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2 A 'Limitations' section should be mandatory in all scientific papers.
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17
18 **Keywords**

19
20 Reproducibility Crisis, Quality of Research, Limitations, Openness, Transparency.

21
22 **Abstract**

23 It is unusual, and can be difficult, for scientists to reflect in their publications on any limitations their
24 research had. This is a consequence of the extreme pressure that scientists are under to 'publish or
25 perish'. The inevitable consequence is that much published research is not as good as it could, and
26 should, be, leading to the current 'reproducibility crisis'. Approaches to address this crisis are
27 required. Our suggestion is to include a 'Limitations' section in all scientific papers. Evidence is
28 provided showing that such a section must be mandatory. Adding a 'Limitations' section to scientific
29 papers would greatly increase honesty, openness and transparency, to the considerable benefit of
30 both the scientific community and society in general. This suggestion is applicable to all scientific
31 disciplines. Finally, we apologise if our suggestion has already been made by others.

32

33 **Introduction**

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35 All research involves compromise. It is impossible to think of research ever having unlimited time
36 or resources available. In theory, the scientist uses a statistically sufficient number of samples or
37 tests to falsify their hypothesis. A positive result can support but not prove their hypothesis. That
38 being the case, scientists must make trade-offs based on the time and finance available to their project.
39 There is no shame in these compromises, it is the common reality of science. Despite this necessity
40 to compromise, scientific publications do not ask their authors to reflect on the limitations inherent
41 in their study. Whether it be enthusiasm or the need to improve their prominence in the field, it has
42 become common for scientists to minimise or not mention the limitations of their work. This lack
43 of reflection on the limitations can lead to scientists misleading others, as well as themselves. The
44 need for humility is no more evident than in the fields of medical research (e.g. Ioannidis, 2005;
45 Begley and Ioannidis, 2015) and psychology (e.g. Tackett et al., 2019), where a significant
46 proportion of influential, high profile research claims have been demonstrated not to be repeatable.
47 This situation is often called the replication crisis. In the fields in which the authors of this paper
48 work - namely environmental toxicology and chemistry - the situation is less well known, although
49 it seems likely that it is no different than in any other field of science. Aside from repeatability
50 concerns, there are fears that authors are over-selling their work and certainly not drawing attention
51 to limitations (Brain and Hanson, 2021; Hanson and Brain, 2020), which is a form of misleading
52 your audience. There are certainly many examples in our field where there are major differences in

53 opinions; where results from one laboratory are very different from those from another laboratory
54 (see, for example, Sumpter et al., 2021). In addition, there are a few examples where it has been
55 shown directly that previously published results are not repeatable. Probably the most well-
56 documented example was the inability to reproduce the results published in a series of papers that
57 claimed that ocean acidification (caused by rising atmospheric carbon dioxide concentrations)
58 profoundly affected the behaviour of many species of coral reef fish (see Munday et al., 2010 and
59 Clark et al., 2019).

60 Despite growing awareness of this problem of lack of reproducibility for quite some time, it is not
61 apparent that the quality of published research has improved. An indication of this is that the number
62 of retractions of research papers continues to increase (retractionwatch.com). A variety of different
63 strategies have been proposed to address the replication crisis, ranging from how to recognise
64 potentially problematic papers (e.g. Begley, 2013) through to suggestions on how to conduct
65 reproducible research (e.g. Munafo et al., 2017). Most of these recommendations relate to medically-
66 orientated research, although even in relatively small, specialized, fields like ecotoxicology, advice
67 on how to conduct good research is available (e.g. Harris et al., 2014). However, these various
68 recommendations on how to improve the quality of research often tend to focus on a single field of
69 science (e.g. medical research, psychology, ecotoxicology), whereas because lack of reproducibility
70 is a systemic problem throughout science, what is ideally needed are approaches that would be
71 relevant to all fields of science. In this article we suggest that all published scientific papers must
72 include a 'Limitations' section that clearly states the limitations of the study being reported.

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74 **The present situation regarding acknowledging the limitations of a piece of**
75 **research**

76 To gauge how common it is for papers to explicitly state any limitations of the research being
77 reported, we investigated what was stated in the 'Guidelines to Authors' about potential limitations
78 in a group of respected journals in our own research fields. We scanned the 'Guidelines for Authors'
79 for the word limitations. We found that only two of the eleven journals we investigated requested
80 that authors should include mention of any limitations in the papers they submit for publication
81 (Table 1). One journal – Environmental Health Perspectives – ask authors to “Provide a frank
82 discussion of study limitations”. The other journal - Chemosphere - ask authors to "include a
83 paragraph dealing with study limitations in the Discussion section" of their papers. How often
84 authors comply with these requests, or if reviewers are aware of this recommendation to include
85 such statements, or if papers are rejected by these journals because they do not include a statement
86 covering limitations, is unknown (but see below). The other nine journals do not currently refer to
87 potential limitations in their guidance to authors. However, as a move towards openness and
88 transparency, most of these journals encourage authors to submit their 'raw' data (although this is not
89 always mandatory), but this recommendation does not really address the issue of study limitations,
90 because a reviewer or reader would need to form their own opinions on any limitations based on
91 their own assessment of the supplementary information containing the 'raw' data. We have not
92 attempted to determine if journals of other fields of science request statements from authors covering
93 the limitations of their studies, but we suspect that what we discovered in our own research field

94 (Table 1) is likely to be similar to that across all fields of research.

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Journal	Publisher	Limitations explicitly requested	Uncertainty/weakness explicitly requested
Aquatic Toxicology	Elsevier	No	No
Chemosphere	Elsevier	Yes: Include a paragraph dealing with study limitations in the discussion section	No
Environmental Science & Technology	American Chemical Society	No	Yes : An assessment of uncertainty or sensitivity analysis should be included in reported data where applicable
Environment International	Elsevier	No	No
Environmental Health Perspectives	Environmental Health Perspectives	Yes: Provide a frank discussion of study limitations	No
Environmental Toxicology and Chemistry	Environmental Toxicology and Chemistry	No	No
Science of the Total Environment	Elsevier	No	No
Water Research	Elsevier	No	No
Environmental Pollution	Elsevier	No	No
Journal of Environmental Monitoring	Royal Society of Chemistry	No	No
Journal of Hazardous Materials	Elsevier	No	No

97 **Table 1: The results of a search of the guidelines to authors of some respected journals covering**
 98 **the fields of ecotoxicology and environmental analytical chemistry, focused on their advice**

99 **regarding the necessity to explicitly state the limitations of a study.**

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101 We conducted a preliminary assessment of how often authors complied with guidance that requested
102 discussion of the limitations of a study. We investigated a batch of papers published recently by the
103 journal Chemosphere. This journal request that authors include in the Discussion of their papers a
104 paragraph dealing with any study limitations. We investigated 18 papers published in the October
105 2022 issue of the journal (Volume 305). That issue contains 92 original research papers. We selected
106 the first 6 research papers in each of the three main sections of the journal: Environmental Chemistry,
107 Toxicology and Risk Assessment, Treatment and Remediation. Those 18 papers were read by one
108 author of this paper, and independently scanned electronically by another author; their conclusions
109 were identical. Sixteen of the 18 papers made no mention of limitations; the word ‘limitations’ did
110 not occur anywhere in those 16 papers. Two papers did use the word ‘limitations’. One paper used
111 the word just once, and not in a manner that highlighted a limitation of the study being reported. The
112 other paper (Lei et al., 2022) contained a separate section entitled ‘Implications and limitations’,
113 which very openly discussed the limitations associated with the study being reported. Thus, those
114 authors went one step further than the journal guidelines requested, by not hiding any limitations
115 within the Discussion, but instead by highlighting them in a separate section. The authors are to be
116 applauded for doing so.

117 Although journals may not require authors to explicitly mention any limitations their studies had,
118 they might request that authors discuss the uncertainties associated with their studies. Although
119 uncertainties are not necessarily the same as limitations, they could be considered much the same

120 thing. Hence, we also searched the guidelines to authors of the 11 journals listed in Table 1 for use
121 of the word uncertainty (or its plural, uncertainties). Only one of the 11 journals, namely
122 Environmental Science and Technology, included the word in its guidance to authors (Table 1). But
123 doing so was not mandatory; it was required "where applicable".

124 It is also possible that journals would ask authors to discuss any weaknesses to their studies. However,
125 this seems less likely, partly because journals do not want to publish 'weak' studies, and partly
126 because authors would probably be unlikely to admit that their studies contained significant
127 weaknesses. Nevertheless, we also searched the guidelines to authors for use of the word weaknesses.

128 We found that none of the 11 journals utilised this word in their guidance to authors (see Table 1).

129 We realise that authors can be aware of the limitations of their studies, and in their papers might
130 mention these without specifically calling them limitations. Often this is done under the guise of
131 "further research is required".

132

133 **The advantages of making a 'Limitations' section mandatory in all published** 134 **research papers**

135 There would be many advantages to making a 'Limitations' section mandatory in research papers.

136 Some, but by no means all, of these advantages are provided in Table 2. Most of them are equally
137 applicable to all fields of science. They range from improving the training of scientists (both young
138 and established), through to helping reviewers assess the quality of papers, to allowing users of the
139 research reported to determine how much confidence they can have in the results and conclusions of
140 that research.

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1. The training of scientists would be improved.
2. Study design may improve, because scientists would know they now have to explicitly confront potential limitations when designing their studies.
3. Scientists would become much more aware of the concept that all research has limitations, and that discussing these should be a normal part of science.
4. The quality of reviewing papers would improve, because reviewers would be made aware of any limitations authors acknowledged.
5. The quality of published papers would improve, because reviewers could request additional limitations (and their consequences) be added to papers prior to publication.
6. Regulation of chemicals would be both simplified and improved, because regulators could more easily assess the reliability, and hence likely repeatability, of scientific claims.

143 **Table 2: Some advantages of including a 'Limitations' section in all published research papers.**
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145 In addition to those wide-ranging advantages, there will be advantages specific to each field of
146 science. For example, in our field, which covers both the presence of chemicals in the environment
147 and their possible adverse effects on biodiversity, regulators use our research to identify chemicals
148 of concern, then regulate their use if appropriate. Regulations can range from introducing mitigation
149 strategies through to outright, international bans on the use of chemicals. Protecting the environment
150 from chemicals relies on sound science; hence, regulators would benefit greatly if authors had to
151 explicitly state any limitations to their published research.

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153 **Examples of common limitations**

154 Although each field of research is likely to require a unique set of potential limitations, there are

155 likely to be some limitations that apply very widely across most fields of science. For example, the
156 limitation 'sample size was small' will be relevant to many fields, ranging from our own through to
157 psychology and medical research. Table 3 provides some examples of limitations that should be
158 included where applicable in a 'Limitations' section of a paper within the fields of environmental
159 toxicology and environmental chemistry. As research scientists working in those fields, we are
160 acutely aware that many published papers within ecotoxicology and environmental chemistry suffer
161 from one, or more, of those limitations.

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A: Ecotoxicology

1. Only one experiment was conducted
2. Only one concentration was tested
3. The effects were not concentration-related
4. Only nominal concentrations, not actual concentrations, are available
5. The test organism is not well studied
6. Sample size was relatively small
7. Insufficient positive or negative controls
8. Bias was associated with scoring the severity of the effect/end-point
9. These laboratory results should not be readily extrapolated to the natural world
10. Only concentrations higher than those present in the environment were tested
11. There is uncertainty over how meaningful the end-points are to individual health or population success

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B: Analytical Chemistry

1. Lack of reference materials for comparison
2. No analytical standards were used in quality assurance
3. The limits of detection were quite high
4. Matrix effects cannot be excluded
5. The samples analyzed were not representative of the wider environment
6. Lack of unequivocal identification tools (i.e. no NMR data)
7. Use of non-standardized (i.e. not widely accepted) analytical methodology
8. How plausible are my concentrations?
9. Non-target analysis can only tentatively identify unknown chemicals
10. Any claim of novelty should comply with conventional analytical requirements

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C: Field studies

1. The locations sampled may not have been representative
2. The timing of sampling may not have been representative
3. The sampling programme may not have been long enough to encompass natural variation
4. The location was not well characterised
5. Other important stressors or compensating factors were not considered
6. Not enough was reported on the status of local biodiversity or individual populations

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Table 3: Some examples of limitations that should be included where applicable in a 'Limitations' section of a paper within the fields of environmental toxicology and chemistry.

If our suggestion is taken up, and journals begin requesting explicit statements covering the

170 limitations of a study, then it would probably be very helpful if they provided guidance on the most
171 common limitations in each major field of science. Such list would be extremely useful to scientists,
172 especially when they were planning a study. The 'Principles of Sound Ecotoxicology' paper published
173 by Harris et al (2014) provides a list covering many of the general problems encountered in
174 ecotoxicology papers, even if those authors did not explicitly state that their principles could
175 constitute the basis of a list of limitations.

176

177 In order to help scientists, it may be useful to publicise common limitations in specific sub-
178 disciplines. For example, a current hot topic in environmental sciences is that of microplastics, which
179 seem to be ubiquitous contaminants. Already a high number of papers covering microplastic
180 determination and presence in the environment have been published. Yet, a recent study (van Mourik
181 et al., 2021) clearly demonstrates that major problems exist currently with both the identification of
182 the different plastic monomers and the quantification of the number of particles present. The lack of
183 basic analytical standards needed to provide quality assurance was highlighted by Koelmans et al
184 (2019). It is clear that any study on microplastics in the environment needs to include very clear and
185 honest discussion about its limitations. Some of the major limitations that could be considered are
186 offered in Table 4.

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1. What size range are you reporting on, and how does this compare to the literature and indeed the probable natural size range?

2. Have you included positive and negative controls, and published your LOD/LOQ

methodology?
3. How do your analytical methods compare to those used by other scientists?
4. If you used a microscopic system which required the operator to select targets, could operator bias have influenced your results?
5. What software and library did you use? Different software libraries accept greater or lesser matching of spectra with those of standards.

188 **Table 4. A specific example – Limitations relevant to the study of microplastics in the**
189 **environment that merit discussion.**

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191 As an area of science, such as that of microplastics in the environment, matures, problems become
192 known, and ways to tackle them are developed. Put another way, early on in an area of research
193 authors may not realise what the limitations of their studies were, and hence they might
194 unintentionally publish results that subsequently are shown to be questionable. This is probably
195 understandable and inevitable, and is the way that research normally proceeds. Those authors
196 publishing the earliest studies in a difficult field, such as microplastics in the environment, should
197 not be criticized for not being aware of some of the limitations of their studies.

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199 **Implementing a Limitations section**

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201 The authors realise that implementing their suggestion in a fair and balanced way will not be easy.
202 It will be necessary to provide guidelines for both authors and reviewers covering what is required
203 in a 'Limitations' section of a paper. It should be stressed to authors that a fair acknowledgement of
204 limitations is in the author's own interest. This is because it would build trust with the reviewer,
205 reader, and user of the science.

206 Authors (and hence also reviewers) should be asked to reflect on how representative and realistic

207 their studies were. For example, because almost all scientific investigations are limited by the
208 available resources (funds in particular, but also often access to the best equipment), citing "more
209 funds would have led to a more comprehensive study" is not very informative. But if only one river
210 was studied, then stating "only one river was studied, and thus it is not known if our results are more
211 generally applicable" as a limitation is very informative. It may be helpful for the instructions to
212 authors to specify the magnitude and breadth of the limitations that need to be stated in a paper; a
213 list of very minor limitations should not be required.

214 It is likely that journals will need to include a specific question in their reviewer forms that ask
215 whether or not limitations were explicitly discussed by the authors. That section of the review form
216 should also ask the reviewer if the study that they are reviewing has other significant limitations that
217 are not acknowledged by the authors.

218 Ultimately, editors of journals will need to focus on what both authors and reviewers say about the
219 limitations of a study being considered for publication. They will need to decide - as they do to some
220 extent already, of course - which limitations are important and appropriate, and how aware and
221 honest authors have been in covering the main limitations to their studies. Editors may also need to
222 take into account resources (money and staff) and facilities available to authors in order that scientists
223 based in developing countries are not excluded from publishing their research findings. The key
224 ingredient being the degree of transparency and thoughtfulness the authors have shown when they
225 tackled the limitations section.

226 We do not think that bringing in a limitations section will solve all the problems associated with

227 conducting and reporting scientific research. But we do consider that it would be a significant step
228 forward towards improving the openness and honesty (i.e. the integrity) of research.

229 **Conclusions**

230 Improving the quality of published research, in order that it does not mislead, is extremely important
231 to society. We also desire that studies are sufficiently well designed and described that they are
232 repeatable. Although it will not completely solve all these challenges, our recommendation - to
233 make mandatory the inclusion of a 'Limitations' section in all scientific papers – can help this process.
234 We should be frank that all studies have their limitations, and that acknowledging this does not
235 demean the scientists or their research. Openness and transparency is at the heart of science and is
236 central to the confidence that society extends to scientists. For example, a preliminary study (some
237 of which can be very helpful and informative) could be described as such by adding the word
238 'preliminary' to the title of the paper, as was relatively common practice in the past (e.g. Runnalls et
239 al., 2007). As this type of discussion becomes more common, we will have nothing to lose by
240 acknowledging limitations, whilst the scientific community and society would have much to gain.

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246 which allowed us to significantly improve our paper.

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