HYBRIDA

D1.4: Typology for artificial biological entities

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Embedding a comprehensive ethical dimension to organoid-based research and relating technologies

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ABSTRACT:	WP1 is dedicated to the exploration of conceptual uncertainties surrounding organoid research and related technologies. This document proposes a typology of the main concepts used in organoid research, in a way that echoes the specific ethical status of each entity. The purpose of the typology is to provide the HYBRIDA project with a shared vocabulary in order to discuss ethical and legislative issues to be raised by organoid research.

Keyword List:	Stem cells; organoids; embryo models; chimeras; hybrids; biotechnology; bioethics.

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EXECUTIVE SUMMARY

WP1 is dedicated to the exploration of conceptual uncertainties surrounding organoid research and related technologies. The current document proposes a socially robust typology of the main concepts used in organoid research. The typology identifies concepts, which are briefly defined and exemplified in the text. Under the same concept, one will find entities that share common properties and that raise by consequence similar ethical issues. With organoid research and the development of organoid-related technology, we are likely to see the emergence of ethically problematic entities, or entities that raise specific, new ethical issues. For instance, the debate on stem cells was focused for a long time on stem cell procurement, especially human embryonic SC. Now, as the entities in the laboratory turn into more and more complex hybrid structures, the question of the nature (some would say moral status) of the entities developed from stem cells is at the forefront.

The current document tries to capture the main elements of the potential emergence of issues in relation to specific new entities developed in the laboratory. For instance, there are ethical issues with stem cell research in general that are common to all research with SC as biomaterial, and there are also more specific issues that appear when researchers develop complex models of embryonic development, of brain development, when they mix cells from human and animal sources, and so on. This typology is not a scientific nomenclature. While presenting the main concepts, it tries to convey the sense of uncertainty peculiar to ongoing research and evolving research objects.

Also, we discuss here distinctions between concepts only when the distinction is consistent with the delineation of major ethical issues (each distinction should make a difference from an ethical point of view). **The result is a conceptual diagram that unfolds several distinctions and points to relevant ethical issues**. There could have been more distinctions and other specific questions. We tried to build the most robust representation of the concepts used in the field. In doing so, we built on the different literature reviews conducted in the project and the many exchanges between HYBRIDA partners. Nonetheless, we could have drawn other perfectly valid typologies: **the finished product is a partial and incomplete taxonomy that indicates directions for future study**. While a professional bioethics reader might find the delineation of ethical issues too sketchy, this document is intended to pinpoint topics that are to be developed in other places in the HYBRIDA project. This work is at a crossroads in the project, building on what has been done in the first part of the project (exploration of conceptual, epistemological, and ethical issues) and providing a guide for future work.

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1 Context

1.1 Work package 1 of HYBRIDA project

The main goal of the HYBRIDA project is **to build a comprehensive ethical framework for organoid-based research and resulting technologies**. What role do we want organoids to play in the design of a future of cure and care in a democratic society? Organoid research and organoid-related technologies are examples of disruptive research and innovation that challenge our conceptual, epistemological and regulatory frameworks by producing three different kinds of uncertainty: (1) conceptual and ontological uncertainty (how should one conceive these entities called organoids? What are they? How do we know their characteristics?); (2) epistemological and methodological uncertainty (how do we address forms of uncertainty that cannot be evaluated using statistical methods, i.e., risk assessment?); (3) regulatory uncertainty, emerging because parts of regulatory frameworks concerning the rights and duties of persons have been merged with elements of regulation dealing with the stewardship of objects or things.

According to the plan developed in the project proposal, WP1 is dedicated to the identification and discussion of **different forms of conceptual uncertainty pertaining to different organoids, chimeric entities, and hybrids** created for research purposes. WP1 relied on an exploration of the ontological, moral and legal status of organoids, chimeric entities, and hybrids present in different cultures and knowledge traditions. An analysis of mythological representations of chimeras and hybrids has been conducted, with a view to these representations' impact on the conceptual and metaphorical landscape of organoid research (see D1.1: *Mythological and artistic representations of chimeras and hybrids*). The second task was the identification of different forms of conceptual uncertainty pertaining to organoids, chimeras, and hybrids (see D1.2: *Identification and discussion of conceptual uncertainties relating to organoids, chimeric entities, and hybrids*). The third task was an exploration of the use of chimeras, hybrids, and organoids as metaphors and termini technici in the scientific literature (see D1.3: *The challenging history of organoid research and its implications for ontology and ethics*).

According to the proposal, **WP1 culminates with D1.4 in the development of a "socially robust typology for artificial biological entities (organoids and biological-technical hybrids)"** that will nourish other WPs in the project and serve as a basis for the ethical discussion. The purpose of this report is to present this typology, along with contextual elements of its development, and to provide keys for its interpretation and use.

WP1 is part of the "mapping" activities of the HYBRIDA project, along with WP2 (exploration of epistemological uncertainties) and WP3 (exploration of normative, ethical, and legal uncertainties). In this sense, it is intended as a diagnostic approach, while the prognostic and potential therapeutics are to be provided by the co-construction process developed by WP4, WP5, and WP6. While WP2 and WP3 have their own approaches for this exploration and mapping phase**, WP1 draws mainly its methodology from the philosophy of science**.

Philosophy of science should not be conflated with bioethics. Philosophy of science is mainly aimed at contextualizing and discussing scientific methods and scientific concepts (How

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does knowledge progress?), while bioethics starts from normative issues (Which research should be pursued or banned?). Our assumption in this work package is that the focus of philosophy of science can assist bioethics by offering useful, foundational work on definitions, concepts, and so on. That the bioethical discussion should be grounded in a philosophical reflection adopting the style of philosophy of science is a commonsensical claim in the field of bioethics, although not always implemented.[1]

As a consequence, **the goal of WP1 is to provide project members, especially in WP4-6, with reflections on the concepts to be used in the project**: provide building blocks, elements of reflection, and ongoing support. However, an important message is also that the work of exploring and mapping conceptual uncertainties – even if turning into something like a "socially robust typology" – does not mean that all uncertainties have to be settled and resolved by WP1. **Our goal is instead to raise awareness of the existence of conceptual uncertainties and accompany the project as a resource**.

1.2 WP1 among other WPs

As a summary, the present document builds on many pieces of work conducted in the HYBRIDA project (Figure 1.1):

- **From D1.2** (*Identification and discussion of conceptual uncertainties relating to organoids, chimeric entities, and hybrids*): This deliverable of WP1 proposed a discussion of the conceptual confusion that might arise when dealing with biotechnological hybrids that challenge our main categories of thought.
- **From D1.3** (*The challenging history of organoid research and its implication for ontology and ethics*): This second deliverable of WP1 was intended to provide the project with a "sense of history" through a pioneering HPS (history and philosophy of science)-style analysis of the concept of an organoid.
- **From WP2** (Mapping of the organoid field): Task 2.1 provided the project with a precise map of the organoid research field, including a list of terms used in the scientific literature and a discussion on several technical terms. Although it has a broader scope in view, the typology draws heavily on the scientific nomenclature for obvious reasons (scientific and clinical research being the source of organoid technology and concepts and one of the main destinations for HYBRIDA's products, e.g., guidelines).
- **From WP3**: D3.1 (*Map report of normative research ethics and research integrity frameworks*) provided a broad review of ethical issues raised in the field of gene editing, cloning, ESC research, iPSC research, and organoid research. The typology draws also from this work as it aims to connect conceptual distinctions with ethical issues.
- **From WP4**: WP4 is in charge of the engagement process for involving citizens and stakeholders in the project. One of the purposes of this engagement process is "to understand not only the current status and challenges of organoid research, but also the

[1] Lewens, T. (2015). *The biological foundations of bioethics*. Oxford University Press.

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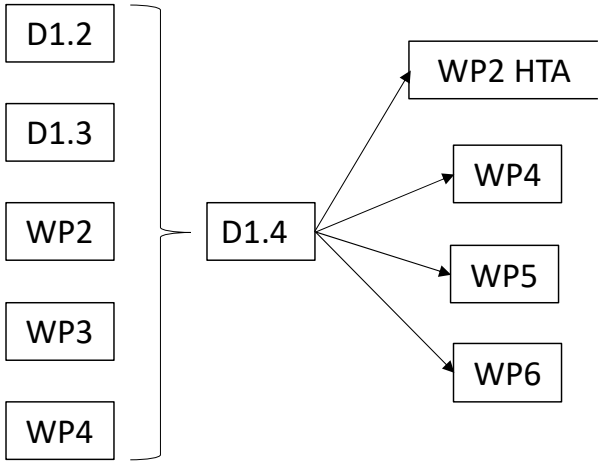
hopes, concerns, expectations and visions for the future of organoid research which are of importance for assessing new organoid technologies and of key ethical importance."[2] The first lessons from the engagement process are taken into account in order to build a "socially robust typology" (see below, methodology).

The following tasks of the HYBRIDA project can put the typology to use according to their various needs and their respective methodologies:

- **For WP2**: The typology can be a guide in the conduct of the amended health technology assessment (HTA). It can help identify where specific assessment is required and delineate problems to divide subtasks in the HTA. This holds more generally for the conduct of the ethical discussion on organoid research and related technologies in the following WPs.
- **For WP4**: The present document could also provide a basis for the discussion during stakeholder workshops, as was D1.2 for previous workshops.
- **For WP5**: WP5 is in charge of drafting the main outputs of the HYBRIDA project (e.g., guidelines for the field, code of responsible conduct). Although the overall architecture of the guidelines might not follow the presentation of the categories that is done in this document, the discussions on the scope of the HYBRIDA project and the nature of entities that we are dealing with imply regular interactions between WP1 and WP5.
- **For WP6**: WP6 is in charge of identifying relevant gaps in existing ethics and normative frameworks and exploring how those gaps can be addressed. The typology will help in defining the scope and delineating the objects of the inquiry.

Figure 1: Previous work and destination of the typology



[2] Ravn, T., Sørensen, M. P. (2021). *HYBRIDA D4.1. Protocol. Organoid-based research: engagement, co-creation and validation: HYBRIDA*. EU Commission.

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2 Conceptual uncertainties

2.1 Conceptual uncertainties in general

In our previous reports, we have pointed to two kinds of conceptual uncertainties. The first one is a series of **general conceptual uncertainties arising in the discussion of common notions such as, e.g., person, subject, nature, artefact, life**. In D1.2, we explored in detail ten axes of hybridity, based on notions borrowed mostly from philosophy or common sense, that can be applied to organoid research and related technologies: person-thing, subject-object, cell culture-organ(ism), life-mechanism, nature-artefact, science-technology, research-clinic, means-ends, actual-becoming, and human-animal. While all the notions are presented as part of a couple, it does not mean that we have to take sides (decide whether the organoid X is a subject or an object). On the contrary, **the hybridity model emphasized in the project proposal suggests that these distinctions are to be overcome**. Hybrids are entities of mixed nature, individuals in which different natural kinds converge, falling under several categories which are often thought of as mutually exclusive.[3] Several dimensions of hybridity have been emphasized in the HYBRIDA project since the project proposal. The HYBRIDA logo, mixing a computer chip and a representation of the brain as a natural organ, put the emphasis on biological/technological hybridity. A biotechnology is a typical case of a hybrid entity – both biological, alive or made from living material, AND artificial, human-made, technological.

Hybridity implies that we usually deal with conceptual categories which are poorly adapted to the ontological status of the entities we need to discuss: if it does not make sense to claim that the entity X is only an object or only a subject, then we have to pick characteristics from the one and the other, or we have to find a new category for X. Taking hybridity seriously means that these distinctions (the basic distinction of the artificial and the biological and all those unfolded in D1.2) do not help us to deal with the entities we want to deal with, because the entities of interest cannot be easily categorized with these conceptual tools. This is exactly the case with so-called chimeras: in a world in which we would not be confused by hybridity, we could delineate clearly distinct categories and make subsequent ethical distinctions, such as research with human material on the one side and research with animal material on the other, but research with chimeric models is precisely blurring the distinction. As a consequence, the typology should not retrace and discuss all these distinctions, it should instead cut across them.

Some of the concepts in use, including in workshops and public discussions such as those taking place in HYBRIDA WP4 and WP5, might suggest a narrative or an interpretative framework. **For the sake of the public discussion, bioethics needs a common language**. However, developing

[3] In this sense, hybridity is a philosophical concept, which is only remotely related to the biological concept of a hybrid. Hybridity, that is, the idea that all sorts of classical dualisms that were the base of our ontology are challenged by contemporary developments, has been a major theme of the philosophical discourse trying to capture the evolution of biotechnologies in the last decades. Haraway, D. (1991). *Simians, cyborgs, and women: the reinvention of nature*. Routledge; Latour, B. (1993). *We have never been modern*. Harvard University Press.

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such a language is a difficult task, and it has not been proven until now that it is even feasible. Language is inevitably a plastic material. As concepts evolve and acquire different meanings according to their context of use, a bioethics project such as HYBRIDA, which includes an extended discussion involving many stakeholders, can only formulate a series of caveats and raise awareness regarding the ambiguity of the concepts of interest.

2.2 Conceptual uncertainties in science

Let us turn now to the second kind of conceptual uncertainty. It might be tempting to acknowledge that general notions such as "nature" or "subject" are polysemous, but to argue nonetheless that scientific concepts, at least, do correspond to a specific definition that we can refer to in order to settle controversies. This, however, is obviously not the case, as shown by the many definitions of "organoid" that can be found in the scientific literature and the current debates in the scientific community on nomenclature issues. Scientific and technological research relies on concepts that are also metaphors or general and vague notions, possibly emotionally laden or misguiding. In other words, **there is conceptual uncertainty even in scientific language**.

This is specifically the kind of uncertainty that the focus of philosophy of science aims at investigating. While a full investigation of the history and philosophy of organoid research was not the purpose of the HYBRIDA project and its WP1, we wanted to explore what philosophy of science could tell us about the concept of an organoid and related terms. According to our analysis (presented in detail in D1.3), a standard narrative dominates the scientific and lay literature, describing the **emergence of organoids as a new biotechnology in the 2010s**. This emergence is supposed to have been followed by the continuous development of organoid research, which includes more organoid types (from gut to liver, brain, etc.) and more complex biotechnological entities (with more properties). The urge for ethical reflection and regulation would then appear as a culmination of the development of a biotechnology – once science has created new entities, these technologies offer new possibilities and it would be time to assess the moral status of these entities. History and philosophy of science suggest that the picture might be in fact more complex.

In this regard, it is interesting to focus on the use of metaphors, analogies, and vague terms in scientific research and communication, especially in biology and biotechnology. **Metaphors are cognitive tools to convey complex and uncertain ideas**. These cognitive tools can be put to use for fostering understanding about complex issues, or for communication in the scientific community and abroad, but they also play a major role in scientific understanding. Many scientific concepts are indeed rooted in metaphors. For instance, following the observation of an unknown entity, the observer will use a metaphor to label the new object and open a field of inquiry that will progressively explore the properties of the entity of interest, notwithstanding the charge of the metaphor, e.g., the "cell" referring to the concrete image of an enclosed space (the cell of a monk).

The discourse surrounding the field of new biotechnologies is especially rich in metaphors. Expressions such as "book of life," "computer code," "blueprint," etc. are omnipresent in the scientific discourse. "Gene editing" refers to the idea of the genome as a text

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that biotechnology can correct or improve at will. Genetic information is seen as a code, a meaningful sequence of material elements, and the editor is often conflated with a person that understands fully the edited text, if not even its creator.[4] It builds on a narrative that the genome is a text and should be read, or interpreted. This language is potentially misguiding as it does not convey the uncertainties of research.[5]

Another example is potentially misleading associations and images associated with terms such as "biobanking," which inappropriately imports an economic connotation in a domain of research for public health.[6] While there would have been plenty of alternative metaphors describing the practice of storing biological samples in public or private depositories, the widespread adoption of "biobanking" as a label should be a matter of concern. In this sense, **scientific metaphors may have an impact on public discourse, bioethics, and regulation**.

2.3 Organoid as metaphor and laboratory object

In this regard, the organoid metaphor is one of the numerous metaphoric and uncertain concepts emerging among other metaphors of genomic research. **Intuitively, "organoid" refers to an entity that is similar to an organ or that has the shape of an organ** (an aster-oid is a celestial body that looks like a planet, but is not a planet). However, there are many uncertainties associated with this definition: what does it mean to be similar to something? When does something begin to look like an organ and when does it stop? As a consequence, **even as a concept forged and used by biomedical researchers, the signification of "organoid" is highly context-dependent**. In this context, vagueness can also be a positive quality: the promotion and expansion of a field of biotechnological research remain possible even if we do not know what organoids exactly are. Researchers definitely need to put labels on the things they observe, and then pursue investigations. In this case, the label refers to a series of objects that putatively share something in common, but are still under investigation.

"Organoid" is arguably a popular label today. Notably, the term was already used in the second half of the twentieth century to designate cancer cell aggregates. Depending on the context, "organoid" could refer to any kind of unusual multicellular structure or malformation that manifests some similarity with an organ, such as teratomas or carcinomas that look like organs instead of looking like an embryo (grounded on the intuitive meaning of –oid as resemblance). Yet the theoretical construct developing in the 2010s is more precise. The claim that organoids are a new concept, or a new kind of entity, goes with a discourse on the novelty of this research and its potentiality. For instance, Simian and Bissell claim that they were building

[4] Fox-Keller, E. (1995). *Refiguring life: metaphors of twentieth-century biology*. Columbia University Press.
[5] O'Keefe, M., Perrault, S., Halpern, J., Ikemoto, L., Yarborough, M. (2015). "Editing" genes: a case study about how language matters in bioethics. *The American Journal of Bioethics,* 15(12), 3-10. https://doi.org/10.1080/15265161.2015.1103804
[6] For an in-depth analysis of the possible role of analogies and metaphors in research biobanking, see Solbakk, J.-H., Holm, S., Hofmann, B., eds. (2009). *The ethics of research biobanking*. Springer Verlag.

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mammary gland organoids in the 1980s, long before the current boom in organoid research.[7] Resolving priority issues does not necessarily make sense when we do not agree on a strict definition. However, it is interesting to observe how a field of research evolves or emerges, how laboratory entities have different significations in different contexts (stem cells in the 1980s are not stem cells now, and the same holds probably for Simian & Bissell's early constructions). In this sense, **the concept of an organoid is a scientific construct, not only because the entities it refers to are laboratory products, but also because the scientific discourse is built around certain assumptions concerning what organoids are and what they can perform in basic and clinical research**.

Organoids are most of the time presented as models of development. At the very least, the public debate should not conflate models, theories, and therapies. As models of development, organoids are tools. Organoids are analogies for organs, or organs' surrogates. A model is always partial, it implies a reduction in complexity and dimensionality. As a consequence, part of the debate around uncertainty originates in the very nature of a scientific model. HYBRIDA project members should reflect on the adequate or inadequate application of metaphors to model or organoids in science and its perception or visions in a given society in time.[8]

In biotechnology, metaphors play a central role in the development of an emerging scientific subject. It is also a sign of the immaturity of the field. **Metaphors contribute to shaping conceptual models for the public and the scientific community**. The public debate is fueled by terms proposed by scientists and then transferred to another context. **There is a responsibility from researchers to use adequate metaphors, as metaphors act as filters highlighting certain aspects and obscuring others**. Is regulation therefore a metaphorical practice? The use of chimera and hybrids led to the possibility to imagine organoids. It is rare for a regulatory frame to be created from scratch. This is a difficulty and a challenge for creativity. Effective regulation in science cannot rely on old metaphors only. The language framework must imply some future applications that will affect governance issues.

An illustration of the conceptual uncertainty surrounding organoid research is the coexistence of many definitions for organoids in the scientific literature. As we have shown in D1.3, there are many definitions of organoids, each insisting on some aspects of the entities of interest and each positing certain requirements. Even if we can still find some common ground, since these definitions all revolve around a set of common ideas, **there is no consensual reference point for including or excluding certain entities from the list of organoids**.

For example, models of embryo development (labelled also embryo models, SHEEF *synthetic human entities with embryo-like features*,[9] blastoids, or gastruloids depending on the stage of development that is modeled) are sometimes implicitly or explicitly assimilated to

[7] Simian, M., Bissell, M. (2017). Organoids: a historical perspective of thinking in three dimensions. *The Journal of Cell Biology*, 216(1), 31-40. https://doi.org/10.1083/jcb.201610056

[8] Solbakk, J.-H. (2022). HYBRIDA D1.1 Mythological and artistic representations of chimeras and hybrids. EU Commission.

[9] Aach, J., Lunshof, J., Lyer, E., Church, G.M. (2017). Addressing the ethical issues raised by synthetic human entities with embryo-like features. *Elife*, 6:e20674, 1-20. https://doi.org/10.7554/eLife.20674

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organoids. But are embryo models organoids? Obviously embryo models do not look like a specific organ of the body. However, one could argue that a similar procedure is used for their construction (they belong to the same family of biotechnologies). The same holds for assembloids or "multi-organ organoids" and tumoroids, that do not look like organs properly speaking.

Another example would oppose organoids as models of development, for developmental biology or (pre)clinical research on the one hand and organoids as mini-organs on the other. Current organoids are not mini-organs and even less organs for replacement (spare parts), but this is obviously part of the public discourse around organoid research, even shared by scientists themselves. How do we keep a clear distinction between models (a cell aggregate in a petri dish) and a pseudo-organ that has the possibility of transplantation in the perspective of regenerative medicine? The bioethical discourse will have to address issues raised from both perspectives, while not conflating them.

The purpose of the typology that we propose here is precisely to go beyond the scientific uncertainties of ongoing research to chart a territory for bioethical discourse that refers not only to metaphors and theoretical constructs, but to actual biotechnological entities developed in the laboratory. As Sheila Jasanoff wrote, "policy makers need ways for accommodating the partiality of scientific knowledge."[10] HYBRIDA project members, and all stakeholders of organoid research, including the public at large, are also in need of a vocabulary that is open to the many possibilities offered by new biotechnological objects.

3 Methodology

3.1 In a nutshell

According to the context described above, the HYBRIDA project cannot be content with a glossary that partially selects some elements of the current scientific debate. **The context of conceptual, epistemological, and regulatory uncertainty underlines the necessity of a consolidated nomenclature that could serve as a guide for ethical discussion**.

This nomenclature is what is referred to in the project proposal as a **"socially robust typology." Such a typology would have to take into account all the elements contributing to conceptual uncertainty** (the dynamics of ongoing research, the fact that we are dealing with living material, etc.). **The distinctions proposed in the typology should be relevant at a social, legal, and ethical level and not only within the context of the laboratory**.

As a consequence, the typology will look like **a well-organized list of notions that are used in organoid research and related technological developments**. Notions are labels that are used to identify entities of the same kind, or that raise similar issues. **Each notion points to specific ethical issues that are raised by these entities, either currently or in the future**. As a typology, it is necessarily partial. Its main goal is to indicate directions for future studies and highlight points of ethical concern. The following two sections develop the main aspects of the methodology.

[10] Jasanoff, S. (2007). Technologies of humility. *Nature,* 450(1), 33

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3.2 What does it mean for a typology to be "socially robust"?

The scientific day-to-day use of words is not necessarily robust in the sense that it is subject to dispute, and because the scientific nomenclature can evolve, simply as a consequence of the fact that we are dealing with ongoing research. Uncertainty is at the core of organoid research, a research with living material. In other words, **our targets are not only hybrids but also moving objects**.

Taking this remark into consideration, the typology must **take into account the dynamic nature of the field while providing some categories that will endure over time**. The typology will try to respect a delicate balance between (i) **establishing categories as snapshots of ongoing research** and the issues it raises, and (ii) **making room for evolution and interpretation**, as we know that the products of research will evolve (we do not want our typology to become obsolete the day we have finished drafting it).

Also, a nomenclature that would strictly follow the scientific literature would not necessarily be *socially* robust in the sense that it might be not understandable for the general public, or in the sense that distinctions might not be socially relevant. **Thus, the typology is not a scientific taxonomy**. We cannot import without further consideration a list of terms used in scientific research. First, the scientific nomenclature is very rich,[11] and there is apparently not a specific ethical problem raised by each entity. A certain distinction can appear to be a very important distinction for researchers, making sense for scientific research, but there might be no point in introducing this distinction at a social, legal, or ethical level. For instance: researchers working on organ A and researchers working on organ B have different expertise, each in their respective fields. Organoids of A and organoids of B are different entities, each might require specific techniques to be developed. But taken at the level of a pathology that would impact both A and B, or at the level of a therapy that would follow a similar procedure whether physicians want to restore the function of A or B, only one category would have to be reported in the typology ("A-B-and the like organoids").

This consideration implies that, for the purpose of our typology, we do not have to look for brand new state-of-the-art scientific distinctions, but for the most significant ones instead. The distinctions proposed in the typology should make sense in society – ethically, for the general public – and not only in the context of the laboratory. The typology has to subsume, that is, put in the same box entities that are individually distinct, and subsumption implies making choices. We try to introduce distinctions between entities only when they make a difference from an ethical point of view. At some point, when introducing details of what this and that compound of stem cells are, we could still argue that entity X and entity Y belong to different kinds of entities, or do not share exactly the same properties, from the viewpoint of science or ontology. Yet there is no need to carve nature at its joints, if the distinction does not correspond to a

[11] According to WP2's map of the field, there are 100+ entities under the label "organoids."

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difference in our ethical treatment of those entities. There is still the possibility that entities will evolve so that a distinction that will be relevant for ethics appears in the near future, and in that case, the distinction deserves to be anticipated and discussed.

As a consequence, the typology is based on the vocabulary used in scientific research: terms such as organoids, embryo models are the building blocks of our conceptual elaboration. It would be counterproductive to build an entire vocabulary from scratch and try to impose it on the many stakeholders who are already familiar with concepts in the field. Yet the discussion of ethical issues sidesteps most of the experimental procedures and technical terms. In the current document, we try to find a balance between scientific concepts and common language. For instance, the typology could be useful for a readership such as ethics committees or review committees. Such committees need tools to identify quickly in a scientific proposal the potential ethical issues that would be raised by the research and its applications.

3.3 Is the typology descriptive or normative?

One classic question in the philosophy of science discourse concerns whether philosophy is (or ought to be) *descriptive* or *normative* with respect to scientific practice. In the former case, we would try to accurately describe the practices of present or historical sciences, and draw from them morals about scientific concepts, theories, progress, etc. In the latter case, philosophers would attempt to change or improve scientific practice, to describe "rational" or "irrational" behaviors in science, etc. To that end, where does the current typology position itself on this descriptive/normative axis?

There are multiple things that one might mean in saying that a philosophical account is "normative." We will draw on a distinction introduced by philosopher of science Marie Kaiser.[12] According to a first perspective, we might mean that being normative is to make claims about what makes a feature of science "good" or "bad." Kaiser calls this *metanormativity*. This is what we have in mind when we talk about "rational" theory choice: some kinds of theory-selecting behaviors are good, or rational, and others are bad, or irrational. A normative philosophy of science would tell scientists what they should do, e.g., what theory they should favor or what concepts they should adopt (or at least, the philosopher could point to a method that the scientist ought to follow in order to favor the "good" theory or adopt the "right" concept). According to another perspective, we might mean that in developing a philosophical account, philosophers have had certain ideas in mind about "good" methodologies – which kinds of evidence about science are "good," which fields in the past have been "successful," and so on. Kaiser calls this *methodological normativity*.

Our work here is largely not metanormative in Kaiser's sense. The field of research in organoids and related technologies is still relatively in its infancy, and it would be premature of

[12] Kaiser, M. (2019). Normativity in the philosophy of science. *Metaphilosophy*, 50(1-2), 36-62. https://doi.org/10.1111/meta.12348

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us to attempt to rewrite biological taxonomy as it is currently used in practice.[13] It is, however, **methodologically normative, in the sense that we have been highly selective about which scientific concepts to include and exclude from the taxonomy**.

Since our goal is to construct a taxonomy for use in understanding the ethical stakes of organoid research, we have kept this as our focus, including in general only concepts that make an ethical difference. Our goal is thus to provide an opinionated map of the territory, highlighting subdivisions in the conceptual space where the ethical landscape is likely to change. In this sense, there is no one "magic bullet" criterion for the selection and definition of concepts in the typology. We stick to a kind of pragmatism, in the philosophical sense: **to be relevant, a conceptual distinction must *make a difference* somewhere**. In other words, there is no point in attributing different categories to X and Y if they are to be dealt with (manipulated, assessed, controlled…) in the same way. We tried our best to make as explicit as possible at each step why we think that the notion X or Y deserves a specific treatment. The work presented below (that is, the picture we arrived at) is based on a literature review on organoids (and biotechnological hybrids in general) and on the ethical issues raised by the entities of interest, and on many discussions with experts and partners in the HYBRIDA project. Our basic hypothesis is that these new laboratory entities are likely to raise issues because they have new biological properties that force us to look differently at them or at their possible uses. This latter claim would be our most disputable ontological premise, but again, it is in the background of the HYBRIDA project and even of the call for projects preceding it. If organoid research were just stem cell research with no added value and nothing more at stake than what is already done in the laboratory, then why all this fuss?

We want the current document to be a support for bioethical discussion, and thus neither a guide for scientific research, nor a set of answers to ethical issues. In this sense, we can note that **the typology does not represent a guideline-style classification**. When referring to guidelines, researchers need to quickly identify the relevant category to which their research belongs. By contrast, we want here a typology that fosters discussion in the project and in the long run. The typology has to stand as a support for discussion, not close the discussion.

As a counter-example, consider for instance the ISSCR guidelines. Such guidelines are proposing a partition of research activities, that is, a typology. Yet this typology starts with the current legal/ethical status of research and the procedure that has to be adopted as a consequence.[14] It distinguishes basically between three categories: (1) research that is exempt from review; (2) research that requires specific oversight; and (3) research that is not allowed. For practical purposes, this classification conflates research objects of different natures, or that may raise different kinds of issues, into the same categories.

For instance, a technology that is highly desirable but not safe in the current state of knowledge and technique will fall under the general category of *research not allowed*. A technology that is ethically not desirable (that is, we do not want it to happen because it would

[13] Of course, instances where we lack conceptual clarity abound, and are even recognized as such within the scientific community; resolving these will be metanormative to some degree, see D1.2 and D1.3.

[14] International Society for Stem Cell Research. (2021). *ISSCR guidelines for stem cell research and clinical translation*. https://www.isscr.org/policy/guidelines-for-stem-cell-research-and-clinical-translation

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contravene certain ethical principles) would fall under the very same category of *research not allowed.* Yet there is a gap between the two technologies from an ethical viewpoint. This distinction matters if we want to identify where we need an ethical discussion or assessment of the technology and what kind of assessment we need.

One issue is that, as research moves forward, some objects might move from one of the ISSCR category to another (this is especially the case of some organoids). Some models of development that were "exempt from further review" in a previous stage of technique would become "in need of a specific review" from a certain degree of complexity and functionality (for instance, one can think of brain organoids). While guidelines can deal with this kind of situation with a recurrent update, **the typology that we propose here emphasizes the dynamics of scientific practice and wants to identify points of discussion**. For some, the typology might seem to be too futuristic when it includes brain organoids and mentions the potential issues surrounding consciousness and sentience, as well as the use of organoids as biological product synthesizers, or even bioartificial organs. These might not be issues of current research, but the purpose of the typology is to anticipate and give directions for future analyses of the legitimacy of certain research projects, based on the maturity of the public debate and clinical expectations, without closing the road to pure research.

4 A typology of biological artificial entities

4.1 How to read the typology?

The typology identifies NOTIONS or CONCEPTS, which are briefly defined and exemplified in the text. Under the same concept, one will find entities of the same kind. We hypothesize that these entities can be **grouped because they share common properties**. As we do not want to reproduce a scientific typology but only to pinpoint major ethical issues, we do not need distinctions between concepts that are too subtle. For instance, we do not introduce a specific distinction between liver organoids, gut organoids, pancreatic organoids, etc., as we make the hypothesis that these organoids will raise the same kind of ethical issues when they fall under our concept "Organoids." Furthermore, other issues will be raised when these organoids are conceived as Bioartificial Organs," but, again, e.g., liver, gut, pancreas bioartificial organs will probably raise the same family of issues, even if an in-depth review could argue for more concepts. We stick here to the most significant distinctions so that we obtain a general map of the field.

As notions are defined by the fact that the entities behind a concept share common properties, **each notion is defined in relation to a SPECIFIC QUESTION, that is, a question that points out the property that makes relevant the definition of a new notion**. In other words, a specific question appears wherever we need to introduce a distinction in the "socially robust typology" because a new remarkable property emerges. If the entity has the property in question, it becomes subject to a new set of ethical issues. For instance, if something falling under

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our concept of "Organoids" also has the property "matured into organs," then it also falls under our concept of "Bioartifical Organs," and further ethical issues will thus be relevant (see below).

Following this delineation of specific questions and concepts, **we obtain a certain hierarchy of terms and questions that are presented below**. Each entity inherits some properties of its predecessors (and shares some of its ethical concerns), but it might also exhibit some new properties.[15] We acknowledge that other valid typologies could have been proposed by posing different specific questions. Our main concerns are those of consistency and of completeness, as we want to take the main problematic entities into account while we do not want to see labels proliferate. Some entities are also still under development, or will be a target for research in the near future, so we also want to keep the typology open for the dynamics of biomedical research.

Each concept is followed by a short mention of ETHICAL ISSUES related to this concept. To keep the typology informative, we try to describe at each stage a specific series of ethical issues raised by a given concept. For instance, there are ethical issues with stem cell research in general that are common to all research with SC as biomaterial, and there are also more specific issues that appear when researchers develop complex models of embryo development, of brain development, when they mix cells from human and animal sources, and so on.

This does not mean that ethical issues raised at some points are exclusive to one given concept. We cannot be content with a partition in the mathematical sense, where each entity belongs to only one concept and no entity falls outside the domain. Ethical issues are not isolated; they interact. Furthermore, concepts themselves overlap. For instance, an entity can be at the same time a chimera and an embryo model, each raising distinct, but cumulative and potentially interacting, ethical issues. From the viewpoint of a bioethicist, the brief mention that we make of some ethical issues will probably be too succinct. Further exploration of ethical issues is to be conducted in WP2, 5 and 6.

4.2 Flowchart

The following flowchart presents in the most succinct way the list of concepts and how they articulate. Each arrow is marked by one or more specific questions, that is, significant specifications of the concept. Dotted boxes represent entities that are more future visions than current laboratory developments.

[15] While organoids derive from stem cells, our current problem, and the very existence of the HYBRIDA project, is based on the fact that organoids might have new features that make them somehow ethically different from other kinds of entities developed by stem cell research in the past. We might want to apply the rules of stem cell research to organoids as far as they belong to stem cell descent (consent, control over data, issues in biobanking…), but we also might want to find new rules that apply specifically to organoids when relevant

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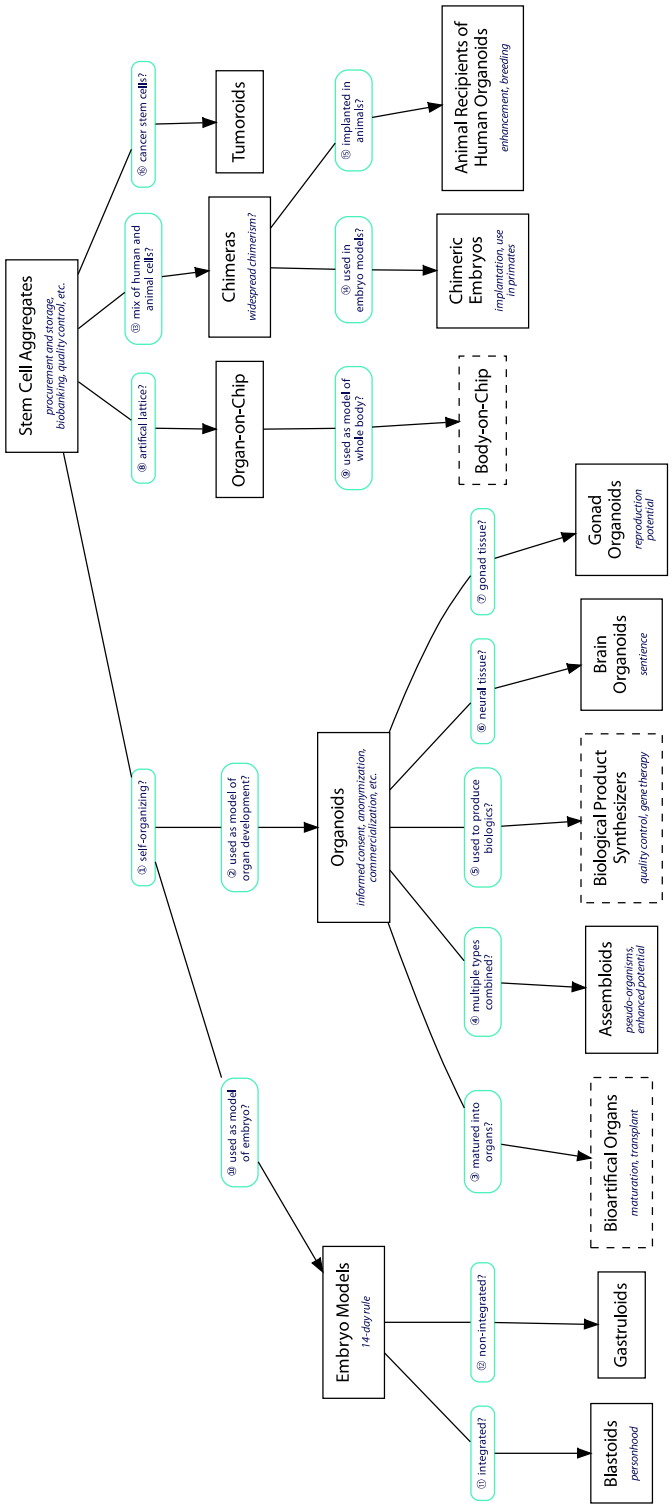
Figure 2: The typology as a flowchart



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4.3 Narrative

Following most accounts, organoid research is one of the latest developments of *stem cell research*, especially research on and with human stem cells, that has seen continuous progress since James Thomson produced the first human embryonic SC line in 1998.[16] Since then, there have been debates on stem cell research, stem cell procurement and the possibility of new treatments through cell therapy and regenerative medicine. SC research has been a controversial field from its inception, a field for political interventions and scientific promises.[17] Sociologist Sheila Jasanoff, in a comparison of the relation of bioethics to politics across countries, goes even on to say that the SC debate played a role in the institutionalization of bioethics.[18]

If organoid research is an expansion of SC research, then, as a simple consequence, organoid ethics should be seen as an expansion of SC ethics. An illustration of this claim is that the most comprehensive set of ethical rules of play developed to date by professionals providing a framework for organoid research is the ISSCR guidelines, that is, the SC research community.[19] Developing an *organoid ethics* means being aware of the background and all the discussions that occurred in the context of stem cell research. After all, if organoids are made from stem cells (even if SC might not be present in organoids any longer, as they would have differentiated), one has to know the stem cell (ethics) basics in order to develop organoids in accordance with ethical standards. This would be a minimal requirement.

At this point, it would be informative to introduce a distinction between "stem cell" as a concept, "a stem cell" as a discrete entity that could be identified as *one* stem cell, and "stem cell culture," the pieces of tissue that scientists are dealing with every day in their laboratories. An egg is literally one stem cell, with the capacity to potentially develop into an entire organism. But most of the time scientists do not work with unique cells: they remove tissue, work with cells in suspension, let them divide and assemble *in vivo* or *in vitro*. In this sense, researchers work with SC aggregates, or SC cultures. The concept of stemness in itself should not be taken too rigidly, as there is still uncertainty over what this concept refers to: Does it make sense to speak of one SC in isolation as if stemness were "sticking" to the cell whatever happens? What about niche properties? Reprogrammation of cells into SC by environmental changes?

The first, and, in a sense, most primitive, kind of entity that is obtained when culturing stem cells in three dimensions, is what we call "**stem cell aggregates**." By this term we refer to entities such as teratomas, embryoid bodies, and spheroids which are composed (or derived) from SC that are differentiating (or have differentiated) until they develop certain properties – a ball of cells for spheroids, different tissue types for embryoid bodies and teratomas. However,

[16] Thomson, J.A., Itskovitz-Eldor, J., Shapiro, S.S., Waknitz, M.A., Swiergiel, J.J., Marshall, V.S., Jones, J.M. (1998). Embryonic stem cell lines derived from human blastocysts. *Science*, 282(5391), 1145-1147. https://doi.org/10.1126/science.282.5391.1145

[17] For an overview, see for instance Scott, C.T. (2005). *Stem cell now.* Penguin.

[18] Jasanoff, S. (2007). *Designs on Nature: Science and Democracy in Europe and the United States*. Princeton University Press.

[19] ISSCR guidelines, op.cit.

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these entities do not exhibit a complex spatial layout and an organization that is typical of the actual organ. These entities have been present in the laboratory for decades. For instance, teratomas are an assay for stemness, and the development of embryoid bodies is a source of SC for all ranges of studies, including the development of organoids. The culture of SC aggregates raises ethical and practical issues on its own, such as quality control and genetic drift of cultures, before their potential storage or commodification as laboratory products.

This remark calls for an important distinction between ethical issues related to the procurement of SC and the development of SC-derived entities in the laboratory. Bioethicist Insoo Hyun insists on maintaining the distinction between the "embryo debate" and the "stem cell controversy."[20] The embryo debate focuses on the fate of embryos and asks whether researchers should be allowed to work with embryonic SC. In this framework, embryonic SC are theoretically seen as potential persons. However, the actual scope of the SC debate is much larger and the bioethical discussion over the future of SC research has already moved forward. According to Hyun, SC research interrogates our ability to control life, to master biological development and to play with its plasticity. In the end, our power over nature and our attitudes toward aging and disease are at stake through the expected contributions of stem cell research and biotechnologies derived from stem cells to regenerative medicine and human enhancement. SC research ethics deals, therefore, not only with the issue of whether scientists should act upon embryonic SC or not, but also a whole range of possibilities and procedures for producing laboratory entities derived from SC (from all kinds of SC: cancer, adult, embryonic, or induced).

The typology starts with the most basic entity from which organoids are built, that is, stem cells, or more precisely in terms of laboratory research, stem cell aggregates, and it expands as biotechnological hybrids develop into more complex entities.

One of the entities derived from SC that is used in labs is **tumoroids**, made from cancer SC. These models can be used to understand cancer development from a basic research perspective or to test drugs according to the paradigm of personalized medicine. Issues raised with this kind of entities – probably the most important ones in terms of volume if we look at actual laboratory research today – are issues raised in oncology and personalized medicine: epistemological uncertainties of this new model and the challenges it poses for translational research, difficulty to assess the clinical efficiency of procedures, respect of and communication to patients regarding prospects of the treatment and potential therapeutic misconception, etc.

Tumoroids are still a rather simple entity. Whether a particular tumoroid reproduces the tumor of origin is even still a matter of dispute due to the difficulty of assessing tumor heterogeneity and developmental randomness. Tumors are by definition proliferating entities that disturb the structure of the organism in which they develop. By contrast, a major property of a healthy tissue/organ is its organization. It is commonly held that organoids are self-organizing biological entities derived from stem cells: once the cells are in the right culture conditions, they will spontaneously assemble into a complex entity. In a sense, this property of self-organization or self-assembly, however difficult it might be to interpret,[21] is foundational to the concept of an

[20] Hyun, I. (2013). *Bioethics and the future of stem cell research*. Cambridge University Press.

[21] Whitesides, G., Grzybowski, B. (2002). Self-assembly at all scales. *Science,* 295(5564), 2418-2421. https://doi.org/10.1126/science.1070821; Turner, D.A., Baillie-Johnson, P., Martinez Arias, A. (2016).

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organoid. In this sense, self-organization, or self-assembly, is said to be the major event that characterizes the development of organoids in vitro. The definition of self-organization might not be as easy and consensual, but the general idea is that the entity, by itself (although in an artificial laboratory context and under specific environmental constraints), is going to manifest a specific shape that it is bound to manifest. This property will probably matter when considering ethical issues, as they make these models of development very close to biological entities that develop in a natural context.

Here, we have to introduce a distinction between two main kinds of self-organizing SC-derived entities: organoids and embryo models. **Embryo models** are in vitro models of embryonic development. Given the history of debates surrounding the status of natural embryos, it is fair to expect embryo models (some say "synthetic embryos") to attract attention and debate in the field of bioethics. Issues to be discussed would include: To what extent do these entities mimic real embryos? Can they develop like real embryos? Could we envision implantation?

By contrast, the typical concept of an **organoid** is a model of development of a particular organ (liver, gut, brain…). Organoids are a family of entities that manifest the spatial organization of the early organ (vs. simpler in vitro 3D constructs such as spheroids) and that are used as models of development for basic research in embryology and disease modeling. This is where the metaphor started and, as the term suggests, it is included in the concept that the organoid should look or function like an organ – and that is the reason why we suggest putting embryo models under a separate concept. However, the term does not immediately suggest that we are speaking most of the time of an embryonic organ (a model of development, not a model of a fully grown, mature organ). Now, does the property of self-organization entail by itself ethical issues, beyond the "plasticity of the living" mentioned above? Unlike embryo models, organoids might not raise issues at this early stage of research: the fact that researchers obtain in vitro something that looks like an early liver or an early gut does not raise the same potential objections as having an embryo in a dish. But the organoid category can be unfolded in many directions, and we should look into the details for each. We do not need to detail all the types of organoids that have been described (and created) by laboratory research; we will just regroup them into distinct categories when they are likely to raise specific ethical issues.

The first kind of entity that we can envision is **bioartificial organs**. The long-term goal (only a vision at this point) would be to supply organs (or parts of organs) for transplants, in strong contrast with current organoids, which function rather as models of development for disease modeling, drug screening, and so on. The way biomedical research would be able to provide such "spare parts" should of course be the matter of a bioethics debate in the years to come. In this regard, it is important not to conflate issues and interpret all organoids (mostly, models of development) through the prism of regenerative medicine. The category can stop here (for instance, we do not introduce a specific distinction between bioartificial livers, guts, pancreases, etc.) given its level of abstraction and because the same kind of issues will probably be raised for all types of organoids, except those listed below that could raise specific issues in the lab.

Organoids and the genetically encoded self-assembly of embryonic stem cells. *Bioessays*, 38(2), 181–191. https://doi.org/10.1002/bies.201500111

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One organ at least is susceptible to raise particular ethical issues even at the stage of models of development: **brain organoids**. The nervous system being key in sentience and consciousness, a brain organoid could acquire some properties that would make it qualify for a certain moral status, even as a research object.

Gonad organoids might not raise issues as models of development but questions about their use, particularly the possibility to use them as fertility treatment, might appear.

Assembloids, as a combination of several organoids, might also raise specific issues. Problematic assembloids include at least those involving brain organoids and reproductive tract assembloids, but the list might not be limited to them.

Finally, we could envision the use of organoids not as models of development but as **biological product synthetizers**, to be used as part of an industrial process. This use would of course be subject to debate and regulation.

Taking one step back – leaving the level of organoids and going back to SC aggregates – we have two other possibilities for the development of entities that deserve ethical discussions.

The first would be **chimeras**, that is, the mixing of cells from different genetic origins, especially chimeras mixing human cells with animal genetic material. Entities such as chimeric embryo models, chimeric organoids deserve a specific treatment from ethical committees as they can manifest certain emerging properties and to the extent the chimerism should stay under control (avoid widespread chimerism or reproduction).

The second one would be **organs-on-chip (OOC)**. OOC differ from organoids in that they do not self-organize but are instead built on an artificial lattice. But they raise similar issues to those raised by organoids. For instance, as organoids can be combined into assembloids, OOC could, in the future, be combined into a **body-on-chip**.

4.4 Typology

Concepts or notions are in CAPITALS. Specific questions have a number that has been attributed according to their position in the flowchart. The *ethical issue*s mentioned below (*in italics, preceded by an arrow*) are mainly suggestions for points of concern and do not represent an extensive review of the field.[22]

The typology is focused on stem-cell derivatives. Stem cells are indeed the primary source of biological material from which organoids are built. Stem cells are usually characterized by two properties: 1/ the ability to self-renew, that is, a stem cell produces by division cells that are still stem cells; and 2/ the ability to differentiate, that is, generate different, specialized cells. A cell is an individual entity with closed boundaries (a membrane) and identifiable parts (e.g., nucleus, organelles). A stem cell (SC) cannot be easily identified as an individual among a population of 'regular' cells. A stem cell is a dynamic entity: stemness is a property of the cell that can be

[22] This would be the purpose of WP6.

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realized only when the cell generates offspring by dividing itself or by becoming a specialized cell.[23]

In the biology of development, stem cells are classified by reference to their differentiation potential, or "potency": totipotent SC can differentiate in any kind of cell and tissue, up to an entire organism, pluripotent SC can differentiate in all cell types of an adult organism, multipotent SC can differentiate in some cell types, and unipotent SC will produce one cell type. Additionally, progenitor cells will transform into a specific type of cell (without self-renewing). SC can also be categorized by their source (which overlaps with the above classification): embryonic stem cells (eSC) are collected from a 5-day-old embryo inner cell mass while adult stem cells (aSC) are located in more mature organisms (adult or at a certain stage of foetal development). The former ones are pluripotent and the latter ones are typically multipotent or unipotent (or oligopotent, generating a small variety of different cell types). Other types of SC are induced pluripotent stem cells (iPSC), which result from a bioengineering procedure allowing a differentiated cell to be turned into a stem cell, and cancer stem cells (CSC), the SC that give rise to tumors. These types of SC do not always have the same properties. According to the source of SC, the derivatives will also manifest different properties, or different risks (e.g., the risk of tumor development is increased with the use of iPSC by contrast with eSC).

The ethical issues raised by the procurement of stem cells have been largely debated in the last twenty years. Most of the debate focused on the collection of human embryonic stem cells. The use of excess in vitro fecundation embryos for research purpose has raised a public discussion about the moral status of the embryo and whether researchers should be allowed to use them as a source of material (some of these issues have been reviewed in D3.1).[24] Oocyte procurement is also a controversial practice, giving rise to issues with egg donor compensation. As hormonal induction and procedures required to collect oocytes are painful, if women are not remunerated for undergoing it, this can be seen as exploitation; if they are remunerated this can be seen as an unfair inducement. The issue of the creation of so-called synthetic embryos as a source of human eSC is also a matter of debate and subject to different regulations. Induced pluripotent stem cells are an object of the ethical discussion since their emergence in 2007.

"Stem cell" is a concept that has been formulated in the context of developmental biology and which is still the subject of many disputes and research. The discovery of "a stem cell" is nothing like the discovery of a new planet. We have to propose an ontology of research objects (organoids and related entities) adapted to the constraints of the "stem cell theory" (and there is even no such thing as a unified theory of SC). For instance, philosopher of biology Melinda Fagan underlines that stemness is context-dependent. That is, a stem cell is a SC because of its location in a certain environmental niche during the course of development and its position

[23] "To be a stem cell is to be the origin of a cell lineage, either actually or potentially. Another way to put the point is that stem cells are essentially generative: they can produce other cells, both like and unlike themselves… Because they are defined in terms of their descendants, stem cells are essentially future-oriented." Fagan, M. (2017). Stem cell lineages: between cell and organism. *Philosophy Theory and Practice in Biology,* 9(6), 1-23. http://dx.doi.org/10.3998/ptb.6959004.0009.006

[24] Kavouras, P., Spyrakou, E., Stavridi, V., Deligiaouri, A. (2021). *HYBRIDA D3.1 Map report of Normative, Research Ethics and Research Integrity frameworks.* EU Commission.

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regarding neighboring cells. Even if many unknowns remain, there seems to be a consensus that stemness is not an intrinsic property of certain cells.[25] One thing that we need to avoid in the construction of our bioethical discourse is thus the reification of SC as a certain type of product that would have the intrinsic power to cure / develop into an organ / become a person. As a consequence, we take "stem cell aggregates," as the point of departure for our typology and specific questions.

STEM CELL AGGREGATES

In our understanding, stem cell aggregates are the material from which stem cell researchers work. They may be distinguished from a bunch of cells that happen to have been collected together and that may contain stem cells or manifest the property of stemness if put in the right conditions. Stem cell aggregates appear after a period of in vitro cultivation. At some point, the cell culture develops into a small colony, and if the cells composing this colony come from the same stem cells that are differentiating, then we can say that we had stem cells in the first place.[26]

Entities that might fall in this category:

- Teratomas/teratocarcinomas/tumoroids

Teratomas are human SCs that grow as tumors in mice. As pluripotent SC, they are the source of all kinds of tissues of the three germ layers (but without organization, as will be seen in organoids). Teratocarcinomas are a type of tumor that grows naturally in testes and that have the specific ability to develop cells that belong to many body parts (e.g., muscle, hair, teeth, etc., in a tumor). This family of entities was fundamental to the development of SC research since the mid-twentieth century.

- Embryoid bodies[27]

Embryoid bodies (EB) are products of 3D cell culture: when cultured in a hanging drop instead of a Petri dish, SC grow until they form a small sphere with SC at the core and more differentiated cells at the periphery. These structures are called embryoid bodies because they look like an early stage of embryo development when the three germ layers appear.[28] Cells are then committed to a lineage. In that sense, EB are used as a source for the neural SC used to grow brain organoids.

- Spheroids

Spheroids is a generic label for entities obtained through three-dimensional cell cultures (such as hanging-drop culture). The concept of a spheroid would deserve an extensive inquiry per se, as we did with the concept of organoid in D1.3. Some ground the existence of spheroids in the principle of self-assembly:[29] cells in a non-adherent environment aggregate together thanks to spontaneous cell-cell interaction. Spheroids are very simplified models, but, because "life is 3D,"

[25] Laplane, L. (2016). *Cancer Stem Cells: Philosophy and Therapies*. Harvard University Press.
[26] This is following Melinda Fagan's account of "lineage model" for the ontology of SC.
[27] Davies, R. (2021) HYBRIDA WP2 document *Embryoid Bodies*.
[28] Ectoderm (neural SC), endoderm (internal organs: e.g., liver, heart), mesoderm (e.g., muscles, bones).
[29] Laschke, M., Menger, M. (2017). Life is 3D: boosting spheroid function for tissue engineering. *Trends in Biotechnology,* 35(2), 133-144. https://doi.org/10.1016/j.tibtech.2016.08.004

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these models are still better than 2D cell cultures for some uses. Compared to the former, spheroid aggregates show an increase in activity and metabolic function and manifest a primitive level of spatial arrangement.

Stem cell aggregates have a wide range of uses in biomedical research. At the most basic level, embryoid bodies and teratomas are used as tests to determine whether a given sample of cells is indeed composed of stem cells. Spheroids are mainly used for drug discovery in personalized medicine and toxicology screening, and they offer also some perspectives in regenerative medicine (e.g., cartilage repair).

➔ *Ethical issues: as SC are not "any kind of cell" but can be the source of other kinds of cells, they have a potential that has drawn the attention of personalized and regenerative medicine. Prospects in SC research are linked to the special abilities of these cells and their position in the developmental process of the organism. Along with the ethical issues related to SC procurement, there are issues related to their becoming and potential.*

- *Issues related to biobanking and its regulation (depository of biological specimens). Risk of breach of privacy: Issue of anonymization/identification (anonymization as a way to prevent a privacy breach, but what if researchers want to contact the donor to collect supplementary information, collect informed consent, or share information about a finding potentially relevant for the individual's health). In the case of iPSC: issues in contact and tracking of donors, dealing with incidental findings, the scope of the consent, commercial benefits. Issues of commodification and property, potential benefits made through research using donated biomaterial.*
- *Standards in production and dissemination of SC: Quality control (genetic drift, mutations in cell culture).*
- *Regulation of biomedical research in general, from bench to bedside (all the intermediate steps: preclinical tests…).*
- *Specific issues related to cancer research (the main domain of application of these entities): patients as vulnerable populations, issues of patients' consent, therapeutic misconception, etc.*

SPECIFIC QUESTION 1 Does the stem cell aggregate exhibit emergence of architecture and functionality (i.e., *in vitro* differentiation of tissues, similarity with organs or stages of development) through self-organization or "self-assembly"?[30]

[30] It has to be noted that the definition of self-assembly and self-organization are particularly unclear from a philosophy of science point of view. What about the constraints imposed by the experimental context, e.g., biochemical factors? This specific question is intended to highlight the ontological difference between organoids and OOC, whether there is a significant difference in the ethical issues is a topic for another work (WP2, HTA?).

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SPECIFIC QUESTION 2
Is the stem cell aggregate used as a model of organ development?

ORGANOIDS

An organoid is a three-dimensional stem cell culture that differentiates into a physiological system that looks like an organ of the body or tissue at a specific stage of development. The organoid shares at least some structural/spatial/architectural properties of the target organ (crypts for intestines, cups for retinas…) and manifests also some function(s) of the target organ. As we said in D1.3 (a discussion of the history and the consistency of the organoid concept): notwithstanding the conceptual uncertainty in this ongoing field of research and the sometimes rather loose understanding of terms such as organoids/embryoids/spheroids, there is obviously something more in the current concept of an organoid compared to simple embryoid bodies and spheroids. While the latter does not exhibit a stable structure, the former are spatially organized and look like organs in development. In a sense, starting with the same biological material (differentiating stem cells), **embryoid bodies and spheroids are more primitive entities or less specific models of development**.

➔ *Ethical issues directly related to the existence of organoids are:*

- *Informed consent regarding existing biological samples. Even when broad consent is given, it is given in a certain context of research and some possibilities of what is going to be done with the collected cells are explained. As organoids are new entities, could researchers assume that donors of SC gave their consent to the development of organoids from their SC?*
- *Now that researchers are aware of the possibility of developing organoids from collected biomaterial, should we ask for broad consent? dynamic consent? etc.*
- *Issues with anonymization and incidental findings. If organoids render anonymization more difficult or incidental findings more frequent and accurate, these issues would have to be dealt with somewhere.*
- *However, do organoids, as SC models and before we add any further complexity to the picture (see below for more specific questions), raise specific issues that are not raised by simpler models (such as spheroids)? The answer is not obvious. The ISSCR guidelines stipulate that, in the current advancement of research, most organoid in vitro research does not raise specific ethical issues.*[31]

[31] In the ISSCR classification, this means that most organoid research falls under category 1A: exempt from review by a specialized oversight process.
This being said, the typology that we propose here is based on the idea that some biotechnological entities have properties that raise new ethical issues compared to more basic entities that do not have these 'problematic' properties. As an example (see below for details): if being a brain organoid implies having the property of (possibly) developing a form of sentience, then there is an obvious ethical issue in that sentient entities deserve some kind of consideration. However, if there is not such an identifiable property that is ethically problematic, as it seems to be the case for many organoids, this does not preclude the

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SPECIFIC QUESTION 3
Organoids are defined as models of development: they do not look like fully developed organs and cannot become functional organs in an organism. What if an entity originating in organoid research develops into a potential organ for transplantation?

BIOARTIFICIAL ORGANS

In this configuration, organoids reach an advanced stage of development, so that they look like, or present functional similarities with, real organs.[32] This is not something that the current state of research in biotechnology allows but this vision is present in many discourses around organoids as mini-organs. In medicine, there is a long tradition of tentative development of artificial organs.[33] The idea of "spare parts" is also quite present in the reports from the deliberative mini-publics workshops (see D4.3). Yet we have to be careful not to conflate issues raised by current research on organoids with prospective insights into the future of regenerative medicine. Our typology offers the possibility to delineate problems: we can at the same time affirm that organoids as models of development are not mini-organs, and open space for discussion on bioartificial organs at another level.

How would these artificial organs be developed? There are two options: a full maturation in vitro may be possible for simpler organs, whereas for more complex organs a maturation in vivo, in animal recipients, is more likely. Bioartificial organs developed from organoids would be a subclass of bioartificial organs trying to remedy the shortage of organs for transplants (such as those potentially obtained through bioprinting, xenograft…). Such bioartificial organs would be typical hybrids: neither natural nor artificial, neither biological nor technological (in the restricted, common sense of these terms). A pending issue would be the determination of the boundary between cell therapy and prosthetics and whether this boundary still makes sense.

existence of second-order properties that might imply that the current state of ethical reflection or the current regulation should evolve. Organoids as models of developments are in a sense just better models than non-self-organizing or non-3D SC culture, but with better models comes the possibility of a better understanding of diseases, better anticipation of toxicity, a better prediction of success of personalized treatment, a higher chance of significant incidental findings, and so on. The higher probability of commercialization and making benefits from SC derivatives is a particularly interesting second-order property. This is not a property of the organoid as a biotechnology like a new function: the organoid does not have the property of raising benefits as it has the property of sharing some function with the target organ. However, the fact that the organoid shares some function with the target organ will lead to better models, better predictions of drug efficiency, etc. which, in turn, increase the probability of making benefits from these entities. To which extent does this improvement forces us to reconsider existing rules (e.g., for biomaterials depositories)?

[32] Or parts of organs. An organ can have several metabolic functions. If the patient's organ is deficient in one specific function (e.g., the synthesis of the molecule X), we can envision the graft of an organoid that provides replacement for this very specific function (molecule X synthesis), even if the full organ in normal conditions is supposed to produce molecules X, Y, and Z.

[33] Duguet, A-M. (2021). Artificial organs. In H. ten Have (Ed.), *Encyclopedia of Global Bioethics* (pp. 176-185). Springer. https://doi.org/10.1007/978-3-319-09483-0_27

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Bioartificial organs would be somewhere in between. A helpful analogy can be the recent pig heart transplant[34] in that the procedure implies genetic modification (hence, artificial, technological), but the object is still, until the beginning of the transplant procedure, a functioning heart in the pig, exactly like a "regular" organ transplant from a human donor. Regulation (including safety and ethical issues) will also imply a systematic comparison and classification of artificial organs (e.g., mechanical valves) and between different types of bioartificial organs. As it is likely that organoid research will contribute to the development of bioartificial organs in the long run, we will have to ask where bioartificial organs derived from organoids fit within the general landscape of bioartificial organs and where to locate them regarding the regulatory classification of e.g., medicinal product vs. medicinal device.

➔ *Ethical issues:*
- *Artificial organ development: anticipation of issues in safety/procedures/clinical research… Are we going to transplant them at some point?*
- *Issue with in vivo maturation: see "chimeras/animal hosts"*

SPECIFIC QUESTION 4 Have several organoids (or functional substructures) been merged to form a single, interacting physiological system?

ASSEMBLOIDS

Assembloids are assemblies of organoids: several functional organ-like structures are developed separately and linked together afterward. An assembloid is a 3D stem cell culture evolving by itself (in which connections between organoids are not forced and are left to biological development), but it lacks the organ-like character of an organoid (see D1.3 for an analysis). The purpose of growing assembloids is to study the development of connections between organs or to study organs with complex structures. One can also hope to model more realistic physiological systems, as organs in the body do not function in isolation. For instance, there have been many organoids of reproductive organs (e.g., fallopian tubes, cervix), but what if these organoids are grown together and connected? Would they be able to mimic, in some sense, the female reproductive tract as a whole? The brain is composed of many subsystems processing specific information domains, and most brain organoids today are organoids of particular brain regions. Now, researchers are trying to produce brain organoids themselves composed of several brain region organoids.

At the extreme point, the emerging properties of an assembloid would be "organismal" properties, provided that some essential functions of an organism or a critical number of essential organs are present in the physiological system. The tipping point where an assembly of organs becomes a potential organism remains to be determined.

[34] Reardon, S. (2022). First pig-to-human heart transplant: what can scientists learn? *Nature,* 601, 305-306. https://doi.org/10.1038/d41586-022-00111-9

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➔ *Ethical issues: if the purpose of assembloids is to study the interactions of organs in an organism, we could ask how far the model should stay from becoming a real organism. This is an adapted version of the "onrushing ethical dilemma": when the surrogate is getting closer to the real thing, it raises more questions.*[35] *So, ontologically speaking: how many organs does it take to make an organism? When would an assembloid develop some properties of an organism? Should we impose a limit on the development of assembloids? There is a specific issue with assembloids that include brain organoids: either because we could produce enhanced brain models by connecting organoids representing several parts of the brain*[36] *(which would raise issues related to sentience and consciousness that simpler brain models/organoids cannot raise), or because connecting brain organoids to other organoids (such as the gut-brain axis) is a way to "embody" brain organoids or to give them a body, a significant step in the development of potentially sentient entities. Indeed, many arguments deflating the concern surrounding brain organoids today rely on the idea that a nervous system in a dish – that is, without a body – cannot develop a sense of consciousness that has a chance to become ethically relevant. With assembloids comes the possibility of endowing brain organoids with input or output organs by connecting them to organs of the senses, muscle tissue, gut, or even artificial bodies.*

SPECIFIC QUESTION 5 Are organoids used as a source of biological products?

BIOLOGICAL PRODUCT SYNTHESIZERS

In this configuration, organoids are used as tools to obtain derived products. Such an organoid is intended neither as a model of an organ, nor as a clinical surrogate for an organ, even in a futuristic vision of organ replacement, but as a tool for the production of biomaterial (e.g., viruses for gene therapy).

➔ *Ethical issues:*
- *Quality control for production, standards and steps of clinical research (how are we going to test & use these products?)*
- *Issues with gene therapy: safety, efficiency, cost control and fair access to gene therapy.*

SPECIFIC QUESTION 6 Are the building blocks for organoid development neural stem cells?

BRAIN ORGANOIDS/CEREBROIDS

[35] Greely, H.T. (2021). Human brain surrogates research: the onrushing ethical dilemma. *The American Journal of Bioethics,* 21(1), 34-45. https://doi.org/10.1080/15265161.2020.1845853

[36] The assumption being here that the brain is an "organ of organs" that could reach its full functionality (i.e., thought, or consciousness) only if a significant number of components are present.

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Made from the 3D culture of neural stem cells, brain organoids are models of the early stages of development of the nervous system or of some parts of the early brain. We introduce a distinction between brain organoids and other organoids as a consequence of the properties of the nervous system or of tissue composed of neural cells compared to other organs/kinds of tissue. Neurons have the ability to send electrical signals that are used to exchange information in an organism (e.g., send a motor command to muscles) or to process information or compute (through a complex network of subcomponents integrated into a "nervous system"). The ability to perform this kind of computation is generally seen as the condition of possibility for the emergence of cognition, or thought. The view can be contested in the details and many concepts would have to be refined, yet, to stick to a minimal theoretical commitment, we could say that an entity endowed with a functional nervous system is much more likely to manifest something like cognition, thought, or even consciousness, than an entity deprived of a nervous system. In other words, to borrow the language of contemporary cognitive neuroscience, if "the brain is the organ of thought" (or "the function of the brain is to think"), then sufficiently developed brain organoids might be the site of some kind of thinking. The property that would matter ethically is not computation in itself but rather some sort of sentience, experience, awareness, pain, or consciousness. This would be especially the case for brain organoids developed from human stem cells, but the same issue could arise with stem cells from other species, provided that the model is sufficiently complex.

We could introduce a further distinction between brain organoids as generic models of early development and specialized models of consciousness. The latter would be developed in the context of the study of consciousness and mental/neurological pathologies, with the explicit aim of producing brain organoids that are specific models of consciousness (aiming at producing some kind of consciousness in the model) or brain organoids that will exhibit some form of cognitive impairment, pathology of consciousness, and so on.[37] While one could mitigate the ethical issues by avoiding the possibility of sentience and consciousness on many organoids (by imposing limits to the complexity of the model or depriving it of nociceptors), research on consciousness would inevitably face the dilemma of brain surrogates mentioned earlier:[38] the better the model, the closer it is to the kind of consciousness that will preclude us from conducting experiments on it.

There is currently no consensus with regard to what phenomena such as sentience, experience, awareness, pain, and consciousness actually entail and very few insights in the way researchers could produce operational procedures to measure these phenomena in various brain models.[39]

➔ *Ethical issues:*

[37] Borrowed from Lavazza, A. (2021). 'Consciousnessoids': clues and insights from human cerebral organoids for the study of consciousness. *Neuroscience of Consciousness,* 7(2), 1-11. https://doi.org/10.1093/nc/niab029

[38] Greely, op. cit.

[39] Gaillard, M., Botbol-Baum, M. (2022). Pursuit of Perfection? On Brain Organoids as Models, *AJOB Neuroscience*, 13(2), 79-80. https://doi.org/10.1080/21507740.2022.2048735

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- *Should we attribute a moral status to potentially sentient entities? How should researchers in the laboratory treat entities with the potential for sentience? Should we have standards to ensure non-sentience or minimal sentience? Should we implement procedures for assessing potential sentience in brain organoids and assembloids? Should we refuse the development of such entities or accept them under certain conditions?*
- *Subsequent issue of consciousness assessment (i.e., the assessment of the possibility that a form of experience emerges in a brain organoid/assembloid implies that researchers agree on a definition of experience/consciousness and on some operational criteria to detect signs of consciousness). The issue of "detection of consciousness" applied to organoids seems to be a research question rather than an ethical issue, but the ethical treatment depends on our ability to find such criteria.*

SPECIFIC QUESTION 7 Are the cells used for organoids development germinal SC?

GONAD ORGANOIDS

Researchers are developing gonad organoids.[40] For instance, in the case of testicular organoids,[41] this basic research is motivated by growing concerns over infertility and male reproductive disorders (testicular cancer, decrease in natural sperm production). From a biotechnology viewpoint, this could lead to the development of in vitro gametes, opening a perspective toward regenerative medicine. Given the in vivo functional dependence of gonads on hormones produced by distant organs, gonad organoids are likely to be integrated into larger assembloids or developed as organs-on-chip instead.

A pending question is whether this technology – generating gametes from organoids – is fundamentally different from the production of gametes directly from pluripotent SC[42] or other forms of artificial gametogenesis (in particular from an ethical point of view[43]).

[40] For a review see Gargus, E.S., Rogers, H.B., McKinnon, K.E., Edmonds, M.E., Woodruff, T.K. (2020). Engineered reproductive tissues. *Nature Biomedical Engineering,* 4, 381-393. https://doi.org/10.1038/s41551-020-0525-x who suggests that "New [ethical] concerns will arise as gametes and niches in which gametes develop are created."

[41] Baert, Y., De Kock, J., Alves-Lopes, J.P., Söder, O., Stukenborg, J-B., Goossens, E. (2017). Primary human testicular cells self-organize into organoids with testicular properties. *Stem Cell Reports,* 8(1), 30-38. https://doi.org/10.1016/j.stemcr.2016.11.012

[42] Easley, C.A., Phillips, B.T., McGuire, M.M., Barringer, J.M., Valli, H., Hermann, B.P., Simerly, C.R., Rajkovic, A., Miki, T., Orwig, K.E., Schatten, G.P. (2012). Direct differentiation of human pluripotent stem cells into haploid spermatogenic cells. *Cell Reports,* 2(3), 440-446. https://doi.org/10.1016/j.celrep.2012.07.015

[43] Mathews, D.J.H., Donovan, P.J., Harris, J., Lovell-Badge, R., Savulescu, J., Faden, R. (2009). Pluripotent stem cell-derived gametes: truth and (potential) consequences. *Cell Stem Cell,* 5(1), 11-14. https://doi.org/10.1016/j.stem.2009.06.005

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➔ *Ethical issue:*

- *Potential use for reproduction of the gametes obtained this way has to be discussed. If such a technology could reach maturity and safety rather soon, might this end the debate on reproductive cloning or on oocytes procurement for research?*

SPECIFIC QUESTION 8 Are SC aggregates mounted on artificial devices (instead of self-organizing)?

ORGANS-ON-CHIP

An organ-on-chip (OOC) refers to a small microphysiological system developed in a microfluidic cell culture device with perfused chambers. The artificial structure allows the reproduction and control of the chemical environment and mechanical constraints of the target organ. By contrast, organoids self-organize in an extracellular substrate (the matrix) [44]. These are two distinct varieties of a 3D stem cell culture. OOCs are a way to enhance the functionality of the model. A manifest dimension of hybridity in OOCs, that is visible also in other products of bioengineering such as bioprinting, is the mix of biological material (living cells) and artificial components, such as human-made lattice. In that sense, there would be an important distinction between OOCs and organoids, and organoids would appear more natural in that they develop by themselves and do not contain artificial components. However, the "degree of naturalness" is itself a complex notion and the fact that organoids develop "by themselves" can be disputed (after all, their development requires the fine tuning of a complex chemical environment by researchers).

OOCs are used as clinical tools for screening and testing. In this sense, they are models of development (in contrast with bioprinting, which is using technology/a scaffold for building artificial organs). OOCs sometimes have better functionality than organoids but might not, in general, manifest exactly the same potentialities and properties. This is the reason why they deserve a specific attention in our typology, although the question of their moral status is one to be discussed later in the project. In other words, OOCs are entities that are distinct from organoids, and while we can already see that the two fields of research are overlapping but not

[44] The European Innovation Council 2021 Pathfinder Challenge made the distinction between *biological engineered living materials* and *hybrid living materials*: "Engineered living materials (ELMs) are composed, either entirely or partly, of living cells. ELMs entirely composed of living cells are called biological ELMs and they self-assemble via a bottom-up process – e.g. synthetic morphogenesis for organoids' production. ELMs only partly composed of living cells are called hybrid living materials (HLMs) and are built with a top-down process with integrated polymers or scaffolds. In both cases, the cellular components extract energy from the environment to form or assemble the material itself, and to adapt its morphology and function to environmental stimuli. This endows these materials with a combination of properties not present in any non-living material: self-regeneration, adaptation to environmental clues, longevity and environmental sustainability. By being alive, ELMs represent a fundamental change in materials' production and performance, enabling new, better or similar functionalities, compared to traditional materials but with decreased costs and environmental impact." European Innovation Council. (2021). *EIC Work Programme 2021* (p.61). European Commission. https://ec.europa.eu/info/funding-tenders/opportunities/docs/2021-2027/horizon/wp-call/2021/wp_horizon-eic-2021_en.pdf

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identical, at this point it remains too early to say whether there are essential differences between organoids and OOCs at the ethical/regulatory level.

➔ *Ethical issues:*

- *It remains to be determined whether there are specific ethical issues with OOCs that are not covered by a reflection on organoids.*
- *Issues in standardization, good manufacturing practice, etc. might be slightly different in the sense that the OOC includes some inert material (the "chip") and not only biological material.*
- *Far from clinical applications and inapt to transplants, OOCs might be even more susceptible to biomedical hype and false therapeutic expectations than organoids.*[45]

SPECIFIC QUESTION 9 Are organs-on-chip combined into complex organism-like systems?

BODY-ON-CHIP

OOCs of many organs of the body have been developed. When combined, these entities may exchange fluids in a set-up that makes possible a kind of blood-like circulation and body physiology. These "multi-organ body-on-chips systems"[46] form complex physiological models that might develop properties akin to an organism (or a subsystem of it), raising issues similar to ASSEMBLOIDS. They offer more realistic testing conditions for the absorption and distribution of drugs in the body than other models: in terms of scientific and clinical modeling, these systems are closer to physiological conditions, offering a possibility to bridge the gap between human cell-based but unrealistic cell cultures and animal models that are physiologically relevant but far from the specificities of the human species. The development of patients' "avatars" in vitro opens a perspective towards personalized diagnostic tools or treatments for a specific disease that are often advertised as "precision medicine" or "personalized medicine," although the responsibility of such a vision should be further assessed (see WP2).

➔ *Ethical issues:*

- *Ethical issues would be similar to the issues raised by assembloids. We would have to determine what kind of properties these entities precisely have.*
- *What would it take to replace animal models? What are the ethical odds regarding the balance between OOC development and animal models?*

[45] Mastrangeli, M., Millet, S., Mummery, C., Loskill, P., Braeken, D., Eberle, W., Cipriano, M., Fernandez, L., Graef, M., Gidrol, X., Picollet-D'Hahan, N., van Meer, B., Ochoa, I., Schutte, M., van den Eijnden-van Raaij, J. (2019). Building blocks for a European Organ-on-Chip roadmap. *ALTEX - Alternatives to Animal Experimentation*, *36*(3), 481–492. https://doi.org/10.14573/altex.1905221

[46] Ingber, D.E. (2022). Human organs-on-chips for disease modelling, drug development and personalized medicine. *Nature Reviews Genetics*. https://doi.org/10.1038/s41576-022-00466-9

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SPECIFIC QUESTION 10
Are stem cells not induced to differentiate into the model of one particular organ, but rather into all cell types, leading to an embryo model?

EMBRYO MODELS

Embryo models are 3D SC entities that replicate in vitro the early stages of development (not of a specific organ or type of tissue). The embryo can refer to entities from the first cleavage of the ovum or the implantation stage[47] until 8-9 weeks of development, that is, until all major structures appear, after that we have a foetus (for the human species at least). Different models correspond to different stages of development. There is no "morula model" as we are just dealing with a couple of cells (totipotent SC) with neither organization nor differentiation, but the following stages represent significant changes in the biological features of the embryo, with emerging properties at each stage, especially in terms of structure/architecture.[48] The blastocyst (around 5 days) is the first stage of development in which there is cell differentiation, with the distinction between the inner cell mass and the outer cells. At the stage of implantation, there is a separation of the embryo (from the former) and extra-embryonic tissues (e.g., placenta, from the latter). Then, during gastrulation (around 3 weeks), SC differentiate in the three germ layers and the axes of the body plan are forming (a step also known as symmetry breaking). Embryo models share (some of) the properties of the stages of development that they want to mimic.

➔ *Ethical issues:*

- *The 14-day rule applies to embryos cultivated in vitro, not to embryo models. It remains to be discussed whether a rule of this kind should be applied to these entities as well.*
- *Embryo models can be chimeric embryo models, which might raise further issues (see below).*

SPECIFIC QUESTION 11
Does the embryo model integrate extra-embryonic tissue?

BLASTOIDS

There could potentially be as many embryo models as there are stages of embryo development that biologists think are worth identifying. We will only consider here the criterion of the

[47] For a clarification, see the glossary in ISSCR Guidelines, op. cit., p.64-65.

[48] Note that these entities are isolated from the process of development, which is a continuum. D1.2 already commented on that. At this point, we could remark that we are looking for the emergence of notable properties, that will be the basis for the qualification for a sort of moral status, and that we have to highlight some breaking points as a consequence. Understanding these entities as part of a pure continuum will imply more efforts to mitigate the consequences for bioethics but that might not be impossible (see Meincke, A-S. (2018). Persons as biological processes. In D. Nicholson & J. Dupré (Eds.), *Everything flows: towards a processual philosophy of biology* (pp. 357-378). Oxford University Press. https://doi.org/10.1093/oso/9780198779636.001.0001).

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dissociation (or not) of extra-embryonic tissues from the embryo.[49] A blastoid is a model of the blastocyst, which includes tissues that will become the embryo properly speaking and extra-embryonic tissues. This is what the ISSCR guidelines call "integrated stem cell-based embryo models."

➔ *Ethical issues:*

- *If one follows the potential viewpoint, this entity has, in a way, the potential to become a fetus if cultured for additional time and transplanted in vivo. An integrated model has the potential to develop into a "full person" (while the non-integrated model cannot). The ISSCR guidelines suggest that this kind of model requires a specialized overview. It should also not be transplanted into a uterus. This can be justified for the same reasons that prevent reproductive cloning: those are research models, neither intended nor fit for reproductive purposes.*

SPECIFIC QUESTION 12 Does the embryo model *not* integrate any extra-embryonic tissue?

GASTRULOIDS

Gastruloids are models that replicate the embryo after gastrulation, with three germ layers, spatial organization, and a determined cell fate. The ISSCR guidelines labels these entities "non-integrated stem cell-based embryo models," because there are no extraembryonic tissues (or at least, "not all aspects of the peri-implantation embryo"). Such a model cannot lead to the development of a fetus. Some properties of the embryo (at the gastrulation stage) will emerge as replicas in the embryo model, but they are unlikely to be ethically problematic.

➔ *Ethical issues:*

- *For how long can an embryo model develop in culture? What kind of property could emerge if cultivated for a longer time?*

SPECIFIC QUESTION 13 Is the stem cell aggregate composed of cells with genetic material from two distinct species, especially a mix of human and animal stem cells?

CHIMERAS

[49] This distinction, introducing a major difference between blastoids and gastruloids, is particularly emphasized in the ISSCR guidelines. There is a strategic reason for that, because under this interpretation, gastruloids are not likely to raise the issue of organismal potential (which can be seen for some as a rebuttal to the development of these models). But if we do not take the potentiality argument for granted, it might be more interesting to look at the emerging structures and functions, and in that case gastruloids are obviously more complex than blastoids. But this complexity is not necessarily synonym of the emergence of ethical points of concern.

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A chimeric individual is an organism with genetic material from two distinct sources, in particular, sources from two distinct species (in the case of interspecies chimeras). Of specific interest or concern are chimeras that include human cells.

➔ *Ethical issues:*

- *Attention to risks of widespread chimerism, cell migration.*

SPECIFIC QUESTION 14 Are the chimeric models produced by mixing animal and human SC at the embryo level?

CHIMERIC EMBRYOS/ORGANOIDS

This concept concerns the introduction of human SC into animal embryos. The properties of chimeric embryos (and the ensuing ethical issues) may vary according to the degree of chimerism, or in other words, the proportion of human cells in the mix.

➔ *Ethical issues:*

- *Potential implantation/gestation of chimeric embryos.*
- *Specific case of chimeras with non-human primates.*

SPECIFIC QUESTION 15 Are the chimeric models produced by grafting SC derivatives in a postnatal animal host?

ANIMAL RECIPIENTS OF HUMAN ORGANOIDS

Transfer of organoids derived from human SC into animal hosts.

➔ *Ethical issues:*

- *The rules of animal research ethics still apply. Consideration of the best standards of animal research ethics.*
- *In the case of brain organoid grafts, possible induction of behavioral/cognitive change: to be assessed and monitored. To what extent does it produce the intended effects (according to the purpose of the model)? To what extent does it alter animal welfare?*
- *Putative enhancement of the animal host: how to deal with it? Is there a risk in the development of "humanized animal models"?*
- *What about non-human primates as animal hosts?*
- *Chimeras that might produce human gametes and their ability to breed.*

SPECIFIC QUESTION 16 Are the cells used cancer stem cells?

TUMOROIDS

Tumoroids are in vitro entities derived from tumors, starting from a biopsy sample removed from a patient. As tools for drug testing, they offer an alternative to 2D cultures and mouse models. Tumoroids are a major tool of personalized oncology in that they offer the possibility to develop a complex model of an individual patient's tumor. A tumoroid is thus a SC-derived entity that shares some properties with the tumor of origin, allowing, for example, individual drug screening.

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A pending question is the extent to which a given tumoroid is a "good" model of a given patient's tumor. While tumoroids offer an interesting option to analyze the tumor as an individual, tumor heterogeneity and the development procedures for growing organoids represent major challenges. The development of tumor biobanks containing a representative sample of the genetic variations of tumors is also a prerequisite to any systematic application in health care as *individual* screening means relying on a large amount of *collective* data (e.g., molecular, genetic).[50]

→ *Ethical issues:*

- *Obviously, tumoroids are not candidates for a particular moral status such as blastoids or brain organoids. They raise nonetheless issues related to the emerging field of "personalized medicine" in general (e.g., statistical assessment of drug efficacy, issues in reimbursement when dealing with individual evidence).*
- *How could this research be transferred to clinics on a massive scale and how to guarantee a fair allocation of resources in the medical paradigm shift? There would be a tension between positive research prospects in the long run and immediate clinical outcomes: scarcity of the biological material that has to be used for tumoroid development and many analyses (genetic, screening…), the difficulty to grow enough tumoroid material in a time window that is clinically relevant, the issue of involving patients who may take risks while not be able to benefit the treatment, etc.*

5 Concluding thoughts

The draft of this document is the result of a long-standing discussion with many partners in the HYBRIDA project. Several options have been considered for the graphic representation that goes with the text of the typology. While the flowchart is useful in its function of guidance, a more compact representation of the typology as a compass rose was envisioned at first. A compass rose is indeed a navigation tool, a guide for general directions. It would have told us in a snapshot what distinct entities we have in organoid research and what are their respective positions in the field. Without being equivalent to a detailed map of a territory, a compass gives directions and a direction leads somewhere (once one chooses a direction, there are consequences: there are roads, we can follow them or not, bifurcate…). In this sense, different concepts would be represented by different directions on the map: if research goes this way, it will develop the entities X, which will raise issues A and B, but if research goes that way, it will develop the entities Y which raise issues C and D, and so on. The reader can find here a representation of the typology as a compass rose (Figure 3).

[50] Green, S., Dam, M.S., Svendsen, M.N. (2022). Patient-derived organoids in precision oncology – towards a science of and for the individual? In C. Beneduce, M. Bertolaso (Eds.), *Personalized medicine in the making: philosophical perspectives from biology to healthcare* (pp. 125-146). Springer. https://doi.org/10.1007/978-3-030-74804-3

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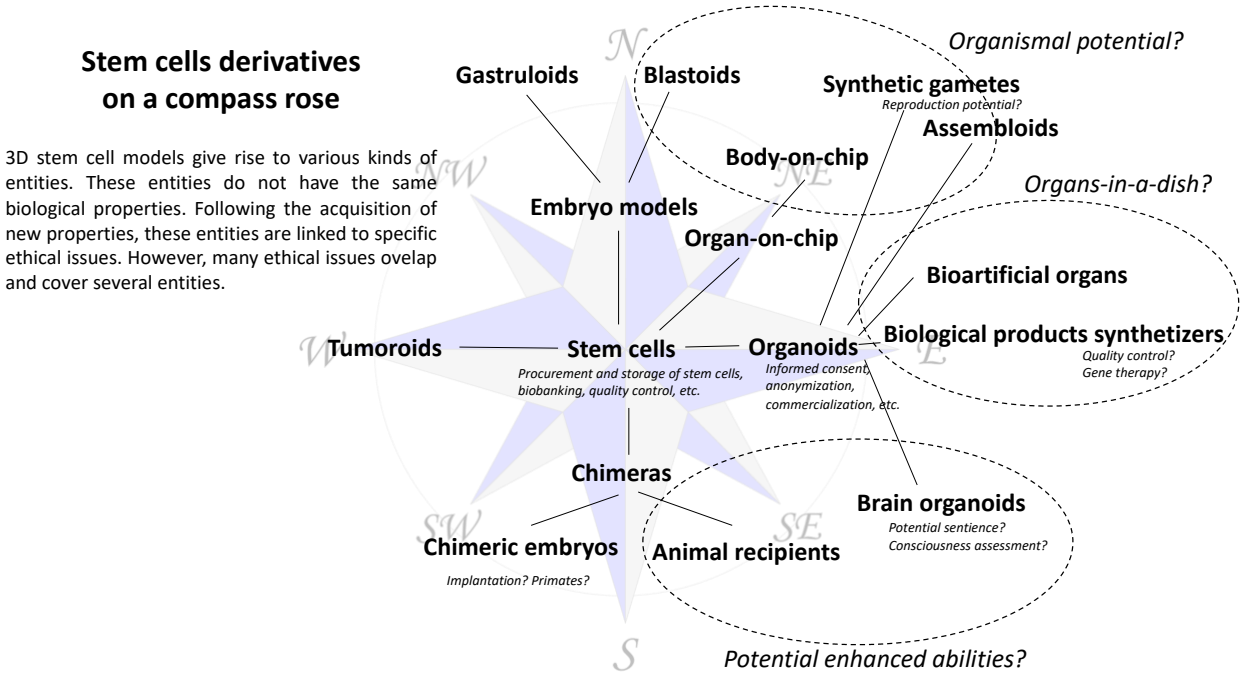
Figure 3: Stem cells derivatives on a compass rose



We have to be careful that the spatial layout may be misleading as well. At times, spatial proximity between entities in the diagram implies that some entities raise similar issues, such as that blastoids and assembloids both have something like a potential to be developed into an organism; while elsewhere the spatial location is purely arbitrary (tumoroids are not the "opposite" of organoids, chimeras are not "closer" to organoids than to embryo models…). Such issues are still pending issues that deserve to be discussed in details. This is also a limitation of the flowchart: the reader should not be inclined to think that the specific questions raised at each branch is to be taken in isolation, or that specificity equates singularity. Some of these entities might be regrouped when they deserve similar treatment, and when they are put to use in the same way. Other could be divided again into more specific entities, with even more specific questions. Taking this into consideration would require a complete assessment of each entity and a list of all its issues of concern. This work begins precisely where the work on the typology stops. The typology is aimed at fostering debate and offer a guidance to explore ethical issues raised in many directions by the biotechnologies that we discuss here, it cannot offer a complete assessment and discussion of these issues.

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