

Checklist¹ of Cumulative Required Evidence for a Prebiotic²

Criterion	Description	Check	Comments
Defined structure, purity and stability	Substance is sufficiently described - e.g., source, purity, chemical and structural characterisation , stability - to enable robust data comparisons and reproducible manufacture.	<input type="checkbox"/>	
Proposed mechanism linking microbiome changes to health benefit	Mechanistic rationale to explain how microbiota changes elicit the health benefit can be informed by many different experimental tools and model systems, such as genome mining, <i>in silico</i> screening, <i>in vitro</i> tests, animal models and machine learning.	<input type="checkbox"/>	
Selective utilization in target host	Substance is selectively utilised by the microbiota as a substrate. In addition substance must elicit detectable changes in microbial taxonomic composition (one or more taxa) and/or function in one or more well-conducted trials in the target host.	<input type="checkbox"/>	
Health benefit in target host	Health benefit demonstrated in one or more controlled, well-conducted trials in the target host.	<input type="checkbox"/>	
Selective utilisation and health benefit demonstrated concurrently in target host	Selective utilisation as measured by microbiota modulation (composition and/or function) and health benefit are observed concurrently in the same study.	<input type="checkbox"/>	
Safe for intended use	Safety data and adverse events tracked in studies conducted in target host with acceptable safety record.	<input type="checkbox"/>	
Dose and formulation	Recommended dose and parameters for use are supported by evidence from a study in the target host for both microbiota modulation and health benefit. The formulation used to deliver the prebiotic should maintain integrity and functionality throughout shelf-life.	<input type="checkbox"/>	
Confirmatory evidence (optional)	Additional randomized controlled trials to increase confidence in findings. Demonstration of causal link between prebiotic consumption, microbiota modulation and health benefit via statistical approaches useful but not essential.		

¹This checklist is derived from principles expounded in [Gibson et al. 2017](#) and [Hutkins et al. 2024](#), and should be used in conjunction with these papers.

²Definition of prebiotic - *a substrate that is selectively utilized by host microorganisms conferring a health benefit*