

The Taxonomic Stability Imperative

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ABSTRACT

Taxonomic stability is essential if the requirements of a host of stakeholders - health professionals, farmers, environmental consultants and public servants, to name just a few occupations – who need advice on particular organisms are to be satisfied. In many cases this involves the production of species lists. Reliably identified lists of species of one or more groups of taxa are desirable if not mandatory for decision making by stakeholders in environmentally-based long-term research. Both straightforward factors (i.e., quality of the taxonomy that is associated with the species list) and more complex factors behind the compilation (the philosophical undergirding that determines how the species are differentiated in the first place) can often make its interpretation a problematic undertaking, particularly when taxonomic changes occur during the course of long-term studies. Concepts behind the naming of species can deal with patterns or with underlying processes. Whichever way it is undertaken, traditional taxonomy is often slow. This paper evaluates recent attempts to devise cheap, cost effective methods (i.e., barcoding, metagenomics) that will make the taxonomic process more uniform, less labor intensive and hopefully more stable over time.

Key words: taxonomic listings, taxonomic theory, species concepts, COI barcoding, MOTUs, metagenomics

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Introduction

Compared with 'sexier' disciplines such as epigenetics, examination of properties of nanoparticles and environmental ecology, traditional taxonomy tends to be seen as a slightly embarrassing elderly relative; a relative who is stuck with practices that don't seem to have changed much in decades, who is glacially slow in their operations and who seems not to have adapted well to the phrenetic pace of the present. However, the world is in need of taxonomists, trained in traditional morphological as well as molecular techniques, as never before. Global warming is accelerating the spread of vectors of disease such as mosquitoes and tropical flies as well as herbivorous pests that threaten vital broadacre crops. However, pest species are often easily confused with their inoffensive near relatives, resulting in loss of money and time as well as of public health and primary industry productivity. Along with the pest aspects, there is the accelerating loss of biodiversity on our planet; often we do not know what is being lost because organisms have never been described. This is of particular importance in the case of invertebrates that may play a keystone role in the environment. With incessant funding cuts and elimination of places in universities, departments of primary industry and research centres, there is a risk that taxonomic expertise might disappear altogether. We argue here that taxonomy has a much underappreciated place in the modern world – but it needs to be done well. Taxonomy that is done well has a firm, appropriate and reliable epistemology and

methodology. This has not always been the case, as we argue in this paper. If it is to regain lost kudos, taxonomy must show itself to be both stable and reliable – thus the 'Taxonomic stability imperative.'

Before embarking on the main theme of the paper, we offer this personal perspective. Both of us have worked on long-term insect projects for over 20 years, and we here outline some of the taxonomic issues that have affected our assessments. Much of this work has been done in collaboration, hence the joint authorship. As this paper, however, is primarily based on the personal experiences and observations of the principal author, discussion of the work focuses on his experiences.

One of the customary roles of a specialist taxonomist is to compile inventories of species (ants, in this instance) for use by environmental consultants working on a particular project. Ants are particularly useful for the formicological taxonomist, as ant species tend to form associations called 'functional groups' that are habitat-determined. Thus, they are seen as important bioindicators of rehabilitation success by mining companies and similar organisations (e.g., Andersen 1990; Majer *et al.* 2007). The degree of credibility attached to such a list often reflects the degree of scientific sophistication of a reader. However, most would regard it as highly desirable that such lists – or, indeed, citations of taxonomic authors in other written

formats (e.g., reports, minutes, websites, etc.) – be as authoritative as possible. Ideally, the species names on the list should reflect a positive consensus of scientific opinion, and be acceptable by publishing bodies as well as the scientific community generally. Along with a reasonable consensus, the itemized lists should reflect soundly-based research, solid scholarship and minimal subjectivity in their compilation. However, this is not easy to achieve – and has become less so in recent years. This difficulty in producing good taxonomic lists is due to a host of reasons. Broadly, these can be put in the category of the simple, insofar as they affect ordinary, basic morphological taxonomy, or they can be more complex, insofar as they touch on methods and understanding (i.e., the ontology) in a more philosophical sense. In either case, if badly done, the taxonomy becomes effectively unstable and of limited value to the end users.

Confounding factors

Simple factors

In general, the main issues are threefold: (i) the quality of the taxonomy that has led to the erection of a scientific name in the first place; (ii) the taxonomic sophistication of the person who compiles the list, often as a result of a field project (frequently this is an ecologist without specialised taxonomic training), and (iii) the intensity of taxonomic effort (see explanation below).

- i. Any person who has been obliged to read original species diagnoses will be aware of the vast increase in sophistication since the time of Linnaeus. To illustrate from our own field of expertise, a description of the Southern meat ant, *Iridomyrmex purpureus*, by Frederick Smith in 1858 (F. Smith 1858) takes just twelve lines of print in a narrow page. The length of the specimen(s) examined is given in lines (an outmoded unit of length), and for the rest the description is a very generalized coverage of colour and shape – far less than what we now know to be required to identify this ant. The habitat is simply ‘Melbourne.’ There is no illustration. By way of contrast (without being exhaustive), a modern taxonomic description is often computer standardized in keeping with the treatment of other species in the revision, there is a range of measurements of body parts deemed critical, there is almost always a map showing the provenance of specimens examined, type material is discussed, photographs as well as drawings are made available and the morphological recognition of the species is often accompanied by a discussion of its molecular features based on mitochondrial and nuclear genes. This is typically the case when phylogenetic figures are included. This unevenness of taxonomic treatment is hidden in the bare contents of the average listing or in the citations in an article. Added to the differences due to the presuppositions and conventions of the era in which the name first appeared are preferences of

individual taxonomists working in the same period, some of whom were prone to see differences between specimens they examined (i.e., ‘splitters’) and others who tended to find unifying features (i.e., ‘lumpers’).

- ii. Differences in the inherent unevenness of the original taxonomy are often compounded by the degree of recognition of specimens collected by the field worker who has compiled the list. They may be (and often are) a student who may still be ‘learning the ropes.’ They can be completely or largely lacking in taxonomic training. This problem may be obviated if the researcher draws on available taxonomic expertise, but such expertise is often lacking or is patchy.
- iii. Even if the researcher has excellent training, if his or her list comprises multiple categories of organisms, some groups will be well researched through recent monographic revisions while others will be almost unknown. This asymmetry of taxonomic knowledge is almost invariably the case with listings of invertebrates. Names in the well-known groups may be highly accurate due to excellent keys, while those of poorly-known groups may be almost pure guess-work...that is, if the individuals examined are not left despairingly as ‘[Order or Family] sp. 1’, etc.!

Complex factors

The simple factors mentioned above operate on a straight-forward morphological recognition of species, i.e., organisms that have one or more discrete morphological characteristics that enable them to be separated from all other taxa throughout their range. However, increased philosophical examination of the very concept of ‘species’, together with new molecular techniques, have considerably broadened the vexed issue of taxonomic stability. Prior to the work of theoreticians such as Dobzhansky (1937) and Mayr (1942; 1963), species were usually described without specific reference to the philosophy underlying the descriptive process. Now, however, species are often more self-consciously recognized on the basis of differing species concepts, and are consequently defined by different criteria.

Once upon a time it was much easier. Very ancient names actually predate Darwin’s (1859) epochal *The Origin of Species*, with their authors assuming all plants and animals had been fixed by God as unchanging ‘kinds’. These authors’ approach was typology driven: i.e., there was one ‘good’ species with which apparent variants might be compared. Later names reflect an awareness of evolutionary processes but it was not until 1889 (the year in which De Vries introduced the concept of genetic units) that inheritance via molecular particles was introduced to the scientific community. Until the 1940’s taxonomy, at least in our field of Myrmecology, still continued to reflect a typology-driven and confused mixture of ‘typical species’, ‘sub-species’, ‘races’, ‘varieties’, ‘stirps’ and other often vague terminology peculiar to an individual author.

The ground-breaking reconstruction of DNA by Watson and Crick (1954) resulted in a completely different way of looking at evolution in phylogenetics.

Alongside the technical work of identifying how individual organisms pass on their inherited characters has been a parallel process of philosophical examination of 'species'. Numerous definitions of 'species' have been suggested by scientific philosophers since the early 1960's but in general they can be characterized by 'process' (i.e., 'conceptual' theories) or 'pattern' (i.e., 'methodological' theories). We will not labour over these theories here, but a useful coverage can be found in Groves (2001) and Heterick (2009). A very readable online overview of the main species concepts and their strengths and weaknesses can also be found in Zelnio (2012).

'Process-driven' theories

These constructs look to a process such as reproductive isolation, shared mate recognition system or some similar reproductive or ecological factor that is shared by a cluster of organisms, and thus define it as a species in opposition to other, similar organisms that do not share the factor. The most frequently cited theories include the Evolutionary Species Concept (Simpson 1961), the Biological Species Concept (Mayr 1942; 1963), The Recognition Concept of Species (Paterson 1984; 1985) and the Cohesion Species Concept (Templeton 1986). These theories, though often elegant, are, however, difficult to apply to actual field situations; e.g., clinal zones of hybridization do occur in nature, so posing difficulties for Mayr's view (though see de Queiroz 2005, for a way through this difficulty); Paterson's model does not include asexual organisms and Templeton's definition is vague and difficult to apply in the real world.

'Pattern-driven' theories

While this group of species-concepts does not dismiss process, in practice these species-concepts chiefly focus on recognizing a substantive species-delimiting pattern, e.g., in output in terms of a gene tree or similar figure. Cracraft's (1983) Phylogenetic Species Concept is perhaps the earliest of these. More recently, Mallett's (1995) 'Genotypic Species Cluster Definition' is crafted with molecular advances in mind. However, approaches like Mallett's may be overly emphasizing the heuristic (i.e., technical problem-solving) to the detriment of underlying principles required in good theorizing. There are also technical difficulties with Mallett's theory if such factors as long branch attraction that causes wrong association of lineages go unnoticed (Philippe *et al.* 2005). Species theories, based on molecular output, also need to account for factors like incomplete lineage sorting and homoplasy.

Personal experiences resulting from these issues

The issues raised above have had a very practical outworking in our experience. In a companion paper

(Majer and Heterick 2017), we discuss the value of long-term monitoring in jarrah woodland near Perth, Western Australia, using ants as bioindicators. In order to properly evaluate how the woodland had altered over a period of more than 10 years, we first had to reconcile several lists of ant species. Not only was this a matter of assigning names to species originally left undetermined or undescribed (and given a Curtin University voucher number – a number preceded by the identifier 'JDM' - in lieu of a name), but it required emending species and even genus names that had changed over the intervening years. Indeed, over this relatively short period one taxon had changed its generic name twice! Some of the original genus or species names had long been recognized as dubious by researchers, based on the quite different morphologies of the ants within that particular group. Ordinary morphological taxonomic revision had been sufficient to pull out such obvious anomalies. Other ants had been placed in the same category as very similar genera or species because of convergent evolution, and their separation was a surprise. However, revisionary activity using more up-to-date cladistics and molecular methods had confirmed the difference between these similar-looking taxa. Alongside these alterations were names that were left unchanged. Thus, the final list of species found in successive surveys in the same patch of woodland contained names arrived at by different taxonomic methods and, arguably, an increasingly refined systematics approach.

Listings of organisms beside ants can pose additional issues. For example, some fish, such as wrasse, can change sex during their development, resulting in different names originally being given to males and females of the species. Others are highly variable in colour. The brown-spotted wrasse (currently *Notolabrus parilus*) both varies in colour and changes sex during in its lifetime. As a result, this common southern Australian shallow water species has had five genus-level names and seven species-level names in various combinations since its discovery (<http://www.iucnredlist.org/details/187535/0> - Accessed 12 January 2017). Apart from scientific confusion, listings can also invoke opposition and possibly rejection because of sentimental or commercial considerations. In such an instance, the well-known native shrub formerly in genus *Dryandra* was synonymised under *Banksia* by Mast and Thiele (2007), drawing an outcry among some botanists, horticulturalists and nursery owners. Given all these problems, what is the best way forward? We now discuss some of the possibilities.

COI barcoding – the 'magic bullet'?

While traditional morphological taxonomy is still practised (being relatively easy and cheap, although slow), universities, in particular, encourage their taxonomically aligned staff and doctoral students to adopt an integrated molecular and morphological taxonomic approach. Such researchers often routinely use a suite of genes, both mitochondrial and nuclear, that reflect differing rates of evolution. Nowadays,

various phylogenetic programs are available to assess the best trees that result from the combination of multiple gene sequences. In this integrated approach both morphological and molecular taxonomists may combine their distinctive approaches to appraise the output. The determination of species thus becomes a cooperative effort. However, in the interests of efficiency, and, allegedly, cost, barcoding with a single gene has come into prominence within the last fifteen or so years; especially where there is a need to process many different organisms urgently rather than a small group of them at leisure.

Since 2003 (Hebert et al. 2003) DNA barcoding has been promulgated as an ideal way of quickly identifying animal species. (The term 'barcoding' has been coined because the DNA segment obtained after running amplified copies through an agarose gel can be scanned analogously to a bar code being passed through a check-out in a store.) As it was first envisaged, this technique utilizes a standardized region of the mitochondrial cytochrome oxidase 1 (COI) gene, which contains 648 base pairs. While mutations in COI in animal subjects are sufficiently frequent to make this generally a very useful gene for identifying unique patterns that are peculiar to a given species, the evolution of mutations is not sufficiently speedy in plants to make it a similarly useful tool for members of the Kingdom Plantae. The same applies for Fungi (Pires and Marinoni 2010). Barcoding with COI is not only used for pure research but has many applications that make it attractive to private companies and philanthropic organizations; e.g., research workers with the Bill Gates Foundation are using barcoding to identify cryptic species of whitefly (Family Aleyrodidae) that cause havoc in subsistence crops such as cassava grown in Sub-Saharan Africa (<http://www.news.uwa.edu.au/201412187274/international/ted-fellowship-australian-computational-biologist> - accessed 12 August, 2016). Barcoding is no longer limited to COI; more recently, other genes have been preferred for the intractable groups (e.g., see tables in Purty and Chatterjee 2016).

So, is barcoding the answer to the taxonomic stability imperative? Possibly, yes - but with certain important caveats. Ciliates, an important group within the Protista, reveal an extremely high rate of mutation within the COI gene and a rate that is also uneven. Admittedly, the resolution that it brings is also high (a positive here), but standardization within the Ciliates is still beset by many problems (Strüder-Kypke and Lynn 2010). Other barcoding genes may be more useful for protists. However, there are broader issues that make barcoding, whether with COI or with other genes, a tool to be used with care. These include problems in distinguishing intraspecific from interspecific genetic variation (since we don't know the rates of variation and how they differ between taxa), the need to have an already established reference collection of taxonomic species, and the very short lengths of most barcode sequences (which renders them of little assistance in establishing deep nodes in phylogenies)

(Purty and Chatterjee 2016). Other difficulties include introgression, incomplete lineage sorting, and pseudogenes in the mitochondrial genome (Note: pseudogenes can also occur in the nucleus) (https://www.researchgate.net/post/Why_cant_traditional_taxonomy_be_replaced_by_DNA_barcoding - Accessed 12 August 2016). A problem with archived material, in which DNA is lacking or is too denatured to produce a sequence, is another major issue. However, there have been advances in this area in recent years with improved sequencing techniques and other DNA technologies (e.g., Rizzi et al. 2012).

Nonetheless, barcoding is seen as a more attractive instrument than traditional taxonomy for cash-strapped governments beset with global warming, the need to conserve rapidly shrinking natural habitats, the issue of bioprospecting and with all the other influences that require rapid assessments of biodiversity. Indeed, replacing the painstaking process of identification with easy recognition of rafts of species through clusters based on divergence within a barcoding gene (such species being termed 'molecular taxonomic units' or MOTUs) is now being championed by various researchers (e.g., Taberlet et al. 2012). Taxonomy involving MOTUs is often a multiphasic operation involving a team of researchers. (As an example of the new integrated taxonomy, Woodcock et al. (2013), who examine a North American beetle fauna, present a table of established names and also an additional file (File 4) that includes MOTUs as both recognisable and potentially new species.) Proponents of mass barcoding argue that there is nothing about this work that threatens taxonomists as a group. Traditional taxonomists, however, cognisant of already drastically shrinking funds, are wary of good intentions and we have already outlined above the limitations of using a single barcoding gene.

A very new approach (at least for animals) is a 'metagenomic' one that envisages identifying genomic material (that includes multiple molecular markers) derived from large-scale processing of environmental samples - thus avoiding some of the drawbacks of single-gene barcoding - and also the digital capture of multiple phenotypes for pooled biota (Didham *et al.* 2013). Although the authors confess that this process cannot compensate for the lack of integrated taxonomy, they endorse its use as a 'genomic triage' process. The information obtained has low resolution but high coverage, and would be useful as a pointer to lineages of particular interest. Undoubtedly, this approach could theoretically generate useful innovations and enhance the importance of existing ones, e.g., in microscopy or photographic techniques. However, the ability to process huge numbers of specimens with a resultant industrial-level increase in genomic data and images founders if the all-important gatekeepers who can properly evaluate this vast amount of data - the trained molecular and morphological taxonomists - do not experience a corresponding increase in both numbers and institutional support.

Some species are more equal than others

All of these changes to taxonomic methodology that have accrued over the many decades since Linnaeus have left their permanent mark. When people like ourselves inspect a recent taxonomic list compiled by a field worker or other researcher, we often find the species names attract differing levels of reliability, and have to be treated accordingly. These names may result from: (a) assessment by a trained taxonomist; (b) an identification arrived at through use of a morphological taxonomic key; (c) identification by revisionary work undertaken by the researcher themselves; (d) a monophyletic clade in a gene tree; (e) identification confirmed by comparison with a type specimen Automontaged on 'AntWeb' (<https://www.antweb.org/>); and so it goes. As many as half-a-dozen means of identification may be involved. MOTU's may be indicated by a 'species' or 'sp.' sign followed by some signifier (e.g., an alpha or numeric symbol, a voucher number, a Genbank sequence, etc.)

In view of the above complexities, one might be left feeling depressed and dubious of accepting any comprehensive listing that has names supplied by very early taxonomists through to those of recent times, and resulting from different *modi operandi*. The proper response, however, is not despair: although structural taxonomic imperfection may be a given, that does not render a taxonomic listing useless – it simply requires that it be treated with caution and critical scrutiny. (We have also found, in practice, most morphological species are also monophyletic clades in phylogenetic trees based on multiple genes.)

There is more that can be done: scientific bodies and corporates can be lobbied to promote greater recognition of the need to fund sound taxonomic research, and taxonomic awareness can be promoted among civic groups and online. Also, we need to realize that while taxonomic progress seems to be unacceptably slow-moving at times, it is progressing and being ever refined as it moves into the future. However, the need for action is urgent. If we are to achieve a taxonomic environment where the nomenclature is based on a robust and modern framework, taxonomic funding needs to be assigned to institutions and, through means of

grants, to individuals, particularly young students who may be drawn to the field of taxonomy. Sadly, most taxonomy of the traditional variety in Australia is done by retirees (the reader is referred to observations made at the end of the chapter entitled 'A Last Word' in Gunawardene *et al.* 2013). We are in desperate need of youth and enterprise of the sort undertaken by Dr. Brian Fisher in California. Along with his colleagues, Dr. Fisher has produced 'AntWeb', a major database that incorporates data and images of ants from all over the world, including most of the primary types in existence. This has profoundly changed the course of recent ant taxonomy for the better. Taxonomic stability and adequate funding are an inseparable partnership in these rapidly changing times!

A Final note: some practical suggestions

This paper has touched mainly on theoretical and philosophical issues related to taxonomic listings. However, there are also some quite practical ways that taxonomic uniformity can be enhanced. One of these is ensuring that, as far as possible, all collections undertaken by academic staff and students, researchers and employees of large organisations (e.g., environmental consultants) be eventually deposited in a regional depository. A State Museum these days is preferable to a University or State Departmental Collection because continuity of curatorial expertise and attention is virtually assured in the former. Along with this, it is highly desirable that morphospecies names and codes accompany the specimens so that they can be associated together by a researcher revising a taxonomic group. There is a sad history of private and departmental collections – and we have seen some of these – that have been inadequately housed, and have become badly damaged over time by pests or other causes to the point where they have become worthless. There is clearly a need for these collections to be deposited in secure facilities. In Western Australia, in fact, the permit to collect fauna (issued by the Department of Parks and Wildlife) stipulates that significant specimens (i.e., all primary types, a half-share of species or subspecies paratypes, and duplicates [one pair] of species collected that represent a significant extension of geographic range) be donated to the WA Museum upon request.

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