Australian Government
Department of Health and Aged Care
Therapeutic Goods Administration

Requirements for microorganism characterisation in Listed Medicines and Registered Complementary Medicines

Guidance for applicants

Version 1.0, February 2023

Contents

Abbreviations

ASTAG	Australian Strategic and Technical Advisory Group on Antimicrobial Resistance
CFU	Colony Forming Units
EFSA	European Food Safety Authority
GCP	Good Clinical Practice
GLP	Good Laboratory Practice
MoA	Mechanisms of Action
MIC	Minimum Inhibitory Concentration
NATA	National Association of Testing Authorities, Australia
QPS	Qualified Presumption of Safety
RCM	Registered Complementary Medicine
TAMC	Total Aerobic Microbial Count
TYMC	Total Yeast and Mould Count
TGA	Therapeutic Goods Administration
VBNC	Viable But Non-Culturable
WGS	Whole Genome Sequencing

Overview

This guidance provides information for applications relating to microorganisms as active ingredients for use as new substances in listed medicines, or as active ingredients in registered complementary medicines (RCM). Listed medicines and RCM containing microorganisms as active ingredients are generally referred to as probiotics or postbiotics.

This document provides tailored guidance that is not covered in the Australian Regulatory Guidelines for listed medicines and RCM and ensures that microorganisms, when identified and characterised, are safe for their intended use in keeping with modern best practice.

Scope

For the purpose of this guidance, microorganisms are whole and intact cells of bacteria and fungi (including yeasts) that are live or non-viable. This guidance is intended for the pre-market assessment of new live and whole/intact non-viable microorganisms potentially used as probiotics and postbiotics and should be considered in conjunction with the Mandatory requirements for an effective application to vary the Permissible Ingredients Determination or Mandatory requirements for an effective registered complementary medicine application. This guidance is intended to supplement the existing quality and safety information requirements.

Substances consisting of non-viable microorganisms (e.g. dead, inactivated, deactivated, heat-killed, tyndallised) where the membrane integrity has been compromised such that the cells are no longer whole/intact, bacterial endospores, and viable but non-culturable (VBNC) cells, are not within the scope of this guidance. VBNC cells require different considerations to standard microorganisms, and they may pose unexpected risks to public health that have not been fully considered, and are therefore generally not suitable for listed medicines or RCM. When a live microorganism has been approved for use in listed medicines, this excludes the non-viable form and vice versa. Specific parts of this guidance are referred to in the Application requirements for new substances in listed medicines – Australian regulatory guidelines where relevant information also applies to microorganisms.

Efficacy data requirements and taxonomic changes in microorganisms are not considered in this guidance.

Information required to demonstrate QUALITY for microorganisms not subject to a monograph in a default standard

Microorganisms that are subject to a specific monograph in a default standard (*British Pharmacopoeia*, *European Pharmacopoeia* and the *United States Pharmacopoeia – National Formulary*), are not within the scope of the quality requirements in this section of the guidance, as those substances must comply with the requirements of that specific monograph.

Where a microorganism is not subject to a specific monograph in a default standard, a comprehensive list of specifications must be provided for that strain under evaluation. The core information requirements for quality in this section encompasses the description, manufacturing details, characterisation, specifications and stability testing in microorganisms that will be evaluated as part of the application. For more information, read ‘SECTION B –

Information requirements' in Application requirements for new substances in listed medicines – Australian regulatory guidelines.

All testing that is not described in a default standard, should be validated to ensure performance and reliability of that method according to Note for guidance on validation of analytical procedures: Text and Methodology (CPMP/ICH/381/95) Rev 1. Laboratories should hold Good Laboratory Practice (GLP) or accreditation to ISO/IEC 17025 or equivalent, e.g. National Association of Testing Authorities, Australia (NATA), United Kingdom Accreditation Service, American Association for Laboratory Accreditation.

Summary of information required to demonstrate quality

Table 1 summarises the information required to demonstrate the quality of microorganisms as active ingredients. Note the quality requirements specified in the Mandatory requirements for an effective application to vary the Permissible Ingredients Determination also apply for listed medicine substances. For RCM, the Mandatory requirements for an effective registered complementary medicine application and the relevant Therapeutic Goods Orders also apply.

For listed medicine substances, please read 'SECTION B – Information requirements' in Application requirements for new substances in listed medicines – Australian regulatory guidelines. A reference to this document is provided for microorganisms under the relevant headings in SECTION B – Information requirements. The core information requirement headings in Table 1 also correspond to the same headings in SECTION B – Information requirements.

RCMs must provide quality information consistent with CTD format. As such, the headings below for quality should be read in conjunction with the relevant parts of CTD Module 3 in CTD modules 2, 3, 4 and 5 for registered complementary medicine applications – Guidance for applicants.

Table 1 – Information required to demonstrate QUALITY for microorganisms not subject to a monograph in a default standard

Core information requirement	
Description	Description of the microorganism. Taxonomy information including strain name/code/culture collection reference, culture origin and method of manufacture. State if the microorganism is derived from or contains genetically modified substances. • If the substance is derived from a genetically modified organism, or genetically modified organism is used during manufacture, demonstrate absence of this in final substance. • If the substance is a live microorganism that has been genetically-modified, provide a declaration that the organism is exempt under Schedule 2 of the *Gene Technology Regulations 2001*.
Manufacturing details	Description of manufacturing process, control of materials, critical steps & intermediates, process development, process validation.

Characterisation	General properties	A list of physico-chemical properties of the microorganism and acceptance criteria. For live microorganisms, also provide: • Antimicrobial resistance and susceptibility profile, antimicrobial resistance genes and horizontal resistance transferability of the microorganism to be characterised using pharmacopeial, or in silico search methods. • Information to address absence of virulence factors of the microorganism using in silico search methods. • Information to address absence of toxigenic and pathogenic attributes using in silico search methods.
	Identity	Strain identification and verification of the microorganism using the most relevant and current valid methodology.
	Assay	Test(s) using pharmacopeial or validated methods, and acceptance criteria that determine the enumeration/viable count of the microorganism.
	Impurities and incidental constituents	A list of contaminant microorganisms that may be present and critical for the quality assurance of the substance, using pharmacopeial or validated methods, and acceptance criteria.
	Reference standard	Information about reference standards for use as the standard in tests, such as identity, assay and impurities testing.
Specifications	Certificates of analysis for minimum two recent commercial-scale batches or three pilot-scale batches, to demonstrate compliance with the proposed specifications. For listed medicines substances, a compositional guideline comprising of a list of tests, reference to validated analytical procedures and appropriate acceptance criteria using the compositional guideline template for microorganisms.	
Stability test	For listed medicine substances, real-time stability testing data for two commercial scale batches or three pilot scale batches. OR For RCMs, stability data to demonstrate the specifications, tests and acceptance criteria in the control of drug substance/product are met for the proposed shelf life.	

Description

The description of the microorganism must be provided as outlined in Mandatory requirements for an effective application to vary the Permissible Ingredients Determination or Module 3 of Mandatory requirements for an effective RCM application.

Provide the taxonomy information (including genus, species and strain name/code/culture collection accession number), culture origin and method of manufacture of the microorganism. The Approved Biological Name (ABN) should be provided, otherwise correspondence regarding naming application must be provided.

Substances derived from or containing genetically-modified organism(s) (GMO) are regulated under the *Commonwealth Gene Technology Act 2000* and *Gene Technology Regulations 2001* which includes regulating import, manufacture, transport, storage and disposal. You should contact the Office of the Gene Technology Regulator (OGTR) early in the process of considering importing, manufacturing or supplying a GMO in a therapeutic good. Refer to Types of GMO dealings on the OGTR website or contact OGTR.CDES@health.gov.au. It is the responsibility of applicants to ensure genetically-modified substances comply with the provisions of all relevant legislation.

It is necessary to state if the substance is derived from or contains GMO. If the substance is derived from a GMO, or a GMO is used during manufacture, demonstrate (by assay or assessment) absence of this in the final microorganism.

If the live microorganism that has been genetically-modified, provide a declaration that the organism is exempt under Schedule 2 of the *Gene Technology Regulations 2001*. Please note that a live GMO that is to be used in a listed medicine does not classify as an 'exempt dealing' because an essential criterion for exemption is that it must be contained. As such, applicants need to seek OGTR approval to be able to import, manufacture or supply the GMO in a therapeutic good. Evidence of OGTR approval is not required in your application however you may provide this information or may be asked to provide this during the evaluation.

Manufacturing details

Description of manufacturing process and process controls

Information about culture establishment, master and working seed cultures, culture media, storage conditions, incubation, batch size should be provided using pharmacopoeial or other validated procedures. It is desirable for the applicant to provide the source of the microorganism and its modification history (including genetic engineering steps) during strain development. Some examples of genetic engineering steps include the insertion or deletion of antibiotic resistance gene(s), reduction of toxigenic and/or pathogenic attributes, use of a recombinant protein expressing virulence factor of a pathogen. If an applicant would like to request that data contained in an application remain commercially confidential, see Treatment of information provided to the TGA.

For non-viable microorganisms, the inactivation method must also be described. As different inactivation methodologies and processes may affect the integrity of the cellular membrane, applicants should include steps taken to ensure and demonstrate that the cells remain whole and intact after inactivation.

For listed medicines substances, other manufacturing details should be provided as per the headings ‘Control of materials’, ‘Controls of critical steps and intermediates’, ‘Manufacturing process development’ and ‘Manufacturing process validation and/or evaluation’ of ‘SECTION

B – Information requirements' in Application requirements for new substances in listed medicines – Australian regulatory guidelines.

For RCMs, other manufacturing details should be provided as outlined in relevant parts of CTD Module 3 in CTD modules 2, 3, 4 and 5 for registered complementary medicine applications – Guidance for applicants.

Characterisation

Genetic differences from strain to strain of a species translate to coding of different proteins; such differences may be functional and have implications on the safety and quality of the microorganisms as active ingredients. Characterisation of the strain under evaluation should entail elucidation of its identity, establishing a suitable assay test, and identifying impurities and incidental constituents.

For live microorganisms, characterisation should also include details of the antimicrobial resistance and susceptibility profile, and information to address the absence of virulence factors, and toxigenic and pathogenic attributes.

General properties

Physico-chemical properties

Information in relation to the physico-chemical properties relevant to the characterisation of the microorganism should be provided. Examples include appearance, colour, state, texture, smell, solubility, loss on drying, sulphated ash, pH, microscopic and macroscopic morphology etc.

Antimicrobial resistance and susceptibility

Therapeutic goods containing live microorganisms as active ingredients should not increase the risk of transfer of antibiotic resistance to the host. Antibiotics considered should be those relevant to use in human medicines in Australia, and classified by the Australian Strategic and Technical Advisory Group on Antimicrobial Resistance (ASTAG) as high, medium or low in terms of importance (ASTAG, 2018).

Although antifungal drug resistance has not developed at the same rate as antibiotic resistance, therapeutic goods containing live microorganisms as active ingredients should not exert selective pressure on fungal resistance in the host.

Antimicrobial resistance and susceptibility profile

Provide information to demonstrate the antimicrobial resistance and susceptibility of the intended proposed substance or RCM containing live microorganisms as active ingredients. Antimicrobial susceptibility to therapeutic concentrations of at least two commercially-available antibiotics from the low and medium categories (e.g. ampicillin, gentamicin, streptomycin, erythromycin, clindamycin, tetracycline, chloramphenicol) defined by ASTAG should be demonstrated.

Using internationally recognised standard methods, the Minimum Inhibitory Concentrations (MIC) should be determined. The microbiological cut-off values (mg/L) for antimicrobials provided in Table 2 of EFSA's Guidance for characterisation of microorganisms based on published data (EFSA, 2018). For fungi and bacteria not listed in Table 2 of EFSA (2018), validated MIC cut-off values in the literature should be referenced. When the growth of a strain is inhibited at a concentration of a specific antimicrobial equal to or lower (≤) than the established MIC cut-off value, it is classified susceptible. When the strain is able to grow at a

concentration of a specific antimicrobial higher than the established MIC cut-off value (>), then the strain is classified resistant.

In silico search for antimicrobial resistance genes

Where genomic data is available, it is recommended to perform an *in silico* search for antimicrobial resistance genes in at least two maintained databases to have a comprehensive view of relevant safety aspects of the live microorganism under evaluation. *In silico* analyses using databases that capture antimicrobial resistance genes from whole genome data sets enable the identification of any potential for genetic resistance. In general, query sequence hits with at least 80% (at the protein level or nucleotide level) and 70% length of the subject sequence should be reported (EFSA, 2021).

Lack of horizontal resistance transferability

The endogenous microbiome in the host functions as a baseline defence against pathogens. It is therefore not desirable to horizontally transfer a gene that codes for resistance to an antimicrobial agent to the host microbiome. The genome of the live microorganism under evaluation should be free of functional and transferable antimicrobial resistance gene DNA that may translate to an antimicrobial resistance phenotype. Where genomic data is available, use of *in silico* search for mobile or transposable genetic elements such as plasmids, resistance transposons, integrons, genomic islands and insertion sequences is desirable.

Absence of virulence factors

Where genomic data is available for the live microorganism strain under evaluation, *in silico* search for genes encoding for known virulence factors (toxins, cell surface proteins that mediate microbial attachment, cell surface carbohydrates and proteins that protect a microorganism, hydrolytic enzymes that may contribute to the pathogenicity of the microorganism, invasion and adhesion factors) should be undertaken using computational comparisons in published databases, applying the minimum available threshold in the database for the length of coverage. In general, query sequence hits with at least 80% (at the protein level or nucleotide level) and 70% length of the subject sequence should be reported (EFSA, 2021). The analysis outcome should include a list of complete genes encoding recognised virulence factors. The list should also include the function of the encoded protein, percentage of identity and e-value of each gene identified. Equivalent methods are acceptable provided they are scientifically accurate and sound.

If a virulence factor gene is detected, details of any strain development strategy (including genetic engineering) intended to reduce the virulence of the strain used should be provided. Data to demonstrate the resultant reduction in virulence of the microorganism under evaluation should also be provided in the application (EFSA, 2018).

Absence of toxigenic and pathogenic attributes

To ensure the live microorganism under evaluation does not have deleterious effect on the consumer, an *in silico* search for presence/absence of known metabolic pathways involved in toxigenicity should be performed, applying the minimum available threshold in the database for the length of coverage. In general, query sequence hits with at least 80% (at the protein level or nucleotide level) and 70% length of the subject sequence should be reported (EFSA, 2021). These include enterotoxins (haemolysin BL, non-haemolytic enterotoxin, enterotoxin T, enterotoxin FM, enterotoxin K), endotoxins, haemolysins and lecithinase. The history of use of the strain or any close relative should also be provided in the form of published literature, particularly those that have been reported as toxigenic and harbour pathogenic attributes.

If there is a toxigenic or pathogenic attribute, details of any strain development step (including genetic engineering) intended to reduce the toxigenic and/or pathogenic attributes of the strain under evaluation should be provided. Data to demonstrate the resultant reduction in toxigenicity and/or pathogenicity of the microorganism under evaluation should also be provided in the application (EFSA, 2018).

Identity

Identification of the genus, species and strain name/code/culture collection accession number is required for all microorganisms as active ingredients in listed medicines or RCM. The microorganism under evaluation should be deposited in a culture collection that is recognised as an International Depositary Authority under the Budapest Treaty. The applicant should provide evidence for deposition of the microorganism strain issued by the culture collection.

Information for elucidation of strain identity should be submitted using genomic methodology, provided the dataset is complete and the methodology is scientifically robust in identifying the strain. Proteomic or phenotypic identification, if available, can be submitted as supporting data. However, proteomic or phenotypic methods alone are not sufficiently robust to unambiguously identify a microorganism to strain level.

Genomic methods

The genome sequence for a microbial strain is a biological fingerprint for unambiguous identification. Genome identification of the microorganism under evaluation should be established using relevant and current valid methodology. In the event that identification by a specific method is incomplete, an alternative method should be used to complement the incomplete data.

Whole Genome Sequencing (WGS) enables the unambiguous taxonomic identification of microorganisms by comparing sequences to relevant databases or via an *in silico* method for taxonomic assignments. WGS also allows for characterisation of the microbial strain with respect to antimicrobial resistance and susceptibility, virulence factors and toxigenicity/pathogenicity. A WGS report should be prepared by a bioinformatician (or equivalent) after genomic analyses have been performed. The report should contain information such as the DNA extraction method, sequencing platform, strategies and annotation protocol used, genome assembly method used for alignment, reference sequence used for alignment, sequence quality assessment and validation (EFSA, 2021). Equivalent methods are acceptable provided they are scientifically accurate and sound.

Proteomic and phenotypic methods

Proteomic methods primarily based on mass spectroscopy enable the identification of microorganisms to genus, species or strain levels. Phenotypic identification of microorganisms relies upon morphological, physiological and biochemical properties that can yield variable results. Proteomic or phenotypic methods are not required if the identity of the microorganism under evaluation is sufficiently addressed using genomic methodologies. Proteomic or phenotypic methods may be used to provide supplementary data to complement genomic methodologies.

Assay

For live microorganisms used as substances in listed medicines, assay tests provide a validated specification to determine the presence and quantity (content) of the microorganism under evaluation. The enumeration or count of an individual microorganism strain, expressed in ‘colony forming units’ (CFU) or ‘number of viable organisms’, should be determined by a suitable microbial enumeration test.

For non-viable microorganisms used as substances in listed medicines, express the enumeration or count of an individual microorganism strain in 'number of deactivated organisms' using a suitable test for control of microbiological quality. Where the enumeration or cell count is performed prior to the inactivation step, then the assay should be expressed as 'deactivated CFU' determined by a suitable microbial enumeration test.

For RCMs, other information addressing assay should be provided as per relevant parts of CTD Module 3 in CTD modules 2, 3, 4 and 5 for registered complementary medicine applications – Guidance for applicants.

Impurities and incidental constituents

Impurities and incidental constituents are constituents that may be present in a microorganism as contaminants, as by-products of production or arise during processing or storage of a microorganism.

Microbial limit testing is an important attribute for quality assurance of a microorganism. The acceptance criteria for the total aerobic microbial count (TAMC) and total yeast and mould count (TYMC) for microbiological quality of the microorganisms as active ingredients may be provided using suitable pharmacopoeial or validated methods that are applicable to seed lots (as opposed to intermediate or final product). Testing for enumeration and absence of other microbial contaminants should be undertaken using pharmacopoeial procedures or other validated procedures.

For listed medicines substances, other applicable impurities and incidental constituent testing should be provided as per the headings 'Residual solvents', 'Elemental impurities' and 'Other organic or inorganic impurities or toxins' of 'SECTION B – Information requirements' in Application requirements for new substances in listed medicines – Australian regulatory guidelines.

For RCMs, other information addressing testing and characterisation of impurities should be provided as per relevant parts of Module 3 in CTD modules 2, 3, 4 and 5 for registered complementary medicine applications – Guidance for applicants.

Reference standard

A reference standard is a reference material prepared for use as the standard in tests, such as identification, assay and impurities testing. Information should be provided about how these reference standards were established. Identification using genomic methods should include the reference sequence used for alignment.

Specifications

Requirements for specifications for listed medicines substances are described under heading 'Specifications' of 'SECTION B – Information requirements' in Application requirements for new substances in listed medicines – Australian regulatory guidelines. You should provide specifications in the form of a compositional guideline using the compositional guideline template specific for microorganisms. The information in the compositional guideline will be derived from the information you provide during the application such as the description, characterisation (identity, assay, impurities) analytical methods and acceptance criteria.

For RCMs, requirements for specifications of active ingredients and finished product are outlined in relevant parts of Module 3 in CTD modules 2, 3, 4 and 5 for registered complementary medicine applications – Guidance for applicants.

Stability test

For listed medicines substances, the requirements for stability test are outlined under heading 'Stability test' of 'SECTION B – Information requirements' in Application requirements for new substances in listed medicines – Australian regulatory guidelines.

For RCMs, information addressing stability of active ingredients and finished product should be provided as specified in relevant parts of Module 3 in CTD modules 2, 3, 4 and 5 for registered complementary medicine applications – Guidance for applicants.

Information required to demonstrate SAFETY

Summary of information required to demonstrate safety

The safety of a substance for use in listed medicines may be supported by history of use, published literature and/or original study data. A combination of data from human exposure information and *in vivo* and *in vitro* nonclinical studies can be used to address the toxicological profile data requirements. Pre-clinical studies should be performed in laboratories that hold GLP or accreditation to ISO/IEC 17025 or equivalent, e.g. NATA, United Kingdom Accreditation Service, American Association for Laboratory Accreditation. Clinical trials should be undertaken in accordance with Good Clinical Practice (GCP) or generally accepted scientific standards. Clinical and other efficacy data, while not evaluated from an efficacy perspective, often include information on tolerability and adverse events that are useful in the safety evaluation.

Table 2 summarises the information required to demonstrate the safety of microorganisms as active ingredients for use in listed medicines or RCM. Note the safety requirements specified in the Mandatory requirements for an effective application to vary the Permissible Ingredients Determination and Mandatory requirements for an effective registered complementary medicine application also apply.

For listed medicine substances, please read 'SECTION B – Information requirements' in Application requirements for new substances in listed medicines – Australian regulatory guidelines. A reference to this document is provided for microorganisms under the relevant headings in SECTION B – Information requirements. The core information requirement headings in Table 2 also correspond to the same headings in SECTION B – Information requirements.

RCMs must provide safety information consistent with CTD format. As such, the headings below for safety should be read in conjunction with the relevant parts of CTD Modules 4 and 5 in CTD modules 2, 3, 4 and 5 for registered complementary medicine applications – Guidance for applicants.

Table 2 – Information required to demonstrate SAFETY

Core information requirement (required for each route of administration proposed)	
Systematic literature search	A systematic literature search on the microorganism; with the search strategy and results with justification for inclusion/exclusion of data.

History and pattern of human use	Information on: • Use in therapeutic goods (Australian and International) • Use in food • Traditional use • History of safe use • Summary of overall human exposure from all sources.	
Biological activity	Pharmacokinetics	Demonstrate absence of microorganism in the systemic circulation of the host
	Pharmacodynamics	• Information on mechanisms of action • Pharmacodynamic interactions with: - Antibiotics/antifungals (only applicable to live microorganisms) - Host immunity
Toxicological data	Information from *in vitro* studies, animal studies, human clinical studies or other information (or a combination) addressing: • Maximum daily dosage • Duration of use • Carcinogenicity (if continuous use of at least 6 months intended) • Reproductive and developmental toxicity (if there are no restrictions proposed in the application that limit use of the microorganism for use in pregnant or lactating females, or in a paediatric population < 18 years) • Local tolerance	
Adverse reactions	A list of the nature, severity and frequency of adverse reactions from adverse event databases, clinical trials, or case reports of human infection.	

Systematic literature search

A systematic literature search on the microorganism strain under evaluation should be provided as outlined under heading ‘Systematic literature search’ of ‘SECTION B – Information requirements’ in Application requirements for new substances in listed medicines – Australian regulatory guidelines or Module 4 of Mandatory requirements for an effective RCM application. The search strategy and results should be provided with justification for inclusion or exclusion of data.

History and pattern of human use

The information in this section should determine the existing exposure of the consumer population to the microorganism. For both listed medicines substances and RCMs, information should be provided as outlined under heading ‘History and pattern of human use’ of ‘SECTION B – Information requirements’ in Application requirements for new substances in listed medicines – Australian regulatory guidelines to address the following:

- Use in therapeutic goods (Australian and International)
- Use in food
- Traditional use
- History of safe use (discussed below)
- Summary of overall human exposure from all sources

History of safe use

Many microorganisms have been in use in food for a substantial length of time, and they usually belong to species with a documented history of safe use. The Qualified Presumption of Safety (QPS) procedure was developed to provide a harmonised generic pre-evaluation to support safety risk assessments of biological substances performed by scientific panels of the European Food Safety Authority (EFSA). EFSA's list of microorganisms classified QPS is founded on assessment of the taxonomic identity, body of knowledge, safety concerns and intended use (EFSA, 2022). The QPS status of a microorganism can be used as information to validate its history and pattern of safe human use. For both listed medicines substances and RCMs, discussion should address the length of time the microorganism has been in use, the level of human exposure from the existing use, the frequency of use in the current setting, and what existing safety monitoring or mechanisms to detect safety signals are available to provide an assurance of safety.

If relying on or leveraging on safety data from other strains that are not the strain under evaluation (if for example you are referring to the QPS status of a microorganism to validate the history of safe use), applicants should establish the equivalence of the microorganism strain under evaluation with data for other microorganism strains by comparing the aspects covered under headings 'Antimicrobial resistance and susceptibility', 'Absence of virulence factors', 'Absence of toxigenic and pathogenic attributes' and 'Identity' under 'Information required to demonstrate QUALITY for microorganisms not subject to a monograph in a default standard'. This can be provided in the form of a comparison table as per Table 3 below.

Table 3 – Table of comparison to establish equivalence between microorganism strains

	Strain under evaluation	Comparable strain A	Comparable strain B	Implication of differences on safety
Identity				
Method of identification % similarity in identities compared with the strain under evaluation				
Characterisation				
Antimicrobial resistance/susceptibility profile				

Antimicrobial resistance genes				
Horizontal resistance transferability				
Virulence factors				
Toxigenic and pathogenic attributes				

Biological activity

Data in this section should be provided for each route of administration proposed for listed medicine substance or RCM in the application.

Pharmacokinetics

Pharmacokinetic studies describe the mechanisms by which the active ingredient is processed by the host and are crucial in assessing systemic exposure. It is prudent that the microorganism strain under evaluation does not reach the systemic circulation of the host post-administration. Appropriate studies, such as human exposure, animal *in vivo* and *in vitro*, and/or alternative studies using appropriate experimental models and routes of administration, should be provided to demonstrate the absence of the microorganism under evaluation in the systemic circulation of the host post-administration. It is not sufficient to state that the microorganism is not absorbed in the gastrointestinal tract or has low bioavailability.

For novel microorganisms, studies using the strain under evaluation should be provided to demonstrate its absence in the systemic circulation of the host.

For microorganisms with a history of safe use, a literature search of the microorganism species should be provided to demonstrate that the strain under evaluation does not enter the systemic circulation. The relevance of any data from microorganisms that are not the strain under evaluation should be comprehensively established as outlined in Table 3.

Pharmacodynamics

The objectives of pharmacodynamics (mechanisms of action, MoA) studies are to identify undesirable pharmacodynamic properties of microorganisms that may have relevance to its human safety. They also facilitate the evaluation of adverse effects of the microorganism observed in toxicology and/or clinical studies and therefore assist in investigating the mechanism of adverse effects observed and/or suspected. Appropriate studies, including human exposure, animal *in vivo* and *in vitro*, and/or alternative studies using appropriate experimental models and routes of administration, should be provided on:

- Mechanisms of action: e.g. antimicrobial activity, immunomodulating activity, inhibition of growth of pathogens, effects on epithelial barrier function, anti-inflammatory activity etc.

For novel microorganisms, pharmacodynamic studies using the strain under evaluation should be provided to address its MoA.

For microorganisms with a history of safe use, a literature search of the microorganism species should be provided to demonstrate the MoA of the strain under evaluation. The relevance of any data from microorganisms that are not the strain under evaluation should be comprehensively established as outlined in Table 3.

Known interactions

Known interactions of the microorganism under evaluation should specifically address interactions with antibiotics/antifungals (applicable to live microorganisms only) and the host immunity as detailed below.

Interactions with antibiotics/antifungals (only applicable to live microorganisms)

Antibiotics/antifungals may be affected by medicines that consist of live microorganisms when co-administered. Therefore, interactions with antibiotics/antifungals should be addressed by means of a literature search for interaction with antibiotics/antifungals for the live microorganism under evaluation. On a case-by case-basis, warning statements 'To be administered X hrs before or after [list antibiotics]' for bacteria or 'To be administered X hrs before or after [list antifungals]' for fungi, or words to that effect may be included depending on the microorganism under evaluation.

Interactions with the host immunity

Therapeutic goods containing microorganisms as active ingredients have been shown to modulate immunological markers. Therefore, the interactions with the host immunity should be addressed by means of a literature search for immunomodulatory properties of the microorganism under evaluation. This is especially important for patients who use an agent/treatment that stimulates or suppresses the immune system (immunomodulators). If the strain under evaluation demonstrates undesirable effects on the immune system, a warning statement may be required such as 'May not be suitable for someone taking immunomodulators. Consult your health professional before taking with other medicines', or words to that effect depending on the microorganism under evaluation.

Toxicological data

The core principles of each toxicological endpoint defined as per heading 'Toxicological data' of 'SECTION B – Information requirements' in Application requirements for new substances in listed medicines – Australian regulatory guidelines or relevant parts of Modules 4 and 5 in CTD modules 2, 3, 4 and 5 for registered complementary medicine applications – Guidance for applicants should be addressed in all applications.

The following toxicological endpoints determine if safety can be established for the intended dosage, duration and population for the microorganism under evaluation. These should be addressed by a combination of *in vitro* studies, animal studies, other suitable alternative and/or clinical trials in humans:

- Maximum daily dosage
- Duration of use aligned with proposed use
- Carcinogenicity (if continuous use of at least 6 months intended)
- Reproductive and developmental toxicity (if there are no restrictions proposed in the application that limit use of the microorganism for use in pregnant or lactating females, or in a paediatric population < 18 years)
- Local tolerance

For novel microorganisms, information using the strain under evaluation should be provided to address the above toxicological endpoints.

For microorganisms with a history of safe use, a literature search for *in vitro* studies, animal studies, other suitable alternatives and/or clinical trials in humans undertaken using equivalent strains of the microorganism should be provided to address the above toxicological endpoints . The relevance of any data from microorganisms that are not the strain under evaluation should be comprehensively established as outlined in Table 3.

Adverse reactions

All reports, published and unpublished and individual case reports relevant to the safety of the microorganism strain under evaluation should be submitted and addressed. These can be from adverse event databases, from clinical trials using the strain under evaluation, or case reports. For both listed medicines substances and RCM, include information on the nature, severity and frequency of adverse reactions as outlined under heading 'Adverse reactions' of 'SECTION B – Information requirements' in Application requirements for new substances in listed medicines – Australian regulatory guidelines.

References

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Version history

Version	Description of change	Author	Effective date
V1.0	Original publication	Complementary and Over the Counter Medicines Branch	February 2023

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Reference/Publication # D22-5888157