Comments submitted February 7, 2020

In response to: **Comments from Suresh Korpole,** Head, Microbial Type Culture Collection (MTCC) in India.

The suggestion by Suresh Korpole raises an interesting potential solution for countries like India, Brazil, Bolivia, Colombia, etc., which have installed stringent restrictions on transfer of their genetic resources.

At first consideration, adopting the proposed changes to the International Code of Nomenclature of Prokaryotes (The Code) may seem to solve some problems of national restrictions on transport of resources out of the countries of origin.

Two points:

1) If the Indian regulations governing transport of national resources out of India are based on the Nagoya Protocol (NP) for Access and Benefit Sharing (ABS), the issue described may NOT be solved by the proposed rule changes.

That is because the regulations on ABS govern "genetic resources". This includes, of course, DNA sequence data.

Does India not restrict WGS data as they restrict biological materials? If not, why not?

That is, if the WGS data provides all relevant phenotypic, metabolic, etc. information (I suggest that it does NOT), then not restricting WGS data defeats the purpose of restricting transport of strains.

But, then, it is not necessarily expected that the national regulations of any country will be completely logical!

2) We have worked for many years with Indian microbiologists.We receive many strains without restrictions for deposit in an international collection.

It is my understanding that transport of strains out of India for taxonomic studies is NOT restricted.

I have copied this mail to colleagues in India with whom we have worked for many years. I ask any of them to provide clarification on national restrictions on transport of bacterial strains out of India, i.e., for taxonomic purposes.

If the only problem for Suresh Korpole is a clause in the IJSEM agreement that does not allow a stipulation on commercial development, the IJSEM agreement should be considered, rather than immediately changing The Code.

If such a stipulation are not allowed by IJSEM, I suggest that the IJSEM may be in violation of European law.

The NP for ABS states that the individual countries regulate the sampling, handling, transport and, particularly, commercial development of their national genetic resources.

Any IJSEM restriction on national regulations of commercial development of nomenclatural type material is most likely illegal – such restrictions certainly make no sense, from point of view of taxonomy.

In order to consider the argument of Suresh Korpole in favour of adopting the proposed changes of The Code, I suggest that clarification is needed on the Indian laws regulating microbial strains that are used for taxonomic purposes and also on the IJSEM restrictions on accepting nomenclatural type material.

In any case, changing The Code to try to accommodate the national laws of all countries is illogical.

Thank you for your consideration. Ed Moore, ICSP – SFM, Sweden

Edward Moore, PhD CCUG - Avd. Klinisk Bakteriologi Sahlgrenska Universitetssjukhuset Göteborgs Universitet

Comments submitted February 8, 2020

Dear Colleagues

In this email I want to address the restrictions on the export of cultures and respond to the request by Ed Moore to provide clarity.

Although I do not know the details of the Indian regulations my understanding from discussions with colleagues during the BISMIS meeting in Pune in 2016 was that their regulations are similar to what we have in countries such as South Africa and Brazil. But let me provide clarity by explaining the South African situation.

It is possible to export biological material (in my case bacteria that need to be deposited in a culture collection) for "*research purposes other than bioprospecting*" once you have obtained a permit from the provincial authorities from where the culture was obtained. The export permit requires that every time this culture is supplied to a third party (e.g culture collection to client) permission should again be obtained from the same provincial authority (9 different departments in the case of SA). This restriction is not acceptable under the current regulation of the Code. This restriction is still required even though the MOUs of many culture collection exclude the commercial use of their cultures.

So why have we been able to still describe new species? Although the Nagoya Protocol is only effective after 12 October 2014, our first national regulations were already published in 2008 and European culture collections will not accept cultures isolated after 2007 without the necessary permits. We are "lucky" that cultures isolated before 2008 can still be used as type material as they are not subjected to the conditions of the South African regulations. This is often a fact we take into account when selecting the type strain but as it is now more than 12 years ago, it becomes more difficult and our work is slowly coming to a stand still unless we can get the regulation amended.

For this scenario having DNA sequences as type material (obtained from an existing strain) will be of great benefit for countries known for their biodiversity.

I will address the use of Digital Sequence Information under the Nagoya Protocol in another email.

Regards Fanus Venter Professor: Department of Biochemistry, Genetics and Microbiology University of Pretoria

Comments submitted February 8, 2020

Dear Colleagues

Email to provide some context to an issue raised by Ed Moore:

That is because the regulations on ABS govern "genetic resources". This includes, of course, DNA sequence data.

The issue of how "Digital Sequence Information" should be treated under the Nagoya Protocol is not clear and is currently one of the major issues which will be discussed at COP 15 (Conference of the Parties to the Convention on Biological Diversity) in Kunming, China in October 2020.

Details on the history and current process leading up to COP 15 as well as the views of a number of countries and organisations can be found at <u>https://www.cbd.int/dsi-gr/</u>

In short:

The issue of how "Digital Sequence Information (DSI) on Genetic Resources" should be regulated under the Nagoya Protocol was first raised at COP 13 in 2016. The matter was not resolved at COP 15 in 2018 and is now one of the main issues that will have to be negotiated at COP 15 later this year. Although there is a common understanding among the country representatives that it would not be ideal to restrict the use of sequence data for research purposes, there are concerns related to how sequence data used for commercial applications could be traced back to the country of origin to ensure benefit sharing.

The negotiation on DSI will certainly be linked to the renegotiation of the Convention of Biological Diversity, (the so called post 2020 framework) and it is important that biologists interact with their respective government delegations long before October 2020 to ensure an agreement that would not restrict our research efforts.

Regards

Fanus Venter Professor: Department of Biochemistry, Genetics and Microbiology University of Pretoria South Africa Comments submitted February 9, 2020

Dear Ed, dear collegues,

I would like to contribute to this particular point of the discussion by pointing out some legal facts:

1. According to Indian Legislation (BD Act, 2002), Indian Researchers can apply to deposit bacterial strains in public collections outside of India using Form C. HOWEVER, all non-Indian persons or entities that would like to subsequently access this strain MUST OBTAIN prior approval of NBA according to Section 3 of the BD Act (see attached note, point 3.). This means that Indian strains are NOT publicly accessible even if deposited in international public collections and any access not authorized individually by the Indian NBA is illegal.

2. Given the current state of discussion regarding the inclusion of Digital Sequence Information into the ABS regime of the Nagoya Protocol, it can be expected that the access to genome sequence information will be regulated soon as well. Even at present, certain countries have legislation and/or policies in place that do not permit the free exchange of sequence information. Next October, the COP will probably decide on new regulations that likely will impose severe restrictions on the exchange of DSI on a multilateral international level. That is, it is well possible that from next year on, a deposit of DSI in public databases that potentially could serve as type for the description of a new species will not be legally possibly.

It is obvious, that the amendment of the Code to include genome sequences as type material will not solve any of the above problems.

Best wishes, Jörg Overmann

February 10, 2020

Dear Colleagues,

Thanks to Jörg and Fanus for describing the current status of the issues and activities regarding the legislation and regulation on the use of Digital Sequence Information related to the Convention on Biological Diversity and the Nagoya Protocol on Access and Benefit Sharing.

When I wrote my comment (Feb. 07) to those of Suresh Korpole (Feb. 07), I was not aware of the planned schedule of the COP for specifically addressing the issues of DSI later this year.

Since then, through private correspondence with colleagues and now from the e-mails of Jörg and Fanus, it seems clear that the proposal to use DSI to 'bypass' national regulations on genetic resources, will be facing new coming international regulations.

Important note: It is clear, as has been pointed out, that some countries already have their own national laws in place that regulate the use and distribution of the associated DSI of national genetic resources, even though India does not at this time.

The point of my response to Suresh Korpole and other colleagues who are proponents of the new proposals to change The Code was that adopting the proposals to allow genome and gene sequence data to serve as nomenclatural type material will NOT solve their problems – as they have suggested.

The issues of the problems facing researchers who must include restrictions on commercial or other developmental applications on nomenclatural type material need to be addressed as a separate issue, perhaps with the ICSP, certainly coordinated with the IJSEM. The only restriction I can find on nomenclatural type material (strains), comes from a 2008 revision of The Code (Tindall et al., 2008; Labeda & Oren, 2008), adopted by the ICSP and the JC, that restricts patent strains from serving as type material. In fact, please note the following: "On deposit of the type strain..." minimal conditions must be met but, restrictions on type material are allowed, i.e., "On deposit of the type strain,

Certain rights may be specifically denied, such as commercial exploitation." (Tindall & Garrity, 2008).

In any case, there are other ways of addressing the problems of researchers doing taxonomic work in countries with strict regulations on transport and distribution of their genetic resources.

Most specifically, in the case of these issues, negotiations to amend the policy of the IJSEM should be considered.

Changing The Code is not the solution to these problems.

Thank you for your consideration. Ed Moore, ICSP – SFM, Sweden

Comments submitted February 10, 2020

In reply to Dr. Korpole' s e-mail.

The deposit of strains that serve as nomenclatural types in at least two collections in two different countries (Rule 30 3b) was introduced because there was at least one scientist depositing strains in two collections in the same country and explicitly required that the depositor be consulted before strains could be released. In essence a "safe deposit".

Rule 30 4 also states:

Organisms deposited in such a fashion that access is restricted, such as safe deposits or strains deposited solely for current patent purposes, may not serve as type strains.

Here emphasis is on "such as" in the knowledge that there may be reasons why a person or institute may not be supplied with a particular organism (could include plant, animal or human pathogens where the appropriate laboratory facilities are not available). A discussion document was made available to the ICSP executive board and there were extensive

correspondence with Editors-in-Chief and those responsible for publishing the IJSEM over a period of 18 years.

The deposit of a strain in more than one collection is also a form of "back up". Imagine the scenario should GenBank be on one server without any back ups.

While strains deposited solely for current patent purposes were clearly not permitted, there are numerous examples where strains deposited solely for current patent purposes have been accepted as nomenclatural types (including very recent instances): https://doi.org/10.1099/ijsem.0.003527

In essence any strain originating in India that is deposited in conformity with the requirements laid down by the National Biodiversity Authority and is deposited either in India or in a foreign country will be subject to the same restrictions. <u>http://nbaindia.org</u>

Placing a blanket ban on not accepting collection accession numbers from collections located in India, but then allowing the deposit of the same strains in collections outside of India to serve as nomenclatural types does not solve the problem of "restrictions".

There would appear to be a number of misunderstandings that have arisen over the years that need to be clarified.

Dr. Brian J. Tindall

Comments submitted February 10, 2020

In reply to Dr. Patil, The first part of point 1) is best answered by Principle 1 (4) of the International Code of Nomenclature of Prokaryotes https://doi.org/10.1099/ijsem.0.000778

Nothing in this Code may be construed to restrict the freedom of taxonomic thought or action.

There is nothing to stop you from employing your genome sequence based classification approach based on a scientific justification of the way this is done. However, you must also accept that others may take a different approach and treat your data/taxa in a different fashion and come to different conclusions. They may also use exclusively other data, or combine additional data with yours. In your case you have used genome sequences, whereas previous work has centred on other data.

Point 2) if you have isolates then one of them would be designated the nomenclatural type. At the same time Rule 27 2c and 2d states:

c) The properties of the taxon being described must be given directly after (a) and (b). This may include reference to tables or figures in the same publication, or reference to previously effectively published work.

d) All information contained in (c) should be accessible.

In other words the digital genome sequence information should be "accessible" when included. This was introduced because one particular lab. had not been making digital sequence information available. If you study the growth properties of the nomenclatural type one would select the appropriate (culture collection) strain, whereas if you are comparing genomes you would access the digital genome sequence information that is documented as being derived from the strain that is the designated nomenclatural type. This guarantees a link between the nomenclatural type (as a strain) and the data derived from studying it.

Thank you for also pointing to your paper: https://doi.org/10.1101/2020.02.04.933507

This highlights two issues. Firstly your evaluation is based on a POCP value of 50% as the lower threshold for delineating the genus, while you seem to be using 60% AAI as the lower threshold for genus delineation. There are, however discussions that indicate 50% POCP may be too low, raising it to 60, 65 or 70% would provide a different interpretation of the same data. Similarly, if an AAI value between 60-80% delineates a genus then this could be taken to mean "anywhere" between 60-80%. In the absence of the similarity values it is not easy to interpret the coloured heat map, but again raising the AAI value to 70, 75 or even 80% would allow a different interpretation of the same data. However, the data indicates that one should go back and look at the classification of the group, which is often the case as new data or new taxa become available.

Dr. Brian J. Tindall

Comments submitted February 10, 2020

Dear microbial taxonomists

My passion for the field of microbial diversity and taxonomy of cold habitats dates back to the early 1980s and over the years my lab has published several papers including the description of about a hundred new bacterial and fungal species. It has always been the endeavour of the international community to bring in stringency while describing a new species like data on DNA-DNA hybridisation, lipid profiles, fatty acid profile, 16S rRNA gene sequences apart from all the other classical data and conventional growth, physiological and biochemical data. With the advent of genome sequencing there is a need to relook at the criteria fo4r describing a new species.

I would like to make the following suggestions for a new species, genus, and higher taxonomic level identification. :

1. Retain all the above especially phenotypic and chemotaxonomic characteristics.

2. Adopt whole genome sequence as mandatory including bioinformatic analysis with respect to whole genome similarity, resistome, unique pathways etc.

3. Candidate species where convincing phenotypic data is available along with genomic data.

4. Deposition of the type strain in a recognised culture collection centre any where in the world including the country of origin.

5. Valid certificate of deposition, viability and availability.

I am confident that this would facilitate the work in the exciting area of microbial diversity

and taxonomy without any hurdles.

Thanking you, yours sincerely

S Shivaji

Dr S Shivaji, FASc, FNASc, FTASc, FAMI Director of Prof. Brien Holden Eye Research Centre Prof D Balasubramanian Chair of Research L V Prasad Eye Institute L V Prasad Marg Banjara Hills Hyderabad 500034 India

Comments submitted February 8, 2020

Dear all,

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

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
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 the attached opinion article that gives more details for the responses above if you have the time to read.

in short, i personally remain convinced that the arguments against using genome sequence as Type are rather weak overall. i hope you all have a great weekend! kostas

Kostas Konstantinidis, Ph.D.

Maulding Faculty Fellow and Professor

School of Civil & Environmental Engineering and School of Biological Sciences (Adjunct) Georgia Institute of Technology

311 Ferst Drive, ES&T Building, Room 3202 Atlanta, GA 30332-0512

Comments submitted

Dear Colleagues,

It would appear that publications are appearing that make reference to current Requests for an Opinion and the "loss" of nomenclatural types. Unfortunately, a closer examination highlights other issues that need to be clarified. I have taken 4 examples at random.

In the case of:

Enterobacter siamensis

1) the sequence available from the NBRC (from NBRC 107138) is not identical with HQ888848 (documented as being obtained from the type strain).

2) there are two deposits of HQ888848, HQ888848.1 and HQ888848.2. These two sequences are clearly not identical.

3) neither HQ888848.1 nor HQ888848.2 are identical with the sequence available from the NBRC website, making it difficult to assess whether either HQ888848.1 or HQ888848.2 were ever obtained from the designated type strain or the strain that was deposited. Consequently one cannot rely on the 16S rRNA sequence data and one should check all other data published to see whether it was derived from the strain currently available.

In the case of:

Seliberia and Seliberia stellata.

This organism was first described in 1963 and Mortimer P. Starr obtained a strain that was mentioned in a publication in 1974 from one of the authors of the original description (via G. A. Zavarzin) that was held in the International Collection of Plant Pathogenic Bacteria (a collection that appears no longer to exist). It is unclear whether the current strain in circulation is the original strain, since it appears to come from D. Nikitin rather than the original authors. When originally described 5S/16S rRNA cataloguing/gene sequencing technology was not available and the Request for an Opinion relies solely on these results,

without making any reference to other properties of the strain from the original publication. Just as Pseudomonas radiora has been shown to be a member of the genus Methylobacterium, or that Brevibacterium halotolerans is a member of the genus Bacillus, no other evidence has been presented that Seliberia stellata is not in the same genus as species, currently in the genus Bradyrhizobium. Consider also Hydrogenomonas eutropha moving via Alcaligenes, Ralstonia, Wautersia and Cupriavidus.

Schmidt and Starr make reference to polar growth and the formation of rosettes (not uncommon in members of the Alphaproteobacteria) as well as similarities to members of the genera Nitrobacter and Rhodopseudomonas.

In the case of:

Moorella thermoautotrophica

An extensive publication deals with this issue and other issues that also arise that also relate to the accuracy of deposited digital sequence information:

https://doi.org/10.3389/fmicb.2019.03070

In the case of

Spirillum volutans

Originally described by Ehrenberg in 1832, no strains were isolated at the time. The designated type strain ATCC 19554 appears to be longer viable, but a 16S rRNA sequence has been deposited as GU585672. A second strain, from Pringsheim, ATCC 19553 might be a suitable candidate as a neotype. This also illustrates the wisdom of "back ups"in more than one collection.

Applying good scientific practice it would seem appropriate to assume that those who deposit digital sequence information or prokaryote strains would take appropriate measures to ensure that what is being deposited is authentic.

Dr. Brian J. Tindall

Comments submitted February 14, 2020

Dear Colleagues,

To put things into perspective when evaluating comments made in current publications on the number of Requests for an Opinion dealing with "problematic" nomenclatural types I would like to refer to two publications:

1) Sequencing orphan species initiative (SOS): Filling the gaps in the 16S rRNA gene sequence database for all species with validly published names.

https://doi.org/10.1016/j.syapm.2012.12.006

Among other aspects the project identified some 230 16S rRNA gene sequences that "had to be discarded due to bad sequence quality". These were "replaced" (ie "neo-type" sequences) by better versions. There are additional 16S rRNA gene sequences there were not picked up in that project that have needed to be replaced and a conservative estimate is that this would total 250. If we had to write a Request for an Opinion or propose neotypes to correct each of these sequences then this would mean 250 such publications. "Updating" digital sequence information would require similar mechanisms.

I note also that Alterococcus agarolyticus AF075271 started out its life as a member of the family Enterobacteriaceae (AF075271.1) as indicated in the original publication, but now enjoys a re-incarnation in the family Opitutaceae (AF075271.2) where it seems to rightfully belong. This is an "update" that few people are aware of.

2) Meeting report: GenBank microbial genomic taxonomy workshop (12–13 May, 2015) https://dx.doi.org/10.1186%2Fs40793-016-0134-1

This touches on the issue of the authenticity of strains for which deposited digital sequence information is available and includes data from type strains that are held in culture collections. Sifting through the different databases indicates that there are instances where the genome comes from a strain of a species that is not the species (sometimes also genus, family, order or even class) that the Latin binomial attached to it appears to claim.

See also:

Phylogenomics and systematics in Pseudomonas

https://www.frontiersin.org/articles/10.3389/fmicb.2015.00214/full

Re-evaluation of the taxonomy of the Mitis group of the genus Streptococcus based on whole genome phylogenetic analyses, and proposed reclassification of Streptococcus dentisani as Streptococcus oralis subsp. dentisani comb. nov., Streptococcus tigurinus as Streptococcus oralis subsp. tigurinus comb. nov., and Streptococcus oligofermentans as a later synonym of Streptococcus cristatus.

https://doi.org/10.1099/ijsem.0.001433

Expression of Concern: Micromonospora craniellae sp. nov., isolated from a marine sponge, and reclassification of Jishengella endophytica as Micromonospora endophytica comb. nov. https://doi.org/10.1099/ijsem.0.003487

Dr. Brian J. Tindall Comments submitted 14.02.2020

Comments submitted February 14, 2020

Dear Colleagues,

In reply to Prof. Wink's comments on an "alternative" system. Those familiar with the NCBI taxonomy section, Prokaryote nomenclature up-to-date or NamesforLife will be aware that

behind all Latin names or in the NCBI for names such as "SAR11 cluster bacterium JGI ETNP_125m_186_B03" there are numerical Codes and as such the system referred to by Prof. Wink is already available, perhaps with the one small issue that appropriate reference points (ie nomenclatural types) are not currently defined.

While there may appear to be advantages of using Latin names that refer to "meaningful" ecological or metabolic properties the Code states:

Principle 4

The primary purpose of giving a name to a taxon is to supply a means of referring to it rather than to indicate the characters or the history of the taxon.

General Consideration 8

The International Code of Nomenclature of Prokaryotes is an instrument of scientific communication. Names have meaning only in the context in which they were formed and used.

However, Rhodococcus equii makes no exclusive claim that it is the only red coccus or that there may not be non-pigmented strains, nor does it preclude the fact that it can be isolated from sources other than horses. Removing it to another genus where the name makes no reference to red or coccus would then destroy the information contained in the name, but not the fact that among its properties it may be a red coccus. Latin names may be easier for us to remember, but do not appear to be suitable for bioinformatics work.

Current numerical nomenclatural systems already exist (but without nomenclatural types designated for names not covered by the ICNP), can be easily implemented, dovetail immediately with names validly published under the ICNP and would not interfere with Latin names as currently used. Perhaps one of the major issues is to educate those working outside of taxonomy at present to implement a nomenclatural type based system and to be consistent in the use of nomenclatures (whether Latin based or numerical), including the principle of propriety that is also not always applied consistently in the Latin based system.

Dr. Brian J. Tindall Comments submitted 14.02.2020