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Establishing allometric relationships between microsomal protein and cytochrome P450 content with body weight in vertebrate species

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ABSTRACT

Data from *in vitro* studies are routinely used to estimate *in vivo* hepatic clearance of chemicals and this information is needed to parameterise physiologically based kinetic models. Such clearance data can be obtained from laboratory experiments using liver microsomes, hepatocytes, precision-cut liver slices or recombinant enzymes. Irrespective of the selected test system, scaling factors are required to convert the *in vitro* measured intrinsic clearance to a whole liver intrinsic clearance. Scaling factors such as the hepatic microsomal protein per gram of liver and/or the amount of cytochrome P450 per hepatocyte provide a means to calculate the whole liver intrinsic clearance. Here, a database from the peer-reviewed literature has been developed and provides quantitative metrics on microsomal protein (MP) and cytochrome P450 contents in vertebrate orders namely amphibians, mammals, birds, fish and reptiles. This database allows to address allometric relationships between body weight and MP content, and body weight and cytochrome P450 content. A total of 85 and 74 vertebrate species were included to assess the relationships between log10 body weight *versus* log10 MP, and between log10 body weight and log10 cytochrome P450 content, respectively. The resulting slopes range from 0.76 to 1.45 in a range of vertebrate species. Such data-driven allometric relationships can be used to estimate the MP content necessary for *in vitro* to *in vivo* extrapolation of *in vitro* clearance data. Future work includes applications of these relationships for different vertebrate taxa using quantitative *in vitro* to *in vivo* extrapolation models coupled to physiologically based kinetic models using chemicals of relevance as case studies including pesticides, contaminants and feed additives.

1. Introduction

For xenobiotics that are not or only partly metabolised, allometric scaling provides an efficient method to quantify and predict interspecies differences in kinetic processes (Huang et al., 2015; Hunter, 2010; Poggesi, 2004). However, despite the fact that the biotransformation enzyme family cytochrome P450 (CYP450) is well conserved phylogenetically, relatively small differences in the primary amino acid sequences between CYP isoforms across species give rise to profound differences in substrate specificity and catalytic activity for xenobiotics. Thus, differences in CYP isoforms across taxa represent a major variable in inter-species differences in relation to xenobiotic metabolism and allometric scaling of CYP-mediated clearances across species remains a challenge (Martignoni et al., 2006).

Data from *in vitro* studies are now routinely used to estimate hepatic clearance of chemicals. These data can be obtained with liver microsomes, fresh or cryopreserved hepatocytes, precision-cut liver slices or recombinant enzymes. Regardless of the *in vitro* method applied to estimate clearance, scaling factors are needed to convert units of *in vitro*-derived intrinsic clearance (Clint) to estimate the whole organ intrinsic clearance (Barter et al., 2007). In humans, a combination of microsomal protein per gram liver (MPPGL) and hepatic enzyme abundance (amount of cytochrome P450 (CYP450)) is used to scale data from human liver fractions and recombinantly expressed enzyme systems, respectively (Hakooz et al., 2006; Smith et al., 2008; Wilson et al., 2003). The Clint values expressed on a per gram liver basis can then be scaled to a whole liver, based on the liver weight of the species and/or individual of interest.

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For humans and several laboratory species including the rat, hepatic cell lines and test systems based on primary cells or tissue are available, as well as established scaling factors from the peer-reviewed literature and these allow to scale *in vitro* liver Clint data to the *in vivo* situation (Chiba et al., 2009). For farm animals or wildlife, it is common to obtain microsomes from fresh liver samples to measure the Clint of chemicals, as they are generally not commercially available. Unfortunately, very often the MPPGL values are not reported in the literature, and, as indicated before, this parameter value is a critical scaling factor to extrapolate *in vitro* clearance data to an *in vivo* clearance. The *in vitro-in vivo* extrapolation method allows generation of quantitative data on metabolism of chemicals prior to studying kinetics *in vivo* and the opportunity to compare inter-species differences in metabolism, especially when such data are being applied in physiologically based kinetic (PBK) models. This is particularly valuable for further development of approaches to derive quantitative chemical-specific assessment factors for species-differences in kinetics, as alternative to the use of the proposed default uncertainty factor, allowing for a better application of inter-species differences in kinetic processes in safety assessment (Dorne, 2010; WHO, 2005). In animal-free toxicity testing strategies, the results of *in vitro* Clint and *in silico* derived parameters such as partition coefficients, are needed for the development and application of PBK models and allow the prediction of (species-specific) dose-dependent internal concentrations in a range of biological fluids and tissues (Kasteel et al., 2021; Punt et al., 2022a; Punt et al., 2022b; Punt et al., 2021).

Over the past century, hundreds of allometric relationships have been derived for physiological and ecological parameters amongst virtually all vertebrate groups. There is ample evidence that processes such as RNA and DNA levels, plant and animal metabolic rates, cancer metastasis, organ weights, and blood flow scale with body size over a range of organism sizes (Burness et al., 1999; Davies and Moyes, 2007; Edwards, 1975; Else and Hulbert, 1985; Hendriks, 1999; Hendriks, 2007; Lindstedt and Schaeffer, 2002; Martinez et al., 2006). The dependence on body size, is usually described by the equation $y = aM^b$, where "y" is the parameter of interest, "a" is a normalisation coefficient, "M" is body mass, and "b" is the scaling coefficient. Allometric scaling is an important methodology which is applied often for the prediction of the body clearance in a given species for which data are lacking while using available body clearance data from another species (Huang et al., 2015). However, while liver weight and hepatic blood flow are also well known for scaling allometrically (*e.g.*, (Boxenbaum, 1980)), hepatic metabolism markedly differs between species (Hunter and Isaza, 2008; Nebbia et al., 2001), both in phase I and phase II metabolism (Dacasto et al., 2005; Giantin et al., 2008; Jancova et al., 2010; Matal et al., 2008; Nebbia, 2001). Information on such species differences can be chemical-dependent and is often lacking, and *in vitro* clearance studies obtained with liver tissues can be performed to quantify such species differences *in vitro*. However, application of such data in PBK models is hampered by the lack of established *in vitro-in vivo* scaling factors, including MPPGL and or cytochrome P450 content. So far, allometric relationships have not been reported for liver metabolism scaling factors in a wide range of species. Given the lack of such scaling factors for different taxa, it is therefore of interest to establish such allometric relationships.

Hence, this study aimed to perform a systematic data collection on biochemical and physiological parameters namely microsomal protein (MP) content, CYP450 content, liver weight and body weight for a wide range of vertebrate species, in order to establish allometric relations between MP content, CYP450 content and body weight. Based on this data collection exercise, inter-species differences in MPPGL contents across the vertebrate class are presented and discussed.

2. Materials and methods

Literature searches were conducted in Scopus and Google Scholar until 1st of June 2022 to identify relevant peer-reviewed publications reporting microsomal protein content (mostly reported as MPPGL) and cytochrome P450 content (mostly reported in nmol/mg microsomal protein) in vertebrates (amphibians, birds, fish, mammals and reptiles) as measured experimentally by the authors. In order to derive allometric relationships between MP, CYP450 content and body weight, data collection for body weights for the different species was also required. Another parameter for which information was collected is liver weight, which ideally, should have been measured and cited in the extracted papers. When the liver weight was not provided, existing allometric relationships were used to derive this value based on the reported body weight (Crile and Quiring, 1940; Else et al., 2004; Lindstedt and Schaeffer, 2002; Withers and Hillman, 2001). Table 1 summarises the keywords applied in the respective literature searches. Data were excluded when the animals were intentionally exposed to chemicals because this may have led to an increase or reduction of the CYP450 and MP contents. Since the MP content is reported as MPPGL, this value was multiplied by the liver weight to derive the MP content. The database was then split further in two datasets by random allocation if a species was mentioned more than once: a first dataset to derive the allometric relationship and a second dataset for evaluation purposes. Both datasets as well as the associated individual references can be found in the Supplementary Information (Table S1 and S2).

The first dataset provides data on recorded body weight, liver weight, and MP or CYP450 content, which were then used to assess in Microsoft Excel, the allometric relationships (Eq. 1) between body weight and MP and body weight and CYP450 content:

$$y = a * M^b \quad (1)$$

where y is the parameter of interest, in this case either MP (g) or CYP450 content (μmol), a is the normalisation coefficient, M is body weight in kg, and b is the scaling coefficient. Finally, the coefficient of determination (r^2) of each regression line is provided and varies between 0 and 1.0 as a measure of the variance in y that is attributable to the variance in M. If r^2 equals to 1, the correlation is absolute whereas for a value of 0, there is no correlation observed. In addition, the p-values were calculated. The second dataset was used as a training set to calculate the prediction error, which was estimated as the ratio between observed and predicted values. This training set contained data for a range of species with a wide range of body weight values. To assess variability, a dataset for a wide range of vertebrate species across a wide range of body weights was included.

3. Results

The values for MPPGL levels in a wide range of vertebrate species, at least available from two different experiments, are presented in Fig. 1 for amphibians, mammals, birds and fish. All values are reported in the supporting information (Table S1). Reptiles are not shown in Fig. 1 due to the small sample size. MPPGL values for species that are mentioned only once in the database are not provided, but can be extracted from the database itself. A 2- to 4-fold variation in MPPGL was observed for most of the included species. The only species where a larger differences in MP content between 5 and 7-fold was observed, were *Cortunix japonica, Pseudopleuronectes americanus, S. gairdneri, Canis lupus familiaris,*

Table 1
Individual keywords applied to the literature searches for the data collection of microsomal protein content and cytochrome P450 content in vertebrate species.

Species	fish, amphibians, reptiles, mammals, birds, wildlife, cattle, cow, bovine, sheep, goat, horse, human, rat, mouse, rabbit, pig, swine, porcine, minipig, guinea pig, hamster, cat, dog, monkey, deer, salmon, trout, carp, flounder, scup, chicken, turkey, goose, quail, pigeon, duck, pheasant, owl, turtle, snake, frog, toad
Other	MPPGL, microsomal protein per gram liver, microsomal yield, microsomal protein liver, cytochrome P450 content, CYP450 content

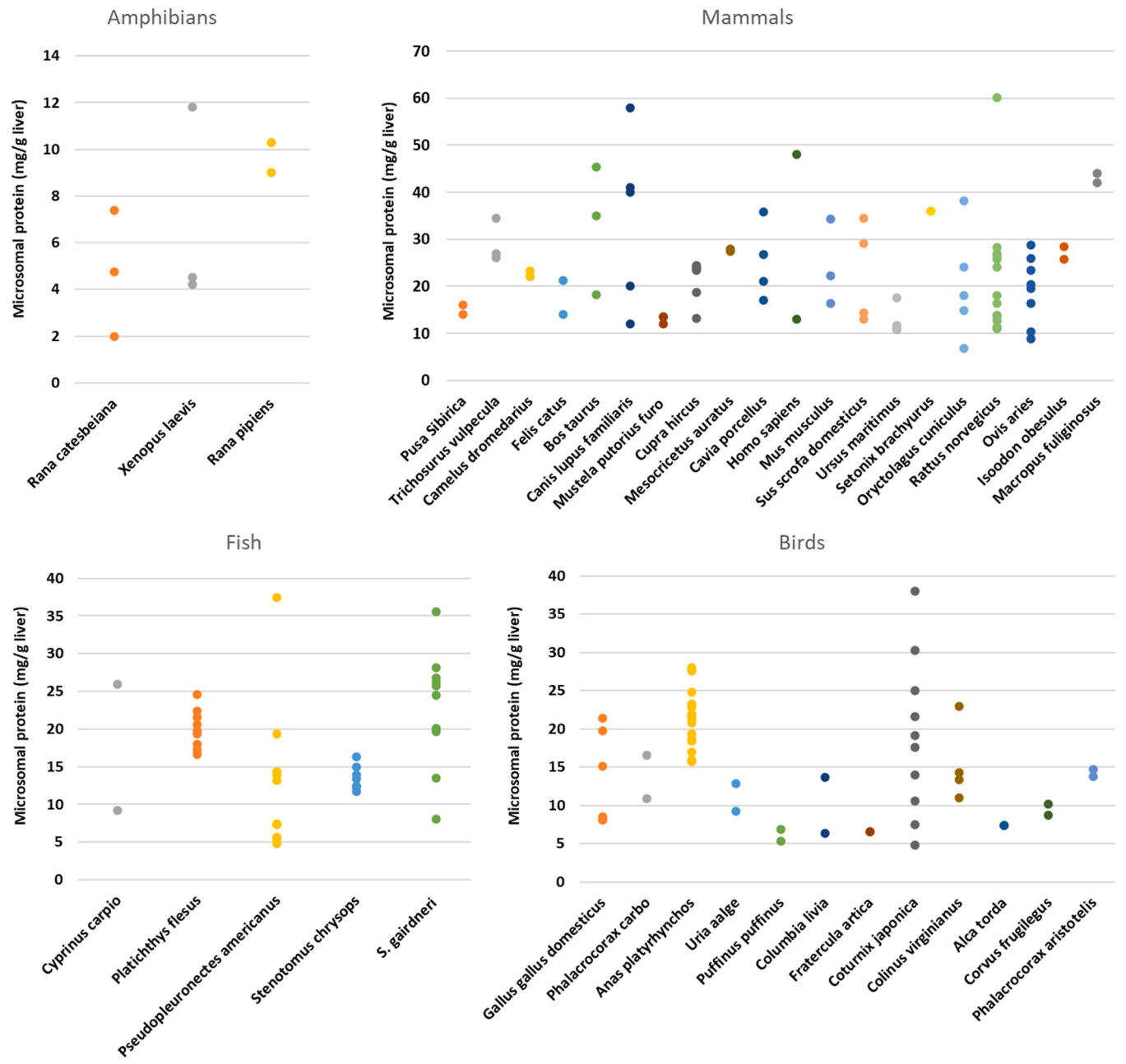

Fig. 1. Variation of microsomal protein per gram liver in various species. Only species are presented for which more than 1 value is available. The full dataset, including species for which only one value is available is presented in the Supporting information.

Oryctolagus cuniculus and Rattus norvegicus. Human studies for MPPGL values included in this database also showed differences up to 5-fold.

The complete database is provided in the Supporting information (Table S1). A total of 85 different species were included to depict the allometric relationship between body weight, expressed as log10 M (kg) and MP content, expressed as log10 MP content (g), and is shown in Fig. 2A. The r^2 is equal or above 0.75, which indicates satisfactory relationships between body weight and MP content (Table 2). An exception applies to the small number of values for reptiles (n = 3) for which only an r^2 of 0.6 was observed. Due to this sample number the slope is also not significant, in contrary to those for the other taxa (Table 2). For amphibians, the MP content scales close to 3/4 of the body weight (b=0.76, Table 2), while for mammals, birds and reptiles the MP content scales sub-linearly ($0.94 < b \geq 0.97$ Table 2). Conversely, fish scaling is super-linear (b=1.4), meaning that the MP content increases with a near 1.5-fold increase in body weight. In addition, Fig. 2B depicts the observed and predicted MP content using the obtained equations from the first dataset. Values are within a 5-fold factor for 98 % of the datapoints.

For CYP450 content, 74 different species were included to derive the allometric relationship between body weight, expressed as log10 M (kg) and CYP450 content, expressed as log10 liver CYP450 content (μmol) (Table S2). Fig. 3A shows the allometric relationships between body weight and liver CYP450 content. In mammals, birds and reptiles, liver CYP450 scales as a nearly linearly proportional to body weight ($0.91 < b < 0.97$ Table 2). For fish, a similar slope as for the MP content is observed, meaning that the liver CYP450 content increases with a near 1.5-fold increase in body weight. The same can be observed for amphibians. With the exception of fish and amphibian taxa, r^2 was higher than 0.9. All derived slopes are significant, due to the low p-value (<0.05) (Table 2). Fig. 3B depicts the observed values from the second dataset and predicted liver CYP450 content using the obtained

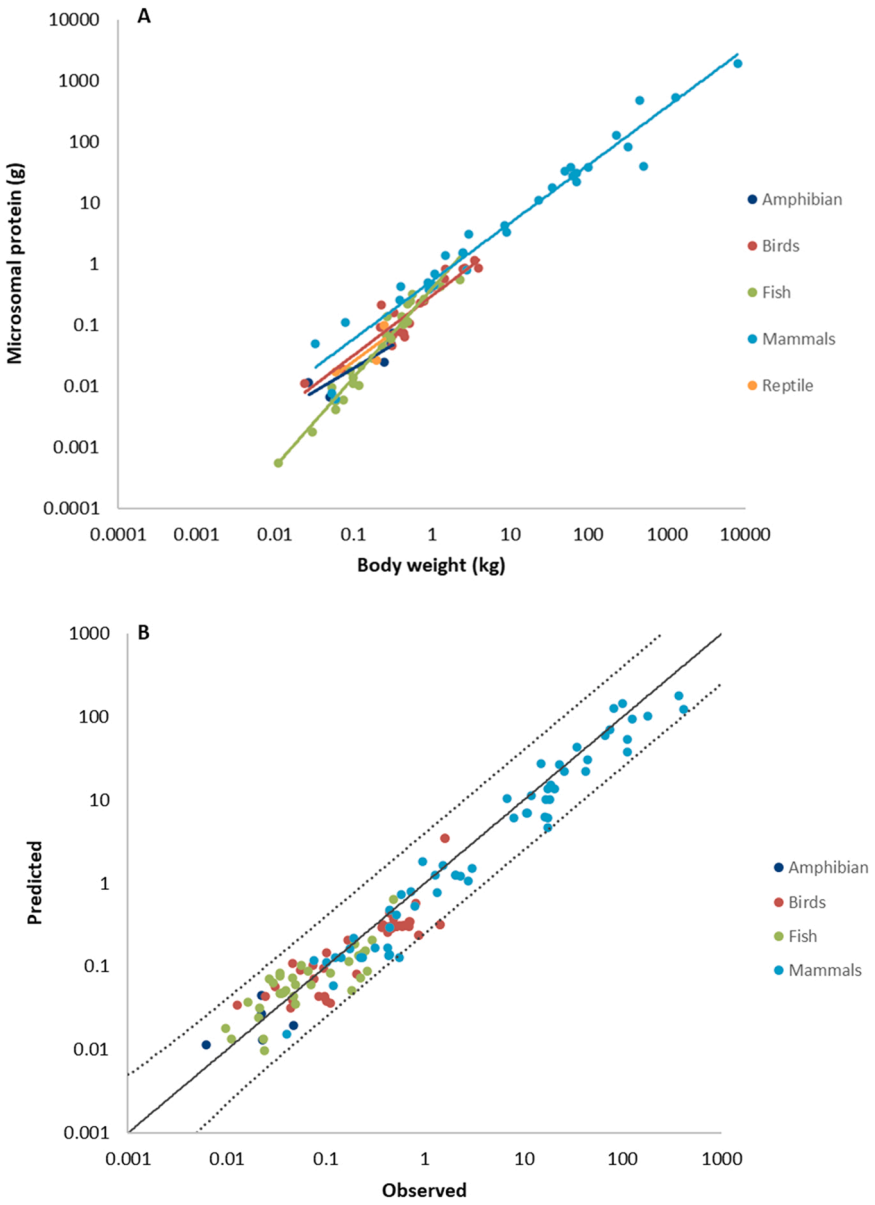

Fig. 2. A) Allometric scaling of liver microsomal protein for amphibians, bird, fish, mammalian, and reptile species. B) Measured liver microsomal protein content *versus* predicted content using regression equations.

Table 2
Linear regression equations for MP content and cytochrome P450 liver content for amphibians, mammals, birds, fish and reptiles.

	N	Intercept a	Slope b	r^2	p-value
Microsomal protein content (g)					
Fish	26	0.3885	1.4569	0.76	<0.05
Amphibians	5	0.1118	0.7615	0.77	<0.05
Reptiles	3	0.2193	0.9420	0.60	0.4
Birds	20	0.2974	0.9750	0.88	<0.05
Mammals	31	0.5210	0.9516	0.95	<0.05
Cytochrome P450 content (μmol)					
Fish	20	0.1238	1.4495	0.58	<0.05
Amphibians	5	0.1927	1.6708	0.83	<0.05
Reptiles	4	0.0558	0.9332	0.99	<0.05
Birds	18	0.0721	0.9722	0.95	<0.05
Mammals	27	0.3483	0.9109	0.97	<0.05

regression equations from the first dataset. Values were within a 5-fold factor for 94 % of the datapoints.

4. Discussion

This manuscript describes data collection for available MP and liver CYP450 content for vertebrate taxa namely amphibians, birds, fish, mammals and reptiles. In addition, an inter-species database providing MPPGL and liver CYP content and its corresponding variability, expressed as standard deviation is also presented. Subsequently, allometric relationships were derived using a set of regression equations which can be used to estimate MP and or liver CYP450 content since such parameter values represent a critical scaling factor to extrapolate *in vitro* metabolism rates to an *in vivo* clearance in vertebrate species.

Inter-species differences in MPPGL showed a maximum of 7-fold variation for a few species, while most species showed only up to 2-fold differences. For this manuscript, MPPGL content in humans was collected from two studies for which body weight of the participants was described and could subsequently be used for allometric scaling (Knaak

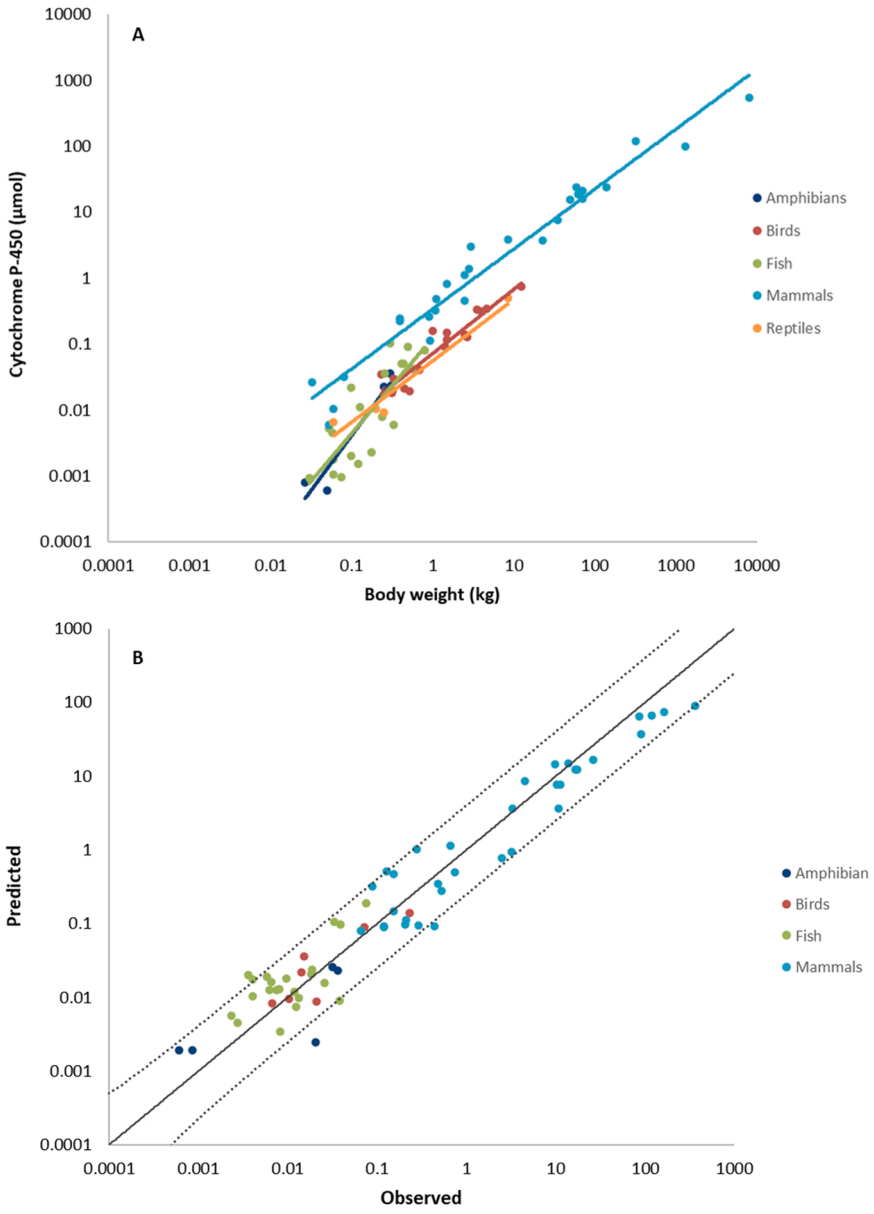

Fig. 3. A) Allometric scaling of liver CYP450 content for amphibian, bird, fish, mammalian and reptile species. B) Measured liver CY P450 content in the liver *versus* predicted content using regression equations.

et al., 1993; Lipscomb et al., 2003). However, a 19-fold variation in human MPPGL content was reported, which resulted from MPPGL levels in 128 samples, with a mean MPPGL content ± SD of 39.5 ± 21.6 mg protein/g liver (Zhang et al., 2015). Since the human MPPGL samples showed no particular characteristics, it was inferred that individuals with extreme values of MPPGL may exist within human populations (Zhang et al., 2015). Such large variation was not observed in this dataset for other species, which might be due to the lower number of available MPPGL sample values from the peer-reviewed literature. This indicates that large variations cannot be ruled out for the other species.

The high scaling coefficient in fish may reflect age differences, which was not observed in the other vertebrate orders. Liver MP and CYP450 contents in fish were obtained from very small and/or juvenile fish and only two values were reported for fully grown fish. It is possible that the CYP enzyme system is immature in those juvenile fish species which may also impact on the MP content. The higher variation in liver CYP450 content for the different fish species might be also due to environmental contamination, which could lead to CYP induction or inhibition. Both reasons can have an impact on the liver MP and CYP450 content and on the derived regression. This can be illustrated for humans in which liver metabolic activity differences between neonates, children and adults are well characterised (Anderson, 2002; Sadler et al., 2016). In this case, allometric scaling accounts for the development of body size and function but not for the fact that liver xenobiotic-metabolising enzyme activities are generally low at birth (Björkman, 2006). This reasoning may also apply to fish, but this would not explain the high variation in liver CYP450 content. Since data from intentionally exposed animals were excluded from this study due to possible CYP450 induction or inhibition can lead to higher or lower CYP and MPPGL contents, it cannot be excluded that animals are unintentionally exposed in their environment, causing variation in the CYP content. This may also play a role in the large slope (b>1) of the CYP content in amphibians and the observed variability.

Previously, scaling has been attributed to bio-physical constraints, volumes, control organs and frequencies or biological times (Temerin, 1985). It has been reported that volumes or capacities scale linearly to the body weight and the number of cells increases with an increase in body weight, while the number of (cellular) functions controlled does

not (Lindstedt and Schaeffer, 2002). A relevant example includes ATPase, which displays a scaling coefficient close to 0.75, while the mammals citrate synthase scales with the slope value (b) equal to approximately 0.9 (Darveau et al., 2002). Furthermore, studies confirm differential scaling of various organs and their mitochondrial membrane content, both of which undoubtedly contribute to the scaling of the metabolic rate (Hulbert and Else, 2000). Liver weight in mammals scales with 0.87 power of the body weight and total liver protein in mammals was reported to scale with 0.84, while the slopes for MP and CYP450 content for mammals in this study range from 0.91 to 0.95 (Lindstedt and Schaeffer, 2002; Munro, 1969; Prothero, 1982). Liver weight in fish scales with 0.887–1.03, while the slopes in this study for MP and CYP450 content scale with both around 1.45 (Crile and Quiring, 1940; Fitzsimmons et al., 2018). Also in other vertebrates the slopes for liver weight with body weight were lower than those reported for MP and CYP450 content in this study (Else et al., 2004; Withers and Hillman, 2001). So, MP and CYP450 contents tend to increase more with body weight than liver weight. Cytochrome c was reported to scale with 0.84 or 0.70 power of the body weight (Adolph, 1949; Munro, 1969). Hence, it is not surprising that the liver MP and CYP contents do not fit the three-quarters power of body weight patterns observed for many other allometric relationships. For liver CYP content and activities, variations is an intrinsic factor which may impact substantially scaling coefficients. In addition, variability in the expression of specific CYP isoforms may also lead to either higher or lower protein contents and this may be impacted by extrinsic factors such as chemical exposure.

Here, a database and reference values for liver MP and CYP450 content as well as respective allometric relationships with body weight for a range of vertebrate species was presented. These values provide data-driven values when direct measurements are impractical or not available from the peer-reviewed literature. In addition, these liver MP and CYP450 datasets have great potential applications. Future work includes applications of such reference values to predict *in vitro* to *in vivo* clearances for different vertebrate taxa using quantitative *in vitro* to *in vivo* extrapolation models coupled to PBK models using chemicals of relevance as case studies including pesticides, contaminants and feed additives.

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CRediT authorship contribution statement

L.S. Lautz: Conceptualization, Investigation, Formal analysis, Visualization, Writing - Original Draft, Writing - Review & Editing; **A.J. Hendriks**: Writing - review & editing; **J.L.C.M. Dorne**: Writing -review & editing, Project administration; **J. Louisse:** Writing -review & editing, Project administration. **N.I. Kramer**: Writing - review & editing, Conceptualization, Supervision.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data Availability

All data used are in the supporting information.

Appendix A. Supporting information

Supplementary data associated with this article can be found in the online version at doi:10.1016/j.tox.2023.153429.

References

Adolph, E.F., 1949. Quantitative relations in the physiological constitutions of mammals. Science 109, 579–585. https://doi.org/10.1126/science.109.2841.579.
Anderson, G.D., 2002. Children versus adults: pharmacokinetic and adverse-effect differences. Epilepsia 43 (Suppl 3), 53–59. https://doi.org/10.1046/j.1528-1157.43.s.3.5.x.
Barter, Z.E., Bayliss, M.K., Beaune, P.H., Boobis, A.R., Carlile, D.J., Edwards, R.J., Houston, J.B., Lake, B.G., Lipscomb, J.C., Pelkonen, O.R., Tucker, G.T., Rostami-Hodjegan, A., 2007. Scaling factors for the extrapolation of in vivo metabolic drug clearance from in vitro data: reaching a consensus on values of human microsomal protein and hepatocellularity per gram of liver. Curr. Drug Metab. 8, 33–45. https://doi.org/10.2174/138920007779315053.
Björkman, S., 2006. Prediction of cytochrome p450-mediated hepatic drug clearance in neonates, infants and children: how accurate are available scaling methods? Clin. Pharm. 45, 1–11. https://doi.org/10.2165/00003088-200645010-00001.
Boxenbaum, H., 1980. Interspecies variation in liver weight, hepatic blood flow, and antipyrine intrinsic clearance: extrapolation of data to benzodiazepines and phenytoin. J. Pharm. Biopharm. 8, 165–176. https://doi.org/10.1007/bf01065191.
Burness, G.P., Leary, S.C., Hochachka, P.W., Moyes, C.D., 1999. Allometric scaling of RNA, DNA, and enzyme levels: an intraspecific study. Am. J. Physiol. 277, R1164–R1170. https://doi.org/10.1152/ajpregu.1999.277.4.R1164.
Chiba, M., Ishii, Y., Sugiyama, Y., 2009. Prediction of hepatic clearance in human from in vitro data for successful drug development. AAPS J. 11, 262–276. https://doi.org/10.1208/s12248-009-9103-6.
Crile, G.W., Quiring, D.P., 1940. A record of the body weight and certain organ and gland weights of 3690 animals. Ohio J. Sci. 40, 219–260.
Dacasto, M., Eeckhoutte, C., Capolongoa, F., Dupuy, J., Carletti, M., Calléja, C., Nebbia, C., Alvinerie, M., Galtier, P., 2005. Effect of breed and gender on bovine liver cytochrome P450 3A (CYP3A) expression and inter-species comparison with other domestic ruminants. Vet. Res. 36, 179–190. https://doi.org/10.1051/vetres:2004066.
Darveau, C.A., Suarez, R.K., Andrews, R.D., Hochachka, P.W., 2002. Allometric cascade as a unifying principle of body mass effects on metabolism. Nature 417, 166–170. https://doi.org/10.1038/417166a.
Davies, R., Moyes, C.D., 2007. Allometric scaling in centrarchid fish: origins of intra- and inter-specific variation in oxidative and glycolytic enzyme levels in muscle. J. Exp. Biol. 210, 3798–3804. https://doi.org/10.1242/jeb.003897.
Dorne, J.L., 2010. Metabolism, variability and risk assessment. Toxicology 268, 156–164. https://doi.org/10.1016/j.tox.2009.11.004.
Edwards, N.A., 1975. Scaling of renal functions in mammals. Comp. Biochem Physiol. A Comp. Physiol. 52, 63–66. https://doi.org/10.1016/s0300-9629(75)80128-9.
Else, P.L., Hulbert, A.J., 1985. Mammals: an allometric study of metabolism at tissue and mitochondrial level. Am. J. Physiol. 248, R415–R421. https://doi.org/10.1152/ajpregu.1985.248.4.R415.
Else, P.L., Brand, M.D., Turner, N., Hulbert, A.J., 2004. Respiration rate of hepatocytes varies with body mass in birds. J. Exp. Biol. 207, 2305–2311. https://doi.org/10.1242/jeb.01017.
Fitzsimmons, P.N., Hoffman, A.D., Fay, K.A., Nichols, J.W., 2018. Allometric scaling of hepatic biotransformation in rainbow trout. Comp. Biochem. Physiol. Part C: Toxicol. Pharmacol. 214, 52–60. https://doi.org/10.1016/j.cbpc.2018.08.004.
Giantin, M., Carletti, M., Capolongo, F., Pegolo, S., Lopparelli, R.M., Gusson, F., Nebbia, C., Cantiello, M., Martin, P., Pineau, T., Dacasto, M., 2008. Effect of breed upon cytochromes P450 and phase II enzyme expression in cattle liver. Drug Metab. Dispos. 36, 885–893. https://doi.org/10.1124/dmd.107.019042.
Hakooz, N., Ito, K., Rawden, H., Gill, H., Lemmers, L., Boobis, A.R., Edwards, R.J., Carlile, D.J., Lake, B.G., Houston, J.B., 2006. Determination of a human hepatic microsomal scaling factor for predicting in vivo drug clearance. Pharm. Res. 23, 533–539. https://doi.org/10.1007/s11095-006-9531-2.
Hendriks, A.J., 1999. Allometric scaling of rate, age and density parameters in ecological models. Oikos 86, 293-293 10.10.2307/3546447.
Hendriks, A.J., 2007. The power of size: a meta-analysis reveals consistency of allometric regressions. Ecol. Model. 205, 196–208. https://doi.org/10.1016/j.ecolmodel.2007.02.029.
Huang, Q., Gehring, R., Tell, L.A., Li, M., Riviere, J.E., 2015. Interspecies allometric meta-analysis of the comparative pharmacokinetics of 85 drugs across veterinary and laboratory animal species. J. Vet. Pharm. Ther. 38, 214–226. https://doi.org/10.1111/jvp.12174.
Hulbert, A.J., Else, P.L., 2000. Mechanisms underlying the cost of living in animals. Annu Rev. Physiol. 62, 207–235. https://doi.org/10.1146/annurev.physiol.62.1.207.
Hunter, R.P., 2010. Interspecies allometric scaling. Handb. Exp. Pharm. 139–157. https://doi.org/10.1007/978-3-642-10324-7_6.
Hunter, R.P., Isaza, R., 2008. Concepts and issues with interspecies scaling in zoological pharmacology. J. Zoo. Wildl. Med. 39, 517–526. https://doi.org/10.1638/2008-0041.1.
Jancova, P., Anzenbacher, P., Anzenbacherova, E., 2010. Phase II drug metabolizing enzymes. Biomed. Pap. Med Fac. Univ. Palacky. Olomouc Czech Repub. 154, 103–116. https://doi.org/10.5507/bp.2010.017.

Kasteel, E.E.J., Lautz, L.S., Culot, M., Kramer, N.I., Zwartsen, A., 2021. Application of in vitro data in physiologically-based kinetic models for quantitative in vitro-in vivo extrapolation: a case-study for baclofen. Toxicol. Vitr. 76, 105223 https://doi.org/10.1016/j.tiv.2021.105223.

Knaak, J.B., Albayati, M.A., Raabe, O.G., Blancato, J.N., 1993. Development of in vitrovmax and km values for the metabolism of isofenphos by P-450 liver enzymes in animals and human. Toxicol. Appl. Pharmacol. 120, 106–113. https://doi.org/10.1006/taap.1993.1092.

Lindstedt, S.L., Schaeffer, P.J., 2002. Use of allometry in predicting anatomical and physiological parameters of mammals. Lab Anim. 36, 1–19. https://doi.org/10.1258/0023677021911731.

Lipscomb, J.C., Teuschler, L.K., Swartout, J.C., Striley, C.A., Snawder, J.E., 2003. Variance of microsomal protein and cytochrome P450 2E1 and 3A forms in adult human liver. Toxicol. Mech. Methods 13, 45–51. https://doi.org/10.1080/15376510309821.

Martignoni, M., Groothuis, G.M., de Kanter, R., 2006. Species differences between mouse, rat, dog, monkey and human CYP-mediated drug metabolism, inhibition and induction. Expert Opin. Drug Metab. Toxicol. 2, 875–894. https://doi.org/10.1517/17425255.2.6.875.

Martinez, M., Mahmood, I., Hunter, R.P., 2006. Interspecies allometric scaling: prediction of clearance in large animal species: part II: mathematical considerations. J. Vet. Pharm. Ther. 29, 425–432. https://doi.org/10.1111/j.1365-2885.2006.00787.x.

Matal, J., Jancova, P., Siller, M., Masek, V., Anzenbacherova, E., Anzenbacher, P., 2008. Interspecies comparison of the glucuronidation processes in the man, monkey, pig, dog and rat. Neuro Endocrinol. Lett. 29, 738–743.

Munro, H., 1969. Evolution of protein metabolism in mammals. Mamm. Protein Metab. 3, 133–182.

Nebbia, C., 2001. Biotransformation enzymes as determinants of xenobiotic toxicity in domestic animals. Vet. J. 161, 238–252. https://doi.org/10.1053/tvjl.2000.0561.

Nebbia, C., Ceppa, L., Dacasto, M., Nachtmann, C., Carletti, M., 2001. Oxidative monensin metabolism and cytochrome P450 3A content and functions in liver microsomes from horses, pigs, broiler chicks, cattle and rats. J. Vet. Pharm. Ther. 24, 399–403. https://doi.org/10.1046/j.1365-2885.2001.00362.x.

Poggesi, I., 2004. Predicting human pharmacokinetics from preclinical data. Curr. Opin. Drug Disco Devel 7, 100–111.

Prothero, J.W., 1982. Organ scaling in mammals: the liver. Comp. Biochem Physiol. A Comp. Physiol. 71, 567–577. https://doi.org/10.1016/0300-9629(82)90205-5.

Punt, A., Pinckaers, N., Peijnenburg, A., Louisse, J., 2021. Development of a web-based toolbox to support quantitative in-vitro-to-in-vivo extrapolations (QIVIVE) within nonanimal testing strategies. Chem. Res. Toxicol. 34, 460–472. https://doi.org/10.1021/acs.chemrestox.0c00307.

Punt, A., Louisse, J., Beekmann, K., Pinckaers, N., Fabian, E., Van Ravenzwaay, B., Carmichael, P.L., Sorrell, I., Moxon, T.E., 2022a. Predictive performance of next generation human physiologically based kinetic (PBK) models based on in vitro and in silico input data. Altex 39, 221–234. https://doi.org/10.14573/altex.2108301.

Punt, A., Louisse, J., Pinckaers, N., Fabian, E., van Ravenzwaay, B., 2022b. Predictive performance of next generation physiologically based kinetic (PBK) model predictions in rats based on in vitro and in silico input data. Toxicol. Sci. 186, 18–28. https://doi.org/10.1093/toxsci/kfab150.

Sadler, N.C., Nandhikonda, P., Webb-Robertson, B.J., Ansong, C., Anderson, L.N., Smith, J.N., Corley, R.A., Wright, A.T., 2016. Hepatic cytochrome P450 activity, abundance, and expression throughout human development. Drug Metab. Dispos. 44, 984–991. https://doi.org/10.1124/dmd.115.068593.

Smith, R., Jones, R.D., Ballard, P.G., Griffiths, H.H., 2008. Determination of microsome and hepatocyte scaling factors for in vitro/in vivo extrapolation in the rat and dog. Xenobiotica 38, 1386–1398. https://doi.org/10.1080/00498250802491662.

Temerin, L.A., 1985. Size, function, and life history. By W.A. Calder III, 1984. xii + 431 pp. In: American Journal of Physical Anthropology, 66. Harvard University Press, Cambridge, pp. 340–342. https://doi.org/10.1002/ajpa.1330660312, 1984. xii + 431 pp.

WHO, 2005. Chemical-specific adjustment factors for interspecies differences and human variability: guidance document for use of data in dose/concentration-response assessment. 9241546786, World Health Organization, Geneva.

Wilson, Z.E., Rostami-Hodjegan, A., Burn, J.L., Tooley, A., Boyle, J., Ellis, S.W., Tucker, G.T., 2003. Inter-individual variability in levels of human microsomal protein and hepatocellularity per gram of liver. Br. J. Clin. Pharm. 56, 433–440. https://doi.org/10.1046/j.1365-2125.2003.01881.x.

Withers, P.C., Hillman, S.S., 2001. Allometric and ecological relationships of ventricle and liver mass in anuran amphibians. Funct. Ecol. 15, 60–69. https://doi.org/10.1046/j.1365-2435.2001.00495.x.

Zhang, H., Gao, N., Tian, X., Liu, T., Fang, Y., Zhou, J., Wen, Q., Xu, B., Qi, B., Gao, J., Li, H., Jia, L., Qiao, H., 2015. Content and activity of human liver microsomal protein and prediction of individual hepatic clearance in vivo. Sci. Rep. 5, 17671. https://doi.org/10.1038/srep17671.