

Minutes of the online meeting of the International Committee on Systematics of Prokaryotes [DRAFT of February 8, 2020]

5th January 2020: Iain Sutcliffe, Chair of the ICSP, Northumbria University, Newcastle upon Tyne, U.K.

To the members of the International Committee on Systematics of Prokaryotes

In keeping with Article 4 of the ICSP Statutes, the Editorial Board of the International Code of Nomenclature of Prokaryotes (ICNP) is conducting an open electronic meeting concerning proposals for changes in the ICNP.

The first phase of the meeting will take place from January 5, 2020, until March 1, 2020. It is intended to allow open discussion of the proposals as an email chain among the members of the ICSP and other interested parties. Comments may be made by the 'reply-all' option on your email server. **Comments should be less than 500 words in length** and should identify the author's name(s) and affiliation(s). Comments should be respectful, and ad hominem comments will be deleted from the record. As comments accumulate, the Editorial Board will transfer them to the ICSP website, and the edited comments will serve as the minutes of the meeting. **Please feel free to add interested parties to the email recipient list and solicit comments from interested parties outside the ICSP.**

The second phase of the meeting will consist of voting and will take place from March 1 to March 31, 2020. Only members of the ICSP may vote.

The issues for the current discussion are the "Modest proposals to expand the type material for naming of prokaryotes" made by Whitman (2016; IJSEM 66: 2018-2112; <https://doi.org/10.1099/ijsem.0.000980>) and a related proposal by Whitman et al. (2019; IJSEM 69: 2174-2175; <https://doi.org/10.1099/ijsem.0.003419>) concerning granting priority to *Candidatus* names. To simplify the discussion, the ICSP and contributing colleagues are asked to give particular consideration to the following statements, which represent the central concepts. Should they be passed at the voting stage, other rules will be changed as described in Whitman (2016) and Whitman et al. (2019) to make the remainder of the Code consistent with these changes.

Proposal 1 (Whitman 2016). Extend the nature of the type acceptable for valid publication of a species or subspecies name to allow the use of complete or partial genome sequences as type (Whitman 2016). The new rules would be worded [new text is underlined]:

Rule 18a. The type of a species or subspecies must unambiguously identify the taxonomic group and is a designated strain or other material. Whenever possible, the type of a species or subspecies is a designated strain.

(3) *[first section]* As from 1 April 2020*, sequences of genomic DNA may also serve as the type when it unambiguously identifies the species. When possible, it should be a high quality draft or better genome sequence.

Rule 30.3.c. *[new rule]* When a sequence is the type, the accession number in a publically available database or the sequence must be given. It is recommended that, when possible, a sample of the DNA be deposited in at least two publically accessible service collections in different countries and the catalog numbers be indicated.

*The original date of January 2016 proposed in Whitman (2016) is changed to reflect the time necessary to bring this matter to a vote. All of the other proposals in Whitman (2016) will be taken as originally worded.

Proposal 2 (Whitman 2016). Articulates a general concept for what can serve as type for a species.

Rule 18a (3). *[second section]* As new methods are developed, they may serve as the type material so long as they unambiguously identify the species or subspecies and can be readily archived and compared.

Proposal 3 (Whitman 2016). Allows valid publication of the name of a genus in the absence of a type species if the type is too ambiguous to circumscribe a species.

The rule would be:

"Rule 20a. The nomenclatural type (see Rule 15) of a genus or subgenus is the type species or the sequence of one or more genes that unambiguously identifies the genus or subgenus. The type species is the single species or one of the species included when the name was originally validly published. Only species whose names are legitimate may serve as types."

Proposal 4 (Whitman et al. 2019). Upon acceptance of Proposal 1, the priority of the names of *Candidatus* taxa published before 1 April 2020* which are otherwise in accordance with the rules of the Code will have priority based upon their date of publication in the IJSEM unless a synonymous name already exists based upon deposition of type cultures.

Whitman et al. (2019) also provides a simple nomenclature for identifying the nature of the type material:

'When the type is a culture, the superscript "T" will be used immediately following the name or strain identifier. If the type is a sequence, the superscript "Ts" will be used. If the type is a description, preserved specimen or illustration, the superscript "Td" will be used. If a representative of a taxon is brought into culture, the type strain is then designated as described in Rule 18f. The name may be emended by the new authors, and the superscript "Ts" or "Td" is replaced by the superscript "T".'

*The original date of 1 January 2020 is changed to reflect the time necessary to bring this matter to a vote.

For further guidance, major publications that discuss these proposals include:

(in favour)

Whitman 2015. Syst. Appl. Microbiol. 38: 217-222 (<https://doi.org/10.1016/j.syapm.2015.02.003>)

Konstantinidis et al. 2017. ISME J 11: 2399-2406
(<https://www.nature.com/articles/ismej2017113>)

Rossello-Mora and Whitman 2019. Syst. Appl. Microbiol. 42: 5-14
(<https://doi.org/10.1016/j.syapm.2018.07.002>)

(against)

Overmann et al. 2019. Syst. Appl. Microbiol. 42: 22-29.
(<https://doi.org/10.1016/j.syapm.2018.08.009>)

Bisgaard et al. 2019. Diagn. Microbiol. Infect. Dis. 95: 102-103.
(<https://doi.org/10.1016/j.diagmicrobio.2019.03.007>)

COMMENTS:

Comments are present in the order they were received and may have been lightly edited. Please email Barny Whitman [whitman@uga.edu] or Lenie Dijkshoorn [L.Dijkshoorn@lumc.nl] for questions, suggestions, errors and omissions.

January 13

Henrik Christensen, Member of Judicial Commission, University of Copenhagen, Copenhagen, Denmark

We have recently published a note that presents a warning (Bisgaard et al. 2019) about the proposal of using DNA sequences as type material to name new species (Whitman). If implemented the proposal to use DNA sequences as type material may have far-reaching consequences for all microbiologists, ID specialists, vets and other specialists dealing with bacterial names, not to speak about the companies that develop species identification tools and strains for biotech production of probiotics, vaccines and enzymes. The risk is an unstable nomenclature violating Principle 1 of the “code” (“.. 1) Aim at stability of names, 2) Avoid or reject the use of names which may cause error or confusion 3) Avoid the useless creation of names .. “). I have become involved in this problem as an active scientist working with bacterial taxonomy at the university. I will contact other taxonomic colleagues as well to revive the discussion.

You are of course welcome to contact me for further explanations and discussions of the problem.

January 13

Lenie Dijkshoorn, Executive Secretary ICSP, Leiden University Medical Center, Leiden, The Netherlands

I fully support the letter from Henrik. There is an urgent need for contemplation for workers in the field who use names in daily work.

January 13

William B Whitman, ICSP Delegate, University of Georgia, Athens USA

Iain Sutcliffe, Chair of ICSP Northumbria University, Newcastle upon Tyne, U.K.

Ramon Rossello-Mora, Vice-Chair of Judicial Commission of the ICSP, Grup de Microbiologia Marina IMEDEA, Illes Balears, Spain

Genome sequencing has revolutionized prokaryotic systematics by greatly improving the identification of species, elucidating the functional properties of taxonomic groups, and resolving many of the ambiguities in the phylogeny of the higher taxa. Following from the principles described in the International Code of Nomenclature of Prokaryotes, gene sequences are also suitable type material for the description of prokaryotic species. As put forth in principle 4 of the Code, the primary purpose of naming is to supply a means of referring to specific prokaryotes. The Code possesses two mechanisms to insure uniqueness and stability of names. First, it gives priority to the earliest name of the entity. Second, each name is associated irrevocably with some type material. The only name that can be used that includes this type material is the name with priority. The relationship of the name to the type material is further determined by the formal description (also called the protologue), which defines how a taxon is delineated in reference to the type material. Gene sequences clearly possess sufficient specificity and information to serve as type material and delineate taxa. In fact, it has been the common practice to differentiate species based upon sequence similarity since the mid-sixties and formally recommended by Wayne et al. (1987).

A stable nomenclature is essential for all scientific disciplines. While this need was met with the adoptions of the Approved Lists in 1980 and the Code of 1990, subsequent changes in 2001 restricted the Code to organisms that can be deposited as pure strains in culture collections. These changes removed the protection of the Code from the names of prokaryotes that cannot be easily cultured. Proposal 1 would restore the original intent of the Code. By allowing gene sequences to serve as type material for prokaryotic species, this simple change will create stability in naming of *Candidatus* taxa, endosymbionts, and many uncultivated prokaryotes. It is already well established that the use of sequence data, increasingly in the form of whole genome sequences, produces reliable and stable classifications. Thus, proposal 1 will meet an important need within microbiology and allow the creation of a unified nomenclature for all prokaryotes, in contrast to the current "International Code of Nomenclature of *Cultivated* Prokaryotes". Proposal 2 states the rationale for Proposal 1. Proposal 4 implements proposal 1 for *Candidatus* taxa and provides a simple system for identifying the nature of the type material.

Proposal 3 recognizes that on some occasions, the sequence data may be of sufficient quality to delineate a genus but not a species. An example might be 16S rRNA sequences, but it is

inevitable that larger amounts of genome sequence data will also be used. In these cases, a genus name may be validly published without designated type species. Because the genus name provides the root for higher taxa, genus names are required creation of stable higher taxonomies.

Wayne et al. 1987. Int J Syst Bacteriol 37:463-464.

January 14

Kostas Konstantinidis, Member of Judicial Commission, Georgia Institute of Technology, Atlanta Georgia, U.S.A.

In response to Christensen and Dijkshoorn:

I do NOT share the same view with you on this issue but before i offer my arguments for this, i would like to ask Henrik (and/or Lenie):

Why you believe the genome/DNA sequence as Type would make for an unstable system OR will make the identification of taxa of medical importance more challenging (since almost all these taxa are known by cultures and, hence, there will be no change to them really if genome sequences are accepted as Type)? Could you offer a couple specific examples to back up these claims?

I would argue that the Bisgaard et al. 2019 paper was vague about these key points, so the underlying rationale is not clear to me yet.

January 15

Edward Moore, ICSP delegate, University of Gothenburg, Gothenburg, Sweden

The proposals formulated by Iain (Jan 05) to modify the International Code of Nomenclature of Prokaryotes (The Code) have been percolating for a number of years now. They have been presented in publications and discussed as 'considerations' or 'modest proposals', etc. However, prior to January 5, no formal proposals were presented to ICSP for consideration to vote for adopting. Now, the ICSP Executive Board has formulated formal proposals for consideration – below. This is an essential step forward, which is good to try to resolve the issues, as well as the concerns of the proponents and opponents of the proposals.

My comment here is to the proposed or designated schedule (it is not clear to me) for the 'second phase' of the open electronic meeting, i.e., for voting (March 01-31) on the proposals. I point out that the members of the ICSP are representatives of the various Microbiological Societies of the different countries. As such, our decisions and votes on issues should reflect the considerations or, the consensus – in the best cases, of our respective national Societies. Particularly, this issue of revising The Code warrants informing member microbiologists of the national Societies, and their consideration, as well, rather than only of the individual ICSP members.

In any case, now we have formal proposals to be considered for voting. Unfortunately, the proposed/designated schedule for the 'first phase of the meeting, i.e., open discussion, etc.,' does not allow for consideration by the overall members of the Societies. I know of presentations of some of these issues, as concepts, only in sessions at the last 2 FEMS

meetings. I do not know if the issues have been presented and discussed within the different Society meetings – they have not been discussed before within the Swedish Microbiological Society (SFM).

Now, the problem is that the dates of Societies' meetings, where these proposals could be brought up and debated, are after the proposed 'deadline' for voting on these proposals, in most cases. The annual meeting of the VAAM in Germany is in the second week of March; they do have a Fachgruppe für Systematik und Identifizierung – I do not know if they are considering the proposals in their session. However, the SFM in Sweden is meeting in May; the MS in the UK is meeting in April; the ASM in the USA is meeting in June; the Spanish Society is meeting in July 2021, although a Systematics and Taxonomy meeting is scheduled for April, 2020! These meetings, and the annual meetings of other Societies, as well, are after the 'deadline' for ICSP voting on the proposals.

I propose that the dates of the 'first phase' of the open electronic meeting for open discussion be prolonged, to allow communication of the formal proposals for revising The Code to be circulated to the members of the national Societies. In any case, I commend the ICSP Executive Board for presenting the formal proposals and initiating the open electronic meeting.

January 15

Iain Sutcliffe, Chair of the ICSP, Northumbria University, Newcastle upon Tyne, U.K.

I should like to reply promptly to these comments since they relate to the timeline & decision making process rather than the scientific issues under discussion.

Firstly, my apologies for any ambiguity in my email of Jan 5th: This is now a 'designated' schedule i.e. voting by ICSP members will begin on March 1st and close on 31st.

Secondly, regarding the timeline and current 'window' for discussion. It is important to stress that the Whitman (2016) proposals were published online on 1st May 2016 i.e. 3 years 8 months ago, which I would have thought is more than sufficient time for interested parties to have encountered these proposals (it is also unambiguous in the original text that these are formal proposals to amend the Code that require an ICSP vote).

Notably, the paper has attracted 26 citations according the IJSEM website (34 by googlescholar), including at least one dedicated commentary outside of the specialist systematics literature (e.g. Bisgaard et al. 2019 in a clinical journal). Moreover, as you note, these issues were highlighted in the last two FEMS meetings and they have also been addressed in specialist meetings (e.g. BISMIS, Bergeys International Society for Microbial Systematics). Thus it is demonstrable that the proposals have had 'reach'.

My personal view is that this is a more than sufficient time for these proposals to have come to the attention and gather responses of the scientific community. Moreover, ICSP members and other interested parties have had the past 44 months to engage in discussions with colleagues and 'gauge the mood'. There are still 12 weeks for further activities of this sort and I am pleased that you have widened the debate by adding recipients to this email trail.

Thus, I believe that the majority vote decision of the ICSP Executive Board to now bring this matter to the vote is the correct one.

January 16

Mei-Chin Lai, ICSP delegate, National Chung Hsing University, Taichung, Taiwan

I agree with "Proposal 1" that genome sequences should be included and suggest that the "completeness" of genome sequences need to be over or around 97%.

January 16

Henrik Christensen, Member of Judicial Commission, University of Copenhagen, Copenhagen, Denmark

In response to Konstantinidis's comments of January 14

Unfortunately there was a space limitation with the paper of Bisgaard et al., and we also wanted to keep the text short. I agree that it would have been relevant to give some examples.

Question:

Why you believe that the genome/DNA sequence as Type would make for an unstable system OR will make the identification of taxa of medical importance more challenging (since almost all these taxa are known by cultures and hence, there will be no change to them really if genome sequence is accepted as Type)? Could you offer a couple specific examples to back up these claims?

I would argue that the Bisgaard et al. 2019 paper was vague about these key points, so the underlying rationale is not clear to me yet.

Answer:

On behalf of my co-authors I will try to give a more extended answer here.

1. Risk for an increase in the number of heterotypic synonyms. A new species B is proposed (validly published) and only one DNA sequence serves as type material and only one sequence is known from the species. B is closely related to an existing well known species A of high clinical importance. This can happen since species of type A have a high diversity at the population level and, in such cases, ANI can be lower than 0.95 for some populations of the species. If only comparisons between A and B are based on type strains (or type DNA material), less than 0.95 ANI can be obtained, and a claim made for a new species. If a medical clinical microbiologist identifies an isolate by whole genomic sequencing as species B, this species is not known to be of clinical importance to him, and he might get confused about the disease associated, how the infection can be treated with antibiotics, and how it can be prevented. The consequence can be a wrong treatment of the patient. The problem already exists with cultured type strains, and it is expected to increase if the proposal of using DNA sequences as type material is adopted.

2. Identification of clinically important streptococci. An example provided by a co-author of the Bisgaard paper is related to the problems in the clinic to differentiate *Streptococcus pneumoniae* (important pathogen) from *S. pseudopneumoniae* and *S. mitis* (commensals). These species are closely genetically related, and their virulence can only be established based on cultivation. This co-author even extends the case to ANY bacteria for which vaccines are being developed. Strain material is essential to test for the specificity of vaccines, for strains of existing species as well as for strains of new species in the future.

A more general statement was made by another co-author: It is only possible clinically to link a name of a culture-positive organism to additional data available through publications, to subjects such as diagnosis, prognosis, and treatment. Allowing for species to be name based upon DNA alone will not be helpful from a clinical standpoint.

An even more general statement made by the same co-author reaches beyond the clinical field relates to the scientific demand for reproducibility of experiments. The deposit of genomic DNA or, worse, simple submission of wgs data to a public database does not allow reproduction and confirmation of the conclusions of the authors about the taxonomic status of new isolates and strains simply because there will be no proof that the wgs data are coming from the proposed species.

January 17

Edward Moore, ICSP delegate, University of Gothenburg, Gothenburg, Sweden

In response to Lai's comments of January 16

Thank you for your mail and your comment.

Please note: The proposals would establish new rules that would substitute whole genome sequence data for the strain as the type material required for the valid publication of names of new bacterial species. IJSEM already requires including whole genome sequence data for the valid publication of new species names, since 2018.

The proposed rule changes do not make a proposal for the coverage or the quality of the whole genome sequence data that would serve as the type material.

Question: Why do you propose 97% 'completeness' or coverage of genomes; why not 20%, i.e., the amount necessary for ANI analyses, or complete genomes, including plasmids?

Thank you in advance for your consideration.

If proponents of the new rules proposals disagree with my assessment, please correct my response to Mei-Chin.

January 17

Frans Reubsaet, Diagnostic Laboratory for Bacteriology and Parasitology (BPD), Center for Infectious Disease Research, Diagnostics and laboratory Surveillance, National Institute of Public Health and the Environment (RIVM), The Netherlands

In response to Moore's comments of January 17

At this moment most genomes are analyzed by Illumina platforms. We experienced that pollution with other DNA is no hypothetical. Second, even the de novo created sequences are artificial. So if the decision is made in favour of whole genome sequences, sooner or later it will become clear that poor data will not prevail.

January 17

William B Whitman, ICSP Delegate, University of Georgia, Athens USA

Regarding the discussion between Ed and Mei-Chin, I'd like to clarify a few points regarding proposal 1, which would allow gene sequences to serve as type. If passed, strains would still remain the preferred type [see highlighted text below]. Thus, sequence data would only substitute for strains when strains are unavailable.

Rule 18a. The type material of a species or subspecies must unambiguously identify the taxonomic group and is a designated strain or other material. **Whenever possible, the type of a species or subspecies is a designated strain.**

This proposal also does not require a whole genome sequence but only enough sequence to unambiguously identify the species. This wording was chosen to allow naming of endosymbionts where the whole genome sequence is not available. There are many examples of this in IJSEM, but a recent one describes a *Borrelia* species, Loh et al. 2017 [doi.10.1099/ijsem.0.001929] where the diagnosis was made on the basis of the sequences of five genes: 16S rRNA, *flaB*, *groEL*, *gyrB* and *glpQ*.

(3) As from April 2020, sequences of genomic DNA may also serve as the type material when it unambiguously identifies the species. **When possible, it should be a high quality draft or better genome sequence.**

The second sentence [highlighted] constitutes a recommendation stating a preference for genome sequences. There is substantial precedence for the Code to make recommendations as well as rules. For instance, the 1990 Code recommended deposition of strains as type material. The current Code recommends the descriptions should conform to the minimum standards for the group (Recommendation 30). Because minimum standards for whole genome sequences were proposed in IJSEM in 2018 [Chun et al. 2018. Proposed minimal standards for the use of genome data for the taxonomy of prokaryotes. Int J Syst Evol Microbiol 68: 461-466. doi.org/10.1099/ijsem.0.002516], clear directions regarding implementation of this recommendation already exist.

January 18

Kostas Konstantinidis, Member of Judicial Commission, Georgia Institute of Technology, Atlanta Georgia, U.S.A.

In response to Christensen's comments of January 16

In my view, there are two distinct issues, one is the concept of using the DNA as an alternative type material and the other is the technical aspect on what the minimum standards will be for this. My read of Whitman's comment of January 17 is: no change in how we do business for cultured organisms (that is, depositing an isolate to 2 culture collections is the recommended way to name/describe new taxa) and genome sequence becomes an equally appropriate alternative type material for uncultured taxa and fastidious organisms that are difficult to be maintained in culture collections or get lost (if I have misread this, please somebody to correct me!). This way, we will be able to start describing the "uncultivated majority" using similar standards to those used for cultures, and I would argue that this would even promote the culturing of the important uncultivated taxa because of more interest/attention to them once they are described taxonomically. Karthikeyan et al. 2019 is a great recent example of this from my

group (we are now in the process of depositing the isolate to culture collections); i am aware of several similar examples if you want to see more. I short, i see this as a win-win situation for all of us and no threat whatsoever for the culture collections. To the contrary, I think allowing genome sequence to serve as type material may further promote culturing efforts! So, I am in favor of Whitman's proposal personally as I understand the proposal.

As far as the technical standards, be sure that all (or most, at least) of us that would like to have genome sequence as type material, do NOT want to do this with lower standards. We want to have as high standards as the isolate genomes, if not higher. I do believe it is doable. I explain a bit more below for those that want to read more on the technical issues and then address your specific concerns further below. I will also try to publish officially the points below in peer-reviewed press so you can refer to them and offer your arguments in favor or against toward helping to establish, hopefully, the standards that we can all adopt and use in practice soon! But the essence of what i am writing below can also be found in the paper cited by Sutcliffe above, Konstantinidis et al. 2017 (<https://www.nature.com/articles/ismej2017113>).

On the technical standard issue:

Several scientists have argued that the MAG and SAG information is not of similar quality to the information derived based on isolate-based experiments in the lab and thus, does not represent well the organisms under investigation (Bisgaard 2019, Overmann 2019). While this is, at least partly, true, it is not critical enough to prevent progress towards cataloguing the taxonomic diversity of uncultivated organisms, for several reasons. First, prokaryotic taxonomy has always relied on imperfect methods; MAGs/SAGs are not an exception to this. Take, for instance, the DNA-DNA hybridization (DDH) method, the "golden standard" for species demarcation. The genome-aggregated average nucleotide (ANI) value of shared genes among two related genomes (Konstantinidis and Tiedje 2005) has been shown to correlate well with their DDH values, and deviations in the values were common and largely attributable to the experimental noise of the former as opposed to the latter method (Goris 2007). Second, there are approaches to assess quality beyond reasonable doubt such as visual examination of read-recruitment plots (Rodriguez-R 2016) in combination with the quality checking pipelines (Parks 2015, Rodriguez 2018), and in our view only genomes of high enough quality based on these tests should be taxonomically described (Konstantinidis 2017). Third, the standards to use have been outlined already previously by us (Konstantinidis 2017) and others (Bowers 2017), and are of similar stringency to those used for isolate genomes. Further, long-read sequencing for routine taxonomic descriptions, even on environmental samples, is coming up soon [e.g., (Andersen 2019)], and is strongly expected to circumvent several of the low quality issues reported for MAGs and SAGs in the literature, e.g., identify and fix genome sequences that may be chimeric. It has been argued that when DNA sequence type material is replaced by new versions due to new sequencing technologies and/or tools for genome assembly, the species descriptions would have to be consequently revised, resulting in an unstable classification (Bisgaard 2019). However, this is unlikely to be true for most -if not all- taxa because such new versions will mostly affect only a small number of genes or nucleotide substitution positions in the genome as analysis of mock datasets of known composition has revealed (Sczyrba 2017) or the sequencing of the isolated *Candidatus* *Macondimonas diazotrophica* that was almost identical to its corresponding MAG (e.g., ANI >99.9%) (Karthikeyan 2019). It is even less likely that the affected genes by new genome versions would represent the species-diagnostic traits because these genes are often the hypothetical, mobile or prophage-associated genes found in multiple copies (and short contigs) in the genome (Pena-Gonzalez 2019). Hence, the genealogy of the genome and thus, its nomenclature and classification, will remain unaffected in the great

majority of cases where new versions of the genome become available. In a few cases that the new genome version will include major changes in gene content, the old version could be replaced by the new version in a process analogous to replacing the (usually lost) type strain of a (named) species by a neotype strain for isolated organisms.

References cited:

- Andersen et al. 2019. Syst Appl Microbiol **42**: 77-84.
Bisgaard et al. 2019. Diagn Microbiol Infect Dis **95**: 102-103.
Bowers et al. 2017. Nat Biotechnol **35**: 725-731.
Goris et al. 2007. Int. Syst Evol Microbiol **57**: 81-91.
Karthikeyan et al. 2019. ISME J **13**: 2129-2134.
Lawrence and Ochman. 1998. Proc Natl Acad Sci U S A **95**: 9413-9417.
Overmann et al. 2019, Syst Appl Microbiol **42**: 22-29.
Parks et al. 2015. Genome Res **25**: 1043-1055.
Pena-Gonzalez et al. 2019. Appl Environ Microbiol **85**(24).
Rodriguez et al. 2018. Nucleic Acids Res **46**(W1): W282-W288.
Rodriguez-R and and Konstantinidis. 2016. PeerJ Preprints(e1900v1).
Sczyrbaf et al. 2017, Nat Methods **14**: 1063-1071.

January 20

Ramon Rossello-Mora, Vice-Chair of Judicial Commission of the ICSP, Grup de Microbiologia Marina IMEDEA, Illes Balears, Spain

In response to Christensen's comments of January 16

I recall Cowan 1965 [3]: *The adequacy of characterization of a bacterium is a reflexion of time; it should be as full as modern techniques make possible. Unfortunately, one now regarded as a adequate is likely, in ten years time, to be hopelessly inadequate.* I think taxonomy must adapt to the modern times. To Christensen concerns:

1. Risk for an increase in the number of heterotypic synonyms.

This is independent of genomes as type material. Close to 90% of the species descriptions in IJSEM are single strains [18], mostly without genome provided, nor DDH, using 98.7% 16S rRNA threshold. The use of strict or narrow values (e.g. 70% DDH) has been incorrectly used to force unnecessary classifications [13, 14]. I anticipate that with the genome sequencing, the recognition of heterotypic synonyms will increase. However, genome sequence as a reference will provide a much more stable framework than the simple use of 16S and API strips. The evidences of an evolutionary gap between species ([7, 17], will facilitate circumscriptions as the database grows.

Diseases are not always linked to species identity. Just looking to e.g. *Bacillus cereus* group [8], some traits are linked to a strain and could even be horizontally transferable (e.g. cry genes diagnostic of *B. thuringiensis*). Other clinically relevant traits as e.g. hemolysin or enterotoxin genes could be genus widely distributed [9]. For instance, sequencing *B. toyonensis* genome allowed the (i) detection of clinically relevant genes and (ii) understanding of their non-functional nature. This is a good example of the contrary of what is mentioned.

There are many other cases in where it is clear a strain-specific and not species-specific virulence factors e.g. *Legionella pneumophila* [2], *Vibrio toranzoniae* [11], *Pseudomonas aeruginosa* [4], *Streptococcus uberis* [20], *Ralstonia solanacearum* [21], and so on...

Treatments against clinical infections are mostly done using antibiotic treatment, and sensitivity may be (i) strain specific, (ii) susceptible of horizontal gene transfer and/or (iii) susceptible of spontaneous mutation. Unstable characters, as linked to plasmids (e.g. degradation of naphthalene; [15]) have always been considered not suitable for taxonomic purposes. It is known that characters like phage sensitivity, immunoreactivity [16] and antibiotic susceptibility are often strain-specific and, may be of a lot of relevance for clinical issues but not for taxonomy.

2. Identification of clinically important streptococci bacteria for which vaccines are being developed.

I agree that for vaccine development living material is needed, but immunoreactivity may be strain specific, and virulence factors that can be horizontally transferred. I doubt that clinical microbiologists will abandon cultivation just because the reasons to isolate an organism are very distant from those of the classification purposes.

It would be good to check how many new descriptions in IJSEM are related to clinical cases and with medical relevance. And how many of them have their virulence factors elucidated. I anticipate that if any, very few.

We never underestimated the value of cultivation and evaluation of clinical relevant traits, but the investigation in infection and disease's research is significantly different from classification. I trust that if a study reveals a clinically relevant yet uncultivated organism, this will lead to focus efforts in obtaining pure cultures as occurred with *Salinibacter* [1], *Macondimonas* [10]; and many more examples of ecologically relevant organisms [6, 7, 12, 19].

References:

- 1- Anton et al. 2002. IJSEM. 52:485-491
- 2- D'Auria et al., 2010. (<https://bmcmgenomics.biomedcentral.com/articles/10.1186/1471-2164-11-181>)
- 3- Cowan. 1965. J Gen Microbiol 39: 143-153
- 4- Choi et al, 2002 (<https://jb.asm.org/content/184/4/952>)
- 5- Harbison et al., 2016 (<https://academic.oup.com/femsle/article/363/15/fnw151/2197705>)
- 6- Henson et al., 2018 (<https://www.nature.com/articles/s41396-018-0092-2.pdf>)
- 7- Jain et al., 2019 (<https://www.nature.com/articles/s41467-018-07641-9>)
- 8- Jiménez et al. 2013a. Syst Appl Microbiol. 36: 383-391.
- 9- Jiménez et al. 2013b. Genome Announcements 1: e01080-13
- 10- Karthikeyan et al. 2019. ISMEJ 13: 2129–2134
- 11- Lasa et al., 2017 (<https://www.frontiersin.org/articles/10.3389/fmicb.2017.00086/full>)
- 12- Lee etl al., 2019 (<https://link.springer.com/content/pdf/10.1007%2Fs12275-019-9001-2.pdf>)
- 13- Rosselló-Mora, R. (2006). In: Molecular identification, systematics, and population structure of prokaryotes (Stackebrandt, ed). Springer Verlag, Heidelberg (Alemania). Pp 23 -50
- 14- Rosselló-Móra. 2012. Environ Microbiol. 14:318-334
- 15- Rosselló et al. 1994. Appl Environ Microbiol. 60:966-972
- 16- Rosselló et al. 1992. Syst Appl Microbiol. 15:617-623

- 17- Rossello-Mora and Amann, 2015 (<https://www.ncbi.nlm.nih.gov/pubmed/25747618>)
18- Rossello-Mora and Whitman WB. 2019. Syst Appl Microbiol 42: 5-1
19- Stott et al., 2008 (<https://sfamjournals.onlinelibrary.wiley.com/doi/epdf/10.1111/j.1462-2920.2008.01621.x>)
20- Tassi et al., 2013
(<https://www.sciencedirect.com/science/article/pii/S0022030213004475>)
21- Yang et al, 2013
(<https://www.microbiologyresearch.org/content/journal/micro/10.1099/mic.0.064915-0>)

January 20

Anne Willems, Ghent University, Ghent, Belgium

In response to Christensen's comments of January 16

I have a question regarding what would be the consequences of the newly proposed rules in the following situation: In case a species would be described with a genome sequence as type material, for example in the absence of a culture or in case of a MAG, and later on cultures belonging to that species do become available: can and should a type strain be designated then for that species even though its genome sequence may not perfectly match with the one first proposed? Would the type strain replace the type genome that was first proposed?

January 20

Iain Sutcliffe, Chair of the ICSP, Northumbria University, Newcastle upon Tyne, U.K.

In response to Willem's comments of January 20

This circumstance is directly addressed in the Whitman (2016) proposals, by minor amendment to Rule 18f. which would allow for the replacement of a type sequence of genomic DNA with a type strain (see text in blue below).

This is one of the ancillary changes referred to in my original email. Apologies for not being clearer.

Rule 18f. If a sequence of genomic DNA, description or illustration constitutes, or a dead preserved specimen has been designated, the type of a species [Rules 18a(1) and 18a(3)] and a later strain of this species is cultivated, then the type strain may be designated by the person who isolated the strain or by a subsequent author. This type strain shall then replace the sequence of genomic DNA, description, illustration or preserved specimen as the nomenclatural type. The designation of a type strain in this manner must be published in the IJSEM, the authorship and date of priority of publication being determined by the effective and valid publication of the name by the original authors (Rule 24b). (underlined text are new additions to the current rule)

January 21

Pierre-Edouard Fournier, ICSP Delegate, UMR VITROME, Marseille, France

As a clinical microbiologist, I have been using partial, and then complete genomic sequences for bacterial identification on a routine basis for diagnostic purposes for many years. As a consequence, I support the proposal to use genomic sequences as type material for new taxa when a culture cannot be obtained.

However, and as discussed recently with Iain, I have a few concerns that include:

- Defining quality criteria that will be applied to DNA sequences prior to being used as type material is crucial and may be very difficult for metagenomic data. There are few sequencing systems commercially available currently, but so many sequence analysis softwares and strategies...
- There is a risk of discouraging culture efforts, and notably the deposit in two type culture collections, of strains of previously described *Candidatus* species whose type material is a DNA sequence. There is a risk that microbiologists who cultivate strains belonging to previously described *Candidatus* species only deposit them in a single culture collection, as requested for publishing in most journals, and do not make the effort to publish them as type strains in IJSEM as described in Iain's message below. To avoid this, maybe the cultivators' names should be added to validation lists, not as "discoverers" of the new species but as the first "cultivators".
- New *Candidatus* species will be proposed mainly on the basis of DNA sequence data, as no strain will be available at the time of description. Currently, many new species descriptions use overall genome relatedness indexes and "universal" thresholds such as 70% for dDDH and 95-96% for OrthoANI. However, these thresholds do not apply to all taxonomic groups and may, therefore over- or underestimate the biodiversity of some groups of prokaryotes. When cultivable strains are available, phenotypic data may help with a more precise classification. With a reduced number of phenotypic characteristics evaluable, which will be the case with uncultivated species, this may not be possible.

January 21

William B Whitman, ICSP Delegate, University of Georgia, Athens USA

In response to Fournier's comments of January 21

With regard to Pierre-Edouard's comment about recognition being given to the cultivators, a mechanism already exists to do just that. Changing the type from a sequence to a strain should be recognized as a change in the species circumscription, which would be recognized by an emendation of the species description [see Rule 35]. Emendations are indicated in the defining publication that accompanies the species name. This has already been done for at least one species whose type was a description.

February 6

**Comments from Suresh Korpole, Head, Microbial Type Culture Collection (MTCC)
CSIR-Institute of Microbial Technology, Chandigarh**

I am Suresh Korpole, working at Microbial Type Culture Collection, CSIR-Institute of Microbial Technology would like to make a submission pertaining to microbial systematic studies. We have been experiencing problems in submission of strains at foreign culture collections with the implementation of Biodiversity Act and Nagoya Protocol. India is a participating country of Budapest Treaty. Though our National Biodiversity Authority (NBA) allows us to deposit the proposed type strains and type strains at abroad culture collections, there are certain issues that are preventing the free supply of microbial strains (as NBA request to provide intimation on further supply of strains for any commercial exploitation). We can submit the strains with terms such as any commercial exploitation involving the deposited microbe must be shared equal benefits. In fact, it must be informed to the depositor, which I think is correct as per IPR related regulations. However, editors at IJSEM insist not to add any conditions during the deposition of strains at culture collections, which is in contradiction to the rules of Government at Indian territory. Therefore, it is becoming very difficult to practice microbial taxonomy related research in India that habitat various biodiversity hotspots. As proposed by Prof. William B. Whitman (Whitman 2016; IJSEM, 66; 2018-2112), we sincerely request to amend the rule for the description of novel species and allow use of complete or draft genome sequence as type description. Since, the genome sequence provides all information (including the in silico DNA identity) on phenotypic features, the requirement of essential deposition of strains in two different countries culture collections may be discontinued and request to allow the publication with strain deposited in a single culture collection in the country of researcher residing with genome sequence available for global researchers. This will certainly boost the research ability of enthusiastic researchers residing in countries like India.

Thank you all for going through my views and looking forward to hear a positive news on amending the strain deposition requirements and accepting genome sequence as type description.

February 6

Comments from A. Nemec, Professor of Medical Microbiology, Laboratory of Bacterial Genetics, National Institute of Public Health, Prague, Czech Republic

I do not support Whitman's proposal. If accepted, this change will further broaden room for proposals for novel names with little or no biological meaning. Labelling single isolates with nomenclatural tags has already become a common practice, which is supported by the majority of bacterial taxonomists but considered meaningless or even ridiculous by many non-taxonomists. It is foreseeable that if the proposal is approved, any novel (partial) genome sequence showing ANI values of <95% against those of type strains associated with validly published names will have a chance to become easily a type for a novel species name. And it will be even possible to automate it as there will be no need for analysis of live cultures, e.g. just using publicly available sequences. As even a single cell can be sequenced, it will change taxonomy to a digital form. Although this progress must be expected, I do not understand why sequences should be labelled by formal binomial names, which definitely will occur given this practice for single isolates. I believe that formal binomial names should be reserved for biologically well-defined discrete and internally coherent population entities. I dislike how statistical thresholds (ANI etc.) are universally/ technocratically applied to natural bacterial communities in the absence of a universal concept of bacterial species. People are just labelling

taxonomically unique (in terms of the quantitative thresholds) singletons without any idea about the taxonomic/population nature of what they are labelling. I can repeat here a comment used in my nomenclatural reviews: The nomenclatural code does not explicitly define how many strains are needed for such a purpose, but it states (Rule 27) that the valid publication of a name must be accompanied by a description of a taxon. However, every description of a general category (species) based on a single individual (strain) is in principle meaningless, providing no information about species-specific or diagnostic traits. Furthermore, in the absence of a generally accepted biological concept of species for bacteria, a bacterial species is defined only stochastically, i.e. as a cluster of highly similar/related individuals in the multidimensional phylogenetic space, which are separated (in terms of quantifiable similarity/ relationship) from other such clusters. The analysis of a taxonomically new single organism then cannot give any information about the nature of a new hypothetical cluster or position of the strain within that cluster.

February 7

Comments from Ulrich Nübel, Leibniz Institute DSMZ-German Collection of Microorganisms and Cell Cultures, Germany

I share many of the concerns raised previously about the proposal to allow sequences as types for bacterial nomenclature and I sincerely hope for a wise decision of the ICSP to reject that proposal in its present form. From my perspective, changing the International Code of Nomenclature for Prokaryotes in accordance to the proposal would primarily result in a relaxation of scientific standards, rather than any 'modernization'. Such a change is likely to discredit bacterial taxonomy in the long run, and would probably damage the science of microbiology in general.

The reproducibility of experimental results is a fundamental requirement of any scientific approach. Therefore, throughout the life sciences, making available investigated materials to peers that raise a valid interest is a mandatory requirement as soon as a manuscript gets published in a scientific journal. This ensures that results can be double-checked and reproduced by colleagues, and complemented by additional analyses in the future. The field of molecular microbial ecology may be unique in that this requirement is rarely enforced and it is uncommon to share or exchange environmental samples across laboratories. Transferring this unique negligence to bacterial taxonomy by abolishing the formal requirement to share the underlying investigated material upon describing a novel species will reduce reproducibility severely. These concerns are not addressed by the deposition of DNA (which is not even mandatory in the proposal) and certainly not by depositing sequences in public databases.

While the genome is a part of an organism, a genome sequence is not. Rather, a genome sequence is an experimental result derived from a sample of that organism. In this respect, a genome sequence is even comparable to a microscopical drawing. For good reasons, drawings are not permitted as nomenclatural types any more. While a genome sequence may well contain more information than a drawing, it is still not even guaranteed that all information present in a microscopical drawing can be derived from a genome sequence. Much like a drawing, a genome sequence can be derived from an organism, but not vice versa, and this non-reversibility is due to an inherent loss of information during the sequencing process. I do not question the value of genomic information in general, but for the study of an organism's biology or phenotype, the physical material is indispensable.

The discussion on the replication crisis in science is still ongoing. It thus can only be detrimental to microbiology if a system is deliberately generated that is prone to artifacts and that decreases reproducibility.

Note: I am not an expert in taxonomy. My current research interests are the genomic epidemiology of pathogenic bacteria and the genetic determinants of bacterial secondary metabolite synthesis.

February 7

Comments of Kostas Konstantinidis, Member of Judicial Commission, Georgia Institute of Technology, Atlanta Georgia, U.S.A.

Allow me a few short comments on the issues raised in today's emails against DNA/genome sequence serving as Type material:

1. A genome sequence is indeed required to be publicly deposited as part of the new proposal for validation/checking purposes (see Whitman 2015). I would also argue that checking/validating a genome sequence can be more accurate/precise and more high-throughput than validating a culture; e.g., the latter is typically done by checking i) the 16S sequence, which has low resolution at species level, and ii) the diagnostic phenotype, which is often lab-specific, and not necessarily representative of a major in-situ activity.

2. The single-strain species description issue is NOT specific to DNA/genome sequence but applies the same to cultures. In fact, I would argue that a MAG that represents an abundant population is NOT a single-strain description but the average genome of the population/many cells and thus, carries much more weight than a single strain for identifying diagnostic traits etc. A SAG (single-cell amplified genome) is similar to a single strain and descriptions based on single SAGs should be discouraged, in my view.

We recently published an opinion article that gives more details for the responses above if you have the time to read [Konstantinidis et al. 2020. Environ Microbiol:

<https://sfamjournals.onlinelibrary.wiley.com/doi/full/10.1111/1462-2920.14934>]

In short, I personally remain convinced that the arguments against using genome sequence as type are rather weak overall.

February 8

Comments from Prabhu Patil, Institute of Microbial Technology, Chandigarh, India

I am Prabhu Patil, Scientist working in the Institute of Microbial Technology, Chandigarh, India that hosts MTCC. My training is in bacterial genetics and never knew what is type strain, type species and what is bacterial taxonomy. But because of my association with MTCC, my group is doing core genome-based taxonomic and phylogenetic studies of bacteria, particularly members of *Xanthomonas* genus and the order *Xanthomonadales*. In earlier studies we reported that even clones have been reported into different species! And in the latest study, which is in the biorxiv preprint server, our analysis revealed that *Xyella*, even though a highly reduced genome, is a variant lineage of genus *Xanthomonas* using deep genome-based phylogenetic and taxonomic analysis.

The advent of the web or the internet and genomics era has transformed the field of bacteriology. There is an urgent need and scope to come with terms before things go out of control. Also, considering the way bacterial evolve and regulate the genes, I have two suggestions

1) To allow the use of genome sequence-based approaches to delineate and proposal a strain into new species, genus, and higher taxonomic levels. Hence use genome sequence, as type or reference material (submitting raw reads and assembly in NCBI or EMBL or DDBJ)

2) Allow the proposal of a novel species just based on genome sequence analysis (like ANI, dDDH, AAI), if a researcher has the genome sequence of two or more non-clonal or diverse isolates belonging to the proposed species!

This will democratize plus avoid bureaucracy and also make the field of taxonomy cross-disciplinary and attractive to a new generation of researchers from both basic and applied areas.

February 8

Comments of Markus Göker, Leibniz Institute DSMZ -- German Collection of Microorganisms and Cell Cultures, Braunschweig, Germany

First, I would like to draw the attention of the ICSP to the discussion in mycology about exactly the same kind of proposal (*IMA Fungus* volume 9, pages167–175(2018). The publication available at:

<https://eur02.safelinks.protection.outlook.com/?url=https%3A%2F%2Fdx.doi.org%2F10.5598%2Fimafungus.2018.09.01.10&data=02%7C01%7Ciain.sutcliffe%40northumbria.ac.uk%7C4a79512564f948ede37808d7ac291316%7Ce757cfdd1f354457af8f7c9c6b1437e3%7C0%7C0%7C637167164730146992&data=BzjQSZlrICCBqK7br2%2BEV1FiUWRf8jdaFbHDE3qYys%3D&reserved=0> is critical, negative response to the proposal that sequences can serve as nomenclatural types in mycology that was signed by more than 400 mycologists.

Second, I would like to emphasize that the ballot is not just about the use of genome sequences as types. The forthcoming decision may indeed be regarded as a decision about whether or not genome sequences are permitted as nomenclatural types. This is inaccurate, however, because the decision is about a specific implementation of this idea by specifically modifying the ICNP. Even researchers sympathetic towards genome sequences as nomenclatural types must consider the consequences of these specific modifications.

The suggested phrasings are presented as modest changes which only increase the options of microbiologists. But the proposed changes will not only cause genome sequences to be used as types of microorganisms that cannot be cultivated but also as types of microorganisms that could well be cultivated. Because journals like IJSEM now require a genome sequence for proposals of new taxa there is no extra effort needed to use this sequence as a type. But deposits may be a burden sometimes. The procedures are time-consuming and have become even more bureaucratic lately due to the need for compliance with the Nagoya protocol. If sequences are accepted as types, these efforts shall no longer be necessary. Rather, you only need to sequence an isolate's genome before you can leave it to moulder in a private collection (or autoclave it right away), and then go ahead and validly publish a species description anyway. Predictably, authors will then in most cases take the line of the least resistance and not deposit. Journals cannot effectively control whether or not it would have been possibly to obtain

a pure culture and deposit it in two collections. Thus the net effect is the large-scale replacement of strains as types by genome sequences as types. This holds although it does not seem to be the intention of the authors of the proposal.

The proposed modifications include ambiguous clauses ("whenever possible", "when possible", "when it unambiguously identifies") in relatively huge numbers and at crucial positions. Similarly, the term to "unambiguously identify" is also used but remains undefined. It appears to be dependent on empirical results and on taxonomic opinion, which is subject to change and must not be governed by the Rules of nomenclature and must not govern them.

The Proposal for Rule 18a (3) appears to imply that methods are material. I am not sure whether this makes any sense. All in all it seems to me that these modifications would introduce ambiguity into the ICNP that would make it increasingly difficult to determine whether or not certain taxonomic proposals are in accordance with the Rules. Again, this may not be the intention of the authors of the proposed changes of the ICNP.

February 8

Comments of Joachim Wink, Working Group Microbial Strain Collection, Helmholtz Centre for Infection Research, Germany

Higher benefit from a separate naming system for uncultivated microorganisms

The use of genome sequences as types of validly published names under the ICNP is sometimes regarded as a necessity for microbial ecology. However, it is unclear whether and if so to which extent ecology could actually benefit. Ecologists were always able to name isolates or sequences quite independently of the ICNP and such names acquired a certain stability simply by their reuse in the literature and in databases. The status of being validly published according to the ICNP does not necessarily increase the stability of naming because taxa with validly published names can be reclassified, yielding other validly published names. Names such as SAR11 for a group of uncultivated bacteria were used stably, were easily recognizable and supported the communication of scientific results.. Such names are not even formed in Latin, let alone validly be published.

While SAR11 was discovered in 1990, the first cultivated representative was not available before 2002 and could prominently be published in Nature. Once a (pure) culture is not a prerequisite for assigning a validly published name any more obtaining a (pure) culture will not be interesting any more and thus hardly ever pursued.

In 2012 Brinkhoff and coworkers (DOI: 10.1038/ismej.2011.190) identified and described the Marine Myxobacterial Cluster (MMC) which includes non cultivated Myxobacteria from sediments. The cluster was found on many different places and was described by partial genome sequences. The many efforts in trying to cultivate these organisms failed. For everyone working with Myxobacteria it's clear what the MMC and it is important to be able to directly separate them from the cultivable ones.

The proposal by Whitman included special annotation for distinct kinds of nomenclatural types. But these are not a part of the taxon name. Since taxonomic literature is hardly read, most people only deal with names. Thus the proposed approach would create a lot of confusion by mixing distinct kinds of types. Previous revisions of the Code have intended to reduce this kind of confusion by restricting the kinds of nomenclatural type that can be used.

A separate formal registry system for names of uncultivated microorganisms is clearly preferable. Such a dual nomenclature is often criticized for creating confusion. Yet an informal way of naming clades in ecology always existed in parallel to the valid publication under the ICNP. Significant confusion cannot arise if the kind of name can easily be inferred from the name itself. Names for uncultivated organisms should simply avoid using Latin Linnaean binomials. This may even be advantageous because Latin is nowadays hardly known and non-Latin names such as SAR11 and MMC are already in use. Confusion that arose in cyanobacterial taxonomy under two codes or in mycology when distinct names for anamorphs and teleomorphs coexisted could not occur in such a system.