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Authors

Rocha, Joana L
Brito, José C
Nielsen, Rasmus
et al.

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REVIEW

Convergent evolution of increased urine-concentrating ability in desert mammals

Joana L. ROCHA* *CIBIO/InBIO, Centro de Investigação em Biodiversidade e Recursos Genéticos, Universidade do Porto, Campus de Vairão, Vairão 4485-661, Portugal and Departamento de Biologia, Faculdade de Ciências, Universidade do Porto, Porto 4169-007, Portugal.*
Email: joana.laranjeira.rocha@gmail.com
José C. BRITO *CIBIO/InBIO, Centro de Investigação em Biodiversidade e Recursos Genéticos, Universidade do Porto, Campus de Vairão, Vairão 4485-661, Portugal and Departamento de Biologia, Faculdade de Ciências, Universidade do Porto, Porto 4169-007, Portugal. Email: jcbrito@cibio.up.pt*
Rasmus NIELSEN *Department of Integrative Biology and Department of Statistics, University of California Berkeley, Berkeley, CA, USA and Globe Institute, University of Copenhagen, Copenhagen DK-1165, Denmark. Email: rasmus_nielsen@berkeley.edu*
Raquel GODINHO* *CIBIO/InBIO, Centro de Investigação em Biodiversidade e Recursos Genéticos, Universidade do Porto, Campus de Vairão, Vairão 4485-661, Portugal and Departamento de Biologia, Faculdade de Ciências, Universidade do Porto, Porto 4169-007, Portugal and Department of Zoology, University of Johannesburg, PO Box 534, Auckland Park, 2006, South Africa. Email: rgodinho@cibio.up.pt*

ABSTRACT

1. One of the most celebrated textbook examples of physiological adaptations to desert environments is the unique ability that desert mammals have to produce hyperosmotic urine. Commonly perceived as an adaptation mainly observed in small rodents, the extent to which urine-concentrating ability has evolved independently in distinct mammalian lineages has not previously been assessed using modern phylogenetic approaches.

2. We review urine-concentrating ability data from the literature in 121 mammalian species with geographic ranges encompassing varying climatic conditions. We explicitly test the general hypothesis that desert-dwelling mammals have evolved greater ability to concentrate urine than non-desert species, controlling for body mass, phylogenetic affinity and other covariates.

3. Ancestral state reconstruction across our dataset's phylogeny shows that the ability to produce hyperosmotic urine, measured as maximum urine osmolality, has evolved convergently in mammalian species with geographic ranges characterised by low mean annual aridity index.

4. Phylogenetic generalised least-squares (PGLS) models show that the mean annual aridity index of a species' geographic range largely predicts its urine-concentrating ability, even when accounting for body mass differences, phylogenetic correlations, the specific condition under which urine osmolality was measured, the method used to measure urine osmolality, and the species' diet.

5. In contrast, we find much weaker correlations between mass-adjusted basal metabolic rate and environmental variables when analysing 84 of the species included in the urine osmolality analysis.

6. Taken together, our results not only show that desert mammals effectively concentrate more urine than non-desert mammals, but further suggest that aridity is likely to have been one of the main selective pressures leading to increasing maximum urine-concentrating ability and driving its repeated evolution in different desert mammalian lineages.

INTRODUCTION

Deserts are defined as regions with an aridity index, a ratio of annual precipitation and potential evapotranspiration, below 0.20 (Ward 2016). Desert species independently evolved striking adaptations to cope with the water scarcity and extreme climatic and physical conditions that characterise such habitats (Nagy 2004, Willmer et al. 2005, Schwimmer & Haim 2009, Pannabecker 2013, Fuller et al. 2014, Rymer et al. 2016). For the last fifty years, this has captured the attention of many eco-physiologists who turned to desert biology to study the physiological mechanisms that help desert animals to maintain their body temperature and retain water. Remarkably, most of the classic works on mammalian desert physiology, pioneered by Knut Schmidt-Nielsen (Schmidt-nielsen et al. 1948, Schmidt-Nielsen et al. 1964, Schmidt-nielsen 1965), have survived the test of time, as researchers have applied new perspectives and tools to test specific hypothesis regarding the evolution of adaptive traits (Walsberg 2000, Tracy & Walsberg 2002).

Among the classical findings of physiological adaptations that minimise water loss is the ability of some desert mammals to produce highly concentrated urine (Sands & Layton 2014). This phenotype is mostly associated with desert 'evaders', such as the Australian spinifex hopping mouse *Notomys alexis*, which holds the record for highest hyperosmotic urine (above 9000 mOsm/kg; Schwimmer & Haim 2009, Donald & Pannabecker 2015). Evaders, part of a classification system proposed by Willmer et al. (2005), are small animals that are able to evade extreme conditions through behaviour (Willmer et al. 2005). By contrast, large mammals unable to shelter from extreme climates that are forced to withstand heat, are called 'endurers', and medium-sized mammals unable to evade or withstand extremes as efficiently as evaders and endurers are called 'evaporators' (Willmer et al. 2005). Mammalian urine-concentrating ability is negatively correlated with body mass (Beuchat 1990), so the small desert evaders stand out in this capacity (Donald & Pannabecker 2015). However, when mass-adjusted, the capacity to produce highly-concentrated urine seems to have evolved independently in desert mammals (Beuchat 1990, 1996).

Classical comparative physiology studies have provided evidence for convergent adaptive evolution of ecologically relevant phenotypes in multiple desert mammals. A textbook example includes basal metabolic rate (BMR), which is predicted by latitude and zonal climate, even when accounting for phylogeny and body mass (Lovegrove 2000, Fristoe et al. 2015). Desert mammals evolved lower production of metabolic heat to maintain body temperatures and reduce evaporative water loss (Elgar & Harvey 1987, Lovegrove 2000, 2003, Clarke et al. 2010, Fristoe et al.

2015). Surprisingly, and to the best of our knowledge, no study to date has attempted to identify continuous climatic or environmental variables that help explain variation in urine-concentrating ability, one of the most celebrated examples of mammalian desert adaptation. Moreover, previous studies concerning urine-concentrating ability have ignored phylogenetic correlations when analysing associations between this trait and the environment (Beuchat 1990, 1996).

We explicitly test the hypothesis that the aridity index of a species' geographic range predicts its maximum urine-concentrating ability, even when accounting for body mass and phylogenetic relationship, and provide statistical evidence that the ability to avoid water loss by producing hyperosmotic urine has evolved independently in multiple phylogenetic lineages of desert mammals. Additionally, we use similar models to reanalyse mass-adjusted BMR for species in which urine osmolality was obtained, and test aridity index and temperature variables as predictors for metabolism in desert mammals.

METHODS

Phenotypes were obtained by extensive web-based literature search and by taking advantage of large datasets from previously published revision studies (Beuchat 1990, 1996, Lovegrove 2000, White & Seymour 2003). A total of 108 mammalian species' maximum urine-concentrating abilities came from two previous studies by Beuchat (1990, 1996), in which the author recorded the specific condition (C) under which urine osmolality (mOsm) was measured, including dehydrated (D), given salt (S) or protein (P) loading, combinations of these conditions, or treatment not specified (X); the method (M) used to measure urine mOsm, as well as the study-species' diet (Di; see Beuchat 1990, 1996 and Appendix S1 for more information). We searched Google Scholar (http://scholar.google.com) for the keywords 'urine osmolality', 'mammals', 'urine-concentrating ability', and recorded the highest mOsm values for 13 additional mammalian species from other studies (Nagy et al. 1976, Maloiy et al. 1979, Downs & Perrin 1991, Kronfeld & Shkolnik 1996, Diaz & Ojeda 1999, Ostrowski et al. 2006). For these studies we also noted the experimental conditions the study-cases were subjected to, the method used to measure urine mOsm, and species' diet type, following Beuchat (1990, 1996). For species with more than one record of urine mOsm, we opted to keep values taken from captive study-subjects under controlled or experimentally induced dehydration, as opposed to measurements of unknown condition taken in the field. Only one measurement was used per species. All species in our dataset were classified according to the endurer-evader-evaporator concept proposed by Willmer

et al. (2005). A full detail of the conditions and studies can be found in Appendix S1.

As mOsm is a plastic trait, the experimental conditions under which this parameter is measured can greatly affect estimates, particularly the degree of hydration of study-subjects. Therefore, we curated two datasets: 1) a larger dataset including all 121 reported measurements of mOsm (see Appendix S1); and 2) a subset containing only 87 observations in which the study-subject, in captivity, was dehydrated and not given food or other treatments. However, we note that there is still some residual variation in the number of days under water-deprivation for an animal to be considered fully dehydrated, and variation in the method used for measuring mOsm. Also, for the 121-species dataset, 24 records came from studies that did not report the procedures used to measure mOsm. Downstream analyses were performed on both datasets.

The geographic range polygons of each species for which physiological data were available were downloaded from the website of the International Union for Conservation of Nature (IUCN 2019, 2020). We also downloaded the following environmental variables at about 1 km spatial resolution: annual aridity index (AI) from the CGIAR-CSI Global Aridity database (Trabucco & Zomer 2009), and annual mean temperature (BIO1), maximum temperature of the warmest month (BIO5), minimum temperature of the coldest month (BIO6) and temperature annual range (BIO7) from the WorldClim database (Fick & Hijmans 2017). We then used a Geographical Information System to calculate mean values for each environmental variable within the geographic range polygon of each species. For

species with global geographic ranges, and for which phenotypes under review were sampled from a specific region, we partitioned the original distribution polygon accordingly and calculated mean values for the sampled zone. A full detail of the mean annual aridity index (AI) and mean temperature variables (BIOS) for each species' geographic range can be found in Appendix S1. To avoid the effects of collinearity, we tested for correlations between pairs of variables using a Spearman correlation test before testing them together as predictors for mOsm and BMR in downstream analyses. This test was performed using the 'ggscatter' function implemented in the R package 'ggpubr' with the correlation method option set to 'Spearman'. We used a previously published mammalian phylogenetic tree estimated using both nuclear and mitochondrial genes (Upham et al. 2019), and pruned the tree to retain only the species included in our study. For visualisation of trait evolution on the tree, we projected states for maximum mOsm and temperature variables, estimated using maximum-likelihood methods, onto the internal edges and nodes of the tree using a colour gradient. The estimation was done using the 'fastAnc' option in the 'contMap' function implemented in the R package 'phytools' (Revell 2012).

Phylogenetic generalised least squares (PGLS) as implemented in 'phytools' were performed using the 'gls' function with the model of Pagel (1999) to test different linear models with increasing complexity. Briefly, the model of Pagel adjusts the off-diagonal elements of variance-covariance matrix in a Brownian evolution model with a multiplicative factor, λ, such that when $\lambda = 0$, a star phylogeny is obtained representing no phylogenetic
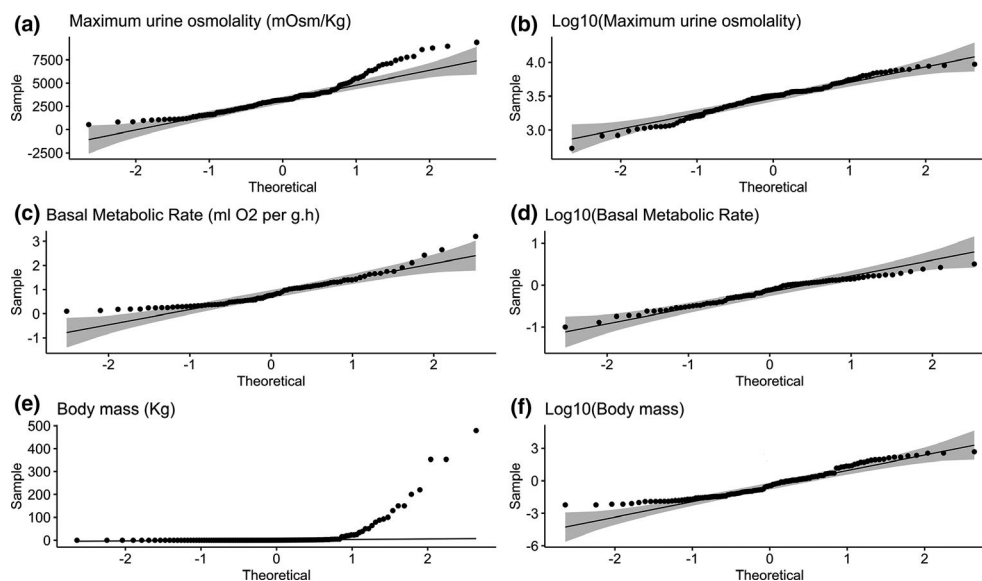

Fig. 1. Quantile–quantile plots for the normal distribution of variables (a, c, e) and log10 of variables (b, d, f) used in this study of urine-concentrating ability of desert mammals.

Fig. 2. Ancestral state reconstruction for mean annual aridity index (AI, left) and maximum urine osmolalities (mOsm/kg; right) in a phylogenetic tree with 121 mammalian species. [Colour figure can be viewed at wileyonlinelibrary.com]

signal, while when λ = 1, a standard Brownian motion process on the reference tree is obtained. As residuals of maximum mOsm are not normally distributed (Fig. 1), we performed PGLS analyses on log10 (maximum mOsm) using log10 (body mass) as covariate. Mean AI

and/or mean temperature variables were used in PGLS models as potential predictors of urine-concentrating ability. Condition (C), method (M) and diet (Di) were also included as covariates. For the dataset including only dehydrated individuals, the same PGLS models were

Table 1. Phylogenetic signal (λ) for variables with significant *P*-values. Variables for which λ is close to 1, and with *P*-values < 0.05, have significant phylogenetic signals

Variable	Pagel's λ (*P*-value)
Log_{10} (maximum urine osmolality)	0.988 (3.26e-12)
Log_{10} (body mass)	1.000 (1.26e-41)
Mean AI (annual aridity index)	0.854 (0.0002)
Mean BIO1 (annual mean temperature)	0.668 (0.0026)
Mean BIO6 (minimum temperature of the coldest month)	0.772 (0.0002)
Mean BIO7 (temperature annual Range)	0.840 (4.90 e-06)

tested with AI converted to a categorical variable using the following binning (defined by Trabucco & Zomer 2009): humid for AI > 0.65, dry sub-humid for $0.5 < AI < 0.65$, semi-arid for $0.2 < AI < 0.5$ and arid for $AI < 0.2$. To test for phylogenetic signal for specific traits, we calculated Pagel's λ on urine-concentrating ability, body mass and AI using the function 'phylosig' as implemented in 'phytools'. All statistics were performed in R v. 4.0.1 and RStudio v. 1.3.1073. Detailed code implementing these analyses is available at https://github.com/joanocha/GIS_PGLS_MetaAnalysis.

RESULTS

We analysed the dataset with information on maximum urine-concentrating abilities, mean annual aridity index (AI) and WorldClim temperatures (BIOS) for a total of 121 mammal species with available DNA sequence information. Ancestral state reconstruction across our dataset's phylogeny shows that the ability to produce hyperosmotic urine has evolved multiple times in mammalian species with geographic ranges with low mean annual aridity index (Fig. 2).

When testing for phylogenetic signal using Pagel's λ, we found significant phylogenetic signal for most variables, validating the relevance of adjusting the linear models for phylogenetic covariance (Table 1). All PGLS models tested in this study point to mean AI as a significant and important predictor of mammalian maximum mOsm (Table 2; see also Appendix S2 for all PGLS models tested). The lower the mean annual aridity index, the more efficiently mammals are able to concentrate urine (Fig. 3), even when accounting for the differences in body mass, diet, experimental condition, methodology and phylogeny (Table 2, Appendix S2). Overall, species inhabiting arid and semi-arid environments can maximise urine concentration well above

Table 2. PGLS models for predicting mammalian log10 (maximum urine osmolality). Model notation refers to the variables used as predictors: mean AI = annual aridity index, BM = log10 body mass (kg); C = condition (fed-F, given salt load-S, protein load-P, unknown condition-X, and combinations of these, all relative to dehydrated-D); M = method (addition of solutes-A, freezing point-F, vapour pressure-V, and combinations of these, all relative to unknown method-?); Di = diet (frugivorous-Fr, granivorous-G, herbivorous-H, insectivorous-I, omnivorous-O, and CI-both carnivorous and insectivorous, all relative to strictly carnivorous-C). Significant model variables and *P*-values are highlighted in bold; AIC = Akaike information criterion, SE = standard error.

| Model	AIC	Variable	Coefficient	SE	t-value	*P*-value
AI + BM (best model)	−74.4	Intercept	3.632	0.059	61.64	**0.0000**
		AI	−0.373	0.040	−9.20	**0.0000**
		BM	−0.092	0.014	−6.43	**0.0000**
AI + BM + C + M + Di	−16.3	intercept	3.703	0.097	37.99	**0.0000**
		AI	−0.368	0.043	−8.47	**0.0000**
		BM	−0.082	0.015	−5.54	**0.0000**
		$DF_{condition}$	0.023	0.080	0.28	0.7763
		$DP_{condition}$	−0.081	0.108	−0.75	0.4537
		$DPS_{condition}$	0.020	0.178	0.11	0.9104
		$DS_{condition}$	−0.047	0.106	−0.45	0.6573
		$P_{condition}$	−0.006	0.077	−0.08	0.9341
		$S_{condition}$	0.049	0.065	0.75	0.4536
		$SP_{condition}$	0.148	0.148	1.00	0.3202
		$X_{condition}$	−0.049	0.047	−1.06	0.2911
		A_{method}	−0.068	0.153	−0.44	0.6592
		F_{method}	0.016	0.039	0.41	0.6801
		FV_{method}	0.058	0.097	0.60	0.5530
		V_{method}	0.106	0.055	1.93	0.0570
		CI_{diet}	−0.158	0.141	−1.12	0.2649
		Fr_{diet}	−0.533	0.149	−3.57	**0.0005**
		G_{diet}	0.013	0.103	0.13	0.8980
		H_{diet}	−0.132	0.088	−1.50	0.1366
		I_{diet}	−0.130	0.107	−1.22	0.2260
		O_{diet}	−0.055	0.095	−0.58	0.5634
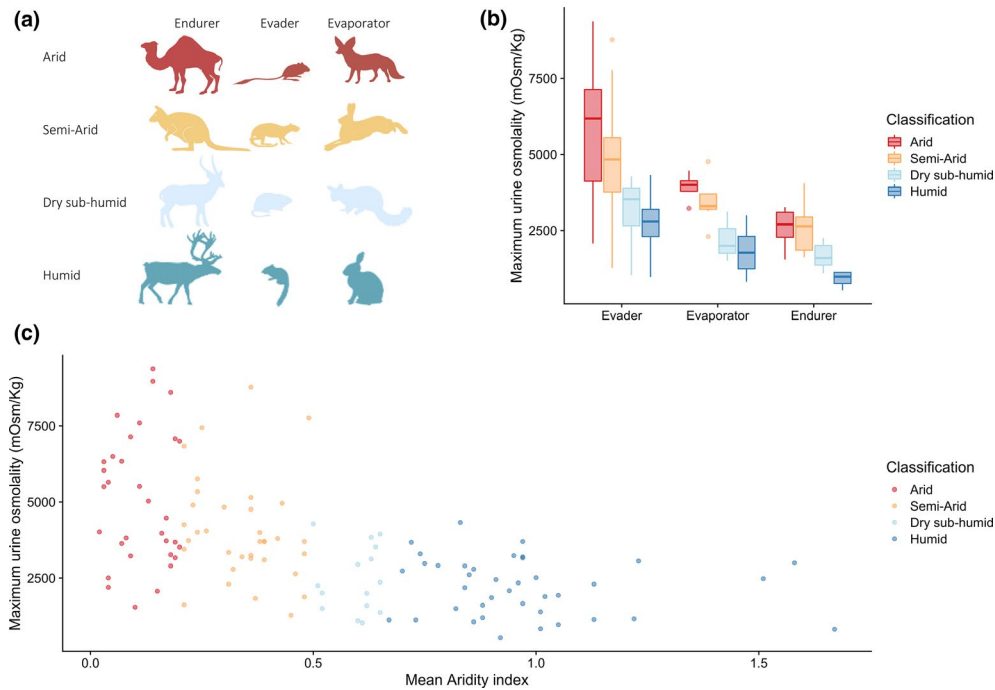
Fig. 3. Maximum urine osmolality (mOsm/kg) in mammals and mean aridity index (AI). (a) Schematic illustration of the endurer-evader-evaporator concept proposed by Willmer et al. (2005) and classification of mammals into arid, semi-arid, dry sub-humid and humid habitat occupiers according to Trabucco and Zomer (2009). (b) Boxplot for maximum mOsm in 121 endurers, evaders, and evaporators with ranges with different AIs. (c) Scatterplot for maximum mOsm in 121 mammal species. [Colour figure can be viewed at wileyonlinelibrary.com]

the levels of their dry sub-humid and humid counterparts (Fig. 3). In congruence with Beuchat (1990), our results also show that log-transformed body mass is also negatively correlated with log-transformed maximum mOsm, though not to the same extent as mean annual aridity index (Table 2, Fig. 4). We also find that urine-concentrating ability is significantly lower in species with a frugivorous diet than in strictly carnivorous species (Table 2). Mean annual aridity index or mean AI correlates with temperature variables,

except mean BIO7 (Appendix S3). Overall, no strong correlations were found between urine-concentrating ability and temperature variables (see Appendix S2 for PGLS using temperature variables as covariates, and Appendix S4 for visual illustration of the relationship between mOsm and WorldClim temperatures). When experimental condition and methodology were used as covariates in the PGLS analysis, their role in predicting maximum mOsm was not significant (Table 2, Appendix S2).
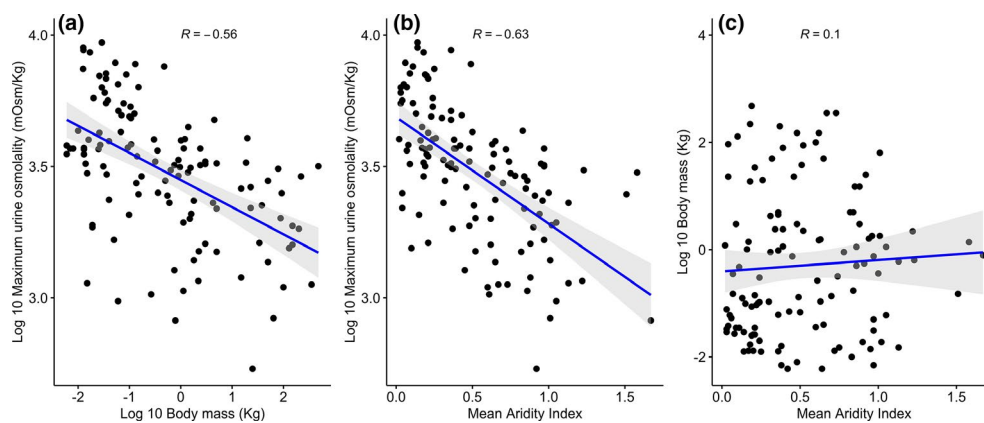

Fig. 4. Spearman correlations for: (a) log10 maximum urine osmolality (measured in mOsm/kg) and $\log_{10}$ body mass (kg); (b) $\log_{10}$ maximum urine osmolality (measured in mOsm/kg) and mean annual aridity index; and (c) $\log_{10}$ body mass (kg) and mean annual aridity index. P-values are not presented, as the plots do not take phylogenetic affinity into account. [Colour figure can be viewed at wileyonlinelibrary.com]

Table 3. PGLS models for predicting mammalian log10 maximum urine osmolality (mOsm/kg) in the dataset including only dehydrated individuals. Model notation refers to the combination of variables used as predictors while taking phylogeny into account: mean AI = annual aridity index; BM = log10 body mass (kg); M = method (addition of solutes-A, freezing point-F, vapour pressure-V, and combinations of these, all relative to unknown method-?); Di = diet (frugivorous-Fr, granivorous-G, herbivorous-H, insectivorous-I, omnivorous-O, and CI-both carnivorous and insectivorous, all relative to strictly carnivorous-C). Significant model variables and *P*-values are highlighted in bold; AIC = Akaike information criterion, SE = standard error.

Model	AIC	Variable	Coefficient	SE	t-value	*P*-value
AI + BM (best model)	−62.3	Intercept	3.572	0.167	21.33	**0.0000**
		AI	−0.309	0.047	−6.51	**0.0000**
		BM	−0.091	0.022	−4.13	**0.0004**
AI + BM + M + Di	−34.7	intercept	3.679	0.132	27.83	**0.0000**
		AI	−0.344	0.043	−7.96	**0.0000**
		BM	−0.067	0.016	−4.18	**0.0001**
		A $_{method}$	−0.107	0.141	−0.75	0.4528
		F $_{method}$	−0.007	0.042	−0.16	0.8702
		FV $_{method}$	0.096	0.104	0.93	0.3563
		V $_{method}$	0.029	0.070	0.42	0.6758
		CI $_{diet}$	−0.098	0.165	−0.60	0.5536
		Fr $_{diet}$	−0.406	0.178	−2.28	**0.0254**
		G $_{diet}$	0.090	0.142	0.63	0.5281
		H $_{diet}$	−0.106	0.135	−0.79	0.4333
		I $_{diet}$	−0.049	0.140	−0.35	0.7261
		O $_{diet}$	0.002	0.139	0.02	0.9873
cAI + BM + M + Di	−24.99	intercept	3.663	0.137	26.71	**0.0000**
		Dry sub-humid	−0.242	0.051	−4.72	**0.0000**
		Humid	−0.342	0.045	−7.58	**0.0000**
		Semi-arid	−0.105	0.045	−2.37	**0.0207**
		BM	−0.068	0.016	−4.32	**0.0000**
		A $_{method}$	−0.154	0.143	−1.08	0.2829
		F $_{method}$	−0.035	0.042	−0.83	0.4094
		FV $_{method}$	0.084	0.105	0.80	0.4246
		V $_{method}$	0.034	0.068	0.50	0.6208
		CI $_{diet}$	−0.116	0.167	−0.70	0.4880
		Fr $_{diet}$	−0.438	0.179	−2.45	**0.0166**
		G $_{diet}$	0.105	0.144	0.73	0.4653
		H $_{diet}$	−0.083	0.137	−0.61	0.5454
		I $_{diet}$	−0.011	0.143	−0.08	0.9384
		O $_{diet}$	0.036	0.140	0.26	0.7971

We tested similar PGLS models on the subset of the data that only included dehydrated study-subjects, resulting in a smaller dataset including 87 species (18 arid, 26 semi-arid, 13 dry sub-humid and 30 humid). The results from this analysis (illustrated in Table 3) are similar to those of the larger dataset conditions (Table 2). When using AI as a discrete variable, we found that mOsm was significantly higher in species from arid environments than in species from semi-arid, dry sub-humid and humid regions (Table 3). In all analyses, the mean AI of the species' geographic range is a stronger predictor of maximum mOsm than body mass and diet, and the condition of study-subjects and methodology used to measure this trait are not significant predictors (Tables 2 and 3; see also Appendix S2).

We obtained mass-adjusted BMR for 84 of the 121 species from our dataset. BMR was weakly correlated with bioclimatic variables (Appendix S5), and variation in this parameter is better explained by log10 body mass (Table 4).

DISCUSSION

The extent to which different mammalian species from desert environments have significantly higher urine-concentrating abilities than mammals from other environments, thus exhibiting phenotypes enabling better water retention in extreme conditions of water-deprivation and extreme temperature ranges, has not previously been subject to rigorous statistical analyses that account for phylogeny (Beuchat 1990, 1996). We performed ancestral state reconstruction on a molecular phylogeny to show that increased ability to concentrate urine, measured as maximum mOsm, has evolved convergently in many desert mammals.

Table 4. PGLS models for predicting log10 mass-adjusted basal metabolic rate (ml of O$_2$ per g per day) ranked by AIC (Akaike information criterion). Model notation refers to the combination of variables used as predictors while taking phylogeny into account. Variables are BM = log10 body mass (g); mean AI = annual aridity index (continuous), cAI = categorical AI (arid, semi-arid, dry sub-humid, humid), Di = diet (carnivorous-C, frugivorous-Fr, granivorous-G, herbivorous-H, insectivorous-I, omnivorous-O, carnivorous and insectivorous-CI); and mean temperature values for BIO1 = annual mean temperature, BIO5= maximum temperature of warmest month, BIO6 = minimum temperature of coldest month, BIO7 = temperature annual range. Significant variables are highlighted in bold (P-value < 0.05). When λ is zero, the covariance between species is zero and the linear model approaches that of a non-phylogenetic regression, while when λ is 1, the evolution of the residual error follows a Brownian model.

Model	AIC	Pagel's λ
BM	−27.28	0.7601046
BM + AI	−23.61	0.7503346
BM + BIO1 + BIO5 + BIO6 + BIO7	−6.51	0.6825869
BM + AI + BIO1 + BIO5 + BIO6 + BIO7	−1.69	0.6759448
BM + AI + Di	−1.68	0.6288732
BM + **BIO1** + Di	1.73	0.5731028
BM + BIO5 + Di	1.77	0.6274593
BM + **BIO6** + Di	1.91	0.5383454
BM + BIO7 + Di	4.05	0.5487208
BM + cAI + Di	5.69	0.5982846
BM + BIO1 + BIO5 + BIO6 + BIO7 + Di	12.28	0.4437627

Phylogenetic generalised least squares analysis on a multi-species dataset revealed that desert-dwelling species effectively concentrate more urine than non-desert species, even when accounting for both body mass and ancestry, and, furthermore, that differences in mammalian maximum mOsm can be predicted, to some degree, by the aridity of species' range. Our measurement of environmental aridity is inherently imprecise as it does not account for spatial and temporal changes in the environmental conditions or in the species' geographic range, and it implicitly assumes a uniform density distribution across the species' range (Trabucco & Zomer 2009, Fick & Hijmans 2017). Still, we find it more rigorous than the often-used approach of classifying species as 'mesic' or 'xeric' based on habitat descriptions or information on whether the species' survival depends on standing water (Beuchat 1996, Lovegrove 2000). Despite the challenges, and while acknowledging additional noise added by varying experimental conditions and methodologies, we find a strongly-significant correlation between maximum mOsm and mean AI. This observation is similar to what has been previously reported in other desert-adaptive phenotypes, such as BMR (Lovegrove 2000, Fristoe et al. 2015). However, we find a much weaker correlation for BMR and environmental variables when analysing 84 of the same species as in the mOsm analysis, suggesting that, in contrast to our results for mOsm, differences in BMR among species are mostly explained by body mass

differences and are only weakly predicted by environmental variables (Genoud et al. 2018).

A contentious debate in the fields of ecological and evolutionary physiology is whether phenotypic differences between species and populations inhabiting contrasting environments are due to genetic adaptations or due to plastic responses, and difference in the ability to concentrate urine is no exception. Although all mammals, when water-deprived, are able to increase urine concentration in relation to their optimal hydrated states (Sands & Layton 2014, Knepper et al. 2015), our results, from a dataset in which subjects were all captive and water-deprived, provide strong evidence that increased maximal urine concentration is an adaptation to high aridity and has evolved multiple times in different phylogenetic lineages. Notably, increased urine-concentrating ability correlates with increased renal medullary thickness in desert mammals (Beuchat 1990, Schwimmer & Haim 2009).

Our PGLS analyses also suggest that, among the many physical and climatic challenges faced by desert species, aridity, more than temperature, has been one of the main selective pressures leading to increasing maximum urine-concentrating ability and driving its repeated evolution in different desert mammalian lineages. Future comprehensive statistical studies investigating correlations of bioclimatic variables with more of the classical desert phenotypes, using multiple covariates including phylogeny, will provide further elucidation on the extent to which climate is predictive for mammalian phenotypes. As population and comparative genomics studies in desert environments continue to search for the genetic basis of desert adaptation (e.g Wu et al. 2014, Kim et al. 2016, Yang et al. 2016, Bittner et al. 2020), maximum urine-concentrating ability might be one of the primary physiological traits that lends itself to more detailed genetic analyses.

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SUPPORTING INFORMATION

Additional supporting information may be found in the online version of this article at the publisher's website.

Appendix S1. Data collection from past studies concerning 121 species listed alphabetically by scientific name, followed by family and order.

Appendix S2. PGLS models for predicting log10 mammalian maximum urine osmolality (mOsm/kg) ranked by AIC (Akaike information criterion).

Appendix S3. Spearman correlations for mean annual aridity index and four measurements of temperature variation. *P*-values are not presented, as the plots do not take spatial correlation into account.

Appendix S4. Spearman correlation for log10 maximum urine osmolality (measured in mOsm/kg) and four measurements of temperature variation. *P*-values are not represented, as the plots do not take phylogenetic affinity into account.

Appendix S5. Spearman correlation for log10 mass-adjusted basal metabolic rate (ml of O_2 per g per day) and four measurements of temperature variation. *P*-values are not represented, as plots do not take phylogenetic affinity into account.