All information regarding future IHI Call topics is indicative and subject to change. Final information about future IHI Calls will be communicated after approval by the IHI Governing Board.

Topic 1: Expanding translational knowledge in minipigs: a path to reduce and replace non-human primates in non-clinical safety assessment

Expected impacts to be achieved by this topic

EU legislation[1] makes it a legal obligation to replace, reduce and refine the use of animals in research (the '3Rs' principle), including a specific focus on restricting the use of the non-human primates (NHPs) unless scientifically justified. The development of *in vitro* models for human safety assessment is still challenging due to complex biological responses in various organ systems following drug treatment. Therefore, laboratory animals will still be requested in the safety testing of new therapeutics and innovative medical technologies until non-animal approaches have reached the necessary level of maturity and validation to ensure that only safe treatments reach patients, and that patients get timely access to the most innovative therapeutics.

A substantial amount of work has already been conducted to increase the scientific knowledge and understanding of the role of minipigs in toxicity testing[2] and the pig is often used e.g., in the toxicological evaluation of small molecules. Replacing NHPs with minipigs in the safety testing of new therapeutic modalities has been, however, more difficult, due to the lack of translational knowledge, but will be an important ethical step towards minimising the use of NHPs. New drug modalities are often designed to engage human targets with high specificity, which is the rationale for selecting NHPs in the safety testing of this kind of drug candidates. By expanding the translational knowledge in minipigs versus NHPs and humans, the scientific justification for selecting pigs as an alternative to NHPs can be improved.

The project funded under this topic adheres to the principles of the 3Rs by: i) closing the current translational knowledge gaps regarding minipigs versus NHPs and humans, offering the opportunity to replace NHPs with pigs, improve the reproducibility of pig studies, and advance the underlying knowledge of biological processes to facilitate the development of non-animal alternatives (reduce, refine and replace); ii) creating scientific and technological opportunities in animal housing facilities to collect, digitalise and generate more reproducible data in freely moving, undisturbed animals with the potential to reduce the total number of animals, and improve animal welfare and data quality (reduce, refine).

Closing the translational knowledge gap regarding minipigs versus NHPs and humans will enable the development of new, refined, and digital research tools, which will contribute to:

- reducing and replacing the overall number of NHPs in research without compromising human safety.
- improving disease understanding that will open up new research pathways, and enhanced use of non-invasive digital technologies that can improve animal welfare (refinement), and furthermore, are potentially applicable to humans.

[1] DIRECTIVE 2010/63/EU OF THE EUROPEAN PARLIAMENT AND OF THE COUNCIL of 22 September 2010 on the protection of animals used for scientific purposes; https://eur-lex.europa.eu/legal-content/EN/TXT/?uri=celex%3A32010L0063
[2] The RETHINK project on minipigs in the toxicity testing of new medicines and chemicals: Conclusions and recommendations. https://doi.org/10.1016/j.vascn.2010.05.008

- improving the sustainability and quality of biomedical research and development (R&D) in areas of unmet medical need by ensuring access to well-characterised minipig models in R&D of new therapeutics and innovative medical technologies.
- optimising knowledge sharing between academia, regulators, and the health care industry to accelerate the generation of knowledge and medical innovation.
- fostering the development and validation of non-animal models and approaches by implementing translational data obtained in the future project, which could pave the way to such models. Data generation will be based on early discussions with regulatory authorities and academic partners, thereby ensuring the contribution to the development and validation of non-animal approaches.

Expected outcomes

- Obtain and share biological knowledge of minipigs, thereby facilitating the development of innovative solutions by improving the translational understanding between minipigs *versus* NHPs and humans, including further understanding of the minipig immune system, with the overall aim to replace, reduce and refine the use of animals in non-clinical safety assessment.
- A regulatory pathway for nonclinical safety assessment of biologicals and other new therapeutic modalities in minipigs with the potential to impact regulatory strategies.
- Publicly available databases and software for physiological, genomic, transcriptomic, metabolomic, proteomic and epigenetic minipig data to understand underlying mechanisms of disease/toxicities and find new mode of actions for pharmaceutical interventions.
- Characterised and validated genetically modified minipig models:
  - genetically modified minipig models based on the CRISPR/Cas9 gene-editing technology.
  - minipigs with ‘humanised’ immune system components and effectors for testing biologicals.
  - small-sized micropig for efficacy/safety assessment to facilitate compound availability in pharmaceutical R&D.
- Assessment of the utility of the minipig as a relevant toxicology species for immuno-safety testing using therapeutics which have been tested preclinically and clinically. Assisting and synergising the already existing translational and regulatory efforts related to immunological safety evaluation. Developing validated antibodies and *in vitro* immunoassays to characterise the immune system and assess the immuno-safety of therapeutics in minipigs.
- Minipig-specific technology for automated study data: validated medical devices, biosensors, algorithms, software, and digital animal housing. Machine learning and artificial intelligence (AI)-based tools to monitor abnormalities in behaviour and physiological systems in undisturbed animals.

To ensure long-term sustainability, all the interdisciplinary science-based knowledge obtained and generated in the project arising from this topic will be shared, integrated, digitalised, and published in peer-reviewed journals, encouraging industry and academia to develop innovative medical science solutions and technologies, such as scientifically and ethically sound animal models, assays, biomarkers, monitoring devices, biosensors for normal physiological behaviour, and algorithms. Based on the close collaboration with regulatory bodies, the knowledge generated in the project is further expected to impact regulatory guideline strategies. All outputs will require long-term sustainability and maintenance to fulfil the scope of the project.

Scope

Challenges

- Increasing need to find alternatives to testing in NHPs in line with EU legislation.
- Almost no precedence in minipig use for safety testing of biologicals and new therapeutic modalities [e.g., oligonucleotides, small interfering RNAs (SiRNAs), crystallisable fragments (Fcs), antigen-binding fragments (Fabs), single-chain variable fragments (scFvs), monoclonal antibodies (mAbs), vaccines, gene-editing and cell-based therapies].
- Lack of scientific knowledge to scientifically justify a de-selection of NHPs in the non-clinical safety assessment of new therapeutics. Lack of public minipig reference 'omics' with good quality annotation: Full genome sequencing, in parallel with baseline transcriptomics, proteomics, metabolomics and epigenetic information.
- Lack of 'humanised' and genetically modified models available for efficacy/safety testing, including genetically modified smaller micropigs to address cases of limited substance supply.
- Significant knowledge gap on the minipig immune system and reduced number of laboratory tools and reagents when compared to other toxicology species (rodent and non-rodent).
- Lack of widespread use of biosensors, medical devices, 'intelligent' animal housing for automated data collection and analysis in minipig studies.

Objectives

The overall objective of this topic is to characterise the minipig for use in R&D of new therapeutics and innovative medical technologies. The knowledge generated in this proposal may facilitate innovative health solutions and improve disease understanding and human predictions. The goal is to advance biomedical R&D by generating background scientific data to evaluate if the minipigs could be a viable and feasible alternative to NHPs in key therapeutic areas, with a special focus on translatability from minipigs to humans.

Key activities

Compile and publish existing historical safety data in minipig biomedical R&D and discuss data with regulators.

- Evaluate the translatability of minipigs in human risk assessment following treatment with biologicals and new therapeutic modalities, and discuss future perspectives of the minipigs with regulatory agencies, e.g., by requesting regulatory interactions with European Medicines Agency (EMA) such as scientific advice and/or novel methodology qualification advice to understand possible regulatory hurdles in using minipigs for safety assessment.
- Minipigs multi-omics and imaging: Generate omics reference data (genomics, transcriptomics, proteomics, metabolomics, and epigenetic information) to enable translational research in minipigs. To further characterise the minipig, imaging technologies such as magnetic resonance imaging (MRI), computed tomography (CT) scans and positron emission tomography (PET) scans are also of interest.
- Genetically modified pig models including the micro-pig: Characterise and validate humanised and genetically modified minipig models, including the micropig to generate translatable animal models in non-clinical safety assessment.
- iPig: Digital technologies, clinical data collection and AI: Create, validate, qualify, and benchmark digital solutions that can objectively measure clinically relevant and functional biomarkers in minipigs for use in preclinical toxicity studies in line with the regulatory agencies' requirements.

- Minipig immune system: validate reagents, assays, and biomarkers for immunological investigations: Conduct investigative studies in minipigs to support their translational significance in immuno-safety assessments and validate reagents/assays.
- Project management: Compile, digitalise, and publish existing and newly-produced data.

Why the expected outcomes can only be achieved by an IHI project

Generating and compiling comprehensive and complex biomedical datasets within various therapy areas, some of which will be for AI purposes, requires the involvement of multidisciplinary skills across several industry sectors (pharmaceuticals, medical technologies, biotech, vaccines, etc.) including small and medium-sized Enterprises. Previous examples of precompetitive public-private projects (SAFE-T and eTRANSAFE) within the Innovative Medicines Initiative (IMI) and private multi-company initiatives (such as BioCelerate) demonstrated the value of a neutral broker to facilitate precompetitive sharing of proprietary information. Expanding such collaborations beyond one sector to integrate tools, data and know-how from the technology and biotechnology sectors, and joining forces with academic partners from various sectors in unprecedented collaborations, requires exploring new precompetitive grounds and calls for this neutral brokerage to continue.

The involvement of regulatory authorities at all stages of the project generated by this topic is essential considering its objective to develop alternatives that can be used to generate data for regulatory purposes. Close collaboration will contribute to accelerating the development of new knowledge; align validation processes with regulatory requirements; and ultimately, lead to the implementation of new solutions in regulatory practice and their deployment in research practice.

Pre-identified industry consortium and contributing partner(s)

The pre-identified industry consortium that will contribute to this cross-sectoral IHI proposal is composed of the following industry partners:

Pharmaceutical/biotech/vaccine companies:
- Bayer
- Boehringer Ingelheim
- Bristol Myers Squibb
- Lundbeck
- Merck KGaA
- Novo Nordisk (Lead)
- Novartis
- Pfizer
- Roche
- Sanofi

Other companies:
- LabCorp
- Charles River

In addition, the following contributing partners will participate to the IHI project:
- VeriSim Life
- JDRF

In the spirit of partnership, and to reflect how IHI two-stage call topics are built upon identified scientific priorities agreed together with a number of proposing industrial beneficiaries, it is envisaged that IHI proposals and projects may allocate a leading role within the consortium to an industrial beneficiary. Within an applicant consortium discussing the full proposal to be submitted for the second stage, it is expected that one of the industrial beneficiaries may become the coordinator or the project leader. Therefore, to facilitate

the formation of the final consortium, all beneficiaries are encouraged to discuss the weighting of responsibilities and priorities with regards to such leadership roles. Until such roles are formalised by execution of the Grant Agreement, one of the proposing industrial leaders shall facilitate as project leader an efficient drafting and negotiation of project content and required agreements.

Indicative budget

The maximum financial contribution from IHI is up to EUR 8 500 000.

The indicative in-kind contribution from industry partners is in total EUR 8 910 000.

The indicative in-kind and financial contribution from IHI JU contributing partners is EUR 492 000.

Due to the global nature of the participating industry partners, it is anticipated that some elements of the contributions will be in-kind contributions to operational activities from those countries that are not part of the EU nor associated to the Horizon Europe programme.

The indicative in-kind contribution from industry partners may include in-kind contributions to additional activities.

Indicative duration of the action

The indicative duration of the action is 60 months.

This duration is indicative only. At the second stage, the consortium selected at the first stage and the pre-identified industry consortium and contributing partners may jointly agree on a different duration when submitting the full proposal.

Phase 1: Evaluation of existing minipig data, develop databases, develop bio sensors and algorithms. Data will be published in peer reviewed journals. Knowledge gaps will be identified, and the development of minipig models will be initiated. Molecule selection and investigations for Phase 2 will be planned and slots for the studies will be booked.

Phase 2: Adolescent and adult male and female minipigs will be treated with modalities e.g., oligos, SiRNAs, Fcs, Fabs, scFvs, mAbs, vaccines, gene-editing, or cell-based therapies, as an alternative to the current precedence of safety testing in NHPs.

Phase 3: Biomaterial from the minipig studies in Phase 2 will be distributed to various work package members (iPig, multi-omics, immuno-safety) for further evaluation. Mechanisms and translational aspects will be explored.

Phase 4: Database scrutinisation, compile, discuss and distribute new knowledge, publication in peer reviewed journals, propose regulatory recommendations, and promote digital solutions.

Contribution of the pre-identified industry consortium and contributing partners

The pre-identified industry consortium and contributing partners expect to contribute to the IHI proposal by providing the following expertise and assets:

- Experimental settings: Pharmaceutical drug candidates, drug products, animals including genetically modified animals, animal units, experimental equipment, laboratories.
- Data: access to standard toxicology and clinical safety endpoints, historical data, gene expression, immunosafety biomarkers and assays.
- Expertise: nonclinical expertise, data science, regulatory expertise, immunosafety, ‘omics’ evaluation, disease models, devices.

- Technology: Standard for Exchange of Nonclinical (SEND) databases and SEND visualisation systems, implants, device software.

The allocation of the EUR 200 000 financial contribution will be decided by the full consortium at the second stage when preparing the full proposal.

Applicant consortium

Public partners:

- Database constructors: merging large databases from different sectors (various public and industry partners) containing complex biological datasets e.g., genomic, transcriptomic, metabolomic, proteomic and epigenetic data.
- Suppliers of genetically modified minipigs and tissue samples.
- Partners experienced with, and suppliers of, MRI, CT and PET scanning in pigs.
- Academic partners developing and validating biomarkers to ensure human translatability.
- Inventors of technologies for automated digital data collection in patients and pigs: validated medical devices and biosensors to measure normal physiological behaviour.
- Inventors of validated algorithmic tools for machine learning and artificial intelligence for automated digital animal housing and prediction of toxicities in minipig vs. human.
- Inventors of validated antibodies and *in vitro* immunoassays to characterise the immune system and assess immuno-safety of therapeutics in minipigs.
- Project administration with experience in public-private partnerships.

Regulatory authorities: Advisors.

Dissemination and exploitation obligations

The specific obligations described in the Conditions of the calls and call management rules under ‘Specific conditions on availability, accessibility and affordability’ do not apply.