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## **Report of ThermApt Grant Thermoregulation in relation to body size and melanin pattern in Skyros wall lizard *Podarcis gaigeae***

### **Purpose of the visit**

The purpose of the visit at University of Athens was to investigate the link between morphology and thermoregulation in morphologically strongly diverged populations of Skyros wall lizard, *Podarcis gaigeae*. Our aim was to combine data on thermoregulatory properties of these physiologically diverged populations with thermal preferences and field data on activity patterns and temperatures of these populations in collaboration with the ecophysiologicals Efstratios Valakos and Panagiotis Pafilis.

### **Work carried out during the visit**

During the visit lizards from two islet populations exhibiting island gigantism as well as the populations on the respectively most proximate main island locals were captured in order to create a parallel design with two independent islet-main island pairs. All captured individuals were mature males or females. We recorded eight standard morphological measures with callipers and body weight with a pesola scales for all individuals. The colour was recorded with an RGB imager in a box with standardized light environment, a white background reference and constant flash illumination. The flash was covered with polarizing film to avoid specular reflexes. Our previous studies has shown that *P. gaigeae* does not have UV coloured backs ( $n > 800$ ), and hence the use of a spectrometer would not add any information that would not be captured by an RGB CED of a camera.

To investigate the effect of morphology on the heating rate, the lizards were cooled down to 15 degrees centigrades and placed in an open terrarium in a homogeneous light field, illuminated by six 50 W halogen spotlights with reflectors. We used a thermal image camera (TIR) to film the heating and extracted a thermal image every 10 seconds with a Matlab-script especially developed for the purpose. The trials were stopped when the temperature of the lizards reached 40 degrees centigrades, or as soon as the lizards tried to escape from the

terrarium. In addition, the same trials were done with the same individuals but without the lamps, in order to be able to disentangle the passive heating from the active heating of the lamps through subtracting the latter from the former. We investigated the heating rate of twelve individuals, six males and six females, from each population.

In addition, we studied the time and the temperature when the lizards from the different populations became active in the morning. All lizards had an individual compartment with a stone to hide under, coarse grained sand, and a water bowl. The terraria were kept in homogenous shade and as soon as an individual lizard emerged from under its' rock we took a thermal image of the lizard. The time, temperature of the lizard and temperature of the background at that moment was recorded. This lowest preferred activity temperature is meant to be a complement to the thermal preference data from a temperature gradient experiment already conducted by our Greek collaborators.

The last, but only preliminary part of the project was to measure the field temperatures of the lizards. Thermal images of lizards in their field environment were taken from 8 am (prior to the first lizards emerging, personal observation) until the hottest hour of the day (approximately 2 pm) for one islet- and one main island population. Complementing this data with more days, longer hours and more populations in the future would be interesting.

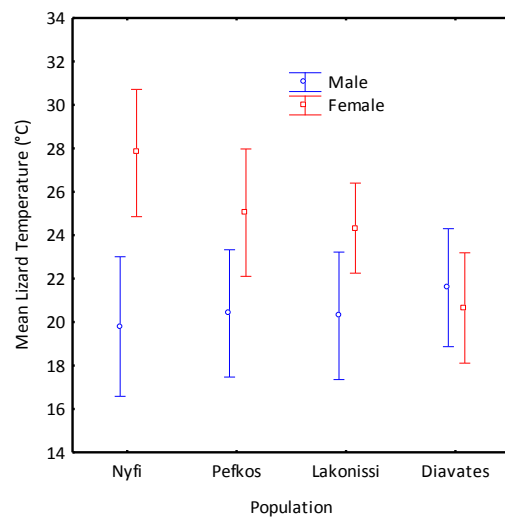
## **Main results**

We performed General Linear Model (GLM) to investigate how different factors affect heating rate in *P. gaigeae*. The time it takes for a lizard to increase in temperature from one given value to another is significantly affected by sex ( $p=0.023$ ), body weight ( $p=0.016$ ) and how dark the lizard is ( $p=0.049$ ; judged as mean  $R*B*G/3$  for all pixels of the back of the lizard). In addition, the interactions habitat\*sex ( $p=0.014$ ; where habitat is islet- or mainland population) is significant, due to smaller sex differences in the mainland populations. The interactions body weight\*dark ( $p=0.029$ ), sex\*greenness ( $p=0.014$ ; where greenness is the ratio  $G/R$  and a measure of how green the lizard is on a red-green scale) and weight\*greenness ( $p=0.034$ ) are also significant. A surprising result was that there was a significant interaction between throat colour morph and body weight ( $p<0.001$ ), where the orange morph differs from the other. There are six different throat colour morphs in *P. gaigeae*, white, yellow, orange and all combinations involving two of these colours, and the morphs are probably determined by a gene of major effect with three different alleles, although there is still no breeding data to confirm this (Runemark et al. *in revision*). Throat colour morphs have been shown to correlate with immune response in *Podarcis muralis* (Sacchi *et al.* 2007). The results of the standardised heating rate experiments are presented in Table 1.

**Table 1**, best fit model of the time it takes for the lizard to heat from 22 to 32 °C.

	SS	Degr. of	MS	F	p
<b>Intercept</b>	5496	1	5496	5.2	0.031
<b>Habitat of origin</b>	760	1	760	0.7	0.403
<b>Sex</b>	6229	1	6229	5.9	0.023
<b>Throatmorph</b>	12970	5	2594	2.5	0.061
<b>Weight</b>	7039	1	7039	6.7	0.016
<b>Dark</b>	4511	1	4511	4.3	0.049
<b>Greenness</b>	1026	1	1026	0.98	0.332
<b>Habitat*Sex</b>	7447	1	7447	7.1	0.014
<b>Habitat*Weight</b>	1039	1	1039	1.0	0.329
<b>Throatmorph*Weight</b>	36552	5	7310	7.0	< 0.001
<b>Habitat*Dark</b>	1507	1	1507	1.4	0.242
<b>Sex*Dark</b>	2913	1	2913	2.8	0.109
<b>Weight*Dark</b>	5644	1	5644	5.4	0.029
<b>Sex*Greenness</b>	7376	1	7376	7.0	0.014
<b>Throatmorph*Greenness</b>	10837	5	2167	2.1	0.105
<b>Weight*Greenness</b>	5284	1	5284	5.0	0.034
<b>Error</b>	25175	24	1048		

There was a clear sexual dimorphism in the temperature of the lizards when they emerged from their shelters, with the females being warmer than the males (GLM;  $F = 203$ ;  $df = 1$ ;  $p < 0.001$ ) as well as a significant interaction between population and sex (GLM;  $F = 3.48$ ;  $df = 3$ ;  $p = 0.023$ ), where islet populations tend to have smaller sex differences, see Fig. 1.



**Fig. 1.** Temperatures of the lizards when they emerge by sex and population. The populations to the left, Nyfi and Pefkos are mainland populations whereas Lakonissi and Diavates are islet populations.

The preliminary field experiments revealed larger fluctuations in body temperature over the day in the mainland populations than in the islet populations. This would be interesting to follow up on with more field experiments.

### **Future collaboration with host institution and projected publications**

The analysis of the data from this collaboration is not yet completed, but some promising results have already emerged (see main results). We hope to combine this data with the data of thermal preferences in a temperature gradient experiment performed by Dr. Valakos and Dr. Pafilis to write a paper on how physiology affects thermal preferences, thermoregulatory behaviour and the thermal niche of the different populations and sexes of *P. gaigeae* together. In face of an increasing rate of extinction in lizards coupled to the global heating (Sinervo *et al.* 2010), we hope that such a paper could be a valuable contribution. We also hope to find the funding to continue our work on field activity temperatures and sex- and population differences in activity patterns to increase our understanding of how changes in the available thermal niche affects this species.

### **References:**

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