1 AmphiTherm: a comprehensive database of amphibian thermal tolerance

2 and preference

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49 Abstract

50 Thermal traits are crucial to our understanding of the ecology and physiology of ectothermic animals.

51 While rising global temperatures have increasingly pushed research towards the study of upper thermal

52 limits, lower thermal limits and thermal preferences are essential for defining the thermal niche of

53 ectotherms. Through a systematic review of the literature in seven languages, we expanded an existing

database of amphibian heat tolerance by adding 1,009 estimates of cold tolerance and 816 estimates of
 thermal preference across 375 species. *AmphiTherm* is a comprehensive and reproducible database that

56 contains 4,899 thermal trait estimates from a diverse sample of 659 species (\sim 7.5% of all described

amphibians) spanning 38 families. Despite its broad geographic coverage, we report evident gaps across

amphibian biodiversity hotspots in Africa, most regions of Asia, central South America, and Western

59 Australia. By providing a more holistic understanding of amphibian thermal tolerance and behavioural

60 preferences, *AmphiTherm* is a valuable resource for advancing research in evolutionary biology,

61 ecophysiology, and biogeography, offering insights that are increasingly needed in a changing climate.

63 Background & Summary

64 Thermal trait data are crucial to our understanding of the biology and physiology of ectotherms. The 65 recent increase in broad-scale syntheses of ectotherm thermal physiologies demonstrates recurring interest in how these organisms respond to changing thermal environments¹⁻¹⁰. Much of this work has 66 focused on traits relating to heat tolerance, reflecting the urgency to predict the impacts of climate 67 warming on natural populations^{11–17}. However, climate change also brings an increased probability of 68 extreme weather events, including negative temperature anomalies^{18,19}. As such, a sole focus on heat 69 70 tolerance provides an incomplete picture of ectotherm responses to climate change. A comprehensive 71 understanding of heat tolerance, cold tolerance, and thermal preference is necessary to fully define 72 ectotherm's thermal niches and predict their responses to climate change. Below, we briefly emphasise 73 the importance of these thermal traits for amphibians (see ^{8,20,21} for more in depth discussion).

74 While the significance of heat tolerance in predicting species' responses to warming climates 75 is well documented¹, data on lower thermal limits are equally vital yet often understudied, especially in amphibians, an at-risk, data-deficient group of ectotherms²². Lower thermal limits represents the lower 76 boundary of an organism's thermal niche and have been included in several data syntheses^{5,6,8,10}. This 77 78 trait provides key insights into how species might respond to increasing frequency of extreme cold weather events, which can lead to significant population reduction events known as winterkills^{18,19,23}. 79 80 Gaining understanding of lower thermal limits can thus help predict the sensitivity and resilience of amphibian populations to extreme cold weather events. Moreover, data on lower thermal limits can 81 82 inform conservation and management strategies, for instance, by identifying microhabitats buffering 83 the effects of extreme cold on activity and survival²⁴.

84 Preferred body temperatures reflect the temperature optimising overall performance and the 85 most favourable microhabitat in the absence of other biotic and abiotic factors^{25,26}. Knowledge of preferred body temperatures can thus help predict how climate change will affect species distributions 86 and activity patterns^{24,27–29}. In particular, thermal preference data can be used to infer behavioural 87 thermoregulation patterns and the microhabitats available for crucial physiological processes^{30–32}. While 88 89 upper thermal limits can help predict acute survival in the face of extreme heat, gradual warming below 90 the upper thermal limit thresholds can make some areas unsuitable for amphibians' activity needs²⁴. On 91 the contrary, warming can benefit some amphibians in historically cooler climates by increasing activity windows or reducing hibernation times³³. As such, thermal preference data can help predict the sublethal 92 effects of climate change. Thermal preferences also, for instance, affect susceptibility to pathogens^{34–36}, 93 94 shape the composition of commensal microbes^{37,38}, and mediate interactions between host and microbial communities³⁹. 95

96 Although investigating thermal traits separately provides important knowledge, the study of a 97 combination of thermal traits provides deeper and more comprehensive insights. A simultaneous 98 analysis of upper and lower thermal limits is particularly interesting as it provides an estimation of 99 thermal tolerance breadth²¹—a measure of the thermal envelope ectotherms can occupy in the absence 100 of other abiotic or biotic factors (e.g., competition, resource availability). When thermal preference is 101 integrated with upper and lower thermal limits, the thermal envelope gains shape, providing additional 102 insights to parameterise models and predict activity and survival in changing environments. For instance, leveraging data on thermal limits and preferred temperatures can help infer past, current, and 103 future distributions of ectothermic species^{15,40-43}. Parametrising biophysical models with data on 104 thermal limits and preference now also allow more accurate predictions of overall performance, activity 105 windows^{17,44} and microhabitat heterogeneity^{17,44,45}, strengthening our ability to predict the impacts of 106 climate change on natural populations^{46,47}. From an evolutionary perspective, the integration of different 107

- thermal traits can also advance our understanding of the (co)evolution of these traits, and how climate
 change may shape evolutionary pressures on thermal tolerance and preference^{6,29,48-51}.
- 110 Thermal trait data can also be used to inform conservation efforts. Comparing thermal niche envelopes 111 among amphibians, their microbiota, and potential pathogens can help predict changes in the microbiome and disease risk⁵²⁻⁵⁷. Amphibians are often accessioned into captivity to establish assurance 112 colonies for breeding and eventual reintroduction⁵⁸. Exposure to environmental conditions via "soft 113 release" or mesocosms prior to reintroduction may influence survival and success of reintroduction 114 115 efforts^{59,60}. Knowledge of the thermal tolerance and preference of a broad range of species from different 116 habitats can help inform the design of enclosures that better simulate natural thermal variability^{61–63}. In 117 addition, understanding thermal constraints on activity, demography, and disease risk enhances our 118 ability to identify habitats suitable for repatriation and reintroduction efforts in endangered
- 119 species^{58,64,65}.
- 120 Therefore, a holistic understanding of amphibian thermal tolerance limits and preferences is essential
- 121 for defining the fundamental thermal niche of ectotherms and to project their activity, distribution and
- 122 survival in rapidly changing environments. Here, we expand an existing database on amphibian upper
- 123 thermal limits¹. We aggregated lower thermal limits and thermal preference data from the global
- 124 literature published in seven languages. In doing so, we expanded thermal niche data for 378 species,
- 125 providing a stronger foundation for research on amphibian ecology, evolution, and conservation.

126 Methods

127 Reporting

We reported the contributions of each author using the CRediT (Contributor Roles Taxonomy)
 statement⁶⁶, and MeRIT (Method Reporting with Initials for Transparency) guidelines⁶⁷. We also

130 followed recommendations to maximise the indexing of titles, abstracts, and keywords in databases⁶⁸.

131 *Literature Searches*

132 We adapted methods from ¹ to search the literature on thermal physiological traits. We aimed to compile 133 a comprehensive and representative sample of the experimental literature on lower thermal limits and 134 thermal preference in amphibians, complementing the data on upper thermal limits compiled previously 135 (see ¹ for methods specific to upper thermal limits). PPottier accessed Scopus, ISI Web of Science (core 136 collection), Lens, and ProQuest (dissertation & theses) on 01 November 2022 using The University of 137 New South Wales' institutional subscriptions (full search strings are available in Supplementary 138 Information (Table S1). For studies in English, PPottier modified search strings to accommodate the structure of each database (Table S1) and performed backward searches of previously published reviews 139 of amphibian thermal preference and tolerance^{1,4–6,10}. This resulted in a total of 1676 unique documents. 140 141 We limited the timespan of our searches to 31 May 2021 to match with the timespan from ¹. This 142 decision was made to normalise all searches to a single timespan to simplify future database updates. 143 PPottier also performed Traditional and Simplified Chinese, French, Japanese, Portuguese, and Spanish searches in Google Scholar using search strings translated by native speakers (NR, PPottier, PPollo, 144 145 SN, YY, and RRYO). The searches contained translated singular and plural forms of the following: "amphibian", "frog", "toad", "salamander", "newt", "tadpole", "preferred temperature", "selected 146 temperature", "thermal preference", "Tpref", "Tsel", or "CTmin". Due to search string limitations in 147 148 Google Scholar (256 characters), each term was assessed in its singular or plural form, and the search 149 producing the largest number of results was selected. We performed searches following the format of 150 ("preferred temperature" OR synonyms) AND (amphibian OR synonyms). We also performed separate

151 searches with "CTmin" and ("Tpref" OR "Tsel"), as these terms are commonly used in the literature to 152 refer to lower thermal limits and preferred temperatures. We (PPottier, NC) opted not to use "thermoregulation" as a synonym for "thermal preference" in our search strategy, as pilot searches 153 revealed that this term often returned studies that did not present experimentally-derived thermal 154 155 preference values, or studies that were already captured by the other search terms. PPottier used Publish or Perish⁶⁹ to extract bibliographic records from Google Scholar. We also reused studies (** in Fig. 1) 156 157 from non-English searches conducted in ¹ as the key terms used successfully retrieved results on cold 158 tolerance. However, we limited the inclusion of studies to those meeting our first two criteria (i.e., 159 studies on amphibians, and published in the targeted language) to reduce the volume of screening. We 160 acknowledge that our searches do not encompass all languages relevant to amphibian thermal 161 physiology research and invite speakers of languages not represented in the current version to contribute 162 to future updates of the database.

163 Eligibility criteria

164 We considered studies that empirically tested lower thermal limits or thermal preference in wild or 165 laboratory amphibians. We only included studies on larval, juvenile, and adult amphibians, excluding 166 studies on embryonic stages due to the lack of comparable methods in embryos. For lower thermal 167 limits, we included studies that measured critical thermal minimum (CTmin)⁷⁰, median lethal temperature (LT50)⁷¹, or presented data that were convertible to these metrics (e.g. % survival of cohorts 168 169 tested at different temperatures). CTmin represents the temperature at which a specific physiological or 170 behavioural endpoint is observed—such as the loss of righting response or a lack of response to 171 prodding—when an organism is exposed to progressively decreasing temperatures (e.g., 1°C/min). It 172 does not represent the lowest temperature an organism can tolerate, but rather the onset of functional 173 stasis, the point at which the organism is unable to move and incapable of essential survival behaviours such as thermoregulation or predator evasion⁷⁰. This distinction is important, because many ectotherms 174 175 can recover from temperatures below their CTmin. For instance, some species can recover from freezing 176 to later resume normal function⁷². In contrast, LT50 is the temperature that is lethal to 50% of animals 177 tested and is derived through statistical interpolation from survival rates across a range of 178 temperatures⁷¹. For thermal preference, we included studies that empirically tested amphibian 179 temperature selection in a thermal gradient or shuttlebox via measures of body temperature (or inferred 180 body temperature from the position in the gradient or shuttlebox). We did not include data reported on amphibian body temperatures from uncontrolled (wild) conditions because available environmental 181 182 temperatures were not standardised. We only included studies where thermal history (acclimatisation 183 or acclimation temperature) was reported or could be inferred from the dates and coordinates of sampling. Detailed inclusion criteria and decision trees are presented in Supplementary Information 184 185 (Fig. S1-2, Tables S3-4). PPottier, RRO, PPollo, ANRV, YY, SV, AVL, and NC screened articles for eligibility using Rayyan QCRI⁷³. This software facilitated the identification of key terms in titles, 186 187 abstracts, and keywords to streamline the screening process for large volumes of literature. During data 188 extraction, 47 papers were ultimately excluded for either lacking extractable data or for not complying 189 with our inclusion criteria (43 English, 1 Traditional Chinese, 3 Simplified Chinese). A total of 184 190 studies were deemed eligible for inclusion in the database. Of these, 157 were identified through formal 191 database searches (comprising one study published in simplified Chinese, and another study in French), 192 while 20 non-English studies and an additional 7 English-language studies were retrieved through 193 Google Scholar (Fig. 1). Therefore, nearly 15% (27/184) of the included studies were retrieved through 194 non-English literature searches. Our literature search methods and screening process is summarised in a PRISMA flowchart⁷⁴ (Fig. 1). 195



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Figure 1 | PRISMA Flowchart delineating the databases used to retrieve studies on lower thermal limits
and preferred body temperatures, the number of studies obtained at each stage of the screening process,
and the reasons for excluding studies. * Two studies published in languages other than English (French,
simplified Chinese) were retrieved through English searches. ** Studies from non-English searches
done in Google Scholar from ¹. For the workflow used to obtain data on upper thermal limits, see ¹.

202 Data Extraction

Data extractions were performed by PPottier (7.7% of estimates extracted), R.R.Y.O. (6.7%), PPollo 203 204 (5.3%), A.N.R.V (6.2%), Y.Y. (3.0%), A.V.L. (11.0%), S.V. (20.5%), and NC (45.0.%). Note that these 205 values do not add to 100% because some data entries were extracted by two authors. Data were extracted 206 following the protocols described in ¹. We extracted data directly from text and tables, and primarily used metaDigitise⁷⁵ (version 1.0.1) in R⁷⁶ to extract data presented in figures (although note that some 207 authors have used WebPlotDigitizer⁷⁷ (version 4.7). When data were available in multiple formats (e.g., 208 209 text and figure), we extracted it from the format with the highest resolution (e.g. data stratified by sex 210 or location rather than aggregated across species). Where possible, we extracted measures of data 211 dispersion (i.e. standard deviation, standard error) to accompany mean estimates. In cases where the raw data was available, we calculated summary statistics (means, standard deviation, sample size) to enhance the accuracy of the analysis. For studies reporting survival rates at different temperatures, we

- 214 predicted the temperature at which 50% mortality occurred using logistic regression from the *dose.p*
- 215 function from the MASS package⁷⁸.

216 We also extracted all additional information presented in the studies to allow investigations of the 217 sources of variation in the data and account for non-independence. We assigned identification numbers 218 to each study, and assigned unique identifiers within each study for each estimate, species, population 219 (individuals of the same species sampled from the same geographical location), and cohort (independent 220 group of individuals within a study). Additional variables included sampling coordinates, acclimation 221 temperatures, ramping rates, life stages, endpoints used to infer cold tolerance, or the duration of 222 exposure to experimental treatments. Additional notes were also taken by each researcher extracting the 223 data to facilitate technical validation. The full list of variables is described in Supplementary 224 Information (Table S2). Species names were standardised during the extraction to match 225 AmphibiaWeb⁷⁹ and further standardised to match the most comprehensive phylogenetic tree to date⁸⁰ 226 (see Data Curation).

227 Data Curation

228 To ensure consistency in data extraction across all studies, PPottier and NC extensively 229 reviewed the extracted data to correct typological errors and resolve uncertainties identified during the 230 extraction process. PPottier then curated the data in R⁷⁶ (version 4.4.2), merging the newly extracted 231 data with the previously compiled dataset from ¹. This process involved standardising publication 232 information (publication year, source name) and other variables (e.g., geographical coordinates, IUCN 233 threat status⁸¹) to ensure uniformity across both datasets. PPottier also standardised species names and 234 taxonomy with phylogenetic information from ⁸⁰, which is primarily based on AmphibiaWeb⁷⁹. The combined dataset comprises 324 publications⁸²⁻⁴⁰⁵. Note that 53 of these publications were taken from 235 university dissertations, and some of this work may have now been published^{e.g.,406,407}. 236

237 We also provide a curated version of the database (n = 4,401 estimates), where PPottier excluded data 238 with procedural inconsistencies (e.g., additional stressor, data collected from a single individual), 239 incomplete species information (e.g., Hyloscirtus sp.), and studies involving animals exposed to 240 toxicants, hormones, high levels of UV radiations, or infected with a pathogen. A script detailing the 241 data curation steps is available at https://github.com/p-pottier/AmphiTherm. This data curation step 242 removed 498 estimates from 15 studies and 45 species. However, we believe that this curated dataset 243 offers broader usability. Nevertheless, we also provide the uncurated version of AmphiTherm for users 244 interested in addressing more specific questions (e.g., how toxicants affect thermal tolerance and 245 preference) or identifying existing research gaps within the field.

246 Data Records

247 AmphiTherm encompasses 4,899 thermal physiological trait estimates, derived from 324 248 studies and covering 659 species across 38 families across a broad geographical coverage (Fig. 2-3). This sample represents $\sim 7.5\%$ of all described amphibian species to date⁷⁹ (Fig. 4). According to the 249 IUCN red list⁸¹, most species (79.2%) are either not threatened or data-deficient (Fig. 2), yet 47 species 250 251 are classified as near threatened (NC), 43 as vulnerable (VU), 29 as endangered (EN), 14 as critically 252 endangered (CR), and one species now extinct. Considering that over 40% of amphibians are globally 253 threatened²², this suggests that research on amphibian thermal physiology is predominantly conducted on non-threatened species, likely due to the invasive (or lethal) nature of some thermal tolerance 254

- experiments and the associated conservation concerns for threatened species. This database contains substantial within-species variation, with an average of 7.43 ± 19.5 (mean \pm s.d.) estimates per species, spanning a range of 1 to 292 estimates, with species sampled from an average of 2.51 ± 3.29 populations. Approximately 81% of these estimates include a measure of statistical dispersion (standard deviation, standard error), facilitating their use in weighted (meta-)analyses⁴⁰⁸.
- This database update adds thermal data for 375 species, including lower thermal limits for 300 amphibian species and thermal preference data for 137 amphibian species (n = 1,825 estimates; Fig 3-4). The majority (98%) of lower thermal limit data are derived from CTmin estimates (990 estimates), with roughly 2% of estimates (19 estimates) derived from lethal limits (LT50). Thermal preference data represent 44% of the database update (816 estimates). This update has a relatively broad phylogenetic coverage, spanning 32 families, with 19.2% of records from salamanders (Fig, 3-4).
- 266 Approximately 62.7% of this database is comprised of upper thermal limit estimates (3074 267 estimates from 616 species and 212 studies; Fig. 3-4), highlighting a significant bias towards responses 268 to heat extremes relative to lower thermal limits (1,009 estimates, 300 species, 88 studies) and thermal 269 preferences (816 estimates, 137 species, 114 studies). We found that only 16 studies measured all three 270 thermal traits, covering 59 species (~9% of the species in the dataset; Fig. 3-4). Upper and lower thermal 271 limits were studied more frequently in tandem (60 studies), allowing to calculate the thermal tolerance 272 breadth (i.e., difference between upper and lower thermal limits) of 276 species (~42% of the species 273 in the dataset; Fig. 3-4).
- 274 Geographically, data were collected on all continents where amphibians occur yet exhibit a 275 strong bias towards Nearctic and European regions (Fig. 2). Large geographic gaps in thermal data are 276 evident across Africa, most regions of Asia, Western Australia, and central South America-regions that 277 are biodiversity hotspots for amphibians (Fig. 2). This is particularly concerning as they constrain our 278 understanding of how species from these underrepresented yet extremely diverse regions⁴⁰⁹ might 279 respond to climate change. We also identified taxonomic gaps in existing sampling where an entire 280 order of amphibians, Gymnophiona, remained unrepresented in the database (Fig. 4). In addition, 1 of 281 10 families of Caudata and 7 of 36 families of Anura lack thermal limits or preferred body temperature 282 estimates (Fig. 4). This suggests that further efforts are needed to broaden the research scope and better represent the thermal niche of amphibians. 283
- 284 We found that the majority (88.7%) of the literature on amphibian thermal physiological traits 285 was published in English (4,343 estimates). However, non-English language literature contributes a notable and important portion of the knowledge base, accounting for approximately 11.3% of the 286 287 data. Notably, this includes 289 estimates from publications in traditional Chinese (23 species, 7 288 studies), 131 estimates from Spanish (40 species, 11 studies), 82 estimates from simplified Chinese (12 289 species, 10 studies), 28 estimates from Portuguese (10 species, 3 studies), and 26 estimates from French 290 publications (4 species, 3 studies). Including more languages, such as Afrikaans, Arabic, Bengali, 291 Dutch, German, Hindu-Urdu, Korean, Russian, or Swahili in the screening process may help fill some gaps in future updates to the database⁴¹⁰. Given the historical bias of higher impact publishing outlets 292 against studies on herpetofauna⁴¹¹, there are likely a number of studies in non-indexed journals or 293 regional journals in local languages that were not retrieved using our methods. 294
- The *AmphiTherm* database is stored and archived at <u>https://github.com/p-pottier/AmphiTherm</u>. These contain the metadata (.csv), raw, cleaned, and curated data (.csv), code for data curation and for producing the figures (.Rmd), supplementary data (.csv) and phylogenetic tree (.tre) for producing the figures, and bibliographic files (.ris and .bib) with all the references in the database. Data records are under a CC-BY license, enabling reuse with attribution. Therefore, database users must cite this study

as well as the primary data sources to attribute the original authors and comply with copyrightregulations.



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Figure 2 | Geographic distribution of thermal tolerance and preference data. a) World map showing the
distribution of lower thermal limits (LTL), preferred body temperatures (PBT), and upper thermal limits
(UTL) for anurans (circles) and salamanders (triangles). The shaded area represents the tropics. Note
that coordinates were unavailable for 775 (15.8%) estimates. b) Latitudinal distribution of estimates for
LTL, PBT, and UTL. c) Threat status of species, classified according to the International Union for the
Conservation of Nature (IUCN⁸¹). One species (not displayed) is now extinct.

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Figure 3 | Distribution of mean estimates for three key thermal traits: lower thermal limits (inner heat 312 313 map), thermal preference (central heat map), and upper thermal limits (outer heat map). The number of 314 estimates for each species is displayed as histograms, scaled on a log2(x+1) axis for clarity. The 315 histograms are colour-coded according to the life stage assessed in the experiments, with the category 316 "juveniles" comprising larvae, metamorphic, and juvenile stages. Gray colour represents missing data. The phylogeny relationships are based on the consensus tree from ⁸⁰. a. *Notophthalmus viridescens*, b. 317 318 Dendropsophus ebraccatus, c. Hyla cinerea, d. Pleurodema thaul, e. Ceratophrys cranwelli, f. 319 Craugastor longirostris, g. Rana pipiens, h. Xenopus laevis, i. Plethodon cylindraceus.

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Figure 4 | Distribution of thermal trait estimates across the phylogeny of most extant amphibians.
 Thermal limits and preferences are mapped onto a comprehensive phylogeny of 7,238 species
 (consensus tree, ^{cf.80}) to identify taxonomic biases in existing knowledge. The outer circle presents
 family names, adapted from ⁸⁰.

327 Technical Validation

328 We employed a transparent and reproducible workflow to systematically review over 4,000 studies from 329 five databases and across seven languages. The potential limitations of this database are similar to those 330 described in ¹. First, the methods used for indexing and retrieving studies in Google Scholar are not publicly disclosed⁴¹², which may undermine reproducibility. However, given the limited coverage of 331 332 non-English studies in other databases (with 95% and 93% of references in Scopus and Web of Science indexed in English, respectively), Google Scholar remains one of the most suitable tools to synthesise 333 across multiple non-English languages at present^{413,414}. Second, different authors extracted data from 334 the original studies, introducing the possibility of individual errors. To ensure consistency and accuracy, 335 all extracted data were subsequently cross-checked and standardised by NC and PPottier (see Data 336 337 *Curation*) to minimise the risk of bias and strengthen the reliability of the dataset.

We aim to conduct updates at regular five-year intervals, following the same systematic methods, to maintain the database as an up-to-date resource for amphibian thermal envelope data. We encourage researchers who possess relevant thermal data not included in the current version to contact us so that the database can be updated to continuously reflect the most comprehensive and current body of knowledge.

343 Usage Notes

We anticipate that this database will facilitate a wide range of novel analyses and investigations that may be difficult to foresee at this time, but we are excited to see how these data advance research in amphibian biology and conservation. Our recommendations for using this resource are straightforward: we encourage researchers to have strong foundations in thermal ecology and amphibian biology, and to carefully consider the best approaches for integrating these data into their own investigations.

349 The database represents a comprehensive compilation of studies employing diverse approaches and 350 experimental designs. Given that we cannot anticipate the full scope of research applications, we have 351 made the entire dataset available to allow users to filter and customise the data as needed. We strongly 352 encourage users to clearly document their analytical steps to ensure reproducibility. However, we 353 emphasise that this database version includes data from animals tested under atypical conditions (e.g., 354 amputations, chemical exposure), or from experiments without replication (e.g. data from a single 355 individual). To accommodate most research needs, we therefore also provide a curated version of the 356 database where we excluded data with procedural inconsistencies, incomplete species-level 357 information, and data involving animals exposed to toxicants, hormones, excessive UV radiation, or 358 pathogens. This curated version of the database is likely more suited for research in ecophysiology, 359 though users with more specialised research questions may find value in the complete dataset. Scripts 360 detailing the data cleaning and curation processes are available at https://github.com/p-361 pottier/AmphiTherm and should provide further guidance for researchers in tailoring the dataset to their 362 specific research needs.

363 As described in the first iteration of this database¹, the data contain inherent sources of non-364 independence as multiple estimates were extracted from each study, species, population (multiple 365 sampling locations from each species), and cohort (e.g., repeated measures on the same individuals). 366 We recommend that users use phylogenetically-informed statistical models with hierarchical randomeffect structures to account for and partition sources of variation^{408,415}. Users should also account for 367 368 variations in sampling effort (sample size differences), for instance, by weighting estimates by the 369 inverse of their sampling variance⁴⁰⁸. Employing hierarchical models that incorporate sampling 370 variance can help address issues of biological and methodological non-independence, enabling more 371 accurate inferences of the factors driving variation in the data⁴⁰⁸. Most (81%) estimates are associated 372 with a measure of dispersion (standard deviation or standard error), species information is standardised to published phylogenetic information⁸⁰, and unique identifiers have been assigned to each study, 373 374 species, population, and cohort. These features make *AmphiTherm* readily applicable for use in complex 375 statistical models aimed at uncovering the drivers of thermal tolerance and preferences in amphibians.

As described in previous studies, thermal traits in amphibians are influenced by multiple variables, including acclimation temperature, acclimation time, ramping rate, endpoint metrics, body size, sex, assay duration, and geographic origin, among others^{8,20,21}. We recommend careful attention to these variables, with consideration of incorporating sources of methodological or biological variation as covariates in statistical models, to better capture the complexities of amphibian thermal ecology. Finally, it is important to note that due to the data gaps in hotspots of amphibian diversity, the data herein represent only a subset of total amphibian diversity, and subsequent analyses should acknowledge thislimitation.

384 Code Availability

The code used to process the data and produce the figures for this manuscript is available at <u>https://github.com/p-pottier/AmphiTherm</u>, and the repository will be archived to Zenodo upon acceptance.

388 Author Contributions

- 389 Conceptualisation: PPottier (lead), NC, SB, TA, SMD, SN.
- 390 Methodology: PPottier (lead), NC, TA, SMD, SN
- 391 Software: PPottier
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- 393 Investigation: NC (lead), PPottier, RRYO, PPollo, ANRV, YY, SV, AVL, SB, H-YL, JOV, SV
- 394 Data Curation: PPottier (lead), NC
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- 396 Writing (Original Draft): NC (lead), PPottier
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- 398 Project administration: NC (lead), PPottier
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- 400 All authors gave final approval for publication.

401 **Competing Interests**

402 The authors declare no conflict of interest or competing interests.

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Figure S1: Decision tree used to screen titles, abstracts, and keywords. Additional details can be
found in Table S3.

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Figure S2: Decision tree used to assess full articles for eligibility. Additional details can be found
in Table S4.

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42 Table S1: Search strings used for the different databases.

Database	Search strings
Scopus	TITLE-ABS-KEY("temperature*" OR "thermal" OR "cold*" OR "cool*") AND TITLE-ABS-KEY("cold tolerance*" OR "tolerance* to cold" OR "thermal min*" OR "CTmin" OR "CT min" OR "chill coma" OR "cold stress tolerance*" OR "tolerance to cold stress" OR "cold stupor" OR "cold resistance" OR "resistance to cold stress" OR "supercooling point" OR "SCP" OR "crystal* temperature*" OR "cold hardiness" OR "freez* tolerance" OR "tolerance to freezing" OR "preferred temperature*" OR "preferred body temperature?" OR "temperature preference*" OR "selected temperature* OR "selected body temperature*" OR "thermal prefer*" OR "temperature* prefer*" OR "temperature* select*" OR "thermal selection") AND TITLE-ABS-KEY("amphibia*" OR "frog*" OR "toad*" OR "salamand*" OR "newt" OR "newts" OR "tadpole*" OR "metamorph" OR "metamorphs" OR "caecili*" OR "therpelidae" OR "typhlonect*" OR "indotyphlid*" OR "dermophi*" OR "siphonop*" OR "caudata" OR "urodela" OR "cryptobranch*" OR "proteidae" OR "rhyacotriton*" OR "amphium*" OR "lethodon*" OR "proteidae" OR "rhyacotriton*" OR "amphium*" OR "alyt*" OR "hinophryn*" OR "pipidae" OR "kenopus" OR "cayptobranch*" OR "alyt*" OR "hinophryn*" OR "pipidae" OR "kenopus" OR "scaphiop*" OR "alyt*" OR "hinophryn*" OR "pipidae" OR "heleophryn*" OR "lethodon*" OR "myobatrach*" OR "rhinoderma*" OR "leptodactyl*" OR "hylod*" OR "atrachyl*" OR "cycloramph*" OR "telmatob*" OR "caratophry*" OR "hemiphract*" OR "hyla*" OR "hylidae" OR "heleophryn*" OR "cauptotoephalell*" OR "hyla*" OR "hylidae" OR "heleophryn*" OR "netohophryn*" OR "leptodactyl*" OR "leptodactyl*" OR "notohophryn*" OR "pipidae" OR "hereophaliae" OR "caugastor*" OR "strabomantidae" OR "pipidae" OR "hereiphidae" OR "corauagastor*" OR "strabomantidae" OR "piristimantis" OR "nasikabatrach*" OR "corauaa" OR "microhyl*" OR "athroleptid*" OR "hyloreof*" OR "hemisus" OR "odontobatrach*" OR "phrynobatrach*" OR "notycibatrach*" OR "microhyl*" OR "athroleptid*" OR "hyperol*" OR "hereiptidae" OR "hemisus" OR "odontobatrach*" OR "phrynobatrach*" OR "nyctibatrach*"
Web of Science (core collection)	TS=("temperature*" OR "thermal" OR "cold*" OR "cool*") AND TS=("cold tolerance*" OR "tolerance* to cold" OR "thermal min*" OR "CTmin" OR "CT min" OR "chill coma" OR "cold stress tolerance*" OR "tolerance to cold stress" OR "cold stupor" OR "cold resistance" OR "resistance to cold stress" OR "supercooling point" OR "SCP" OR "crystal* temperature*" OR "cold hardiness" OR "freez* tolerance" OR "tolerance to freezing" OR "preferred temperature*" OR "preferred body temperature*" OR "temperature*" OR "selected temperature*" OR "selected body temperature*" OR "thermal prefer*" OR "temperature*" OR "temperature*" OR "thermal selection") AND TS=("amphibia*" OR "frog*" OR "toda*" OR "salamand*" OR "newt" OR "newts" OR "tadpole*" OR "metamorph" OR "salamand*" OR "caceili*" OR "thermal*" OR "isiphonop*" OR "caudata" OR "indotyphild*" OR "dermophi*" OR "sirenidae" OR "amphibia*" OR "leicamptodon*" OR "cryptobranch*" OR "hypobiid*" OR "sirenidae" OR "amphibia*" OR "leicamptodon*" OR "anura*" OR "hepelodat*" OR "hepelodat*" OR "hepelodat*" OR "hepelodat*" OR "hepelodat*" OR "hepelodat*" OR "sizenidae" OR "salamand*" OR "hypobiid*" OR "sizenidae" OR "sizenidae" OR "sizenidae" OR "hop "hippidae" OR "hepelodat*" OR "hippidae" OR "hippidae" OR "hypobiid*" OR "sizenidae" OR "amphibia*" OR "leicophry*" OR "pelodyt*" OR "hippidae" OR "heleophry*" OR "calyptocephalell*" OR "hypobiry*" OR "heleophry*" OR "calyptocephalell*" OR "hypobiry*" OR "hemperature*" OR "hepelodat*" OR "hepelodat*" OR "hepelodat*" OR "hepelodat*" OR "hepelodat*" OR "hepelodat*" OR "heleophry*" OR "hemiphract*" OR "hyla*" OR "heleophry*" OR "calyptocephalell*" OR "hyla*" OR "heleophry*" OR "hemiphract*" OR "hyla*" OR "heleophry*" OR "hemiphract*" OR "hyla*" OR "heleophry*" OR "hemiphract*" OR "hyla*" OR "hyla*" OR "heleophry*" OR "hemiphract*" OR "hyla*" OR "hyla*" OR "hemiphract*" OR "hyla

	"dendrobat*" OR "ceuthomanti*" OR "eleutherodactyl*" OR "brachycephalidae" OR "craugastor*" OR "strabomantidae" OR "pristimantis" OR "nasikabatrach*" OR "soogloss*" OR "microhyl*" OR "arthroleptid*" OR "hyperol*" OR "brevicipitidae" OR "hemisus" OR "odontobatrach*" OR "phrynobatrach*" OR "ptychaden*" OR "conraua" OR "petropedet*" OR "pyxicephal*" OR "micrixalus" OR "nyctibatrach*" OR "ranixalidae" OR "ceratobatrach*" OR "dicroglossidae" OR "rana" OR "ranidae" OR "rhacophor*" OR "mantellidae") NOT PY=(2022)
Lens	("temperature*" OR "thermal" OR "cold*" OR "cool*") AND ("cold tolerance*" OR "tolerance* to cold" OR "thermal min*" OR "CTmin" OR "CT min" OR "cold stupor" OR "cold stress tolerance*" OR "tolerance to cold stress" OR "supercooling point" OR "SCP" OR "crystal* temperature*" OR "cold hardiness" OR "freez* tolerance" OR "tolerance to freezing" OR "preferred temperature*" OR "preferred body temperature*" OR "temperature preference*" OR "selected temperature*" OR "selected body temperature*" OR "thermal prefer*" OR "temperature* prefer*" OR "temperature* select*" OR "thermal selection") AND ("amphibia*" OR "frog*" OR "toad*" OR "salamand*" OR "newt' OR "newts" OR "tadpole*" OR "metamorph" OR "metamorphs" OR "caecili*" OR "rhinatrema*" OR "tothyophi*" OR "scolecomorph*" OR "chikil*" OR "herpelidae" OR "typhlonect*" OR "indotyphild*" OR "chikil*" OR "sirenidae" OR "ambystoma*" OR "cyptobranch*" OR "proteidae" OR "thyacotriton*" OR "ambystoma*" OR "lethodon*" OR "proteidae" OR "thiadcom "ambystoma*" OR "lethodon*" OR "anura*" OR "ascaph*" OR "leiopelma*" OR "bombina*" OR "lyte* OR "hinophry*" OR "pelobat*" OR "heleophryn*" OR "lyte* OR "hinophry*" OR "pelobat*" OR "heleophryn*" OR "hemiphract*" OR "hylabetrach*" OR "heleophryn*" OR "hemiphract*" OR "hylabetrach*" OR "heleophryn*" OR "cuthomanti." OR "allophryn* OR "centroler*" OR "lentobat*" OR "asogloss*" OR "bodontophryn*" OR "ghrynobatrach*" OR "bombina*" OR "strabomantidae" OR "pristimantis" OR "anskabatrach*" OR "sogloss*" OR "microhyl*" OR "arthroleptid*" OR "hylidae" OR "brevicipitidae" OR "hemisus" OR "doantophryn*" OR "ghrynobatrach*" OR "dendrobat*" OR "caugastor*" OR "hemiphract*" OR "phynobatrach*" OR "anaskabatrach*" OR "sogloss*" OR "microhyl*" OR "arthroleptid*" OR "hylidae" OR "brevicipitidae" OR "hemisus" OR "doantobatrach*" OR "phynobatrach*" OR "dendrobat*" OR "caugastor*" OR "hemiphract*" OR "phynobatrach*" OR "dendrobat*" OR "caugastor*" OR "hemiphract*" OR "hyle* OR "hylidae" OR "brevicipitidae" OR "hemisus" OR "dotontobatrach*" OR "phynobatrach*
Proquest (Dissertation and Theses)	(noft(cold tolerance*) OR noft(CTmin*) OR noft(preferred temperature*) OR noft(selected temperature*)) AND (noft(amphibia*) OR noft(frog*) OR noft(toad*) OR noft(anura*) OR noft(tadpole*) OR noft(salamand*) OR noft(newts))
Google Scholar (French)	("température préférée" OR "température sélectionnée" OR "température choisie" OR "préférences thermiques") AND (amphibiens OR grenouille OR crapaud OR salamandres OR triton OR têtards OR Amphibia OR Caudata OR Anura OR batracien OR anoure)
	CTmin AND ("amphibiens" OR grenouille OR crapaud OR "salamandres" OR triton OR têtards OR batracien OR anoure)
	(Tpref OR Tsel) AND ("amphibiens" OR grenouille OR crapaud OR "salamandres" OR triton OR têtards OR batracien OR anoure)

Google Scholar (Japanese)	 (好適温度 OR 選択温度 OR 温度嗜好性) AND (両生類 OR カエル OR ヒキガ エル OR サンショウウオ OR イモリ OR おたまじゃくしOR "Amphibia" OR "Caudata" OR "Anura") CTmin AND (両生類 OR カエル OR ヒキガエル OR サンショウウオ OR イモ リ OR オタマジャクシ) (Tpref OR Tsel) AND (両生類 OR カエル OR ヒキガエル OR ヒキガエル OR サンショウウオ OR イモリ OR オタマジャクシ)
Google Scholar (Portuguese)	 ("temperatura preferida" OR "temperatura selecionada" OR "preferência termal") AND (anfibio OR "rã" OR sapos OR salamandra OR tritão OR girino OR Amphibia OR Caudata OR Anura OR anuros) CTmin AND (anfibio OR "rã" OR sapos OR salamandra OR tritão OR girino) (Tpref OR Tsel) AND (anfibio OR "rã" OR sapos OR salamandra OR tritão OR girino)
Google Scholar (simplified Chinese)	 (合适温度 OR 选温度 OR 耐热程度) AND (两栖动物 OR 青蛙 OR 蛤蟆 OR 蝾 螈 OR 蝌蚪 OR 小鲵 OR 大鲵 OR Amphibia OR Caudata OR Anura) CTmin AND (两栖动物 OR 青蛙 OR 蛤蟆 OR 蝾螈 OR 蝌蚪 OR 小鲵 OR 大鲵) (Tpref OR Tsel) AND (两栖动物 OR 青蛙 OR 蛤蟆 OR 蝾螈 OR 蝌蚪 OR 小鲵 OR 大鲵)
Google Scholar (traditional Chinese)	 (偏好溫度 OR 溫度選擇OR 熱偏好) AND (兩棲類 OR 蛙青蛙 OR 蟾蟾蜍癩蝦 蟆 OR 螈蠑螈OR 蝌蚪 OR 鯢小鯢山椒魚 OR 鯢大鯢娃娃魚OR Amphibia OR Caudata OR Anura) CTmin AND (兩棲類 OR 蛙青蛙 OR 蟾蟾蜍癩蝦蟆 OR 螈蠑螈OR 蝌蚪 OR 鯢 小鯢山椒魚 OR 鯢大鯢娃娃魚) (Tpref OR Tsel) AND (兩棲類 OR 蛙青蛙 OR 蟾蟾蜍癩蝦蟆 OR 螈蠑螈OR 蝌 蚪 OR 鯢小鯢山椒魚 OR 鯢大鯢娃娃魚)
Google Scholar (Spanish)	 ("temperatura preferida" OR "temperatura seleccionada" OR "preferencias térmicas") AND (anfibio OR rana OR sapo OR salamandra OR tritón OR renacuajo OR Amphibia OR Caudata OR Anura OR anuros) CTmin AND (anfibio OR rana OR sapo OR salamandra OR tritón OR renacuajo or Anuros) (Tpref OR Tsel) AND (anfibio OR rana OR sapo OR salamandra OR tritón OR renacuajo or Anuros)

45 Table S2: Metadata.

Data	Description
unique_ID	Unique identifier for each row in the data.
name	Name of the researcher who performed the data extraction.
ref	Abbreviated reference for the study.
title	Title of the paper or thesis.
pub_year	Publication year of the paper or thesis.
thesis_chapter	If the study is a thesis, the chapter the data is taken from (e.g., 2).
chapter_title	The title of the thesis chapter the data is taken from.
peer-reviewed	Whether the study was peer-reviewed or not (i.e., thesis). Factor with
	two levels: "peer-reviewed", "not_peer-reviewed".
doi	DOI of the paper.
language	Language of the paper (main text). Factor with seven levels:
	"English", "traditional Chinese", "simplified Chinese", "French",
	"Japanese", "Portuguese", "Spanish".
population_ID	Unique identifier for each population. Note that populations were
	considered individuals of the same species taken from different
	geographical locations. For studies without geographical coordinates,
	populations were assigned based on descriptions made by the authors
	(e.g., "Northern population" vs. "Southern population").
cohort_ID	Unique identifier for each cohort. By "cohort", we refer to
	independent groups of animals. In some cases, traits were measured
	multiple times on the same cohort of animals (e.g., using different
	endpoints, or at different life stages). As such, the same cohort_ID
	was assigned to repeated measures. Note that cohort_ID was assigned
	at the trait-level; as it was not always possible to assign whether
	multiple traits (e.g., CTmin and CTmax) were measured with the
	same, or independent groups of animals.
notes_ID	General notes related to population_ID and cohort_ID.
order	Species order, according to Jetz and Pyron (2018).
family	Species family, according to Jetz and Pyron (2018).
species	Species name, according to Jetz and Pyron (2018).
strain	The strain, variety, subspecies, or morph of the species, as reported in
	the study.
IUCN_status	International Union for the Conservation of Nature (IUCN) threat
	status. Factor with 7 levels: "DD", "LC", "NT", "VU", "EN", "CR"
	and "EX", for "data-deficient", "least-concern", "near threatened",

	"vulnerable", "endangered", "critically endangered" and "extinct",
	respectively.
origin	Origin of studied animals. Factor with four levels: recently collected
	from the wild (i.e., "wild"), eggs laid in the laboratory (i.e., "lab"),
	animals provided from a supplier (i.e., "supplier") or "unclear". For
	studies collecting eggs from the wild and testing the same generation
	of animals, animals were considered as "wild".
n_generations_lab	Number of generations spent in the laboratory, if reported in the
	study.
latitude	Latitude from which animals were collected (decimal degrees).
	Latitudes presented in degrees/minutes/seconds were converted to
	decimal degrees. When geographical coordinates were not presented,
	the coordinates were estimated using Google Maps.
longitude	Longitude from which animals were collected (decimal degrees).
	Longitudes presented in degrees/minutes/seconds were converted to
	decimal degrees. When geographical coordinates were not presented,
	the coordinates were estimated using Google Maps.
elevation	Elevation from which animals were collected (meters above sea
	level), as reported in the study. When not reported, elevation was
	estimated using latitude and longitude and freemaptools.com.
date_sampling	Date at which the animals were sampled (format
	YEAR/MONTH/DAY, e.g., "2020/07/26").
month_sampling	Month from which the animals were collected.
year_sampling	Year from which the animals were collected.
start_range_sampling_dates	The beginning of the range of dates over which animals were
	collected. Indicated are both the month and the year of collection
	(e.g., "January_2015").
end_range_sampling_dates	The end of the range of dates over which animals were collected.
	Indicated are both the month and the year of collection (e.g.,
	"September_2015").
notes_sampling	General notes regarding the sampling of the animals.
ambient_temp	For animals recently sampled from the wild (eggs not laid in the
	laboratory), the mean ambient temperature (°C) in the month of
	collection, if reported in the study. If animals were collected over a
	range of months, the mean temperature across this sampling period
	was reported.
substrate_temp	For animals recently sampled from the wild (eggs not laid in the
	laboratory), the mean temperature of the substrate (°C) in the month
	of capture. If animals were collected over a range of months, the
	mean temperature across this sampling period was reported.

water_temp	For animals recently sampled from the wild (eggs not laid in the
	laboratory), the mean water temperature (°C) in the month of
	collection. If animals were collected over a range of months, the mean
	temperature across this sampling period was reported.
field_body_temp	For animals recently sampled from the wild (eggs not laid in the
	laboratory), the mean body temperature (°C) measured in the field
	when animals were collected. If animals were collected over a range
	of months, the mean temperature across this sampling period was
	reported.
notes_env_temp	General notes regarding the sampling of animals in the field.
acclimated	Whether the animals were maintained in the laboratory for >12h or
	tested shortly after collection. Factor with two levels: "acclimated" or
	"field-fresh".
incubation_temp	For animals born in the laboratory, the mean temperature (°C) at
	which the embryos were incubated.
sd_incubation_temp	Variability (standard deviation) in incubation_temp (°C).
life_stage_acclimated	For acclimated animals, the life stage acclimated prior to the upper
	thermal limit assessment. Factor with five levels:
	"embryos_and_larvae", "larvae", "juveniles", "metamorphs" or
	"adults". Larval stages of salamanders and tadpoles were referred to
	as "larvae". Animals between Gosner stages 42 and 45 were
	considered "metamorphs", while those between Gosner stage 45 and
	sexual maturity were considered "juveniles".
gosner_acclimated	For acclimated animals, the Gosner stage when the acclimation
	started, if reported in the study.
acclimation_temp	For acclimated animals, the mean temperature of acclimation (°C).
	Note that "acclimation" refers to a prolonged (>12h) exposure to a
	new temperature. Therefore, cold/heat shocks or housing conditions
	just prior to assessing thermal tolerance or preference (e.g., 2 hours at
	25°C) were not considered as "acclimation" conditions. If animals
	were exposed to multiple acclimation conditions (e.g., 15°C for 1
	month, and then re-acclimated to 25°C for 7 days), we took the latest
	acclimation condition as the "acclimation_temp".
sd_acclimation_temp	Variability (standard deviation) in acclimation_temp (°C).
acclimation_time	The duration of acclimation (days).
notes_acclimation	General notes regarding the laboratory acclimation of animals.
life_stage_tested	The life stage tested for thermal tolerance or preference. Factor with
	four levels: "larvae", "metamorphs", "juveniles" or "adults". Larval
	stages of salamanders and tadpoles were referred to as "larvae".
	Animals between Gosner stages 42 and 45 were considered

	"metamorphs", while those between Gosner stage 45 and sexual
	maturity were considered "juveniles".
gosner_tested	Gosner stage when the animals were assessed for thermal tolerance or
	preference.
SVL	Mean snout-vent length of the animals (mm) when assessed for
	thermal tolerance or preference, if reported in the study. Note that
	SVL data was often taken from Rohr et al. (2018).
body_mass	Mean body mass of the animals (g) when assessed for thermal
	tolerance or preference.
age_tested	The age (days-post-hatching) at which the animals were tested for
	thermal tolerance or preference.
sex	The sex of the animals. Factor with four levels: "male", "female",
	"mixed", "unknown". The "mixed" category was used when authors
	clearly stipulate that they mixed males and females.
metric	The metric used to assess thermal tolerance (CTmax, LT50_hot,
	CTmin, LT50_cold) or preference (Tpref). Factor with two levels:
	"CTmax", "LT50_hot", "CTmin", "LT50_cold", "Tpref".
endpoint	The endpoint that was used for assessing thermal tolerance (loss or
	righting response, loss of equilibrium, onset of spasms, no response to
	prodding, supercooling point, death). Factor with seven levels:
	"LRR", "LOE", "OS", "prodding", "SCP", "death", "other". If
	"other", details are reported in "notes_test" (see below).
medium_test_temp	Whether the temperature measured during the test was the ambient,
	the water, the substrate, or the body temperature. Factor with three
	levels: "ambient", "substrate", "water", "body".
start_temp	If the metric was CTmax, the starting temperature used in the upper
	thermal limit assay (°C).
ramping	If the metric was CTmax, the ramping (heating) rate applied to the
	animals (°C/min).
set_time	If the metric was LT50, the time the animals spent at the test
	temperature (the time after which the animals the survival was
	assessed, in hours). If the authors report e.g., 96h-LT50, then set_time
	would be 96.
n_test_temp	If the metric was LT50, the number of temperatures tested to assess
	upper thermal limits. E.g., if authors measured survival at 36, 38, 39,
	and 41° C, n_test_temp = 4.
n_replicates_per_temp	If the metric was LT50, the number of replicates used at each test
	temperatures. E.g., if authors used 5 test temperatures and measured
	the survival of three independent cohorts of animals at each test
	temperature, then n_replicates_per_temp = 3 .

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preference	

n_animals_per_replicate	If the metric was LT50, the number of animals in each replicate.
duration_measurement	If the metric was Tpref, the duration of the assay to measure thermal
	preference (hours).
rate_measurement	If the metric was Tpref, the rate at which body temperature was
	measured (measurements/hour).
gradient_type	If the metric was Tpref, the type of thermal gradient used. Factor with
	two levels: "linear", "shuttlebox".
gradient_low_temp	Lowest temperature in the thermal gradient (°C).
gradient_high_temp	Highest temperature in the thermal gradient (°C).
notes_test	General notes regarding the thermal tolerance or preference assays.
humidity	Humidity at which animals were acclimated or tested (% relative
	humidity). If the humidity during the acclimation and the test were
	different, priority was given to the conditions of the test.
oxygen	Oxygen at which animals were acclimated or tested (mg.L ⁻¹ dissolved
	oxygen). If the oxygen concentration during the acclimation and the
	test were different, priority was given to the conditions of the test.
salinity	Salinity at which animals were acclimated or tested (parts per
	thousands). If the salinity during the acclimation and the test were
	different, priority was given to the conditions of the test.
pH	pH at which animals were acclimated. If the pH during the
	acclimation and the test were different, priority was given to the
	conditions of the test.
photoperiod	Photoperiod at which animals were acclimated (number of hours of
	light per day).
chemical	If any, which chemical (e.g., pollutant, toxin) was added to the
	animals' environment. If animals were in a control group (i.e., only
	supplemented with a solvent), "control" was indicated.
hormone	If any, which hormone (e.g., corticosterone, thyroid hormone) was
	added to the animals' environment. If animals were in a control group
	(i.e., only supplemented with a solvent), "control" was indicated.
concentration_chemical_hormone	If any, the concentration of the hormones or chemicals used. If
	animals were in a control group, "0" was indicated.
unit_chemical_hormone	The unit used to quantify the chemical or hormonal concentration
	administered (e.g., g/L, ng/g of sediment).
infected	Whether the animals were infected with a pathogen. Indicate
	"infected" if the animals were infected with a pathogen. Otherwise,
	leave the field blank.
pathogen	If the animals were infected with a pathogen, the name of the
	pathogen (e.g., Batrachochytrium dendrobatidis).

-	notes_supplements	General notes regarding the addition of chemicals, hormones, or
		pathogens.
	data_source	Where the upper thermal limit data is reported (main text, table,
		figure, published data).
	data_url	If the data was published in a repository, the url link to the repository
		containing the data.
	flag	Whether the study has procedural concerns (with details).
	mean_trait	Mean thermal tolerance or preference (°C).
	error_trait	Standard deviation or standard error of mean_trait (see error_type)
	n_trait	Sample size of mean_trait. When the metric was LT50, the sample
		size was taken as the number of test temperatures ("n_test_temp") *
		the number of replicates per test temperature (n_replicates_per_temp).
	error_type	Whether the error is presented as standard deviations (i.e., "sd") or
		standard errors (i.e., "se").
	notes_trait	General notes about thermal tolerance or preference estimates
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TABLE S3: Inclusion criteria used to screen abstracts, titles and keywords. Numbers match those

64 used in in Figure S1 (decision tree).

	Descr	iption
1	Studies not published in French, Japanese, Portuguese, simplified Chinese, traditional Chinese or Spanish were excluded.	
2	We on simular	ly included empirical studies presenting original data. Therefore, we excluded reviews, syntheses, tions, theoretical studies, and conference abstracts, unless supplemented with original data.
3	3 "Amphibians" refer to frogs, toads, salamanders, newts, and caecilians. We only included studies on whole organisms.	
	Desired	d measures of cold tolerance included the:
	i)	critical thermal minimum (CTmin), where animals are subject to incremental decreases in temperature until an endpoint (e.g. loss of righting response, onset of spasms, supercooling point, crystallisation) is reached;
	ii)	the temperature lethal for 50% of the animals (LT50; sometimes referred to as the "incipient lethal temperature"), where survival is recorded after animals are abruptly transferred to a set of cold temperatures for a given period of time (e.g. 24 hours) and LT50 is interpolated from the survival curve; and
4	iii)	the death time or chill coma time where animals are abruptly transferred to cold temperatures and the time needed for animals to reach an endpoint (e.g. immobilisation, death) is recorded as the response. With the latter measure, the thermal tolerance limit can be inferred from the relationship between chill coma time and the temperature of the knockdown assay. Therefore, cold knockdown times must have been measured at >2 temperatures (e.g. chill coma times at 10, 15, and 17°C).
	We exe (e.g. ch critical	clude alternative measures of cold tolerance which cannot be converted to the temperature scale nill coma recovery time) or CTmin extrapolated from physiological performance curves (e.g. temperature for ATPase activity).
	Desired	d measures of preferred temperature were:
	i)	where animals are placed in a temperature gradient and the body temperature of the animals is measured at regular intervals, or inferred from photography. The mode or median body temperatures animal select is usually defined as the preferred (or selected) body temperature.
	ii)	where animals are placed in an experimental set up with levers that trigger the warming or cooling of the surface or experimental chamber (shuttlebox). Similarly to above, the body temperatures of animals is tracked, and the mode or median body temperature animals experienced is usually defined as the preferred (or selected) body temperature.
5	We focused our search on juveniles (i.e., tadpole, metamorph, froglet) or adults. Hence, we excluded studies only measuring the cold tolerance of embryos.	

70 TABLE S4: Inclusion criteria used to assess full articles for eligibility. Numbers match those used

71 in in Figure S2 (decision tree).

	Descri	ption	
1	Studies not published in French, Japanese, Portuguese, simplified Chinese, traditional Chinese or Spanish were excluded.		
2	We only included empirical studies presenting original data. Therefore, we excluded reviews, syntheses, simulations, theoretical studies, and conference abstracts, unless supplemented with original data.		
3	Amphibians" refer to frogs, toads, salamanders, newts and caecilians. We only included studies on juveniles (i.e. tadpole, metamorph, froglet) or adults. Hence, we exclude studies only measuring the cold tolerance of embryos.		
	Desired measures of cold tolerance included the:		
4	iv)	critical thermal minimum (CTmin), where animals are subject to incremental decreases in temperature until an endpoint (e.g. loss of righting response, onset of spasms, supercooling point, crystallisation) is reached;	
	v)	the temperature lethal for 50% of the animals (LT50; sometimes referred to as the "incipient lethal temperature"), where survival is recorded after animals are abruptly transferred to a set of cold temperatures for a given period of time (e.g. 24 hours) and LT50 is interpolated from the survival curve; and	
	vi)	the death time or chill coma time where animals are abruptly transferred to cold temperatures and the time needed for animals to reach an endpoint (e.g. immobilisation, death) is recorded as the response. With the latter measure, the thermal tolerance limit can be inferred from the relationship between chill coma time and the temperature of the knockdown assay. Therefore, cold knockdown times must have been measured at >2 temperatures (e.g. chill coma times at 10, 15, and 17°C).	
	We exclude alternative measures of cold tolerance which cannot be converted to the temperature scale (e.g. chill coma recovery time) or CTmin extrapolated from physiological performance curves (e.g. critical temperature for ATPase activity).		
	Desired measures of preferred temperature were:		
	iii)	where animals are placed in a temperature gradient and the body temperature of the animals is measured at regular intervals, or inferred from photography. The mode or median body temperatures animal select is usually defined as the preferred (or selected) body temperature.	
	iv)	where animals are placed in an experimental set up with levers that trigger the warming or cooling of the surface or experimental chamber (shuttlebox). Similarly to above, the body temperatures of animals is tracked, and the mode or median body temperature animals experienced is usually defined as the preferred (or selected) body temperature.	
5	To be included, the study must have reported the temperature at which animals were maintained in the laboratory (i.e. temperature of acclimation), the temperature of the environment from which animals were captured (i.e. temperature of acclimatization), or the geographical coordinates and dates of capture.		

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83 TABLE S5: Summary of procedural concerns found in some studies. Note that estimates having

⁸⁴ procedural concerns were excluded during the data curation (see main text).

Procedural concerns	Number of estimates concerned
Data from a single individual	70
Uncommon or inconsistent methodology	55
Unclear/uncommon acclimation conditions	53
Animals were starved prior to testing	34
Animals underwent surgery or amputation	16
Highly uncertain estimates	14
Animals were dehydrated prior to testing	14
Animals were exposed to hypoxic or hypercapnic conditions	13
Animals were exposed to high levels to UV radiation	5
Animals were perfused with pH solution	3
Statistical dispersion and sample sizes not reported	3
Animals were exposed to predators	2

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