AmphiTherm: a comprehensive database of amphibian thermal tolerance and preference

Authors

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Abstract

Thermal traits are crucial to our understanding of the ecology and physiology of ectothermic animals. While rising global temperatures have increasingly pushed research towards the study of upper thermal limits, lower thermal limits and thermal preferences are essential for defining the thermal niche of 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 contains 4,899 thermal trait estimates from a diverse sample of 659 species (~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 Australia. By providing a more holistic understanding of amphibian thermal tolerance and preferences, *AmphiTherm* is a valuable resource for advancing research in evolutionary biology, ecophysiology, and biogeography of amphibians, offering insights that are increasingly needed in changing climates.

Background & Summary

Thermal trait data are crucial to our understanding of the biology and physiology of ectotherms. The recent increase in broad-scale syntheses of ectotherm thermal physiologies demonstrates recurring interest in how these organisms respond to changing thermal environments[1–10]. Much of this work has focused on traits relating to heat tolerance, reflecting the urgency to predict the impacts of climate warming on natural populations[11–17]. However, climate change also brings an increased probability of extreme weather events, including negative temperature anomalies[18,19]. As such, a sole focus on heat tolerance provides an incomplete picture of ectotherm responses to climate change. A comprehensive understanding of heat tolerance, cold tolerance, and thermal preference is necessary to fully define ectotherm's thermal niches and predict their responses to climate change. Below, we briefly emphasise the importance of these thermal traits for amphibians (see [8,20,21] for more in depth discussion).

While the significance of heat tolerance in predicting species' responses to warming climates is well documented[1], data on lower thermal limits are equally vital yet often understudied, especially in amphibians, an at-risk, data-deficient group of ectotherms[22]. Lower thermal limits represents the lower boundary of an organism's thermal niche and have been included in several data syntheses[5,6,8,10]. This 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]. 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 inform conservation and management strategies, for instance, by identifying microhabitats buffering the effects of extreme cold on activity and survival[24].

Preferred body temperatures reflect the temperature optimising overall performance and the 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 and activity patterns[24,27–29]. In particular, thermal preference data can be used to infer behavioural thermoregulation patterns and the microhabitats available for crucial physiological processes[30–32]. While upper thermal limits can help predict acute survival in the face of extreme heat, gradual warming below the upper thermal limit thresholds can make some areas unsuitable for amphibians' activity needs[24]. On the contrary, warming can benefit some amphibians in historically cooler climates by increasing activity windows or reducing hibernation times[33]. As such, thermal preference data can help predict the sublethal effects of climate change. Thermal preferences also, for instance, affect susceptibility to pathogens[34–36], shape the composition of commensal microbes[37,38], and mediate interactions between host and microbial communities[39].

Although investigating thermal traits separately provides important knowledge, the study of a combination of thermal traits provides deeper and more comprehensive insights. A simultaneous analysis of upper and lower thermal limits is particularly interesting as it provides an estimation of thermal tolerance breadth[21]—a measure of the thermal envelope ectotherms can occupy in the absence of other abiotic or biotic factors (e.g., competition, resource availability). When thermal preference is integrated with upper and lower thermal limits, the thermal envelope gains shape, providing additional 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 future distributions of ectothermic species, including assessing potential invasion risk[15,40–43]. Parametrising biophysical models with data on thermal limits and preference now also allow more accurate predictions of overall performance, activity windows[17,44] and microhabitat heterogeneity[17,44,45], strengthening our ability to predict the impacts of climate change on natural populations[46,47]. From an evolutionary perspective, the integration of different 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].

Thermal trait data can also be used to inform conservation efforts. Comparing thermal niche envelopes among amphibians, their microbiota, and potential pathogens can help predict changes in the microbiome and disease risk[52–57], providing key information on the environment component of the host-pathogen-environment interactions emphasized with the disease triangle concept[54]. Amphibians are often accessioned into captivity to establish assurance colonies for breeding and eventual reintroduction[58]. Knowledge of the thermal tolerance and preference of a broad range of species from different habitats can help inform the design of enclosures that better simulate natural thermal variability[59–61]. It is well established that temperature influences captive breeding success of amphibians[60]. Amphibian husbandry guides recommend setting thermal environments near the preferred temperature of a species, and when practical, providing thermal gradients and thermal cycling[60,62]. Exposure to environmental conditions via “soft release” or mesocosms prior to reintroduction can also influence survival and success of reintroduction efforts[63,64]. In addition, understanding thermal constraints on activity, demography, and disease risk enhances our ability to identify habitats suitable for repatriation and reintroduction efforts in endangered species[58,65,66].

Therefore, a holistic understanding of amphibian thermal tolerance limits and preferences is essential for defining the fundamental thermal niche of ectotherms and to project their activity, distribution and survival in rapidly changing environments. Here, we expand an existing database on amphibian upper thermal limits[1], which has already facilitated global assessments of vulnerability to climate warming[17]. We conducted systematic searches and aggregated lower thermal limits and thermal preference data from the global literature published in seven languages. In doing so, we expanded thermal niche data for 378 species, providing a stronger foundation for research on amphibian ecology, evolution, and conservation.

Methods

Reporting

We reported the contributions of each author using the CRediT (Contributor Roles Taxonomy) statement[67], and MeRIT (Method Reporting with Initials for Transparency) guidelines[68]. We also followed recommendations to maximise the indexing of titles, abstracts, and keywords in databases[69].

Literature Searches

We adapted methods from the previous version of the database on amphibian upper thermal limits[1] to search the literature on thermal physiological traits. We aimed to compile a comprehensive and representative sample of the experimental literature on lower thermal limits and thermal preference in amphibians, complementing the data on upper thermal limits compiled previously (see [1] for methods specific to upper thermal limits). PPottier accessed Scopus, ISI Web of Science (core collection), Lens, and ProQuest (dissertation & theses) on 01 November 2022 using The University of New South Wales' institutional subscriptions (full search strings are available in Supplementary Information (Table S1). Briefly, the search strings were built using as a combination of relevant terms, including "temperature" (and synonyms) and "thermal tolerance or preference" (and synonyms) and "amphibian" (and synonyms) (see Table S1 for details). 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 of amphibian thermal preference and tolerance[1,4–6,10]. This resulted in a total of 1676 unique documents. We limited the timespan of our searches to 31 May 2021 to match with the timespan represented in the upper thermal limit data[1]. This decision was made to normalise all searches to a single timespan to simplify future database updates. 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, SN, YY, and RRYO). The searches contained translated singular and plural forms of the following: "amphibian", "frog", "toad", "salamander", "newt", "tadpole", "preferred temperature", "selected temperature", "thermal preference", "Tpref", "Tsel", or "CTmin". Due to search string limitations in Google Scholar (256 characters), each term was assessed in its singular or plural form, and the search producing the largest number of results was selected. We performed searches following the format of ("preferred temperature" OR synonyms) AND (amphibian OR synonyms). We also performed separate searches with "CTmin" and ("Tpref" OR "Tsel"), as these terms are commonly used in the literature to 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 revealed that this term often returned studies that did not present experimentally-derived thermal preference values, or studies that were already captured by the other search terms. PPottier used Publish or Perish[70] to extract bibliographic records from Google Scholar. We also reused studies (designated with ** in Fig. 1) from non-English searches conducted in [1] as the key terms used successfully retrieved results on cold tolerance. However, we limited the inclusion of studies to those meeting our first two criteria (i.e., studies on amphibians, and published in the targeted language) to reduce the volume of screening. We acknowledge that our searches do not encompass all languages relevant to amphibian thermal physiology research and invite speakers of languages not represented in the current version to contribute to future updates of the database.

Eligibility criteria

We considered studies that empirically tested lower thermal limits or thermal preference in wild or laboratory amphibians. We included studies on larval, juvenile, metamorphic, and adult amphibians, and distinguished studies that acclimated both embryos and larvae to those acclimating single life stages. We excluded studies solely on embryonic stages due to the lack of comparable methods in embryos—for example, embryos are not capable of movement in a thermal gradient to assess thermal preference, and it is not possible to assess the righting response of embryos. For lower thermal limits, we included studies that measured critical thermal minimum (CTmin)[71], median lethal temperature (LT50)[72], or presented data that were convertible to these metrics (e.g. % survival of cohorts tested at different temperatures). We distinguish these metrics in the database. CTmin represents the temperature at which a specific physiological or behavioural endpoint is observed—such as the loss of righting response or a lack of response to prodding—when an organism is exposed to progressively decreasing temperatures (e.g., 1°C/min). It does not represent the lowest temperature an organism can tolerate, but rather the onset of functional stasis, the point at which the organism is unable to move and incapable of essential survival behaviours such as thermoregulation or predator evasion[71]. This distinction is important, because many ectotherms can recover from temperatures below their CTmin. For instance, some species can recover from freezing to later resume normal function[73]. In contrast, LT50 is the temperature that is lethal to 50% of animals tested and is derived through statistical interpolation from survival rates across a range of temperatures[72]. For thermal preference, we included studies that empirically tested amphibian temperature selection in a thermal gradient or shuttlebox via measures of body temperature (or inferred 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 temperatures were not standardised. We only included studies where thermal history (acclimatisation 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 (Fig. S1-2, Tables S3-4). PPottier, RRO, PPollo, ANRV, YY, SV, AVL, and NC screened articles for eligibility using Rayyan QCRI[74]. This software facilitated the identification of key terms in titles, abstracts, and keywords to streamline the screening process for large volumes of literature. During data extraction, 47 papers were ultimately excluded for either lacking extractable data or for not complying with our inclusion criteria (43 English, 1 Traditional Chinese, 3 Simplified Chinese). A total of 184 studies were deemed eligible for inclusion in the database. Of these, 157 were identified through formal database searches (comprising one study published in simplified Chinese, and another study in French), while 20 non-English studies and an additional 7 English-language studies were retrieved through Google Scholar (Fig. 1). Therefore, nearly 15% (27/184) of the included studies were retrieved through non-English literature searches. Our literature search methods and screening process is summarised in a PRISMA flowchart[75] (Fig. 1).

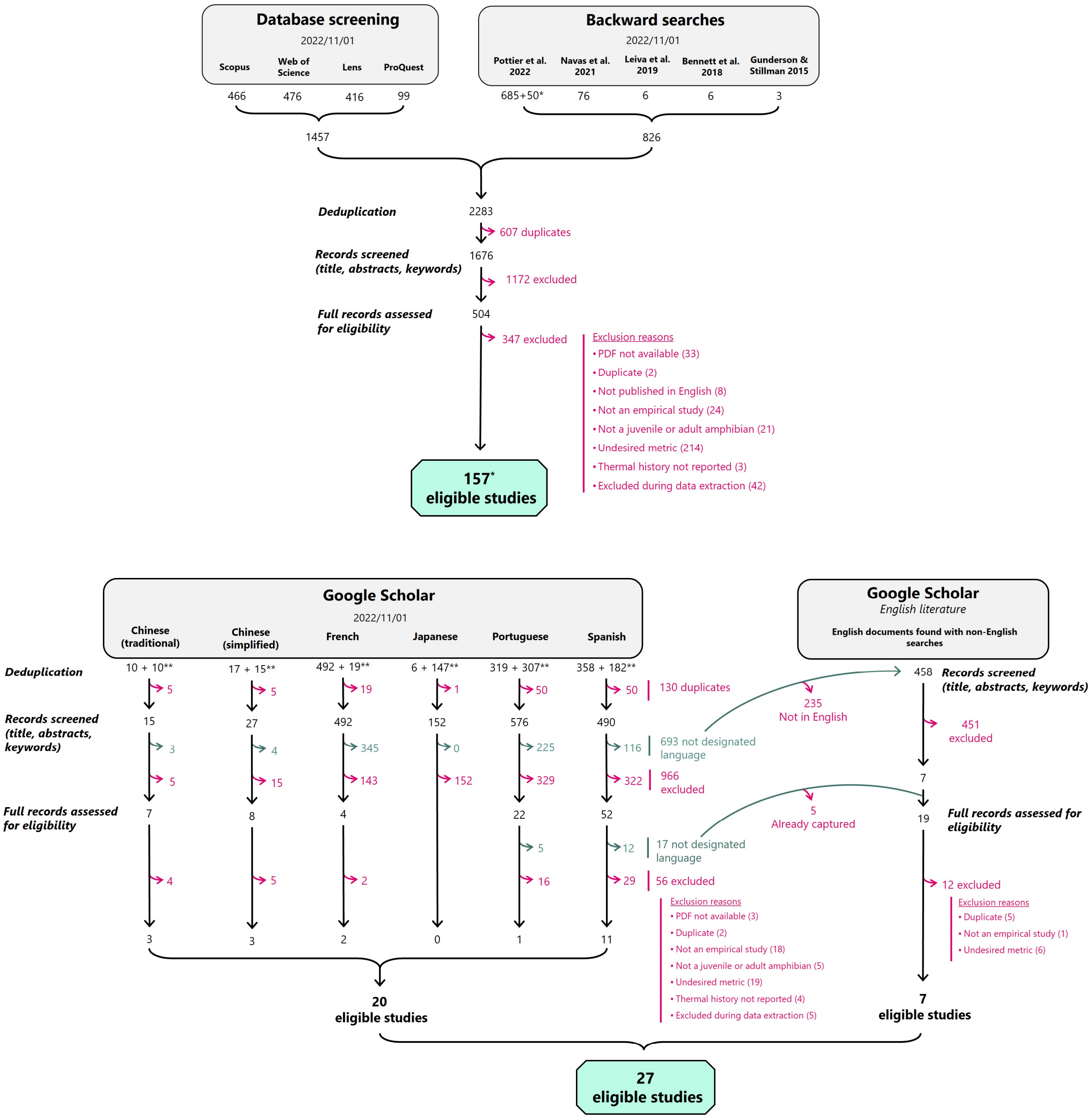

Figure 1 | PRISMA Flowchart delineating the databases used to retrieve studies on lower thermal limits and preferred body temperatures, the number of studies obtained at each stage of the screening process, and the reasons for excluding studies. * Two studies published in languages other than English (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].

Data Extraction

Data extractions were performed by PPottier (7.7% of estimates extracted), RRYO (6.7%), PPollo (5.3%), ANRV (6.2%), YY (3.0%), AVL (11.0%), SV (20.5%), and NC (45.0%). Note that these values do not add to 100% because some data entries were extracted by two authors. Data were extracted following the protocols described in the previous version of the database[1]. We extracted data directly from text and tables, and primarily used *metaDigitise*[76] (version 1.0.1) in R[77] to extract data presented in figures (although note that some authors have used WebPlotDigitizer[78] (version 4.7). When data were available in multiple formats (e.g., text and figure), we extracted it from the format with the highest resolution (e.g. data stratified by sex or location rather than aggregated across species). Where possible, we extracted measures of data 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 predicted the temperature at which 50% mortality occurred using logistic regression from the *dose.p* function from the MASS package[79].

We also extracted all additional information presented in the studies to allow investigations of the sources of variation in the data and account for non-independence. We assigned identification numbers to each study, and assigned unique identifiers within each study for each estimate, species, population (individuals of the same species sampled from the same geographical location), and cohort (independent group of individuals within a study). Additional variables included sampling coordinates, acclimation temperatures, ramping rates, life stages, endpoints used to infer cold tolerance, or the duration of exposure to experimental treatments. Additional notes were also taken by each researcher extracting the data to facilitate technical validation. The full list of variables is described in Supplementary Information (Table S2). Species names were standardised during the extraction to match AmphibiaWeb[80] and further standardised to match the most comprehensive phylogenetic tree to date[81] (see *Data Curation*).

Data Curation

To ensure consistency in data extraction across all studies, PPottier and NC extensively reviewed the extracted data to correct typological errors and resolve uncertainties identified during the extraction process. PPottier then curated the data in R[77] (version 4.4.2), merging the newly extracted data with the previously compiled dataset from [1]. This process involved standardising publication information (publication year, source name) and other variables (e.g., geographical coordinates, IUCN threat status[82]) to ensure uniformity across both datasets. PPottier also standardised species names and taxonomy with phylogenetic information from [81], which is primarily based on AmphibiaWeb[80]. The combined dataset comprises 324 publications[83–406]. Note that 53 of these publications were taken from university dissertations, and some of this work may have now been published[e.g.,407,408].

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

Data Records

AmphiTherm encompasses 4,899 thermal physiological trait estimates, derived from 324 studies and covering 659 species across 38 families across a broad geographical coverage (Fig. 2-3). This sample represents ~7.5% of all described amphibian species to date[80] (Fig. 4). According to the IUCN red list[82], most species (79.2%) are either not threatened or data-deficient (Fig. 2), yet 47 species are classified as near threatened (NC), 43 as vulnerable (VU), 29 as endangered (EN), 14 as critically endangered (CR), and one species now extinct. Considering that over 40% of amphibians are globally threatened[22], 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 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 ± 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-)analyses[409].

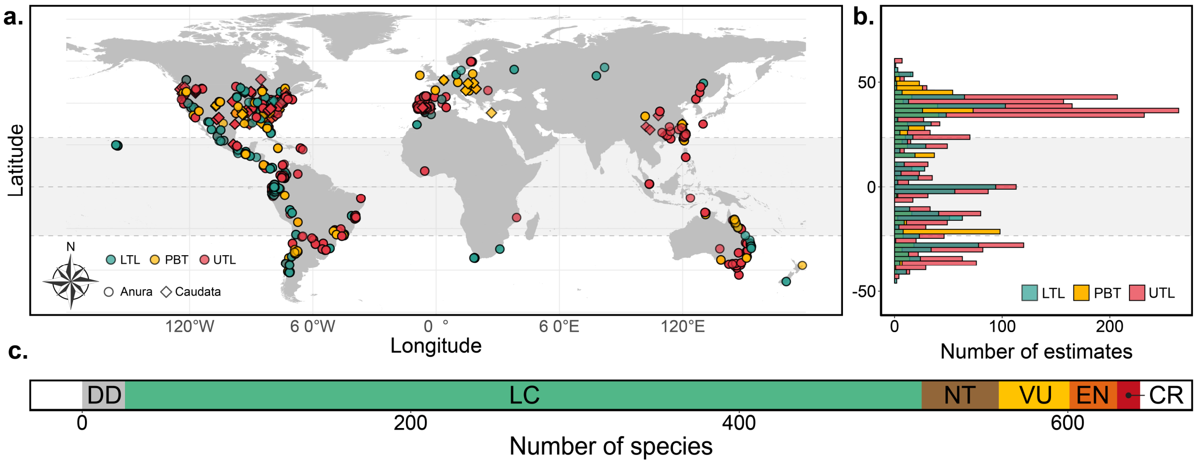

Figure 2 | Geographic distribution of thermal tolerance and preference data. a) World map showing the distribution of lower thermal limits (LTL), preferred body temperatures (PBT), and upper thermal limits (UTL) for anurans (circles) and salamanders (triangles). The shaded area represents the tropics. 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 (IUCN[82]). One species (not displayed) is now extinct.

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).

Approximately 62.7% of this database is comprised of upper thermal limit estimates (3074 estimates from 616 species and 212 studies; Fig. 3-4), highlighting a significant bias towards responses to heat extremes relative to lower thermal limits (1,009 estimates, 300 species, 88 studies) and thermal preferences (816 estimates, 137 species, 114 studies). We found that only 16 studies measured all three thermal traits, covering 59 species (~9% of the species in the dataset; Fig. 3-4). Upper and lower thermal limits were studied more frequently in tandem (60 studies), allowing to calculate the thermal tolerance breadth (i.e., difference between upper and lower thermal limits) of 276 species (~42% of the species in the dataset; Fig. 3-4).

Geographically, data were collected on all continents where amphibians occur yet exhibit a strong bias towards Nearctic and European regions (Fig. 2). Large geographic gaps in thermal data are evident across Africa, most regions of Asia, Western Australia, and central South America—regions that are biodiversity hotspots for amphibians (Fig. 2). This is particularly concerning as they constrain our understanding of how species from these underrepresented yet extremely diverse regions[410] might respond to climate change. We also identified taxonomic gaps in existing sampling where an entire order of amphibians, Gymnophiona, remained unrepresented in the database (Fig. 4). In addition, 1 of

10 families of Caudata and 7 of 36 families of Anura lack thermal limits or preferred body temperature estimates (Fig. 4). This suggests that further efforts are needed to broaden the research scope and better represent the thermal niche of amphibians.

We found that the majority (88.7%) of the literature on amphibian thermal physiological traits 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 data. Notably, this includes 289 estimates from publications in traditional Chinese (23 species, 7 studies), 131 estimates from Spanish (40 species, 11 studies), 82 estimates from simplified Chinese (12 species, 10 studies), 28 estimates from Portuguese (10 species, 3 studies), and 26 estimates from French publications (4 species, 3 studies). Including more languages, such as Afrikaans, Arabic, Bengali, Dutch, German, Hindu-Urdu, Korean, Russian, or Swahili in the screening process may help fill some gaps in future updates to the database[411]. Given the historical bias of higher impact publishing outlets against studies on herpetofauna[412], there are likely a number of studies in non-indexed journals or regional journals in local languages that were not retrieved using our methods.

The *AmphiTherm* database is stored at https://github.com/p-pottier/AmphiTherm, and regularly archived to Zenodo[413]. The repository is organised into four main folders: “data”, “R”, “references”, and “shiny_app”. These contain the metadata (.xlsx), raw, cleaned, and curated data (.csv), code for data curation and for producing the figures (.Rmd), code for deploying the Shiny web application (.R), supplementary data (.csv) and phylogenetic tree (.tre) for producing the figures, and bibliographic files (.ris and .bib) with all the references in the database. The metadata file describes all columns (n = 84) of the *AmphiTherm* database in detail. We have also launched a Shiny web application to help navigate and visualise the database. The Shiny app is available at https://p-pottier.shinyapps.io/AmphiTherm-Explorer/.

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 copyright regulations.

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.

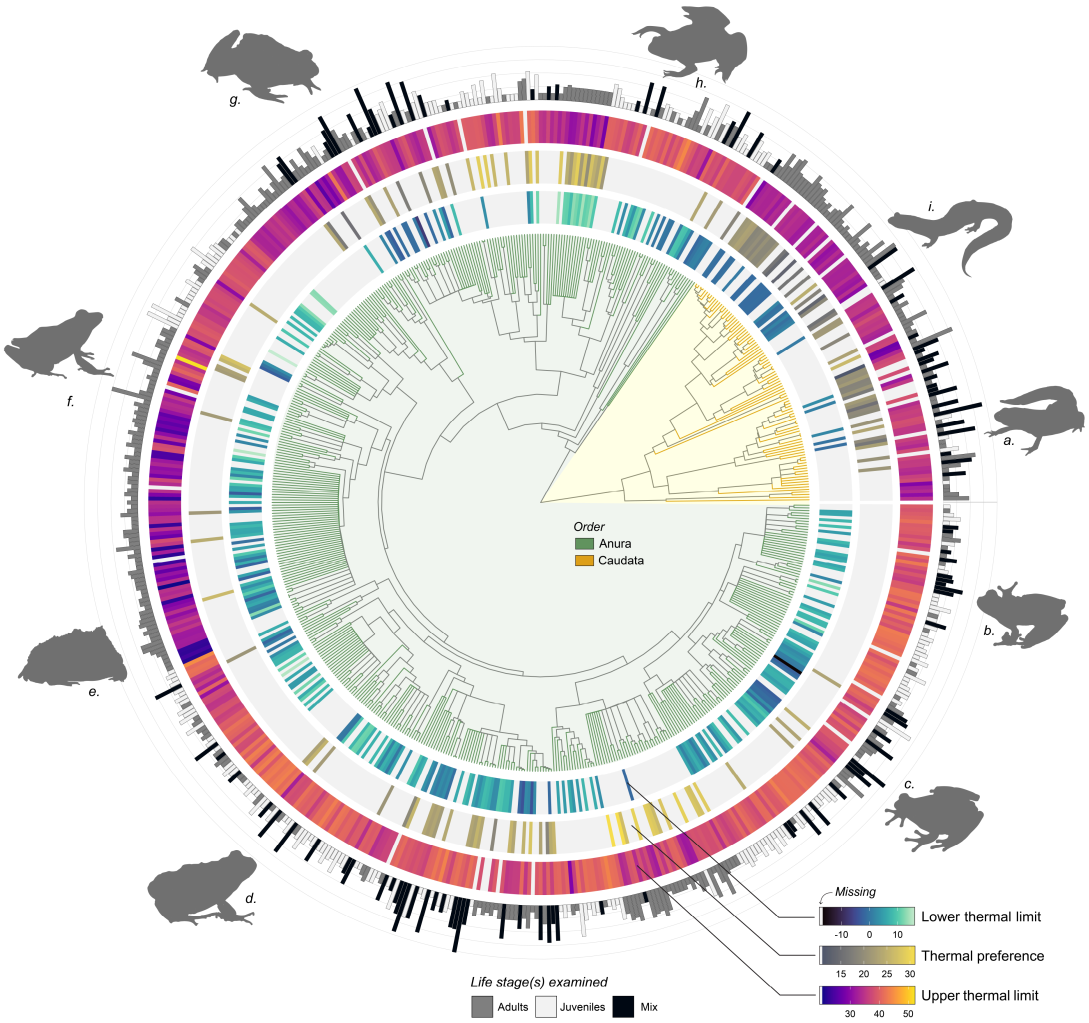

Figure 3 | Distribution of mean estimates for three key thermal traits: lower thermal limits (inner heat map), thermal preference (central heat map), and upper thermal limits (outer heat map). The number of estimates for each species is displayed as histograms, scaled on a log2(x+1) axis for clarity. The histograms are colour-coded according to the life stage assessed in the experiments, with the category "juveniles" comprising larvae, metamorphic, and juvenile stages. Gray colour represents missing data. The phylogeny relationships are based on the consensus tree from [81]. a. *Notophthalmus viridescens*, b. *Dendropsophus ebraccatus*, c. *Hyla cinerea*, d. *Pleurodema thaul*, e. *Ceratophrys cranwelli*, f. *Craugastor longirostris*, g. *Rana pipiens*, h. *Xenopus laevis*, i. *Plethodon cylindraceus*.

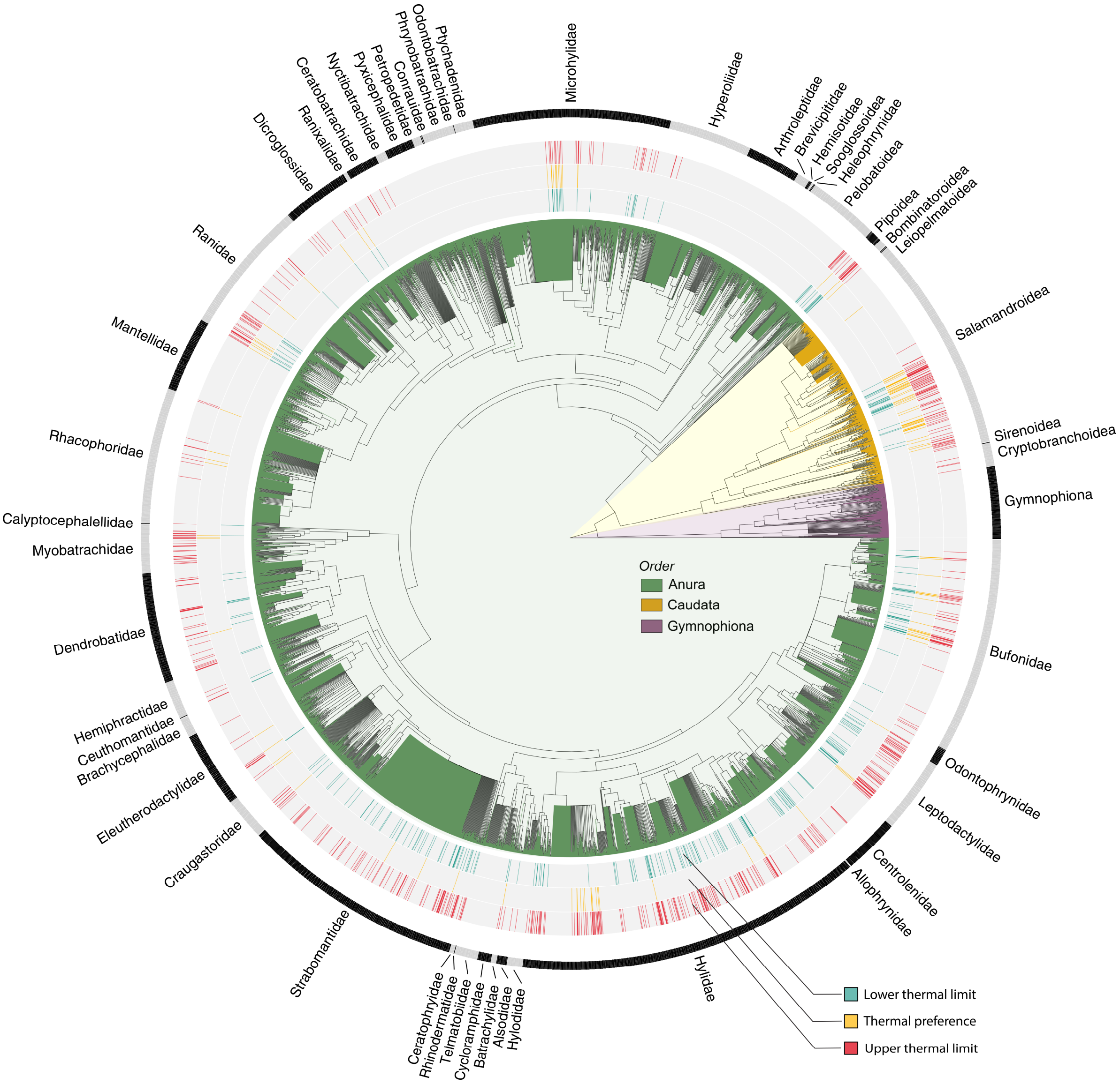

Figure 4 | Distribution of thermal trait estimates across the phylogeny of most extant amphibians. Thermal limits and preferences are mapped onto a comprehensive phylogeny of 7,238 species (consensus tree, cf.[81]) to identify taxonomic biases in existing knowledge. The outer circle presents family names, adapted from [81].

Technical Validation

We employed a transparent and reproducible workflow to systematically review over 4,000 studies from five databases and across seven languages. The potential limitations of this database are similar to those described in [1]. First, the methods used for indexing and retrieving studies in Google Scholar are not publicly disclosed[414], which may undermine reproducibility. However, given the limited coverage of 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 across multiple non-English languages at present[415,416]. Second, different authors extracted data from the original studies, introducing the possibility of individual errors. To ensure consistency and accuracy, all extracted data were subsequently cross-checked and standardised by NC and PPottier (see *Data Curation*) to minimise the risk of bias and strengthen the reliability of the dataset.

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.

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

As described in the first iteration of this database[1], the data contain inherent sources of non-independence as multiple estimates were extracted from each study, species, population (multiple sampling locations from each species), and cohort (e.g., repeated measures on the same individuals). We recommend that users use phylogenetically-informed statistical models with hierarchical random-effect structures to account for and partition sources of variation[409,417]. Users should also account for variations in sampling effort (sample size differences), for instance, by weighting estimates by the inverse of their sampling variance[409]. Employing hierarchical models that incorporate sampling variance can help address issues of biological and methodological non-independence, enabling more accurate inferences of the factors driving variation in the data[409]. Most (81%) estimates are associated with a measure of dispersion (standard deviation or standard error), species information is standardised to published phylogenetic information[81], and unique identifiers have been assigned to each study, species, population, and cohort. These features make *AmphiTherm* readily applicable for use in complex 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 this limitation. Formally assessing the extent to which geographic and taxonomic biases may influence ecological inferences is an important avenue for research.

Code Availability

The code used to process the data and produce the figures for this manuscript is available at https://github.com/p-pottier/AmphiTherm, which is archived permanently in Zenodo[413].

Author Contributions

Conceptualisation: PPottier (lead), NC, SB, TA, SMD, SN.

Methodology: PPottier (lead), NC, TA, SMD, SN

Software: PPottier

Formal Analysis: PPottier

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Data Curation: PPottier (lead), NC

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The authors declare no conflict of interest or competing interests.

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Supplementary Information

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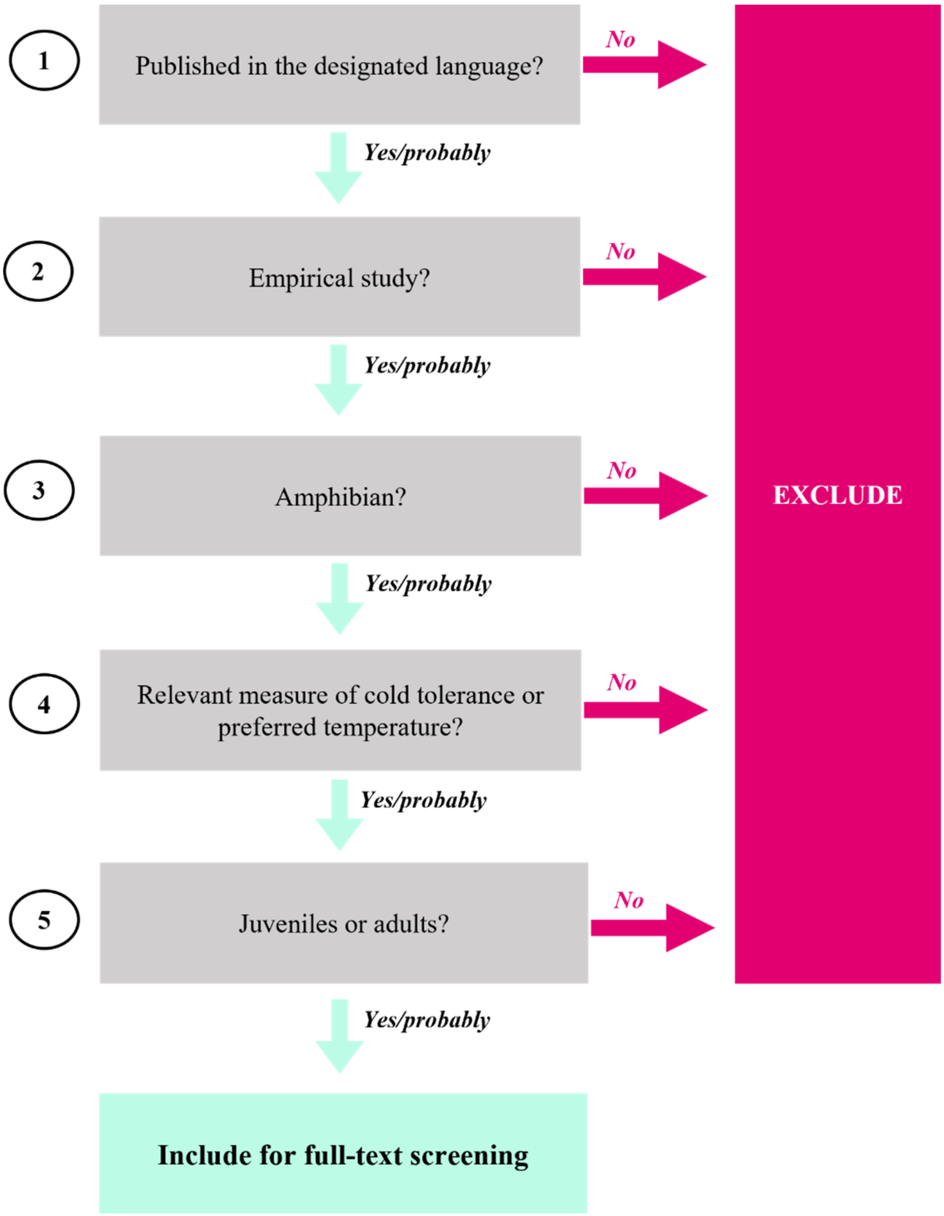

Figure S1: Decision tree used to screen titles, abstracts, and keywords. Additional details can be found in Table S3.

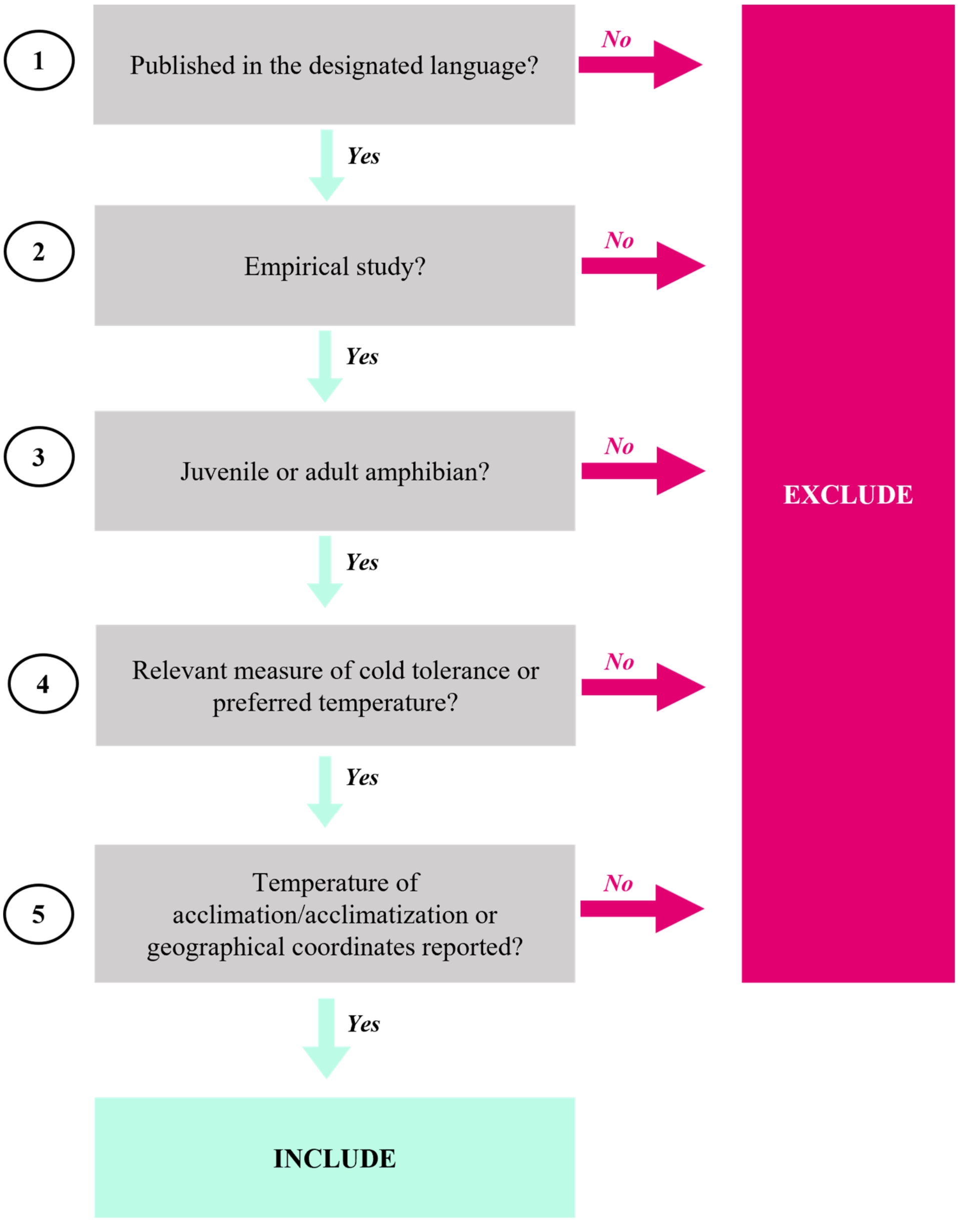

Figure S2: Decision tree used to assess full articles for eligibility. Additional details can be found in Table S4.

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 "rhinatrema*" OR "ichthyophi*" OR "scolecomorph*" OR "chikil*" OR "herpelidae" OR "typhlonect*" OR "indotyphlid*" OR "dermophi*" OR "siphonop*" OR "caudata" OR "urodela" OR "cryptobranch*" OR "hynobiid*" OR "sirenidae" OR "ambystoma*" OR "dicamptodon*" OR "proteidae" OR "rhyacotriton*" OR "amphium*" OR "plethodon*" OR "anura*" OR "ascaph*" OR "leiopelma*" OR "bombina*" OR "alyt*" OR "rhinophryn*" OR "pipidae" OR "xenopus" OR "scaphiop*" OR "pelodyt*" OR "megophry*" OR "pelobat*" OR "heleophryn*" OR "calyptocephalell*" OR "myobatrach*" OR "rhinoderma*" OR "alsod*" OR "hylod*" OR "batrachyl*" OR "cycloramph*" OR "telmatob*" OR "ceratophry*" OR "hemiphract*" OR "hyla*" OR "hylidae" OR "bufo*" OR "leptodactyl*" OR "odontophryn*" OR "allophryn*" OR "centrolen*" OR "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") AND (EXCLUDE(PUBYEAR , 2022))
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 preference*" OR "selected temperature*" OR "selected body temperature*" OR "thermal prefer*" OR "temperature* prefer*" OR "temperature* select*" OR "thermal selection") AND TS=("amphibia*" OR "frog*" OR "toad*" OR "salamand*" OR "newt" OR "newts" OR "tadpole*" OR "metamorph" OR "metamorphs" OR "caecili*" OR "rhinatrema*" OR "ichthyophi*" OR "scolecomorph*" OR "chikil*" OR "herpelidae" OR "typhlonect*" OR "indotyphlid*" OR "dermophi*" OR "siphonop*" OR "caudata" OR "urodela" OR "cryptobranch*" OR "hynobiid*" OR "sirenidae" OR "ambystoma*" OR "dicamptodon*" OR "proteidae" OR "rhyacotriton*" OR "amphium*" OR "plethodon*" OR "anura*" OR "ascaph*" OR "leiopelma*" OR "bombina*" OR "alyt*" OR "rhinophryn*" OR "pipidae" OR "xenopus" OR "scaphiop*" OR "pelodyt*" OR "megophry*" OR "pelobat*" OR "heleophryn*" OR "calyptocephalell*" OR "myobatrach*" OR "rhinoderma*" OR "alsod*" OR "hylod*" OR "batrachyl*" OR "cycloramph*" OR "telmatob*" OR "ceratophry*" OR "hemiphract*" OR "hyla*" OR "hylidae" OR "bufo*" OR "leptodactyl*" OR "odontophryn*" OR "allophryn*" OR "centrolen*" OR

	"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 "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 ("amphibia*" OR "frog*" OR "toad*" OR "salamand*" OR "newt" OR "newts" OR "tadpole*" OR "metamorph" OR "metamorphs" OR "caecili*" OR "rhinatrema*" OR "ichthyophi*" OR "scolecomorph*" OR "chikil*" OR "herpelidae" OR "typhlonect*" OR "indotyphlid*" OR "dermophi*" OR "siphonop*" OR "caudata" OR "urodela" OR "cryptobranch*" OR "hynobiid*" OR "sirenidae" OR "ambystoma*" OR "dicamptodon*" OR "proteidae" OR "rhyacotriton*" OR "amphium*" OR "plethodon*" OR "anura*" OR "ascaph*" OR "leiopelma*" OR "bombina*" OR "alyt*" OR "rhinophryn*" OR "pipidae" OR "xenopus" OR "scaphiop*" OR "pelodyt*" OR "megophry*" OR "pelobat*" OR "heleophryn*" OR "calyptocephalell*" OR "myobatrach*" OR "rhinoderma*" OR "alsod*" OR "hylod*" OR "batrachyl*" OR "cycloramph*" OR "telmatob*" OR "ceratophry*" OR "hemiphract*" OR "hyla*" OR "hylidae" OR "bufo*" OR "leptodactyl*" OR "odontophryn*" OR "allophryn*" OR "centrolen*" OR "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") Year Published = (1900 - 2021) Field of Study = (excl Botany , excl Physics , excl Geology , excl Materials science , excl Context (language use) , excl Geochemistry , excl Internal medicine , excl Mutant , excl Arabidopsis , excl Condensed matter physics , excl Computer science , excl Biophysics , excl Biotechnology , excl Computational biology , excl Horticulture , excl Aquaporin , excl Crop , excl Mineralogy , excl Nanotechnology , excl Arabidopsis thaliana , excl Chemical physics)
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 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 (anfíbio OR “rã” OR sapos OR salamandra OR tritão OR girino OR Amphibia OR Caudata OR Anura OR anuros) CTmin AND (anfíbio OR “rã” OR sapos OR salamandra OR tritão OR girino) (Tpref OR Tsel) AND (anfíbio 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 蠑螈 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 蠑螈 OR 蝌蚪 OR 小鲵 OR 大鲵) (Tpref OR Tsel) AND (两栖动物 OR 青蛙 OR 蛤蟆 OR 蠑螈 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

Table S2: Metadata table and column descriptions.

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"). Note that when multiple data points were extracted from the same population, they shared the same population_ID. The population_ID is structured as a combination of the species name and the population number (e.g., Eupemphix_nattereri_pop1).
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. For “wild” animals, this was left as NA.
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”). If the precise day is not reported, the month was reported in “month_sampling”. If a range of dates was presented (e.g., “June to August 2020”), details were reported in the “start range_sampling_dates” and “end_range_sampling_dates” columns.
month_sampling	Month from which the animals were collected. If the sampling spans two or more months, details were reported in the “start_range_sampling_dates” and “end_range_sampling_dates” columns.
year_sampling	Year from which the animals were collected. If the sampling spanned two years, details were reported in the “start_range_sampling_dates” and “end_range_sampling_dates” columns.
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, if reported in the study. For the temperature at which the larvae and adults were acclimated, see “acclimation_temp”.
sd_incubation_temp	Variability (e.g., +/- 1C) in incubation_temp (°C), if reported.
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. If a range of Gosner stage was presented (24-27), the mean of the range was taken.
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 (e.g., +/- 1C) in acclimation_temp (°C), if reported in the study.
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 heat tolerance, if reported in the study. If a range of Gosner stage was presented (24-27), the mean of the range was taken.
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 five 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 CTmin or CTmax, the starting temperature used in the thermal tolerance assay (°C).
ramping	If the metric was CTmin or CTmax, the ramping (cooling or 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.
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. Infected animals were marked as "infected". Otherwise, this field was left 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 data presents procedural concerns. The exact issue is listed in this column. Note that these observations were removed from the curated version of the database.
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

TABLE S3*: Inclusion criteria used to screen abstracts, titles and keywords.* Numbers match those used in in Figure S1 (decision tree).

	Description
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 whole organisms.
4	Desired 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 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 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: 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.

TABLE S4*:* Inclusion criteria used to assess full articles for eligibility. Numbers match those used in in Figure S2 (decision tree).

	Description
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.
4	Desired measures of cold tolerance included the: 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.

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