**Results**

We set out to test the effect of seasonality on leporid brain evolution, and more specifically, on the variation of OB and ROB volumes. We did not find any support for such effect (see Table XX). We tested 5 other models related to hypotheses explaining the evolution of brain size variation (shown in Table X) and we only found support for two of them: the pGLS models indicated that leporids with larger home ranges and such that exhibit burrowing behaviour (graph 1) have larger brains than non-burrowers or species with smaller home ranges. These results all included lambda values (indicating the strength of phylogenetic signal in the residuals) that were negative or were higher than 1, which violates the definition of lambda per se (see Discussion). Moreover, these results could not be replicated using MCMCglmm modelling.

Graph 1



|  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- |
|  | **MCMCglmm** | | | | **pGLS** | | | |
| **Model** | **Beta** | **SE** | **Posterior distribution > 0 (%)** | **DIC** | **t-value** | **p-value** | **Lambda** | **DF** |
| Seasonality  **ROB**  Temperature  Precipitation  **OB**  Temperature  Precipitation | 0.0140  0.0001  0.0527  0.0039 | 0.0544  0.0050  0.0801  0.0075 | 61  51  76  71 | -19.8  -17.1 | 0.48  1.05  0.07  0.01 | 0.63  0.31  0.49  0.59 | -0.33\*  0.58 | 18 |
| Maternal investment  **ROB**  Litter Size  Gestation Length  **OB**  Litter Size  Gestation Length | 0.0833  -0.0069  0.1313  -0.1612 | |  | | --- | |  | |  |   0.0871  0.2757  0.1325  0.3674 | 84  48  85  35 | -19.8  -16.2 | 1.05  -1.80  0.05  -0.29 | 0.31  0.1  0.32  0.78 | -0.38\*  0.58 | 17 |
| Spatial  **ROB**  Home Range  Geographic Area  **OB**  Home Range  Geographic Area | 0.0243  -0.004  0.0093  -0.0145 | 0.0167  0.0142  0.0256  0.0232 | 94  38  66  25 | -28.7  -21.6 | 7.42  -0.1  1.56  -0.55 | **<0.001**  0.85  0.15  0.6 | 1.12\*  0.93 | 14 |
|  |  |  |  |  |  |  |  |  |
| **Model name** | **Beta** | **SE** | **Posterior distribution > 0 (%)** | **DIC** | **t-value** | **p-value** | **Lambda** | **DF** |
| Diet  ***ROB***  Diet Breadth  ***OB***  Diet Breadth | 0.0159  -0.0106 | 0.0939  0.1347 | 58  47 | -21.5  -19.8 | -0.91  -0.76 | 0.39  0.46 | 1.05\*  0.72 | 14 |
| Behavioural (activity)  ***ROB***  Locomotor mode Generalised  Locomotor mode Saltatorial  Activity cycle  Burrow  ***OB***  Locomotor mode Generalised  Locomotor mode Saltatorial  Activity cycle  Burrow | 0.0527  0.0507  -0.1682  -0.0520  -0.0179  0.0816  0.1460  0.0424 | 0.1595  0.1156  0.1396  0.0788  0.2518  0.1842  0.2234  0.1281 | 64  68  11  24  47  68  24  65 | -21.7  -13.0 | 0.25  0.71  1.25  -0.44  -0.26  0.28  1.02  -0.29 | 0.81  0.49  0.24  0.67  0.8  0.79  0.33  0.78 | -0.27\*  0.55 | 18 |
| Burrowing  ***ROB***  Burrow  ***OB***  Burrow | 0.0258  0.0458 | 0.0684  0.1007 | 65  69 | -20.1  -20.7 | 2.57  1.4 | **0.02**  0.17 | -0.33\*  0.72 | 18 |

Table XX, Results from the MCMCglmm and pGLS analysis, ROB – Rest of brain, OB – Olfactory bulb, Asterisk indicates lambda values <0 and >1.

**Discussion**

We did not find support for any effect of seasonality on the evolution of brain size variation in leporids. Seasonality in temperature or precipitation has previously been shown to have both positive (REFS) and negative (REFS) effects on the evolution of brain size in vertebrates. One reason for the lack of such effect in leporids might be due to the fact that burrowing behaviour compensates for such climatic variation, and as such appears to be a selection force behind increase in brain size. This is in line with the cognitive buffering hypothesis (REF) and is also additionally supported by our finding that species with larger home ranges have larger brains. (ELABORATE ON BURROWING IN LEPORIDS)

We did not find support for any maternal investment effect limiting brain size evolution in leporids, as measured by litter size and weaning age. This might be due to the fact that our sample is relatively homogenous in terms of these two reproductive variables, and whilst such effect has been shown in larger clades (marsupials, etc REF), focusing on a family level, such effect might not be detectable. (ELABORATE ON THIS WITH A FEW MORE SENTENCES)

Additionally, our activity model was not supported either (including locomotor mode and activity cycle) indicating that leporid species, being mainly terrestrial are unable to buffer any environmental effects relating to brain size, besides using burrowing behaviour.

The size of the olfactory bulb in leporids did not seem to be related to any variable in any of our models. (EXPLAIN)

One observation in our pGLS models was the fact that in all models including ROB the lambda estimates were unrealistic. This is a common situation in many phylogenetic comparative studies (lizard paper, other REFS) that does not receive enough attention from scholars in the field. Lambda, as a measure of phylogenetic signal in the residuals, can only take values between 0 (indicating lack of phylogenetic signal), and 1, so any value larger than 1 or lower than 0 is a result of a flawed maximum likelihood estimation and is unrealistic. As shown on graph XXX, in all pGLS models including ROB the estimated lambda values’ maximum likelihood always falls outside the 0-1 interval. This is most probably due to the small sample size, which renders all lambda estimates unreliable. This and other cases of the same issue (REFS) warrant caution when using small sample sizes in phylogenetic comparative studies, as then, maximum likelihood estimations become unreliable. Such suggestion is supported by the fact that none of our MCMCglmm models confirmed the results obtained by pGLS, as MCMCglmm uses similar maximum likelihood algorithm to obtain probability densities of the posterior distribution. Obviously, with a sample size of 18 species, the available data were not enough for conclusive analysis using Bayesian inference. Unfortunately, the limitations of sample size are common in many comparative studies (REFS), and while such analyses lay a foundation for further inquiries after more extensive data collection, they should be taken with a grain of salt. Authors of such studies should pay more attention to the statistical artifacts arising from sample size limitation and analyse them in more details before reporting their results. PROPOSE A METHODOLOGICAL STEP BY STEP SOLUTION.

Conclusion