STAR-IDAZ
International Research
Consortium on Animal Health

Research Roadmap Development for Alternatives to Antibiotics

Report 2022

The Secretariat for the STAR-IDAZ IRC (SIRCAH) is funded from the European Union's Horizon 2020 research and innovation programme under grant agreement No 727494

Acknowledgments

This report has been produced by

STAR-IDAZ International Research Consortium on Animal Health

In collaboration with

IDRC · CRDI
International Development Research Centre
Centre de recherches pour le développement international

Moreover, we wish to thank each of the following experts, who participated in the process for roadmap development, without their expertise and efforts this report could not have been produced:

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Abstract

The document highlights research needs for developing innovative Alternative to Antibiotics (ATA) discussed during the STAR-IDAZ IRC workshops for the development of research roadmaps along the period 2019-2022.

Research roadmaps support researchers and funders to visualize complex systems and focus research on where it is most needed.

Contents

Introduction

STAR-IDAZ International Research Consortium (IRC) is a global initiative to address the coordination of research programmes at an international level in the area of animal health and in particular infectious animal diseases including zoonoses (STAR-IDAZ – Global Strategic Alliances for the Coordination of Research on the Major Infectious Diseases of Animals and Zoonoses) – for details see **http://www.star-idaz.net/**.

Antimicrobial resistance (AMR) and the Development of Innovative Alternatives to Antibiotics (ATA) was identified by STAR-IDAZ partners as one of the priority issues for collaborative activities.

Roadmapping alternatives to antibiotics

STAR-IDAZ aims to advance a global research agenda on the development of alternatives to antibiotics (ATA) by exploring experts' views on research priorities. Several workshops were held from 2019 to 2022 to develop research roadmaps on ATA.

The workshops built on the outputs of earlier expert engagement starting with a gap analysis workshop held alongside the *3rd International Symposium, Alternatives to Antibiotics, Challenges and Solutions in Animal Health and Production*, Bangkok, Thailand, on December 16 -18, 2019 after which there was electronic engagement leading to three webinars in October/November 2020 when the development of roadmaps was initiated.

Experts were initially invited to comment on a draft overarching logical framework defining the goal, purpose, outputs and activities of the working group (**Annex VII: Logical Framework**). The overall goal was determined as 'development of alternatives to antibiotics and to reduce/rationalise the use of medically important antimicrobials in livestock so as to safeguard their effectiveness, decreasing the level of use in livestock and the development of resistance to antibiotics, while maintaining/enhancing production levels, and control of animal diseases'.

Based on the results of the ATA symposium, the working group of ATA experts focused on highlighting promising research results and novel technologies that provide alternatives to antibiotics for use in animal health and production, assessing challenges associated with their development and commercialisation. The symposium focused on five product categories that could reduce the use of medically important antibiotics in animal health and production: 1) vaccines; 2) microbial derived products; 3) phytochemicals; 4) immune-derived products; and 5) innovative drugs, chemicals and enzymes. Alternatives to antibiotics were broadly defined as any substance that can be substituted for therapeutic drugs that are increasingly becoming ineffective against pathogenic bacteria, viruses or parasites.

The subsequent research gap-analysis workshops focused on phage technology, immunomodulators, the microbiome and how it might be manipulated and how antibiotics promoted growth. Vaccines chemotherapeutics (including phytochemicals and antimicrobial peptides) and disease control were not considered in details, as these are covered by the **vaccine**, **therapeutics** development and **disease control** research roadmaps respectively.

In particular, for each of the key areas discussed, what we are trying to achieve was agreed in terms of specific target product profiles. Working back from these targets, the challenges in delivering them were discussed and the possible solution routes and their dependencies were identified. A prioritisation of research priorities was also conducted during the final workshops (see Table 1).

In October and November 2021, **STAR IDAZ IRC** and **IDRC** (International Development Research Centre, Canada) organized 5 on-line workshops on Alternatives to Antibiotics (ATA) (see Annex I,II, III, IV and V). More than 60 experts from 15 countries across the world actively participated in those workshops to develop research roadmaps by identifying key areas of research to develop alternative to antibiotics in livestock production and discuss current challenges.

A number of possible approaches targeting either the pathogen, the host, or both were considered, without covering in detail genome/genetic editing, which was outside of the WG scope. While in a separate workshop held in October 2021 'Antibiotics as growth promoters: research needs on how antibiotics work as growth promoters' (Annex I) were discussed.

Research roadmaps identifying the critical knowledge gaps needing to be addressed to deliver a range of possible alternatives to antibiotics, targeting either the pathogen or the host or both, were finalised into the following 3 workshops:

1. **Phage technologies: Alternatives to antibiotics acting directly on the pathogen, including establishing their mode of action- 12 October 2021 (Annex II)**
2. **Immunomodulators: Agents and compounds for their ability to enhance the hosts resistance to disease, including establishing their mode of action – 15 October 2021 (Annex III)**
3. **The role of the microbiome in the maintenance of health, and how it can be manipulated- 18 October 2021 (Annex IV)**

A fifth and final workshop, was then organised to discuss current challenges and opportunities for **'Taking new alternatives to antibiotics to market'** 9 November 2021.

After the workshops, the roadmaps were revised by electronic consultations and an overarching roadmap was proposed to link the different research roadmaps already published (e.g. **vaccine**) or drafted during the workshops (e.g. Phages, Immunomodulators and Microbiome/microbiota optimisation) (Figure 1). This overarching roadmap also highlights transversal aspects of testing and tools for all ATA including: safety, delivery routes, delivery platforms and efficacy in challenge models.

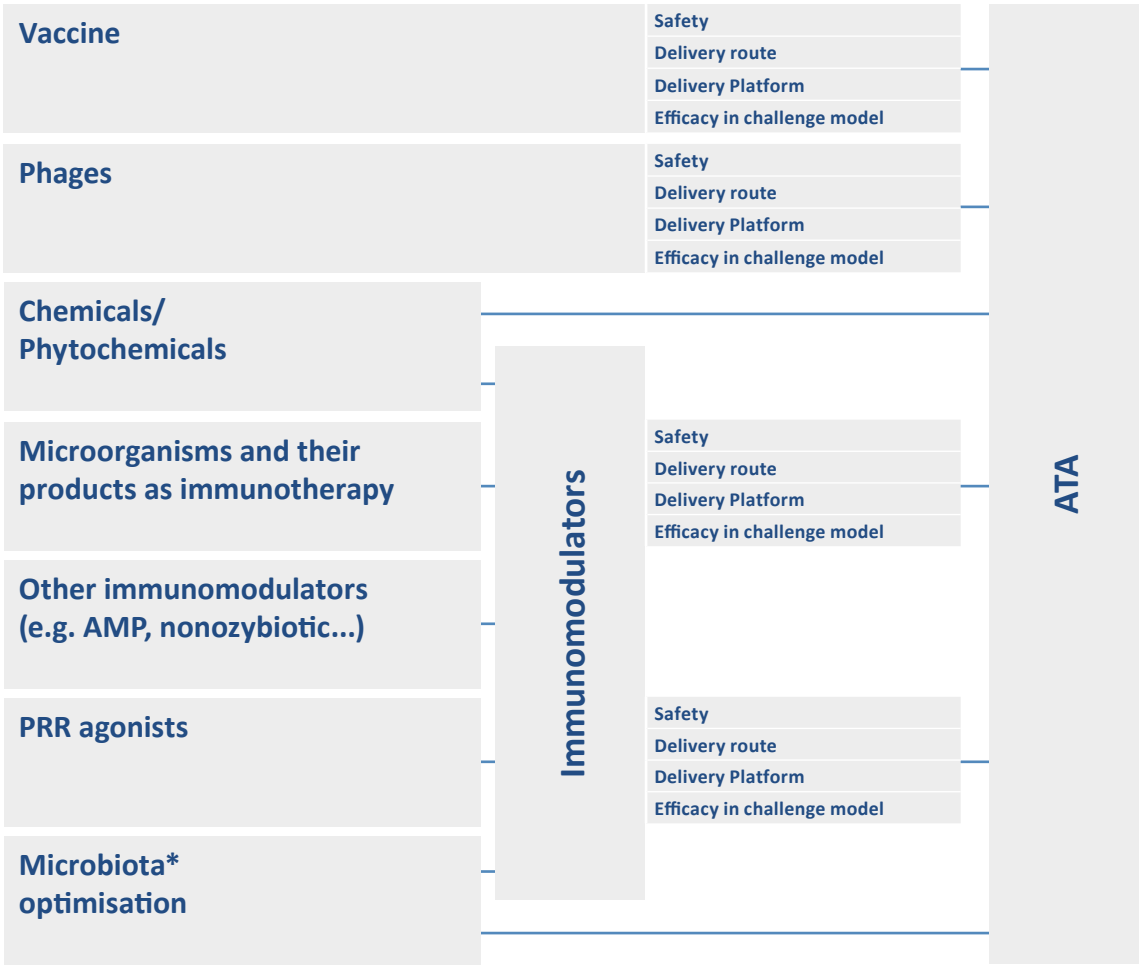


Figure a: Diagram representing the overarching roadmap for ATA linked to others roadmaps developed or under constructions

Main research priorities

During each workshop the research priorities within the main topics discussed were identified by experts utilising an on-line toll for voting. In the table below, the main research priorities identified are summarised (Table 1).

Table 1: Main research priorities identified

Topic:	Research priorities
Mechanisms behind antibiotics as growth promoters:	• Understand mechanisms of how AB work as growth promoters, to develop other alternatives • Create appropriate in-vivo/ex-vivo/in-vitro models • Basic research to better characterise microbiota • Defined standardized methods to test mechanism of subAbx and defined goal (growth vs feed conversion rate)
Phage technologies:	• Phage-bacteria interaction • In-vivo models and trials • Investigate phage survival in the animal and in the environment • Synthetic biology for retargetable phage-based platforms • Interaction between phage and the immune system • High throughput screening platforms for phage isolation/characterisation
Immunomodulators:	• Understand interaction between immune responses and inflammation • Mechanisms of host-microbial interaction • Kinetics and quantification of innate response stimulated by immunomodulators or by vaccines (non-specifically) • Functional studies of microbiota • Clearly define desired outcomes and best practices in testing immunomodulators
Microbiome:	• Increase knowledge on 'the microbiome', particulrly in different production forms/age-groups • Understanding mode of action of effective probiotics • Functional studies on the microbiome, linking taxonomy with function • Determine the impact microbiome shaping on vaccine efficacy and basic metabolic turnover

The next steps will be to validate the research roadmaps and the prioritised research gaps identified at the workshops. The roadmaps will be published on the **dedicated area** of the STAR-IDAZ IRC website, together with a recommendation on key research priorities areas.

Annex I: Antibiotics as growth promoters: research needs on how antibiotics work as growth promoters

Workshop 22 October 2021

Knowing how antibiotics work as growth promoters can help in finding alternatives to antibiotics as growth promoters. The overall goal remains to reduce the use of antibiotics in livestock production, decreasing the development of resistance to antimicrobials in livestock, while maintaining/enhancing production levels (increasing feed conversion rates or increasing food intake or both) and controlling the occurrence of disease. A number of options for the control of disease targeting either the pathogen or the host or both are possible while the basis by which antibiotics promote enhanced growth also needs to be investigated.

A questionnaire was developed and shared with a group of selected experts to explore four main areas: Questions 1 and 2 aimed at identifying the sort of products required and the associated research needs while Questions 3 and 4 focused on scientific areas where development could help in reducing the need to use antimicrobials (mode of action of growth promoters and microbiota). Based on the inputs collected through the survey, a deeper exploration into the research needs was conducted at a workshop that was held in Bangkok, immediately after the 3rd Alternatives to Symposium (19 December 2019). Expert contributions received both electronically and during the meeting were merged in the consolidated report.

The document below was revised during a final workshop the 22 October 2021 and provides a framework for identifying the critical gaps in knowledge needed to understand how antibiotics act as growth promoters, and to help sustain and better coordinate research on the area.

a) Research needs to establish how antimicrobials work as growth promoters

	What areas need to be considered to identify the role of antimicrobials in promoting growth - what are the research questions?	What are the main research needs requiring to be addressed to establish the role of antimicrobials in promoting growth?	What are the possible solution routes to establish the role of antimicrobials in promoting growth?	What else needs to be considered before we can solve this question
1	**Identifying the role of antimicrobials in promoting growth.**	Identify bio-markers (tissue or peripheral ones) for effect of AB. Characterise interactions between host, microbiome and low antimicrobials concentrations. Identify the antimicrobials and dosages that have the best growth-promoting effects. Understanding microbiota situation. Understanding correlation with diseases and herd management level - benefits less apparent when good disease management. Important to know which organisms affected - Broad vs Narrow spectrum antimicrobials. Identify the different effect of antimicrobials on diverse species and stages of development of animal.	Use OMICs to understand host responses (e.g. metabolomics, metaproteomics, glycomics) and impact on microbiome (shotgun metagenomics, metabolomics). Determine host immune response to different antimicrobials. Assess the growth-promoting effects of different antibiotic drugs and dosages. Impact of different antimicrobials on clostridia. (Impact of feed additive eg. copper and zinc in feed-investigation on how they are used) Determine how metals function as antimicrobials and their resistance development process – they also have some effect on the immune system. Patent search for Infection Prevention (IP)	2a - Investigating the impact of antimicrobials on gut microbiota. 2b - Investigating the impact of antimicrobials on metabolic processes. 2c - Investigating the impact of antimicrobials on immune system and inflammation. 2d - Investigating the impact on subclinical infections. 2e - Investigating how antimicrobials modulate animal environment microbiota (i.e. litter). 2f - Defining the role of antimicrobials on gut epithelial homeostasis. 2 g - Defining the differential impact of different antimicrobials as growth promoters.

	What areas need to be considered to identify the role of antimicrobials in promoting growth - what are the research questions?	What are the main research needs requiring to be addressed to establish the role of antimicrobials in promoting growth?	What are the possible solution routes to establish the role of antimicrobials in promoting growth?	What else needs to be considered before we can solve this question
2a	**Investigating the impact of antimicrobials on gut microbiota.**	Establish if normal commensals or protozoa or parasites are reduced as collateral damage and the advantage/ disadvantages resulting from this. Investigating antimicrobials and feed competition routes. Understand how very low antibiotic concentrations alter the structure of the commensal microbiome. Demonstrate cause-effect relationships between antibiotic-induced microbiome structures and health. Develop alternative methods to induce and maintain 'healthy' microbiome structures (e.g. diet, pre/probiotics, faecal transplantation, improving animal husbandry, reducing stress, etc.) Investigate correlation between stress and dysbiosis. Impact of antimicrobials on (the resistome) mobile genetic elements and their ecology.	Examine many of the normal protozoa and bacteria for their sensitivity to antimicrobials. Validate hypotheses generated by omics studies by microbiome modulation studies or by population studies correlating microbiome structure to growth/health. Metagenomic analysis and transcriptomics and proteomics to understand what is happening to the system - Establishing how the whole system is responding and not just 16s profiling but looking at the pathways and what is happening to the physiology of the bacteria - Host and bacteria multi-omics. Develop analytical methods for quantification of antimicrobials in the gut (pharmacokinetics, activity, etc..).	Better characterisation of microbiota especially for anaerobes - because when we look at the microbiota, the number of OTUs or hits that have no characterisation or cannot be tracked to any organism, is a major shortcoming – we need to be able to characterise the microbial members (especially anaerobes) and their function, and also the phageome of the gut. Microorganism physiology. Strategies to incorporate antimicrobials in feeds and stability Delivery systems for protection and controlled release of antimicrobials

	What areas need to be considered to identify the role of antimicrobials in promoting growth - what are the research questions?	What are the main research needs requiring to be addressed to establish the role of antimicrobials in promoting growth?	What are the possible solution routes to establish the role of antimicrobials in promoting growth?	What else needs to be considered before we can solve this question
2b	**Investigating the impact of antimicrobials on metabolic processes.**	Establish standardised methodologies and tissues to study host responses to antimicrobials. Understanding the effect of antimicrobials on different body/system tissues (e.g. liver and gut) in relation to metabolism.	Comparative studies on the physiological effect of different antimicrobials. Toxicity studies of new antimicrobials using both in vitro and in vivo models. Look at which metabolites (in blood) are produced in antimicrobial-treated animals compared to controls. Tissue transcriptomic. Impact of antimicrobials on host versus microbiota metabolic processes and use of gnotobiotic animals looking at the effect of AB out with the context of the microbiota or in conjunction with the microbiota. Kinome analysis.	

	What areas need to be considered to identify the role of antimicrobials in promoting growth - what are the research questions?	What are the main research needs requiring to be addressed to establish the role of antimicrobials in promoting growth?	What are the possible solution routes to establish the role of antimicrobials in promoting growth?	What else needs to be considered before we can solve this question
2c	**Investigating the impact of antimicrobials on immune system and inflammation.**	Use standardised methodologies and tissues, cell types to study host responses to antimicrobials. Identify reagents (e.g. if looking at protein expression there is a lack of well-defined target antibodies) Investigating direct vs indirect effects of antimicrobials on the immune system and inflammation Understanding the linkages between normal flora and inflammation.	Gene/protein expression in immune/ inflammatory cells from gut, systemically and in epithelial cells. Need to focus on the cell types (e.g. neutrophils) involved rather than the tissues	Available reagents and annotation of genomes. Share resources (networking, increasing trust).

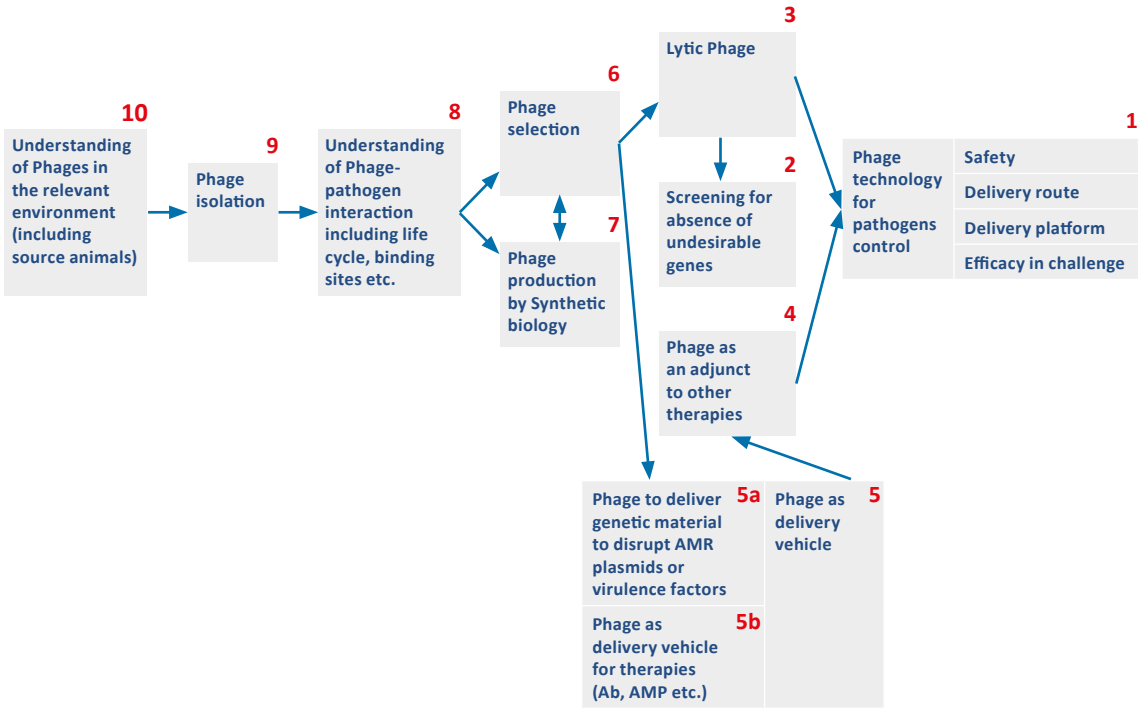
	What areas need to be considered to identify the role of antimicrobials in promoting growth - what are the research questions?	What are the main research needs requiring to be addressed to establish the role of antimicrobials in promoting growth?	What are the possible solution routes to establish the role of antimicrobials in promoting growth?	What else needs to be considered before we can solve this question
2d	**Investigating the impact on subclinical infections.**	Establishing if there is compartmentalisation of resources so if less immune system activation resources can go to growth. Establishing a definition of subclinical infection. Understanding the impact of subclinical/ clinical infection. Establishing if reduced subclinical infections results in improved gut function.	Measurement (Respiration) chambers (lack of these for large animals) to measure energy expenditure and challenge the animals. Observational studies on aquatic animals and poultry (large populations) under various levels of antimicrobials in the environment and infection levels. Comparison gnotobiotic animals and SPF animals and animals with reconstitute microbiota. Mock communities. Develop and characterize animal models to simulate specific infections.	3a - Identifying key limiting nutritional components for maintaining growth and immune functions.
2e	**Investigating how antimicrobials modulate animal environment microbiota (i.e. litter).**	Establishing the modulation of antimicrobials in environmental microbiota and later impact in animal gut microbiome.	Observational studies at farm level	

	What areas need to be considered to identify the role of antimicrobials in promoting growth - what are the research questions?	What are the main research needs requiring to be addressed to establish the role of antimicrobials in promoting growth?	What are the possible solution routes to establish the role of antimicrobials in promoting growth?	What else needs to be considered before we can solve this question
2f	**Defining the role of antimicrobials on gut epithelial homeostasis.**	Understanding the different antimicrobial classes affecting epithelium homeostasis and pathways directly or indirectly involved. Understanding the physiology and nutrition of host. Is it increased food intake or improved feed conversion rate or both?	Organoids cell culture, using chambers. Use of intestine/gut loop (with a focus on epithelial cells). In vivo, in vitro, ex vivo studies. Precision cut gut slices for HPI studies looking at the epithelial layer. Study oral tolerance. Defined standardized methods to test mechanism of subAbx and defined goal (growth vs feed conversion rate- FCR).	Mechanisms and/ or cellular action (direct or indirect) via microbes, of antimicrobials used as AGP, on epithelial cell homeostasis.
3a	**Identifying key limiting nutritional components for maintaining growth and immune functions.**	Comparing nutrients and food components with immunological effects in standard settings including studies on different status of the animal.	Observational studies on growth and subclinical disease control.	

Annex II: Phage technologies: Alternatives to antibiotics acting directly on the pathogen, including establishing their mode of action

Workshop 12 October 2021

Roadmap for Phage technologies



Lead Summary [1]

Pathogen control using phage technology

Research Question

What are we trying to achieve and why? What is the problem we are trying to solve?

Use of virus technology to deliver improved control of infectious (bacterial, protozoal, helminths) disease.

Challenge(s)

What are the scientific and technological challenges (knowledge gaps needing to be addressed)?

Consistent research methodologies to enable evaluation of efficacy and safety on animals and comparison between groups and technologies.

Socioeconomic evaluations of industry uptake.

Efficacy - Establish range of target organisms.

Safety – for the animal as well as containment (especially in a farm environment).

Equity and equality of phage products (affordability in different countries).

Increase public acceptability.

Acceptability to the end user and understanding how they are going to be applied.

How they differ from other phage products on the market.

Regulatory challenges.

Need for dedicated phage manufacturing facility and production line of phages.

Investigating phage-kinetic (how they are distributed) in the host (especially for systemic use), with various delivery systems.

Better understanding the dynamics of the interaction between phage (pure and mixed culture), plasmids (sometimes more than one in a bacterial cells), suicide systems and bacteriocins. This for in vitro and different in vivo systems (septicaemia and gut colonisation).

Research into their ability to survive in animal and in the environment is needed.

Dosing, timing (i.e. Understanding the impact of administration in different immunity states of host, age groups) and administration for each disease (systemic versus localised).

Target and Route of administrations.

Immune response and effects of several administrations.

Understanding the relation between phage and immune system (two sides – immune system kills the phages and/or the phages also kill useful microbiota).

Knowing the target population.

Specificity of phages.

Cross reactivities between phages, target bacteria, other bacteria.

Intracellular pathogens (extra delivery systems?) and biofilms.

Complexity of phage and delivery systems.

Nutrition and survival of phages in GUT environments (host/nutrition).

Solution Routes

What approaches could/should be taken to address the research question?

Trials using standardised model systems, dosage, and controls.

Field trials.

Determining how phages behave after they have been shed by animals.

Engagement with competent authorities (EMA/FDA) to define efficacy and safety requirements, and to validate clinical protocols for market authorisation.

Cost and benefits analysis.

Producer and producer group engagement.

Marketing and benchmarking campaigns to increase acceptability of users.

Increasingly complex systems for modelling, starting with one phage and one bacterium then adding a plasmid, then two plasmids and bacteriocins but also modelling transposons.

Encapsulations for delivery to appropriate site.

Storage and application methods (on-farm).

Study interaction of phages and the immune system using organ cultures/organ on a chip.

Organ cultures could also be used for screening in vivo using phages and the microbiota

Dependencies

What else needs to be done before we can solve this need?

Studies on the use of phages which have a direct effect, killing pathogens (lytic and lysogenic) or other effects such as selecting for AMR plasmid loss.

Using phages as a delivery system.

Investigating the use of antimicrobials peptides (i.e. bacteriocins, tailosins...)

Better mathematical models that can incorporate several sub-bacterial entities (phages- plasmids and bacteriocins - and possibly transposons).

Establishing best phages as the different types of phages (e.g Myoviridae) have different kinetics (very complicated phages may not be as easy to dry as they have more parts to break off).

State of the Art

Existing knowledge including successes and failures

Extensive (recent) literature going back to 1982.

Projects

What activities are planned or underway?

AVANT project (WG on phages)

InnoVet-AMR funding two projects developing phage.

Lead Summary [2]

Screening for absence of undesirable genes

Research Question

What are we trying to achieve and why? What is the problem we are trying to solve?

Increasing product safety.

Challenge(s)

What are the scientific and technological challenges (knowledge gaps needing to be addressed)?

Understand direct and indirect effects on non-targeted commensal bacteria

Consistent research methodology to enable evaluation of efficacy and safety.

Genomic information missing (70% of their genomes is hypothetical proteins)

Detect elements that can allow transfer of AMR genes.

Solution Routes

What approaches could/should be taken to address the research question?

An alternative to engineering from scratch is to modify existing phages to improve their safety profile.

Identify Phage “virulence determinants.

Improving bioinformatic based on structures rather than on sequence. similarities by using structural prediction tools (e.g. alpha fold-2).

Dependencies

What else needs to be done before we can solve this need?

Understanding the link of phages with healthy animal.

State of the Art

Existing knowledge including successes and failures

Projects

What activities are planned or underway?

Lead Summary [3]

Phage as an adjunct to other therapies

Research Question

What are we trying to achieve and why? What is the problem we are trying to solve?

To understand if phage and antibiotics can be used together

to generate synergistic effect and reduce/counteract resistance.

Challenge(s)

What are the scientific and technological challenges (knowledge gaps needing to be addressed)?

Identify receptors sites (e.g. binding efflux pumps).

Investigate interaction among phages and antibiotics.

Understanding the adaptation of phages to immune systems.

Characterization of phage resistance in bacteria and fitness costs of bacteria (resistant bacteria may be more susceptible to AB and the immune response).

Use of phages to block transmission of AMR plasmids (phages could be used to cut antibiotic resistant genes and plasmids).

Solution Routes

What approaches could/should be taken to address the research question?

In vitro and in vivo evaluation.

Crisp-Cas system in the bacteria – can they show immunity to the phages.

Dependencies

What else needs to be done before we can solve this need?

State of the Art

Existing knowledge including successes and failures

Phages and antibiotics both induce resistance.

Projects

What activities are planned or underway?

Lead Summary [4]

Lytic Phages

Research Question

What are we trying to achieve and why? What is the problem we are trying to solve?

Using phages which have a direct effect killing bacteria/pathogens (lytic and lysogenic).

Challenge(s)

What are the scientific and technological challenges (knowledge gaps needing to be addressed)?

Suppress development of Phage resistance and enhancing susceptibility.

Select both the 'best' and most aggressive phages and use them in combination to ensure phages with good synergies are used.

Finding phages against which bacteria struggle to develop resistance.

Limits of their use (depending on environment - under agriculture conditions). e.g., stability, dosage.

Develop better strategies to manipulate lytic phages – although techniques exist for E. coli phages and others with good genetic systems -these are still slow.

Lytic phages may produce lots of LPS toxins (bad = proinflammatory).

Lytic phages maybe ok for acute phase treatments, phage cocktails needed to avoid resistance.

Solution Routes

What approaches could/should be taken to address the research question?

Passage of lytic phage combinations in bacterial cultures.

Metagenomics analysis of phage effects on microbial diversity, richness, and stability.

Identify phages specific for surface virulence determinants.

Investigate phages that bind with the sex pili produced by antimicrobial resistance plasmids – the phage may select for resistance in the bacterium because they have lost the plasmid that produce the pili.

Dependencies

What else needs to be done before we can solve this need?

Understanding phage-pathogen ecology.

Establishing a better ecological and bioinformatic framework to inform isolation.

Better understanding the biology of the bacterium to identify areas the bacteria in question live and we can find more phages.

Establishing better informed strategies to isolate or choose relevant phages.

Determining the phageome (basic studies are needed to determine the diversity of phages).

Investigating phage-kinetics (how they are distributed) in the host (especially for systemic use), with various delivery systems.

Investigating evolution of phages and their targets.

Developing new methods for detection and quantification of lytic phages.

Investigating new ways of selecting sequences to be kept (positive as well as negative screens).

State of the Art

Existing knowledge including successes and failures

Projects

What activities are planned or underway?

Lead Summary [5]

Phage as delivery vehicle

Research Question

What are we trying to achieve and why? What is the problem we are trying to solve?

Using phages as a delivery system (including to deliver i) factors that modulate AMR by targeting AMR plasmids ii) technology such as CRISPR-Cas including for modulation of virulence factors, iii) antibodies and iv) antimicrobial peptides).

Challenge(s)

What are the scientific and technological challenges (knowledge gaps needing to be addressed)?

Stability of phage genetics.

Mechanisms of function.

Plasmid stability and retransformation.

Bacterial selection and phage resistance.

Efficacy in pure culture and complex communities (i.e., microbiome).

Better engineering needed to construct mutants – A robust retargetable phage platform is needed for that.

Phages used to deliver plasmid disrupting genes may be considered GM organisms.

Solution Routes

What approaches could/should be taken to address the research question?

High throughput systems – CRISPR-Cas technology to edit out parts of the phages would be useful.

Improvement in the application of synthetic biology to phage biology and for example exploiting Gibson assemblies and rebooting or CRISPR technology could help considerably.

Experimental design requires sufficient replication to account for variation.

Projects could include a testing pipeline of increasing complexity: pure culture > artificial or ex vivo communities > animal studies.

Experimental design requires sufficient replication to account for variation.

Cell free platform.

Might use simple phages such as small filamentous phage (e.g. M13) for delivery.

Dependencies

What else needs to be done before we can solve this need?

[Need to have good genetic systems.] Development of genetic systems for all phages.

[Need to genuinely compare good native phages to engineered ones.]

Mechanics of more widely applying editing systems.

State of the Art

Existing knowledge including successes and failures

Projects

What activities are planned or underway?

Lead Summary [5A]

Phage to deliver genetic material to disrupt AMR plasmids or virulence factors

Research Question

What are we trying to achieve and why? What is the problem we are trying to solve?

Using phages as a delivery system for factors that modulate AMR by targeting AMR plasmids or targeting plasmid-mediated virulence factors.

Challenge(s)

What are the scientific and technological challenges (knowledge gaps needing to be addressed)?

AMR and virulence gene stability.

Better understanding of AMR and virulence plasmids and phages' ability to selectively target plasmid-containing cells also needed.

One main problem is that phages may also increase horizontal gene transfer (they do so in their natural environments). Survey horizontal gene transfer (HGT).

Solution Routes

What approaches could/should be taken to address the research question?

Targeting the sex pili to eliminate AMR transfer.

Dependencies

What else needs to be done before we can solve this need?

State of the Art

Existing knowledge including successes and failures

Projects

What activities are planned or underway?

Lead Summary [5B]

Phage as delivery vehicle for therapies (Ab, AMP etc)

Research Question

What are we trying to achieve and why? What is the problem we are trying to solve?

Can we use phage capsids to deliver material toxic to bacteria?

Challenge(s)

What are the scientific and technological challenges (knowledge gaps needing to be addressed)?

Technological challenges – capsid construction and encapsulating delivery material.

Timely administration of phage for therapy.

Solution Routes

What approaches could/should be taken to address the research question?

Delivery material can be chemically conjugated to capsid surface, if 'natural'' phage is identified.

Dependencies

What else needs to be done before we can solve this need?

Prompt diagnosis.

State of the Art

Existing knowledge including successes and failures

See work of George Lomonossoff at John Innes.

Projects

What activities are planned or underway?

Lead Summary [6]

Phage selection

Research Question

What are we trying to achieve and why? What is the problem we are trying to solve?

Determining end point of application for each phage.

Challenge(s)

What are the scientific and technological challenges (knowledge gaps needing to be addressed)?

Developing new methods for detection and quantification of phages.

Identify suitable, stable and robust phages: it is very difficult to isolate either any, or strictly virulent phages for some bacterial species – so effort should be put into this area.

Not just lytic phages but phages with desirable properties e.g., for delivery or which target key surface structures on bacteria.

Selection of specific phages that do not attack useful bacteria.

Effect of phages within microbiota that affect production –the use of phages to target gut microorganisms to favour increased production.

Understanding of phages and genetics of animals (host/genetic).

Better understanding of phage life cycle - to identify genes linked with different lifecycles as it is not a clear-cut situation between lytic and lysogenic phages.

Solution Routes

What approaches could/should be taken to address the research question?

Could include initial DNA/RNA detection systems.

Large scale screening - utilising machine learning to identify which phages may be the most effective at targeting key pathogens -

(this would include looking to see if all those that are effective have particular genes associated with them but could also include integration of data on the genome of the bacteria).

Viral tagging.

Metagenomics to understand where phages may be in the specific environment.

Organ cultures could also be used for screening in vivo using phages and the microbiota.

Dependencies

What else needs to be done before we can solve this need?

Suitable range of bacteria against which phages can be sought for targeting.

State of the Art

Existing knowledge including successes and failures

Gram negative bacteria more difficult to kill than Gram positive ones (with bacteriocins – 1:00).

Projects

What activities are planned or underway?

Lead Summary [7]

Phage production by Synthetic biology

Research Question

What are we trying to achieve and why? What is the problem we are trying to solve?

Construction new phages de novo with well understood characteristics and genome structure.

To construct a retargetable platform that can be tweaked to meet different needs.

Challenge(s)

What are the scientific and technological challenges (knowledge gaps needing to be addressed)?

Lack of knowledge on mechanisms of action

Investigating chemicals/phytochemicals that modulate immune responses as well as attack bacteria including host antimicrobials peptides (AMP).

Enhancing natural production in host.

Scaling up production.

Establishing specificity.

Study toxicity, safety and environmental impact of products

Immune response against agent.

Mechanics of more widely applying editing systems that will enable development of phages with desirable characteristics such as expanded host range.

Genetically retarget the phages, orthogonally functionalize the capsid surface, get possibly rid of the phage nucleotide core (genome).

Need to ensure avidity is not reduced, if affinity is increased.

Regulatory challenges.

Solution Routes

What approaches could/should be taken to address the research question?

Explore de novo construction of phages with desirable attributes, cloning and expression systems.

Investigating new ways of selecting sequences to be kept (positive as well as negative screens).

Dependencies

What else needs to be done before we can solve this need?

Long-stretch PCR to help in de novo construction of larger viruses

Targeting moieties that are selective enough (peptides, Abs, natural ligands

Knowledge can fuel synthetic biology approaches to engineer and retarget phage delivery platforms

State of the Art

Existing knowledge including successes and failures

Methods for de novo construction of small viruses already exist

Some selective targeting molecules reported in literature

Projects

What activities are planned or underway?

Lead Summary [8]

(Understanding of) Phage-pathogen interaction including life cycle, attachment sites etc

Research Question

What are we trying to achieve and why? What is the problem we are trying to solve?

Investigating basic phage-pathogen interaction, kinetics and evolution of phages and their targets.

Challenge(s)

What are the scientific and technological challenges (knowledge gaps needing to be addressed)?

In vitro study for phage-pathogen interaction

For many bacteria need to know physiological state it is in when it causes infection

Determining which resistance or counter-defence processes will take place at any given moment depends on multiple factors, e.g., mutation rate, bacterial population diversity, nutrient availability, spatial structure etc

How do phages interact with plasmids and their suicide systems.

Solution Routes

What approaches could/should be taken to address the research question?

In vitro modelling over time with sequencing analysis.

Integration of epidemiology and ecology of bacteria and phages into disease management.

Need more in silico and in vivo studies to investigate co-evolutionary dynamics - short versus long term.

Identifying phage binding sites/surface virulence determinants: map the adhesion sites and determine the common binding tags of the adhesion with the receptor. This may lead to a better understanding of the binding and engineer a phage with the desired spectrum - Identify tail fiber determinants and specificity and attachment site.

X-ray and other 3D analysis systems for receptor study.

Dependencies

What else needs to be done before we can solve this need?

Modelling systems probably already exist.

State of the Art

Existing knowledge including successes and failures

Projects

What activities are planned or underway?

Lead Summary [9]

Phage Isolation

Research Question

What are we trying to achieve and why? What is the problem we are trying to solve?

Establishing better informed strategies to isolate or choose relevant phages.

Challenge(s)

What are the scientific and technological challenges (knowledge gaps needing to be addressed)?

Establishing a better ecological and bioinformatic framework to inform isolation.

Better understanding of the bacterial structures that are receptors for phages.

Explore relevant/specific environments to isolate them: most phages are isolated from sewage, rivers or farms.

More work could be carried out on isolation procedures to not just enrich and thus find the common phages but to identify less common phages that could have better properties.

Establishing phage collections for all the major pathogens and main commensals especially those bacteria which harbour AMR plasmids. This will include numerically dominant anaerobes from the gut.

Biases associated with the key isolation steps, e.g., from sample handling, VLP extraction, nucleic acid extraction protocol & library preparation.

Solution Routes

What approaches could/should be taken to address the research question?

Use of AI and machine learning with the increasing information on phage genomes to identify characteristics optimal for the use to which they may be put.

Dependencies

What else needs to be done before we can solve this need?

Larger accessible genome libraries.

Software adapted to use with phages many of which are AT rich.

State of the Art

Existing knowledge including successes and failures

Projects

What activities are planned or underway?

Lead Summary [10]

Understanding of Phages in the relevant environment/ phage ecology

Research Question

What are we trying to achieve and why? What is the problem we are trying to solve?

Better understanding the biology of the bacterium/phage interrelationship within the host and in the wider environment which will enhance our knowledge of where to find new and improved phages.

Determining the phageome (basic studies are needed to determine the diversity of phages).

Phage ecology: better understanding phage stability/fragility in the environment.

Challenge(s)

What are the scientific and technological challenges (knowledge gaps needing to be addressed)?

Better understanding of the "life within bacteria" the genomes that parasitise them including phages. Plasmids and some bacteriocins.

Impact of phages on the microbiome of animals.

What are the most effective intervention points?

Use of MDA prior to sequencing preferentially amplifies small and circular ssDNA–, sequencing depth etc… complicates comparison of phageome

Gut phageomes in particular are dominated by uncharacterised sequences - Viral dark matter.

Requires a better understanding of phage survival in environment (also in building and on surfaces) and genetic exchange in the environment.

Requires better understanding of relationship, if any, between phages in the environment and those in the human and animal phageome.

Solution Routes

What approaches could/should be taken to address the research question?

More research on the dynamics of phage.

Plasmids, bacteriocins and their suicide systems.

Funding to establish comprehensive collections. Done with time to explore phage evolution.

Studies in vitro on phage evolution.

Standardised protocols help to promote comparison across studies – challenge is to ensure that novel phages are not missed by these standardised methodologies.

Increased efforts in increasing representation of phage within databases – both animal and human focused.

Further development of alignment-free bioinformatic methods for identifying phage sequences.

Difficult in situ studies without access to sensitive PCR and RT-PCR systems. Controlled in vitro studies will help.

Dependencies

What else needs to be done before we can solve this need?

Methods already there for in vitro work.

State of the Art

Existing knowledge including successes and failures

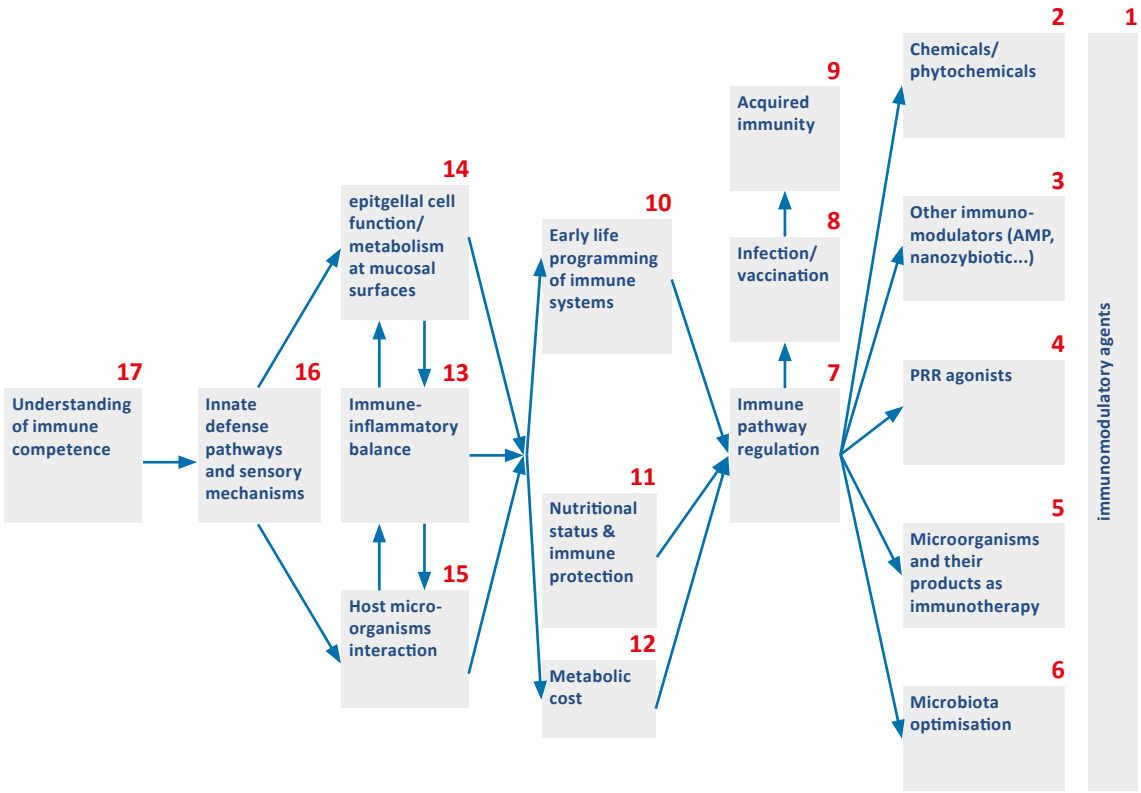
Projects

What activities are planned or underway?

Annex III: Immunomodulators: Agents and compounds for their ability to enhance the hosts resistance to disease, including establishing their mode of action

Workshop 15 October 2021

Roadmap for Immunomodulators



Lead Summary [1] .

Immunomodulatory agents

Research Question

What are we trying to achieve and why? What is the problem we are trying to solve?

Establishing the potential of the following approaches to enhancing the non-specific resistance of animals:

The use of bacteria that stimulate/reprogram the immune response; Probiotics; Prebiotics that stimulate growth of immunostimulatory bacteria; AMP; Phytochemicals; Other immunogenic compounds (e.g. nutraceuticals, biomolecules); Develop C-terminal VH region of camelid heavy chain antibodies (VHH) that target pattern recognition receptors;

Passive immunisation strategies; Antibodies derived from blood used as additives/for nutrition; Microbiota modification.

Challenge(s)

What are the scientific and technological challenges (knowledge gaps needing to be addressed)?

Consistent research methodologies to enable evaluation of efficacy and comparison between groups and technologies.

(Animal factors: different batches of animals give different results - different models for species?/ environment).

Consistent production of the immunomodulator.

Interaction among different immunomodulators (investigate mode of actions)

Interaction with nutrients.

Ensuring efficacy (at least up to 30% as compared to antibiotics).

Robust response without over stimulation of the immune system.

Reducing unwanted inflammation (in mucosal surfaces).

Consistent research methodologies to enable evaluation of efficacy and comparison between groups and technologies (e.g., what defines ‘resilience’).

Identification of host factors that determine tolerance (proof of cause-effect relationship).

Pointing out the critically important elements of nutrition and the way feeding is performed that prevent diseases, and if the elements differ in relation to the cause of the disease.

Understanding reasons behind failure of immunomodulators that were brought to the market earlier.

Routes of administration (oral vs parenteral and impact on other compartments).

Frequency of administration.

Dosage.

Resistance to the technological process when delivered by feed (e.g. temperature when preparing feed).

Type of formulation and shelf-life of the product.

Genetic and age of the host.

Palatability of the formulation.

Storage requirements in different environments.

Solution Routes

What approaches could/should be taken to address the research question?

Field studies to establish the efficacy and safety of:

Bacteria that stimulate/reprogram the immune response.

Probiotic.

Prebiotics that stimulate growth of immunostimulatory bacteria.

AMP.

Phytochemicals

Nutrition.

Other immunogenic compounds (e.g., nutraceuticals, biomolecules).

C-terminal VH region of camelid heavy chain antibodies (VHH) that target pattern recognition receptors.

Passive immunisation strategies.

Microbiota modification.

Antibodies derived from blood used as additives/for nutrition

Investigate cross immunomodulations among different compartments)

Dependencies

What else needs to be done before we can solve this need?

Establishing mode of action of the various candidates

Investigating early life programming of the immune system from a Th2 to a Th1 response.

Investigating innate defence pathways (e.g., interferons) and how they are impacted by specific agents.

Studying the impact of probiotics on the immune response.

Investigating AMP that modulate immune responses as well as attack bacteria.

Investigating nutrition requirements to maintain optimal immune response.

Using cytokines.

Using phytochemicals.

Better understanding interaction between immune responses and inflammation.

Investigating competitive exclusion.

Using enzymes able to break down the toxins produced from pathogens.

Studying innate immune memory.

Investigating epithelial cells metabolism and proliferation.

Investigating down-regulation of receptors for pathogens in animals.

Understand the metabolic cost of this approach.

Investigating impact of live vaccines (competitive colonisation)

Evaluating nutritional compounds (e.g., metals, vitamins)

State of the Art

Existing knowledge including successes and failures

Projects

What activities are planned or underway?

Lead Summary [2]

Chemicals/phytochemicals

Research Question

What are we trying to achieve and why? What is the problem we are trying to solve?

Investigating chemicals/phytochemicals that modulate immune responses as well as attack bacteria.

Challenge(s)

What are the scientific and technological challenges (knowledge gaps needing to be addressed)?

Lack of knowledge on mechanisms of action

Investigating chemicals/phytochemicals that modulate immune responses as well as attack bacteria including host antimicrobials peptides (AMP).

Enhancing natural production in host.

Scaling up production.

Establishing specificity.

Study toxicity, safety and environmental impact of products

Immune response against agent.

Solution Routes

What approaches could/should be taken to address the research question?

Develop recombinant systems to produce and purify AMPs at scale.

Develop conjugation chemistries and release technologies (e.g., self-immolating, enzyme/microbial mediated).

Dependencies

What else needs to be done before we can solve this need?

Understanding mode of action of the AMPs

State of the Art

Existing knowledge including successes and failures

Projects

What activities are planned or underway?

Lead Summary [3]

Other immunomodulators (e.g. AMPs, nanozybiotic…)

Research Question

What are we trying to achieve and why? What is the problem we are trying to solve?

Investigating other new class of potential immunomodulators (peptides, nanozybiotics, metal–sulfide/oxides, carbon-based nanostructures…).

Challenge(s)

What are the scientific and technological challenges (knowledge gaps needing to be addressed)?

AMPs challenges:

Stability in vivo environment;

Administration route: -low oral bioavailability due to pre-systemic enzymatic degradation and poor penetration of the intestinal mucosa -short life in blood.

Enzybiotics (e.g. peptidoglycan hydrolases, proteases, and nuclease) challenges:

Short half-life and raise of neutralizing antibodies in vivo due to their proteinaceous nature;

Lack of stability outside optimum operating conditions.

Solution Routes

What approaches could/should be taken to address the research question?

Develop recombinant systems to produce and purify AMPs at scale.

Minimal inhibitory concentration (MIC) or minimal microbicidal concentration (MMC) assays.

Investigate modification that can improve stability, safety, long term storage and reduce the cost of their productions.

Explore new biocompatible nanozybiotics using enzyme-like nanozymes.

Develop combination therapy using nanozybiotic.

Develop conjugation chemistries and release technologies (e.g., self-immolating, enzyme/microbial mediated).

Dependencies

What else needs to be done before we can solve this need?

Understanding mode of action of the AMPs.

Nanocarriers developments.

Cost-benefit of the preparations.

State of the Art

Existing knowledge including successes and failures

Projects

What activities are planned or underway?

Lead Summary [4]

Pattern Recognition Receptors (PRRs) agonists

Research Question

What are we trying to achieve and why? What is the problem we are trying to solve?

Identifying PPRs agonists as immunostimulants.

Challenge(s)

What are the scientific and technological challenges (knowledge gaps needing to be addressed)?

Understanding the TLR signalling and its function.

Understand how to antagonize the harmful effects without affecting host defence functions.

Solution Routes

What approaches could/should be taken to address the research question?

Identify a panel (an array) of PRR

Identify trigger and functions.

Identify PRR, test for function and look for selection.

Dependencies

What else needs to be done before we can solve this need?

State of the Art

Existing knowledge including successes and failures

Projects

What activities are planned or underway?

Lead Summary [5]

Microorganisms and their products as immunotherapy

Research Question

What are we trying to achieve and why? What is the problem we are trying to solve?

Define mechanisms of interaction between microorganisms and the host immune system.

Challenge(s)

What are the scientific and technological challenges (knowledge gaps needing to be addressed)?

Influence of anti-inflammatory or pro-inflammatory molecules on receptor expression.

Include antibiotic sensitive bacterial strains similar to those in the gut which are AMR since this generates an exclusion effect and stimulates immunity.

Understanding ecology in terms of microbes

Product with competitive exclusion properties with attention of the immune system activation. Is expectant that adhesion of product activates the local and systemic immunity. Is important to avoid over inflammation.

Product with competitive exclusion properties that bind to adhesions on surface of bacterial pathogens and prevent interactions with host tissues.

To produce stable molecule or sub-unit structures that mimic the adhesins of gut bacteria to epithelial cells and mucin.

Solution Routes

What approaches could/should be taken to address the research question?

Bacteria.

Glycoproteins.

Mimetics from agricultural residues.

Tools for quantitative expression of receptors based on PCR, proteomics, glycomics, and or immunoassays.

Identify products with affinity receptors of interest. Other think the competitive exclusion, the product should stimulate a targeted immune response against specific pathogen (there is already a vaccine based on this mechanism of action. This product is more than a vaccine).

Glycomic analyses of surface glycans and glycoproteins to inform the structure of soluble decoys.

Determine sources of soluble decoys (e.g., dairy waste) for extraction.

Either take an approach such as this or seek to analyse the basis of adhesion.

Dependencies

What else needs to be done before we can solve this need?

Investigating products, bacteria and/or glycoproteins (e.g., lectins) able to interfere with bacterial colonisation (e.g., occupying the same site of adhesion/niche of pathogens, such as receptor on the small intestine for E. coli F4 and F18).

Investigating interaction between host and the selected product.

Investigating the timing of administration to boost the effect:

- dose effect;
- age effect of animal adhesion/niche of pathogens, such as receptor on the small intestine for E. coli F4 and F18).

Developing soluble decoys to prevent adherence/colonisation of bacterial pathogens.

Identifying the receptors recognised by pathogens.

Much better understanding of adhesins and also 3D interaction with the host receptors and effect of adhesion on gut physiology.

(Investigating competitive exclusion- Investigating products, bacteria and/or glycoproteins (e.g., lectins) able to interfere with bacterial colonisation (e.g., occupying the same site of adhesion/niche of pathogens, such as receptor on the small intestine for E. coli F4 and F18).

Developing soluble decoys to prevent adherence of bacterial pathogens.)

State of the Art

Existing knowledge including successes and failures

Projects

What activities are planned or underway?

Lead Summary [6]

Microbiota optimisation (including microbiome) – see roadmap on Microbiota optimisation

Research Question

What are we trying to achieve and why? What is the problem we are trying to solve?

Challenge(s)

What are the scientific and technological challenges (knowledge gaps needing to be addressed)?

Solution Routes

What approaches could/should be taken to address the research question?

Dependencies

What else needs to be done before we can solve this need?

State of the Art

Existing knowledge including successes and failures

Projects

What activities are planned or underway?

Lead Summary [7]

Immune pathway regulation

Research Question

What are we trying to achieve and why? What is the problem we are trying to solve?

Identifying how the immune response can been modified to enhance resistance.

Challenge(s)

What are the scientific and technological challenges (knowledge gaps needing to be addressed)?

Lack of specific reagents.

Lack of knowledge.

Solution Routes

What approaches could/should be taken to address the research question?

Cytokine responses.

Cellular responses.

System approach.

MOA (Mechanism of Action).

Multi-omic-approach.

(interaction with receptors TRLs).

Dependencies

What else needs to be done before we can solve this need?

State of the Art

Existing knowledge including successes and failures

Projects

What activities are planned or underway?

Lead Summary [8]

Infection/vaccination

Research Question

What are we trying to achieve and why? What is the problem we are trying to solve?

Investigating impact of live vaccines.

Challenge(s)

What are the scientific and technological challenges (knowledge gaps needing to be addressed)?

Live vaccines delivered to the gut can induce an immune response in addition to generating an exclusion effect.

Live vaccines delivered parenterally can also induce innate immunity which can be cross protective against unrelated bacterial pathogens.

Adjuvant as innate modulator for training or tolerance.

Integrated immunology needed rather than separation of innate and adaptive

understanding of ecology and evolution and pathogens.

Solution Routes

What approaches could/should be taken to address the research question?

Further experimental in vivo studies.

Dependencies

What else needs to be done before we can solve this need?

Safety - vaccine might be damaging in the long term and should not select for escape variants- understanding of ecology and evolution and pathogens in a vaccinated and unvaccinated or partially vaccinated world.

State of the Art

Existing knowledge including successes and failures

Projects

What activities are planned or underway?

Lead Summary [9]

Acquired immunity – see Vaccinology Roadmap

Research Question

What are we trying to achieve and why? What is the problem we are trying to solve?

Challenge(s)

What are the scientific and technological challenges (knowledge gaps needing to be addressed)?

Solution Routes

What approaches could/should be taken to address the research question?

Dependencies

What else needs to be done before we can solve this need?

Vaccinology roadmap.

State of the Art

Existing knowledge including successes and failures

Projects

What activities are planned or underway?

Lead Summary [10]

Early life programming of immune system

Research Question

What are we trying to achieve and why? What is the problem we are trying to solve?

Investigating early life programming of the immune system

(from a Th2 to a Th1 response).

Regulatory responses (including Th17).

Can we train the immune system to produce a more favourable response – whether it is Th1, Th2, Treg or others– are there early life events that can influence vaccination and subsequent ability to respond?

Challenge(s)

What are the scientific and technological challenges (knowledge gaps needing to be addressed)?

Identifying suitable products.

Role of stress on immune responses.

Improved understanding of the immune system maturation of new-born.

Solution Routes

What approaches could/should be taken to address the research question?

Characterise and produce synthetic or purified compounds produced by mammalian mothers (colostrum, milk glycans, secondary metabolites).

Investigating Ifn gamma levels.

Investigating PRR agonists.

Antigens (or pathogens) at low dosage.

Dependencies

What else needs to be done before we can solve this need?

Improved understanding of the immune competence of the new-born.

Investigating what pathways are upregulated in the host when probiotics/bacterial agents/milk compounds are administered.

Confirmation that the same Th1/Th2 divide occurs in the species of concern.

State of the Art

Existing knowledge including successes and failures

Projects

What activities are planned or underway?

Lead Summary [11]

Nutrients & immune protection

Research Question

What are we trying to achieve and why? What is the problem we are trying to solve?

Establishing the nutrition requirements to maintain optimal immune response.

Evaluating nutritional compounds (e.g., metals, vitamins).

Challenge(s)

What are the scientific and technological challenges (knowledge gaps needing to be addressed)?

Define the nutrient requirements in sub-optimal sanitary conditions (species-specific).

- boosting the immune response after vaccination or immune stimulation (e.g., amino acids like Trp of Thr in pigs are primarily involved in the immune response and their utilisation are prioritised for this function than for the animal growth).

Solution Routes

What approaches could/should be taken to address the research question?

Define nutrients and the appropriate dosage to sustain immune response.

Dependencies

What else needs to be done before we can solve this need?

Measuring the improvement of the immune response with specific supra-nutritional nutrients supplementation in specific conditions.

State of the Art

Existing knowledge including successes and failures

Projects

What activities are planned or underway?

Lead Summary [12]

Metabolic cost

Research Question

What are we trying to achieve and why? What is the problem we are trying to solve?

Understand the metabolic cost of interventions.

Challenge(s)

What are the scientific and technological challenges (knowledge gaps needing to be addressed)?

Solution Routes

What approaches could/should be taken to address the research question?

Lead Summary [13]

Immune-inflammatory balance

Research Question

What are we trying to achieve and why? What is the problem we are trying to solve?

Better define how inflammation negatively impact on the animals.

Challenge(s)

What are the scientific and technological challenges (knowledge gaps needing to be addressed)?

Establish incitants and pathways of inflammation to abiotic and biotic factors in livestock animals.

Explore if reducing inflammation has a negative impact on the disease outcome on the animals.

Role of stress on immune response/inflammation.

Role of hormones on immune response/inflammation.

Solution Routes

What approaches could/should be taken to address the research question?

Platforms for monitoring host responses (e.g., PCR, ELISA, OMICs).

Measurement of cytokine/chemokine expression is key.

Monitoring interactions between immunomodulators and microbiota.

Histology or tissue effects.

Dependencies

What else needs to be done before we can solve this need?

State of the Art

Existing knowledge including successes and failures

Projects

What activities are planned or underway?

Lead Summary [14]

Epithelial cell function/metabolism at mucosal surfaces

Research Question

What are we trying to achieve and why? What is the problem we are trying to solve?

Investigating epithelial cells metabolism and proliferation.

Challenge(s)

What are the scientific and technological challenges (knowledge gaps needing to be addressed)?

Identify products that improve epithelial cells activities in vitro and in vivo, under normal or stress conditions (oxidative imbalance, toxins, infections, general mucosal damage, undernourished animals, etc).

Solution Routes

What approaches could/should be taken to address the research question?

Identify and classify products that can stimulate several/different metabolic pathways and proliferative phases of epithelial cells, particularly at small intestine.

Dependencies

What else needs to be done before we can solve this need?

Investigating the action with cell lines.

Characterising the impact on intestinal epithelial cells and overall epithelium physiology in healthy and challenged animals.

Investigating the action with cell lines or organoids.

Role of nutrients (e.g. Vit A).

State of the Art

Existing knowledge including successes and failures

Projects

What activities are planned or underway?

Lead Summary [15]

Host-microorganism interaction

Research Question

What are we trying to achieve and why? What is the problem we are trying to solve?

Establishing the impact of probiotics/prebiotics on the immune response.

Establishing down-regulation of receptors for pathogens in animals.

Establishing what pathways are upregulated in the host when probiotics/bacterial agents/milk compounds are administered.

Challenge(s)

What are the scientific and technological challenges (knowledge gaps needing to be addressed)?

Microorganisms and TLR signalling

Tolerance of immune response of the host

Study on “functional response of the host” Functional study of microbiota

Identify probiotics/prebiotics that deliver specific required outcomes.

Insight into dosage, storage, and application of probiotics/prebiotics on farm (delivery).

Advantages-disadvantages of host-adapted and broad-spectrum probiotics/prebiotics.

Include antibiotic sensitive bacterial strains similar to those in the gut which are AMR since this generates an exclusion effect and stimulates immunity.

Define mechanisms of interaction between pathogens and host.

Solution Routes

What approaches could/should be taken to address the research question?

Standardisation of methods (e.g., delivery method, dosage, animal age) to determine host-response.

Determining factor produced by microorganisms (e.g. butyrate).

Design platforms for monitoring host responses (e.g., PCR, ELISA, OMICs).

Targeted interventions to tune host responses.

Dependencies

What else needs to be done before we can solve this need?

Basic immunology

Functional microbiota studies- microbiome roadmap

State of the Art

Existing knowledge including successes and failures

Projects

What activities are planned or underway?

Lead Summary [16]

Innate defence pathways

Research Question

What are we trying to achieve and why? What is the problem we are trying to solve?

Improved understanding of the innate defence mechanisms, including sensory mechanisms, and how they can be modulated by specific agents.

Challenge(s)

What are the scientific and technological challenges (knowledge gaps needing to be addressed)?

Determine host responses to diet and microbiome interactions.

Define mechanisms of interaction between pathogens and host.

Understand how long (or short) the innate memory lasts. Identify products that can modify the duration.

Solution Routes

What approaches could/should be taken to address the research question?

Coupling OMICs of microbiome-host (agonist and antagonist) interaction to immune responses: High-resolution metagenome analyses (shotgun metagenomics, MAGs), metabolomics, metaproteomics, glycomics.

Developing experimental models and assays (screening systems for potential agonists).

Define/Identify vaccines that can induce and increase duration of innate immune memory (e.g., target monocytes/macrophages).

Dependencies

What else needs to be done before we can solve this need?

State of the Art

Existing knowledge including successes and failures

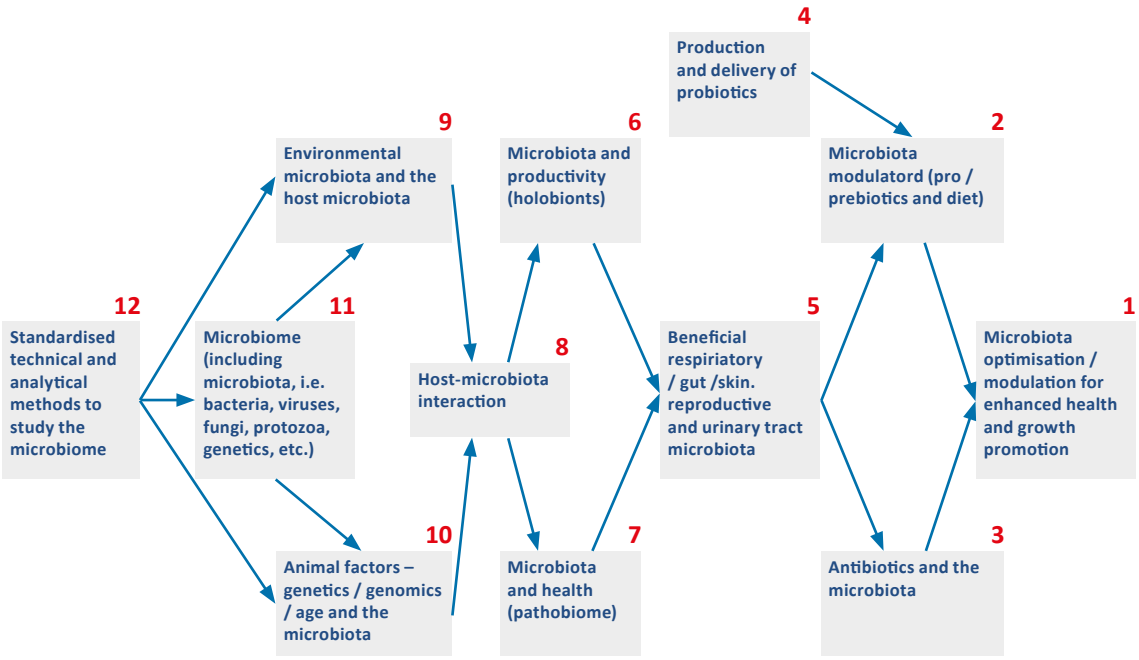
Projects

What activities are planned or underway?

Annex IV: The role of the microbiome in the maintenance of health, and how it can be manipulated

Workshop 18th October 2021

Roadmap for Microbiota optimisation



Lead Summary [1]

Microbiota optimisation/modulation for enhanced health and growth promotion

Research Question

What are we trying to achieve and why? What is the problem we are trying to solve?

Manipulation of the host microbiota (MB) to improve health outcomes without the use of antibiotics.

Microbiome transplantation after antibiotic oral therapy to replace with a flora that might be better than the host's original flora?

Challenge(s)

What are the scientific and technological challenges (knowledge gaps needing to be addressed)?

Ensuring efficacy and safety of interventions.

Determine sufficient level of depth required to measure positive responses.

Ensuring sharing of data and technologies.

Identifying bacteria that enhance health and growth promotion and which bacteria can collaborate with each other – interaction between microbiota components.

Determining effect of faecal transplantation for specific issue and species

Immune response to transplanted microorganisms negating desired effect of faecal transplantation or other manipulations.

Establishing a desirable microbiota may need to be carried out in neo-nates (– unless re-establishing a healthy microbiota after a crises or AB therapy).

Solution Routes

What approaches could/should be taken to address the research question?

Developing tools that can help predict impact of interventions.

Use of probiotics/prebiotics and other dietary supplements.

Dependencies

What else needs to be done before we can solve this need?

Investigating how the MB affects AH and productivity – what is in the microbiota that allows it to do that.

Defining the effect of pro/prebiotics, diet, etc on MB.

Investigating the impact of antibiotics on the microbiota.

Correlating gut/respiratory microbiome with faecal microbiome or biomarkers

Investigating the role of environmental microbiota (i.e., poultry litter) on animal performance and health.

State of the Art

Existing knowledge including successes and failures

Projects

What activities are planned or underway?

Lead Summary [2]

Microbiota modulators (pro/prebiotics and diet)

Research Question

What are we trying to achieve and why? What is the problem we are trying to solve?

Defining the effect of pro/prebiotics, diet, etc on MB.

Understanding mode of action of effective probiotics.

Establishing analytical methods to standardise the pro/prebiotics used.

Disease prevention.

How probiotics can be used to enhance response to vaccination (like an adjuvant effect).

Challenge(s)

What are the scientific and technological challenges (knowledge gaps needing to be addressed)?

Controlled clinical trials involving pro/prebiotics, antibiotics, diet, MB transplants, etc.

Determine sufficient level of depth required to measure positive responses.

Impact on gut wall.

Impact on bacteria in gut – protecting gut epithelia. How the gut environment affects expression of bacterial beneficial antigens, some might be expressed in vitro but not in vivo.

Availability of what we consider as controls (identify controls)

Develop challenge models.

Establishing the mix of organisms in probiotics (probiotics cocktail).

Need to standardise how (functional) probiotics are grown, stored and delivered as this can affect surface molecules presented.

Establishing standards for trials so as to allow comparisons.

Need to study the purity and absolute structures of prebiotics in order to define mechanisms. There is often a difference in the purity of prebiotics (I.e. a treatment may contain many different carbohydrates).

Solution Routes

What approaches could/should be taken to address the research question?

Establishing what is the expected effect (functional effect) of the pre/probiotic to be used (what are the expected outcomes?).

Establishing what aspects need to be considered to assess effectiveness (e.g., daily gain, uniformity, challenge models).

Therapeutic use vs disease prevention vs growth promotion.

Defining microbial functionality affecting uniformity.

Is there a microbial basis to the differences in uniformity.

Dependencies

What else needs to be done before we can solve this need?

Understanding mode of action of effective probiotics.

Establishing analytical methods to standardise the pro/prebiotics used.

State of the Art

Existing knowledge including successes and failures

Projects

What activities are planned or underway?

Lead Summary [3]

Antibiotics/vaccines and the microbiota

Research Question

What are we trying to achieve and why? What is the problem we are trying to solve?

Investigating the impact of antibiotics/vaccines on the microbiota.

How microbiota affect vaccination.

Challenge(s)

What are the scientific and technological challenges (knowledge gaps needing to be addressed)?

Understand if antibiotic treatment alters the core microbiota, opening a “door” for pathogen infection (as observed for Salmonella infection in mice and C. diff in humans).

Dose-dependent effects.

Drug-dependent effect.

Age-dependent effect.

Administration route- dependent effect

Identify commonalities among different antibiotics in their effect on metabolic pathways of different bacteria. – as ones with a broad range can have a similar impact on growth as narrow spectrum ones.

Epigenetic of the host (consequence of early antibiotic treatment in the long term in the host) and possible effects on the next generations

Solution Routes

What approaches could/should be taken to address the research question?

Study microbiome before and after AB use in different age groups of animals.

Determine which organisms are affected by which antibiotics

Randomised controlled trials comparing impact of different antibiotics.

Dependencies

What else needs to be done before we can solve this need?

State of the Art

Existing knowledge including successes and failures

Administration route- dependent effect: evidence from pigs suggests no difference between oral and systemic administration.

AB use has an impact on the microbiome and therefore on the short chain fatty acids which has a knock-on effect on the microbiome as well as growth performance of the host.

Projects

What activities are planned or underway?

Lead Summary [4]

Production and delivery of probiotics

Research Question

What are we trying to achieve and why? What is the problem we are trying to solve?

Establishing analytical methods to standardise the pro/prebiotics used

Challenge(s)

What are the scientific and technological challenges (knowledge gaps needing to be addressed)?

Standardised technical and analytical methods to produce, deliver and use of pro/prebiotics.

Growing anaerobic bacteria (a lot of the bacteria associated with health are strict anaerobes and fermentation companies are not set up to grow these).

Regulatory framework (what organisms can one use and standardisation of the product). Classification as therapeutic or nutritional will impact on this.

Standardisation of dose – may depend on strain of organism.

Develop a stable way to delivery and store probiotics – especially anaerobes

Purity and genomic stability of bacteria grown.

Screen for unwanted bacteria/genomic elements.

Production of all the different types (cocktail) of microorganisms to achieve effect.

Creating an effective ecological system in which other wanted organisms thrive.

Solution Routes

What approaches could/should be taken to address the research question?

Study on effective methods to store and deliver probiotics to target maintaining their viability.

Exploration of further biocompatible materials for delivery of probiotics.

Study on efficacy of delivery routes / effects on performance for different products.

Dependencies

What else needs to be done before we can solve this need?

State of the Art

Existing knowledge including successes and failures

Microencapsulation technologies are available

Several biomaterials for encapsulation are available (e.g. gelatin, chitosan, whey proteins, cellulose acetate, locust bean gum…)

Projects

What activities are planned or underway?

Lead Summary [5]

Beneficial respiratory/gut/skin/reproductive and urinary tract microbiota

Research Question

What are we trying to achieve and why? What is the problem we are trying to solve?

Need to define a healthy/dysfunctional microbiome.

Challenge(s)

What are the scientific and technological challenges (knowledge gaps needing to be addressed)?

Definition of what dysbiosis means is contentious -are we talking about, e.g., loss of keystone taxa, loss of diversity, shifts in metabolic capacity, or blooms of pathogen.

Defining what is dysfunctional needs to be set in the context of variation for a specific situation.

Understanding what mechanisms are important to develop and maintain gut microbiome homeostasis.

Identifying what is a “good” microbiota in different species and situations.

Establish standard sampling techniques (small animals, pigs.) for respiratory tract.

Solution Routes

What approaches could/should be taken to address the research question?

Combination of readouts/tools to measure the microbiome:

DNA, RNA, metabolomics…- combine compositional resolution with functional tools

Case controlled studies that delineate the real from spurious disease associations.

Move beyond statistical associations and define causal relationships

Interventional study designs.

Dependencies

What else needs to be done before we can solve this need?

Baseline composition of healthy respiratory microbiota.

Understanding of anaerobic/aerobic condition.

Culturomics (Standard process for cultivating anaerobic/unculturable bacteria, fungus, protozoa, archaea).

State of the Art

Existing knowledge including successes and failures

Projects

What activities are planned or underway?

Lead Summary [6]

Microbiota and productivity

Research Question

What are we trying to achieve and why? What is the problem we are trying to solve?

Understanding how microbiomes (holobionts) can increase performance.

Challenge(s)

What are the scientific and technological challenges (knowledge gaps needing to be addressed)?

Microbiota functionality.

Phenotype definitions.

Establish a correlation between microbiota and phenotypes.

Standard studies for different species.

Differences of immunity system among species (e.g. monograstrics, ruminats, fish…).

Solution Routes

What approaches could/should be taken to address the research question?

Technologies to study microbiota.

Modelling for data analysis.

Case control-studies.

Deep phenotyping of the population.

Minimal unit for defining microbiota in different species.

Impact of diet on microbiome that affect productivity.

Dependencies

What else needs to be done before we can solve this need?

State of the Art

Existing knowledge including successes and failures

Projects

What activities are planned or underway?

Lead Summary [7]

Microbiota and health

Research Question

What are we trying to achieve and why? What is the problem we are trying to solve?

Understanding how the MB affects AH.

Can we develop prognostic tests for various conditions based on the microbiome?

Appropriate microbiome can replace antibiotic treatment?

Challenge(s)

What are the scientific and technological challenges (knowledge gaps needing to be addressed)?

Developing an understanding of niche ecology.

Relating MB components to health status.

Determining functions of microbiome metabolites (what animal sees).

Identify MB biomarkers that predict disease/health.

Investigating real MB function as compared to predicted one.

Identifying the parameters associated with a healthy microbiome.

Linking taxonomy with function.

Defining cut off values to predict disease/health.

How the changes of the environment influence the genes of host and microorganisms (epigenetics).

Solution Routes

What approaches could/should be taken to address the research question?

Using proper study design (which shall include an appropriate sample size).

(Defining appropriate sample size for experiments.)

Meta-analysis of existing datasets.

(Data analysts at functional level and whole animal physiologists).

Deep phenotyping of the population.

Develop prognostic tests to identify individuals predisposed to disease.

Artificial intelligence/machine learning approaches

Prognostic tests

Dependencies

What else needs to be done before we can solve this need?

Investigating how the microbiota interacts with the host.

Defining what is a dysfunctional microbiome.

Understanding the functional capacity of microbiota (including better annotation and characterisation of the organisms present).

Determine what is shaping the microbiome including the interactions within the microbiome with bacteria, viruses etc... - we don't have a good understanding how it is interacting with itself.

State of the Art

Existing knowledge including successes and failures

Projects

What activities are planned or underway?

Lead Summary [8]

Host-microbiota interaction

Research Question

What are we trying to achieve and why? What is the problem we are trying to solve?

Investigating how the microbiota interacts with the host.

Investigating capacity of MB in accelerating immunity development (adaptive and innate).

Investigate implications of early-life modification

Understanding mode of action of effective probiotics

Challenge(s)

What are the scientific and technological challenges (knowledge gaps needing to be addressed)?

Determine which GIT section is the most predictive of the host response under investigation e.g., microbiota in the small intestine may have a greater impact on the immune response while microbes in the large intestine may have a greater impact on feed efficiency.

Improve understanding of the host's response to microbial metabolites.

Determine how early-life microbiota and subsequent succession affect host performance.

Investigating what is shaping MB (including nonbacterial factors)

Correlating the composition of the microbiota with its activity and link that with the host response – very different organisms may be affecting the host in the same way so it is their functionality that is important but also the same microbe might be having a different function under different circumstances.

Improving understanding of different compartments (interactions between different physiological compartments of the host and barriers e.g. blood udder barrier, gut-brain barrier).

Understand to what extent and how gut microbiota communicates with the brain, and which fraction of the microbiota plays an important role in this communication.

How to integrate all knowledge especially from murine models where there is the ability to disrupt function.

Study the transfer of plasmids and the blocking of their transfer.

Solution Routes

What approaches could/should be taken to address the research question?

For field studies, considering animal in cohorts, as to reduce external confounding factors (e.g., antibiotic treatments, diet) and then see the differences in responses to the common stressors.

Using suitable approaches to investigate the host-pathogen-interaction-HPI (including, but not limited to, -omics techniques) to perform the studies including looking at how it interacts with the immune system - glycomics is often ignored but it is the surface of the microbe and the host as this could be where the interactions are taking place.

Functional models

Integrating the results of functional models, with studies utilising the diversity in the livestock host population.

Dependencies

What else needs to be done before we can solve this need?

Investigating animal genomic and impact on microbiome.

Studying systems biology.

Investigating capacity of MB in accelerating immunity development (adaptive and innate).

Investigate implications of early-life modification.

State of the Art

Existing knowledge including successes and failures

Projects

What activities are planned or underway?

Lead Summary [9]

Environmental microbiota and the host microbiota

Research Question

What are we trying to achieve and why? What is the problem we are trying to solve?

Investigating the role of environmental microbiota (i.e., poultry litter/hatchery) on animal performance and health.

Impact of alternatives on the interaction of animal microbiota and environmental microbiota.

Challenge(s)

What are the scientific and technological challenges (knowledge gaps needing to be addressed)?

Developing an understanding of niche ecology.

Defining the temporal microbiota changes.

Linking the animal microbial ecosystem with the environment.

Global climate change and microbiome survival in farm settings (e.g. in manure).

Solution Routes

What approaches could/should be taken to address the research question?

Defining the gut microbiome of animals on farm (not faecal microbiome).

Characterise microbiota in experimental and productive environments.

Define the role of alternatives products/strategies in surrounding environmental microbiota.

Study mother hen/egg shell/ hatchery impact on adult microbiome.

Dependencies

What else needs to be done before we can solve this need?

State of the Art

Existing knowledge including successes and failures

Projects

What activities are planned or underway?

Lead Summary [10]

Animal factors - genetics/genomics/age and the microbiota

Research Question

What are we trying to achieve and why? What is the problem we are trying to solve?

Investigating animal genomic and impact on microbiome.

Challenge(s)

What are the scientific and technological challenges (knowledge gaps needing to be addressed)?

Delineating the role of host genetic factors on the microbiome.

Solution Routes

What approaches could/should be taken to address the research question?

Investigate a larger population in animals.

Dependencies

What else needs to be done before we can solve this need?

State of the Art

Existing knowledge including successes and failures

Projects

What activities are planned or underway?

Lead Summary [11]

Microbiome (including microbiota, i.e. bacteria, viruses, fungi, protozoa, genetics)

Research Question

What are we trying to achieve and why? What is the problem we are trying to solve?

Understanding the functional capacity of microbiota (including better annotation and characterisation of the organisms present).

Determine what is shaping the microbiome including the interactions within the microbiome with bacteria, viruses etc… - we don't have a good understanding how it is interacting with itself.

Challenge(s)

What are the scientific and technological challenges (knowledge gaps needing to be addressed)?

Developing computation tools, databases (unique database to collect data on animal microbiome), and metabolic modelling.

Diet or food affects the microenvironment on different compartments, for example, by changing the PH.

Culturomics.

Solution Routes

What approaches could/should be taken to address the research question?

Culturomics.

Intestine loops.

Organ-on-a chip.

Organoids from organs with microbiomes.

Dependencies

What else needs to be done before we can solve this need?

State of the Art

Existing knowledge including successes and failures

Projects

What activities are planned or underway?

Lead Summary [12]

Standardised technical and analytical methods to study the microbiome

Research Question

What are we trying to achieve and why? What is the problem we are trying to solve?

Ensuring scientific soundness of technical and analytical approaches for studying the host microbiome (approaches need to be justified and metadata made available).

Challenge(s)

What are the scientific and technological challenges (knowledge gaps needing to be addressed)?

Harmonised technical and analytical methods for studying host microbiome that are accessible.

Standardised techniques and systems (paying attention to don't be too picky.

To define standards for assessing antibiotic impact on gut microbiome/dysbiosis, allowing comparative analysis between drug types/classes.

Big data sharing and analysis.

Solution Routes

What approaches could/should be taken to address the research question?

Transparency and sharing in the selection/justification of technical and analytical approaches for studying the host microbiome (metadata needs to be made available).

Organ-on-a chip.

Organoids.

Software/supercomputers? For big data analysis.

Networking.

Dependencies

What else needs to be done before we can solve this need?

State of the Art

Existing knowledge including successes and failures

Projects

What activities are planned or underway?

Annex V: Taking new alternatives to antibiotics to market

Workshop 9 November 2021

The workshop 'Taking new alternatives to antibiotics to market', organised by STAR-IDAZ IRC and IDRC, was opened by Renée Larocque, senior Program Specialist in Animal Health at IDRC. Thirty-three participants belonging to the public research sector, industry and biotech companies attended the workshop, allowing a great exchange of information among the public and the private sector.

During the opening, the two organisers provided some background information on their activities to develop ATA. IDRC presented their animal health research programs covering several countries around the world, particularly LMIC. The 'Livestock Vaccine Innovation fund' and the 'Innovative Vaccine Veterinary solution for AMR, moving respectively 57 and 28 million Canadian dollars, were introduced. STAR-IDAZ IRC, represented by Maddy Newman, introduced the international network of research funders and programme owners on Animal Health and its priorities for research coordination for a range of diseases and cross-cutting issues, among which research on ATA.

Armando Heriazon, Senior Program Specialist in Animal Health at IDRC, provided an overview of the different stages of a product research development in industrial settings, namely research, development and marketing/commercialisation, and the relative decreasing possibility for successful commercialization of the product at each consequential stage (Figure 1).

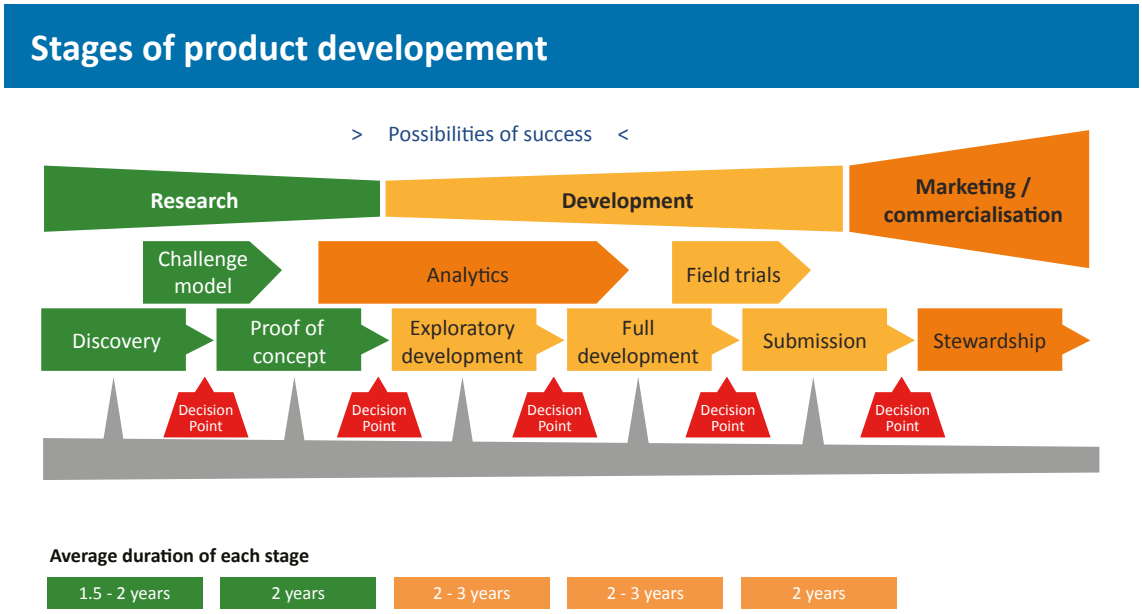


Figure 1: Stages of products developments

There are two main routes for industry to scale-up products: internal knowledge base (internal research project teams) or external knowledge base. In some industries, internal project teams will take care of all the development pipeline from the discovery and proof of concept phases. Nevertheless, as the number of projects that fail to progress is higher at the start of the pipeline, industries often buy external knowledge, e.g. at the stage of proof of concept, from academia and biotech. This is different for 'Developing companies', that do not carry out their own basic research but instead always buy proof of concepts bringing them forward to exploratory development. This opens for continuity of work and greater collaboration between public and private sectors.

Decision points, where it is determined if the product will be brought forward to the next step of development or if the product will be rejected and not progress along the development pipeline, are present at each stage as bottlenecks of the process. After discovery and proof of concept phases of product development, the analytics stage begins as efficacy and effectiveness analysis need to be explored (known as exploratory development). After exploratory development, full development and field trials precede the submission of products to regulators. Before any products get into the development process, it is important to build a product profile. In industry, teams study the product profiles where main characteristics of the product are listed together with targets and top critical features which could be, for example, the presence of similar products in the market, problems with acceptance, blocking points in regulations, minimum effectiveness acceptability. The product profile is important for determining the business strategy regarding the product and the approach to market if any is possible. Once the product passes to the exploratory development phase it is forwarded to a cross-functional project team that aims to deliver projects in a competitive timeline and with costumer focus.

Javier Pozo González, Scientific Administrator, Veterinary Biologicals and Emerging Therapies at the European Medicines Agency (EMA), presented the current EU challenges in taking new alternatives to antibiotics to market from a regulatory perspective. The current challenges are the lack of harmonised definitions for the classification of products. There is currently a wide veterinary range of ATA products with different mechanisms of actions that can be classified as medicinal products, feed additives and biocides. In particular, probiotics, organic acids and phytochemical could be claimed as feed additives or veterinary medicines. Even if the final marketing authorisation is granted by the EU Commission, different regulatory frameworks are applied to each of them, which in turn are evaluated by different competent authorities, e.g.:

- veterinary medicinal products: Directive 2001/82 (Reg EU 2019/6 from 28.01.22) – evaluated by EMA,
- feed additive: Reg EU 1831/2003 - evaluated by EFSA,
- biocide: Reg EU 528/2012 - evaluated by ECHA.

Furthermore, there are novel borderline products, e.g. bacteriophages that could be considered either as feed additives or as veterinary medicines (phage therapy), depending on their application.

Due to these regulatory challenges, it will be important to provide support to developers and applicants from the earliest stages of the development pipeline. SME, spin-off and academia are often not familiar with regulatory framework and requirements. For these reasons, EMA is currently supporting developers and applicants with an innovative task force, providing advice free of charge at any stage.

Moreover, there is a need for financial models and incentives, e.g. pull incentives for most promising products and activities for strategic collaboration and stakeholder engagement, increase communication, tailored advice, roadmaps for target developments and need to increase international collaboration.

Michael Shaw, Founder of Wellspring, a firm that provides market research analysis and investment advice in the agricultural value chain, presented the commercialization part of agri-innovation. He highlighted the importance of market-led innovation value based, which considers the added value of an innovation for the final target, the price that the target is willing to pay and, only after these considerations, if the cost of the production can balance the accepted price. Market intelligence is a fundamental step in product development. A new technology without market would have little or no value. Thus, before proceeding with product development, is important to understand the market, supply chain and customer preferences. Additionally, the Intellectual Property (IP) implications should be considered from the outset of a project, i.e. which type of IP may be generated and how it can be managed.

Finally, the recommendations developed for funders regarding considerations for the ultimate impact of R&D projects were outlined, where applicants should demonstrate an understanding of market analysis, the deliverables related to the needs of both costumers and commercial partners, identify a route to market, IP management and consider the overall impact of technology from the proposal stage.

Simon Labrie, CSO and co-founder of SyntBioLab Inc, a Canadian bioengineering company that develops alternative antimicrobials, presented the lesson learnt from commercialising bacteriophages. He provided first-hand experience of the difficulties faced by start-ups in determining the product patent claim, making sure that the product is appropriate for field applications, understanding the process of registering products, and demonstrating its efficacy and safety, not only for the animal, but also for humans and the environment, which could be prohibitive studies for a start-up.

As regulation issues should be considered when developing a product, some current trade-offs were also discussed regarding the difference in registering products as feed additives rather than veterinary medicines. For example, feed additives can be registered more easily than medical products, however they can't be prescribed as therapeutics, resulting in difficulties marketing them and possible legal implication for farmers and veterinarians in case of disease outbreaks.

Regarding marketing issues, social sciences studies could facilitate understanding the reasons that move farmers to utilise particular products over others and find the best ways to increase farmers trust in new products. This could increase the success of ATA.

The discussion highlighted that antibiotics are competitors of alternatives to antibiotics. The only way to substitute the usage of antibiotics with their alternatives is to produce them in a cost-effective way. Thus, it is important to build financial models and evaluate incentives to bring ATA to market in different settings.

Finally, the panel discussion focussed on how to accelerate the process of product development. Ideas incubators and events that increase communication between researchers and industries were identified as routes to support researchers. Moreover, international collaboration among regulators for facilitating authorisation of borderlines products could speed up authorisation to market.

Annex VI: Agendas of the workshops

Alternatives to Antibiotics Research Gap Analysis Workshop 19 December 2019

The Berkeley Hotel Pratunam, Bangkok, Thailand
Chelsea A, 5th Floor

AGENDA

9:00: Meeting start

1. Welcome and overview of the workshop objectives
2. STAR-IDAZ International Research consortium
   a. Research Roadmaps
3. Structuring the research needs on Alternatives to Antibiotics
   a. Logical Framework

10:45-11:00 Tea/coffee

4. Research needs relating to alternatives to antibiotics that act directly on the pathogen
   a. Bacteriophage Phytochemicals
   b. Phytochemicals
      c. Antimicrobial Peptides
      d. Others?
5. Research needs relating to agents/compounds that enhance the hosts' resistance to disease
      a. Chemicals/phytochemicals
      b. Bacteria/bacterial products
      c. Others?

13:00-14:00 Lunch

6. Research needs to establish how antibiotics work as growth promoters
7. Research needs relating to the role of the microbiome in the maintenance of health

15:30-15:45 Tea/coffee

8. Cross-cutting issues relating to the
9. Research priorities
10. General Discussion and next steps

17:00: Meeting end

Goal of STAR-IDAZ IRC:

To coordinate research at the international level to contribute to new and approved animal health strategies for at least 30 priority diseases/infections/issues.

Objectives of the meeting:

To identify the most promising approaches as alternatives to antibiotics

To identify and prioritise the research needs relating to taking new alternatives to antibiotics forward to market.

To identify the research needs to how antibiotics worked as growth promoters.

Alternatives to Antibiotics Webinar 1

Alternatives to antibiotics acting directly on the pathogen, including establishing their mode of action (with a focus on phage technologies)

Tuesday 12th October 2021
16:00 – 18:00 CET
AGENDA

1. Welcome and overview of the workshop objectives
2. Background to IDRC and STAR-IDAZ International Research consortium
   a. Overview
   b. Research roadmaps
   c. Logical Framework for structuring research needs on ATA
3. Target Product Profile
4. Validate previous consultation of research needs
5. Prioritisation of research gaps

Goal of STAR-IDAZ IRC:

To coordinate research at the international level to contribute to new and approved animal health strategies for at least 30 priority diseases/infections/issues.

Objectives of the meeting:

To identify and prioritise the research needs relating to identify alternatives to antibiotics acting directly on the pathogen, including establishing their mode of action (with a focus on phage technologies) and taking these to market

To validate the previous consultations relating to new alternatives to antibiotics acting directly on the pathogen, with a focus on phage technologies.

Alternatives to Antibiotics Webinar 2

Agents and compounds for their ability to enhance the hosts resistance to disease, including establishing their mode of action, with a focus on immunomodulators

Friday 15th October 2021
15:00-17:00 CET
AGENDA

1. Welcome and overview of the workshop objectives
2. Background to IDRC and STAR-IDAZ International Research consortium
   a. Overview
   b. Research roadmaps
   c. Logical Framework for structuring research needs on ATA
3. Target Product Profile
4. Validate previous consultation of research needs
5. Prioritisation of research gaps

Goal of STAR-IDAZ IRC:

To coordinate research at the international level to contribute to new and approved animal health strategies for at least 30 priority diseases/infections/issues.

Objectives of the meeting:

To identify and prioritise the research needs to identify agents/compounds for their ability to enhance the hosts resistance to disease, including establishing their mode of action, with a focus on immunomodulators, and how these could be taken to market.

To validate the previous consultations relating the research needs required to identify agents/compounds with the ability to enhance the hosts resistance to disease, including establishing their mode of action, with a focus on immunomodulators.

Alternatives to Antibiotics Webinar 3

The role of the microbiome in the maintenance of health, and how it can be manipulated

Monday 18th October 2021
15:00-17:00 CET
AGENDA

1. Welcome and overview of the workshop objectives
2. Background to IDRC and STAR-IDAZ International Research consortium
   a. Overview
   b. Research roadmaps
   c. Logical Framework for structuring research needs on ATA
3. Target Product Profile
4. Validate previous consultation of research needs
5. Prioritisation of research gaps

Goal of STAR-IDAZ IRC:

To coordinate research at the international level to contribute to new and approved animal health strategies for at least 30 priority diseases/infections/issues.

Objectives of the meeting:

To identify and prioritise the research needs relating to the role of the microbiome in the maintenance of health, and how it can be manipulated and taking these to market

To validate the previous consultations relating to the role of the microbiome and how it can be manipulated.

Alternatives to Antibiotics Webinar 4

Antibiotics work as growth promoters

Friday 22nd October 2021
15:00-17:00 CET
AGENDA

1. Welcome and overview of the workshop objectives
2. Background to IDRC and STAR-IDAZ International Research consortium
   a. Overview
   b. Research roadmaps
   c. Logical Framework for structuring research needs on ATA
3. Validate previous consultation of and further consider research needs
4. Prioritisation of research gaps

Goal of STAR-IDAZ IRC:

To coordinate research at the international level to contribute to new and approved animal health strategies for at least 30 priority diseases/infections/issues.

Objectives of the meeting:

To identify and prioritise the research needs to establish how antibiotics work as growth promoters

Alternatives to Antibiotics Webinar 5

Taking new alternatives to antibiotics to market

Tuesday November 9th, 2021
15:00-17:00 CET
AGENDA

1. Welcome and overview of the workshop objectives
2. Background to IDRC and STAR-IDAZ International Research consortium
3. Presentation around Bringing alternatives to antibiotics to Market
   a. Armando Heriazon (IDRC): Product development – private sector approach
   b. Javier Pozo Gonzalez (EMA): Perspectives from a regulatory agency
   c. Michael Shaw (Wellspring Inc.): Commercialization and Knowledge of markets
   d. Simon Labrie (SyntBioLab): Commercialization of bacteriophages – lessons learned
4. Discussion and identification of challenges and solutions

Annex VII: Logical Framework

LOGICAL FRAMEWORK – A focussed approach to research on alternatives to antibiotic use in livestock production

Narrative Summary	Objectively Verifiable Indicator(s)	Means of Verification	Important Assumptions
Goal			
To reduce the use of antibiotics in livestock, decreasing the development of resistance to antimicrobials in livestock, while maintaining/enhancing production levels and controlling the occurrence of disease.	Reduction in the prevalence of resistant organisms/AMR genes. Livestock production data - feed conversion rate. Data on prevalence of diseases. Antibiotics sales data.	Public and private sector monitoring reports on animal production, antibiotic usage and prevalence of resistant organisms.	That verification data is available at relevant scales.
Purpose			
New non antibiotic-based products and approaches for controlling infections and enhancing productivity while maximising the life of the therapeutics: • New technologies that allow for herd-specific vaccines; • Immunomodulators; • Pre- and probiotics or other nutritional supplements; • Bacteriophages; • Antimicrobial peptides; • New delivery systems for precision therapy.	Product registration.	Data produced by regulators.	That the various products are effective in controlling infections. That the various agents can be licensed (e.g. as veterinary medicines or feed additives). That the various agents are acceptable to livestock producers. That the various agents can be produced/ marketed at a cost competitive to use of antibiotics. Cost benefit analysis supports their uptake.

Outputs

Improved understanding of the role of the microbiome in maintenance of health. Improved understanding of the role of antibiotics in growth promotion. Improved understanding of the relationship between control of sub-clinical infection and growth. Identity and mode of action of putative new products– targeting the pathogen and/or the host and/or the microbiome. New technological systems developed for vaccination and/or precision therapy.	Peer reviewed scientific papers	Pubmed, WoS, CABI, Google Scholar databases	No harmful residues That administration of the various agents is practicable in terms of volume required and route and frequency of administration. That anti-infective peptides can be mass-produced synthetically in an active form. That the immunostimulants and bacteriophages have sufficient spectrum width to make their use practicable. That bacterial evolution doesn't rapidly render bacteriophages ineffective. That manipulation of the gut microbiome is practicable and doesn't conflict with other reasons for its manipulation (e.g. to reduce methane production).

Activities

Establishing the makeup of the microbiome in health and different disease situations. Screening target agents/ compounds for their ability to enhance the hosts resistance to disease. Establishing how antibiotic growth promoters work (e.g. compartmentalisation of resources). Development of alternatives to antibiotics, including establishing their mode of action. Developing new technological solutions for precision therapy and vaccination.	Funded research projects	STAR-IDAZ research project database	That animal derived anti-infective peptides can be isolated and produced in an active form. That animal derived anti-infective peptides can reach infection sites. That probiotic and prebiotic agents can be established at target sites in sufficient numbers to competitively exclude a pathogen. That bacteriophages can be delivered in an active form to infection sites.

STAR-IDAZ
International Research
Consortium on Animal Health