples and associated data from a biobank, the latter should only release the sample(s) after a review and validation of the scientific pertinence of the research project and in full respect of the informed consent of the donors, including their TRUSTED list, concerning potential applications. The biobank should thus ask its scientific committee about the scientific pertinence of the project; if this receives approval, it should then verify whether it is acceptable in light of the TRUSTED list. We therefore recommend that each biobank should constitute a review board composed of scientists, clinicians, ethicists and representatives of patient organisations, in order to fulfil this critical mission. Furthermore, and regardless to the type of consent adopted, the biobank must inform the donor of any new requests from researchers regarding their projects (for example, by sending an email or an SMS inviting the donor to visit an interactive online site). Information could thus be provided via a dedicated website using a video explanation. By placing this procedure online under entirely anonymous conditions, information and exchanges (questions and answers between the parties to ensure that donors fully understand the explanations given) would be accelerated through a dynamic electronic consent/refusal process. In addition, this website would enable donors to be informed about the progress of research using their donation; as indicated above, this is frequently requested by them.

After approval by a scientific committee, in all cases where cell lines, primary cells and organoids associated with personal data are distributed by biobanks, commercial companies or between groups of researchers, the distribution of biological samples should be accompanied by an “anonymized passport” which includes the patient information sheet, a blank version of the informed consent form and the TRUSTED list. On their side, researchers must ensure that the donor’s information, consent procedure and TRUSTED list have been complied with and will be respected during their project, and they must seek the approval of a REC before starting the project. Cf Figure 1: Informed Consent Procedure.

6.3 Providing information beforehand and what to consider during the consent process

According to the findings of the deliberative workshop conducted by HYBRIDA WP4 in Italy, “participants point to rigorous consent procedures and Ethics Committees as two avenues for

This project has received funding from the European Union’s HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

control. Despite inconclusiveness in terms of governance responsibilities, unanimity exists as to the position that the **ethical use of cell donations must be guaranteed through strict governance structures, control and ethical oversight procedures** to ensure the ethically responsible, transparent and safe storage and use of cells, tissues or organoids in biobanks."[36]

Taking account of this and of the recommendations of the I-Consent Guidelines, three principles need to be considered regarding sound informed consent:

1. The donors are able to understand the information delivered orally and in writing by the physician or medical advisor and to recall it, including details on the withdrawal procedure and its consequences regarding the safeguarding of their biological samples and personal data.

2. The donors are capable of making an independent decision to authorise the retention and use(s) of their biological materials and personal data or to refuse any or all of the studies described.

3. Consent is voluntary, and obtaining it involves no manipulation, abusive incentives or promises. It should be noted that in some jurisdictions, no financial inducements or other personal benefits (except for financial compensation for travel expenses and working hours lost) should be offered to research participants, but financial inducement is legal in some other jurisdictions.

Consent must be prior, free and informed, and must be obtained for the collection by invasive or non-invasive procedures of biological samples and associated data of any kind, and their subsequent processing: the culture of cells for the construction of an organoid (including through the reprogramming of cells into iPSCs), use and storage.

Counselling can be an important part of the communication process; "counselling should be non-directive, culturally adapted and consistent with the best interests of the person concerned"[37]. This may include providing assistance during the reading of the consent form. The person responsible for requesting consent should encourage an open dialogue and potential participants must be offered an opportunity to ask questions about the project and protection of their data.

4. In the context of a clinical trial: the physician or medical advisor must "provide information on the treatment to be followed and on the course of the study, including the expected personal benefits and risks", and the likelihood of such benefits and risks (and their severity and/or frequency) occurring. It is desirable that an independent clinician should carry out the consent procedure.

6.4 Construction and use of organoids

6.4.1 Typology of organoids

Specific aspects of consent concerning the future use of the organoids are addressed in the following sections (See Figure n°2). Organoids are simplified models of organs, composed of

[36] Please go to Deliverable 4.3, *Public attitudes, understandings and perspectives on organoid research*, HYBRIDA Project. Authors' highlighting.

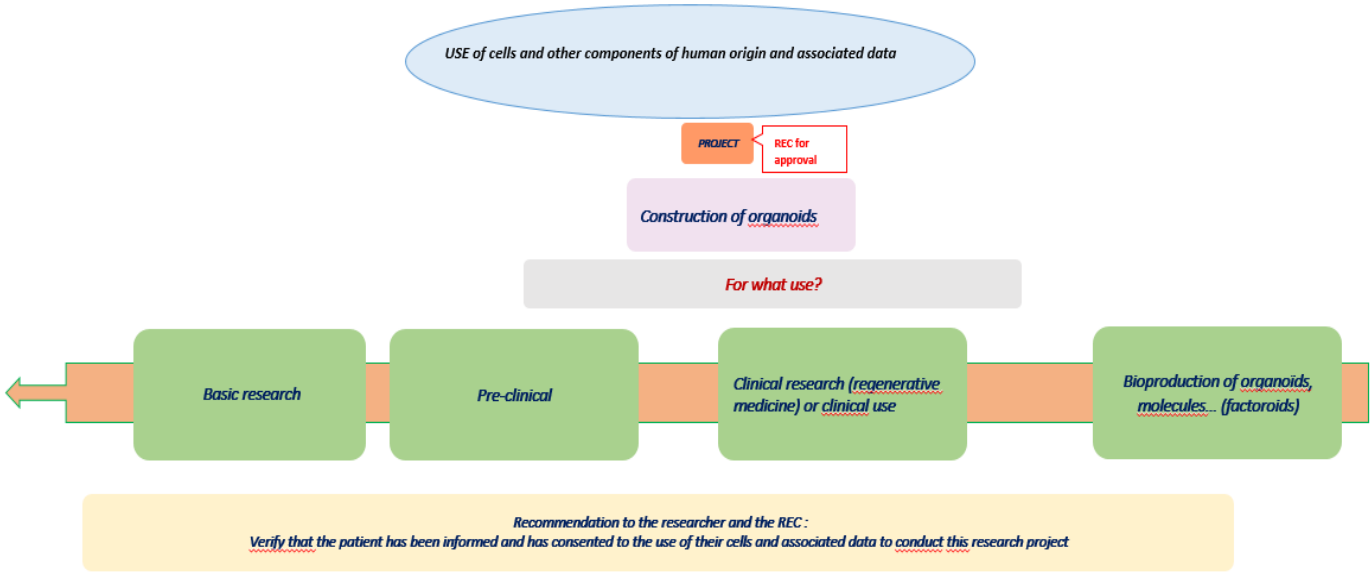
[37] *International Declaration on Human Genetic Data*, 2003, UNESCO, Article 11. Accessible at: https://www.ohchr.org/en/instruments-mechanisms/instruments/universal-declaration-human-genome-and-human-rights#:~:text=groups%20of%20people.-,Article%2011,beings%2C%20shall%20not%20be%20permitted.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

cells specific to the organ which, when placed in suitable culture and environmental conditions, self-organise into three dimensions. The origins of cells necessary to construct an organoid may vary:

- Embryonic pluripotent stem cells (ESCs) or induced pluripotent stem cells (iPSCs) are particular because they can give rise to more than 400 cell types representative of all tissues of the body and they are capable of infinite self-renewal. They also can give rise to EMSU (embryonic models for scientific use).
- Multipotent stem cells, or "adult" stem cells, are derived from foetal or adult tissues. They have a limited self-renewal capacity and display the particularity of giving rise to one or more cell types specific to an organ. In grafts, they ensure tissue renewal.
- The tumour stem cells collected from biopsies or during the surgical removal of tumours, are the source of tumoroids, which do not mimic organs but the patient's specific tumour.

6.4.2 Checklist for the type of information to be supplied to donors: three cases depending on the use of organoids



N° 3 - Tree describing the different uses of organoids

1. In the context of basic or pre-clinical research	**2. In the context of clinical research or clinical use**	**3. In the context of the industrial and/or commercial bioproduction of organoids, molecules, proteins, etc.**
Inform the participant at least in general terms about organoids and related ethical issues.	Inform the participant at least in general terms about organoids and related ethical issues.	Inform the participant at least in general terms about organoids and related ethical issues.
A statement that the study involves the collection of biological samples and associated data, and an explanation of the	A statement that the study involves the collection of biological samples and associated data, an explanation of the general framework within which they will	A statement that the study involves the collection of biological samples and associated data, an explanation of the general

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

general framework within which they will be used to construct an organoid.	be used to construct an organoid, and an explanation of the objectives of the study or the clinical use of the constructed organoid.	framework within which they will be used/commercialised to build organoids/factoroids and an explanation of the objectives of the study or use.
Precise information on the storage of biological samples: where, how and under whose responsibility. Additional information should be provided on the pseudo-anonymization of samples, including the possibility of destruction or anonymization (no destruction)	idem	idem
The expected duration of participation: o active (at the time of sampling with a description of the modalities), o duration of storage (samples and data), o duration of the organoid construction project, o duration of organoid storage.	idem	idem
A description of any reasonably foreseeable risks, discomfort or inconveniences associated with the collection of cells/tissues prior to construction of an organoid.	A description of any reasonably foreseeable risks, discomfort or inconveniences associated with the clinical trial.	A description of any reasonably foreseeable risks, discomfort or inconveniences associated with cell/tissue harvesting prior to the construction of organoids/factoroids.
A statement describing the procedures adopted to ensure the protection/confidentiality of privacy and data (personal, health, genetic, biological, etc.).	idem	idem
A statement that participation is free and voluntary and a refusal will have no impact on medical follow-up.	idem	idem
A statement that provides the subject with an opportunity to ask questions, to be informed about the use of the organoid and to be able to object to it without any consequences.	idem	idem
Information regarding the possibility and conditions of withdrawing consent.	idem	idem
Information on the persons and/or organisations organising and financing the storage and/or investigation.	idem	idem
A description of the expected benefits of this research.	idem	idem
A description of any planned genetic testing.	idem	idem
The contact details of the referring person who can be contacted to obtain appropriate answers to the participant's questions.	idem	idem

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

NA	For investigations involving risk, an explanation of the availability of treatment or compensation for harm. The insurance cover taken out by the sponsor must be mentioned, as well as contact details for the person to be contacted in the event of injury or adverse reactions.	NA
An explanation of what will happen to the data/samples and the constructed organoid at the end of the research period and whether the data/organoid will be kept or sent/sold to a third party in France or abroad for further research.	idem	idem
Information on what will happen to the results of the research and the conditions under which donors could access this information.	idem	idem
Information indicating the possibility of future re-use of the biological samples, organoid and associated data in new projects.	idem	idem
Special case of organoid biobanks: Indicate the website set up to provide patients with real-time information on any new projects.	idem	idem

6.5 Typologies of consent

6.5.1 Advantages and drawbacks of various types of consent

The consent form that must be signed by a donor of cells or tissue after receiving clear and honest information is mandatory to protect the rights of the donor regarding their biological material and personal information, but it also needs to facilitate research, not hinder it. With these two requirements in mind, four types of informed consent can be proposed, each with its own advantages and drawbacks:

1. SPECIFIC CONSENT: this type of informed consent can be defined as traditional and specific when it concerns a single project with no intended reuse of biological samples and derived organoids. This is the most commonly used type of consent because it is easy to implement. It has the advantage of offering full control by the donors over their biological samples and associated data. In this case, the researcher is limited to a single research project and cannot reuse the samples and derived biological materials for future studies without new informed consent being obtained for the new project. It should be noted that trying to contact a donor again is frequently unsuccessful. Specific consent has the drawback for research because of the destruction of samples and derived biological materials and associated data at the end of the study, thus preventing any chance of acquiring other promising results or requiring repeated requests for additional consent.

2. BROAD CONSENT: Consent can be defined as broad when it provides for the re-use of biological samples. In this context, consent must include a clear description of the

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

research areas concerned and future directions of the research (for example to perform genetic analysis), as well as the possibility of commercialisation and/or export of the organoids if this is pertinent. This type of consent is very popular in the international research community because it safeguards biological samples and associated data. On the other hand, it does not provide donors with visibility regarding the nature of future research studies or future risks and benefits at the time when they give their consent. This lack of visibility does not allow donors to fully exercise their autonomy in accepting or refusing to participate.

3. OPEN CONSENT or BLANKET CONSENT: Consent can be defined as general; biological samples and associated data are stored in a biobank for unrestricted use without transparency for the donors, who thereby waive their rights except for the right of withdrawal. Requests for samples in this context do not require prior review of the biomedical research project by an Ethics Committee. This type of uninformed consent offers no visibility to donors regarding the reuse of their samples and raises the question of protection against value violations and protection of their autonomy. This type of highly questionable consent is mentioned here because it is encountered with some commercial providers.

4. DYNAMIC CONSENT: Consent can be defined as dynamic when the scope or purpose of the project changes over time and when two-way communication is established between the donor and the user via an interface that respects the pseudonymisation principle. In the specific case of organoids, where research is rapidly evolving, a dynamic, multi-option procedure involves initial consent from the patient for the initial collection and storage of their samples/tissues and associated data in a biobank, plus continuous information on any new projects using the samples. It is an opt-out solution: the donor must make a proactive decision to withdraw. It should be noted that dynamic consent requires highly organised collection procedures, mostly developed in not-for-profit biobanks.
A biobank could thus act as a one-stop shop for the collection of information for researchers while helping to inform patients along the way via an interactive platform. In the specific case of a research project that aims to construct organoids from donor cells or cell lines obtained from donor cells (e.g. iPSC), the researcher would be asked to describe the constitution of the organoids produced and the related research. For each project, written information and a consent form will be offered online to the patient. Consequently, this type of consent requires a high degree of involvement by the donor.
The advantage of dynamic consent is that it allows donors to retain control over the use of their samples and associated data. It offers them some control over exercising their right to opt out of certain uses, but it does not guarantee that their decisions were informed by a sufficient level of understanding and information. On the other hand, with respect to biobanks, it may be an expensive undertaking to maintain and update the level of information necessary for each donation of biological samples, as well as being time-consuming.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

The HYBRIDA consortium explored which type of consent might be the best fit for organoids. Given the complexity of organoid research and its potential applications, none of the existing consent models is a perfect fit, leading to the proposal of a new type of consent implementation: the Consent For Governance Model (CFGM) [38]. In this framework, the entire consent process, including reuse, can be entrusted to a third party. An independent intermediary body is responsible for reviewing the use and reuse of samples, with the mandate to represent the rights and interests of donors, addressing issues related to specific applications. Donors retain the right to withdraw from the study but delegate consent for both initial and new research projects to this independent intermediary body. We recommend that the Ethics and Scientific Board of the Biobank either be or hire this independent committee. Alternatively, donors could be included in a governance structure to act as representatives for all donors. To facilitate the implementation of this type of consent in the organoid field and assist the third party in making informed decisions on behalf of donors regarding future use, we propose the inclusion of a "Donor's Tissue Research Under Secure Transparent Ethical Donation" (TRUSTED, see section 6.7). In this scenario, donors would have the opportunity to express their concerns and specify limits on the use of their cells.

Summary Table of the advantages and drawbacks of various types of consent

Types of Consent	Donor rights and visibility	Research/ researcher interests	HYBRIDA evaluation Adaptation/Hurdles
Traditional/Specific	Provide transparent information to the patient about: the use of their samples, the duration of the study, the destruction of their samples, associated data at the end of the study, their right to withdraw. Good protection of donor rights	Major drawback of samples and associated data being destroyed at the end of the study. Impossible for the researcher to make secondary investigations (except in the case where samples have been anonymised).	Not well suited to organoid research: * difficult to foresee how research might evolve * Need to contact the donor again if the purpose of the research is to change or if the cells are to be used in a way not foreseen in the signed consent form.

[38] Boers SN, van Delden J M, Bredenoord AL, Organoids as hybrids: ethical implications for the exchange of human tissues, *Journal of Medical Ethics*, p. 136-139, 2019. Accessible at: 10.1136/medethics-2018-104846.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

Broad	Does not ensure that donors have visibility regarding the nature of future research studies or future risks and benefits at the time they give consent. Does not protect donor withdrawal rights.	Ensures flexibility to conduct secondary research	Adapted to organoid research but does not ensure public trust.
Open or Blanket	No visibility for donors regarding reuse of their samples Raises questions of protection against value violations and the protection of donor autonomy.	Allows unrestricted use of biological samples and associated donor data. Flexible for multiple research types	Adapted to organoid research but does not ensure public trust. It is not a really informed consent
Dynamic	Ensures good visibility for donors, protects their rights, but requires significant investment, reactivity, time, and technological resources.	Flexible for multiple research types.	Adapted to organoid research but requires complicated logistics and highly educated donors with an interest in the research.
Consent For Governance Model	The trusted third party ensures the level of donor understanding and capacity to consent and guarantees	Provides mediation between the donor and researcher to enable further research.	Adapted to organoid research but requires organisation of a third-party process: time, logistics, archives, interactions, etc.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

	protection of their rights.		Combines public trust, no need for high investment from donors and flexibility of research.

6.5.2 General considerations

Several points need to be considered when choosing any form of consent, such as constant attention to data confidentiality, constant public engagement in the whole process, measures to ensure fairness and ethical institutions being involved to give ethical advice on design of a project involving organoids. Please see the Table below:

Informed Consent Principles[39]	
Privacy by design	The incorporation of privacy measures in the entire infrastructure of organoid exchange. The most appropriate privacy standards apply by default; for example, the coding of samples, governance of IT and data-sharing policies.
Participant engagement	The substantial engagement of (groups of) donors and/or the wider public in the design and continuous adaptation of biobank governance.
Benefit sharing	The fair sharing of monetary and non-monetary benefits generated through organoid exchange among all parties involved, including donors, patients and society.
Ethical oversight	The involvement of ethical oversight bodies in different contexts of organoid exchange.

The list above could be supplemented and adapted to the cultural and evolving context of organoid research and commercialisation, and several comments and improvements could be added to the proposed model.

Further developments will be needed to deal with questions regarding the use of cells or tissue derived from initial cell donations (such as iPSCs or organoids) and will depend on the choices made regarding the conditions for withdrawal of consent.

6.5.3 Duration of consent

The duration of consent may vary depending on the type of informed consent that is proposed, but this must be explicit in the document to be signed.

[39] *Organoids as hybrids: ethical implications for the exchange of human tissues*, 2019, Boers SN, van Delden JJM, Bredenoord AL, Journal of Medical Ethics p. 137.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

Information on the duration of secondary research to be supplied to the donor is complex insofar as the cell lines are immortal and may be distributed by a biobank (see paragraph 6.3.2). For example, the original hESC lines have now been in use for almost 25 years. There are often reasons to return to archived samples after many years; for example, to check for genetic drift or to study a rare disease.

In a situation where donors have agreed to the use of their samples for secondary studies for which there is not yet clear visibility, differential storage would be recommended, with options ranging from 5 years, 10 years to a lifetime to which the donor would give advance agreement.

6.6 Anticipating donor's issues: the TRUSTED questionnaire for Tissue Research Under Secure Transparent Ethical Donation

To combine our commitment to informed consent and to the respect of research conditions, the anticipation of potential uses and a follow-up procedure (third party responsible for representing the donor's interests) should be ensured.

In addition to information on the future use of the organoids being communicated to the donor, we propose that the consent form should include a questionnaire with checkboxes so that donors can decide to consent or not to different potential uses of their cells, tissues and the organoids to be generated. Once completed, this questionnaire must be pseudonymized in the same way as the donor sample and be linked to it and to the derived avatars (i.e. iPSC lines, organoids, etc.). The resulting overall "passport" should be distributed with them to any users or to structures responsible for storing and preserving the samples and/or avatars so that each user is fully aware of the authorisations and restrictions stipulated by donors regarding use of their donation. We propose to name this questionnaire TRUSTED for Tissue Research Under Secure Transparent Ethical Donation. This acronym, TRUSTED, highlights the critical elements of the donation process; Tissue Research: Specifies the type of research, emphasizing the scientific focus on tissues and cells; Under Secure Transparent Ethical Donation: Reflects the core values of security, transparency, and ethics in the donation process. It assures donors that their wishes will be respected, and their contributions will be used responsibly and with integrity.

The following is a model for a preliminary, non-exhaustive checklist:

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

Do you consent to:			
Genomic studies on your cells - limited to your mutation		yes* no	not applicable
- on your whole genome		yes*	no
Genetic modification on your cells or on their products		yes*	no
limited to your mutation		yes*	no
on your whole genome		yes*	no
Generating iPSC from your cells		yes	no
Generating organoids from your cells or from derived-iPSCs			
Organ to be mimicked	- heart	yes	no
	- lung	yes	no
	- intestine	yes	no
	- kidney	yes	no
	- liver	yes	no
	- skin	yes	no
	- pancreas	yes	no
	- brain/nervous system	yes	no
	- reproductive tract	yes	no
	- embryonic models	yes	no
	- others	yes	no
Specify which organ(s) must not be mimicked using your cells:			
The transfer of your cells, tissues or organoids to laboratory animals?		yes	no
If yes, to which species?			
All ☐ fish ☐ rodents ☐ pig ☐ dog ☐ non-human primates ☐			
The transfer of your cells, tissues or organoids to other laboratories?			
in your country yes no		to European countries	yes no
to any country yes no			
country(ies) to be excluded:			
for which type of research purposes	Fundamental	yes	no
	Applied	yes	no
	Clinical	yes	no
in which type of research structures			
	academic laboratories	yes	no
	private laboratories	yes	no

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

	any	yes	no
	Biotech companies	yes	no
	Industry	yes	no
Commercialisation		yes	no

Duration of sample storage along with associated data: 5 years ☐ 10 years ☐
Unlimited ☐
Other duration:

* Genomic testing would require specific informed consent in consideration of particular issues (transparency, privacy, unexpected findings, etc.). It might be possible to specify that if the donor responds "yes" to genomic testing, specific consent for this will follow.

Some worries about the TRUSTED list adding complexity to the system have been expressed. Some fear that listing numerous unforeseen potential uses could be seen as a false promise of organoids development that may never take place. Finally, some may question the system for handling the TRUSTED list and what might happen in the event of its enforcement. However, we have already seen that these types of restriction already exist in the field of embryonic stem cells: University of Wisconsin WiCell listed a certain number of restrictions on the use of donated materials, with annual checks that donor requests have been respected. To facilitate research and at the same time respect donor wishes, WiCell has recently changed its policy regarding Annual Certifications of the use of donated stem cells. This change is said to be consistent with the obligations of WiCell towards donors. WiCell will now be sending out annual reminders about the restricted uses of cell lines (see WiCell conditions in Annex 7). A similar process could thus be adapted to organoids and related fields.

6.7 Withdrawal of consent

As a basic principle, a donor can withdraw their consent to the use of their biological samples to construct organoids for medical and scientific research purposes. In the event of withdrawal, the initial biological sample used to construct organoids must be destroyed and all personal data deleted. Withdrawal of consent must not result in any inconvenience to the donor.

However, in view of the many practical difficulties raised by withdrawal of consent, the International Society for Stem Cell Research guidelines point out that many researchers and institutes use Informed Consent documents that only allow donor withdrawal up to the moment when the cells enter processing; for example, in the case of hiPSC generation, until the time the cells are thawed for reprogramming. Once the cells have been transformed, reprogrammed or used to generate organoids, the costs and number of samples derived from the initial tissue grows exponentially, multiple banks with various (genetic) modifications may have been generated and the cells have been widely distributed to users under MTAs that have been approved and signed in good faith by research institutions or biobanks.

Indeed, if a donor withdraws after several years while a project is still ongoing, the task of tracking down all the biological (sub)samples for destruction would be extremely costly and

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

highly unrealistic. A recommendation to avoid this difficulty is to propose possible withdrawal until the cells from the donor's biological sample are initially thawed and/or cultured but no more after their engineering.

However, from an ethical standpoint, the question arises as to how TRUSTED can be respected in such a context. As mentioned in other HYBRIDA D4 reports, public trust is strongly dependent on respecting the wishes of the regarding organoid use and the prevention of misuse. The organoid community therefore needs to engage reflection to find an acceptable consensus that on the one hand can respect scientific work and the advancement of knowledge and on the other hand implementation of the TRUSTED list.

6.8 Need for a taskforce to further analyse the new forms of consent required for research on organoids and related fields

The type of consent needs to be adapted to the context of research and its evolution, and be sensitive to both donor rights and research continuity. Following our analysis, we consider that consent entrusted to a third party (CFGM) should be further explored, in view of its respect for donor privacy and rights and also for the work of researchers in a time-related perspective, thus ensuring continuity of their efforts.

Because of the complexity of defining the best strategy to achieve informed consent (for both participants and researchers), the best type of consent, as well as the numerous questions regarding the withdrawal of consent and the specificity of a rapidly changing field such as organoid research, the HYBRIDA consortium considers that this task goes beyond its remit and suggests the creation of an EU taskforce to focus on these very particular aspects. We suggest EU to name it the Public Advisory Committee for Organoid Research (PACOR) and to set it inside an existing agency to provide oversight and input into the direction of organoid research. The PACOR should monitor the implementation of current recommendations and pursue reflection and analysis regarding the typology and particularities of informed consent in the organoid field. The PACOR should implement transparent research agendas and reporting. The PACOR should be a multidisciplinary working group that can guide discussions and represent the various interests in the organoid community; for example, it should include biobank board members, BBMRI-ERIC members (European research infrastructure for biobanking), representatives of patient associations, expert scientists working on organoids, legal experts and ethicists.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

7 OPEN ETHICAL QUESTIONS

In the rapidly evolving field of organoids, we have identified a certain number of open questions (i.e. there is no easy answer or practical process that can be proposed immediately) that raise specific ethical dilemmas and that will require further elaboration. However, in the framework of our ethical analysis we feel it is unavoidable to address such questions. Further ethical concerns will arise as technologies evolve towards more sophisticated and integrated systems involving diverse interconnected organoids and their hybridization with non-biological systems. At the date of Operational Guidelines publication, four open questions are already clearly identified:

- *7.1 Nomenclature and moral status of embryonic models*
- *7.2 Functions of nervous system organoids and assembloids and their relationship with sentience, pain, consciousness*
- *7.3 Naming organoids according to specific fields*
- *7.4 Organoids as marketable commodities: the commercial status and ownership of cells/tissues.*

7.1 Embryonic models ethical questioning

Recent years have seen the development of embryo models generated from aggregates of pluripotent stem cells that exhibit aspects of embryonic development. Such embryo models are not models of organs like organoids but models of organisms. They are useful to study mammalian development. We consider that they are covered by the work of the HYBRIDA project for two main reasons:

1. From a biological point of view, they share many commonalities with organoids.
2. From an ethical point of view, they raise important moral issues that need to be addressed, especially as they are of concern to the legislators and the public.

We take here the definitions developed in the most recent ISSCR recommendations concerning stem cell-based embryo models:

> "Advances in cellular engineering make possible the assembly, differentiation, aggregation, or re-association of cell populations in a manner that models or recapitulates key stages of embryonic development. Such experimental systems can provide essential insights into embryo and tissue development but raise concerns when such structures achieve complexity to the point where they might realistically manifest the ability to undergo further integrated development if cultured for additional time in vitro. There are two types of stem cell-based embryo models.
>
> Non-integrated stem cell-based embryo models: These stem cell-based embryo models will experimentally recapitulate some, but not all aspects of the peri-implantation embryo, for example differentiation of the embryonic sac or embryonic disc in the absence of extraembryonic cells. These stem cell-based embryo models do not have any reasonable expectations of specifying additional cell types that would result in formation of an integrated embryo model. Gastruloids are an example of a non-integrated stem cell-based embryo model.
>
> Integrated stem cell-based embryo models: These stem cell-based embryo models contain the relevant embryonic and extra-embryonic structures and could potentially achieve the complexity where they might realistically manifest the ability to undergo further integrated development if cultured for additional time in vitro. Integrated stem cell-based embryo models could be generated from a single source of cells, for example expanded potential human pluripotent stem cells capable of coordinately differentiating into embryonic and

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

> extraembryonic structures. Alternatively, integrated stem cell-based embryo models could also be generated through the formation of cellular aggregates where extraembryonic/embryonic cells from one source are combined with embryonic/extraembryonic cells from different sources to achieve integrated human development. This might include using non-human primate cells as one of the sources. Previous restrictions on preimplantation human embryo culture (the "14-day/primitive streak rule") were not written to apply to integrated stem cell-based embryo models. Thus, these guidelines specify the imperative for specialized review when such research is designed to model the integrated development of the entire embryo including its extraembryonic membranes. A guiding principle of review should be that the integrated stem cell-based embryo models should be used to address a scientific question deemed highly meritorious by a rigorous review process. Blastoids are an example of an integrated stem cell model."[40]

The recent development of so-called "synthetic mouse embryos" by the teams led by Jacob Hanna and Magdalena Zernicka-Goetz has raised major ethical concerns among the scientists, particularly because one biotech company plans to make "human synthetic embryos" in the near future[41]. As there are no precise regulations which cover this emerging "synthetic" technology, the field of bioethicists must move to encompass issues concerning natural human embryo research and related questions and regulations. Debating the moral status of a human embryo might prove a valuable first step to consideration of their surrogates and subsequent regulatory framework.

In this sense, the HYBRIDA Consortium decided to dedicate several debates and a joint working group to the development of different scientific embryo models. In order to better understand the use and development of various embryo models, please refer to the typology below.

7.1.1 When studying the properties of embryos, several types of entities are likely to be involved:

a. **Embryonic models for scientific use** (EMSUs.) They can be created because stem cells are capable of forming structures that recapitulate aspects of embryo organisation and development. Some authors such as John Aach have referred to the emergence of "synthetic embryology".

b. **Chimeras.** These are organisms that contain at least two groups of genetically different cells that come from individuals or different species (intraspecies or interspecies chimeras). They are obtained by introducing pluripotent stem cells, embryonic stem cells (ESC) or iPS cells into an embryo (blastocyst). Each cell population retains its own genetic character and the result is a mosaic. Interspecies chimeras notably include human-animal chimeras (human embryo into which animal cells are introduced) and animal-human chimeras (animal embryo comprising human cells).

---

[40] International Society for Stem Cell Research, *Guidelines for Stem Cell Research and Clinical Translation, Laboratory-based Human Embryonic Stem Cell Research*, Embryo Research and Related Research Activities Chapter, 2021. Accessible at: isscr-guidelines-for-stem-cell-research-and-clinical-translation-2021.

[41] CBN News. *Israeli Biotech Firm Plans to Create Human Embryos to Harvest Organs, Field Experts Say There are Ethical Concerns*, 16/09/2022. Accessible at: https://www1.cbn.com/cbnnews/health/2022/september/israeli-biotech-firm-plans-to-create-human-embryos-to-harvest-organs-field-experts-say-there-are-ethical-concerns.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

c. **Hybrids and cybrids**. A hybrid is formed when a spermatozoa from one individual is used to fertilise an ovum from another individual of a different species. As a consequence, each cell of the hybrid organism contains chromosomes from both species. A cybrid is a cytoplasmic hybrid created when the nucleus of a cell from an organism is introduced into an enucleated ovum of an individual from another species or the same species. A cybrid is a virtual-clone of the organism whose nucleus has been transferred. Hybrids and cybrids are often, inaccurately, referred to as chimeras. We should emphasise once again that many countries ban the creation of entities which combine human and animal genetic heritage.

d. **Cloning** is a procedure used to identically reproduce the initial biological entity (e.g. monoclonal antibodies) so that all are identical. Nuclear transfer may be one technical stage in cloning if the purpose is reproductive and transfer is between two syngeneic animal cells (with the same genome). This is not the case for scientific research where the embryo must be destroyed at the end of the experiment and the embryonic entity constructed is not similar to the embryo that supplied the transferred nucleus.

e. **Parthenotes** are embryos obtained through parthenogenesis, i.e. through the division of an unfertilised female gamete.

f. **Embryos constituted through the micromanipulation** of constituent cell elements (e.g. mitochondrial donation) or by eliminating some of their constituents (e.g. restoration of diploidy). Mitochondrial replacement therapy is a case of intraspecies cybrid.

g. **Embryos created for research by IVF**.

7.1.2 Two principal ethical issues can be identified

A) Inappropriate naming

As the writer and philosopher Albert Camus said, "to misname an object is to add to the misfortune of this world *"mal nommer un objet, c'est ajouter au malheur de ce monde"*. In the case of new technologies, the terms "synthetic", "embryo-like", "human entities", etc., should be prohibited. As mentioned in several publications quoted below, such as the ISSCR Guidelines or INSERM Ethics Committee articles[42] on the ethics of embryo models from 2019, the use of "synthetic embryos" or "stem cell-derived embryos" does not help to convey a clear message to the public reflecting the state of scientific innovations. An accurate description of the models described in Hanna and Zernicka-Goetz's works would be "E8.5 mouse embryo models".

For an ethical overview to be intelligible, transparent and accountable, researchers must report on their research using clear and well-defined terms[43]. At present, the models are rudimentary and imperfect. They only partially reflect the *conceptus* and they lack the ability to develop

[42] Bernard Baertschi, Marc Brodin, Christine Dosquet, Pierre Jouannet, Anne-Sophie Lapointe, Jennifer Merchant, Grégoire Moutel, Research on Embryos and Embryonic Models for Scientific Use (EMSUs), Accessible at: https://www.hal.inserm.fr/inserm-02373609/document.

[43] Kirstin R W Matthews, Daniel S Wagner, Aryeh Warmflash, Stem cell-based models of embryos: The need for improved naming conventions, *Stem Cell Reports*, 2021. Accessible at: https://pubmed.ncbi.nlm.nih.gov/33770498/.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

into a living organism[44]. Because of these limitations, these models are typically not included, either biologically or legally, in the class of embryos in the great majority of jurisdictions. The term "synthetic" or "artificial" embryo is sometimes used to refer to these structures. However, in order to accurately reflect the state of research, the ISSCR suggests using the term "embryo model" instead[45]. The reason is that these models form from stem cells that spontaneously but imperfectly unleash their intrinsic potential to re-enact developmental processes. They are thus neither synthetic nor artificial but rather reflect potent cells attempting to "act naturally" by expressing their potential.

Moreover, terms such as "synthetic" indicate the use of non-natural elements (obtained by synthesis) while, at the same time, erroneously indicating that we are dealing with an embryo proper. Such inadequate terminology implies a value judgment that these are embryos, although of an outlandish type. The terms "synthetic" and "artificial" also belie the historically evolving understanding of the embryo by suggesting that the same structure is "natural" or "unnatural" solely because of its origin[46]. Furthermore, "synthetic" is an adjective that often has negative connotations. The Merriam-Webster definition of synthetic[47] is:

a (1)**:** of, relating to, or produced by chemical or biochemical synthesis especially**:** produced artificially synthetic drugs, synthetic silk

(2)**:** of or relating to a synfuel

b**:** devised, arranged, or fabricated for special situations to imitate or replace usual realities

c**:** factitious, bogus.

John Aach also proposed the term Synthetic Human Entities with Embryo-like Features (SHEEFs)[48]. To speak of "human entity" brings to mind a human being, which these models are not. They are human in the sense that the genome of their cells is human, but they do not have all the characteristics that make a human embryo a human being.

Worst of all should be "Mini-Mes". All authors rightly consider that this expression is meaningless to characterise these entities. Moreover, even natural embryos are not mini-mes, which implies a personal identity that does not yet exist.

Finally, to clarify and facilitate an understanding of the difference between a naturally occurring phenomenon and the scientific artefact used to model it, in its 2019 article[49] the INSERM Ethics Committee proposed "embryonic model" rather than "embryo model". Indeed

[44] Eszter Posfai, John Paul Schell, Adrian Janiszewski, Isidora Rovic, Alexander Murray, Brian Bradshaw, Tatsuya Yamakawa, Tine Pardon, Mouna El Bakkali, Irene Talon, Natalie De Geest, Pankaj Kumar, San Kit To, Sophie Petropoulos, Andrea Jurisicova, Vincent Pasque, Fredrik Lanner, Janet Rossant, Evaluating totipotency using criteria of increasing stringency, *Nature Cell Biology*, 2021. Accessible at: https://www.nature.com/articles/s41556-020-00609-2.

[45] Amander, T Clark, Ali Brivanlou, Jianping Fu, Kazuto Kato, Debra Mathews, Kathy K Niakan, Nicolas Rivron, Mitinori Saitou, Azim Surani, Fuchou Tang, Janet Rossant, Human embryo research, stem cell-derived embryo models and in vitro gametogenesis: Considerations leading to the revised ISSCR guidelines, *Stem Cell Reports*, 2021. Accessible at: https://www.ncbi.nlm.nih.gov/pmc/articles/PMC8190666/.

[46] Ball, P. Synthetic biology: starting from scratch, *Nature* 431, 624-626, 2004. Accessible at: 10.1038/431624a.

[47] Merriam Webster Dictionary. Accessible at: https://www.merriam-webster.com/dictionary/synthetic.

[48] J. Aach, J. Lunshof, E. Iyer and G. Church, Addressing the Ethical Issues Raised by Synthetic Human Entities with Embryo-like Features, eLife, 2017: 1-20.

[49] Bernard Baertschi, Marc Brodin, Christine Dosquet, Pierre Jouannet, Anne-Sophie Lapointe, Jennifer Merchant, Grégoire Moutel, *Research on Embryos and Embryonic Models for Scientific Use* (EMSUs), Accessible at: https://www.hal.inserm.fr/inserm-02373609/document.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

"embryonic" indicates both "relating to an embryo" and being in an early stage of development**:** *incipient*, *rudimentary* (adapted from Merriam-Webster).

B) The moral status of embryonic models

The use of embryonic models enables the gathering of scientific knowledge and eventually societal benefits, while avoiding the constraints normally associated with the use of embryos. Embryonic models, integrated and non-integrated are not embryos: this is their interest from an ethical point of view, as they make it possible to avoid the still-debated issue of embryo research and the status of the embryo (remember, however, that in our countries, it is legal to carry out experiments on spare embryos).

Scientific advances are nevertheless narrowing the gap between models and embryos and, consequently, the moral and legal gap between models and embryos. For example, Australia now considers that embryonic models are similar to embryos. Embryonic models will eventually pass a tipping point after which there is no reason to value and regulate them differently from embryos: they will become the scientific and moral equivalent of embryos. Put differently, at a certain point in experimental sophistication, embryonic models could pass a "Turing Test", which entails that an evaluator who lacks information about the origins of the embryonic model would not be able to tell it apart from a human embryo.[50] To our mind, from this point on any meaningful distinction between the embryonic model and the embryo, biologically as well as morally, has disappeared. We therefore propose that a more truthful path for terminology is to name them *embryonic models*, thus recognizing their current limits, but that once they are assumed equivalent, they will become fully entitled as embryos regardless of the way they were formed.

The degree to which embryonic models approach this tipping point is of course dependent on how an embryo is defined. In other words, an accurate description of scientific advances, the adequate formulation of clinical research aims and the quality of moral reasoning and policymaking pertaining to embryo models will require a definition of the embryo that is appropriate for these purposes. For the time being, legislations adopt varying definitions of what an embryo is, which need to be overcome. A general definition has been used by the ISSCR: "In this document, the term 'embryo' is used generically to describe all stages of development from the first cleavage of the fertilized ovum to nine weeks post fertilization in the human, including the placenta and other extraembryonic membranes."[51]

Once this point of semantics has been settled, the trickiest question will be to determine the criteria for determining the conditions under which an embryonic model will have become equivalent to an embryo, at which point the tipping point will have been reached. A direct

[50] Rivron NC, Martinez Arias A, Pera MF, Moris N, M'hamdi HI. An ethical framework for human embryology with embryo models. Cell. 2023 Aug 17;186(17):3548-3557. doi: 10.1016/j.cell.2023.07.028. The expression "Turing test" is used by analogy with the test that Alan Turing devised in the field of artificial intelligence: "Turing envisioned a game in which he can communicate with a teletype in another room. Controlling this second console would be either another human or a digital computer. The player could ask any question he wished through his console in order to determine whether he was in contact with a man or a machine" (D. Bolter, *Turing's Man*, Penguin Books, 1986, p. 191). With the progress of algorithms, there will come a time when it will no longer be possible to tell whether we are in contact with a human being or a machine. Applied to the case of embryonic models, the test amounts to determining a tipping point at which we can no longer distinguish, by virtue of its properties and behavior, an embryonic model from an embryo.

[51] ISSCR, *Guidelines for Stem Cell Research and Clinical Translation*, 2021, p. 64.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

Turing test would simply be to transfer such an embryonic model into a uterus. However, such a procedure would be legally and morally unacceptable. Nicolas Rivron and his colleagues therefore propose two indirect tests. The first is to evaluate its development in vitro over a certain period of time (for the embryo, the countries that accept the conclusions of the Warnock report limit this to 14 days; there are proposals to extend this period up to 28 days).[52] The second is to examine how a similar animal embryo model develops when transferred to an animal uterus, and whether it gives rise to a living and fertile animal.

In the meantime, we think it's worth reiterating the importance of the principle of subsidiarity, as the Agence de la biomédecine does; it stipulates that "the means used be strictly necessary to achieve the intended objectives", which means that "it should be conducted on the minimum structure necessary to answer the question: [for example] a cardiac organoid, not an embryoid."[53]

7.2 Nervous system functions: organoids and assembloids and their relationship with sentience, pain, consciousness

7.2.1 Does a neural organoid have a moral status?

As specified by Marie-Anne Warren: "to have moral status is to be morally considerable, or to have moral standing. It is to be an entity towards which moral agents have, or can have, moral obligations. If an entity has moral status, then we may not treat it in just any way we please."[54] We have just seen what this means for embryonic models; but the question also arises for neural organoids: do they have a moral status, and if so, what is it?

As previously said[55], according to the scientific literature and the results of stakeholder consultations, there is at this stage no reason to believe, or evidence to suggest, that the neural organoids that have been created resemble a fully functioning brain or integrated parts of the brain. The ISSCR stresses also this point.[56] There is therefore no reason to believe that such organoids possess sentience or will achieve a level of consciousness that warrants special ethical or legal concern (i.e. that confers on them a moral or legal status). Nevertheless, certain neural organoids can mature and become more complex when combined with other organoids in complex neural assembloids. Regulatory questions regarding the normative status of these entities, and user's obligations to them, may therefore arise.

The arguments

Present-day neural organoids only replicate a particular region of the brain, and not the brain in its entirety;[57] in particular, only two types of cells are found: neurons and astrocytes. Even

[52] Agence de la biomédecine, *Opinions of the Conseil d'orientation: stem cell-based embryo models*, september 21, 2023, p. 7, available at: https://www.agence-biomedecine.fr/IMG/pdf/22-06_avis_du_co_embryoi_des_eng-2.pdf
[53] Agence de la biomédecine, *ibid.*, p. 6.
[54] Mary Anne Warren, *Moral Status*, Oxford, OUP, 1997, p. 3.
[55] WP6, HYBIDA Project, D 6.1, p. 10.
[56] ISSCR, *Guidelines for Stem Cell Research and Clinical Translation*, 2021, p. 10.
[57] Chen & al., "Transplantation of Human Brain Organoids: Revisiting the Science and Ethics of Brain Chimeras", *Cell Stem Cell*, 2019, vol. 25, p. 463.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

when considering whole brain organoids, their volume is only 1/1"000 of a mouse brain and 1/1"000"000 of that of a human being. Moreover, they have no mature neural networks and so are unable to interact with their environment.[58] Consequently, neural organoids cannot possess mental states such as sentience and consciousness.

More generally and importantly, neural organoids are not functioning organs within an organism. This implies that even a brain (a fully functioning brain or integrated parts of the brain) cannot think, because thinking is a function of the organism as such, when it is situated in an environment that supplies it with stimuli through receptors. A brain cannot be conscious of anything, nor can it have the slightest sentience that could be translated at a psychic level. The same applies to neural organoids; thus, although it is true that "no one knows how many neurons it would take for a distinctively human thought to emerge",[59] this is because the question does not make sense.

The situation may be different when neural organoids are transplanted into the brains of animals such as rats or pigs, which have mental states and are sentient.[60] Their chimeric brain is then an organ functioning within an organism. It has already been observed that this transplantation normalises expression of the genes of neurons, which is altered in *in vitro* cerebral organoids.[61] But what will be the moral status of these "humanized" animals, i.e. these chimeras? At first glance, there is no answer to this question. Here we are confronted with the following problem: these chimeras will have to be human enough to be used as a research model and then, if all goes well, for therapeutic purposes, but not human enough to fall under the protection enjoyed by human beings.[62]

7.2.2 The nature of consciousness and sentience

If one day organoids, assembloids or organisms constructed out of them become sentient or have consciousness, then they will acquire a moral status.

But an immediate question arises: what does it mean to possess sentience or to be conscious? There is no consensus on the meaning of these terms. For neurologists, consciousness is a two-fold concept, defined by wakefulness and awareness. Éric Racine specifies that "wakefulness is basically equated to arousal […] and consists of mechanisms that keep the patient awake […]. Awareness refers to the content of consciousness or the awareness of self and environment, including functions such as emotions, thoughts, and sensory experience".[63] Ned Block, from a philosophical and psychological point of view, distinguishes two general categories of consciousness, access consciousness and phenomenal consciousness. Access consciousness includes all the representations we have, such as our thoughts or desires, which all have a content, i.e. an intentional object, and which can be used by our executive system

[58] Munsie & al., "Ethical Issues in Human Organoids and Gastruloid Research", *The Company of Biologists*, 2017, vol. 144, p. 943.
[59] W. Cheshire, "Miniature Human Brains: An Ethical Analysis", *Ethics and Medicine*, 2014, vol. 31/1, p. 9.
[60] Yeager, "As Brain Organoids Mature, Ethical Questions Arise", *The Scientist*, August 1, 2018, https://www.the-scientist.com/features/brain-organoids-mature--raise-ethical-questions-64533.
[61] D. Kwon, "Organoids Don't Accurately Model Human Brain Development", The Scientist, October 23, 2019, https://www.the-scientist.com/news-opinion/organoids-dont-accurately-model-human- brain-development-66629.
[62] U. Lee McFarling, "Near the Campus Cow Pasture, a Scientist Works to Grow Human Organs" – in Pigs, *Stat*, October 20, 2017, https://www.statnews.com/2017/10/20/human-pig.
[63] É. Racine, *Pragmatic Bioethics*, Cambridge MA, MIT Press, 2010, p, 141-142.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

(decisions, actions); it embraces also second-order capacities such as monitoring consciousness (reflective consciousness) and self-consciousness. Phenomenal consciousness is the subjective experience of what we do and what happens to us. As we can see, an entity can very well possess phenomenal consciousness and be sentient (for example, feel pain), without being self-conscious.

Sentience, that is the capacity to experience pleasure and pain[64], is a form of phenomenal consciousness: "Sentient subjects have an interest in avoiding negative conscious states and experiencing positive ones"[65], but there are much less demanding conceptions of it, notably the one adopted by Brett Kagan and his colleagues, in an article entitled: "In vitro neurons learn and exhibit sentience when embodied in a simulated game-world". They claim that "neural cultures meet the formal definition of sentience as being *responsive to sensory impressions*, specifying that two interrelated processes are required for sentient behaviour in an intelligent system. Firstly, the system must learn how external states influence internal states via perception and how internal states influence external states via action. Secondly, the system must infer from its sensory states when it should adopt a particular activity and how its actions will influence the environment"[66].

Such a broad and formal conception, however, seems to amount to a *reductio ad absurdum*, just as when John McCarthy argued that a thermostat had beliefs, that is, mental states: "A simple thermostat that turns off the heat when the temperature is a degree above the temperature set on the thermostat, turns on the heat when the temperature is a degree below the desired temperature, and leaves the heat as is when the temperature is in the two-degree range around the desired temperature. The simplest belief predicate B(s,p) ascribes belief to only three sentences: "The room is too cold", "The room is too hot", and "The room is OK" – the beliefs being assigned to states of the thermostat in the obvious way. We ascribe to it the goal, "The room should be ok". When the thermostat believes the room is too cold or too hot, it sends a message saying so to the furnace".[67] The absurdity of attributing mental states to a thermostat is matched by the absurdity of attributing sentience to a neural culture. When we twist the definitions in this way, anything becomes possible.

7.2.3 Bis repetita…

This is not the first time that mental states have been attributed to entities that cannot possess them, although it is fair to say that this sometimes has a metaphorical flavour. In the 18th century, Maupertuis criticized the Cartesian explanation for the formation of the living in these terms: "This great philosopher [Descartes] in his treatise on man thought he could explain how, by the laws of movement and fermentation alone, a heart, a brain, a nose, eyes, etc. were

[64] A. Jaworska and J. Tannenbaum, "The Grounds of Moral Status", *The Stanford Encyclopedia of Philosophy* (Spring 2023 Edition), Edward N. Zalta & Uri Nodelman (eds.), URL = <https://plato.stanford.edu/archives/spr2023/entries/grounds-moral-status/>.

[65] A. Lavazza & M. Massimini, Cerebral organoids and consciousness: how far are we willing to go?, *Journal of Medical Ethics*, p. 1, 2018. Accessible at: 10.1136/medethics-2018-104976.

[66] B. Kagan & al., "*In vitro* neurons learn and exhibit sentience when embodied in a simulated game-world", *Neuron*, 2022, vol. 110, p. 1-2.

[67] John McCarthy, *Ascribing Mental Qualities to Machines*, University Computer Science Department Stanford, 1979. Accessible at: http://jmc.stanford.edu/articles/ascribing/ascribing.pdf.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

formed".[68] However, the formation of a living being does not occur by a juxtaposition of parts regulated by a mechanical movement, and to account for it, even chemical attraction is insufficient: "If all [the parts of matter] have the same tendency, the same force to unite, why do these form the eye, why these the ear? Why this marvellous arrangement and why don't they all unite in a jumble?" It is therefore necessary, according to Maupertuis, to add a new property to matter, directing the living molecules and uniting them with those that are likely to form with them such an organ, such an organism; it will be "something similar to what we call desire, aversion, memory".[69]

But what is this property and of what does it consist? Maupertuis did not say, but his contemporaries, following the physician Albrecht von Haller, identified it as sensibility or sentience [*sensibilité*].

For Henri Fouquet, author of the article on *Sensibility* in the *Encyclopaedia*, sensibility is nothing else, "in the living body, than a property that certain parts have of perceiving the impressions of external objects, and of producing consequently movements proportionate to the degree of intensity of this perception".[70] And he justifies his assertion by the fact that, in order to contract, a living part, whether or not it is detached from the organism, whether or not it is innervated and linked to a central nervous system, must indeed perceive in some way the irritating substance or body: "This action must necessarily be perceived or felt by the part, and what is more, appropriate to the feeling of that same part; and what other animal power than sensibility will be able to be the judge of sensible bodies applied to a living body?"[71] Fouquet is thinking here of the heart of a frog that continues to beat for some time when immersed in salt water: such a heart perceives the irritant (salt) and reacts to it (it contracts).

Thus described, sensitivity is a quasi-psychic property, which is a way of returning to the origins, since, before Haller – and acknowledging his debt to him – it was Glisson who, as early as 1654, spoke of irritability and explained it in terms of perception, appetite and movement, since it is necessary for the irritated organ to perceive the cause that acts on it in order to defend itself.

7.2.4 Searching for criteria

In order to possess moral status, a neural organoid has to have a form of consciousness, in particular sentience (a kind of phenomenological consciousness). This is not the case for current organoids, but as was the case for embryonic models, the question should come up in the future. So, here too, the question arises in terms of tipping point and Turing test. So, when the question of organoids possessing a form of consciousness really arises in the future, how will it be answered? The difficulty is that we do not have direct access to the consciousness of others, and therefore *a fortiori* to that of animals or any other entity which could possess it. However, indirect means are possible, applicable by analogy to neural organoids; for example, we already have criteria to test the presence of consciousness in animals and in comatose people: "The challenge of detecting consciousness in brain-injured, comatose patients shares

[68] Pierre Louis Moreau De Maupertuis, *Vénus physique*, 1745, Paris, Aubier, 1980, p. 109.
[69] *Système de la nature*, 1756, Paris, Vrin, 1984, p. 146-147.
[70] In D. Diderot D. & J. d'Alembert, Encyclopédie, t. XV, 1765, p. 38.
[71] *Ibid.*, p. 50-51.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

some basic similarities with the issue posed by cerebral organoids".[72] But it should be remembered that, at present, no neural organoid has the size and organization that constitute the conditions of consciousness in animals and human beings.

7.3 Naming of organoids

An organoid does not have the same properties and functions as an organ. It is therefore incorrect to refer to organoids as mini-organs, and communicating incorrect information constitutes misconduct. Misnaming is both prejudicial for the advancement of science and for the trust of the public. The HYBRIDA project recognizes the effort of the scientific community to raise a consensus for an adequate nomenclature for nervous system organoids and assembloids. We endorse the recently proposed nomenclature consensus for nervous system organoids and assembloids[73] . These authors proposed a simple set of guidelines for naming that is rooted in developmental neuroanatomy. We anticipate that this effort will facilitate communication and scientific advancement, promote collaborations, and initiate the development of quality control measures and benchmarking in the field.

Three main classes of 3D models are used in the field at present: organoids, assembloids and grafted organoids. As we do in the HYBRIDA project, authors define organoids as in vitro-generated cellular systems that emerge by self-organization, include multiple cell types, and exhibit some cytoarchitectural and functional features reminiscent of an organ or organ region. Concerning nervous system organoids, they may be constructed from pluripotent stem cells but can also be derived from donor tissues with growth potential (such as glioblastoma organoids). They can be generated as 3D cultures or by a combination of 3D and 2D approaches (also known as 2.5D) that can develop and mature over long periods of time (months to years). Engineered 3D cultures obtained by bioprinting that enable and/or sustain self-organization would also be considered as organoids. Authors refer to nervous system organoids by the major anatomic region they model, such as cortical organoids, retinal organoids, hypothalamic organoids or spinal cord organoids. The terms neural organoids and nervous system organoids each encompass a broader spectrum of cell types, including those from the central and the peripheral, autonomic and enteric nervous systems, and are appropriate as blanket terms or to describe organoids encompassing several regionally diverse cell types. These authors define assembloids as self-organizing cellular systems resulting from the combination of a type of organoids with another type of organoids (for example, dorsal forebrain with ventral forebrain) or with different specialized cell types (for example, cortical organoid with endothelial cells) that result in integration. Finally, organoids or assembloids that have been transplanted into animals should be named grafted organoids or grafted assembloids.

The HYDRIDA project considers of tremendous value that scientists should provide the details of methods to obtain organoids. Accordingly, naming should reflect such details recognizing two main categories of neural organoids on the basis of the level of guidance provided during the differentiation of pluripotent stem cells into organoids. By guidance we mean the addition

[72] A. Lavazza & M. Massimini, Cerebral organoids: ethical issues and consciousness assessment, *Journal of Medical Ethics*, 2018, p. 3.
[73] *Ibid*., p. 7.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

of small molecules or factors intended to generate a particular region or collection of cell types. If organoids are obtained by selecting parts of organoids by using genetic reporters, cell sorting or mechanical dissection they should be named as selected neural organoids.
Interestingly, in their general recommendations, authors of the consensus nomenclature support the elements of MIAOU and Echoes detailed in HYBRIDA section 4. Furthermore, they strongly reject misnaming such as mini-brain, brain-in-a-dish, whole-brain organoids, humanized animals. Finally, they call for a precise description allowing a real dialog between scientists, ethicists and the public « *We do not endorse the use of terms such as fetal-like or the unsubstantiated use of terms describing complex, emergent processes of the nervous system. Regarding the use of terms commonly used to define complex mental, cognitive and behavioural processes, such as sentience, intelligence, learning and pain, we encourage authors to explicitly define cellular activities in the isolated system, rather than using them to infer that organoids are learning or sentient bodies. For example, cells in retinal organoids respond to light and exhibit aspects of phototransduction, but we should not suggest that these organoids have vision or sight.* »

For the Research field

Do not use the term "mini-organ" to designate organoids: A consensus workshop should define the best way to characterize the organoid subtype. An example can be found with neural organoids as developed in 7.3.

Do not use the term human on chip (prefer avatar on chip) or organ on chip (prefer neural circuit on chip, etc., or physioid on chip if there are interconnections between organoids).

For the Bioproduction field

- Do not use the term organoid but rather “factoroid” or “manufacturoid” in the context of bioproduction.
- As for the production facilities, production processes by factoroids must be validated by the regulatory agencies according to what is being produced (medicine, dietary supplements, graft, etc.).
- Apply the mode of thinking already in place for the factory of the future.
- Based on the model of factories of the future, work will be necessary regarding the management of *factoroid* banks. A mission is necessary to visit existing bioproduction centres[74].

For the Pre-clinical field

- Build a statistical toolbox linked to the use of innovative approaches (validation of care protocols rather than of molecules on more or less precisely stratified cohorts).
- Define the advantages and drawbacks (risk/benefit ratio) regarding the use of organoids (e.g., blood-brain barrier or intestinal barrier) in relation to the models currently accepted by the regulatory agencies.
- Based on the model of testing centres, preclinical CRO and the Hubrecht Institute, work will be necessary regarding the management of organoid banks (conservation, storage of organoids and associated data). A mission to visit existing reference centres.

[74] Such as the Yposkesi example of therapeutic vector and cell batch production. Accessible at: https://www.yposkesi.com.

This project has received funding from the European Union’s HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

For the Clinical field

Define GMP quality levels regarding the clinical use of organoids for all three objectives. Be inspired by what exists for cell therapy. Keep in mind that this is a guidelines document.
- Do not use the concept of “symbiote”, but rather ”innovative medical bio-devices" (possibly linked to digital medical devices-DMD-). This relates to the section on the Code of Conduct for Research Integrity (Interaction with the public).

7.4 Ethical and Legal Ownership of Organoids: A complex Landscape

The ethical and legal landscape regarding the ownership of human tissues once they have been removed from the body is complex and varies across jurisdictions. Legal precedents in countries such as Australia, France, the UK, and the USA[75, 76] generally indicate that individuals do not retain ownership rights over their tissues post-removal, particularly when these tissues have undergone significant manipulation by researchers. This principle has significant implications concerning the development and use of organoids derived from patient stem cells.

Intellectual Property Rights and Organoids

When patient-derived cells are used to create organoids, the question arises: who owns the resulting organoid? The answer hinges on the significant manipulation and innovation involved in developing these structures.

1. Transformation and Innovation: under existing legal principles, the entity (usually a research institution or biotech company) that invests resources and expertise into transforming basic cells into complex organoids is typically considered to hold the Intellectual Property (IP) rights. This transformation process adds significant value and complexity to the original biological material.

2. Patentability: organoids, due to their innovative nature and potential applications in drug testing, disease modeling, and personalized medicine, can be patented. However, the patenting process must navigate ethical and legal boundaries set by EU regulations, particularly concerning the origin of the biological material and the rights of the original donors.

3. Balancing Innovation and Ethics: while IP rights incentivize innovation by protecting and rewarding creators, they must be balanced with ethical considerations. This balance is essential in the EU context, where the non-commercialization of the human body and its parts principle is strongly emphasized.

Ownership and Legal Precedents

In numerous legal systems, the act of donating tissues or cells typically transfers ownership to the entity receiving them, often a research institution or medical facility. For example, the landmark case of « Moore v. Regents of the University of California » in the United States set a precedent where the court ruled that a patient did not own the cells removed from his body

[75] Gold E. *Body parts: property rights and the ownership of human biological materials*. Washington, D.C.: Georgetown University Press; 1997.
[76] *Fiduciary Duty of Researchers - the Spleen Case - Moore v. Regents of University of California*, 793 P.2d 479 (Cal. 1990) [Internet]. 1998. Available from: https:// biote ch. law. lsu. edu/ cases/ consent/ Moore_v_ Regents.

This project has received funding from the European Union’s HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

that were subsequently used for profitable research and development. The court's decision hinged on the transformation and value added by the researchers' manipulation of the cells.
Similarly, in the UK, the Human Tissue Act 2004 regulates the removal, storage, and use of human tissues, but it does not explicitly grant ongoing ownership rights to the donor once the tissues have been removed and used for research. Australia's legal stance is also aligned with these principles, where the National Health and Medical Research Council (NHMRC) provides guidelines that typically place ownership with the institution conducting the research.

In France, the Civil Code and the Public Health Code govern the use of human tissues and cells. The principle of "non-patrimonialité" (non-commercialization) of the human body and its parts is enshrined in French law, meaning that human tissues cannot be subject to property rights or commercial transactions. However, once tissues are manipulated for research purposes, the intellectual property and resultant innovations may belong to the researchers or institutions involved.

Do Organoids should be viewed as Body Parts?

The classification as "body parts" introduces another layer of complexity. In many jurisdictions, certain body parts, such as blood and gametes, can be donated and even sold under specific conditions. For instance, the sale of blood and plasma is legal in the USA under regulated circumstances, and gametes can also be sold. If organoids are classified similarly, this could open the door to their commercialization, which may influence public perception and acceptance.
In the European Union, the donation of tissues and cells, including for research, is strictly regulated to ensure ethical standards and the protection of donor rights. The EU Tissues and Cells Directive emphasizes voluntary and unpaid donation, although compensation for expenses and inconveniences incurred is allowed. The potential classification of organoids as body parts could challenge these existing frameworks and necessitate new regulations to address commercialization concerns.

The EU Tissues and Cells Directive

The EU Tissues and Cells Directive (2004/23/EC) sets out standards for the quality and safety of human tissues and cells intended for human application. It aims to ensure that the procurement, testing, processing, preservation, storage, and distribution of tissues and cells are carried out ethically and safely. Key principles include:
1. Voluntary and Unpaid Donation: The Directive emphasizes that donations should be voluntary and unpaid, although compensation for expenses and inconveniences incurred is allowed.

2. Informed Consent: Donors must provide informed consent, understanding the purposes and potential uses of their donated tissues or cells.

3. Traceability and Quality Assurance: Comprehensive records must be maintained to ensure traceability from donor to recipient, and high standards of quality and safety must be upheld.

Implications for Organoids

Given that the creation of organoids involves extensive manipulation and development by researchers, they may be viewed as new entities separate from the original cells donated by the patient.
This perspective aligns with existing legal principles where the transformation process confers ownership to the entity that has invested resources into the development. However, this stance

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

does not entirely resolve the ethical and legal challenges associated with the use of organoids, particularly regarding their classification and the potential for commercialization.

Aligning Organoid IP with EU Ethical and Legal Framework

To harmonize organoid IP with the EU Tissues and Cells Directive, several key aspects need consideration:

1. Informed Consent and Transparency: Ensuring that donors are fully informed about the potential IP implications of their donation is crucial. This includes clear communication about how their cells might be used, the possibility of commercial applications, and the potential for IP generation. This is why we included such an information and an anticipation in the TRUSTED list.

2. Non-commercialization Principle: The EU's stance on the non-commercialization of the human body and its parts implies that while the original biological material cannot be commercialized, the transformed and significantly manipulated organoids can be, provided that ethical standards are met. This distinction must be carefully managed to maintain public trust and adherence to legal norms.

3. Ethical Review and Compliance: Research involving organoids must undergo rigorous ethical review to ensure compliance with EU regulations. This includes assessing the ethical implications of IP claims and ensuring that the rights and dignity of donors are respected throughout the research and commercialization process.

4. Data Protection and Privacy: The General Data Protection Regulation (GDPR) in the EU also impacts organoid research, particularly regarding the handling of personal data derived from donors. Ensuring robust data protection measures aligns with the ethical use of donated tissues and cells.

Legislative Clarity and Regulatory Frameworks

The clinical use of organoids necessitates legislative clarity, particularly concerning the sale and ownership of body parts. Current regulations around organ and tissue donation are not directly applicable to organoids, which are more complex and have broader applications in personalized medicine. Legislative bodies need to address whether organoids can be commercialized and under what conditions.

Moreover, the classification of organoids as "procedures" rather than "medicinal products" could have significant regulatory implications. If clinics present organoid-based treatments as procedures, they might bypass some of the stringent regulatory requirements applied to medicinal products. This approach could expedite clinical use but might also raise concerns about safety, efficacy, and ethical oversight.

Regulatory bodies such as the FDA in the USA, the European Medicines Agency (EMA) in Europe, and similar entities worldwide will need to develop specific guidelines for organoids. These guidelines should address the ethical sourcing of cells, consent processes, ownership rights, and the conditions under which organoids can be commercialized.

The EU may need to update its existing directives or introduce new regulations specifically addressing the unique challenges posed by organoids. This could include guidelines on the ethical commercialization of organoids, informed consent processes, and the handling of IP rights.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

Engaging with stakeholders, including researchers, ethicists, legal experts, and patient advocacy groups, can help shape balanced and effective regulatory frameworks. Public consultations and interdisciplinary dialogues are crucial for developing policies that reflect diverse perspectives and ethical considerations. Such tasks might be entrusted to the PACOR proposed above.

Given the global nature of scientific research and innovation, international collaboration on regulatory standards for organoids can help harmonize practices and ensure that ethical and legal norms are consistently applied across jurisdictions.

Conclusion

The articulation between organoid IP and the EU Tissues and Cells Directive involves navigating a complex landscape of ethical, legal, and scientific considerations. Ensuring that IP rights are managed in a way that respects donor rights, adheres to ethical standards, and promotes innovation requires clear regulatory guidelines and ongoing dialogue among stakeholders. By aligning the transformative potential of organoid research with the principles of the EU's regulatory framework, it is possible to foster responsible and ethically sound advancements in regenerative medicine.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

8 CONCLUDING REMARKS AND RECOMMENDATIONS

The Operational Guidelines for organoids and the Code of responsible conduct are developed by referencing existing frameworks such as the ALLEA European Code of Conduct, the International society for stem cell research (ISSCR) guidelines, European ethical self-assessment questionnaires, and donor consent documentation.

HYBRIDA's Guidelines for the design, production, and use of organoids are organized around four key deliverables crucial to civil society. This approach prioritizes the concerns of individuals, emphasizing ethical self-assessment (Ricocheck), informed consent with a detailed list of donor options (TRUSTED), ensuring responsible and ethically informed organoid research and application.

The recommendations for organoid research cover several critical areas including the Ethics by Design (ED) approach, the Reflexivity, Anticipation, Deliberation (RAD) process, Responsible Research and Innovation (RRI) practice, allowing the integration of ethical considerations throughout the research and development process. These approaches are aimed at ensuring that organoid research is conducted with foresight, inclusivity, and a commitment to ethical integrity, anticipating and addressing potential ethical, social, and technical challenges from the outset.

FINAL RECOMMENDATIONS

1. **Anticipate and Address Ethical Concerns Proactively**: Stakeholders in the field of organoids are called to employ an ED/RAD framework to proactively anticipate ethical concerns, including the social implications of organoid research. This involves engaging in ethical reflection and assessment from the conceptual stage through to clinical applications, considering long-term societal impacts.
   1.1. Utilize the MIAOU/EChOES questionnaires for reporting organoid research to ensure comprehensiveness and standardization. Moreover, these questionnaires will be used to establish a representation of knowledge related to organoids and will allow to detect evolutions both in knowledge and in the use of organoids
   1.2. Put special emphasis on the confidentiality and privacy concerns related to genetic information derived from organoids, aligning with GDPR and other relevant data protection frameworks.
   1.3. Address potential misuse of organoids in research and therapy, such as creating organoids with sentient potential or for purposes not aligned with ethical guidelines and societal values.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

2. **Incorporate Responsible Research and Innovation (RRI) Practices**: Adhere to RRI principles by involving stakeholders early in the research process, promoting interdisciplinarity, including social sciences and humanities, and ensuring that research is conducted for and with society.
   2.1. Use the RICOCheck questionnaire for ethical self-assessment, in line with the European ethical self-assessment document.
   2.2. Update RICOCheck to follow the evolution of the European ethical self-assessment and the evolution of the social impact of organoids.

3. **Ensure Continuous Ethical Engagement**: Maintain ongoing ethical dialogue among researchers, stakeholders, and society, from the initial stages of research to all conceivable applications. This continuous engagement allows for the adaptation of research practices in response to evolving ethical standards and societal expectations.
   3.1. Use HYBRIDA Operational Guidelines on the ethical procurement of human tissues and biological material when creating organoids.
   3.2. Anticipate future use and reuse in accordance to the donor's TRUSTED list, with potential agreement delegation to an independent third party representing the interest of the donors.

4. **Implement Ethical Reflection**: Combine RAD methodology with ED to allow for swift adaptations as ethical considerations and scientific knowledge evolve. This integration facilitates the ethical development of organoids by enabling rapid prototyping, stakeholder feedback, and iterative refinement.
   4.1. Initiate discussions to clarify the status of embryo models
   4.2. Initiate discussions on sentience and consciousness of complex neural organoids
   4.3. Initiate discussions on consent withdrawal
   4.4. Initiate discussions on chimeras involving organoids
   4.5. Monitor all emerging issues, through a task force involving all stakeholders.
   4.6. Prevent as much as possible inequitable access to benefits by recognizing and mitigating risks related to the commercialization of organoids, ensuring equitable access to advancements in organoid-based therapies and diagnostics

5. **Foster Public deliberation and transparent regulation over Organoid Development**: Advocate for transparent oversight and regulation over the development and application of organoid technologies. This involves transparent decision-making processes that include a broad range of stakeholders, ensuring that organoid research aligns with deontological practices and ethics.
   5.1. Establish a Public Advisory Committee for Organoid Research inside an existing agency that includes scientists, ethicists, patient advocates, policymakers, and laypersons to provide oversight and input into the direction of organoid research. Implement transparent research agendas and reporting of this committee.
   5.2. Promote open access to research agendas, funding sources, and findings in the field of organoid science. This includes the requirement for researchers to publicly disclose objectives, methodologies, results, and implications of their work, enabling public scrutiny and input.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

5.3. Promote Initiatives to enhance public understanding of organoid research, including its potential benefits and ethical considerations, fostering an informed dialogue between researchers and the public.

6. **Facilitate Ethical Literacy and Education in the field of organoids**: Develop educational resources and training programs to enhance ethical literacy among organoid researchers, stakeholders, and the broader public. This includes fostering an understanding of the ethical dimensions of organoid research and the importance of ethical design principles.
   6.1. Avoid misnaming such as using terms like "mini-organ" or "synthetic embryo" that might mislead about organoids' capabilities.
   6.2. Prevent exaggerated claims, hype as well as fears, regarding clinical applications to maintain public trust and realistic expectations.
   6.3. Develop an Open-Access Ethical Literacy Platform: Launch a digital platform offering a comprehensive suite of resources on the ethics of organoid research. This platform would host interactive modules, case studies, expert lectures, and guidelines designed to enhance ethical understanding and decision-making skills among researchers and the broader community.
   6.4. Integrate Ethics into STEM Curricula (Science, Technology, Engineering and mathematics): Work with educational institutions to integrate ethics modules specifically tailored to organoid research within existing STEM curricula. This initiative should aim at instilling a foundational ethical mindset from an early stage in scientific education, preparing future researchers to consider ethical implications inherently in their work.

7. **Commit to respecting the informed consent process**:
   7.1. Upstream of any project of organoid derivation, provide both verbal information during consultations with the donor and a comprehensible information letter detailing i) the research topic, ii) objectives, iii) the procedure for sample or tissue collection, handling of biological samples, iv) the fate of biological samples and associated data, in particular if reuse is planed, v) the procedure to protect patient anonymity, vi) the right to withdraw consent without prejudice, and vii) the right to be informed about reuse of their biological samples as well as on the follow-up and the results of the study.
   7.2. Implement and include in the informed consent the "TRUSTED" questionnaire, enabling donors to explicitly authorize or prohibit specific potential uses of their biological material and data.
   7.3. Formalize signed informed consent in a pseudonymized passport-style document - letter of information, informed consent form and TRUSTED list - accompanying biological samples distributed by biobanks.
   7.4. Involve, if applicable, an independent third party responsible for reviewing sample use and reuse, ensuring compliance with donor preferences.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

7.5. Entrust the Public Advisory Committee for Organoid Research, mentioned in recommendation 5.1, to assess various consent forms and define the most appropriate consent options, as well as the modalities and consequences of a possible withdrawal of consent for all parties.

These recommendations emphasize the need for a holistic and anticipatory approach to the ethical challenges of organoid research, integrating ethical considerations throughout the research process, and engaging with a broad range of stakeholders to ensure that organoid technologies develop in a way that is socially responsible, inclusive, and aligned with human values.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

9 GLOSSARY

Ethics Terminology Unwrapped

Accountability:

Anticipation of the positive and negative impacts of research or evaluation work, extending to all roles within a research or institutional context.

Researcher to Evaluator: Anticipate as much as possible the positive and negative impacts of work in a given context and at a given time.

Evaluator to researchers: anticipate the impact of the evaluation performed.

Evaluator towards the institution: anticipate the impacts of evaluation on the functioning of the institution (suffering at work for example)

From the institution to the evaluator: provide the means to carry out the evaluation according to the principles defined, avoid paradoxical injunctions (DORA versus bibliometric index).

From the institution to the researcher: working conditions must be provided that enable researchers to undertake honest, fair and responsible research, in a collaborative rather than a competitive framework.

Advanced therapy products:

Advanced Therapy Medicines (ATMs) are cell or tissue products modified to compensate for functions a patient has lost, to promote repair mechanisms (regenerative medicine) or to stimulate the immune system against viruses or tumours (immunotherapy).

Anonymised (data):

The data has been rendered anonymous in such a way that the subject of the data can no longer be identified (the data are therefore no longer personal and thus fall outside the scope of data protection law).

Applied Research/Engineering:
Research that seeks to solve practical problems and improve the human condition. It focuses on the application of theories, knowledge and methods.

Autonomy:

Evaluator to evaluator: in order to perform an evaluation, there must be consent from the researcher being evaluated and thus a procedure that is sufficiently clear for that person to understand and accept it.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

Benevolence:

Evaluator to evaluator: consistency in follow-up of the evaluation.

Biobanks:

Large collections of biological specimens linked to relevant personal and health information (health records, family history, lifestyle, genetic information) that are held predominantly for use in health and medical research.

Biological Samples:
Any material that is derived from a human, animal or microbial source, such as blood, tissue, cells, DNA, RNA or proteins, and which are used for laboratory experiments and analysis.

Chimera:
In biology, a chimera is defined as an organism or tissue that contains at least two different sets of DNA, usually originating from the fusion of different zygotes (fertilized eggs). The concept is named after the Chimera from Greek mythology, a creature that was part lion, part goat, and part dragon. This biological phenomenon is distinguished from mosaicism, where an organism has genetically distinct cell populations all originating from a single zygote, and from hybrids, which involve genetically identical cell populations originating from the cross of two different species.
In the field of organoid research, the concept of a chimera refers to a biological entity that includes cells from different sources, typically involving human cells introduced into another animal species. These chimeras are created through a process where donor cells are introduced into a non-human animal at an early embryonic stage, allowing for high levels of integration between the donor and host cells.

Clinical Research:
Clinical research corresponds to scientific studies carried out on human beings with a view to developing biological or medical knowledge. This is prospective research, involving the follow-up of patients or healthy volunteers. Such research is essential to better understand and treat diseases, and to identify potential risk factors.

Clinical trial:

A clinical trial is an experimental situation in which a therapeutic hypothesis is tested in humans. A clinical trial on a drug, for example, aims to assess the efficacy and safety of the new molecule.

Consent:

The provision of clear, accurate, and comprehensive information about a research study means that consent can be given voluntarily and can be withdrawn at any time without negative consequences.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

Citizen/patient to researcher: In the context of citizen/patient to researcher, the TRUSTED list - part of the consent form - refers to the specific set of instructions or guidelines provided by an individual or their legally authorised representative which detail how their donated biological samples should be used to create organoids for research. The researcher is expected to adhere to these wishes as closely as possible, ensuring that the individual's autonomy and personal values are respected throughout the research process. This emphasises the importance of transparent communication and ethical practices when handling human-derived samples in the context of scientific studies.

Citizen/patient to Evaluator: In the citizen/patient to evaluator setting, consent refers to the voluntary, informed and unambiguous agreement given by an individual or their legally authorised representative that allows the evaluator to access, review and assess their personal information, data, or any results derived from their participation in a specific research study. This consent is obtained after the individual has been provided with sufficient information about the evaluation process, its objectives, potential risks and benefits, and their rights to privacy and withdrawal. Consent helps to ensure that evaluations involving human subjects are conducted ethically and respect the autonomy of the individuals involved.

Citizen/patient to Institution: In the citizen/patient to institution context, consent refers to the voluntary, informed and unambiguous agreement given by an individual or their legally authorised representative that allows the institution to collect, use, store and share their personal information, data, or biological samples for specific research or healthcare purposes. This consent is obtained after the individual has been provided with sufficient information about the institution's practices, potential risks and benefits, and their rights to privacy, data protection and withdrawal. Consent helps to ensure that the institution's activities involving human subjects are conducted ethically and respect the autonomy and rights of the individuals involved.

Citizen/patient to citizen/patient: In the citizen/patient to citizen/patient context in terms of society, consent refers to the voluntary, informed and unambiguous agreement given by an individual or their legally authorised representative that allows another individual or group of individuals to access, use or share their personal information, experiences or opinions for a specific purpose. This consent is obtained after the individual has been provided with sufficient information about the potential risks and benefits, and their rights to privacy, confidentiality and withdrawal. Consent fosters a respectful and cooperative environment within society, ensuring that personal boundaries and autonomy are honoured when exchanging information or sharing experiences among fellow citizens or patients.

European Code of Conduct:

The European Code of Conduct for Research Integrity serves the European research community as a framework for self-regulation across all scientific and scholarly disciplines and for all research settings.

Factoroid:

A specialised type of organoid engineered to produce a specific biological product in large quantities. Factoroids represent a valuable tool for biological and medical research, enabling the large-scale production of biological substances for various applications such as drug testing and disease modelling.

Fundamental Research:

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

Research carried out to improve our understanding of fundamental principles. It is not necessarily directed towards any specific practical aim or application.

Genome, DNA, RNA, Nucleic Acid:
These terms relate to genetic material:
- Genome: The complete set of genes or genetic material present in a cell or organism.
- DNA: Deoxyribonucleic Acid, the molecule that carries genetic instructions in living organisms.
- RNA: Ribonucleic Acid, a molecule involved in the coding, decoding, regulation and expression of genes.
- Nucleic Acid: A complex organic substance present in living cells, especially DNA or RNA, whose molecules consist of numerous nucleotides linked in a long chain.

Honesty:
Accurate and complete presentation of project details, acknowledging potential biases, conflicts of interest and uncertainties.
Researcher to researcher: honesty refers to the commitment of researchers to uphold ethical standards and principles when communicating and collaborating with their peers. This includes presenting accurate and unbiased data, acknowledging the contributions of others, sharing information transparently, and avoiding any fabrication, falsification or plagiarism. Honesty fosters a culture of trust, integrity, and accountability within the research community, promoting the advancement of knowledge and the development of reliable, high-quality research outcomes.
Researcher to evaluator: In the researcher to evaluator context, honesty refers to the commitment of researchers to present accurate, unbiased and complete information about their work, methodologies, data, and results during the evaluation process. This includes being transparent about any limitations, uncertainties or potential biases in their research, as well as acknowledging the contributions of others. Honesty fosters trust, integrity, and accountability between researchers and evaluators, ensuring that evaluations are fair, accurate and contribute to the improvement of research quality and the advancement of knowledge.

Researcher to Institution: In the researcher to institution context, honesty refers to the commitment of researchers to uphold ethical standards and principles when engaging with their affiliated institutions. This includes transparently reporting research progress, outcomes and any issues that arise, as well as acknowledging the contributions of others and complying with institutional policies and guidelines. Honesty fosters a culture of trust, integrity, and accountability within the institution, promoting a supportive environment for high-quality research and the responsible conduct of research activities.
Researcher to Citizen: In the researcher to citizen context, honesty refers to the commitment of researchers to communicate their research findings, methodologies and their implications in an accurate, transparent and accessible manner to the general public. This includes presenting information without bias or distortion, acknowledging uncertainties, and being open about the limitations of their work. Honesty fosters trust, credibility and accountability between researchers and the wider community, promoting public understanding of science, informed decision-making and constructive dialogue on research-related issues.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

Evaluator to researcher (**equity/fairness**): In the evaluator to researcher context, honesty in terms of equity and fairness refers to the commitment of evaluators to assess research projects, methodologies, data and outcomes impartially, without bias or favouritism. This includes applying consistent evaluation criteria and principles across all researchers, acknowledging the diverse backgrounds and perspectives of researchers, and providing constructive feedback to help researchers improve their work. By upholding honesty, equity and fairness, evaluators contribute to a supportive and inclusive research environment that fosters trust, accountability and the advancement of knowledge.
Institution to researcher: In the institution to researcher context, honesty refers to the commitment of institutions to provide accurate and transparent information, support and the resources necessary for researchers to carry out their work effectively and ethically. This includes setting clear expectations, guidelines and policies, as well as acknowledging researchers' achievements, addressing their concerns, and fostering an environment that promotes open communication and collaboration. Honesty helps to build trust, integrity and accountability between institutions and researchers, ensuring a supportive atmosphere conducive to high-quality research and responsible conduct.

Citizen/patient to Citizen/patient: In the citizen/patient to citizen/patient context, honesty refers to the commitment of individuals to communicate their experiences, opinions and information with one another in an accurate, transparent and unbiased manner. This includes sharing personal experiences or knowledge about a particular topic, medical condition or treatment without distortion or exaggeration, and acknowledging uncertainties or limitations in their understanding. Honesty fosters trust, credibility and accountability between citizens or patients, promoting informed decision-making, mutual support and constructive dialogue within the community.

iPSC (induced Pluripotent Stem Cells):
Cells that are reprogrammed from differentiated adult cells to have the characteristics of embryonic stem cells, i.e. self-renewal and pluripotency (the ability to differentiate into all cell types of the body).

Justice:

Evaluator to evaluator: the principles of justice are explicit and identical for all committees. Serving on a committee means accepting these principles.

Minimisation Measures:
A method or approach implemented to reduce the negative impact or extent of a particular activity or process. In a research context, this could involve measures to minimise the use of animal models or reduce the amount of potentially hazardous materials used in an experiment.

Non-malevolence:

Evaluator to evaluator: encourage supportive evaluation rather than punitive evaluation.

Norm:

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

A norm is a proposition that expresses what must or must not be done. *E.g. you shall not kill!*

Openness:

Institution to institution: a commitment to open science (open access, open data -FAIR- open methodologies and protocols) the promotion of interdisciplinarity, multidisciplinarity and cross-disciplinarity, and the promotion of collective efforts.

Opt-in:
A system or process where individuals must explicitly consent or agree to participate or receive certain services or communications. This is often used in the context of data privacy or research consent.

Opt-out:
A system or process where individuals are automatically included or subscribed but have the option to decline or withdraw. This is often used in the context of data privacy or research consent.

Organoid:

A three-dimensional structure grown from stem cells that mimics an organ and can be used in biological and medical research.

Personalised medicine:
A medical practice that uses information about a person's own genes or proteins to prevent, diagnose, or treat disease.

Precision medicine:
Precision medicine looks at the genetics, environment and lifestyle of a person in order to select the treatment that could work best for them.

Physical, Moral and Social Well-being*:*

The overall health, ethical treatment and social acceptance of citizens or patients in the context of their interactions with researchers.

Researcher to society. In the researcher to society context, Physical, Moral, and Social Well-being refers to the researcher's responsibility to conduct research that contributes positively to overall health, ethical standards and social cohesion within the community or society as a whole. This involves ensuring that the research process and outcomes align with ethical principles, minimise harm, promote health and welfare, respect cultural diversity and foster social equity. By prioritising physical, moral, and social well-being, researchers contribute to the development of a more sustainable, harmonious and inclusive society.

Principle:

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

A principle is a standard that expresses an important moral consideration and serves as a general guide. *E.g. The principle of beneficence in medicine*

Pseudonymised (data and samples):

means to separate the data and samples from their direct identifiers using a code so that linking them to a person is only possible by means of additional information that is held separately. This additional information (code) must be stored separately and securely from the processed data in order to ensure non-attribution.

Rationality of a research work:

Ability to decipher the research strategy, particularly through the proper use of citations and a description of the current state of research and related questions.

Reliability:

Robustness and reproducibility of research work and the assurance of objective, transparent evaluations based on sound methodologies.

Researcher to researcher: In the researcher to researcher context, reliability refers to the consistency, dependability and reproducibility of research findings, methodologies, and data when shared or communicated between researchers. This involves ensuring that research processes and outcomes are well-documented, transparent, and adhere to established standards and practices, making it possible for other researchers to replicate or build upon the work. Reliability fosters trust, collaboration, and scientific rigour within the research community, promoting the advancement of knowledge and the development of high-quality research outcomes.

Researcher to evaluator: In the researcher to evaluator context, reliability refers to the consistency, dependability and accuracy of the research findings, methodologies and data presented by the researcher during the evaluation process. This involves providing well-documented, transparent and verifiable information, as well as adhering to established standards and practices, which enables the evaluator to thoroughly assess the quality and rigour of the research. Reliability fosters trust, accountability and scientific integrity between researchers and evaluators, ensuring that evaluations contribute to the improvement of research quality and the advancement of knowledge.
Researcher to institution: In the researcher to institution context, reliability refers to the dependability, consistency and trustworthiness of the research findings, methodologies and data presented by the researcher to their affiliated institution. This involves adhering to established standards, guidelines and best practices, as well as providing accurate, well-documented and transparent information about research progress, outcomes and any issues that might arise. Reliability fosters trust, accountability and scientific integrity between researchers and institutions, ensuring a supportive environment for high-quality research and the responsible conduct of research activities.

Replicability in research:

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

Ability to reconstruct a set of data from the description of the materials and methods and the research strategy and then from this new set of data to reproduce the results and conclusions if there are no hidden or unaccounted-for variables (e.g. particularities of the working environment, specificity of the animal house, etc.)

Reproducibility in research:

Ability to reproduce figures and a discussion of results and conclusions based on access to raw data and a description of the materials and methods used.

Respect:

Acceptance of protocols, decisions and counter-arguments and the acknowledgment of contributions, achievements and feedback.

Researcher to evaluator: In the researcher to evaluator context, respect refers to the mutual recognition and appreciation of each other's roles, expertise and contributions to the research process. This involves researchers being open to constructive feedback and acknowledging the evaluators' efforts to assess their work, while evaluators approach the research with an open mind, considering the researchers' perspectives and recognizing the limitations and challenges they may face. Respect fosters a positive and collaborative environment between researchers and evaluators, promoting trust, communication and the continuous improvement of research quality and rigour.
Researcher to institution: In the researcher to institution context, respect refers to a mutual recognition and appreciation of each other's roles, expertise and contributions within the research environment. This involves researchers acknowledging the support and resources provided by the institution, adhering to its policies and guidelines, and valuing its mission and goals. Conversely, the institution recognizes the researchers' expertise, work, and the challenges they may face, and supports their professional growth and development. Respect fosters a positive, collaborative and inclusive environment between researchers and institutions, promoting trust, communication, and the pursuit of high-quality research and responsible conduct.

Institution to researcher: Respect in this context involves the institution providing the researcher with an inclusive, diverse and safe environment, encouraging open communication, and recognizing their contributions and achievements. It also requires the institution to protect the researcher's rights, address their concerns, and ensure that they are treated fairly and without discrimination throughout the research process. By demonstrating respect in their interactions with the researcher, the institution cultivates a positive and productive research environment, which contributes to overall success of the scientific project, the personal and professional development of the researcher, and the reputation and integrity of the institution.
Evaluator to researcher: In the evaluator to researcher context, respect refers to a mutual recognition and appreciation of each other's roles, expertise and contributions to the research and evaluation process. This involves evaluators approaching the assessment of research with an open mind, considering the researchers' perspectives, acknowledging the limitations and challenges they may face, and providing constructive feedback. In turn, researchers value the evaluators' expertise and efforts to assess their work, being open to feedback and improvement.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

Respect fosters a positive and collaborative environment between evaluators and researchers, promoting trust, communication, and the continuous enhancement of research quality and rigour.

Evaluator to institution: In the evaluator to institution context, respect refers to the mutual recognition and appreciation of each other's roles, expertise and contributions within the research evaluation process. This involves evaluators acknowledging the institution's mission, goals, and the support it provides to researchers, while approaching the assessment process objectively and fairly. On the other hand, the institution values the expertise and insights of the evaluators and considers their recommendations will improve research quality, policies, and practices. Respect fosters a positive, collaborative and inclusive environment between evaluators and institutions, promoting trust, communication and the pursuit of high-quality research and responsible conduct.
Institution to evaluator: In the institution to evaluator context, respect refers to the mutual recognition and appreciation of each other's roles, expertise and contributions within the research evaluation process. This involves institutions acknowledging the evaluators' expertise, insights and efforts in assessing research quality, being open to their recommendations, and providing necessary support and information for a fair evaluation. Conversely, evaluators approach the assessment process objectively, taking into account the institution's mission, goals, and the support it provides to researchers. Respect fosters a positive, collaborative and inclusive environment between institutions and evaluators, promoting trust, communication and the continuous improvement of research quality and responsible conduct.

Responsibility:

Diligent and impartial conduct of evaluation that considers the potential impacts and implications of the evaluation.

Evaluator to researcher: Responsibility in this context involves the evaluator conducting the assessment with diligence, impartiality and competence, and thoroughly reviewing the project's objectives, methods, results and limitations. It also requires the evaluator to consider the potential impacts and implications of the evaluation on the scientist's work, the institution, and the broader scientific community, taking care not to cause harm or unfairly disadvantage any party involved.
By demonstrating responsibility in their interactions with the scientist, the evaluator contributes to the credibility and trustworthiness of the evaluation process, ensuring that the assessment of the scientific project is rigorous, objective and useful in informing future research, decision-making and policy.
*Institution to researcher: R*esponsibility in this context involves the institution ensuring an environment conducive to research, establishing clear guidelines and expectations for scientific integrity, and implementing fair, transparent and accountable review, approval, and assessment processes. It also requires the institution to address any potential conflicts of interest, misconduct, or ethical concerns promptly and appropriately, taking care not to cause harm or unfairly disadvantage the scientist or other parties involved. By demonstrating responsibility in their interactions with the scientist, the institution contributes to the credibility and trustworthiness of the research process, ensuring that realization of the scientific project is

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

rigorous, ethical and useful in advancing knowledge, informing decision-making, and fostering a culture of integrity and excellence.

Evaluator to researcher: Responsibility in this context involves the scientist presenting the project's objectives, methods, results, limitations and potential implications with honesty and clarity, acknowledging any uncertainties, potential risks and ethical concerns. It also requires the scientist to be open to constructive feedback, criticism and recommendations from the evaluator, using the insights gained to improve the quality, credibility and impact of their research.

By demonstrating responsibility in their interactions with the evaluator, the scientist contributes to the trustworthiness and integrity of the evaluation process, ensuring that the assessment of the scientific project is informative, useful and ultimately beneficial to the advancement of knowledge and the broader scientific community.

Evaluator to institution: Responsibility in this context involves the institution providing the evaluator with accurate, complete and transparent information about the project, including its objectives, methods, results, limitations and potential implications. It also requires the institution to address any potential conflicts of interest, biases or ethical concerns that may arise during the evaluation process and to ensure that the evaluation is conducted fairly and with respect for the parties involved.

By demonstrating responsibility in their interactions with the evaluator, the institution contributes to the trustworthiness and integrity of the evaluation process, ensuring that the assessment of the scientific project is informative, useful and ultimately beneficial to the advancement of knowledge, the institution's reputation and the broader scientific community.

Institution to researcher: Responsibility in this context involves the scientist designing and executing the research with rigor, integrity and transparency, ensuring that the project complies with ethical guidelines, legal requirements and the institution's research policies. It also requires the scientist to communicate effectively with the institution, reporting any challenges, uncertainties, potential risks and ethical concerns that may arise during the project, and seeking guidance and support when needed.

By demonstrating responsibility in their interactions with the institution, the scientist contributes to the credibility and trustworthiness of the research process, ensuring that realization of the scientific project is ethical, rigorous and in line with the institution's values and standards, ultimately benefiting the advancement of knowledge and the broader scientific community.

Institution to evaluator: Responsibility in this context involves the evaluator thoroughly reviewing the project's objectives, methods, results and limitations, using appropriate evaluation criteria and techniques and considering the potential impacts and implications of the evaluation on the project, the institution and the broader scientific community. It also requires the evaluator to maintain open and effective communication with the institution, reporting any challenges, uncertainties, potential biases or ethical concerns that may arise during the evaluation, and seeking clarification and additional information when needed.

By demonstrating responsibility in their interactions with the institution, the evaluator contributes to the trustworthiness and integrity of the evaluation process, ensuring that the assessment of the scientific project is rigorous, objective and informative, ultimately benefiting the advancement of knowledge, the institution's reputation, and the broader scientific community.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

Transparency:
Open sharing of, and access to, raw data, methodologies, evaluation procedures, and rules of governance.
Researcher to researcher: In the researcher to researcher context, transparency refers to the open, honest and clear communication of research methods, data, findings and limitations among peers. This involves sharing information and resources, such as data sets, experimental protocols and analytical techniques, so that others can replicate, build upon, or scrutinize the work. Transparency fosters a collaborative and supportive research environment, promoting trust, accountability and scientific rigor, which ultimately contribute to the advancement of knowledge and the development of high-quality research outcomes.

Researcher to evaluator: In the researcher to evaluator context, transparency refers to the open, honest and clear communication of research methods, data, findings and limitations during the evaluation process. This involves providing the evaluator with accurate, well-documented and accessible information about the research, including any challenges or uncertainties, to enable a thorough and fair assessment. Transparency fosters trust, accountability and scientific integrity between researchers and evaluators, ensuring that evaluations contribute to the improvement of research quality and the advancement of knowledge.
Researcher to institution: In the researcher to institution context, transparency refers to the open, honest, and clear communication of research methods, data, findings and limitations between the researcher and their affiliated institution. This involves providing accurate, well-documented and accessible information about research progress, outcomes, challenges and any issues that may arise, as well as adhering to institutional policies and guidelines. Transparency fosters trust, accountability and scientific integrity between researchers and institutions, ensuring a supportive environment for high-quality research and the responsible conduct of research activities.
Evaluator to researcher: In the evaluator to researcher context, transparency refers to the open, honest and clear communication of evaluation criteria, methods, feedback and recommendations during the research assessment process. This involves providing researchers with a clear understanding of the standards and expectations being applied, as well as sharing constructive and evidence-based feedback and justifications for the evaluation outcomes. Transparency fosters trust, accountability and collaboration between evaluators and researchers, ensuring that evaluations contribute to the improvement of research quality, responsible conduct, and the advancement of knowledge.

Evaluator to institution: In the evaluator to institution context, transparency refers to the open, honest and clear communication of evaluation criteria, methods, results and recommendations during the research assessment process. This involves providing the institution with a clear understanding of the standards and expectations being applied, sharing evidence-based feedback and justifications for the evaluation outcomes, and disclosing any potential conflicts of interest. Transparency fosters trust, accountability and collaboration between evaluators and institutions, ensuring that evaluations contribute to the continuous improvement of research quality, policies and practices within the institution.

Institution to evaluator: In the institution to evaluator context, transparency refers to the open, honest and clear communication of institutional policies, guidelines, research activities and any relevant information that may impact the research evaluation process. This involves providing the evaluator with necessary access to data, resources and documentation, sharing information

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

about the institution's mission, goals, and expectations, and disclosing any potential conflicts of interest. Transparency fosters trust, accountability and collaboration between institutions and evaluators, ensuring that evaluations contribute to the continuous improvement of research quality, policies and practices within the institution.

Institution to researcher: In the institution to researcher context, transparency refers to the open, honest and clear communication of institutional policies, guidelines, expectations and any relevant information that may impact the research process. This involves providing researchers with necessary access to data, resources and documentation, sharing information about the institution's mission, goals, and support mechanisms, and disclosing any potential conflicts of interest. Transparency fosters trust, accountability and collaboration between institutions and researchers, ensuring a supportive environment for high-quality research and the responsible conduct of research activities.

Values:

Universal value: A value is a universal value if it has the same value or worth for all, or almost all, people. Spheres of human value encompass morality, aesthetic preference, traits, human endeavour and social order.
Contextual value: as opposed to universal values, contextual values actually depend on the context in which they are looked at and modified.

Standards and values

Norms and values are different moral categories (Max Scheler):

A standard expresses:	**A value is expressed:**
an assignment as a task	an ideal duty to be
the property is imperative (mandatory)	the good is attractive (= object of desire)
e.g.: Human beings must be treated fairly	e.g.: Women should give birth without pain

They determine statements of different types:

Standard	**Value**
deontic or normative statement	axiological statement
e.g.: It is obligatory not to lie	e.g.: It is good not to lie
e.g.: You must not lie!	e.g.: Truthfulness is a good

Standards and values have different characteristics:

Standard	**Value**

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

mandatory, prohibited, must	good, bad, desirable
thin concept (= without descriptive content)	thick concept (with descriptive content: cruel, honest, etc.)
not related to emotions	linked to emotions (admirable, frightening, etc.)
categorical (must / must not)	gradual (more or less admirable)

The basis of morality: norms or values?

Two opposing positions (Kant versus Aristotle):

Standards are primary and foundational to moral values	Values are primary and some are the basis for standards
e.g.: Life has a moral value because it is forbidden to kill	e.g.: It is forbidden to kill because life has great value
In morality, obligation is first	In morality, the desirable is first
Samuel de Puffendorf: "Natural good becomes morally significant when it is enjoined by law and brought about voluntarily because of law."	

Realism and anti-realism of values

"Is that which is pious approved by the gods as being pious, or is it pious because the gods approve it?" (Plato, *Euthyphro*)

Two opposing positions:

Realism (Aristotle)	Anti-realism (David Hume)
The value of something is the value *it has* according to its properties	The value of something is what *we attribute to it* according to its properties
Aristotle: "We desire a thing because it appears to us to be good, rather than because it appears to us to be good because we desire it."	Hume: "It is a common observation, that the mind has a great propensity to spread itself over external objects, and to conjoin with them the internal impressions which they occasion."

The case of dignity

Dignity is presented as a value. It is therefore gradual: a particular behaviour can be more or less dignified. However, it is said that all human beings possess the same dignity, that it does not vary and must be respected, whatever their behaviour. There are two solutions to this problem:

1. Dignity is based on belonging to humanity, which is the same for all human beings, and therefore the dignity of each person is invariable.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

2. Dignity is not a value, but a norm. Affirming the equal dignity of every human being means that we have a duty to behave with the same respect towards each of them.

Learn more about this:

Ruwen Ogien and Christine Tappolet, *Les concepts de l''éthique*, Paris, Hermann, 2008.

Bernard Baertschi, "La place du normatif en morale", *Philosophiques*, vol. 28, 2001, p. 69-86.

Bernard Baertschi, "Valeurs et vertus", in J.-D. Causse & D. Müller, *Introduction à l''éthique*, Geneva, Labor & Fides, 2009, p. 177-197.

Bernard Baertschi, "Dignity," *Encyclopedia of Philosophy*, https://encyclo-philo.fr/item/109, 2017.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

10 REFERENCES

Agence de Biomédicine [Biomedicine Agency], *Consentement Au Don d'Embryons Pour La Recherche* [*Consent to Donation of Embryos for Research*]. Available at: https://www.agence-biomedecine.fr/Consentement-au-don-d-embryons-pour-la-recherche-650?lang=fr.

All European Academies, Available at: https://allea.org/techethos-future-technology-ethics/?cn-reloaded=1.

Allen MJ, Powers MLE, Gronowski KS, Gronowski AM. "Human tissue ownership and use in research: what Laboratorians and researchers should know". *Clin Chem*. Accessible at: 2010;56(11):1675–82. 61.

Amander, T Clark, Ali Brivanlou, Jianping Fu, Kazuto Kato, Debra Mathews, Kathy K Niakan, Nicolas Rivron, Mitinori Saitou, Azim Surani, Fuchou Tang, Janet Rossant, "Human embryo research, stem cell-derived embryo models and in vitro gametogenesis: Considerations leading to the revised ISSCR guidelines", *Stem Cell Reports*, 2021. Accessible at: https://www.ncbi.nlm.nih.gov/pmc/articles/PMC8190666/.

Baertschi, Bernard, Henri Atlan, Mylène Botbol-Baum, Bertrand Bed'hom, Hélène Combrisson, et al., "Organoids Research: What are the ethical issues?" *Memo*., 2020. Available at: inserm-03117706.

Baertschi Bernard, Marc Brodin, Christine Dosquet, Pierre Jouannet, Anne-Sophie Lapointe, Jennifer Merchant, Grégoire Moutel, *"Research on Embryos and Embryonic Models for Scientific Use (EMSUs*)", INSERM Notes, Accessible at: https://www.hal.inserm.fr/inserm-02373609/document.

Baker, L., Cristea, I., Errington, T., et al, *Reproducibility of scientific results in the EU: scoping report*, European Commission, Directorate-General for Research and Innovation, Luxembourg: Publications Office of the European Union, 2020.

Ball, P. "Synthetic biology: starting from scratch". *Nature* 431, 2004, Nature 431(7009):624-6, Accessible at: 10.1038/431624a.

Beauchamp, Tom, James Childress, *Les principes de l'éthique biomédicale*, Médecine & Sciences humaines, Les Belles Lettres, Paris, 2008.

Boers SN, van Delden JJM, Bredenoord AL, *Organoids as hybrids: ethical implications for the exchange of human tissues*, 2019, Journal of Medical Ethics.

Brey, Philip, Brandt Dainow, Yasemin J. Erden, Amal Matar, Philip Jansen, Rowena Rodrigues, Nicole Santiago, et al. 2021. *SIENNA D6.3: Methods for Translating Ethical Analysis into Instruments for the Ethical Development and Deployment of Emerging Technologies*. Available at: https://doi.org/10.5281/zenodo.5541539.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

Chen & al., "Transplantation of Human Brain Organoids: Revisiting the Science and Ethics of Brain Chimeras", *Cell Stem Cell*, 2019, vol. 25.

Cheshire W., "Miniature Human Brains: An Ethical Analysis", *Ethics and Medicine*, 2014, vol. 31/1.

Chirba M, Noble A. *Our bodies, our cells: FDA regulation of autologous adult stem cell therapies*, Bill of Health Boston College Law School Faculty Papers; 2013.

CIOMS, *International Ethical Guidelines for Biomedical Research Involving Human Subjects*, 2016, Prepared by the Council for International Organizations of Medical Sciences (CIOMS) in collaboration with the World Health Organization (WHO).

Diderot D. & J. d'Alembert, *Encyclopédie*, t. XV, 1765.

Emedicinehealth. *Informed Consent Form*. Available at: https://www.emedicinehealth.com/informed_consent/article_em.htm

Ethical Place. Île de France Region. [Espace éthique. Région Île-de-France]. Available at: https://www.espace-ethique.org/ressources/article/de-lethique-vers-la-pratique.

Ethical Review in FP7. Guidance for applicants. Informed consent. European Commission - Research Directorate-General Directorate L - Science, Economy and Society Unit L3 - Governance and Ethics. Accessible at https://ec.europa.eu/research/participants/data/ref/fp7/89807/informed-consent_en.pdf.

Ethics and Research Integrity Sector, DG R&I, European Commission, *Ethics By Design and Ethics of Use Approaches for Artificial Intelligence*. 25 November 2021. Available at: https://ec.europa.eu/info/funding-tenders/opportunities/docs/2021-2027/horizon/guidance/ethics-by-design-and-ethics-of-use-approaches-for-artificial-intelligence_he_en.pdf.

Éthique téléologique | philosophie, 10 October 2020, Available at: https://delphipages.live/fr/divers/teleological-ethics.

European Medicines Agency. Available at: https://www.ema.europa.eu/en/human-regulatory/research-development/scientific-guidelines/quality-guidelines.

Fiduciary Duty of Researchers - the Spleen Case - Moore v. Regents of University of California, 793 P.2d 479 (Cal. 1990) [Internet]. 1998. Available from: https:// biotech.law.lsu. edu/ cases/ consent/ Moore_v_ Regents.

Gold E., *Body parts: property rights and the ownership of human biological materials.* Washington, D.C.: Georgetown University Press; 1997.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

Horizon 2020 Programme, *Guidance: How to complete your ethics self-assessment*, 2019. Accessible at: https://ec.europa.eu/research/participants/data/ref/h2020/grants_manual/hi/ethics/h2020_hi_ethics-self-assess_en.pdf.

HYBRIDA Consortium (2020). Project Description. European Commission.

Kagan B. & al., "In vitro neurons learn and exhibit sentience when embodied in a simulated game-world", *Neuron*, 2022, vol. 110.

Kirstin R W Matthews, Daniel S Wagner, Aryeh Warmflash, "Stem cell-based models of embryos: The need for improved naming conventions", *Stem Cell Reports*, 2021. Accessible at: https://pubmed.ncbi.nlm.nih.gov/33770498/.

Kwon D., "Organoids Don't Accurately Model Human Brain Development", *The Scientist*, October 23, 2019, https://www.the-scientist.com/news-opinion/organoids-dont-accurately-model-human- brain-development-66629.

Lavazza A. & M. Massimini, "Cerebral organoids and consciousness: how far are we willing to go?", *Journal of Medical Ethics*, 2018.

----------, "Cerebral organoids: ethical issues and consciousness assessment", *Journal of Medical Ethics*, 2018.

Lee McFarling U., "Near the Campus Cow Pasture, a Scientist Works to Grow Human Organs" – in *Pigs, Stat*, October 20, 2017, https://www.statnews.com/2017/10/20/human-pig.

McCarthy John, *Ascribing Mental Qualities to Machines, University Computer Science Department Stanford*, 1979. Accessible at: http://jmc.stanford.edu/articles/ascribing/ascribing.pdf.

Moreau De Maupertuis Pierre Louis, Vénus physique, 1745, Paris, Aubier, 1980.

Munsie & al., Ethical Issues in Human Organoids and Gastruloid Research, The Company of Biologists, 2017, vol. 144.

Pașca, S.P., Arlotta, P., Bateup, H.S. *et al.* "A nomenclature consensus for nervous system organoids and assembloids". *Nature* 609,2022. Accessible at: https://doi.org/10.1038/s41586-022-05219-6.

Posfai Eszter, John Paul Schell, Adrian Janiszewski, Isidora Rovic, Alexander Murray, Brian Bradshaw, Tatsuya Yamakawa, Tine Pardon, Mouna El Bakkali, Irene Talon, Natalie De Geest, Pankaj Kumar, San Kit To, Sophie Petropoulos, Andrea Jurisicova, Vincent Pasque, Fredrik Lanner, Janet Rossant, "Evaluating totipotency using criteria of increasing stringency", *Nature Cell Biology*, 2021. Accessible at: https://www.nature.com/articles/s41556-020-00609-2.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

Rossi G. & al., "Progress and Potential in Organoid Research", Nature Reviews, 2018, vol. 19.

Recital 33, 2016, EU General Data Protection Regulation.

Ricœur, Paul, *Soi-même comme un autre* **[***Self as another***]**, Editions Seuils, Paris, 1990.

UNESCO, *International Declaration on Human Genetic Data*, 2003. Available at: https://unesdoc.unesco.org/ark:/48223/pf0000136112.

UNESCO, *Report of the IBC on Big Data and Health*, International Bioethics Committee, 2017.

Wall J. *Being and owning: the body, bodily material, and the law*. Oxford: Oxford University Press; 2015.

Warren Mary Anne, *Moral Status*, Oxford, OUP, 1997.

WMA Declaration of Helsinki, Ethical Principles for Medical Research Involving Human Subjects. General Principles. Accessible at: https://www.wma.net/policies-post/wma-declaration-of-helsinki-ethical-principles-for-medical-research-involving-human-subjects/.

World Health Organization. WHO Expert Advisory Committee on Developing Global Standards for Governance and Oversight of Human Genome Editing, https://www.who.int/teams/health-ethics-governance/emerging-technologies/expert-advisory-committee-on-developing-global-standards-for-governance-and-oversight-of-human-genome-editing.

Yeager, "As Brain Organoids Mature, Ethical Questions Arise", *The Scientist*, August 1, 2018, https://www.the-scientist.com/features/brain-organoids-mature--raise-ethical-questions-64533.

Yousfi, Louisa. "L'éthique des vertus selon Aristote", Nicolas Journet éd., *La Morale. Éthique et sciences humaines*. Éditions Sciences Humaines, 2012, pp. 96-99. Available at: https://www.cairn.info/la-morale--9782361060312-page-96.htm.

International Society for Stem Cell Research, Guidelines for Stem Cell Research and Clinical Translation, https://www.isscr.org/guidelines.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

11 ANNEXES

The basic documents were compiled by a team of seven members with interdisciplinary profiles of the Ethics Committee for INSERM, the only public research organization in France entirely dedicated to human health. Contributions were also received from all HYBRIDA partners.

11.1 Annex 1: INSERM team

Hervé Chneiweiss (MD-PhD) is a neurologist and neuroscientist, Research Director at the CNRS. He has been involved in neurogenetic research on diseases such as cerebellar ataxias and the molecular mechanisms involved in glial plasticity and the development of brain tumours. Technical approaches include proteomics, metabolism, epigenetics, cell cultures, animal models, single cells. He has published over 170 original scientific papers (h=46). He is currently Director of the Neuroscience Paris Seine - IBPS research Centre (CNRS/INSERM/Sorbonne University). HC is also involved in bioethics; first (2000-2002) as an adviser on the life sciences and bioethics to the Minister of Research and Technology, then as member of the Scientific Council for the Parliamentary Office for Scientific and Techniques Assessment (2003-2016), member of the National Consultative Ethics Committee (CCNE; 2013-2017), and currently as Chairman of the INSERM Ethics Committee and EMBL Ethics Board, former chair of the UNESCO International Bioethics Committee. He is a former Editor-in-Chief of Medicine/Sciences (2006-16). He has published several books for the general public (latest: “Notre cerveau”, “L’Iconoclaste”, 2019).

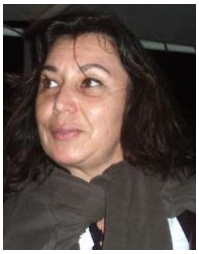
Holding an MD and an MSc in molecular biology, Dr. Anne Dubart-Kupperschmitt has lengthy experience and expertise in the biology of human stem cells, gene transfer and molecular and cellular gene therapies. Her research interests are currently focused on the differentiation of human pluripotent stem cells into hepatic cells (mainly hepatocytes and cholangiocytes) as well as the generation of liver organoids from patient-specific iPSCs in order to model liver diseases, setup gene/cell therapy approaches or for drug screening and toxicology studies. She has published more than 100 peer-reviewed articles. She is a member of the steering committee of the Research Group on Organoids for the French Alliance for Life Sciences (Aviesan) in health technologies and the molecular basis of life, where she is more specifically in charge of the bioethics work package. She is also member of the INSERM Ethics Committee.

A philosopher by profession, Dr. Bernard Baertschi was Senior Lecturer at the Institute for Biomedical Ethics and at the Department of Philosophy in the University of Geneva (Switzerland) until he retired in 2014. His doctoral dissertation was dedicated to a French philosopher from the post-Enlightenment period, Maine de Biran, but he soon became interested in moral philosophy and bioethics. He has been a member of several Ethics Committees in Switzerland: the Federal Ethics Committee on Non-Human Biotechnology (ECNH), the Ethics Committee for Animal Experimentation of the Swiss Academy of Science (ScNat), while in France he is currently a member of the INSERM Ethics Committee, where he leads the Working Group on Organoids. He has published several books on the ethics of

This project has received funding from the European Union’s HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

biotechnologies (genetic engineering and medically assisted procreation), of synthetic biology and of the neurosciences.

Dr. Jean-Luc Galzi graduated in biochemistry before receiving his PhD in biorganic chemistry. He then trained for a further eight years in molecular neurobiology at the Pasteur Institute, Paris, before starting his own research group on the dynamics and pharmacology of G Protein-coupled Receptors in Strasbourg. He is currently Research Director at the CNRS, Director of the Research Institute of the School of Biotechnology and principal investigator in chemical biology of chemokines and their receptors. His research interests focus on pharmacology and all the techniques and tools that facilitate drug discovery and development. He has published more than 100 peer-reviewed articles, mostly in chemistry and pharmacology, filed 10 patents and founded a start-up company in drug discovery. JL Galzi is currently directing a national research infrastructure in bioactive compound discovery (probes and drug candidates) comprising academic chemical libraries, screening platforms, chemoinformatics and preclinical ADME studies. He also coordinates a work group on organoids, the four main topics of which concern design and construction, characterisation, applications and training on organoids.



Corinne Sébastiani holds a PhD in molecular biology, specialised in genomics (sequencing and mapping) and in the ethical and legal aspects of the exploitation of research in the life sciences. She also holds a Master's degree in intellectual property and patent law. As assistant to the Director of the Institute of Technologies for Health, she leads the community and coordinates several national programmes on new technologies for health. She is a member of the steering committee for the Research Group on Organoids of National Centre for Scientific Research (CNRS) including work packages on clinical research and regulation, bioethics and training. She is also member of the INSERM Ethics Committee.

Jacques Haiech is Honorary Professor of Biotechnology at the University of Strasbourg. He holds a master's degree and teaching qualification in Mathematics, and a master's degree and PhD in Biology. He has worked on the understanding of information management by the cell regarding both fundamental research and drug development (oncology and inflammatory pathologies). Through his responsibilities in research administration (Director of the National Genomics Programme, Director of the Biology-Health Department at the French Ministry of Research and Higher Education, Scientific Delegate to the AERES), he has worked on research evaluation methodologies and, more recently, on the links between scientific integrity and methods for the evaluation of scientists.

PhD in Sociology from the École des Hautes Études en Sciences Sociales, Ioana Andreescu is an associated researcher at the Raymond Aron Centre for Sociological and Political Studies. She had previously been in charge of developing and implementing an education strategy to improve the harmonisation of innovative and socially responsible degree programmes within EIT Health. She has contributed to several international projects, such as "Digital Futures" or the European Commission "European Space" programme. In 2015, she won the Henri Desroche Prize for social transformation projects.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

11.2 Annex 2: Ten conceptual uncertainties pertaining to the ontological status of organoids as hybrids

HYBRIDA's WP1 produced an analysis of the principal uncertainties resulting from organoid research. One potential way to clarify the status of organoids given their extensive "hybridity" is to explore explicitly the ways in which common conceptual distinctions break down when applied to this complex case. In this section, we offer a list of ten conceptual distinctions that are briefly defined in very general terms and applied to organoids. By reviewing some of the classical conceptual distinctions applicable to organoids, we have provided an initial conceptual map which could be used to form ontological judgments on organoids and in future discussions on the ethical and regulatory issues raised by these entities.

The conceptual distinctions listed below are based on common sense, on canonical, philosophical concepts, and on more recent theoretical developments in the philosophy of science:

1. From a legal viewpoint, human organoids are things, but they might also be related to persons in specific manners that need to be investigated.

2. Organoids are objects of research and development, yet they might become subjects.

3. For many scientists, organoids are more than a mere cell culture, but they are not full organs, and

even less organisms.

4. Are organoids living entities, or should we identify them as mechanisms?

5. Referring to another classical philosophical distinction, one might ask whether they are natural entities or

artefacts?

6. Organoids belong to science, offering a means to gain knowledge, and they are also technologies i.e.

objects designed to have an impact on the world in which we live.

7. They belong at the same time to the category of research tools and to that of potential clinical devices.

8. As tools for research and clinical use, organoids are mere means, but they can also be seen as ends from the perspective of technological development or regenerative medicine.

9. We tend to think of organoids as actual biotechnological entities that have a certain nature, but most research using organoids is focused on their development and thus sees them as part of a larger process oriented towards the future.

10. Certain types of organoids, such as chimeras, tend to blur the distinction (entrenched in common sense) between humans and animals.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

11.3 Annex 3: What is behind the "ethics by design" requirement?

1. Introduction

Ethics by design (ED) has been developed recently as **an approach to prompt the developers of new technologies to take ethical issues into consideration at the design process stage**. As seen from a short survey of the academic literature and grey literature dedicated to the topic, **the main field for the application of ethics by design is artificial intelligence** (AI). Yet, as is also mentioned, the ethics by design approach refers to a broader perspective regarding the ethics of new technologies that could be applied to many emerging fields, including biotechnologies.

The H2020 SwafS call for proposals on organoids stipulated that: "The work undertaken is expected to produce operational guidelines for the field. The guidelines should ensure "ethics by design" and be drafted to support the work of the research community, research ethics committees and integrity bodies" (p.47). What should be the approach that underpins the drafting of guidelines so that they will "ensure ethics by design"? What is expected in this context? How can we develop a methodology that meets this requirement?

The ED approach is guided by a need to anticipate effectively the ethical issues that will arise with emerging technologies (i.e. technologies in the making that are not yet entrenched in society). This is particularly the case with AI, as major impacts are expected with respect to society, the economy, social life, medicine warfare, and other domains. When the impact of a technology is supposed to be considerable (to impact society as a whole or have important consequences) and almost immediate (there will be no time to mitigate the risk once the technology is disseminated), ethical issues must be assessed and ethical principles implemented at the earliest possible stage of technological development, or in other words during the design process. A technology that is designed to conform to certain ethical standards is more likely to limit the negative consequences that might arise when it becomes widely used.

According to the family of "by-design" approaches, it is possible to include certain options in the technology so that it will conform to the values that matter. For instance, rather than protecting privacy by placing limits on the use of data, privacy-by-design prompts a developer to produce a tool that can be functional while requiring a minimal amount of data. Indeed, this approach ensures the design of a tool that will collect a limited amount of sensitive data rather than building one that collects a large quantity of data and then to impose restrictions on their conservation and use.

The fact that ED has been developed with AI and robotics as the main targets deserves further consideration. AI systems are not only artefacts that shape our environment, or that we use: they contribute to decision-making in very practical ways. As *intelligent* systems, they are asked to sort, discriminate or choose for us in what might be *ethical* judgments (for instance, the debate on self-driving vehicles). The "laws of robotics" proposed by Asimov clearly express the type of constraints that we would wish to impose on technological artefacts that will make decisions. Indeed such decision are primarily the developer's ones.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

The problem is therefore as follows: is this approach fit for our object, organoid technology? Organoids do not make decisions. They are not artificial moral agents. However, they may be considered more generally as artefacts, or technological devices, and ED would apply to them in this general sense. Furthermore organoids are not just any type of artefact, they are *biotechnologies*. Biotechnologies are a family of technological devices that build upon biological materials and possess certain properties of living beings. Biotechnologies can evolve and convey a sense of uncertainty, insofar as researchers do not know of what they are capable. Indeed, this is also true for new forms of generative AI. Further, questions around the development and use of organoids mostly refer to biomedical research. Unlike many emerging technologies that are precisely the target of an ED approach, biomedical research is already regulated, with numerous procedures in place. For instance, research using human material or involving human participants is supervised by laws and guidelines — up to the point that biomedical research is considered as a reference for the development of regulations in other fields. What would we gain by *considering organoids as emerging technologies*? Is there something in the ethics of emerging technologies that might benefit bioethics/biomedical research ethics?

In this document, we will try to answer these questions through an in-depth analysis of ethics by design and its history, before considering its application to biotechnologies.

2. Definition and history of ethics by design

Several contextual elements need to be recalled in order to capture the general ideas behind ED, and before asking how this approach can be put to use in our context. The expression "ethics-by-design" has been put forward in recent years in the EU as a keyword, up to the point that a recent EGE (European Group on Ethics in Science and New Technologies) statement claimed, in a somewhat retrospective reading of history, that the "design turn and the idea of the value-laden nature of scientific and technological innovation has also been foregrounded by EU research funding in the last decades." (EGE 2021) This is especially the case relative to two European SwafS projects: SIENNA & SHERPA. The SIENNA report (Stakeholder-Informed Ethics for New technologies with high socio-ecoNomic and human rights impAct, 2018-2021) provided an ethical framework for human genomics, human enhancement, artificial intelligence and robotics. It also contained reflections on the methodology for the production of guidelines with respect to emerging technologies that are of particular importance to HYBRIDA (Brey et al. 2021). SHERPA (Shaping the ethical dimensions of smart information systems—a European perspective, 2018-2021) addressed smart information systems with specific emphasis on ethics by design. Both projects prompted software designers and developers to follow the ED approach, proposing concrete steps for implementation.

To say of something that it has to be ethical *by design* is equivalent to saying that it has to be designed as ethical. However, if these seems similar they are not the same. The former is keeping ethics in mind through each stage, including the possibilty that something isn't designed, or is pulled at the last minute, while the latter could in principle involve ethics at only the initial and latter stages.The ED approach has notably been developed in the context of the SIENNA and SHERPA EU SwafS projects. One of the objectives of these projects was to produce guidelines that would ensure ED for AI developers. According to these projects, ED starts from the definition of general principles, or ethical values, that we aim to respect (e.g., fairness) plus consultation, agreement and consensus. These values are translated into more specific requisites at the system level (e.g., in order to comply with fairness, the system must

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

not produce discrimination, such as gender or racial bias). **Guidelines are proposed to ensure that the requisites are taken into consideration during development of the product** (e.g., test/screen for bias at steps X and Y) and that the approach can be adapted to numerous development methodologies, up to the most technical level.[77]

Ideally, ED aims to embed ethical values in the system itself. It aims at "guaranteeing ethical behaviour "by design", i.e., embedded in the system's implementation" (Dignum et al. 2018). The translation of ethical values into concrete requisites and actions in the course of development is what ensures that the product (software, algorithm, etc.) complies with certain desiderata and prevents the generation of unethical consequences. On the face of it, ED does not prevent other forms of ethical oversight or follow-up.[78] Furthermore, **ED is neither an off-the-shelf toolbox nor a specific methodology that must be followed strictly, but a general approach**. To adapt the ED approach to different software development methodologies or to different fields of research requires some research (Coeckelbergh 2019; Brey et al. 2021), and this is precisely what we wanted to do here.

One obvious reason for development of the ED approach is **the need to anticipate ethical issues that will arise with emerging technologies** and to build that scope for anticipation into the lifetime of the technology. Emerging technologies are technologies that are not yet fully or at all entrenched in society. We cannot deal with ethical issues raised by emerging technologies (e.g., medical nanotechnologies, internet of things, metaverse) in the same way as with issues raised by entrenched technologies (e.g., automotive technology, antibiotics, nuclear power) (Brey 2017). In this context, the development of a technology can lead very rapidly to unexpected harmful consequences. Nobody wants the technology to cause damage, so good sense commands that we carefully anticipate the potential consequences of developing a technology that will have an impact on society. This is particularly the case with AI, which will have marked expected impacts on society, the economy, social life, medicine and warfare. As the impact is likely to be strong (it will impact society as a whole, or have important consequences) and almost immediate (there is no time to mitigate the risk once the technology has been launched, because as soon as the technology develops, it will start to impact the lives of people), there is obviously a need to anticipate the risks.

As a consequence, ethical issues should be taken into consideration as early as the conception phase. This concern was already expressed in a family of "X-by-design" (Fischer 2019) concepts, ethics by design being only the latest in a series. For instance, "privacy-by-design" aims to protect users from abuse through the unjustified collection of personal data. Rather than protecting privacy by placing limits on the use of data, privacy-by-design prompts the developer to produce a tool that will function but require a minimal amount of data. It offers a greater guarantee of designing a tool that collects a limited amount of sensitive data than one that collects a large quantity of data where restrictions must be imposed on their conservation and use.

[77] This problem is specific to software development (the ED approach should be adapted to, e.g., AGILE), but there is an analogy with our problem: how specific should be the guidelines concerning different methods to develop organoids? Referring to the five layers of SIENNA/SHERPA (values, requisites, guidelines, methodology, tools), we should ask ourselves how many layers exist in biomedical research and how the guidelines produced by HYBRIDA can identify (or target) each of these layers. For instance, SIENNA does not go into detail on each methodology but gives an example of how the ED approach can be implemented in AGILE.

[78] Nobody claims that ED will solve all the ethical issues raised by emerging technologies, and indeed, some documents present both "ethics by design" and "ethics of use" (European Commission 2021).

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

During recent decades, there has been **a general boom in the development of guidelines, charts, codes of integrity, and so on**, in all fields of research (Iphofen 2020). This type of "soft law approach" may be seen as a second best when compared to regulation by law, complementary at first since legal instruments are also imperfect, given that they encourage a 'by the numbers' response, whereas good guidelines can encourage thought. Yet the law is condemned to stay somewhat vague, with a certain level of abstraction, when covering different fields of application, while standards can go into the detail of a technology. Also, guidelines offer a more practical approach than the general reiteration of universal principles or ethical values with which everyone agrees. Again, there is added value in the translation of values into concrete requirements.[79] Soft and hard law can be articulated in several ways. They can be seen as opposites but they appear complementary: "In all cases, the alternatives being considered can be divided into two types: regulation by means of legislation and standards, or design, ensuring that the systems themselves take ethical decisions at all times" (Dignum et al. 2018), or as complementary, as guidelines propose concrete procedures that the law cannot offer.

The ED approach is currently enjoying a certain degree of success among developers and in the context of AI ethics, as can be seen through recent references in the literature (Felzmann et al. 2020; Iphofen and Kritikos 2021; Urquhart and Craigon 2021; Nussbaumer, Pope, and Neville 2021). Although debate continues regarding the best implementation of ED (for instance, the importance of data instead of software development (Gerdes 2021), there seems to be a consensus on the interest of developing such an approach.

3. The philosophy behind ethics by design

3.1. Ethical artefacts?

The principal philosophical hypothesis underlying the ED approach is the idea that ***values are embedded in artefacts***. This is emphasized in the EGE statement: "Everything designed, every artefact, piece of technology and human-made system contains the preferences, values and worldview of its designers and makers" (EGE 2021). This is a longstanding view in the philosophy of technology, akin to the claim that "artefacts have a politics" (Winner 1980). A well-known example taken by Winner is the bridges over the Long Island Highway, built intentionally low so that buses conveying a population of lower social status could not access certain places. Through the constraints that they impose on society, artefacts represent a certain social order or favour certain groups or individuals in society. They constrain the course of our actions, or incline us in certain directions, thus limiting and orienting choices.

Ethics by design builds on this idea to propose that **artefacts are produced so that the constraints they impose are somehow *ethical***. There is an interesting connection with the debate on dual-use (on technology and warfare), as, in a sense, ethics-by-design is symmetric to dual-use. While dual use is the development of a technology (e.g., nuclear power) designed overtly for civilian purposes and transformed quickly into a military device, an artefact designed as ethical offers a guarantee that it cannot be misused. Hence, producing artefacts that embed ethical values is a type of democratic ideal where objects that circulate and disseminate in our society and around the world bring these values to the world and cannot be diverted. If

[79] The same problem occurs with the principist approach of biomedical ethics and has been a subject of debate for decades: with the very same "principles of bioethics," one can defend a position or its opposite, depending on the weighting and interpretation of the principles.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

artefacts behave ethically – or constrain human behaviour so that it is ethical – then in a way we do not have to rely on human judgments that are always susceptible to err.[80]

This being said, **there is a specificity of AI systems that justifies the current trend in ED in this very particular field**. AI systems are not only artefacts that shape our environment, or artefacts that we use, they are **artefacts that make decisions**. As *intelligent* systems, they are asked to sort, discriminate and choose for us. They do not impose passive constraints only, such as the low bridges, but they already can. They make decisions that are at the level of ethical judgments. Here, the debate is more on the side of robotics and autonomous machines (e.g., autonomous weapons) than on the side of general objects. The Asimov laws nicely express the type of constraints that we want to impose on artefacts that can make a decision.

Now we can gradually ask how all this applies to organoids, as organoids are neither simply *artificial* objects such as bridges, nor systems that can make decisions by themselves.[81] This methodological inquiry aims precisely to clarify these points. Before doing just that, let us turn to more general points that can be extracted from the ED approach.

3.2. Anticipating potential ethical issues

Anticipation is a keyword that is crucial to all ED approaches. The adjective of "proactive" (borrowed from administration and business language) is recurrent, as opposed to reactive. "Ethics by Design is intended to *prevent* ethical issues *from arising in the first place* by addressing them during the development stage, rather than trying to fix them later in the process" (European Commission 2021).

The need to anticipate is justified by the idea that emerging technologies will in any case have an impact on society. In the case of AI, there is no window to experiment properly without impacting real people. If we want the AI to work, we need to feed it with real data, and as soon as the algorithm is used, it will have an impact. But we now that unbiased data may be impossible. All artefacts might not exhibit this feature so clearly – a car can be built and then crash-tested with a mannequin in it. For AI systems, ethical issues ought to be properly considered occur at the very start of the process. One obvious example is data collection: if data are biased, then the entire system will be biased. Other formulations stipulate that ED "implies bringing the debate on the ethical and societal implications at the primary stage of the research process" (D"Aquin 2018) or that "ethical reflection is required across the whole product lifecycle, including the early conception phase" (Keber 2021).

The general idea of anticipating as early as possible – and not waiting until products hit the market before regulating – is surely laudable, but how is this specific to the ED approach? In a way, it seems that anticipating the consequences of technologies has been the bread and butter of many scholars for decades, from philosophers of technology to bioethicists. Regulation itself does always intervene *afterwards*, as there are always mechanisms to anticipate the consequences (e.g., market authorizations for drugs).

[80] Which raises further questions about what it means to behave ethically if this behaviour is constrained by technology, but this falls outside the scope of the current discussion.
[81] As far as we know, and in the near future, maybe one can envision a neural organoid so complex that it forms a network on which an artificial intelligence could run.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

In defence of ED, one could say that **this capacity to anticipate precisely concerns the lack of a specific methodology and that there have been many failures of these oversight mechanisms** (e.g., in health, environment). In other words, the anticipatory net has many breaches, and the State, or authorities, are sometimes powerless when it is realised that something that they authorized is doing more harm than good (e.g., pesticides). In this sense, ED would be opposed – at least methodologically – to several current approaches in the ethics of emerging technologies. Ethics committee approaches are often limited in the sense that committees authorize research based on analysing the conduct of research (is data managed properly? does research respect the rights of participants?) but without considering the long-term impacts of the research on society. Furthermore, an ethics committee gives approval at an early stage with no incentive to reflect on the ethical issues that might arise once the project is launched (D"Aquin 2018). ELSI-type approaches have also been frequently criticised as parts of projects that are already launched, so that ethical concerns cannot take the lead over the scientific programme in the event of conflict (see the many objections to the ELSI approach in, e.g., nanotechnology, synthetic biology).[82] There is more than anticipation, there is a concern related to the production process of science (i.e., the role of ELSI, and ethics more broadly, in governance).

We mentioned earlier that one characteristic of emerging technologies is that they will have an impact on society anyway. There is no methodology that can perfectly anticipate all the potential impacts of a technology. Starting from the premise that we cannot foresee everything, the introduction of a new technology into society can be considered as a type of social experimentation. "The question, therefore, is not simply one about determining whether and under what conditions a given technology is to be deemed morally acceptable, but also about whether it is morally acceptable to test a given technology openly within our societies, and under what conditions this can be said to be the case." (Nurock, Chatila, and Parizeau, 2021). Under these circumstances, **the ED approach offers a way to mitigate risks when we cannot avoid uncertainty: experimenting in and with society**.[83] The EGE states that in the absence of anticipation and early engagement, the ethical choices remain in private hands: "We cannot leave the design of our future world to coincidences and to those who design for self-serving purposes outside democratic control. Moral reflection should therefore be situated when and where it can make a difference" (EGE 2021). Ethical regulation is not only about constraints, we need a positive construction of ethics within the system, leading to human flourishing (Coeckelbergh 2019). This means not only drawing red lines but finding a way to empower society through technology.

3.3. Responsible innovation with all stakeholders

Another major element in the philosophy of ED is inherited from the "responsible research and innovation" field. The RRI field has been growing for two decades and the move to ED can be interpreted as an operationalisation of RRI in certain areas of research. In that case, an

[82] For instance, it can be almost impossible to reorient the course of action of a scientific programme once that programme has been launched (Rabinow and Bennett 2012).

[83] The situation differs quite markedly from one domain to another. This discourse is relevant for AI, but we could analyse the situation differently for drug testing. Testing a drug (for safety and efficacy) always involves making a leap, and the protocols for clinical research are intended to ensure that this leap is as circumscribed and reasonable as possible at each step. If there is an issue, then we want to make sure that the issue is circumscribed to the participants, people engaged in the test and covered by medical follow-up.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

interpretation of the "by-design" requirement for organoid guidelines would be simply that **we want organoid research to conform to RRI**.

What does RRI entail? Firstly, the involvement of stakeholders at an early stage in the process. Research is not only *for* society but *with* society. Stakeholders are of course potential users of the technology and private entities with an interest in developing the technology (companies, civil organizations, etc.). Another methodological point is interdisciplinarity: responsible research cannot reach all stakeholders and examine all potential ethical issues without support from social science and the humanities. The ED literature on AI insists that ED is not only a toolbox to be used by AI coders/researchers, it is also a promise of engagement with other disciplines (Coeckelbergh 2019; Gerdes 2021; Nurock, Chatila, and Parizeau 2021). An ethical design should identify the critical steps at which an ethical assessment will be required and who will take part in this assessment.[84] **In this regard, it is worth noting that the ED approach does not propose "injecting" ethics at an early stage and then getting rid of it, it fosters continuous discussion among researchers and stakeholders, at least in theory (EGE 2021)**. According to Coeckelbergh, ED offers a practical solution to bridge the gap between the general principles of ethics and concrete actions.

It should be noted that some authors see a tension between the technicality of the work (implementation in technology – first level of expertise – of ethics – second level of expertise) and the call for external stakeholders and broad participation (Gerdes 2021). The emphasis on stakeholder participation and global discussion may in a way downplay the technical aspects of a project.

The RAD methodology emphasizes quick, iterative development cycles, rapid prototyping, and immediate feedback from end-users, aiming to solve problems by adapting to changing requirements in a fast-paced environment. This approach aligns with ED's proactive stance on anticipating ethical issues by allowing for quick adjustments as ethical considerations evolve during the development process.

Similarities between RAD and ED include their iterative nature, focus on stakeholder feedback, and adaptability. Both approaches prioritize early and continuous involvement of relevant parties (in RAD, the end-users; in ED, stakeholders concerned with ethical implications) to refine the product or technology to better meet its intended objectives and ethical standards.

However, the differences lie in their primary focus and implementation strategies. RAD is primarily concerned with the efficiency and effectiveness of software development, aiming to reduce development time and increase flexibility without a specific emphasis on ethical considerations. In contrast, ED is explicitly focused on embedding ethical principles into the design and development of new technologies, ensuring that products not only meet functional requirements but also adhere to ethical standards from the outset.

By integrating RAD's flexibility and speed with ED's ethical foresight, developers can create technology that is not only rapidly developed and responsive to user needs but also inherently ethical. This hybrid approach ensures that as technologies evolve, they do so in a manner that

[84] For instance, suggesting (as has been done elsewhere; REF), that a cellular biologist with no competence in cognitive science or in philosophy of the mind should *not* decide by herself whether a given brain organoid is conscious or not and how this possibility should be assessed, would correspond to this interdisciplinary-hence-more-likely-ethical-by-design framework.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

is both ethically responsible and adaptable to the fast-paced changes characteristic of modern technological development.

To effectively apply RAD within the ED framework, it's crucial to include ethical considerations as part of the rapid prototyping and feedback cycles. This integration ensures that ethical compliance is not an afterthought but a foundational component of the development process, enabling the creation of technologies that are both innovative and ethically sound.

To discuss the use of the Rapid Application Development (RAD) approach in the organoid field, it is essential to recognize the unique challenges and opportunities presented by this area of research. Organoid technology, which involves growing three-dimensional organ avatars from stem cells, requires a highly adaptive and responsive research and development strategy due to its complexity and the fast pace of scientific advancements.

Integrating RAD with Ethics by Design (ED) in organoid research emphasizes rapid iteration and stakeholder feedback, similar to software development, but with a focus on addressing ethical, biological, and technical challenges inherent to manipulating life-like structures. This combination allows for swift adaptation to new ethical considerations and scientific discoveries, ensuring that organoid research progresses in a manner that is both innovative and ethically responsible.

The primary difference in applying RAD in organoid research compared to software development lies in the tangible ethical implications and biological constraints. The iterative cycles of RAD must be carefully balanced with rigorous ethical oversight, ensuring that each development phase respects the dignity, safety, and rights of all stakeholders involved, including potential therapeutic applications for patients.

In summary, upgrading the RAD approach for organoid research involves a nuanced integration of rapid development cycles with a strong ethical framework, tailored to the specific challenges of this cutting-edge scientific field. This ensures that organoid technologies not only advance quickly but do so with a fundamental commitment to ethical principles.

This section was mainly contributed by Maxence Gaillard with inputs from Jacques Haiech and feedbacks from the focus groups gathered by WP4

References

Brey, Philip. 2017. “Ethics of Emerging Technology.” In *The Ethics of Technology: Methods and Approaches*, by Sven Hansson. Rowman and Littlefield International.

Brey, Philip, Brandt Dainow, Yasemin J. Erden, Amal Matar, Philip Jansen, Rowena Rodrigues, Nicole Santiago, et al. 2021. “SIENNA D6.3: Methods for Translating Ethical Analysis into Instruments for the Ethical Development and Deployment of Emerging Technologies,” October. https://doi.org/10.5281/zenodo.5541539.

Coeckelbergh, Mark. 2019. “Artificial Intelligence: Some Ethical Issues and Regulatory Challenges.” *Technology and Regulation* 2019 (May): 31–34. https://doi.org/10.26116/techreg.2019.003.

This project has received funding from the European Union’s HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

D. Urquhart, Lachlan, and Peter J. Craigon. 2021. “The Moral-IT Deck: A Tool for Ethics by Design.” *Journal of Responsible Innovation* 8 (1): 94–126. https://doi.org/10.1080/23299460.2021.1880112.

D”Aquin, Mathieu. 2018. “Towards an “Ethics by Design” Methodology for AI Research Projects.” In *AIES”18*.

Dignum, Virginia, Matteo Baldoni, Cristina Baroglio, Maurizio Caon, Raja Chatila, Louise Dennis, Gonzalo Génova, et al. 2018. “Ethics by Design: Necessity or Curse?” In *Proceedings of the 2018 AAAI/ACM Conference on AI, Ethics, and Society*, 60–66. AIES “18. New York, NY, USA: Association for Computing Machinery. https://doi.org/10.1145/3278721.3278745.

EGE. 2021. *Values for the Future: The Role of Ethics in European and Global Governance*. LU: Publications Office of the European Union. https://data.europa.eu/doi/10.2777/595827.

European Commission. 2021. “Ethics By Design and Ethics of Use Approaches for Artificial Intelligence.” https://ec.europa.eu/info/funding-tenders/opportunities/docs/2021-2027/horizon/guidance/ethics-by-design-and-ethics-of-use-approaches-for-artificial-intelligence_he_en.pdf

Felzmann, Heike, Eduard Fosch-Villaronga, Christoph Lutz, and Aurelia Tamò-Larrieux. 2020. “Towards Transparency by Design for Artificial Intelligence.” *Science and Engineering Ethics* 26 (6): 3333–61. https://doi.org/10.1007/s11948-020-00276-4.

Fischer, Flora. 2019. “L”éthique by design du numérique : généalogie d”un concept.” *Sciences du Design* 10 (2): 61–67.

Gerdes, Anne. 2021. “A Participatory Data-Centric Approach to AI Ethics by Design.” *Applied Artificial Intelligence* 0 (0): 1–19. https://doi.org/10.1080/08839514.2021.2009222.

Iphofen, Ron. 2020. *Handbook of Research Ethics and Scientific Integrity*. Springer. https://link.springer.com/referencework/10.1007/978-3-030-16759-2.

Iphofen, Ron, and Mihalis Kritikos. 2021. “Regulating Artificial Intelligence and Robotics: Ethics by Design in a Digital Society.” *Contemporary Social Science* 16 (2): 170–84. https://doi.org/10.1080/21582041.2018.1563803.

Keber, Tobias. 2021. “Digital Ethics by Process? Technical Conflicts and Policy Ethics Committees in Europe.” *Informatio. Revista Del Instituto de Información de La Facultad de Información y Comunicación* 26 (1): 216–29. https://doi.org/10.35643/Info.26.1.11.

Nurock, Vanessa, Raja Chatila, and Marie-Hélène Parizeau. 2021. “What Does “Ethical by Design” Mean?” In *Reflections on Artificial Intelligence for Humanity*, edited by Bertrand Braunschweig and Malik Ghallab, 171–90. Lecture Notes in Computer Science. Cham: Springer International Publishing. https://doi.org/10.1007/978-3-030-69128-8_11.

Nussbaumer, Alexander, Andrew Pope, and Karen Neville. 2021. “A Framework for Applying Ethics-by-Design to Decision Support Systems for Emergency Management.” *Information Systems Journal* n/a (n/a). https://doi.org/10.1111/isj.12350.

Rabinow, Paul, and Gaymon Bennett. 2012. *Designing Human Practices: An Experiment with Synthetic Biology*. Chicago, IL: University of Chicago Press. https://press.uchicago.edu/ucp/books/book/chicago/D/bo12986260.html.

This project has received funding from the European Union’s HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

Verbeek, Peter-Paul, 2008, Obstetric Ultrasound and the Technological Mediation of Morality: A Postphenomenological Analysis, *Human Studies* 31(1) 11-26

Winner, Langdon. 1980. “Do Artifacts Have Politics?” *Daedalus* 109 (1): 121–36.

This project has received funding from the European Union’s HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

11.4 Annex 4: SIENNA's Methodological Steps for Ethics by design

According to the SIENNA project, as well as to related biography which provides methodological guidance for the building ethical guidelines in general, there is a 5-layer Model for Ethics by Design[85]:

I. **Principles**: Reach consensus on the key moral values and principles that apply to the technology field and that we want to respect. Establish a list of values and principles that should guide the development process.
II. **Ethical Requirements**: Translate the general values into ethical requisites.
III. **Methodologies**: Translate the ethical requisites into actionable methodological guidelines. The ethics guidelines identify the specific steps in the development process where ethical assessment/intervention should take place. Guidelines are proposed to ensure that the requisites are taken into consideration during development of the product.
IV. **Ethics by Design Guidelines:** Choose and describe an established design methodology.
V. **Tools & Methods**: Develop tools and assessment methods to address specific issues, consider special topics.

It is possible to envisage an understanding of ethics by design in terms of methodological requisites, which can be summarised as follows:

a) **anticipation** of all the consequences of the emerging technology under scrutiny;

b) paying attention to the **evolution** of the technology through a life cycle (ethics is not just a green light at the start of the research project, but should cover all aspects of the technology, as distinct issues might arise at distinct stages of technology development);

c) **inclusion** of all stakeholders potentially concerned when dealing with ethical issues;

d) **interdisciplinarity** (all stakeholders cannot be reached and all potential ethical issues cannot be examined without support from the social sciences and humanities);

e) **the responsibility** of technology developers (they are ultimately responsible for integrating the ethical requisites into the data/software/technology) and, symmetrically, ethics by design as a form of **democratic control** over technology development.

If, in specific contexts the ethics by design approach might not be sufficiently internalised for the research community, it could be supplemented by external audits, IRBs or RECs, and specifically oriented to scrutinise the different steps of the research in question and fill existing gaps.

[85] Brey, Philip, Brandt Dainow, Yasemin J. Erden, Amal Matar, Philip Jansen, Rowena Rodrigues, Nicole Santiago, et al. 2021. *SIENNA D6.3: Methods for Translating Ethical Analysis into Instruments for the Ethical Development and Deployment of Emerging Technologies*, p.53. https://doi.org/10.5281/zenodo.5541539.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

11.5 Annex 5: How these operational guidelines were drawn up within the framework of the HYBRIDA project

HYBRIDA interactions

OGLs were deliverable 5.1 of the HYBRIDA project, under the responsibility of WP5. WP5 was a highly interactive working group, responsible for delivering several HYBRIDA products (Operational Guidelines for Researchers in the Field, the Code of Responsible Conduct for Researchers and eventually the Supplement to the ECoC). In order to achieve these goals, WP5 liaised with both HYBRIDA partners and external experts. WP5 initially organised several meetings and exchanges with WP1. Drafts and written materials were circulated regularly: exchanges between WP5 and WP1 resulted in the document entitled: What is behind the "ethics by design" requirement? (see: Annexes 3 and 4). We also thoroughly considered the ten conceptual uncertainties highlighted by WP1 and how they may inform our operational guidelines and code of conduct. Following the WP1 Brussels workshop on the 30 March 2022, WP5 also provided comments and proofreading on the D1.4 deliverable.

Furthermore, the systematic mappings performed in WP2 and WP3, and the identification of existing gaps in WP6, support drafting of the Operational Guidelines and the Code of Conduct. The WP5 programme manager (Ioana Andreescu until 31 March 2023) was a member of the WP2 Amended Working Group and participated in the discussions and drafting of WP2 documents. Further, in the context of WP3, INSERM conducted expert interviews for the mapping of the normative, REI (research ethics and integrity) framework for organoids and related technologies; these interviews were coded and analysed by WP3. WP5 organized a bilateral meeting with WP6 in order to establish how exchanges and interactions would nourish compilation of the Guidelines and the Code.

Furthermore, cooperation between WP5 and WP4 resulted in the joint organisation of two Co-creation workshops, one in Paris on the 19 May 2022 and the second in Copenhagen on 23 June 2022 (for further details, please check WP4 Deliverables).

Organisation of the Kick-off Meeting for WP5 progressed discussions on the ethics of organoids, and enabled planning of the future steps for drafting of the Operational Guidelines together with HYBRIDA partners. The Kick-off Meeting took place on the 19 January 2022 in Paris and was also accessible via Zoom; it featured lectures by Christine Mummery, François Hirsch and Alexei Grinbaum (representing the H2020 Swafs29 programme TechEthos), and gathered more than 30 HYBRIDA collaborators for discussions. Finally, the general plan (including the Table of Contents) for the Operational Guidelines was presented and discussed at the annual General Assembly of HYBRIDA in Athens on 27-28 May 2022.

An advanced draft (dated 20 April 2023) was the result of several interactions and includes numerous remarks, comments and suggestions collected during the HYBRIDA joint meeting on the 28 December 2022, as well as further suggestions and comments resulting from discussions with the HYBRIDA Advisory Board on 23 February 2023. This draft served as the working document for the HYBRIDA General Assembly on the 27/28 April 2023 in Rome, Italy, thus allowing compilation of a new version of the Guidelines. This revised version was discussed among partners during a videoconference on Thursday 6 July 2023; this led to the

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

draft of version 7 presented to partners on Monday 31 July 2023 and circulated to participants of the focus groups to be gathered in September by WP4.

WP4 organized 6 focus groups gathering in fall 2023. This resulted in the document 4.5 published in the end of December 2023 including 80 recommendations. On such basis, WP5 amended extensively the OGLs to produce version 8, circulated to result in v8.5 presented at the concluding gathering 15 may 2024 in Brussels. An additional presentation was made on-line 19 June 2024, where 112 people registered for the 2h webinar. Feedbacks from these two events were collected and integrated to produce the final document presented here.

Consultations with experts

WP5 developed several expert exchanges and organised several bilateral meetings with experts from the field, focusing among others on GMP, ethics-by-design, ethical principles in organoid research, benchmarking, and the building of protocols, etc.

One of the experts consulted was Dr. Tenneille Ludwig, Senior Scientist and the Director of the WiCell Stem Cell Bank (Wisconsin, USA). The meeting took place on the 4 February 2022 and was dedicated to opportunities for collaboration in the field of stem culture conditions in light of the working group she leads within the framework of future ISSCR recommendations. For the ISSCR, Dr. Ludwig's ongoing work focuses on updating the standards for stem cell banking, their characterisation and distribution for research and cGMP grade materials. Since the beginning of 2022, the ISSCR has been working on an updated version of the ISSCR Standards for Basic Stem Cell Research and Dr. Hervé Chneiweiss was one of the reviewers of the document (review submitted in August 2022).

Another meeting was organised with Dr. Alexei Grinbaum, a researcher working on the ethics of new technologies within CEA (Atomic & Alternative Energies Commission, a French industrial and commercial public establishment) who is involved in several EU projects such as TechEthos, the SwafS29 project dedicated to prioritising ethics and societal values in the design, development and deployment of new and emerging technologies (2021-2024) or *RRI-Practice: Responsible Research and Innovation in Practice* (2016-2019). The meeting with Dr. Grinbaum was very important in terms of developing an efficient ethics by design methodology, as well as adding a chapter dedicated to organoid project evaluators and to the evaluation process.

Several organoid researchers took part in three working sessions focused on planning the content of two main sections in the Operational Guidelines: the *Minimum Information About Organoids and their Use* (MIAOU) (Annex 5 and 6) and *Evaluator Checklist for Organoid Experimental Studies* (ECHOES), as described in Annex 7.

Timeline of WP5 activities

Based on the mappings in WP2 and 3 and the 1st stage of the HYBRIDA Engagement Process (HEP), version 1.0 of the Operational Guidelines and the European Code of Conduct for Research Integrity for Organoids and Related Technologies were drafted and sent to the EC in October 2022. Version 2.0 of both products will be drafted after taking account of the results of the 2nd stage of the HEP. The final version (Version 3.0) will be compiled and delivered

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

based on consultations with the experts and professional stakeholders involved in the 3rd stage of HEP[86].

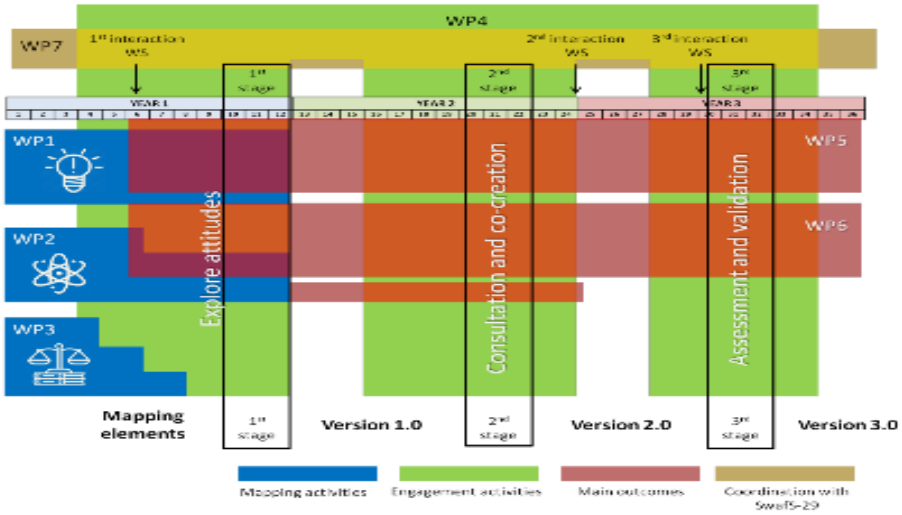


Figure 2: Flow chart and a timeline for HYBRIDA's streamlined activities.

Inputs from other work packages

From WP1: Identification of different forms of conceptual uncertainty (ontological, moral and legal) relating to organoids; development of a socially robust typology for artificial biological entities and analysis of the ethics-by-design concept.

From WP2: Performance of a traditional HTA of organoids by applying existing evidence for efficacy, effectiveness, safety, and cost-effectiveness; definition of basic concepts and perspectives in order to modify HTA to assess organoids, and application of the amended HTA methodology to the assessment of organoids as a health technology.

From WP3: Mapping of ethical dimensions and concerns that have been raised in the past in the context of technologies or families of technologies comparable to organoid research; comparison of relevant regulatory frameworks in Europe and beyond.

From WP4: Organisation of three deliberative (mini-public) workshops, each with 15-20 participants who included representatives of the general public, patients, donors and CSOs. Organisation of two co-creation stakeholder workshops in Paris and Copenhagen involving 15-20 participants who included researchers (academic and industrial), members of RECs and RIOs, policy makers, legal experts, patient organizations and biobanks, in order to explore stakeholder views on the initial and collective elements of the HYBRIDA Operational Guidelines and Code of Responsible Conduct. Organization of 6 focus groups to analyse WP5 and WP6 documents.

From WP6: Analysis of existing, applicable ethics and normative frameworks in Europe and beyond and identification of assessment gaps in relation to donors, patients, civil society, Open Science (OS) regarding organoid generation as a social practice, and assessment of gaps identified through stakeholder engagement.

[86] From the HYBRIDA Project, p.36.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

11.6 Annex 6: Working groups for the design of MIAOU and EChOES

Minimum Information about Organoids and their Use for Researchers (MIAOU)

First working session – Paris, Biopark 11 March 2022

LIST OF PARTICIPANTS

Ioana Andreescu, HYBRIDA WP5 Project Manager, INSERM, Paris.

Celine Cougoule, Researcher at the Institute of Pharmacology and Structural Biology in Toulouse.

Anne Dubart-Kupperschmitt, Director of Research, Pathophysiology and Therapeutics of Liver Diseases, INSERM, Paris.

Jean-Luc Galzi, Director of the Research Institute of the Strasbourg Biotechnology School.

Jacques Haiech, Honorary Professor of Biotechnology at the University of Strasbourg.

Maxime Mahe, Researcher working on the Enteric Nervous System in Gut and Brain Disorders, Nantes.

Laurent Poulain, Researcher at the François Baclesse Centre, Biology and Innovative Therapies of Locally Aggressive Cancers.

Xavier Gidrol, Director Biomics Laboratory & "Large Scale Biology Unit (CEA/Inserm/UGA)", CEA Grenoble.

Vincent Flacher, Research scientist at the Institute of Molecular and Cell Biology, Strasbourg.

Corinne Sébastiani, Deputy Director, INSERM Health Technologies Institute, Paris.

Minimum Information about Organoids and their Use for Researchers (MIAOU)

Second working session – (Online) 29 March 2022

LIST OF PARTICIPANTS

Ioana Andreescu, HYBRIDA WP5 Project Manager, INSERM, Paris.

Celine Cougoule, Researcher at the Institute of Pharmacology and Structural Biology in Toulouse.

Anne Dubart-Kupperschmitt, Director of Research, Pathophysiology and Therapeutics of Liver Diseases, INSERM, Paris.

Jean-Luc Galzi, Director of the Research Institute of the Strasbourg Biotechnology School,.

Jacques Haiech, Honorary Professor of Biotechnology at the University of Strasbourg.

Maxime Mahe, Researcher working on the Enteric Nervous System In Gut And Brain Disorders, Nantes.

Laurent Poulain, Researcher at the François Baclesse Centre, Biology and Innovative Therapies of Locally Aggressive Cancers.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

Xavier Gidrol, Director Biomics Laboratory & "Large Scale Biology Unit (CEA/Inserm/UGA)", CEA Grenoble.

Vincent Flacher, Research scientist at the Institute of Molecular and Cell Biology, Strasbourg.

Corinne Sébastiani, Deputy Director, INSERM Health Technologies Institute, Paris.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

11.7 Annex 7: MIAOU Full questionnaire

Section 1: IDENTIFICATION OF THE PROJECT

This section aims to clearly identify the project, including its applications and their limits. Particular attention should be paid to the use of wording that might induce fear or unrealistic promises.

PROJECT TITLE

…..

ACRONYM OF THE PROJECT (if any)

….

TYPE OF ORGANOID

Organoid for basic research	Yes/No
Factoroid (organoid for bioproduction)	Yes/No
Organoid for preclinical research	Yes/No
Organoid for clinical use	Yes/No
Others	Yes/No
If Yes, Specify	

NAME OF THE ORGANOID (avoid misnaming such as ‘minigut’, ‘minibrain’, ‘synthetic brain’, ‘synthetic embryos’; on the contrary, follow consensus nomenclature when it exists)

…

PURPOSE OF THE PROJECT (describe the aim of the project, including appropriate regulatory documents)

….

Section 2: SOURCE MATERIAL

Critical elements in this section are: 1) stem cell metadata based on the ATCC model (batch, structural, morphological and functional data, maintenance and preservation protocol), 2) declaration of collection (declaration or authorisation of activities relative to the conservation and preparation for scientific purposes of human body elements), mandatory for human samples, 3) monitoring of possible drifts of starting materials, 4) regulatory and medical ethics documents, if any (restrictions on use depending on donor consent).

Does your research involve human material?		YES/NO
If yes	Is the material obtained from volunteers?	YES/NO
	Has informed consent been obtained?	YES/NO
	Provide details of the informed consent:	
	Is the volunteer a patient?	YES/NO
	Is the genetic identity at arrival known?	YES/NO
	If starting material is obtained from a biopsy, are descriptors known (gender, age, anatomical regions, diagnosis, viral status, etc.)?	YES/NO

This project has received funding from the European Union’s HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

	Please list items.	
	Does the sponsor of the research hold clinical data on the patient?	YES/NO
	Is the laboratory authorised to prepare and conserve human body elements for scientific purposes?	
	Give details and references of the collection declaration.	
Is the starting material a cell line?		YES/NO
	Is the genetic identity at arrival known?	YES/NO
	Please specify (DNA sequence, SNPs, PCR, STR, CGH array, etc.)	
	Is there genetic quality control (karyotype, STR, PCR, etc.)?	YES/NO
	Please describe.	
If yes	Is the cell line functionally validated (differentiation test for pluripotency of IPSCs, permeability tests for epithelial cells, etc.)?	YES/NO
	Please describe.	
	Is the cell identity validated after X passages?	YES/NO
	Specify X.	
	Are cell type markers identified (example: marker name, detection method, target value)?	YES/NO
	Please list.	
	Is the number of passages at arrival known.	YES/NO
	Is the number of possible, or required, passages before the genesis of organoids defined?	YES/NO
	Are the storage conditions known?	YES/NO
	Please describe preservation protocol (culture, freezing, thawing protocol, storage modalities)	
	Does the material contain mutations (genetic disease)?	YES/NO
	Is the sanitary status known?	YES/NO
	Please give details of tests (mycoplasma, bacteriological, fungal, etc.).	
Is the starting material primary cells from patients?		YES/NO
	Is the genetic identity at arrival known?	YES/NO
	Please specify (DNA sequence, SNPs, PCR, STR, CGH array, etc.)	
	Is there genetic quality control (karyotype, STR, PCR, etc.)?	YES/NO
	Please describe.	
	Is the cell line functionally validated (differentiation test for pluripotency of IPSCs, permeability tests for epithelial cells, etc.).	YES/NO
If yes	Please describe.	
	Is the cell identity validated after X passages?	YES/NO

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

	Specify X.	
	Are cell type markers identified (example: marker name, detection method, target value)?	YES/NO
	Please list.	
	Is the number of passages at arrival known?	YES/NO
	Is the number of possible, or required, passages before the genesis of organoids known?	YES/NO
	Are the storage conditions known?	YES/NO
	Please describe preservation protocol (culture, freezing, thawing protocol, storage modalities).	
	Does the material contain mutations (genetic disease)?	YES/NO
	Is the sanitary status known?	YES/NO
	Please give details of tests (mycoplasma, bacteriological, fungal, etc.)	
Are the cell culture conditions precisely described?		YES/NO
	Are the culture conditions well defined?	YES/NO
	Provide details of culture conditions (composition of culture media, nature, origin and quantities of supplements used - e.g. glucose, serum, antibiotics, growth factors etc.)	
If yes	Are the nature and treatment of the substrates well described?	YES/NO
	Are the seeding conditions well described?	YES/NO
	Is the frequency of media changes defined?	YES/NO
	Are O2/CO2 concentrations given?	YES/NO
	Provide details of the culture conditions	
What are the storage conditions for the lines or cells?		
	Are there cell master banks?	YES/NO
	Describe protocols and drift control.	
	Are there cell working banks?	YES/NO
	Describe protocols and drift control.	
	Are storage conditions indicated?	YES/NO
	Describe the freezing and thawing protocol.	
	Are the storage modalities given?	YES/NO
	Please specify.	

Section 3: MANUFACTURING OF THE ORGANOID

Critical elements in this section are: 1) differentiation protocol and organoid generation (table of differentiation factors, timelines, culture protocols, purification protocols, and if necessary, maintenance and preservation protocols) 2) design and development of master organoid bank and working organoid bank, 3) monitoring of possible drifts of organoids (genetic, protein post-translational modifications, metabolism, other biomarkers)

Does the project include 2D differentiation?		YES/NO
	Provide details on culture media, nature, origin, supplements used (e.g. growth factors glucose, serum, antibiotics, CO2/O2 concentrations, etc.)	

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

If yes	Describe the sequence and duration of differentiation treatments. Are the culture substrates treated? Describe the seeding conditions and frequency of media changes Is there quality control for the differentiation process? Provide details (e.g. morphology, material homogeneity, max and min confluence, proliferation, functional test, monitoring of markers, possible sorting, mortality rate).	YES/NO YES/NO
Does the project include the generation of (3D) organoids? **General considerations**		YES/NO
If yes	Provide details on culture media, nature, origin, supplements used (e.g. growth factors glucose, serum, antibiotics, CO2/O2 concentrations, etc.). Describe the sequence and duration of differentiation treatments Are the culture substrates treated? Describe the seeding conditions and frequency of media changes. Is there quality control for the differentiation process? Provide details (e.g. morphology, material homogeneity, max and min confluence, proliferation, functional test, monitoring of markers, possible sorting, mortality rate).	YES/NO YES/NO
Does organoid generation make use of matrices?		YES/NO
If yes	Describe the nature of the matrix (matrigel, hydrogels, hyaluronic acid, human decellularized matrix, etc.) Give the matrix concentration. Give details of the preparation method (temperature, polymerization time, drop or layer structure, etc.). Give the seeding density per matrix volume unit. Specify the volume and number of drops of matrix per unit area in the culture medium. Specify the amount of medium depending on the size of the well. Describe the matrix dissociation method for organoid recovery. Describe the organoid dissociation method used for their expansion.	
Does the culture take place on a solid 3D substrate (e.g.: mineral substrate for bones, substrate for liquid-gas interfaces)?		YES/NO
	Describe the preparation method for the 3D solid substrate (composition of the medium to be freeze-dried, freeze-drying conditions). Give details on the seeding method.	

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

If yes	List biocompatible materials used (PDMS, COC, Silicon, etc.): Indicate chip design (provide a map). Provide the physical characteristics of the chip.	
Does the organoid grow in suspension (self-organisation)?		YES/NO
If yes	Specify the type of container. Describe the nature and protocol for agitation. Indicate the nature and concentrations of matrices.	
Does the culture include multiple cell types?		YES/NO
If yes	Describe the sequence for co-culturing and adaptation of the co-culture media. Indicate the proportions of cell types.	

Section 4: ORGANOID CHARACTERISATION

Detailed characterisation is project-dependent and should be assured in line with the proposed use of the organoid (research, bioproduction, preclinical and clinical uses); however, some standards may emerge (omics for structural characterisation, imaging for morphology and specific functional readouts, depending of the expected use of the organoid):

Has a morphological/structural characterisation been performed?		YES/NO
If yes	Describe the appearance, size, shape [circularity, tubularity, regularity of contours (budding), etc.]. Evaluate opacity/ refringency. Quantify intra and inter-organoid homogeneity. Give details of expected morphological, architectural and ultrastructural features, and organisation of cell types (identity, proportions, distribution).	
Has a molecular characterisation been performed?		YES/NO
If yes	Provide elements of genomics, transcriptomics, metabolomics, proteomics. Indicate expected specific molecular markers, epigenetic characteristics.	
Has a functional characterisation been performed?		YES/NO
If yes	What are the qualitative and (if possible) quantitative functional characteristics? If treatments are applied, detail the treatment protocol, response to treatments (pharmacological, chemical, physical, hormonal, etc.), and evaluation (quantitative or qualitative).	
Are traceability and organoid drift evaluated?		YES/NO
If yes	Describe how the traceability of components is evaluated (batches, suppliers etc., environments, complements). Indicate criteria for the traceability of conditioned media (drift of cells used for conditioning, control of lines, such as those at the origin of the organoid), control of at least one of the growth factors).	

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

	Describe the qualitative drift criteria (morphological, structural, functional, molecular, etc.) specific to each organoid. Specify indices if applicable. How is robustness evaluated (same starting cells, same organoid)? Specify indices if applicable.	

Section 5: USE OF ORGANOIDS

The critical element in this section is the robustness of the preparation and characterisation of the organoid. Anticipate the future use of organoids, from basic to the development of innovative applications (for instance the use of good laboratory practices will facilitate the transition from basic to preclinical research).

Are the organoids designed for basic research?		YES/NO
If yes	Is compliance with GLP (good laboratory practice) required for organoid production? Give details, if applicable.	YES/NO
Are the organoids designed for bioproduction?		YES/NO
If yes	Is compliance with GLP (good laboratory practice) required for organoid production? Give details, if applicable.	
Are the organoids designed for preclinical research (pharmacology, toxicology, etc.)?		YES/NO
If yes	Indicate the functional similarity criteria between the organoid and the mimicked organ (battery of controls to be performed, with target values).	
	Is the organoid to be used for the preclinical development of a drug candidate (IND file)?	YES/NO
	Indicate the number of usable passages.	
	Is the organoid used to define predictive signatures of responses (companion test)?	YES/NO
	Indicate the number of usable passages.	
	Is the organoid to be used to validate a care protocol (specific patient) on a cohort: choice of a therapy.	YES/NO
	Indicate the number of usable passages.	
Are the organoids designed for clinical research *(personalized, predictive and regenerative medicine, transplantation, etc.)*?		YES/NO/N.A.
If yes	Is the organoid to be used for: a care protocol? For example, tumoroid that will be used to test the efficacy of a chemotherapy, organoids used to optimize personalised medicine to orient a therapeutic choice.	YES/NO
	Specify the process for GMP certification, the total traceability of components, the qualification of components.	
	Give criteria for the similarity between the organoid and the biopsy (objective elements to support the plausibility of using an organoid for the choice of a therapy).	

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

	Is the organoid to be used in regenerative medicine, as already done for cell and tissue therapies? Specify the process for GMP certification, the total traceability of components, the qualification of components. Specify functionality and safety criteria (derivation of biological material and evaluation of the risk of cancer).	YES/NO
Are the organoids to be used for other purposes?		YES/NO
If yes	Specify other uses of organoids	

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

11.8 Annex 8: EChOES full questionnaire

Section 1: IDENTIFICATION OF THE PROJECT

This section aims to clearly identify the project, including its applications and their limits. Particular attention should be paid to the use of wording that might induce fear or unrealistic promises.

PROJECT TITLE

…..

ACRONYM OF THE PROJECT (if any)

….

TYPE OF ORGANOID

Organoid for basic research	Yes/No
Factoroid (organoid for bioproduction)	Yes/No
Organoid for preclinical research	Yes/No
Organoid for clinical use	Yes/No
Others (If Yes, Specify)	Yes/No

NAME OF THE ORGANOID (avoid misnaming such as 'minigut', 'minibrain', 'synthetic brain', 'synthetic embryos'; on the contrary, follow consensus nomenclature when it exists)

…

PURPOSE OF THE PROJECT (describe the aim of the project, including appropriate regulatory documents)

….

Section 2: SOURCE MATERIAL

Critical elements in this section are: 1) stem cell metadata based on the ATCC model (batch, structural, morphological and functional data, maintenance and preservation protocol), 2) declaration of collection (declaration or authorisation of activities relative to the conservation and preparation for scientific purposes of human body elements), mandatory for human samples, 3) monitoring of possible drifts of starting materials, 4) regulatory and medical ethics documents, if any (restrictions on use depending on donor consent).

Does your research involve human material?		YES/NO
	Is the material obtained from volunteers?	YES/NO
	Has informed consent been obtained?	YES/NO
	Provide details of the informed consent:	
	Is the volunteer a patient?	YES/NO
	Is the genetic identity at arrival known?	YES/NO

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

If yes	If starting material is obtained from a biopsy, are descriptors known (gender, age, anatomical regions, diagnosis, viral status, etc.)?	YES/NO
	Please list items.	
	Does the sponsor of the research hold clinical data on the patient?	YES/NO
	Is the laboratory authorised to prepare and conserve human body elements for scientific purposes?	
	Give details and references of the collection declaration.	
Is the starting material a cell line?		YES/NO
	Is the genetic identity at arrival known?	YES/NO
	Please specify (DNA sequence, SNPs, PCR, STR, CGH array, etc.)	
	Is there genetic quality control (karyotype, STR, PCR, etc.)?	YES/NO
	Please describe.	
If yes	Is the cell line functionally validated (differentiation test for pluripotency of IPSCs, permeability tests for epithelial cells, etc.)?	YES/NO
	Please describe.	
	Is the cell identity validated after X passages?	YES/NO
	Specify X.	
	Are cell type markers identified (example: marker name, detection method, target value)?	YES/NO
	Please list.	
	Is the number of passages at arrival known.	YES/NO
	Is the number of possible, or required, passages before the genesis of organoids defined?	YES/NO
	Are the storage conditions known?	YES/NO
	Please describe preservation protocol (culture, freezing, thawing protocol, storage modalities)	
	Does the material contain mutations (genetic disease)?	YES/NO
	Is the sanitary status known?	YES/NO
	Please give details of tests (mycoplasma, bacteriological, fungal, etc.).	
Is the starting material primary cells from patients?		YES/NO
	Is the genetic identity at arrival known?	YES/NO
	Please specify (DNA sequence, SNPs, PCR, STR, CGH array, etc.)	
	Is there genetic quality control (karyotype, STR, PCR, etc.)?	YES/NO
	Please describe.	

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

	Is the cell line functionally validated (differentiation test for pluripotency of IPSCs, permeability tests for epithelial cells, etc.).	YES/NO
If yes	Please describe.	
	Is the cell identity validated after X passages?	YES/NO
	Specify X.	
	Are cell type markers identified (example: marker name, detection method, target value)?	YES/NO
	Please list.	
	Is the number of passages at arrival known?	YES/NO
	Is the number of possible, or required, passages before the genesis of organoids known?	YES/NO
	Are the storage conditions known?	YES/NO
	Please describe preservation protocol (culture, freezing, thawing protocol, storage modalities).	
	Does the material contain mutations (genetic disease)?	YES/NO
	Is the sanitary status known?	YES/NO
	Please give details of tests (mycoplasma, bacteriological, fungal, etc.)	
Are the cell culture conditions precisely described?		YES/NO
	Are the culture conditions well defined?	YES/NO
	Provide details of culture conditions (composition of culture media, nature, origin and quantities of supplements used - e.g. glucose, serum, antibiotics, growth factors etc.)	
If yes	Are the nature and treatment of the substrates well described?	YES/NO
	Are the seeding conditions well described?	YES/NO
	Is the frequency of media changes defined?	YES/NO
	Are O2/CO2 concentrations given?	YES/NO
	Provide details of the culture conditions	
What are the storage conditions for the lines or cells?		
	Are there cell master banks?	YES/NO
	Describe protocols and drift control.	
	Are there cell working banks?	YES/NO
	Describe protocols and drift control.	
	Are storage conditions indicated?	YES/NO
	Describe the freezing and thawing protocol.	
	Are the storage modalities given?	YES/NO
	Please specify.	

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

Section 3: MANUFACTURING OF THE ORGANOID

Critical elements in this section are: 1) differentiation protocol and organoid generation (table of differentiation factors, timelines, culture protocols, purification protocols, and if necessary, maintenance and preservation protocols) 2) design and development of master organoid bank and working organoid bank, 3) monitoring of possible drifts of organoids (genetic, protein post-translational modifications, metabolism, other biomarkers)

Does the project include 2D differentiation?		YES/NO
If yes	Provide details on culture media, nature, origin, supplements used (e.g. growth factors glucose, serum, antibiotics, CO2/O2 concentrations, etc.) Describe the sequence and duration of differentiation treatments.	
	Are the culture substrates treated?	YES/NO
	Describe the seeding conditions and frequency of media changes	
	Is there quality control for the differentiation process?	YES/NO
	Provide details (e.g. morphology, material homogeneity, max and min confluence, proliferation, functional test, monitoring of markers, possible sorting, mortality rate).	
Does the project include the generation of (3D) organoids?		YES/NO
General considerations		
If yes	Provide details on culture media, nature, origin, supplements used (e.g. growth factors glucose, serum, antibiotics, CO2/O2 concentrations, etc.). Describe the sequence and duration of differentiation treatments	
	Are the culture substrates treated?	YES/NO
	Describe the seeding conditions and frequency of media changes.	
	Is there quality control for the differentiation process?	YES/NO
	Provide details (e.g. morphology, material homogeneity, max and min confluence, proliferation, functional test, monitoring of markers, possible sorting, mortality rate).	
Does organoid generation make use of matrices?		YES/NO
If yes	Describe the nature of the matrix (matrigel, hydrogels, hyaluronic acid, human decellularized matrix, etc.) Give the matrix concentration. Give details of the preparation method (temperature, polymerization time, drop or layer structure, etc.). Give the seeding density per matrix volume unit. Specify the volume and number of drops of matrix per unit area in the culture medium.	

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

	Specify the amount of medium depending on the size of the well. Describe the matrix dissociation method for organoid recovery. Describe the organoid dissociation method used for their expansion.	
Does the culture take place on a solid 3D substrate (e.g.: mineral substrate for bones, substrate for liquid-gas interfaces)?		YES/NO
If yes	Describe the preparation method for the 3D solid substrate (composition of the medium to be freeze-dried, freeze-drying conditions). Give details on the seeding method. List biocompatible materials used (PDMS, COC, Silicon, etc.): Indicate chip design (provide a map). Provide the physical characteristics of the chip.	
Does the organoid grow in suspension (self-organisation)?		YES/NO
If yes	Specify the type of container. Describe the nature and protocol for agitation. Indicate the nature and concentrations of matrices.	
Does the culture include multiple cell types?		YES/NO
If yes	Describe the sequence for co-culturing and adaptation of the co-culture media. Indicate the proportions of cell types.	

Section 4: ORGANOID CHARACTERISATION

Detailed characterisation is project-dependent and should be assured in line with the proposed use of the organoid (research, bioproduction, preclinical and clinical uses); however, some standards may emerge (omics for structural characterisation, imaging for morphology and specific functional readouts, depending of the expected use of the organoid):

Has a morphological/structural characterisation been performed?		YES/NO
If yes	Describe the appearance, size, shape [circularity, tubularity, regularity of contours (budding), etc.]. Evaluate opacity/ refringency. Quantify intra and inter-organoid homogeneity. Give details of expected morphological, architectural and ultrastructural features, and organisation of cell types (identity, proportions, distribution).	
Has a molecular characterisation been performed?		YES/NO
If yes	Provide elements of genomics, transcriptomics, metabolomics, proteomics. Indicate expected specific molecular markers, epigenetic characteristics.	

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

Has a functional characterisation been performed?		YES/NO
If yes	What are the qualitative and (if possible) quantitative functional characteristics? If treatments are applied, detail the treatment protocol, response to treatments (pharmacological, chemical, physical, hormonal, etc.), and evaluation (quantitative or qualitative).	
Are traceability and organoid drift evaluated?		YES/NO
If yes	Describe how the traceability of components is evaluated (batches, suppliers etc., environments, complements). Indicate criteria for the traceability of conditioned media (drift of cells used for conditioning, control of lines, such as those at the origin of the organoid), control of at least one of the growth factors). Describe the qualitative drift criteria (morphological, structural, functional, molecular, etc.) specific to each organoid. Specify indices if applicable. How is robustness evaluated (same starting cells, same organoid)? Specify indices if applicable.	

Section 5: USE OF ORGANOIDS

The critical element in this section is the robustness of the preparation and characterisation of the organoid. Anticipate the future use of organoids, from basic to the development of innovative applications (for instance the use of good laboratory practices will facilitate the transition from basic to preclinical research).

Are the organoids designed for basic research?		YES/NO
If yes	Is compliance with GLP (good laboratory practice) required for organoid production? Give details, if applicable.	YES/NO
Are the organoids designed for bioproduction?		YES/NO
If yes	Is compliance with GLP (good laboratory practice) required for organoid production? Give details, if applicable.	
Are the organoids designed for preclinical research (pharmacology, toxicology, etc.)?		YES/NO
If yes	Indicate the functional similarity criteria between the organoid and the mimicked organ (battery of controls to be performed, with target values). Is the organoid to be used for the preclinical development of a drug candidate (IND file)? Indicate the number of usable passages. Is the organoid used to define predictive signatures of responses (companion test)? Indicate the number of usable passages. Is the organoid to be used to validate a care protocol (specific patient) on a cohort: choice of a therapy.	YES/NO YES/NO YES/NO

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

	Indicate the number of usable passages.	
Are the organoids designed for clinical research *(personalized, predictive and regenerative medicine, transplantation, etc.)*?		YES/NO/N.A.
	Is the organoid to be used for: a care protocol? For example, tumoroid that will be used to test the efficacy of a chemotherapy, organoids used to optimize personalised medicine to orient a therapeutic choice.	YES/NO
If yes	Specify the process for GMP certification, the total traceability of components, the qualification of components. Give criteria for the similarity between the organoid and the biopsy (objective elements to support the plausibility of using an organoid for the choice of a therapy).	
	Is the organoid to be used in regenerative medicine, as already done for cell and tissue therapies?	YES/NO
	Specify the process for GMP certification, the total traceability of components, the qualification of components. Specify functionality and safety criteria (derivation of biological material and evaluation of the risk of cancer).	
Are the organoids to be used for other purposes?		YES/NO
If yes	Specify other uses of organoids	

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

11.9 Annex 9: Working group for RicOCheck design

11 May 2022

Research Integrity Committee check-list

First working session – Paris, Biopark

List of participants

Ioana Andreescu, HYBRIDA WP5 Project Manager, INSERM, Paris.

Celine Cougoule, Researcher at the Institute of Pharmacology and Structural Biology in Toulouse.

Anne Dubart-Kupperschmitt, Director of Research, Pathophysiology and Therapeutics of Liver Diseases, INSERM, Paris.

Jean-Luc Galzi, Director of the Research Institute of the Strasbourg Biotechnology School, Strasbourg.

Jacques Haiech, Honorary Professor of Biotechnology at the University of Strasbourg.

Cecile Legallais, CNRS Research Director, Université Technologique de Compiègne

Maxime Mahe, Researcher on the Enteric Nervous System in Gut and Brain Disorders, Nantes.

Laurent Poulain, Researcher at the François Baclesse Centre, Biology and Innovative Therapies of Locally Aggressive Cancers.

Xavier Gidrol, Director Biomics Laboratory & "Large Scale Biology Unit (CEA/Inserm/UGA)", CEA Grenoble.

Vincent Flacher, Research scientist at the Institute of Molecular and Cell Biology, Strasbourg.

Corinne Sébastiani, Deputy Director, INSERM Health Technologies Institute, Paris.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

11.10 Annex 10: RICOCheck full questionnaire

Project title		
Global purpose of the project		Summarise
Are ethical issues raised by your expected results?	Yes/No	Explain

Section 1: HUMAN EMBRYONIC STEM CELLS (HESCS) AND HUMAN EMBRYOS (HES) (HE, DEP, EU4H AND EDF)	
Research integrity and ethics committees are particularly attentive to the existence of informed consent for the use of donor biological samples, associated personal data and the correct adequacy between consent, the proposed research programme and its applications.	
Does your research involve human embryonic stem cells (hESCs)?	Yes/No
If yes — Will they be derived directly from embryos within this project?	Yes/No
If yes — ⚠ If Yes, research may not be eligible for European funding	
If yes — Are they previously established cell lines?	Yes/no
If yes — If Yes, origin and lines of cells	
If yes — Details of licensing and control measures applied by the competent authorities of the Member States involved	
Does your research involve the use of human embryos?	Yes/No
If yes — Provide the origin of the embryos.	
If yes — Provide details of recruitment, the inclusion and exclusion criteria and of the procedure for obtaining informed consent.	
If yes — Has oral and written information been provided and informed consent obtained?	Yes/No
If yes — Provide the consent form and information sheets	
If yes — Will the research lead to the destruction of the embryos?	Yes/No
If yes — ⚠ If YES, research may not be eligible for European funding	
Does your research involve the use of human foetal tissues/cells?	Yes/No
If yes — Provide the origin of human foetal tissues/cells.	
If yes — Provide details of the procedure followed to obtain informed consent.	
If yes — Has oral and written information been provided and informed consent been obtained?	Yes/No
If yes — If Yes, provide the consent form and information sheets	
Sections 2: HUMANS	Yes/No

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

This section deals with physical, moral and social well-being and with notions of respect and honesty. Research integrity and ethics committees will be particularly attentive to the existence of informed consent and to the correct adequacy between consent, the proposed research programme and its applications. Denomination of the organoids generated should not be misleading or give rise to undue hopes.

Does your research involve human subjects?		Yes/No
If yes	Details of recruitment, inclusion and exclusion criteria and informed consent criteria Are these subjects volunteers for medical studies? Are they healthy volunteers for medical studies? Are they patients? What disease/condition/disability do they have? What is your policy on incidental findings? Are they vulnerable individuals or groups? Provide details on the type of vulnerability Are they persons unable to give informed consent? Are they children/minors? Details of the procedure followed to obtain approval from the guardian/legal representative and the agreement of children or other minors What steps will you take to ensure that participants are not subjected to any form of coercion? For all "Yes" responses in this section, provide copies of ethics approval Provide the informed consent form and information sheets.	
Does the research involve physical interventions on the study participants?		Yes/No
If yes	Does it involve invasive techniques? Does it involve the collection of biological samples? Please describe the risk assessment made for each technique and overall.	
	Does it involve the collection of biological samples?	Yes/No
	What type of samples will be collected? What are the procedures followed for the collection of samples? Please add copies of the ethics approval.	

Section 3: HUMAN CELLS/TISSUES/ORGANOIDS	Yes/No

This section deals with the following values: transparency, honesty and responsibility. Human body elements should be traceable and used in accordance with the conditions of the research, according to the donor's informed consent and responses

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

to the potential future use questionnaire. Denomination of the organoids generated should not be misleading or give rise to undue hopes.

Does the research involve human cells or tissues or organoids (other than from human embryos/foetuses, i.e. the section above)?		Yes/No
If yes	Provide details of the cells, tissue type Provide copies of the relevant ethics approval Provide accreditation/designation/authorisation/licensing for the use of cells or tissues (if required).	
Are they available commercially?		Yes/No
If yes	Details of the supplier. Copies of the import licence, if relevant	
Are they obtained within this project?		Yes/No
If yes	Details on the source of material and procedure for collection. Details on the duration of storage and on what will happen to the material at the end of the project. Confirm that oral information has been provided and informed consent has been obtained. Provide copies of the informed consent form and information sheets	
Are they obtained from another project, laboratory, institution or biobank?		Yes/No
If yes	Country where the material is stored. Details of the legislative framework within which the material is stored. Details on the duration of storage and on what will happen to the material at the end of the project. Name and country of the laboratory/institution/biobank. Confirmation that the material is anonymised. Confirmation that permission for secondary use has been obtained during the consent process.	 Yes/No Yes/no
Provide copies of the import licence (if relevant) and of statement from the laboratory/institution/company that informed consent has been obtained.		

Section 4: PERSONAL DATA	Yes/No

This section deals with the notion of privacy-by-design, i.e. data confidentiality. The acquisition of personal data should be limited to the strict minimum, anonymised or pseudonymised and their use should be limited to the proposed research programme.

Does the research involve the collection and/or processing of personal data?		Yes/No
	Details on the technical and organisational measures implemented to safeguard the rights of research participants (protection policy).	

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

If yes	Has oral and written information been provided and informed consent been obtained? Details of the security measures in place to prevent unauthorised access to personal data. How is the processed data relevant and limited to the purposes of the project (data minimisation principle)? Details of methods used to ensure anonymisation/pseudonymisation. Justification if research data will not be anonymised/pseudonymised (if relevant). Details on data transfers and countries to which they are transferred.	Yes/No

Does the research involve the further processing of previously collected personal data (secondary use)?	Yes/No

If yes	Details on the database used or source of data. Details on data processing operations. How will the rights of the participants be safeguarded? Please explain. How is the processed data relevant and limited to the purposes of the project (data minimisation principle)? Justification if the research data will not be anonymised/pseudonymised (if relevant).

Compliance: upload relevant documents, if any.

Section 5: ANIMALS/CHIMERAS	Yes/No

This section relates to the 3R rule (reduce, replace, refine use of animals in experimentation). The use of animals instead of *in vitro* or *ex vivo* model systems requires justification

Does the research involve animals?	Yes/No

If yes	Details of the species and rationale for their use, number of animals, nature of the experiments, procedures and techniques. Justification of animal use (including the kind of animals) and why alternatives cannot be used.

Are the animal vertebrates?	Yes/No
Are the animals non-human primates (NHP)?	Yes/No

If yes	Why are NHPs the only research subjects suitable to achieve the scientific objectives of the project? What is the purpose of animal testing? Please give details Where do the animals come from?

Are they genetically modified or cloned animals?	Yes/No

If yes	Details of the phenotype and any inherent suffering anticipated.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

	What scientific justification is there for producing such animals? What measures will be implemented to minimise suffering during breeding and maintenance of colonies?

Do the animals belong to endangered species?	Yes/No
If yes	Why is there no alternative to using this species? What is the purpose of the research?
Does the research involve chimeras?	Yes/No
Are these human-animal chimeras?	Yes/No
If yes	What scientific justifications are there for producing such chimeric animals?
Are the recipients non-human primates?	Yes/No
If yes	Why are NHPs the only research subjects suitable to achieve the scientific objectives?
Are the recipients genetically modified or cloned animals?	Yes/No
If yes	What scientific justifications are there for producing such animals?
Are the recipients endangered species?	Yes/No

Compliance: brief description of compliance procedures, upload relevant documents if any.

Section 6: NON-EU COUNTRIES	Yes/No

Regulations differ significantly among countries. The study sponsor should ensure that the export of material to other countries is permitted, and that any exports comply with current national regulations (request for authorisation from the competent authorities).

Are non-EU countries involved?	Yes/No
Specify which countries	
Do the research-related activities undertaken in these countries raise potential ethical issues?	Yes/No
If Yes	Risk-benefit analysis? What activities are carried out in non-EU countries? Provide copies of ethics approval and other authorisations or notifications (if relevant) Provide confirmation that the activity could have been legally carried out in an EU country (for instance, an opinion from an appropriate ethics structure in an EU country)
Is it planned to use local resources (animals, tissue samples, genetic material, endangered fauna)?	Yes/No
If Yes	What type of local resources will be used and how exactly? For human resources: copies of ethics approvals. For animals, plants, microorganisms and associated traditional knowledge: documentation demonstrating compliance with the UN convention on biological diversity (e.g. access permit and any benefit sharing agreements)

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

Do you plan to import any material -including personal data- from non-EU countries?		Yes/No
If Yes	What type of materials will you import? Copies of import licences	
Do you plan to transfer any material -including personal data - from EU-countries to non-EU countries?		Yes/No
If Yes	What type of materials will you transfer, and to which country? Copies of export/transfer licenses	
If the research involves low- or middle-income countries, are any benefit-sharing actions planned?		Yes/No
If Yes	Please provide details of any benefit-sharing measures Please give details of responsiveness to local research needs. Please give details of procedures in place to facilitate effective capacity building.	
Could the situation in the country put the individuals taking part in the research at risk?		Yes/No
If Yes	Please detail any safety measures you intend to put in place, including training for staff and insurance cover.	

Section 7: POTENTIAL MISUSE OF RESULTS

Attention must be paid to any secondary uses of donor samples. The sponsor of the research should ensure that all potential uses are included in the informed consent and allowed for in the donor "TRUSTED list" (restrictions on use), or should obtain a new informed consent.
This section also deals with values such as benevolence and non-malevolence.
Potential military uses are also intended to be considered here.

Does the research involve dual use items in the sense of regulation 428/2009, or other items for which au authorisation is required?		Yes/No
If yes	What goods and information used and produced during the research projects will require an export licence? How exactly will you ensure compliance? How exactly will you avoid negative implications?	
Does the research have potential for the misuse of research results?		Yes/No
	Risk assessment Details of applicable legal requirements Details of measures implemented to prevent misuse	
Could the research raise concerns regarding the exclusive focus on civilian applications?		Yes/No
	Explain the exclusive civilian focus of the research	

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

If Yes	Justify the inclusion of military partners or military technologies (i.e. explain how they relate to civilian applications, e.g. in the context of law enforcement activities).

Compliance: provide copies of authorisations, export licences, security clearances and ethics approvals (if applicable).

Section 8: OTHER ETHICS ISSUES

Please explain out any further ethics concerns that might arise from the proposed project

Are there any other ethics issues that should be taken into consideration? Please specify.	Yes/No

Compliance: upload any relevant documents.

I confirm that I have taken into account all ethics issues described above, and that if any ethics issues apply, I will complete the ethics self-assessment and attach the required documents.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

11.11 Annex 11: Winsconsin-Madison supporting organisation, WiCell, policy for use of cell lines

WiCell has changed its policy regarding Annual Certifications. This change is consistent with our obligations to donors. WiCell will no longer require signatures and the submission of an Annual Certification form. WiCell will be issuing annual reminders regarding the restricted uses of cell lines.

The reminder is shown here:
This notice serves as a reminder that in accordance with the terms of the above-referenced Simple Letter Agreement, and the Memorandum of Understanding between WiCell and your Institution, you have agreed to abide by such terms and conditions regarding the receipt and use of the Wisconsin Materials. By signing the SLA agreement you certified that the research you are engaged in does not and will not include any of following:

(a) mixing of Wisconsin Materials with an intact embryo, either human or non-human;
(b) implanting Wisconsin Materials or products of the Wisconsin Materials in a uterus;
(c) attempting to make whole embryos with Wisconsin Materials by any method; or
(d) using Wisconsin Materials for any therapeutic or commercial purpose, including the performance of services (including diagnostic services) for consideration or the production or manufacture of products for sale or distribution.

Further, in response to numerous enquiries that WiCell received this year about blastoids and other embryo models, the Wisconsin Alumni Research Foundation ("WARF") has formulated the following clarification and guidance regarding the prohibition on attempting to make whole embryos with Wisconsin Materials by any method, as listed above.

- WARF does not believe that using the Wisconsin Materials for blastoid research, given the current developmental potential of blastoids, would be contrary to the prohibition listed above and will therefore allow the Wisconsin Materials to be used for current blastoid research.
- WARF recognizes, however, that the situation may change in the future as research continues and the developmental potential of blastoids becomes more embryo-like. WARF provides notice that stem cell-derived embryo models, including blastoids, may progress to meet the definition of a whole embryo in some of the informed consents (that is, would develop into a fetus if returned to the uterus). If used to make a whole embryo, WARF specifically reserves the right to demand the immediate return or destruction of any such Wisconsin Materials or Modifications to Wisconsin Materials and the retraction of any related articles.
  Please let me know if you are no longer working with WiCell's human embryonic stem

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

cells.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

11.12 Annex 12: Organ-on-Chip versus Organoids-on-Chip

Xavier Gidrol (Xavier.Gigrol@cea.fr) Directeur de Biomics
Directeur de l'Unité 1038 (CEA - Inserm - UGA) « Biologie à Grande Echelle » CEA-Grenoble, France

Organoid technology and organ-on-chip engineering have emerged as two historically distinct research fields, but with the same goal; to better model the tissue under study. We have recently observed a trend towards the fusion of these two fields, which might address the limitations of each approach and, in a synergistic way, may pave the way towards the generation of tissues that are closer and closer to the physiological reality of an organ. These organ substitutes could therefore better respond to the expectations of pharmacology and participative, predictive, personalised and precision medicine.

Organoids have been developed by specialists in human pluripotent stem cells (hPSCs) or adult stem cells (AdSCs) residing in the tissues of virtually all human organs. These three-dimensional (3D) cell models offer a better representation of gene and protein expression, metabolic functions and physiological and functional data than two-dimensional (2D) cell culture models. Patient-derived stem cells cultured as 3D tissue models of human diseases could, by uncovering the mechanisms responsible for a disease, lead to more effective drug discovery. However, obtaining 3D tissues that faithfully reproduce all the properties of an organ derived from stem cells remains a major challenge.

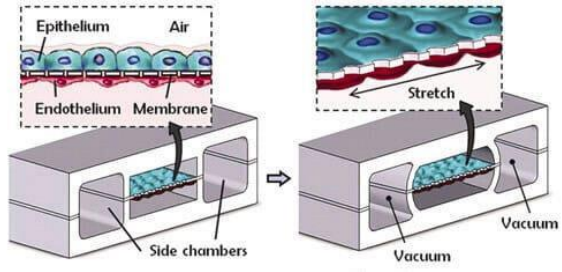
The main characteristic of organoids is the self-organisation of differentiating cells. This has enabled the development of various epithelial and epithelial-mesenchymal organoids from hPSCs such as optic cup, endocrine tissue, intestine, liver, pancreas, brain, kidney, lung, retina, prostate, breast, among others (Lancaster and Knoblich, 2014). An organoid typically has a complex multicellular architecture resembling that of the original tissue in miniature. Hence the name, since the suffix "-oid" means "which resembles ....". However, most organoids generated *in vitro* from hPSCs possess foetal and relatively immature phenotypes, although a series of studies have shown that after transplantation, these organoids reach some level of maturity (Takebe et al., 2013). Organoids derived from hAdSCs have the advantage of establishing and maintaining a more mature phenotype compared to hPSC-derived organoids and have been used to generate models more representative of the physiological reality of the digestive tract, lung, mammary gland or prostate, to name but a few.

Although the physiological relevance of organoids as human models is now accepted, their practical use, especially in pharmacology or regenerative medicine, remains limited by low yield on the one hand, and by the very low reproducibility of the 3D structures obtained on the other.

Organs-on-a-chip were developed by engineers, specialists in microfluidics, working on what was then called Lab-on-Chip. Microfluidic engineering allows the precise control of microchannel geometry, flow volumes and conditions, shear forces, nutrient supply, and local mechanical and electrical properties. Initially, these devices were mainly used to study molecular activities, such as the measurement of metabolites or enzymatic activities in very small volumes, but in the mid-2000s, researchers started to add cells to them. These are mainly combinations of pre-differentiated cells, often cell lines, in certain ratios in order to emulate the composition of native tissue. The first publication to use the term organ-on-a-chip was by Don Ingber's team in Science in 2010 (Huh et al. 2010 and Figure 1), which presented a lung-

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.

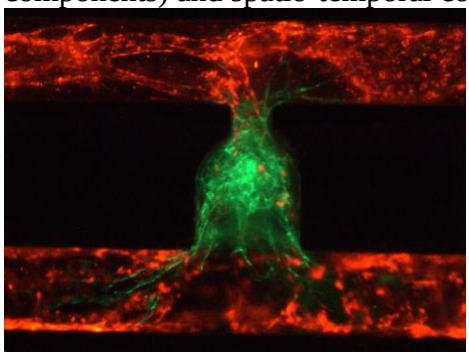
on-a-chip device using lithography and microfluidics to recreate an air/liquid interface similar to that found in lung alveoli.



However, while organs-on-chip allow for a reduction in the variability of the resulting biological objects, this comes at the expense of fidelity to the tissue architecture. The cellular composition in organs-on-chip is oversimplified, as most contain only a few cell types from the main tissue and the cell lines often have limited physiological relevance. Organs-on-a-chip often have very limited structural similarity to the organ. They also do not allow fully mature adult phenotypes.

Organoids-on-a-chip. It seems obvious that neither field alone is able to reproduce exactly the organogenesis and physiology of an organ. It is indeed necessary to increase the spatio-temporal control of 3D tissue generation in order to decrease the excessive variability of shapes and sizes of the organoids created by self-organisation. At the same time, it is essential to control the architecture of the organ thanks to the organ-on-chip, by placing more complex objects that are more representative of the reality of the organ.

To obtain 3D tissues that are increasingly faithful to the physiological reality of an organ, it is essential to introduce more complex cellular interactions (vascularisation, innervation, immune components) and spatio-temporal control (4D) in better defined environments.



Convergence of the two approaches, through the generation of organoids-on-a-chip (Quintard et al. 2021, Figure 2 from my lab, vascularized pancreatic organoids in green, microfluidic channels lined with endothelial cells in red), will enable the improved modelling and screening of complex human pathologies, mimic the dynamic nature of a self-developing system, and thus facilitate the discovery of new treatments and access to 4P medicine.

This project has received funding from the European Union's HORIZON 2020 Research and Innovation programme under Grant Agreement No 101006012.