STATEMENT

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Update of the list of QPS-recommended biological agents intentionally added to food or feed as notified to EFSA 11: suitability of taxonomic units notified to EFSA until September 2019

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Abstract

Qualified presumption of safety (QPS) was developed to provide a generic safety evaluation for biological agents to support EFSA's Scientific Panels. The taxonomic identity, body of knowledge, safety concerns and antimicrobial resistance are assessed. Safety concerns identified for a taxonomic unit (TU) are where possible to be confirmed at strain or product level, reflected by 'qualifications'. No new information was found that would change the previously recommended QPS TUs and their qualifications. The list of microorganisms notified to EFSA was updated with 54 biological agents, received between April and September 2019; 23 already had QPS status, 14 were excluded from the QPS exercise (7 filamentous fungi, 6 *Escherichia coli*, *Sphingomonas paucimobilis* which was already evaluated). Seventeen, corresponding to 16 TUs, were evaluated for possible QPS status, fourteen of these for the first time, and *Protaminobacter rubrum*, evaluated previously, was excluded because it is not a valid species. Eight TUs are recommended for QPS status. *Lactobacillus parafarraginis* and *Zygosaccharomyces rouxii* are recommended to be included in the QPS list. *Parageobacillus thermoglucosidasius* and *Paenibacillus illinoisensis* can be recommended for the QPS list with the qualification 'for production purposes only' and absence of toxigenic potential. *Bacillus velezensis* can be recommended for the QPS list with the qualification 'absence of toxigenic potential and the absence of aminoglycoside production ability'. *Cupriavidus necator, Aurantiochytrium limacinum* and *Tetraselmis chuii* can be recommended for the QPS list with the qualification 'production purposes only'. *Pantoea ananatis* is not recommended for the QPS list due to lack of body of knowledge in relation to its pathogenicity potential for plants. *Corynebacterium stationis, Hamamotoa singularis*, *Rhodococcus aetherivorans* and *Rhodococcus ruber* cannot be recommended for the QPS list due to lack of body of knowledge. *Kodamaea ohmeri* cannot be recommended for the QPS list due to safety concerns.

Keywords: safety, QPS, bacteria, yeast, *Aurantiochytrium limacinum*, *Bacillus velezensis*, *Corynebacterium stationis*, *Cupriavidus necator*, *Hamamotoa singularis*, *Kodamaea ohmeri*, *Lactobacillus parafarraginis*, *Paenibacillus illinoisensis*, *Pantoea ananatis*, *Parageobacillus thermoglucosidasius*, *Protaminobacter rubrum*, *Rhodococcus aetherovorans*, *Rhodococcus ruber*, *Tetraselmis chuii*, *Zygosaccharomyces rouxii*

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Amendment: The hyperlink sending to Appendix A on Zenodo has been corrected on page 3. This editorial correction do not materially affect the contents or outcome of this scientific output. To avoid confusion, the older version has been removed from the EFSA Journal, but is available on request, as is a version showing all the changes made.

Erratum: On page 1 in the Abstract, page 3 Summary, page 13 Section 3.1.1.1, and on page 27, the reference to the qualification of *B. velezensis* has been revised from 'absence of aminoglycoside production including the genes encoding it' to 'absence of aminoglycoside production ability'. To avoid confusion, the older version of the output has been removed from the EFSA Journal but is available on request as is a version showing all the changes made.

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Summary

The European Food Safety Authority (EFSA) asked the Panel on Biological Hazards (BIOHAZ) to deliver a Scientific Opinion on the maintenance of the list of qualified presumption of safety (QPS) biological agents intentionally added to food or feed. The request included three specific tasks as mentioned in the Terms of Reference (ToR).

The QPS process was developed to provide a harmonised generic pre-evaluation procedure to support safety risk assessments of biological agents performed by EFSA's scientific Panels and Units. The taxonomic identity, body of knowledge and safety of biological agents are assessed. Safety concerns identified for a taxonomic unit (TU) are, where possible to be confirmed at strain or product level, reflected as 'qualifications' that should be assessed at the strain level by the EFSA's scientific Panels. A generic qualification for all QPS bacterial TUs applies in relation to the absence of acquired genes conferring resistance to clinically relevant antimicrobials (EFSA, 2008).

The list of microorganisms is maintained and re-evaluated approximately every 6 months in a Panel Statement. If new information is retrieved from extended literature searches that would change the QPS status of a microbial species or its qualifications, this is published in the Panel Statement. The Panel Statement also includes the evaluation of microbiological agents newly notified to EFSA within the 6-month period. The main results of the assessments completed from 2017 onwards were included in the scientific Opinion of the BIOHAZ Panel adopted in December 2019. Until July 2019, as a result of each Panel Statement, the '2016 updated list of QPS status recommended biological agents for safety risk assessments carried out by EFSA scientific Panels and Units' was extended by the inclusion of new recommendations for QPS status and appended to the Opinion adopted in December 2016 (Appendix E). The results of the current Panel Statement have been included in the '2019 updated list of QPS status recommended biological agents for safety risk assessments carried out by EFSA scientific Panels and Units'.

The first *ToR* requires ongoing updates of the list of biological agents notified to EFSA, in the context of a technical dossier for safety assessment. The overall list (https://doi.org/record/zenodo.3607184) was updated with the notifications received since the latest review in March 2019. Within this period, 54 notifications were received by EFSA, of which 27 were for feed additives, 15 for food enzymes, food additives and flavourings, 9 for novel foods and 3 for plant protection products (PPPs). The new notifications received between April 2019 and September 2019 are also included in the current Statement (see Appendix F).

The second *ToR* concerns the revision of the TUs previously recommended for the QPS list and their qualifications when new information has become available, and the updating of the information provided in the previous Opinion adopted in December 2016. According to the articles retrieved through an extensive literature search (ELS) protocol available in Appendix B (see https://doi.org/10.5281/zenodo.3607190 and the search strategies in Appendix C (see https://doi.org/10.5281/zenodo.3607193, for articles published from January until June 2019, no new information was found that would affect the QPS status of those TUs and their qualifications.

The third *ToR* requires a (re)assessment of new TUs notified to EFSA, for their suitability for inclusion in the updated QPS list at the Knowledge Junction in Zenodo (https://doi.org/10.5281/zenodo.1146566, Appendix E). The current Statement focuses on the assessments of the TUs that were notified to EFSA between April and September 2019. Of the 54 notifications received, 23 biological agents already had QPS status and did not require further evaluation in this Statement and 14 were not included because: 7 were notifications of filamentous fungi that were excluded from the QPS exercise; 6 were notifications of *Escherichia coli* that were excluded from further QPS evaluations within the current QPS mandate and *Sphingomonas paucimobilis* which was already evaluated in the previous Panel Statement. Seventeen new notifications, corresponding to 16 TUs, were considered for the QPS assessment within this Statement:

Eight TUs have been recommended for the QPS status. *Protaminobacter rubrum*, was evaluated during the previous QPS mandate and still cannot be assessed for a possible QPS recommendation because it is not a valid species name. *Lactobacillus parafarraginis* and *Zygosaccharomyces rouxii* are recommended to be included in the QPS list. *Parageobacillus thermoglucosidasius* and *Paenibacillus illinoisensis* can be recommended for the QPS list with the qualification 'for production purposes only' and absence of toxigenic potential. *Bacillus velezensis* can be recommended for the QPS list with the qualification 'absence of toxigenic potential and the absence of aminoglycoside production ability'. *Cupriavidus necator, Aurantiochytrium limacinum* and *Tetraselmis chuii* can be recommended for the QPS list with the qualification 'for production purposes only'. *Pantoea ananatis* is not recommended for

the QPS list due to lack of body of knowledge in relation to its pathogenicity potential for plants. *Corynebacterium stationis, Hamamotoa singularis*, *Rhodococcus aetherivorans* and *Rhodococcus ruber* cannot be recommended for the QPS list due to lack of body of knowledge. *Kodamaea ohmeri* cannot be recommended for the QPS list due to safety concerns.

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1. Introduction

The qualified presumption of safety (QPS) approach was developed by the European Food Safety Authority (EFSA) Scientific Committee to provide a generic concept for risk assessment within the EFSA of microorganisms intentionally introduced into the food chain, in support of the respective Scientific Panels and Units in the frame of market authorisations (EFSA, 2007a,b). The list, first established in 2007, has been continuously revised and updated. Each 6 months, a Panel Statement is published. These Panel Statements include the results of the assessment of the relevant new papers related to the TUs with QPS status. They also contain the assessment of newly arrived TUs to the EFSA Units (dealing with feed additives, food enzymes, food additives and flavourings, novel foods or plant protection products. After 3 years, a QPS opinion is published summarising the results of the Panel Statements published in that period.

1.1. Background and Terms of Reference as provided by EFSA

1.1.1. Background as provided by EFSA

A wide variety of microorganisms are intentionally added at different stages into the food and feed chain. In the context of applications for market authorisation of these biological agents, used either directly or as sources of food and feed additives, food enzymes and plant protection products, EFSA is requested to assess their safety.

Several taxonomic units (usually species for bacteria, yeasts and protists/algae,[1] families for viruses) have been included in the qualified presumption of safety (QPS) list either following notifications to EFSA or proposals made initially by stakeholders during a public consultation in 2005, even if they were not yet notified to EFSA (EFSA, 2005).[2] The EFSA Scientific Committee reviewed the range and numbers of microorganisms likely to be the subject of an EFSA Opinion and published in 2007 a list of microorganisms recommended for the QPS list.[3]

In 2007, the Scientific Committee recommended that a QPS approach should provide a generic concept to prioritise and to harmonise safety risk assessment of microorganisms intentionally introduced into the food chain, in support of the respective Scientific Panels and EFSA Units in the frame of the market authorisations. The same Committee recognised that there would have to be continuing provision for reviewing and modifying the QPS list and in line with this recommendation, the EFSA Scientific Panel on Biological Hazards (BIOHAZ) took the prime responsibility for this and started reviewing annually the existing QPS list. The first annual QPS update[4] was published in 2008 and EFSA's initial experience in applying the QPS approach was included. The potential application of the QPS approach to microbial plant protection products was discussed in the 2009 update.[5] Also in 2009, bacteriophages were assessed and were not considered appropriate for the QPS list. After consecutive years of reviewing the existing scientific information, the filamentous fungi (2008–2013 updates) and enterococci (2010–2013 updates) were not recommended for the QPS list. The 2013 update[6] of the recommended QPS list included 53 species of Gram-positive non-spore-forming bacteria, 13 Gram-positive spore forming bacteria (*Bacillus* species), one Gram-negative bacterium (*Gluconobacter oxydans*), 13 yeast species, and three virus families.

In 2014 the BIOHAZ Panel, in consultation with the Scientific Committee, decided to change the revision procedure: the overall assessment of the taxonomic units previously recommended for the QPS list is no longer carried out annually but over 3-year periods. From 2017, the search and revision of the possible safety concerns linked to those taxonomic units start to be done every 6 months period. The revision of the 2013 update (EFSA BIOHAZ Panel, 2013) was updated in 2016

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[1] Included during this 3-year period.

[2] Opinion of the Scientific Committee on a request from EFSA related to a generic approach to the safety assessment by EFSA of microorganisms used in food/feed and the production of food/feed additives. EFSA Journal 2005; 226, 1–12.

[3] Introduction of a Qualified Presumption of Safety (QPS) approach for assessment of selected microorganisms referred to EFSA – Opinion of the Scientific Committee. EFSA Journal 2007; 587, 1–16.

[4] Scientific Opinion of the Panel on Biological Hazards on a request from EFSA on the maintenance of the list of QPS microorganisms intentionally added to food or feed. EFSA Journal 2008; 923, 1–48.

[5] Scientific Opinion of the Panel on Biological Hazards (BIOHAZ) on the maintenance of the list of QPS microorganisms intentionally added to food or feed (2009 update). EFSA Journal 2009;7(12):1431, 92 pp. https://doi.org/10.2903/j.efsa.2009.1431

[6] EFSA BIOHAZ Panel (EFSA Panel on Biological Hazards), 2013. Scientific Opinion on the maintenance of the list of QPS biological agents intentionally added to food and feed (2013 update). EFSA Journal 2013;11(11):3449, 107 pp. https://doi.org/10.2903/j.efsa.2013.3449

(EFSA BIOHAZ Panel, 2017a) and the next update will be published in a scientific Opinion of the BIOHAZ Panel after its adoption in December 2019.[7] The QPS list of microorganisms has been maintained and frequently checked, based on the evaluation of extensive literature searches. In the meantime, and every 6 months, a Panel Statement, compiling the assessments for a QPS status of the microbiological agents notified to EFSA requested by the Feed Unit, the Food Ingredients and Packaging (FIP) Unit, the Nutrition Unit or by the Pesticides Unit, has been produced and published. In the follow up of the 2013 update[5] the Scientific Committee agreed to exclude some biological groups (filamentous fungi, bacteriophages and *Enterococcus faecium*[8]) notified to EFSA from the QPS assessment because it was considered unlikely that any taxonomical units within these groups would be granted QPS status in the foreseeable future. Thus, the assessment of members of these biological groups needs to be done at a strain level, on a case-by-case basis, by the relevant EFSA Unit.

The QPS provides a generic safety pre-assessment approach for use within EFSA that covers risks for human, animals and the environment. In the QPS concept a safety assessment of a defined taxonomic unit is considered independently of any particular specific notification in the course of an authorisation process. The QPS concept does not address hazards linked to the formulation or other processing of the products containing the microbial agents and added into the food or feed chain. Although general human safety is part of the evaluation, specific issues connected to type and level of exposure of users handling the product (e.g. dermal, inhalation, ingestion) are not addressed. In the case Genetically Modified Microorganisms (GMMs) for which the species of the recipient strain qualifies for the QPS status, and for which the genetically modified state does not give rise to safety concerns, the QPS approach can be extended to genetically modified production strains (EFSA BIOHAZ Panel, 2018a).[9] Assessment of potential allergenicity to microbial residual components is beyond the QPS remit; if there is however, science-based evidence for some microbial species it is reported. Where applicable these aspects are assessed, separately by the EFSA Panel responsible for assessing the notification. Antimicrobial resistance was introduced as a possible safety concern for the assessment of the inclusion of bacterial species in the QPS list published in 2008 QPS Opinion (EFSA, 2008)[3]. In the 2009 QPS Opinion (EFSA BIOHAZ Panel, 2009)[4] a qualification regarding the absence of antimycotic resistance for yeasts was introduced.

1.1.2. Terms of Reference as provided by EFSA

The Terms of Reference, as provided by EFSA are as follows:

ToR 1: Keep updated the list of biological agents being notified in the context of a technical dossier to EFSA Units such as Feed, Pesticides, Food Ingredients and Packaging (FIP) and Nutrition, for intentional use directly or as sources of food and feed additives, food enzymes and plant protection products for safety assessment.

ToR 2: Review taxonomic units previously recommended for the QPS list and their qualifications when new information has become available. The latter is based on a review of the updated literature aiming at verifying if any new safety concern has arisen that could require the removal of the taxonomic unit from the list, and to verify if the qualifications still efficiently exclude safety concerns.

ToR 3: (Re) assess the suitability of new taxonomic units notified to EFSA for their inclusion in the QPS list. These microbiological agents are notified to EFSA and requested by the Feed Unit, the FIP Unit, the Nutrition Unit or by the Pesticides Unit.

1.2. Interpretation of the Terms of Reference

A wide variety of microorganisms are intentionally used at different stages of the food chain and are risk assessed in several EFSA areas e.g. feed, food, pesticides, nutrition, on the basis of an application dossier to the European Commission. The qualified presumption of safety (QPS) assessment was developed to provide a safety pre-assessment of microorganisms to support the risk assessments performed by EFSA's scientific panels. The lowest taxonomic unit (TU) for which the QPS status is granted is the species level for bacteria, yeasts and protists/algae, and families for viruses. The safety of unambiguously defined biological TUs and their body of knowledge are assessed.

[7] References updated from the original mandate.

[8] The taxonomic unit was corrected from the original mandate: 'enterococci'. It is only referred to *Enterococcus faecium*, the only species which was evaluated for a possible QSP status.

[9] Sentence included, correcting the previous sentence from the original mandate: 'Genetically modified microorganisms are similarly not taken into account'.

In the case that scientific knowledge identifies a specific hazard related to a TU or more generally applicable, e.g. acquired antimicrobial resistance, which can be tested at the strain or product level, a 'qualification' to exclude that hazard may be established. The details of those qualifications in the microbial strain under investigation are evaluated by the EFSA Unit to which the application dossier has been allocated. Microorganisms belonging to bacterial, yeast and protists/algae species or virus families in the QPS list are still submitted to a safety assessment based on the individual data package, although with lesser requirements. The data required in each application have to confirm the unambiguous identification of the organism and the confirmation that the qualifications are met.

The BIOHAZ Panel confirmed that in the case Genetically Modified Microorganisms (GMMs) for which the species of the recipient strain qualifies for the QPS status, and for which the genetically modified state does not give rise to safety concerns, the QPS approach can be extended to genetically modified production strains (EFSA BIOHAZ Panel, 2018a).

In June 2017 (EFSA BIOHAZ Panel, 2017b), the BIOHAZ Panel has agreed to exclude *Escherichia coli* and any species of the genus *Streptomyces* from QPS evaluation.

In June 2018 (EFSA BIOHAZ Panel, 2018b), the BIOHAZ Panel clarified that the qualification 'for production purpose only' implies the absence of viable cells of the production organism in the final product and can also be applied for food and feed products based on microbial biomass.

2. Data and methodologies

2.1. Data

Only valid TUs covered by the relevant international committees on the nomenclature for microorganisms are considered for the QPS assessment.

In reply to ToR 2, concerning the revision of the TUs previously recommended for the QPS list and their qualifications, an extensive literature search (ELS) was conducted as described in Appendix B – ELS protocol, see https://doi.org/10.5281/zenodo.3607190, and in Appendix C Search strategies – see https://doi.org/10.5281/zenodo.3607193, respectively.

In reply to ToR 3, (re)assessment of the suitability of TUs notified within the time period covered by this Statement (from March to September 2019) is carried out. The literature review considered the identification, the body of knowledge, the potential safety concerns and the knowledge on acquired antimicrobial resistance (AMR). Relevant databases, such as PubMed, Web of Science, Cases Database, CAB Abstracts or Food Science Technology Abstracts (FSTA) and Scopus, were searched. More details on the search strategy, search keys and approach are described in Appendix A.

2.2. Methodologies

2.2.1. Evaluation of a QPS recommendation for taxonomic units notified to EFSA

In response to ToR 1, the EFSA Units were asked to update the list of biological agents being notified to EFSA. A total of 54 notifications were received between April and September 2019, of which 27 were for a feed additive, 15 for food enzymes, 9 for novel foods and 3 for plant protection products (Table 1).

In response to ToR 3, of the 54 notifications received, 23 were related to TUs that already had QPS status and did not require further evaluation. Of the remaining 31 notifications, 14 were related to TUs not evaluated for a QPS status for the following reasons:

- Seven notifications related to filamentous fungi, which were excluded from QPS evaluations in the follow-up of a recommendation of the QPS 2013 and 2016 updates (EFSA BIOHAZ Panel, 2013, 2014, 2016).
- Six notifications related to *E. coli*, which were excluded from the current mandate by the BIOHAZ Panel.
- *Sphingomonas paucimobilis* which was already evaluated in the previous Panel Statement.

The remaining 17 notifications, corresponding to 16 TUs were evaluated for a possible QPS recommendation:

- *Protaminobacter rubrum* already evaluated during the previous QPS mandate;
- *Schizochytrium* sp. which is a genus and not a species and, therefore, not adequate for QPS approach.

- *Aurantiochytrium limacinum, Bacillus velezensis, Corynebacterium stationis, Cupriavidus necator, Hamamotoa singularis, Kodamaea ohmeri, Lactobacillus parafarraginis, Paenibacillus illinoisenis, Pantoea ananatis*, *Parageobacillus thermoglucosidasius, Rhodococcus aetherivorans, Rhodococcus ruber*, *Tetraselmis chuii* and *Zygosaccharomyces rouxii*, evaluated for the first time.

The notifications received by EFSA, per risk assessment area and by biological group from April to September 2019, are presented in Table 1.

Table 1: Notifications received by EFSA, per risk assessment area and by biological group, from April to September 2019

Risk assessment area / Biological group	Not evaluated in this Statement		Evaluated in this Statement[(b)]	Total
	Already QPS	Excluded in QPS[(a)]		
Feed additives	**19**	**5**	**3**	**27**
Bacteria	12	1	3	16
Filamentous fungi	0	4	0	4
Yeasts	7	0	0	7
Novel foods	**1**	**3**	**5**	**9**
Bacteria	0	2	1	3
Filamentous fungi	0	1	0	1
Protists/Algae	0	0	4	4
Yeasts	1	0	0	1
Plant protection products	**2**	**0**	**1**	**3**
Bacteria	1	0	1	2
Filamentous fungi	0	0	0	0
Viruses	1	0	0	1
Food enzymes, food additives and flavourings	**1**	**6**	**8**	**15**
Bacteria	1	4	5	10
Filamentous fungi	0	2	0	2
Yeasts	0	0	3	3
Total	**23**	**14**	**17**	**54**

QPS: qualified presumption of safety.
(a): The number includes 7 notifications of filamentous fungi excluded from QPS evaluation in the 2013 QPS Opinion and 6 notifications of *E. coli* (bacterium) already excluded in the Panel Statement adopted in December 2016 (EFSA BIOHAZ Panel, 2017a) and *Sphingomonas paucimobilis* which was already evaluated in the previous Panel Statement (EFSA BIOHAZ Panel, 2019a).
(b): 17 notifications corresponding to 16 TUs, one was already evaluated in the previous QPS mandate (*Euglena gracilis*), 14 were evaluated for the first time. The TU *Schizochytrium* sp. was notified twice but was not assessed as genus is not adequate for the QPS approach.

2.2.2. Use of MLT in the context of the yeasts and *Bacillus* taxonomic units

To explore the potential application of a machine learning technique (MLT) for screening papers in the context of the QPS project, the performances of such technique were assessed against the previous batch of papers retrieved for the *Bacillus* and yeasts taxonomic units.

To that purpose, the DistillerAI Toolkit included in the DistillerSR online software was used.

DistillerAI 'Preview and Rank' function was used mapping the papers from 'Title screening' to 'Article evaluation'. The SVM algorithm with 100% training set and 100% references to preview was used and the references were subsequently tagged. The algorithm was trained on the combined results of the two reviewers in the QPS rounds from 1 June 2016 to 31 December 2017. This is considered a conservative approach since, in the case of conflicts among the experts, the algorithm considers the paper as relevant.

The MLT predicted screening results on the batch of papers corresponding to the period January–June 2018 were obtained and compared with the results obtained by the two reviewers in the real exercise.

The results of the exercise showed that, in the case of yeasts, MLT had around 88% and 80% of sensitivity and specificity, respectively, while, in the case of *Bacillus*, MLT had 100% and about 82% of sensitivity and specificity, respectively. Moreover, it was found that in case of using the MLT algorithm as a reviewer in parallel with a human reviewer, in both projects no information relevant for the QPS status would have been missed.

On the basis of these results and considering the high number of papers retrieved for both yeasts and *Bacillus* in the context of the QPS exercise, it was decided to use the MLT in one ELS exercise in parallel with two human reviewers to screen one 6-month batch of papers in these two TUs. As expected, considering its specificity, the application of the MLT algorithm resulted in a high number of potentially relevant papers at the end of the screening phase. On the other hand, the algorithm did not miss any paper identified as potentially relevant by the human reviewer. Therefore, in the current ELS exercise, the MLT was used for pre-screening of the large number of papers, followed by a second screening by two experts of the articles retrieved by MLT.

2.2.3. Monitoring of new safety concerns related to the QPS list

The aim of the ELS carried out in response to ToR 2 (review of the recommendations for the QPS list and specific qualifications) was to identify any publicly available studies reporting on safety concerns for humans, animals or the environment caused by QPS organisms since the previous QPS review (i.e. publications from July to December 2018). For a detailed protocol of the process and search strategies, refer to Appendices B and C.

After removal of duplicates, 22,899 records were submitted to the *title screening* step (2,981 if we include all references screened initially by MLT for *Bacillus* and yeasts TUs), which led to the exclusion of 2,170 of them (2,862 if we include all references screened initially by MLT). The remaining 119 records were found eligible for the *Title and abstract screening* step, which led to the exclusion of 52 of these. Of the 67 articles that finally reached the *Article evaluation step* (full text), 36 were considered to be relevant for the QPS project and were deeply analysed.

The flow of records from their identification by the different search strategies (as reported in Appendix C) to their consideration as potentially relevant papers for QPS is shown in Table 2.

Table 2: Flow of records by search strategy step

Species	No of papers			
	Title screening step	**Title/ abstract screening step**	**Article evaluation step (screening for potential relevance)[a]**	**Article evaluation step (identification of potential safety concerns)**
Bacteria	**(2,391) 1,833**	**65**	**31**	**15**
Bacillus spp.	741 (183)[b]	15	9	6
Geobacillus stearothermophilus			0	0
Bifidobacterium spp.	270	14	6	1
Carnobacterium divergens			0	0
Corynebacterium glutamicum	39	0	0	0
Gluconobacter oxydans	164	1	0	0
Xanthomonas campestris			0	0
Lactobacillus spp.	620	23	8	2
Lactococcus lactis	165	2	2	0
Leuconostoc spp.	81	2	1	1
Microbacterium imperiale			0	0

Species	No of papers			
	Title screening step	Title/ abstract screening step	Article evaluation step (screening for potential relevance)[(a)]	Article evaluation step (identification of potential safety concerns)
Oenococcus oeni	39	1	1	1
Pasteuria nishizawae			0	0
Pediococcus spp.	166	3	2	2
Propionibacterium spp.	27	0	0	0
Streptococcus thermophilus	79	4	2	2
Viruses	**99**	**5**	5	0
Alphaflexiviridae	41	2	2	0
Potyviridae				0
Baculoviridae	58	3	3	0
Yeasts	**(491) 357**[(b),(c)]	**49**	**31**	**21**
Debaryomyces hansenii (anamorph=Candida famata)	357	49	31	5
Kluyveromyces lactis (anamorph=Candida spherica)				1
Kluyveromyces marxianus (anamorph= Candida kefyr)				13
Saccharomyces cerevisiae including *Saccharomyces boulardii*				7
Total	**(2,981) 2,289**	**119**	**67**	**36**
Excluded	**(2,862) 2,170**	**52**	**31**	

(a): Relevant references in Appendix D.

(b): The relatively low number of papers allocated to title screening for this TU as compared to the previous statement is due to the application of a pre-screening step that was employing artificial intelligence (AI). A search for papers potentially relevant for the QPS consideration of *Bacillus* spp. and *Geobacillus stearothermophilus* provided 741 references. The MLT analysis left 183 articles.

(c): The relatively low number of papers allocated to title screening for this TU as compared to the previous statement is due to the application of a pre-screening step that was employing artificial intelligence (AI). A search for papers potentially relevant for the QPS consideration of the yeast species included in the QPS list provided 491 references. The MLT analysis left 357 articles.

3. Assessment

3.1. Taxonomic units evaluated during the previous QPS mandate and re-evaluated in the current Statement

3.1.1. Bacteria

3.1.1.1. *Protaminobacter rubrum*

Identity

The species *P. rubrum* is not taxonomically validated according to the List of prokaryotic names with standing in nomenclature (LPSN) (Euzéby, 2013) (http://www.bacterio.net/-allnamesdl.html) and the modifications that appear in the International Journal of Systematic and Evolutionary Microbiology (IJSEM) (Oren and Garrity, 2019 and earlier notifications). The organism has been already considered as not suitable for QPS consideration (EFSA BIOHAZ Panel, 2016) and no new relevant information has appeared that might justify changes in this consideration.

Body of knowledge

Not applicable.

Safety concerns

Not applicable.

Antimicrobial resistance aspects

Not applicable.

Conclusions on a recommendation for the QPS list

P. rubrum cannot be assessed for a possible QPS recommendation because it is not a valid species name.

3.2. Taxonomic units to be evaluated for the first time

The search strategy (key words, literature databases, number of papers found) followed for the assessment of the suitability of TUs notified to EFSA not present in the current QPS list for their inclusion in the updated list (reply to ToR 3) can be found in Appendix A.

3.2.1. Bacteria

3.2.1.1. *Bacillus velezensis*

Identity

Bacillus velezensis was first described as a new species by Ruiz-García et al. (2005) and is considered a bacterial species with standing in nomenclature (LPSN bactero.net).

Body of knowledge

The natural habitats of *B. velezensis* are soil and rhizosphere and the marine environment. It has been involved in the fermentation of foods such as kimchi and fermented soybean paste. This species has been used as a plant growth promoting rhizobacterium, in the biological control of plant pathogens and mycotoxigenic fungi and in the detoxification of mycotoxins. Moreover, studies describe the use of *B. velezensis* as probiotic in fish, being able to control fish bacterial pathogens, and in chickens. This species produces compounds of biotechnological interest, such as β-glucanases, L-asparaginase and surfactins.

Safety concerns

No association of *B. velezensis* to intoxication or infection has been reported in humans or animals.

A strain of this species, isolated from marine environment and identified by 16S rRNA gene analysis, was shown to produce an antimicrobial substance that, based on structural analysis, is classified as an aminoglycoside (Pournejati et al., 2019).

Antimicrobial resistance aspects

A recent study (Agersø et al., 2018) addressed the MIC distribution and the presence of genes coding for antimicrobial resistance in five *Bacillus* species, including *B. velezensis*. The tetracycline efflux gene, *tet*(L), was found in strains with reduced tetracycline susceptibility but not in susceptible strains.

Conclusions on a recommendation for the QPS list

Bacillus velezensis can be recommended for the QPS list with the qualifications 'absence of toxigenic potential' and 'absence of aminoglycoside production ability'.

3.2.1.2. *Corynebacterium stationis*

Identity

Corynebacterium stationis (synonym *Achromobacter* stationis; basonym *Brevibacterium* stationis) is a valid species with standing in nomenclature. It was described by Bernard et al. (2010), grouping *Brevibacterium stationis* strains and *Corynebacterium ammoniagenes* ATCC 6872. Members of the species may alkalinise citrate and may be discriminated from other *Corynebacterium* species by 16S rRNA gene and *rpoB* sequencing. The complete genome of *C. stationis* ATCC 6872 has been determined (Liu et al., 2016).

Body of knowledge

The body of knowledge for *Corynebacterium stationis* is limited. The type strain was isolated from seawater and strain ATCC 6872 from a human infant stool sample.

Safety concerns

Two clinical *C. stationis* isolates were obtained from blood cultures from a 62-year-old male with chest infection and a 66-year-old female; no further clinical information has been provided (Bernard et al., 2010). *C. stationis* has been isolated from raw milk of cows with mastitis (Anaya-López et al., 2006; Leon-Galvan et al., 2015); no invasion in Bovine Mammary Epithelial Cells (BMECs) was observed (Anaya-López et al., 2006).

Antimicrobial resistance aspects

Multiresistant *C. stationis* strains were isolated from raw milk samples of cows with mastitis (Leon-Galvan et al., 2015).

Conclusion on a recommendation for the QPS list

Corynebacterium stationis cannot be recommended as QPS due to a lack of body of knowledge.

3.2.1.3. *Cupriavidus necator*

Identity

Cupriavidus necator was first described by Makkar and Casida (1987) and is confirmed by DNA–DNA hybridisation to be the validated species name with standing in nomenclature (Vandamme and Coenye, 2004). Members of the species were formerly named as *Alcaligenes eutrophus, Ralstonia eutropha* or *Wautersia eutropha* (Vaneechouttes et al., 2004). They are Gram-negative bacteria belonging to the family *Burkholderiaceae* and the class β-proteobacteria. The whole genome sequence of *C. necator* strain NH9 and a set of *Cupriavidus* and *Ralstonia* strains confirmed the clear delineation of both genera (Gan, 2019; Moriuchi et al., 2019).

Body of knowledge

C. necator has been reported to pray upon a wide range of Gram-negative and Gram-positive bacteria (Seccareccia et al., 2016). *C. necator* is used as source of polyhydroxybutyrate (PHB) which can be used for the production of bioplastics after recovery from the cell cytoplasm (Aramvash et al., 2015) or by using the dried biomass (Kunasundari et al., 2013). PHBs and the dried biomass of *C. necator* can be used for its antimicrobial, insecticidal and antiviral activities based on the degradation by bacteria of PHB into β-hydroxybutyrate (van Hung et al., 2019). *C. necator* has been genetically modified to produce several compounds as isopropanol, hydrocarbons, methyl ketones, free fatty acids, alkanes etc. (Marc et al., 2017).

Safety concerns

No safety concerns were reported related to *C. necator.*

Antimicrobial resistance aspects

No papers on antimicrobial resistance of *C. necator* were found.

Conclusions on a recommendation for the QPS list

Cupriavidus necator can be recommended for the QPS status with the qualification for production purposes only.

3.2.1.4. *Lactobacillus parafarraginis*

Identity

Lactobacillus parafarraginis is a valid species name according to the List of Prokaryotic Names with standing in nomenclature. It was first described upon isolation from shochu compost (is a sake derived distilled beverage) (Endo and Okada, 2007) and belongs to the *L. buchneri* group of lactobacilli (Salvetti et al., 2018). The genome of five strains has been sequenced.

Body of knowledge

L. parafarraginis is commonly isolated from plant-based fermentative processes such as those of green-olives (Benitez-Cabello et al., 2016), silage (Liu et al., 2014; Wu et al., 2014; Xu et al., 2018), kefir (Zanirati et al., 2015) and sayur-asin (a mustard-based sauce consumed in Indonesia) (Mangunwardoyo et al., 2016). Its presence is also linked to silage aerobic stability due to the production of benzoic and hexadecenoic acids, which inhibit yeast growth (Liu et al., 2018). For this reason, it has been tested as a silage inoculant (Xu et al., 2017).

Safety concerns

L. parafarraginis is used for fermentation of food and feed. Consequently, it is frequently consumed by humans and livestock. There is no report on safety concerns.

Antimicrobial resistance aspects

Tetracycline resistance (Feichtinger et al., 2016) has been found for all species within the *L. buchneri* group, including *L. parafarraginis*. However, no known gene determinants were detected by PCR or microarray analysis.

Conclusions on a recommendation for the QPS list

Lactobacillus parafarraginis is recommended to be included in the QPS list.

3.2.1.5. *Paenibacillus illinoisensis*

Identity

Paenibacillus illinoisenis, previously known as *Bacillus circulans,* group 6, was described by Shida et al. (1997). It is a valid species with standing in nomenclature.

Body of knowledge

P. illinoisensis was isolated from the rhizosphere of soil and characterised for its siderophore-producing capacity, promoting iron absorption by plants (Liu et al., 2017). Strains of *P. illinoisensis* were reported to secrete cyclodextrin gluconotransferase (Doukyu et al., 2003; Lee et al., 2013, chitinases (Jung et al., 2008) and enzymes degrading methane (Jhala et al., 2014).

Safety concerns

No safety concerns were reported for *P. illinoisensis*.

Antimicrobial resistance

No reports were found related to antimicrobial resistance of *P. illinoisensis.*

Conclusion

Paenibacillus illinoisensis can be recommended for QPS with the specific qualifications for production purposes only and absence of toxigenic potential.

3.2.1.6. *Pantoea ananatis*

Identity

Pantoea ananatis is a Gram-negative, motile rod belonging to the Enterobacterales (Adeolu et al., 2016). Its identity is well established, being initially part of the *Erwinia herbicola–Enterobacter agglomerans* complex and later assigned to the genus *Pantoea* (Mergaert et al., 1983). It is a valid species name with standing in nomenclature. Identification can be performed by multilocus sequencing analysis (Brady et al., 2008; Delétoile et al., 2009).

Body of knowledge

The information available deals with its phytopathogenic characteristics, genomic analysis and presence in a variety of habitats. *P. ananatis* has been isolated from the environment and hosts showing global distribution (Weller-Stuart et al., 2017). The capacity of the bacterium to infect agronomic crops is high (maize, rice) and well documented (Weller-Stuart et al., 2017). Among the pathogenicity determinants, T6SS systems have been putatively involved in pathogenesis in both plant and animal hosts (Coutinho and Venter, 2009; De Maayer et al., 2014; Weller-Stuart et al., 2017). The genome plasticity of this species with integration of mobile genetic elements on both the chromosome and on the plasmid LPP-1 (Coutinho and Venter, 2009; De Maayer et al., 2014; Weller-Stuart et al., 2017) has been associated with the variable phenotypes described (e.g. ability to utilise a wide range of carbohydrate, amino acid and organic acid substrates, pathogenicity features and antibiosis).

Pantoea ananatis has the potential to be used for growth promotion of different plants, as a biological control agent against a range of bacterial and fungal plant pathogens and for bioremediation and biofuel production (Gasser et al., 2012; Hara et al., 2012; Gkorezis et al., 2016).

The presence of an Integrative and Conjugative Element (ICEPan) in some strains was associated with the potential production of a new antibiotic and bacteriocins (De Maayer et al., 2015).

Safety concerns

P. ananatis is a phytopathogen that infects a wide range of crop and forest plants such as maize (Miller et al., 2016), rice (Watanabe et al., 1996), onion (Gitaitis et al., 2002; Weller-Stuart et al., 2014), eucalyptus (Coutinho et al., 2002; De Maayer et al., 2010) and it has occasionally been reported as a clinical isolate infecting workers with plant material or from immunocompromised individuals in hospital settings. The number of reports linking the TU with human disease is scarce (De Baere et al., 2004; De Maayer et al., 2012; Manoharan et al., 2012).

Antimicrobial resistance aspects

No information was found.

Conclusions on a recommendation for the QPS list

Pantoea ananatis is not recommended for the QPS list due to lack of body of knowledge and in relation to its pathogenicity potential for plants.

3.2.1.7. *Parageobacillus thermoglucosidasius*

Identity

Parageobacillus thermoglucosidasius is the valid species name with standing in nomenclature (Oren and Garrity, 2019). The basonym is *Bacillus thermoglucosidasius* (Suzuki et al., 1983); *Geobacillus thermoglucosidasius* can be used as homotypic synonym; the name *Geobacillus thermoglucosidans* (Coorevits et al., 2012) has not been accepted as correct name.

Body of knowledge

P. thermoglucosidasius is a facultative anaerobic thermophilic bacterium which is frequently isolated from high temperature environments including hot springs (Brumm et al., 2015) and compost (Sung et al., 2002; Fong et al., 2006; Brumm et al., 2016). The body of knowledge is mainly related to its biotechnological potential for fermentation of plant biomasses (Iwazaki et al., 2018) to generate bio-ethanol (Zhou et al., 2016) and biohydrogen (Mohr et al., 2018). Also, of interest is the production of (heterologous) thermostable enzymes for various industrial applications (Holland et al., 2019) and the biomineralisation potential of this species (Murai and Yoshida, 2016).

P. thermoglucosidasius has been frequently found as spontaneous contaminant in dairy powder products and is isolated in biofilms from the dairy processing industry (Zhao et al., 2013, 2018).

Safety concerns

No safety concerns were reported.

Antimicrobial resistance aspects

No reports considering antimicrobial resistance were found.

Conclusions on a recommendation for the QPS list

Parageobacillus thermoglucosidasius can be recommended for QPS list with the qualification 'for production purposes only' and the absence of toxigenic potential.

3.2.1.8. *Rhodococcus aetherivorans*

Identity

Rhodococcus aetherivorans is a valid species name (Goodfellow et al., 2004) that belongs to the order *Actinomycetales*, suborder *Corynebacterineae*, family *Nocardiaceae*. As all rhodococci, *R. aetherivorans* possesses mycolic acids in its cell wall and presents remarkable catabolic abilities (Goodfellow et al., 2004). The genomes of two strains of the species have been sequenced.

Body of knowledge

The knowledge on the general biology, habitats, symbiotic relations, etc. of *R. aetherivorans* is very scarce. Most papers on the organism deal with its ability to degrade recalcitrant xenobiotics such as diverse petrol components (Auffret et al., 2009), 1-4 dioxane (Inoue et al., 2018) or reducing arsenic (Firrincieli et al., 2019). In addition, *R. aetherivorans* generates polymeric bioplastic components when grown on toluene (Hori et al., 2009).

Safety concerns

No reports on clinical cases produced by *R. aetherivorans* were found.

Antimicrobial resistance aspects

No information was found in the literature.

Conclusions on a recommendation for the QPS list

Rhodococcus aetherivorans cannot be recommended for the QPS list due to lack of body of knowledge.

3.2.1.9. *Rhodococcus ruber*

Identity

Rhodococcus ruber is a valid species name according to the List of Prokaryotic Names with Standing in Nomenclature, although the specific name changed from the original description of the genus, which included the species as *R. rubrus* (Goodfellow and Alderson, 1977). It belongs to the order *Actinomycetales*, suborder *Corynebacterineae*, family *Nocardiaceae*. As all rhodococci, *R. ruber* possesses mycolic acids in its cell wall. Whole-genome data from 12 strains (October 2019) are available.

Body of knowledge

R. ruber is an environmental organism that presents remarkable bioremediation abilities, as well as the capacity to synthesise polymeric bioplastic precursors. The enzymatic machinery behind these activities comprises dehydrogenases (Wang et al., 2017), sterol hydroxylases (Guevara et al., 2017), alkylsulfatases (Pogorevc and Faber, 2003), production of biosurfactants (Ivshina et al., 2016) and many other activities that allow degradation of polyethylene (Orr et al., 2004), polychlorinated biphenyls (Egorova et al., 2013), polycyclic aromatic hydrocarbons (Ivshina et al., 2016) and other recalcitrant contaminants. *R. ruber* has also been used as a model organism for production of polyhydroxybutyric acid (Pieper and Steinbüchel, 1992).

Safety concerns

R. ruber may act as an opportunistic pathogen (Lalitha et al., 2006) and its mycolic acids were able to generate granulomas in the lungs, liver and spleens of mice (Matsunaga et al., 1996).

Antimicrobial resistance aspects

No relevant information was published.

Conclusions on a recommendation for the QPS list

Rhodococcus ruber cannot be recommended for the QPS list due to lack of body of knowledge.

3.2.2. Yeasts

3.2.2.1. *Hamamotoa singularis*

Identity

The species was first described in 1962 as the basidiomycetous yeast *Sporobolomyces singularis*. The name *Bullera singularis* has also been used, after a suggestion by Rodrigues de Miranda (1984). Recently, rRNA sequencing suggested that the species belong to the *Microbotryomycetes* clade and should be broken out from the genus *Sporobolomyces* (Wang et al., 2015). *Hamamotoa singularis* was introduced as new name for *Sporobolomyces singularis* (Wang et al., 2015). The new genus *Hamamotoa* currently contains two species. They are cream to pale yellowish-brown pigmented and form budding cells and ballistoconidia. Sexual reproduction is not known.

Body of knowledge

The body of knowledge is mainly related to its production of β-galactosidase (Ishikawa et al., 2005; Sakai et al., 2008; Kaneko et al., 2014). The species is known from a single strain isolated from insect frass (manure) of a dead Alaska pine (Kurtzman et al., 2011) and no information is available about its ecology, clinical importance or occurrence in agriculture or food.

Safety concerns

No safety concerns were reported.

Antimicrobial resistance aspects

Not relevant.

Conclusions on a recommendation for the QPS list

Hamamotoa singularis cannot be recommended for the QPS list due to lack of body of knowledge.

3.2.2.2. *Kodamaea ohmeri*

Identity

The genus *Kodamaea* belongs to the *Ascomycetae* class and the *Saccharomycetaceae* family and includes five species *K. anthophila*, *K. kakaduensis*, *K. laetipori*, *K. nitidulidarum* and *K. ohmeri*. The last one is considered the type species of the genus. The synonymous of *K. ohmeri* is *Pichia ohmeri* and the teleomorphic form is *Candida guilliermondii* (Kurtzman et al., 2011).

Body of knowledge

K. ohmeri has been recovered from a broad variety of sources. From a biotechnological point of view, this species is used in the production of xylitol from glucose and the conversion of xylulose to xylitol (Kurtzman et al., 2011).

Safety concerns

K. ohmeri has been added to the growing list of emerging opportunistic pathogens (Al-Sweih et al., 2011). Different publications associated *K. ohmeri* with infection in immunosuppressed patients (Tashiro et al., 2018; Diallo et al., 2019) or in premature newborns (Vivas et al., 2016).

Antimicrobial resistance aspects

No relevant information was published.

Conclusions on a recommendation for the QPS list

Kodamaea ohmeri cannot be recommended for the QPS list due to safety concerns.

3.2.2.3. *Zygosaccharomyces rouxii*

Taxonomy

The genus *Zygosaccharomyces* is a member of the *Saccharomycetaceae* family and most closely related to *Torulaspora*, *Zygotorulaspora*, *Vanderwaltozyma* and *Tetrapisispora.* Six species are present in this genus (*Z. bailii*, *Z. bisporus*, *Z. kombuchaensis*, *Z. lentus*, *Z. mellis* and *Z. rouxii*). *Z. rouxii* is considered the neotype of the genus.

Body of knowledge

Z. rouxii is typically found in highly osmotic habitats. Strains of *Z. rouxii* have been isolated from a wide variety of sources, including cane sugar, chocolate syrup, concentrated black grape must, honey, jam, maple syrup, marmalade, marzipan, miso, red wine, salted beans, soft drinks and soy sauce (for a review, see Kurtzman et al., 2011). *Z. rouxii* is used to ferment a number of salted, oriental fermented foods, the best-known being soy sauce and miso. This species is also important in the early stages in the manufacture of balsamic vinegar.

Z. rouxii is primarily a spoilage yeast of high-sugar or high-salt foods, such as sugar syrups, candied fruit and soy sauce.

Safety concerns

No description of humans or animals' infections produced by *Z. rouxii*, was published.

Antimicrobial resistance aspects

No information are available

Conclusions on a recommendation for the QPS list

Zygosaccharomyces rouxii is recommended to be included in the QPS list.

3.2.3. Protists/Algae

3.2.3.1. *Aurantiochytrium limacinum*

Identity

Aurantiochytrium limacinum is a marine protozoa, belonging to the genus *Aurantiochytrium,* composed of unicellular eukaryotes, belonging to the order Thraustochytrida, class Labyrinthulea within the phylum Bigyra (Catalogue of Life, online). The taxonomic identification is mainly based on life cycle and developmental stages. The whole genome sequence of strain CCTCC M209059 has been determined (Ji et al., 2015). *Schizochytrium limacinum* is considered a synonym (Catalogue of Life, online). *A. limacinum* is most often called a microalga, although it is autotrophic and not photosynthetic.

Body of knowledge

A. limacinum strains are known to produce large amounts of docosahexaenoic acid/docosapenaenoic acid (DHA/DPA), eicosapentaenoic acid (EPA), astaxanthin and β-carotene (Liang et al., 2011; Du et al., 2019; Ye et al., 2015; Zhang et al., 2017; Bindea et al., 2018). It may also produce peptides with antioxidant activity (Hu et al., 2019) and can be used for the production of biofuel (Xu et al., 2018). It is able to grow on saline waste water (e.g. demineralisation water from cheese whey) (Humhal et al., 2017), cull potato (Chi et al., 2007) and biodiesel derived crude glycerol (Ethier et al., 2011) for biomass production. A combined effect of the probiotic *Lactococcus lactis* and the prebiotic *A. limacinum* biomass, fed to fish induced positive effects on their growth and immunity (Sun et al., 2019). *A. limacinum* biomass is commercialised and was successfully used as alternative for fish oil in feeding laying hens for the enrichment of table eggs with n-3 fatty acids (Kralik et al., 2019).

Safety concerns

No safety concerns were reported related to *A. limacinum.*

Antimicrobial resistance aspects

Not applicable.

Conclusions on a recommendation for the QPS list

The species *Aurantiochytrium limacinum* is recommended for the QPS status with the qualification for production purposes only.

3.2.3.2. *Tetraselmis chuii*

Identity

Tetraselmis chuii (also sometimes spelled *Tetraselmis chui*) is a unicellular, planktonic microalga belonging to the phylum Chlorophyta (green algae) and family Chlorodendraceae. Members of the genus (currently around 25 species) have four flagella arranged in two pairs and are capable of active movement. The taxonomy of the genus has been described based mainly on morphological and ultrastructural information. A few studies employed rDNA sequencing for species identification in limited collections of *Tetraselmis* strains (Lee and Hur, 2009; Arora et al., 2013; González et al., 2015), however, systematic taxonomic studies of the genus combining morphological and molecular approaches are lacking.

Body of knowledge

The species was first described in the 1950s (Butcher, 1959) but has since then been found in phytoplankton communities in marine and brackish environments around the world. An interesting ecological observation is that the functional chloroplast of *T. chuii* can be retained within the cell in some ciliates that graze the alga and thereby possibly give the ciliate a nutritional supplement by photosynthesis (McManus et al., 2018).

T. chuii is cultured and refined in large-scale facilities and has a long history of use as an efficient feed in the aquaculture industry (Camus et al., 2009; Galimany et al., 2014). It has a high nutritional value (Tibbetts et al., 2015) and is of considerable interest for biotechnological production of useful compounds, e.g. essential fatty acids, antioxidants (carotenoids and phenolic compounds), starch and bulk lipids and oils (Araujo et al., 2011; Custódio et al., 2012; Gifuni et al., 2018). Genetic tools for transformation of *T. chuii* have been developed (Úbeda-Mínguez et al., 2015). The species has also been used as a test organism in toxicity assays of different types of pollutants (Debelius et al., 2009; Prata et al., 2018; Davarpanah and Guilhermino, 2019).

The alga *T. chuii* has also been used in human food, based on its content of antioxidants (Widowati et al., 2017). Dried biomass of *T. chuii* has been authorised in the EU as a novel food and food supplement (Commission Implementing Regulation (EU) 2017/2470[10]), and it is sold under the name TetraSOD®.

Safety concerns

No safety concern was identified. Cerezuela et al. (2012a, 2012b, 2012c, 2013) reporting effects of different diets, including supplements of *T. chuii*, on expression of genes related to intestinal and immune functions in the fish sea bream (*Sparus aurata* L.). However, no information is given about any effects of the diets on the growth or health status of the fish. One study investigated the toxicity of freeze-dried biomass of *T. chuii* in a rat model (Mantecón et al., 2019). Different doses of *T. chuii* had no effect on growth rate, and no clinical signs or effects on blood parameters, organ weights or histopathology were observed.

Antimicrobial resistance aspects

Not applicable.

Conclusions on a recommendation for the QPS list

Tetraselmis chuii is recommended for the QPS list with the qualification for production purposes only.

3.3. Monitoring of new safety concerns related to organisms on the QPS list

The summaries of the evaluation of the possible safety concerns for humans, animals or the environment described and published since the previous ELS exercise (i.e. articles published between January and June 2019, as described in Appendices B and C) with reference to the articles selected as potentially relevant for the QPS exercise (Appendix D) for each of the TUs or groups of TUs that are part of the QPS list (Appendix E), are presented below.

[10] Commission Implementing Regulation (EU) 2017/2470 establishing the Union list of novel foods in accordance with Regulation (EU) 2015/2283 of the European Parliament and of the Council on novel foods. OJ L 351, 30.12.2017, p. 72–201.

3.3.1. Gram-positive non-sporulating bacteria

3.3.1.1. *Bifidobacterium* spp.

A search for papers potentially relevant for the QPS consideration of *Bifidobacterium* species and *Carnobacterium divergens*[11] provided 270 references. The analysis of their title left 14 articles; the rest were discarded because they did not deal with safety concerns. Six articles were found relevant for the QPS consideration of *Bifidobacterium* spp. at the level of title and abstract screening. Five of these articles were not in English or not dealing with safety concerns. One article was considered for further evaluation (Kothari et al., 2019) but was not considered because it is describing safety concerns related to different probiotics but not pointing out to new specific concern and referring to a paper describing a case of a person with underlying predisposing factors that had a bacteraemia due to *B. longum*.

Based on the available evidence as described above, the QPS status of *Bifidobacterium* spp. is not changed.

3.3.1.2. *Carnobacterium divergens*

A search for papers potentially relevant for the QPS consideration of QPS *Bifidobacterium* species and *Carnobacterium divergens*[10] provided 270 references. The analysis of their title left 14 articles; the rest were discarded because they did not deal with safety concerns. No article was considered relevant at the level of title and abstract screening for this TU. Consequently, the QPS status of *C. divergens* is not changed.

3.3.1.3. *Corynebacterium glutamicum*

A search for papers potentially relevant for the QPS consideration of *Corynebacterium glutamicum* provided 39 references. No paper reached the level of title and abstract screening; therefore, no new safety concerns were identified.

3.3.1.4. *Lactobacillus* spp.

A search for papers potentially relevant for the QPS consideration of any of the 37 *Lactobacillus* species included in the list, provided 620 references. Analysis of their titles left 23 articles; the rest were discarded because they did not deal with safety concerns. Inspection of their abstracts allowed the selection of 23 papers that could raise safety concerns but 15 were not considered relevant at the end. After analysing the full texts of the eight articles that arrived to the evaluation phase, three were not related to this TU, three were referring to articles published before the period being evaluated and had already been subjected to inspection by the working group (WG). The two papers that arrived to final step (Hubbard et al., 2018; Nayeem et al., 2018) described infections putatively ascribed to lactobacilli, although in none of them the methods used for identification of the aetiologic agents are indicated. In Hubbard et al. (2018), a female that suffered from type 2 diabetes and had diabetic ketoacidosis at admission, presented a necrotic ulcer in the left genital labium that was claimed to be produced by *L. acidophilus*. However, primary pathogens, such as *Candida albicans*, *Staphylococcus haemolyticus* and *Klebsiella oxytoca*, were also isolated upon debridement of the lesion. Nayeem et al. (2018) present the case of a patient that suffered from obstruction of the bile duct provoked by a pancreas carcinoma. After percutaneous drainage, *L. rhamnosus* and a fungus were isolated from blood cultures. In both cases, clear predisposing conditions and polymicrobial infections are reported which, together with the absence of indication on the identification methods, make the lactobacillal aetiology of the infections questionable.

Based on the available evidence as described above, the QPS status of the lactobacilli involved in the reported cases and, by extension, of all others included in the QPS list, is not changed.

3.3.1.5. *Lactococcus lactis*

A search for papers potentially relevant for the QPS consideration of *Lactococcus lactis* provided 165 references. Two papers arrived at the title/abstract level, but the possible safety concern was not confirmed due to identification methodological problems. Analysis of their title/abstract/full text left no articles because they did not deal with safety concerns.

Based on the available evidence as described above, the QPS status of *Lactococcus lactis* is not changed.

[11] These 2 TUs were searched together for practical reasons.

3.3.1.6. *Leuconostoc* spp.

A search for papers potentially relevant for the QPS consideration of QPS *Leuconostoc* species and *Microbacterium imperiale*[11] provided 81 references. The analysis of their title/abstract left one article; that was discarded because does not deal with safety concerns. Consequently, the QPS status of *Leuconostoc* spp. is not changed.

3.3.1.7. *Microbacterium imperiale*

A search for papers potentially relevant for the QPS consideration of QPS *Leuconostoc* species and *Microbacterium imperiale*[11] provided 81 references. The analysis of their title/abstract left no article. Consequently, the QPS status of *M. imperiale* is not changed.

3.3.1.8. *Oenococcus oeni*

A search for papers potentially relevant for the QPS consideration of *Oenococcus oeni* and *Pasteuria nishizawae*[11] provided 39 references. The analysis of their title/abstracts left one article for consideration but does not describe any safety concern. Consequently, the QPS status of *O. oeni* is not changed.

3.3.1.9. *Pasteuria nishizawae*

A search for papers potentially relevant for the QPS consideration of *Oenococcus oeni* and *Pasteuria nishizawae*[11] provided 39 references. The analysis of their title/abstracts left one article for consideration, but no new safety concern was found. Consequently, the QPS status of *P. nishizawae* is not changed.

3.3.1.10. *Pediococcus* spp.

A search for papers potentially relevant for the QPS consideration of *Pediococcus* spp. provided 166 references. The analysis of their title/abstract left two articles for the evaluation phase which were not related to this TU or not dealing with safety concerns. Consequently, the QPS status of *Pediococcus* spp. is not changed.

Pediococcus dextrinicus (Coster and White, 1964), included in Back, 1978 (approved list of species) was reclassified as *Lactobacillus dextrinicus* comb. Nov. (Haakensen et al., 2009) according to multilocus sequence analysis. It is updated in the QPS list.

3.3.1.11. *Propionibacterium*

A search for papers potentially relevant for the QPS consideration of *Propionibacterium* spp. provided 27 references. Following the analysis of their title/abstracts, no articles were selected for the evaluation phase; thus, no new safety concerns were identified. Consequently, the QPS status of *Propionibacterium* spp. is not changed.

3.3.1.12. *Streptococcus thermophilus*

A search for papers potentially relevant for the QPS consideration of *Streptococcus thermophilus* provided 79 references. The analysis of their title left four articles. Two reached the evaluation phase, but both of them were not dealing with safety concerns. Therefore, the QPS status of *S. thermophilus* is not changed.

3.3.2. Gram-positive spore-forming bacteria

3.3.2.1. *Bacillus* spp.

A search for papers potentially relevant for the QPS consideration of QPS *Bacillus* spp. And *Geobacillus stearothermophilus*[11] provided 741 references. The MLT analysis left 183 articles. The analysis of their titles by two experts left 15 articles for the title/abstract phase, and from these nine articles passed to the full text phase for further analysis. Three papers (Harwood et al., 2018; Jezewska-Frackowiak et al., 2019, Drillich and Wagener, 2018) were not dealing with safety concerns. Three papers have serious methodological problems in relation to strain identification and source attribution (Joshi et al., 2019; Shah et al., 2019) and are not food related (Aydin et al., 2018). Two papers have a methodological problem in relation to strain identification and present a bacteraemia case caused by *B. licheniformis* in an old and immunocompromised patient (of Li et al., 2016) and a pleuritic caused by *B. megaterium* in an old patient with underlying diseases (Crisafulli et al., 2019).

The paper of Gu et al. (2019) reports the analysis of the whole genome sequence of a *Bacillus* strain indicating a low degree of homology with some virulence determinants of specific pathogens. Moreover, this strain was shown to have haemolitic activity. This pathogenicity feature can be detected by the cytotoxicity test that is required by the current qualification for all *Bacillus* spp*.*

The ELS did not come up with any information that would change the status of the *Bacillus* species included in the QPS list and confirmed the qualification 'absence of cytotoxicity'.

3.3.2.2. *Geobacillus stearothermophilus*

A search for papers potentially relevant for the QPS consideration of QPS *Bacillus* spp. And *Geobacillus stearothermophilus*[11] provided 741 references. The MLT analysis left 183 articles. The analysis of their titles by two experts left 15 articles, for which of nine articles the full texts were analysed. None was dealing with this species. Consequently, the QPS status *G. stearothermophilus* is not changed.

3.3.3. Gram-negative bacteria

3.3.3.1. *Gluconobacter oxydans*

A search for papers potentially relevant for the QPS consideration of *Gluconobacter oxidans* and *Xanthomonas campestris*[11] provided 164 references. The analysis of their titles left one article; the rest were discarded because they did not deal with safety concerns. No paper reached the final selection phase for this TU. Consequently, the QPS status of *G. oxydans* is not changed.

3.3.3.2. *Xanthomonas campestris*

A search for papers potentially relevant for the QPS consideration of *Gluconobacter oxidans* and *Xanthomonas campestris*[11] provided 164 references. The analysis of their titles left one article; the rest were discarded because they did not deal with safety concerns. No paper reached the evaluation phase for this TU. Consequently, the QPS status of *X. campestris* is not changed.

3.3.4. Yeasts

A search for papers potentially relevant for the QPS consideration of the yeast species included in the QPS list provided 491 references. The MLT analysis left 357 articles. The analysis of their titles left 49 articles. Eighteen of these were immediately excluded because they were not in English or because they were not dealing with safety concerns. thirty-one arrived to the article evaluation step. From these, 10 were not in English or not describing a safety concern. Thus, the ELS identified 21 articles relevant to different yeast species with QPS status. Out of these 21 articles, 13 referred to *Kluyveromyces marxianus* (anamorph = *Candida kefyr*) (Altintop et al., 2019; Arastehfar et al., 2019; Espinel-Ingroff et al., 2019; Fay et al., 2018; Ghajari et al. 2018; Hamzehee et al., 2019; Hosain-Pour et al., 2018; Lim et al., 2019; Maheronnaghsh et al., 2019; Mirhendi et al., 2019; Ruosta et al., 2019; Salse et al., 2019; Srivastava et al., 2018), 7 to *Saccharomyces cerevisiae* including *Saccharomyces boulardii* (Davis et al., 2019; Espinel-Ingroff et al., 2019; Fadhel et al., 2019; Landaburu et al., 2019; Oladugba et al., 2018; Pérez-Cantero et al., 2019; Sharma et al., 2019), 5 to *Debaryomyces hansenii* (anamorph=*Candida famata*) (El-Mashad et al., 2019; Espinel-Ingroff et al., 2019; Fay et al., 2018; Karapetsa et al., 2019; Srivastava et al., 2018) and 1 to *Kluyveromyces lactis* (anamorph=*Candida spherica*) (El-Mashad et al., 2019). For the other yeast species with QPS status, no relevant studies were identified through the ELS.

Some of these articles were considered relevant to evaluate since they presented results about the antimycotic susceptibility of specific yeast TU. Espinel-Ingroff et al. (2019) analysed the MIC of triazole in different strains of *C. kefyr* and *S. cerevisiae*. The *in vitro* activity of compounds amphotericin B, voricona-zole and anidulafungin of *S. cerevisiae* has been described in Pérez-Cantero et al. (2019).

Methodological problems were identified in 8 of those 21 studies (Altintop et al., 2019; El-Mashad et al., 2019; Espinel-Ingroff et al., 2019; Fay et al., 2018; Landaburu et al., 2019; Lim et al., 2019; Maheronnaghsh et al., 2019; Oladugba et al., 2018).

Nine described isolations of the QPS yeasts from opportunistic infections in patients and in the majority of them serious predisposing factors were described. Karapetsa et al. (2019) claim to be the first to report septic shock due to *D. hansenii* in an immunocompetent subject, although the patient was characterised as showing 'immunoparalysis'. The young male had serious injuries after a car accident and was admitted to an intensive care unit. Predisposing conditions included a central venous catheter, recurrent bacterial infections and prolonged use of antibiotics. The patient recovered from the fungal infection after treatment with amphotericin B. Six of the studies described infection produced by *K. marxianus* (*C. kefyr*) with low incidence. Ruosta et al. (2019) reidentified a number of isolates from different hospital yeast collections without information about the health status of the patients; Hamzehee et al. (2019) isolated strains from oral candidiasis infection; Mirhendi et al. (2019) from paediatric patients with invasive candidiasis and Hosain-Pour et al. (2018) from oral samples from HIV/AIDS patients. Ghajari et al. (2018) isolated from women with suspected vulvovaginal candidiasis *K. marxianus* but with low prevalence. Finally, two papers were associated with *S. cerevisiae*. Davis et al. (2019) reports hepatosplenic infection by *S. cerevisiae* in a 4-year boy with lymphoblastic leukaemia and Fadhel et al. (2019) report a case of S. cerevisiae fungaemia in a 74-year-old man with predisposing factors in an intensive care unit, who had been taking a probiotic containing *Saccharomyces cerevisiae* (boulardii) for several years.

In short, the ELS did not identify any information that would change the status for the yeast species included in the QPS list.

Table 3: Articles that arrived to the evaluation phase (final step of the extensive literature search) for the QPS status yeasts group (21 articles with 26 studies)

Relevant to the QPS exercise[(a),(b)] 21 articles (26 studies)	Articles not describing safety concerns	1 article (1 study)	**Any methodological problem identified?**	*Yes*	1 article (1 study)	*Methodology used for identity confirmation of the microorganism*	1 article (1 study)	Oladugba et al. (2018)
						Reliability of the source attribution	None	
						Misuse of the microorganism	None	
						Predisposing factors in the exposed subjects	None	
						Other reasons	None	
				No	None			
	Articles dealing with safety concerns	20 articles (25 studies)	**Any methodological problem identified?**	*Yes*	7 articles (11 studies)	*Methodology used for identity confirmation of the microorganism*	7 articles (11 studies)	Lim et al. (2019) Maheronnaghsh et al. (2019) Fay et al. (2018) Altintop et al. (2019) Salse et al. (2019) El-Mashad et al. (2019) Espinel-Ingroff et al. (2019)
						Reliability of the source attribution	None	
						Misuse of the microorganism	None	
						Predisposing factors in the exposed subjects	1 article (2 study)	El-Mashad et al. (2019)
						Other reasons	None	
				No	13 articles (14 studies)	Pérez-Cantero et al., 2019 Srivastava et al. (2018) Landaburu et al. (2019) Ruosta et al. (2019) Hamzehee et al. (2019) Sharma et al. (2019) Arastehfar et al. (2019) Ghajari et al. (2018) Hosain-Pour et al. (2018) Mirhendi et al. (2019) Fadhel et al. (2019) Karapetsa et al. (2019) Davies et al. (2019)		

(a): Please refer to Appendix D for the complete list of references.
(b): Number of references (articles and studies) indicated for each step.

3.3.5. Viruses used for plant protection

3.3.5.1. *Alphaflexiviridae*

A search for papers potentially relevant for the QPS consideration of *Alphaflexiviridae* and *Potyviridae*[11] provided 41 references. No paper dealing with *Alphaflexiviridae* reached the final selection phase; thus, no new safety concern was found.

3.3.5.2. *Potyviridae*

Two papers (Gachara and Wisser, 2018; Cong et al., 2019), dealing with *Potyviridae* reached the final selection phase, but no new safety concern was described.

3.3.5.3. *Baculoviridae*

A search for papers potentially relevant for the QPS consideration of *Baculoviridae* provided 58 references. Three articles reached the final selection phase.

Zhao et al. (2019) is an in-depth study on the fate and consequences of baculovirus infection by intravenous infection of rats. This is an unusual interaction of baculoviruses and vertebrate hosts; normally vertebrates only ingest baculoviruses by food intake. Nevertheless, no pathology and no adverse effects on animal health were observed. Charon et al. (2019) describe largely the regulatory framework for agrochemicals (including baculoviruses) and promote the inclusion of baculoviruses as low risk substance. Nan et al. (2019) note the prion-like properties of one gene product (Late Essential Factor 10) related to the insect pathology of baculoviruses. This has no consequences for the safety of baculoviruses as biocontrol agent of insect pests for vertebrates.

The ELS did not come up with any information that would change the current QPS status of any of the above virus families.

4. Conclusions

ToR 1: *Keep updated the list of biological agents being notified, in the context of a technical dossier to EFSA Units (such as Feed, Food Ingredients and Packaging (FIP), Nutrition Unit and Pesticides Unit), for intentional use in feed and/or food or as sources of food and feed additives, enzymes and plant protection products for safety assessment*:

Between April 2019 and September 2019, the list was updated with 54 notifications that were received by EFSA, of which 25 were for feed additives, 15 for food enzymes, food additives and flavourings, 9 for novel foods and 3 for plant protection products.

ToR 2: *Review taxonomic units previously recommended for the QPS list and their qualifications when new information has become available*:

In relation to the results of the monitoring of possible new safety concerns related to the QPS list, there were no results that justify removal of any TU from the QPS list or changes in their respective qualifications.

ToR 3: *(Re)assess the suitability of taxonomic units notified to EFSA not present in the current QPS list for their inclusion in that list*:

The TUs corresponding to 23 out of the 54 notifications received, already had a QPS status.

Of the 31 notifications without a QPS status, 7 notifications related to filamentous fungi which were excluded from QPS activities in the follow-up of a recommendation of the QPS 2013 update (EFSA BIOHAZ Panel, 2013, 2014, 2016), 6 notifications related to *E. coli*, which was excluded from the current mandate by the BIOHAZ Panel (EFSA BIOHAZ Panel, 2018a) *Sphingomonas paucimobilis* which was already evaluated in the previous Panel Statement.

- The remaining 17 notifications, corresponding to 16 TUs were evaluated:
- *Protaminobacter rubrum* already evaluated during the previous QPS mandate
- *Schizochytrium* sp. which it is a genus and not a species and therefore, not adequate for QPS approach.
- *Aurantiochytrium limacinum, Bacillus velezensis, Corynebacterium stationis, Cupriavidus necator, Hamamotoa singularis, Kodamaea ohmeri, Lactobacillus parafarraginis, Paenibacillus illinoisenis, Pantoea ananatis*, *Parageobacillus thermoglucosidasius, Rhodococcus aetherivorans, Rhodococcus ruber*, *Tetraselmis chuii, Zygosaccharomyces rouxii*, evaluated for the first time.

5. Recommendations

- *Protaminobacter rubrum* cannot be assessed for a possible QPS recommendation because it is not a valid species name.
- *Lactobacillus parafarraginis* and *Zygosaccharomyces rouxii* are recommended to be included in the QPS list.
- *Parageobacillus thermoglucosidasius* and *Paenibacillus illinoisensis* can be recommended for QPS list with the qualification 'for production purposes only' and absence of toxigenic potential.
- *Bacillus velezensis* can be recommended for QPS list with the qualification 'absence of toxigenic potential and absence of aminoglycoside production ability'.
- *Aurantiochytrium limacinum, Cupriavidus necator* and *Tetraselmis chuii* can be recommended for the QPS status with the qualification 'for production purposes only'.
- *Pantoea ananatis* is not recommended for the QPS list due to lack of body of knowledge in relation to its pathogenicity potential for plants.
- *Corynebacterium stationis, Hamamotoa singularis*, *Rhodococcus aetherivorans* and *Rhodococcus ruber* cannot be recommended for the QPS list due to lack of body of knowledge.
- *Kodamaea ohmeri* cannot be recommended for the QPS list due to safety concerns.

This new QPS recommendation will be included as an addition to the list of QPS status recommended biological agents (EFSA BIOHAZ Panel, 2016), published both as an update to the Scientific Opinion (EFSA BIOHAZ Panel, 2016) and as supporting information available on the EFSA Knowledge Junction community on Zenodo at: https://doi.org/10.5281/zenodo.1146566

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Glossary

Anamorph name	second valid name of a fungi based on the asexual state reproductive state (morphologically)
Antimicrobial compounds	Antibiotics, bacteriocins and/or small peptides with antimicrobial activity
Basonym name	the earliest validly published name of a taxon
Synonymous name/Homotypic synonym	have the same type (specimen) and the same taxonomic rank
Teleomorph name	primary name of a fungi based on the sexual reproductive state (morphologically)

Abbreviations

AMR	antimicrobial resistance
BIOHAZ	EFSA Panel on Biological Hazards
LS	extensive Literature Search
FEEDAP	EFSA Panel on Additives and Products or Substances used in Animal Feed (FEEDAP)
FIP	EFSA Food ingredients and packaging Unit
FSTA	Food Science Technology Abstracts

GMM	genetically modified microorganism
IJSEM	International Journal of Systematic and Evolutionary Microbiology
LPSN	List of Prokaryotic Names with Standing in Nomenclature
MLT	machine learning technique
QPS	qualified presumption of safety
PPP	plant protection product
ToR	Terms of Reference
TU	taxonomic unit
WG	Working Group

Appendix A – Search strategy followed for the (re)assessment of the suitability of TUs notified to EFSA not present in the current QPS list for their inclusion in the updated list (reply to ToR 3)

Protaminobacter rubrum

Not done as it is not a valid species name.

Bacillus velezensis

A literature search was performed in the last 5 years in PubMed database, using the search terms described below, retrieved the following number of hits:

- "Bacillus velezensis": 139 hits (several biotech and industrial applications);
- "Bacillus velezensis" AND toxin*: 4 hits (3 papers on B. velezensis used to reduced mycotoxins; no report of intoxication);
- "Bacillus velezensis" AND surfactin: 20 hits (B. velezensis produces surfactins (cyclic non-ribosomally synthetized peptides));
- "Bacillus velezensis" AND probiot*: 13 hits (reports on fish and one on chickens);
- "Bacillus velezensis" AND (diseas* OR infect* OR intox*): 44 (no report on diseases in animals or humans).

Corynebacterium stationis

A literature search was performed in PubMed database, using the search terms "Corynebacterium stationis" retrieved 12 hits from which, 2 were relevant (Bernard et al., 2010; and Liu et al.,2016).

A literature search also performed in PubMed database, using the search terms "Brevibacterrium stationis" retrieved 11 hits.

Cupriavidus necator

A literature search was performed in PubMed database, using the search terms below and retrieved the following number of hits:

- "Cupriavidus necator": 1,034 hits;
- "Cupriavidus necator" AND taxonomy: 66 hits from which, 4 were selected;
- "Cupriavidus necator" AND safety: 5 with nothing relevant;
- "Cupriavidus necator" AND infection: 19 hits with nothing relevant;
- "Cupriavidus necator" AND disease: 6 hits with nothing relevant.

Lactobacillus parafarraginis

A literature search was performed in the Web of Science Core collection, using the search term "Lactobacillus parafarraginis" retrieved 22 articles that were screened and 10 were selected and considered as relevant.

Paenibacillus illinoisensis

A literature search was performed in PubMed database, using the search terms "Paenibacillus illinoisensis" retrieved 16 hits.

Pantoea ananatis

A literature search was performed in PubMed database, using the search terms "Pantoea ananatis" retrieved 185 hits.

Parageobacillus thermoglucosidasius

A literature search was performed in PubMed database, using the search terms described below, retrieved the following number of hits:

- "Parageobacillus thermoglucosidasius": 7 hits;
- "Geobacillus thermoglucosidasius": 69 hits;
- "Geobacillus thermoglucosidans": 19 hits;
- "Bacillus thermoglucosidasius": 42 hits.

Rhodococcus aetherivorans

A literature search was performed in PubMed database, using the search terms "Rhodococcus aetherivorans" retrieved 23 hits.

Rhodococcus ruber

A literature search was performed in the Web of Science Core collection, using the search term "Rhodococcus ruber" provided a total of 35 hits.

A literature search was also performed in PubMed database, using the same search term, retrieved 177 papers.

Hamamotoa singularis

A literature search was performed in PubMed database, using the search terms described below, retrieved the following number of hits:

- "Hamamotoa singularis: 1 hit (Wang et al., 2015);
- "Hamamotoa" AND infection: no hits;
- "Sporobolomyces singularis": 15 hits;
- "Sporobolomyces AND infection: 1 hit;
- "Sporobolomyces" AND safety: 2 hits.

Kodamaea ohmeri

A literature search was also performed in PubMed database, using the search term "Kodamaea ohmeri", retrieved 82 papers.

Zygosaccharomyces rouxii

A literature search was also performed in PubMed database, using the search term "Zygosaccharomyces rouxii", retrieved 282 papers.

Appendix B – Protocol for Extensive literature search (ELS), relevance screening and article evaluation for the maintenance and update of list of QPS-recommended biological agents (reply to ToR 2)

The protocol for extensive literature search (ELS) used in the context of the EFSA mandate on the list of QPS-recommended biological agents intentionally added to the food or feed (EFSA-Q-2016-00684) is available on the EFSA Knowledge Junction community on Zenodo, at: https://doi.org/10.5281/zenodo.3607190

Appendix C – Search strategies for the maintenance and update of list of QPS-recommended biological agents (reply to ToR 2)

The search strategies for each taxonomic unit (TU), i.e. the string for each TU and the search outcome, are available on the EFSA Knowledge Junction community on Zenodo at: https://doi.org/10.5281/zenodo.3607193

Appendix D – References selected from the ELS exercise as relevant for the QPS for searches from January to June 2019 (reply to ToR 2)

Gram-Positive Non-Sporulating Bacteria

Bifidobacterium

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Vandenplas Y and Savino F, 2019. Probiotics and prebiotics in pediatrics: What is new? Nutrients, 11, 431. https://doi.org/10.3390/nu11020431

Carnobacterium divergens

None.

Corynebacterium glutamicum

None.

Lactobacilli

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Shimizu A, Hase R, Suzuki D, Toguchi A, Otsuka Y, Hirata N and Hosokawa N, 2019. Lactococcus lactis cholangitis and bacteremia identified by MALDI-TOF mass spectrometry: a case report and review of the literature on Lactococcus lactis infection. Journal of Infection and Chemotherapy, 25, 141–146.

Leuconostoc

Avand A, Akbari V and Shafizadegan S, 2018. In vitro cytotoxic activity of a lactococcus lactis antimicrobial peptide against breast cancer cells. Iranian Journal of Biotechnology, 16, 213–220.

Microbacterium imperiale

None.

Oenococcus oeni

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Pasteuria nishizawae

None.

Pediococci

Brenciani A, Fioriti S, Morroni G, Cucco L, Morelli A, Pezzotti G, Paniccia M, Antonelli A, Magistrali CF, Rossolini GM and Giovanetti E, 2019. Detection in Italy of a porcine Enterococcus faecium isolate carrying the novel phenicol-oxazolidinone-tetracycline resistance gene poxt. A Journal of Antimicrobial Chemotherapy, 74, 817–818.

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None.

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Gluconobacter oxydans

None.

Xanthomonas campestris

None.

Yeasts

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Viruses used for plant protection

Alphaflexiviridae

None.

Potyviridae

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Appendix E – The 2019 updated list of QPS Status recommended biological agents in support of EFSA risk assessments

The list of QPS status recommended biological agents (EFSA BIOHAZ Panel, 2020) is being maintained in accordance with the mandate of the BIOHAZ Panel (2017–2019), extended for the following years. Possible additions to this list are included around every 6 months, with the latest Panel Statement adopted in December 2019. These additions are published as updates to the Scientific Opinion (EFSA BIOHAZ Panel, 2020); the updated QPS list is available at https://doi.org//10.2903/j.efsa.2020.5966 and, as of January 2018, also as supporting information linked to every Panel Statement available on the Knowledge Junction at https://doi.org/10.5281/zenodo.1146566.

Appendix F – Microbial species as notified to EFSA, received between April 2019 and September 2019 (reply to ToR 1)

EFSA risk assessment area	Microorganism species/strain	Intended use	EFSA Question number[(a)] and EFSA webpage link[(b)]	Additional information provided by the EFSA Scientific Unit	Previous QPS status of the respective TU?[(c)]	To be evaluated? yes or no[(d)]
Bacteria						
Feed additives	*Bacillus amyloliquefaciens* BS918, *Bacillus amyloliquefaciens* BS1013, *Bacillus subtilis* BS3BP5	Zootechnical additives/ Digestibility enhancers	EFSA-Q-2019-00480	Syncra® SWI 201 TPT (*Bacillus amyloliquefaciens* BS918, *Bacillus amyloliquefaciens* BS1013, *Bacillus subtilis* BS3BP5 and Protease)	Yes	No
Feed additives	*Bacillus amyloliquefaciens* PTA-6507, *Bacillus amyloliquefaciens* NRRL B-50013 and *Bacillus amyloliquefaciens* NRRL B-50104	Zootechnical additives/ Gut flora stabilisers	EFSA-Q-2019-00457	Enviva® PRO 202 GT (*Bacillus amyloliquefaciens* PTA-6507, *Bacillus amyloliquefaciens* NRRL B-50013 and *Bacillus amyloliquefaciens* NRRL B-50104)	Yes	No
Feed additives	*Bacillus coagulans* DSM 32016	Zootechnical additive Gut flora stabiliser	EFSA-Q-2019-00313	TechnoSpore® (Bacillus coagulans DSM 32016)	Yes	No
Feed additives	*Bacillus licheniformis* DSM 28710 (BL11)	Zootechnical additives/ Gut flora stabilisers	EFSA-Q-2019-00525	Already authorised for other target species	Yes	No
Feed additives	*Bacillus subtilis* C-3102, DSM 15544	Zootechnical additive Gut flora stabiliser	EFSA-Q-2019-00370/ FAD-2019-0037	Calsporin® (*Bacillus subtilis* C-3102, DSM 15544) Application for renewal	Yes	No
Feed additives	*Bacillus subtilis* PB6 (*Bacillus subtilis* ATCC PTA-6737)	Zootechnical additives/ Gut flora stabilisers *Bacillus subtilis* PB6 (*Bacillus subtilis* ATCC PTA-6737)	EFSA-Q-2019-00410 FAD-2019-0017	Trade name: *Bacillus subtilis* PB6	Yes	No
Plant protection products	*Bacillus subtilis* RTI477		EFSA-Q-2019-00341		Yes	No

EFSA risk assessment area	Microorganism species/strain	Intended use	EFSA Question number[(a)] and EFSA webpage link[(b)]	Additional information provided by the EFSA Scientific Unit	Previous QPS status of the respective TU?[(c)]	To be evaluated? yes or no[(d)]
Plant protection products	*Bacillus velezensis* RTI301	Fungicide *Bacillus velezensis* RTI301 and *Bacillus subtilis* RTI477 are the microbial active ingredients in the formulated product F4034-5, efficacious against seed- and soil-borne pathogens such as *Rhizoctonia solani, Phytium* spp., *Phytophtora capsici, Sclerotinia sclerotiorum, Fusarium* spp., *Phoma lingam* and *Plasmodiophora brassiceae*	EFSA-Q-2019-00363		No	Yes
Feed additives	*Corynebacterium glutamicum* DSM32932	Nutritional additives Amino acids	EFSA-Q-2019-00331/ FAD-2019-0028	L-lysine monohydrochloride produced by fermentation using genetically modified *Corynebacterium glutamicum* DSM32932	Yes	No
Feed additives	*Corynebacterium glutamicum* KCCM 80189	Nutritional additive L-Isoleucine produced by fermentation with *Corynebacterium glutamicum* KCCM 80189es Amino acids	EFSA-Q-2019-00293	L-Isoleucine produced by fermentation with *Corynebacterium glutamicum* KCCM 80189	Yes	No
	Corynebacterium glutamicum KCCM80183	Nutritional additives/ Amino acids	EFSA-Q-2019- 00411 FAD-2019-0016	L-Lysine monohydrochloride/ Concentrated liquid L-Lysine/L-Lysine sulfate produced by fermentation with Corynebacterium glutamicum KCCM80183	Yes	No

EFSA risk assessment area	Microorganism species/strain	Intended use	EFSA Question number[(a)] and EFSA webpage link[(b)]	Additional information provided by the EFSA Scientific Unit	Previous QPS status of the respective TU?[(c)]	To be evaluated? yes or no[(d)]
Feed additives	*Corynebacterium stationis*	Sensory additives Flavouring compounds IMP (disodium 5′-inosinate) produced by fermentation with *Corynebacterium ammoniagenes* KCCM 80161	EFSA-Q-2019-00040	Disodium 5′-inosinate feed grade is a highly purified product and does not contain any microorganisms. After the fermentation, the cells of the production strain Corynebacterium ammoniagenes KCCM80161 are eliminated by filtration and centrifugation from the fermentation broth	No	Yes
Food enzymes, food additives and flavourings	*Cupriavidus necator*	Production of food contact material	EFSA-Q-2017-00412	The strain expresses the enzyme PHBH synthase derived from *Aeromonas caviae,* used for the synthesis of a copolymer	No	Yes
Food enzymes, food additives and flavourings	*Escherichia coli* (Strain K-12 W3110)	Production of food enzyme D-allulose 3-epimerase	EFSA-Q-2019-00445	Genetically modified strain	No	No
Novel foods	*Escherichia coli* (W3110 – TK0) K12 -derivative	D-psicose-3-epimerase, produced by a GMO derivative from *E. coli* K12, is used in the synthesis of allulose (novel food)	EFSA-Q-2019-00	The novel food is allulose. An application for the FE has been submitted to EFSA (EFSA-2016-00211)	No	No
Feed additives	*Escherichia coli* AG3149	Nutritional additives, Amino acids and Sensory additive, Flavouring compound	EFSA-Q-2019-00361/ FAD-2019-0035	L-isoleucine produced by fermentation of *Escherichia coli* AG3149 Application for renewal and extension of use	No	No
Food enzymes, food additives and flavourings	*Escherichia coli* BL21 (DE3)	Production of food enzyme OBT-001	EFSA-Q-2019-00444	Genetically modified strain	No	No

EFSA risk assessment area	Microorganism species/strain	Intended use	EFSA Question number[(a)] and EFSA webpage link[(b)]	Additional information provided by the EFSA Scientific Unit	Previous QPS status of the respective TU?[(c)]	To be evaluated? yes or no[(d)]
Novel foods	*Escherichia coli* BL21 (DE3)	Production of the novel food Lacto-N-neotetraose (LNnT)	EFSA-Q-2019-00448	Extension of the present specifications for LNnT produced by genetically modified (GM) *E. coli* K-12 to include LNnT produced by GM *E. coli* BL21 (DE3)	No	No
Food enzymes, food additives and flavourings	*Escherichia coli* production strains (LE1B1090)	Production of enzymes (UDP-glucosyltransferases and a sucrose synthase) derived from GM strains of *E. coli* K-12	EFSA-Q-2019-00499	Amendment of specification of food additive steviol glycosides produced by enzymatic conversion of highly purified reb A and/or stevioside from stevia leaf extract	No	No
Food enzymes, food additives and flavourings	Genetically modified strain of *Bacillus licheniformis* (strain NZYM-VR)	Production of food enzyme phospholipase C	EFSA-Q-2019-00442		Yes	No
Feed additives	*Komagataella phaffii* appaT75 (CGMCC 12056)	Zootechnical additives/ Digestibility enhancers	EFSA-Q-2019-00461	APSA PHYTAFEED® 20,000 GR, APSA PHYTAFEED® 20,000 L (6-phytase produced by genetically modified *Komagataella phaffi* appaT75 (CGMCC 12056))	Yes	No
Feed additive	*Lactobacillus parafarraginis DSM 32962*	Technological additives/Silage additive	EFSA-Q-2019-00626/ FAD-2019-0062	Non-GMO lactobacillus/WGS received as part of the dossier	No	Yes
Feed additives	*Lactobacillus plantarum* CECT 8350 and *Lactobacillus reuteri* CECT 8700	Zootechnical additives/ Gut flora stabilisers	EFSA-Q-2019-00487	AQ02® (*Lactobacillus plantarum* CECT 8350 and *Lactobacillus reuteri* CECT 8700)	Yes	No

EFSA risk assessment area	Microorganism species/strain	Intended use	EFSA Question number[(a)] and EFSA webpage link[(b)]	Additional information provided by the EFSA Scientific Unit	Previous QPS status of the respective TU?[(c)]	To be evaluated? yes or no[(d)]
Food enzymes, food additives and flavourings	*Paenibacillus illinoisensis*	Production of food enzyme	EFSA-Q-2016-00523	The production strain is used in the manufacturing of cyclomaltodextrin glucanotransferase. The initial submission of the application identified the strain as Bacillus circulans. B. circulans group 6 has been reclassified as the new species P. illinoisensis	No	Yes
Feed additives	*Pantoea ananatis* - Strain NITE BP-02525 strain	Nutritional additive amino acids, their salts and analogues and sensory additive flavouring compound	EFSA-Q-2019-00332/ FAD-2019-0026	L-cystine produced by fermentation using strain NITE BP-02525 strain which has been derived from *Pantoea ananatis*	No	Yes
Food enzymes, food additives and flavourings	*Parageobacillus thermoglucosidasius*	Production of food enzyme alpha-amylase	EFSA-Q-2016-00145	The food enzyme is an Alpha-amylase It was initially designated by the applicant as *Geobacillus stearothermophilus* which is recommended for QPS. However, new data provided after a request for a 16S rRNA analysis, demonstrated quite unambiguously that the production strain is a strain of *Geobacillus thermoglucosidans*, closely related to *G. stearothermophilus* but not on the QPS list	No	Yes
Feed additives	*Pediococcus pentosaceus* DSM 16244	Technological additives Silage additive	EFSA-Q-2019-00369/ FAD-2019-0039	Application for renewal	Yes	No

EFSA risk assessment area	Microorganism species/strain	Intended use	EFSA Question number[(a)] and EFSA webpage link[(b)]	Additional information provided by the EFSA Scientific Unit	Previous QPS status of the respective TU?[(c)]	To be evaluated? yes or no[(d)]
Novel foods	*Protaminobacter rubrum* strain CBS 574.77 us	For the production of a Novel Food 'Isomaltulose syrup'	EFSA-Q-2018-00609	Request for a scientific opinion as an NF; see also short published summary of this application: https://ec.europa.eu/food/sites/food/files/safety/docs/novel-food_sum_ongoing-app_isomaltulose-syrup.pdf	No	Yes
Food enzymes, food additives and flavourings	*Rhodococcus aetherovorans* strain USA-AN012	Production of Food Additives	EFSA-Q-2011-00612 EFSA-Q-2011-00613 EFSA-Q-2011-00614 EFSA-Q-2011-00615 EFSA-Q-2011-00616 EFSA-Q-2011-00617 EFSA-Q-2011-00637	The production strain is used in the manufacturing of tartaric acid	No	Yes
Food enzymes, food additives and flavourings	*Rhodococcus ruber* strain CM001	Production of Food Additives	EFSA-Q-2011-00612 EFSA-Q-2011-00613 EFSA-Q-2011-00614 EFSA-Q-2011-00615 EFSA-Q-2011-00616 EFSA-Q-2011-00617 EFSA-Q-2011-00637	The production strain is used in the manufacturing of tartaric acid	No	Yes
Food enzymes, food additives and flavourings	*Sphingomonas paucimobilis*	Production of Food Additives	EFSA-Q-2011-00517	The production strain is used in the manufacturing of Gellan Gum	No	No
Filamentous fungi						
Feed additives	*Aspergillus aculeatus* CBS589.94	Zootechnical additives/ Digestibility enhancers	EFSA-Q-2019-00528	RONOZYME® VP (CT) and RONOZYME® VP (L) (endo-1,3(4)-β-glucanase (IUB No 3.2.1.6)) produced by *Aspergillus aculeatus* CBS589.94	No	No
Feed additives	*Aspergillus niger* CGMCC No.5751	Zootechnical additives/ Other zootechnical additives Renewal authorisation	EFSA-Q-2019-00590/ FAD-2019-0054	AviPlus® is a preparation of sorbic acid, citric acid, thymol and vanillin. Citric acid is produced by a non-GMO strain of *A. niger*	No	No

EFSA risk assessment area	Microorganism species/strain	Intended use	EFSA Question number[(a)] and EFSA webpage link[(b)]	Additional information provided by the EFSA Scientific Unit	Previous QPS status of the respective TU?[(c)]	To be evaluated? yes or no[(d)]
Novel foods	*Aspergillus oryzae* strain GL 470	Beta-galactosidase as a novel food	EFSA-Q-2019-00257	The production of beta-galactosidase via fermentation of a stable non-GMO *Aspergillus oryzae* strain	No	No
Feed additives	*Trichoderma reesei* RF11556	Zootechnical additives Digestibility enhancers	EFSA-Q-2019-00330/ FAD-2019-0029	ECONASE® XT (endo-1,4-beta-xylanase) produced by fermentation of genetically modified *Trichoderma reesei* RF11556 Change in the production strain	No	No
Feed additives	*Trichoderma reesei* RF7265	Zootechnical additives Digestibility enhancers	EFSA-Q-2019-00333/ FAD-2019-0027	FINASE® EC (6-phytase) produced by fermentation of genetically modified *Trichoderma reesei* RF7265 Application for renewal	No	No
Food enzymes, food additives and flavourings	*Moniliella pollinis*	Production of food flavourings	EFSA-Q-2011-00730	The production strain is used in the manufacturing of erythritol	No	No
Food enzymes, food additives and flavourings	*Moniliella megachilensis*	Production of food flavourings	EFSA-Q-2011-00730	The production strain is used in the manufacturing of erythritol	No	No
Yeasts						
Novel food	*Euglena gracilis*	Biomass of *Euglena gracilis*	EFSA-Q-2019-00593	The novel food is Paramylon, derived from proprietary strain of *Euglena gracilis*	Yes	No
Food enzymes, food additives and flavourings	*Hamamotoa singularis* (homotypic name as *Sporobolomyces singularis*)	Food enzyme: Beta-galactosidase from Sporobolomyces singularis (YIT 10047)	EFSA-Q-2016-00529		No	Yes
Food enzymes, food additives and flavourings	*Kodamaea ohmeri*	Production of food flavourings	EFSA-Q-2011-00729	The production strain is used in the manufacturing of xylitol	No	Yes

EFSA risk assessment area	Microorganism species/strain	Intended use	EFSA Question number[(a)] and EFSA webpage link[(b)]	Additional information provided by the EFSA Scientific Unit	Previous QPS status of the respective TU?[(c)]	To be evaluated? yes or no[(d)]
Feed additives	*Komagataella pastoris* (CECT 13094)	Zootechnical additives Digestibility enhancers, substances which favourably affect the environment	EFSA-Q-2019-00430 FAD-2019-0041	Preparation of 3-phytase produced by *Komagataella pastoris* (CECT 13094) presented in solid (FSF10000) and liquid (FLF1000) forms. The confirmation of the identity of the GMM was conducted using Microbial Identification by DNA sequencing.	Yes	No
Feed additives	*Komagataella pastoris* appaT75 (CGMCC 12056)	Zootechnical additive Digestibility enhancer 6- phytase produced by a genetically modified yeast	EFSA-Q-2019-00312/ FAD-2019-0021	APSA PHYTAFEED® 20,000 GR, APSA PHYTAFEED® 20,000 L (6-phytase)	Yes	No
Feed additives	*Komagataella phaffi* DSM 32854	Zootechnical additives/ Digestibility enhancers	EFSA-Q-2019-00479	OptiPhos® PLUS (6 phytase produced by genetically modified *Komagataella phaffi* DSM 32854)	Yes	No
Feed additives	*Komagataella phaffii* strain BSY-0007 (DSM 32854) (GMO production organism) *Komagataella phaffii* BG10 (Recipient organism)	OptiPhos Plus is a 6-phytase classified under category 4 'zootechnical additives', functional group [(a)] 'digestibility enhancers', subclassification 'enzymes' as specified in Annex I to Regulation (EC) No 1831/2003	EFSA-Q-2019-00303 FAD-2019-0023	GMO production organism: *Komagataella phaffii* strain BSY-0007 (DSM 32854)	Yes	No
Feed additives	*Komagatella phaffii*/*Pichia pastoris* DSM 32159)	Technological additives/Substances for the reduction of the contamination of feed with mycotoxins	EFSA-Q-2019-00624/ FAD-2019-0061	FUMzyme® (fumonisin esterase) produced by a genetically modified strain of *Komagatella phaffii*	Yes	No

EFSA risk assessment area	Microorganism species/strain	Intended use	EFSA Question number[(a)] and EFSA webpage link[(b)]	Additional information provided by the EFSA Scientific Unit	Previous QPS status of the respective TU?[(c)]	To be evaluated? yes or no[(d)]
Feed additives	*Komagatella phaffii* DSM 32854)	Zootechnical additives/ Digestibility enhancers	EFSA-Q-2019-00526	OptiPhos® PLUS (6 phytase) produced by a genetically modified strain of *Komagatella phaffii*	Yes	No
Feed additives	*Saccharomyces cerevisiae* MUCL 39885	Zootechnical additives Gut flora stabilisers Saccharomyces cerevisiae MUCL 39885	EFSA-Q-2019-00219	*Saccharomyces cerevisiae* MUCL 39885	Yes	No
Food enzymes, food additives and flavourings	*Zygosaccharomyces rouxii*	Production of food flavourings	EFSA-Q-2011-00723	The production strain is used in the manufacturing of mannitol	No	Yes
Viruses						
Plant protection products	*Phthorimaea operculella granulovirus* (PhopGV)	Insecticide with activity against larvae of the tomato leafminer *Tuta absoluta* and the potato tuber moth *Phthorimae operculella*	EFSA-Q-2019-00382	Belongs to the group of Betabaculovirus	Yes	No
Algae						
Novel food	*Schizochytrium limacinum* strain WZU477 (synonym of *Aurantiochytrium limacinum*)	Modification to include *Schizochytrium limacinum* strain WZU477 for the production of Docosahexaenoic acid-rich oil	EFSA-Q-2019-00306	Novel food already authorised	No	Yes
Novel food	*Schizochytrium* sp	Oil rich in Docosahexaenoic acid from *Schizochytrium* sp	EFSA-Q-2019-00323	Novel food already authorised	No	Yes

EFSA risk assessment area	Microorganism species/strain	Intended use	EFSA Question number[(a)] and EFSA webpage link[(b)]	Additional information provided by the EFSA Scientific Unit	Previous QPS status of the respective TU?[(c)]	To be evaluated? yes or no[(d)]
Novel food	*Schizochytrium* sp.	Production of DHA-rich oil (BioDHA) from *Schizochytrium* sp. as a novel food	EFSA-Q-2019-00548	Extension of use in infant and follow-on formulae	No	Yes
Novel food	*Tetraselmis chuii*	Dried *Tetraselmis chuii* microalgae as a novel food	EFSA-Q-2019-00535	Modification of the specifications of the dried *Tetraselmis chuii* microalgae, already authorised	No	Yes

(a): To find more details on specific applications please access the EFSA website - Register of Questions: http://registerofquestions.efsa.europa.eu/roqFrontend/ListOfQuestionsNoLogin?0&panel=ALL
(b): Where no link is given this means that the risk assessment has not yet been published.
(c): Included in the QPS list as adopted in December 2016 (EFSA BIOHAZ Panel, 2017a) and respective updates which include new additions (latest: EFSA BIOHAZ Panel, 2019b).
(d): In the current Panel Statement.