



## A plea for preregistration in taxonomy

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### Abstract

There are notoriously many different definitions of species and methods of species delimitation, forcing taxonomists to make a long range of methodological decisions in species delimitation. Because of this, there are sometimes multiple viable competing methodological paths, which could lead to different ranking (or even grouping) decisions. As a result, it is often unclear what it means for a group to be recognized as a species, the groups recognized as species are not always comparable, and some have even called ranking decisions ‘subjective’. To mitigate the problems this causes for users of taxonomy and taxonomists, we propose that taxonomists across the tree of life should start *preregistering* their research design and criteria for species delimitation in advance of their research. We argue that even if it were to require additional effort, preregistering taxonomic research would strongly benefit taxonomy in the long term, by increasing the transparency and usability of taxonomic outcomes and by reducing the need for *ad hoc* methodological decisions.

**Keywords:** open science, species delimitation, methods in taxonomy, replicability, transparency

### I. Introduction

It is well known that species in biology are not always *comparable*. The reason for this is that they do not have some relevant biological property in common simply by virtue of having species-status (Ereshefsky 2016;

Wilson 1999; Zachos 2016). Depending on which definition of species, methods of species delimitation, and evidence types were used, groups recognized as species constitute very different kinds of things. Some represent reproductively isolated groups, some the smallest groups diagnosable by some morphological character, others monophyletic clades in a phylogenetic tree built using molecular markers, and yet others are groups that meet some threshold of genetic similarity. For many groups of organisms, particularly in charismatic taxa such as birds, mammals and orchids, these diverging taxonomic approaches lead to the circulation of multiple, incompatible classifications (Cuypers *et al.*, 2022; Heller *et al.* 2013; McClure *et al.* 2020; Neate-Clegg *et al.*, 2021).

This heterogeneity in what species represent is not a problem *per se*—most probably there simply isn’t one single best way to classify organisms into species (Ereshefsky 2001). However, it causes problems whenever users of taxonomy assume that species are comparable. Unfortunately, this frequently happens when species are adopted in other fields of science or in downstream uses such as conservation, trade regulation or other domains (Faurby *et al.* 2016; Thomson *et al.* 2021). Part of this confusion may be linked to a lack of transparency about the criteria for species delimitation. Taxonomists do not always specify their criteria for assigning the species rank to groups, making it hard to understand what it means that such a group is a species. This lack of transparency has led some to see taxonomy as a form of ‘art’, considering a species to be whatever an experienced taxonomist calls a species and rendering the criteria for ranking hard or

impossible to define (Luckow 1995). For that reason, some have argued that in addition to the species rank being heterogeneous, individual species are also subjective.

Some authors think that the problems listed above are a reason to abandon the species rank (Mishler 1999; Mishler & Wilkins 2018). Others have argued that we should homogenize the species rank, so that all groups recognized as species present the same properties, be it reproductive isolation or something else (Wheeler & Meier 2000). We think the latter is impossible, and the former is unlikely to happen at least in the short term. We instead propose that taxonomists should *preregister* their research in order to become more transparent about why groups are ranked as species, and to reduce potential subjectivity in ranking. As will become clear, preregistration is not intended as a means to impose any material standards about how good taxonomy is done. Rather, it is a tool designed to make taxonomic methods, reasonings, and decisions more transparent and easier to use and interpret.

## II. What is preregistration?

In the title of a recent and oft-cited paper, Nosek *et al.* (2018) speak of the ‘preregistration revolution’ in scientific research. By preregistration, they refer to the practice to archive a detailed description of the hypotheses, design, and analysis methods of a scientific study on a public repository prior to running that study. The term revolution was well chosen: while two decades ago preregistrations were nearly nonexistent, one important platform for preregistration (<https://osf.io/registries>) now has nearly 100k registrations, and another (<https://aspredicted.org>) receives more than 1,200 registrations per month (Simmons *et al.* 2021).

The growing popularity of preregistration is tightly linked to the growing awareness among scientists and other stakeholders of various interrelated problems that threaten the reliability of scientific research. Most famous is the so-called replication crisis: in a wide array of fields, including medicine, psychology, economy, evolutionary biology, and ecology, a substantial proportion of replications of previously conducted studies fail to yield the same results as the original study (Camerer *et al.* 2016, 2018; Kelly 2019; Open Science Collaboration 2015). This replication crisis is likely due to a variety of causes, including in particular a range of questionable research practices such as p-hacking and various biases such as confirmation bias and publication bias (DuBroff 2018; Dwan *et al.* 2013; Fraser *et al.* 2018; Gopalakrishna *et al.* 2022; Lundh *et al.* 2017; Parker *et al.* 2016).

Many believe that preregistration can contribute substantially to limiting the influence of biases and questionable research practices, increasing the replicability of research (Dirnagl 2020; Nosek *et al.* 2018, 2019). This is because preregistration forces researchers to state their hypotheses, how they will test them, and based on which criteria they will reject or confirm them, *before they run the study*. This is important because complex scientific

research typically involves a long range of small decisions about design, sampling, analysis, and reporting that can often push the results and presentation of the research in various ways (Brown 2020; Conix 2020; Willholt 2009). By recording and justifying these decisions before collecting the data, researchers limit the opportunity for biases to subconsciously influence these micro-decisions.

## III. Why taxonomy needs preregistration

Just like scientists in other disciplines, taxonomists have to make a long range of conceptual and methodological choices in their research, and these choices affect the outcomes of their research (Cuypers *et al.* 2022; Faurby *et al.* 2016; Willis 2017; Zachos 2022). Indeed, divergences in these decisions are what makes species often incomparable and what leads to incompatible taxonomies. Taxonomists have to choose a conception of what species are, what kinds of evidence to collect and how to collect it, pick characters on which to base taxonomic assessments, choose methods to analyze the data, and so on (Schlick-Steiner *et al.* 2010). With many different legitimate species concepts, evidence types, analysis methods, and sampling regions and strategies to choose from, it should be no surprise that two taxonomists investigating the same organisms sometimes come to different taxonomic conclusions, particularly if many of these choices remain implicit.

Preregistration is primarily designed for hypothesis-driven research, making the conditions of accepting or rejecting hypotheses explicit before collecting the data. Much of taxonomy, on the other hand, is exploratory—an important task of taxonomists is to scan the world for unknown diversity. However, recognizing what is encountered as a new species (or other taxon) is something that follows a confirmatory logic in a way similar to accepting or rejecting hypotheses. Just as hypothesis-driven research is geared towards a binary decision, taxonomic research typically concludes with the acceptance or rejection of a particular group as a taxon of a given rank (Zachos *et al.* 2020). While the initial recognition of a new species is not the confirmation of a hypothesis—many will argue that it rather becomes a hypothesis that can be confirmed or rejected by further research—the fact that it is a clear binary decision puts it in a confirmatory structure. And just as other scientists ought to set transparent criteria or thresholds that must be met for a binary decision, taxonomists ought to set transparent criteria or thresholds that determine whether one or more specimens can be seen as a new species or other taxon.

In other words, there is a clear and strong analogy between the problems of the replication crisis and some of the problems in taxonomy. Therefore, while we acknowledge that taxonomy often works differently from many other disciplines, we suggest that the solution that works so well in the rest of science might work for taxonomy as well. In the first place, just like preregistration forces scientists to be *transparent* about

criteria for the confirmation of hypotheses, it would help taxonomists to be transparent about the criteria they use for the recognition of species. In addition, preregistration would force taxonomists to specify their research methods and ranking criteria *before conducting their research*. This way, the role of subjective *ad hoc* decisions is minimized, making it more likely that decisions are made in a consistent and traceable manner, in accordance with predefined principles. As such, transparency provided through preregistration is both valuable to allow the correct interpretation and comparison of taxonomic work, and to help taxonomists in making decisions in a clear and consistent manner.

Thus, we propose that taxonomists adopt the practice of preregistration to improve transparency and avoid the pitfalls that come with the combination of exploratory research with binary decision-making. Minimally, such preregistration should consist in recording detailed decision rules for grouping and ranking decisions. That is, before collecting data (or, in case of pre-existing data, before looking at the data), taxonomists should publicly archive in detail in what cases they would recognize a group of organisms as a taxon, and what necessary and/or sufficient criteria such groups must meet to be recognized as a species. Importantly, these criteria should go beyond broad theoretical descriptions and include an as detailed as possible operational description of the criteria as well. For example, if one preregisters that a group within a particular higher taxon will be recognized as a species if it is reproductively isolated from closely related taxa, this should come with a detailed description of how reproductive isolation will be measured and what degree of such isolation (measured in that way) is sufficient for species status.

However, we hope that taxonomists will go beyond the registration of decision rules, and preregister their decisions for the entire research process and methodology: where, when, and how data will be sampled; how these samples will be stored and processed; how characters will be measured and identified; which data will be retained and which will be thrown out; which methods of analysis will be used; and what the rules are for ranking and grouping decisions. Of course, taxonomic research is very diverse, and it may take some trial and error to find suitable preregistration formats for different taxa and different methods. However, when used successfully, we believe preregistration could be an effective tool to improve the methodological rigor and reliability (and thus, also the appreciation!) of taxonomic research. In appendix to this comment, we present a potential template for an extensive preregistration of an entire research process that can serve as a basis for further debate. In this comment, we focus only on preregistration for taxonomic research regarding the delimitation of species and lower taxa. However, insofar as research on supra-specific taxa follows a similar logic, preregistration could be useful there too.

Of course, preregistration is only one among several tools that are advocated to increase transparency and replicability in scientific research and that are part of a

trend towards more ‘open science’, such as open access, open data, preprints, and—in taxonomy—the registration of taxonomic names. Just like these other tools, preregistration is not intended to impose any material views on how good taxonomy ought to be done—which decisions ought to be made—but merely to improve transparency and accessibility. Preregistration is different from these other instruments in that it takes place before research is conducted, which gives preregistration its unique advantages. Note in particular that ‘preregistration’ is different from the ‘registration’ of taxonomic names, which is for example mandatory for fungi. Preregistration concerns classificatory and not nomenclatural decisions, takes place before the research is conducted, and requires transparency on the taxonomic concepts and methods used, all of which is typically not required when registering a taxonomic name.

#### IV. Advantages of preregistration in taxonomy

Various other papers list advantages of preregistration, both for the individuals doing it and the field in which they function (Nosek *et al.* 2019; Simmons *et al.* 2021; Wagenmakers & Dutilh 2016). These benefits mostly concern the quality of preregistered research and improved study design. Here, we want to discuss separately and in more detail four advantages (some of which we have already pointed to above) that are particular to the application of preregistration in taxonomy.

##### a) Transparency and comparability

Philip Kitcher (1984, 308) quipped that ‘species are those groups or organisms which are recognized as species by competent taxonomists. Competent taxonomists, of course, are those who can recognize the true species’. As discussed before, while there are probably very few taxonomists who adhere to this ‘Cynical Species Concept’, taxonomists often do not provide explicit justification for their ranking decisions (Luckow 1995). Coupled with the fact that there are many competing taxonomic ranking and grouping criteria, this means that the mere recognition of a group as a species typically provides very little information about that group (Mayden 1997). Preregistration of ranking and grouping criteria would drastically improve the transparency of taxonomic research by making clear what is meant by the claim that a group of organisms should be recognized as a species. Registering the taxonomic approach and methods followed in research would also increase the probability that authors are explicit about these criteria in eventual publications—something that is often missing at present.

As such, it would be easier for users of taxonomic information to understand why there is disagreement (*e.g.* different species concepts, different evaluation of the same biological phenomena, the use of genetic rather than morphological data, etc.). This would allow these users to find the taxonomic information that is most suitable for their needs.



### *b) Subjectivity*

Because there are many defensible taxonomic views, and as many criteria for ranking species, and also because taxonomic metrics are continuous rather than discrete, ranking sometimes boils down to making an ‘executive decision’ that is partially arbitrary (Zachos *et al.* 2020). While preregistration cannot get rid of this subjectivity in taxonomic research, it can ensure that the executive decision is made on as principled, consistent, and systematic grounds as possible. If ranking decisions are made while analyzing the data, they are more likely to be influenced by undesirable biases and be inconsistent with similar decisions in other cases. As such, preregistration does not only lead to transparency, but also to the consistent application of the principles and methods that are adopted, whatever they consist of.

### *c) Disagreement*

While taxonomic disagreement can be a sign of a healthy taxonomic research community, it also sometimes hinders taxonomic research and leads to duplicated taxonomic work. The transparency that comes with preregistration could make these disagreements more productive. It could encourage reflection on methodological choices—*are we really measuring what we are interested in?*—and foster explicit debate about these criteria within taxonomic communities. While there has been plenty of debate regarding broad conceptual definitions of species (the so-called species concepts) over the past decades, detailed operational discussions are much rarer and often still quite general in that they cover methods and criteria that are taxon-independent (Camargo & Sites 2013; Conix 2018; Zachos 2016). Preregistration could stimulate debates about ranking and grouping criteria tied to particular operational contexts (e.g. particular taxa).

### *d) Credibility*

The cynical species concept cited above reflects badly on taxonomy: if a species is what a taxonomist calls a species, the science of taxonomy is at risk—unwarrantedly, we believe—of being perceived as an ‘art’ (a term G. G. Simpson also used in a similar context; Simpson 1961) rather than as a serious field of investigation. In that sense, preregistration might play a role in safeguarding the credibility of taxonomy as a science, and might come to serve as a quality label that reflects a rigorous methodological attitude, hence improving the reputation, and perhaps the attractiveness towards funders of the discipline.

## **V. Objections and replies**

As some of its opponents have pointed out, preregistration has limitations and downsides that are important to keep in mind. We list the main points here with regard to the application of preregistration in taxonomy, and explain why we think they do not outweigh the benefits.

### *a) Unknown diversity and flexibility*

One might argue that because taxonomic research is typically exploratory research, it is very hard to predict which methodological decisions will have to be made until one gets to those decisions, having material or data on the table. One of the main aims of taxonomy is to unearth patterns of and structure in biodiversity, and to delimit species on the basis of these patterns. As it is impossible to determine in advance what these patterns will look like, it is difficult to predict in what ways taxonomically relevant units will be distinguishable. In some cases, evolutionary differentiation might occur without obvious morphological change, meaning that morphology as a criterion does not delimit what are considered as reasonable taxonomic units. In other cases, there might be evidence that morphological distinctions are relevant, even if they are not confirmed by available molecular data. In these cases, setting rules in advance might be considered too rigid, and difficult to reconcile with the diversity of evolutionary patterns. The sampling strategy, methods of analysis, and grouping and ranking criteria might only really be possible to choose once one has a better view of the subject of investigation. This fundamentally exploratory character of taxonomy seemingly challenges the confirmatory practice of preregistration.

While we agree that preregistration for complex and largely unknown topics of investigation is hard, we think that for taxonomists familiar with a taxon it is typically possible to foresee many of the methodological decisions to be taken. Of course, unforeseen decisions or factors are likely to crop up and may force taxonomists to diverge from their preregistered plans. However, preregistration is still valuable even if one has to do things differently than preregistered or if it lacks detail for some of the research steps. This is because it forces researchers to state and justify precisely and explicitly why it was necessary to alter the methods. This way, the motivation to deviate from the preregistration is clear for both readers and the researchers conducting the research, and the chance for bias to affect the outcomes remains limited (or, at least, is reduced).

It is also worth mentioning that taxonomists can deal with complexity and the unknown nature of their subject by building flexibility into their preregistration. This can be done by preregistering decision trees rather than a single fixed plan. That is, preregistration can take the shape of a series of steps in which the choice at each step depends on the outcomes in previous steps or on precisely described criteria. This way, preregistrations offer the flexibility to deal with the contingencies that are inevitable in research subjects of which little is known before the start of research.

Still, we acknowledge that sometimes taxonomists will work with diversity that is unknown to such an extent that it is impossible to register (flexible) decision rules for ranking. The space of possible variation has so many dimensions that it is not possible to register methods and rules for all variation they might encounter. In such cases, the best one can do is preregister methods and ranking

rules in as much detail as possible. As said, taxonomic research differs in many ways from traditional hypothesis-driven research, so that its preregistration may need to be held to different standards.

#### b) Workload

Only a small proportion of biodiversity on earth has been described so far, and there are relatively few taxonomists compared to the large task of describing all unknown biodiversity (Costello *et al.* 2013; Fontaine *et al.* 2012; Larsen *et al.* 2017). One might argue that it is unwise to add a time-consuming practice like preregistration to the workload of taxonomists. And indeed, a survey shows that preregistration probably increases the total duration of the research process, and there seems to be a weak association between work pressure and preregistration (Gopalakrishna *et al.* 2022; Sarafoglou *et al.* 2021). There are also additional costs for the whole research community, as the platforms for preregistration require maintenance and new preregistrations sometimes require review (Pham & Oh 2021).

At the same time, however, Sarafoglou *et al.*'s (2021) survey shows that nearly all scientists that have experience with preregistration would recommend it to other researchers in their field. Thus, while preregistration takes additional effort, people who do it generally consider it time well spent. It is also worth pointing out that the increase in effort is probably limited. Most of the work that preregistration requires scientists to do—formulating research questions, designing methods, and choosing methods of analysis—is work they would have to do anyway, either in grant applications, during research, or, in most cases, in writing up papers. Instead of burying this information in inaccessible grant applications, preregistration makes it available to all users of taxonomy. And instead of doing it after data has been collected, preregistration moves this step to the start of the research project. Preregistration thus front-loads much of the work, but does not create much extra work.

In general, compared to the time it takes to carefully design a study, the time it takes to describe that design and archive it would typically be limited. This is definitely the case when taking into account that taxonomists can probably use the same preregistration template across their career for different studies. Note also that preregistrations require no peer-review before one can proceed with the intended research (except for registered reports), so that no additional reviewing work is created.

Thus, while preregistration probably comes at the cost of some additional work, it seems reasonable to say that this cost is limited and outweighed by the benefits of improved methods and transparency. In a way, preregistration also need not be difficult: it does not require additional work in the design of a study, apart from thinking through methodological choices and challenges in advance. As such, we believe preregistration should be possible for professional taxonomists and amateurs alike.

#### c) Pre-existing data

Preregistration should typically be done before collecting specimens and data. This way, it is ensured that data collection processes are less likely to be influenced by researchers after they have seen the first results. However, this is not always possible in taxonomy as taxonomists often use pre-existing data (Fontaine *et al.* 2012), or obtain data from existing collections. While it is true that preregistrations are *ideally* archived before data collection, preregistration can still be useful if this ideal is not met. As long as researchers do not observe and explore the data before choosing their methods, most of the benefits of preregistration still apply. Indeed, most preregistration platforms explicitly allow for the option to analyze pre-existing data. In addition, purely exploratory analysis of data remains valuable as well, as long as it is not used to justify the recognition of a new taxon.

#### d) Preregistration is not infallible

Pham and Oh (2021) point out that preregistration is not sufficient for good science, and still leaves room for selective reporting as well as plenty of wiggle-room for scientists to push results in a direction they desire. However, we assume that such intentionally malicious practices are rare in taxonomy, and only play a minor role in how biases and questionable research practices affect science (Gopalakrishna *et al.* 2022). It is true that if scientists want to cheat, preregistration will not stop them. However, the main benefit of preregistration is that it helps scientists avoid *unintended* roles for bias and questionable research practices.

Similarly, Pham and Oh (2021) point out that one can indeed preregister pointless research projects and bad methodologies—the mere fact that a project is preregistered does not make it good science. However, the transparency generated by a culture of preregistration might raise formal methodological standards, and therefore take oddballs some wind out of the sails. Preregistration does not impose methodological standards, but it incentivizes to justify methodological choices. For scientists who want to do good science—and we assume that this covers nearly all of taxonomy—preregistration is simply a useful but fallible tool. Designing methodologies and running analyses will remain difficult and require practice and skill, and preregistrations are a tool that helps scientists avoid some of the pitfalls in this complex process.

## VI. Conclusions

We have argued that taxonomy could benefit substantially from adopting preregistration as a common practice. This should not be taken to imply that taxonomy is in a worse state than other research fields with respect to problems of replicability and bias. Rather, we see no reason to think that taxonomy is invulnerable to these problems which have been well documented in other fields, and we argue that the benefits of adopting preregistration to address

these might be particularly large in taxonomy because this practice would also dramatically increase the transparency about ranking criteria and research methods in this field. We believe preregistration offers an instrument that can reconcile the diversity of approaches and methods in taxonomy with increased transparency and consistency. This should not be taken to imply that preregistration is more important as a solution for these problems than other aspects of the open science movement. For the sake of clarity, we have only focused on preregistration here, but it goes without saying that practices such as open access publishing, sharing of data and other materials, complete reporting, and replication studies are important too (Marshall & Strine 2021).

With this comment, we wish to encourage all stakeholders in taxonomy, working across the Tree of Life, to consider the benefits of preregistration and make an effort to introduce it into taxonomy. In the first place, we invite taxonomists to use the template we have provided here, potentially improve it, and use it to preregister their taxonomic research. Beyond that, we want to call on taxonomic journals to promote preregistration. While we understand that they might not immediately want to go as far as some influential journals in psychology that have made preregistration obligatory, we hope that they will at least recommend authors to preregister their research and offer scientists the option to submit registered reports.

### Author contributions

All authors were involved in the conceptualization of the ideas in this paper. Stijn Conix wrote the original draft, and all other authors substantially reviewed and edited multiple versions of this draft. Tom Artois, Vincent Cuyppers, and Marlies Monnens developed the example in the appendix. Vincent Cuyppers wrote the first draft of the appendix, and all other authors substantially reviewed and edited multiple versions of this appendix.

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## Appendix: A template for preregistration in taxon delimitation research

*Note: This template represents what we consider as a maximalist preregistration effort, i.e. that is used in an ideal situation. The various notes will guide you through the different steps. An example based on a real-world ongoing taxonomic research project, that of the taxonomic revision of the free-living flatworm complex *Gyratrix hermaphroditus* Ehrenberg, 1831, will be developed throughout the template to illustrate which information is exactly required. As an example, it presents some of the challenges we consider typical for the application of preregistration in taxonomy, such as the extensive use of existing collections, and a lack of clarity on the relevance of taxonomic characters. In practice, the information that must be included for adequate preregistration depends on two fundamental aspects. First, whether research is exploratory or hypothesis-based (in the case of taxonomic revisions). In both cases, confirmatory rules for recognizing taxa must be stated, but in the latter case, a hypothesis must be given as well. Second, whether one is planning to do fieldwork and collect new samples, or planning to collect new data on existing samples, or planning to work with existing data(sets). Some elements are optional, and can be completed when the specific context renders it desirable.*

*Note: This template is roughly based on the standard template provided by the Open Science Framework (Bowman et al. 2020). Some of the instructions are directly copied from this template. For more information on creating a preregistration and where to store it, please consult the Open Science Framework (<https://help.osf.io/article/158-create-a-preregistration>).*

### A. Study Information

#### 1. Title (required):

*Note: Provide the working title of your study. This may or may not be the title that you submit for the publication of your final manuscript. The title should be a specific and informative description of your project.*

**Example:** Detangling the species complex: *Gyratrix hermaphroditus* Ehrenberg, 1831 species complex

#### 2. Description:

*Note: This is the abstract of the preregistration. Briefly summarize the research for readers to get a general idea of what this registration is about.*

**Example:** *Gyratrix hermaphroditus* Ehrenberg, 1831, although never formally split, has for long known to be a species complex that comprises substantial phenotypic and genotypic variation, and there is agreement that it can reasonably be split into several species. Recent research by Tessens *et al.* (2021) has shown that diagnosable morphotypes can be delimited within the complex, and that these morphotypes are monophyletic according to a molecular phylogeny. However, with regard to species delimitation, the authors applied multiple taxonomic methods, based on morphology and genetic species delimitation. This resulted in possible taxonomies that recognize 14, 62 and 78 species, leaving the best manner to proceed with a formal split in doubt. Here, we aim to offer and justify a formal taxonomic revision of the species complex, starting with the diagnosable morphotypes defined by Tessens *et al.*, by testing their consistency with additional material from new locations, and by developing a yardstick for ascribing species status to newly discovered morphotypes in the future.

#### 3. Authors (required):

*Note: Cite all authors that will be involved in the research process, not only the authors contributing to the preregistration.*

#### 4. Research aims (required):

*Note: Elaborate the exact research aims of your study, providing as much background as is needed for the interpretation of the information cited in the preregistration. It serves to provide context to what follows. State the exact research questions or objectives in as concise and concrete a manner as possible.*

**Example:** Our concrete research aims consist in (a.) assessing the consistency of the morphotypes within *G. hermaphroditus* as defined by Tessens *et al.* (2021) with new material; (b.) developing a yardstick for the recognition of new, unknown morphotypes; (c.) delimiting new, unknown morphotypes in the additional material used; and (d.) providing a formal taxonomic revision of *G. hermaphroditus* along the different recognized morphotypes.

#### 5. Study type(s) (required):

- o Discovery of new diversity

- o Exploratory taxonomic revision
- o Taxonomic revision with detailed hypothesis
- o Other:...

*Note: Specify the type of research in which your study fits best. The most straightforward types of taxonomic research are, on the one hand, research efforts aimed at the discovery of unknown diversity, for example concerning a particular taxonomic group in a particular region, and, on the other hand, the revision or testing of an existing taxonomy with a new method or data, in which the existing taxonomy serves as a detailed hypothesis. However, in taxonomy, there appear to be many intermediate forms, in which revisions are conducted, for example, without a detailed hypothesis in mind, although there might be intuitions on splitting or lumping, or in which existing taxa and newly described taxa are combined. For such cases, the category of ‘exploratory taxonomic revisions’ was introduced. Although this category does not start with detailed hypotheses, any hypothesis, however vague, requires preregistration in this category as well.*

**Example:** Our example falls in the intermediate category of ‘exploratory taxonomic revisions’, combining the revision of known diversity along new data, with exploratory research (e.g. the description of new morphotypes).

#### 6. Data type(s) (required):

- o Collection of new samples through fieldwork
- o Collection of new data from existing material
- o Use of existing data
- o Other:...

*Note: Specify all sources of data that are used in your study. This is important to determine which methodological information should be given, for instance about fieldwork, but on the other hand also which possible pre-existing biases must be taken into account when using existing material or data. Existing material concerns studied or unstudied museum samples, herbarium material, etc. Existing genetic sequences count as existing data. Of course, more than one data type can be used. In that case, check all boxes that apply.*

**Example:** We will make use of both new data collected from existing material, and existing data.

#### 7. Hypotheses (required for taxonomic revisions):

*Note: List here the hypotheses you have, with as much detail as possible. If you simply think that an existing taxonomic situation will be upheld, or will be rejected, state that. If you have specific ideas (for example that taxon x will have to be split in three), state that as well.*

**Example:** We expect that the morphotypes of *Gyratrix hermaphroditus* as described by Tessens *et al.* (2021) show consistent, diagnosable morphological differences, and will for that reason, as detailed below, merit recognition as formal species. We also expect to encounter other morphotypes not yet described, for which that will be the case.

## B. General Taxonomic Approach

*Note: Given the variety of approaches that exist in taxonomy, both on the conceptual and on the methodological levels, the main aim of preregistration in taxonomy is to fix a method, limiting the room for conscious or unconscious deviations, and to create transparency. First, you are asked to describe the overall taxonomic approach of your study, which will be detailed in the remainder of the preregistration. Ideally, this general description breaks down into a conceptual and a methodological part, but the latter counts as a minimal requirement.*

#### 8. Species (or other taxon) concept (optional):

Select the species concept that fits your study best:

- o Biological Species Concept  
(*Interbreeding natural populations reproductively isolated from other such groups / all individuals capable of producing fertile offspring.*)
- o Evolutionary/General Lineage/Unified Species Concept  
(*Population or metapopulation-level lineages that evolve separately from other such lineages.*)
- o Genotypic Cluster Species Concept  
(*Groups of individuals or populations that have no intermediates when they are in, or come into, contact.*)
- o Phylogenetic Species Concept, diagnosability version  
(*The smallest group of populations diagnosable by a unique combination of character states.*)
- o Phylogenetic Species Concept, monophyly version

(The least inclusive monophyletic taxon worthy of formal recognition because of its biological relevance.)

- o Other:...

*Note: Species concepts, definitions of species, have been argued to play an important role in structuring taxonomic research. If your study design is guided by a certain species concept, select or cite it here. This step is not strictly required to meet the objectives of preregistration, but it counts as good taxonomic practice to be explicit on the concept of species used.*

**Example:** We follow a Phylogenetic Species Concept, diagnosability version.

#### 9. General approach to species delimitation (required):

*Note: Describe here the general approach to species or taxon delimitation your study will follow. If you follow a specific species concept, describe how that species concept will be operationalized.*

**Example:** Our taxonomic approach builds on the morphologically diagnosable clades supported as monophyletic in the molecular phylogeny of Tessens *et al.* (2021), which are treated as representative of good candidates for species status. Based on additional material and additional data, such as morphometric measurements, we will test these candidates and describe them as formal species accordingly. Based on these results, we will use them as yardstick to describe other such diagnosable groups within the species complex based on similarly divergent morphotypes not yet included in existing phylogenies.

### C. Sample/data collection

*Note: In this section, you will be asked to describe how you plan to collect data and samples for new data. The elements that require completion depend on the data types that were selected above. For all selected data types, the required elements need to be completed. If multiple data sources of the same type are used that follow a different methodology (for example multiple fieldwork campaigns), elements need to be completed multiple times.*

#### 10. [If relevant] Existing data (required)

##### 10.1 Scenario of existing data use:

*Note: The aim of preregistration is to provide as much methodological information before any analysis is performed. Of course, when using existing data, part of the work has already been done. Therefore, creating a research plan in which existing data will be used (for confirmatory research) presents challenges to avoid biases. Please select the description that best describes your situation.*

- o Registration prior to any human observation of the data: at the time of submission, the data exist but have not yet been quantified, constructed, observed, or reported by anyone—including individuals that are not associated with the proposed study. For example, data that have been collected by non-human collectors and are inaccessible.
- o Registration prior to accessing the data: as of the date of submission, the data exist, but have not been accessed by you or your collaborators. Commonly, this includes data that have been collected by another researcher or institution.
- o Registration prior to analysis of the data: at the time of submission, the data exist and you have accessed it, though no analysis has been conducted related to the research plan (including calculation of summary statistics). A common situation for this scenario is when a large dataset exists that is used for many different studies over time, or when a dataset is randomly split into a sample for exploratory analyses, and the other section of data is reserved for later confirmatory data analysis.
- o Registration following analysis of the data: at the time of submission, you have accessed and analyzed some of the data relevant to the research plan. This includes preliminary analysis of variables, calculation of descriptive statistics, and observation of data distributions. Please see [cos.io/prereg](https://cos.io/prereg) for more information.

**Example:** Our study falls in the category of ‘Registration following analysis of the data’, because it builds heavily on the data published by Tessens *et al.* (2021), a study to which some co-authors of this preregistration have contributed.

##### 10.2 Rationale of existing data use (optional):

*Note: Explain here why you will be using existing data, and what measures have been taken so that the principles and objectives of preregistration can be safeguarded. For instance, to assure that you are unaware of any patterns or summary statistics in the data. This*

may include an explanation of how access to the data has been limited, who has observed the data, or how you have avoided observing any analysis of the specific data you will use in your study. For example, an appropriate instance of using existing data would be collecting a sample size much larger than is required for the study, using a small portion of it to conduct exploratory analysis, and then registering one particular analysis that showed promising results. After registration, conduct the specified analysis on that part of the dataset that had not been investigated by the researcher up to that point.

**Example:** Our study builds on the work by Tessens *et al.* (2021) and makes use of their data and non-published data related to that work as collected by coauthors of that study. These data, therefore, are known and fully analyzed, and serve, in the context of this study, as the starting point for the elaboration of the research question and aim, and for the setting of hypotheses. These data will not be used to test our hypotheses on the consistency of the morphotypes described by these authors. That will be done with new data collected from existing material.

#### 10.3 Data collection (optional):

*Note:* Describe how existing data will be collected, which datasets will be used and where they will be obtained. For example, if existing DNA sequences or trait data are used, describe where they come from.

**Example:** All data referred to above will be used, no specific procedure of data collection applies.

#### 10.4 Sampling strategy (optional):

*Note:* If relevant, describe any procedure you will use to make samples of data. What will be the sample size? How many units will be analyzed? If your data collection procedures do not give you full control over your exact sample size, specify how you will decide when to terminate your data collection.

**Example:** Not applicable

### 11. [If relevant] Existing material for new data

*Note:* When using existing material for the collection of new data, similar challenges arise as with the use of existing data. The exact nature of the data, and the way in which they will be obtained, will be specified further in the preregistration.

#### 11.1 Scenario of existing material use:

- o Registration prior to any human analysis of the material: at the time of submission, the material exists but has not yet been observed, or studied by anyone—including individuals that are not associated with the proposed study. For example, field samples that have not yet been looked at.
- o Registration prior to accessing the material: at the time of submission, the material exists, but has not been accessed by you or your collaborators. Commonly, this includes material that has been collected by another researcher or institution, such as herbarium material from another collection.
- o Registration prior to analysis of the material: at the time of submission, the material exists and you have accessed it, though no analysis has been conducted related to the research plan (including calculation of summary statistics).
- o Registration following analysis of the material: at the time of submission, you have accessed and analyzed some of the material relevant to the research plan. This includes preliminary analysis of characters.

**Example:** “Our example makes use of material stored in our lab collection, which has been collected, prepared, but not analyzed from a taxonomic viewpoint.” Our research therefore falls in the category of ‘Registration prior to analysis of the material’.

#### 11.2 Rationale of existing material use (optional):

*Note:* Explain here why you will be using existing material, and what measures have been taken so that the principles and objectives of preregistration can be safeguarded. For instance, to ensure no sampling biases of existing collections are imported in your study, or no bias arises from prior interaction with existing material.

**Example:** We want to make use of a large reference collection of *G. hermaphroditus* with specimens obtained throughout field excursions across the years and in many areas, as additional material to test putative species boundaries, their consistency, and diagnosability. While this collection is subject to sampling bias, it is the largest in the world and sufficiently large for credible use in taxonomic work.



### 11.3 Material collection:

*Note: Describe how existing material will be collected, which material will be used and where it will be obtained.*

**Example:** We plan to use all material identified as belonging to the *G. hermaphroditus* complex available in the lab collection.

### 11.4 Sampling strategy (optional):

*Note: If relevant, describe any procedure you will use to take samples of existing material. What will be the sample size? How many units will be analyzed? If your material collection procedures do not give you full control over your exact sample size, specify how you will decide when to terminate your material collection.*

**Example:** Not applicable

## 12. [If relevant] Fieldwork strategy:

### 12.1 Location of fieldwork (required):

*Note: Describe here where fieldwork will be conducted, in as precise a manner as possible.*

**Example:** Not applicable

### 12.2 Sampling methodology (required):

*Note: Describe here how sampling will be conducted, for example what samples exactly are sought, how they will be collected, how they will be stored, etc.*

**Example:** Not applicable

### 12.3 Sample size (optional):

*Note: If relevant, describe here how many samples you aim to collect, or what will count as a stopping rule to end the sampling effort.*

**Example:** Not applicable

## D. Analysis Plan

*Note: In this section, the details of the actual taxonomic analysis will be expounded.*

### 13. Characters (required)

*Note: Describe each character type that you will use, through existing data, or through observation or analysis of existing or new material. Here, it is particularly important to be as precise as possible. For example, 'morphometrics,' 'morphological traits,' 'mtDNA', and 'mating calls' are too vague. Of course, in various taxonomic approaches the relevance of characters is not always clear from the onset. For this reason, it is advisable to include all characters that are potentially relevant. Other characters can be added to the preregistration throughout the research process, should that appear to be necessary.*

**Example:** Our analysis will focus on morphological characters traditionally used in rhabdocoel taxonomy. These include in particular characters related to (sclerotized parts of) the reproductive organs, such as the general form of the male copulatory organ, form and length of the stylet, sheath and stalk, and proportions between them; the presence and absence of distal hooks, flags, and other protruding structures; and the form and size of the potentially sclerotized vagina. Other potentially relevant characters include the presence or absence of eyes, and the coloration of the mesenchyme tissue. This will be linked to molecular data obtained from 18S, 28S, ITS1, and ITS2 ribosomal DNA genes.

#### 14. Transformations (optional):

*Note: If you plan on transforming, centering, recoding character data, or will require a coding scheme for categorical variables, describe that process.*

**Example:** Not applicable

#### 15. Character analysis (required):

*Note: Describe here any analysis that will be performed on your characters, particularly if multiple characters are used. Be as complete as possible.*

**Example:** We do not plan any quantitative or statistical analysis of characters.

#### 16. Inference criteria (required):

*Note: Describe here the criteria you set for species (or other taxon) recognition. Which criteria must the characters or the output of the analysis meet for the recognition? Note that this is probably the most difficult aspect of preregistering taxonomic research, in particular in an exploratory context. In many contexts, patterns of diversity remain unknown, and a degree of flexibility is required in assessing them and translating them into a species classification. The criteria described here should be as precise as possible in the context of the research in question. They can probably be more precise in explicit hypothesis-based revisions than in exploratory work.*

**Example:** Groups will be recognized as species if samples show consistent, diagnosable differences in the studied morphological characters, primarily of the stylets of the male copulatory organ, to an extent similar to or greater than differences between the known monophyletic morphotypes. Previously unknown characters might play a role in the assessment of new species.

#### 17. Data exclusion (optional)

*Note: Describe here how you will determine what samples or data, if any, to exclude from your analysis. How will outliers be handled? Will you use any awareness check?*

**Example:** Not applicable

#### 18. Missing data (optional)

*Note: Describe here how you will deal with incomplete or missing data.*

**Example:** Not applicable (accessed 13/04/2023)

### E. Other

#### 19. Other (Optional):

*Note: If there is any additional information that you feel needs to be included in your preregistration, please enter it here. Literature cited, disclosures of any related work such as replications or work that uses the same data, or other context that will be helpful for future readers would be appropriate here.*

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