

The Society for Integrative and Comparative Biology



with the

**American
Microscopical
Society**

and

**The Crustacean
Society**

Final Program
Oregon Convention Center
Portland, Oregon
3-7 January 2016

- P3-80** Voisinet MP, Vasquez MC, Elowe C, Crocker DE, Tomanek L; California Polytechnic State University, San Luis Obispo, Sanoma State University Proteomic response of elephant seal pups, *Mirounga angustirostris*, to prolonged fasting.
- P3-81** Schmidt JE, Sirman AE, Clark ME, Reed WL, Heidinger BJ; North Dakota State University Telomere length correlations of somatic tissues in juvenile Franklin's gulls
- P3-82** Rahe CE, Neptune TC, Bouchard SS; Otterbein University Metabolic plasticity in red-eyed treefrog larvae
- P3-83** Neptune TC, Rahe CE, Bouchard SS; Otterbein University Predation risk in the face of limited resources: nutritional plasticity in red-eyed treefrogs
- P3-84** Usher CR, Bouchard SS; Otterbein University Growth rate plasticity in larval leopard frogs

Digestive Physiology

- P3-85** Leigh SC, German DP; University of California, Irvine The resource acquisition strategies of seagrass-eating bonnethead sharks
- P3-86** Rott KH, Caviedes-Vidal E, Karasov WH; University of Wisconsin, Madison, Universidad Nacional San Luis & CONICET Intestinal enzyme activity in nestling house sparrows (*Passer domesticus*) not depressed by high dietary lipid content
- P3-87** Nguyen-Phuc BQ, Stewart S, Demetropoulos C, German DP; University of California, Irvine, Southwest Aquatic and Terrestrial Consulting, Thousand Oaks Understanding the digestive physiology of the herbivorous sucker *Catostomus santaanae*
- P3-88** Wehrle BA, Nguyen-Phuc BQ, Dang RK, Krajnovic M, Tadic Z, Herrel A, German DP; University of California, Irvine, University of Zagreb, CNRS/MNHN Seasonal and sex effects on the digestive physiology of a newly herbivorous lizard
- P3-89** Plakke MS, Goetz BJ, Meslin C, Clark NL, Morehouse NI; University of Pittsburgh Stomachs in your butterfly: exploring the identity and specificity of proteases in the reproductive tract of female butterflies
- P3-90** Secor SM, Andrew AL, Castoe TA; University of Alabama, University of Texas at Arlington Single-cell RNAseq differentiates gene expression among cell types of the small intestine
- P3-91** Smoot SC, Smith CR, Halanych KM; Auburn University, University of Hawaii at Manoa Gene expression of cellulose degradation in the wood-eating bivalve *Xylophaga washingtona*

Cardiovascular and Respiratory

- P3-92** Reeve CM, Onthank KL; Walla Walla University Can octopuses breathe air?
- P3-93** Webber MA, Ivanov BM, Johnson MA; Trinity University Interspecific variation in blood physiology in Caribbean *Anolis* lizards
- P3-94** Hou JJ, Cornell A, Williams TD; Simon Fraser University How developmentally mature are chicks at fledging? Variation in hematology during a critical life-history transition
- P3-95** Marshall H, Bernal D, Skomal G, Richard B, Bushnell P, Whitney N; Mote Marine Laboratory, University of Massachusetts Dartmouth, Massachusetts Division of Marine Fisheries, National Marine Fisheries Service, NOAA, Indiana University South Bend Blood stress physiology parameters and mortality rates of sharks after commercial longline capture

Terrestrial Locomotion

- P3-96** Tennett KA, Costa DP, Fish FE; West Chester University, University of California, Santa Cruz Kinematics of terrestrial locomotion of northern elephant seals
- P3-97** Polet DT, Hasaneini SJ, Bertram JEA; University of Calgary Quadrupedal walking revisited: energy minimization strategy of walking dogs
- P3-98** Harvey RJ, Roskilly K, Hubel TY, Evans HE, Wilson AM; Royal Veterinary College A snapshot of the domestic cat's daily life in different environments
- P3-99** Minicozzi M, Finden A, Gibb A; Northern Arizona University Are there performance tradeoffs in the ability to perform the aquatic C-start and terrestrial tail-flip jump in killifishes?
- P3-100** Usherwood JR, McGowan CP; The Royal Veterinary College, London, University of Idaho An energetic account for the higher prevalence of bipedal hopping versus running among smaller animals using intermittent or fast gaits

Seasonal and sex effects on the digestive physiology of a newly herbivorous lizard

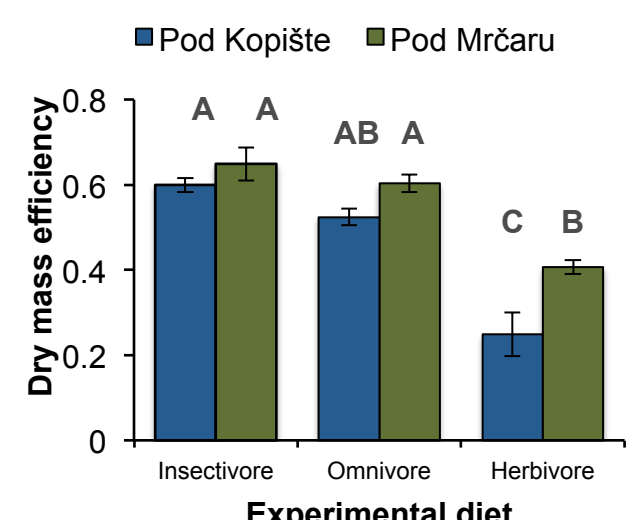
Beck A. Wehrle¹, Bao-Quang Nguyen-Phuc¹, Robert K. Dang¹, Zoran Tadić², Marija Kranjović², Anthony Herrel³, & Donovan P. German¹

¹ University of California, Irvine; ² University of Zagreb; ³ CNRS/Muséum National d'Histoire Naturelle, Paris

Abstract
Few studies of diet incorporate analyses of what an animal is digesting. Knowing what an animal actually digests allows us to understand if its physiology and morphology are optimized for its nutritional source. A population of Italian Wall Lizards (*Podarcis sicula*) in Croatia has become primarily herbivorous and morphologically distinct from its source population in ~30 generations, making it a compelling example of rapid evolution. To characterize the changes that occur on this short timescale, we compared gut structure and enzyme activity across populations of males in spring and summer, and in females in spring. Our previous work documented few biochemical and histological changes in the herbivorous population's guts from male lizards collected in summer. Despite these similarities, the herbivorous population had a higher digestive efficiency of plants than their source counterparts. Stable isotope analyses revealed seasonal dietary differences, but also that the "herbivorous" population had elevated $\delta^{15}\text{N}$ values in comparison to the source population. Although we found no differences in gut length among males, we did find differences in the cross-sectional surface area in some gut regions. Females from the herbivorous population had longer guts and we identified qualitative differences in their gut structure. As such, we expected to find greater differences in gut enzyme activities across seasons and in females. We found no seasonal differences in enzyme activity, but did find higher pancreatic amylase activity in "herbivorous" females. Thus, sex and population each appear to affect the strategy each group uses to accommodate their diets.

System

Podarcis sicula experimentally moved from Pod Kopište to Pod Mrčaru in 1971. In <30 generations Pod Mrčaru found to feed mainly on plants while the Pod Kopište population remained primarily insectivorous.†



Pod Mrčaru population:

- ↑ bite force
- nematodes in their hindguts
- hindgut chambers
- ↑ digestive efficiency of plants

Digestive efficiency: mean ± SEM, N=5, ANOVA_{population}: $F_{1,24}=13.49$, $P=0.001$, ANOVA_{diet}: $F_{2,24}=52.64$, $P<<0.001$.

Diet

Pod Mrčaru lizards consume more plant material:

- than Pod Kopište lizards†
- in summer†

No sex differences for stomach contents†

¹³C and ¹⁵N isotopic analyses of tissue from males in summer

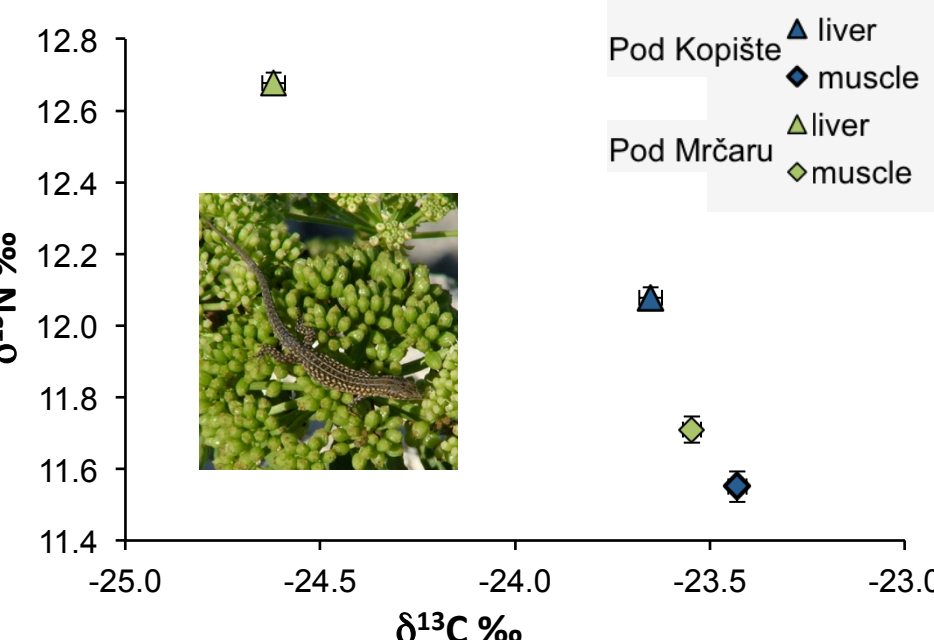
Stable isotope analyses confirm seasonal differences in assimilated diet

- ✗ Pod Kopište population
- ✓ Pod Mrčaru population

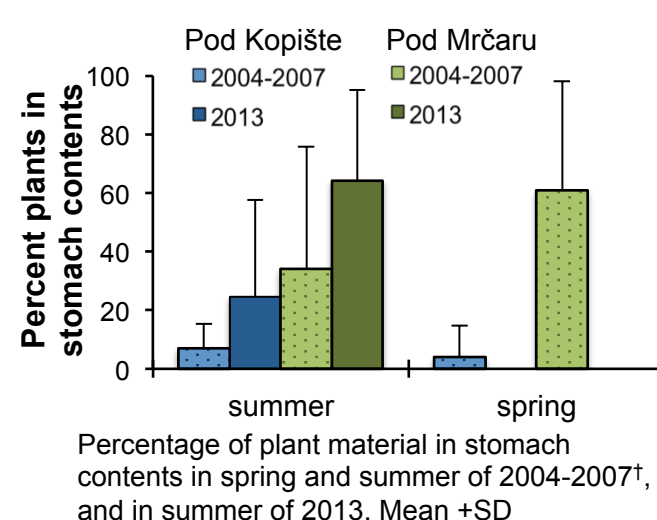
Pod Mrčaru population short term isotope signatures (liver: ~20 days[§]):

- ↑ δN^{15} ($P=0.035$)
 - unusual for an herbivore to be more N^{15} enriched
- ↓ δC^{13} ($P=0.002$)
 - more similar to carbon signature of cellulose (-25‰)

No differences in long term isotope signatures (muscle: ~80 days[§])
 $P_{15\text{N}}=0.56$, $P_{13\text{C}}=0.52$



Carbon¹³ and nitrogen¹⁵ isotopic signatures of *P. sicula* liver and muscle tissues in summer 2013, Mean ±SEM N=7, Analyzed with two-sample t-tests.



Percentage of plant material in stomach contents in spring and summer of 2004-2007†, and in summer of 2013. Mean +SD

Gut Length

Hypotheses:

Pod Mrčaru lizards will have longer guts than:

- guts of Pod Kopište lizards
- in summer than in spring
- No differences in gut length by sex

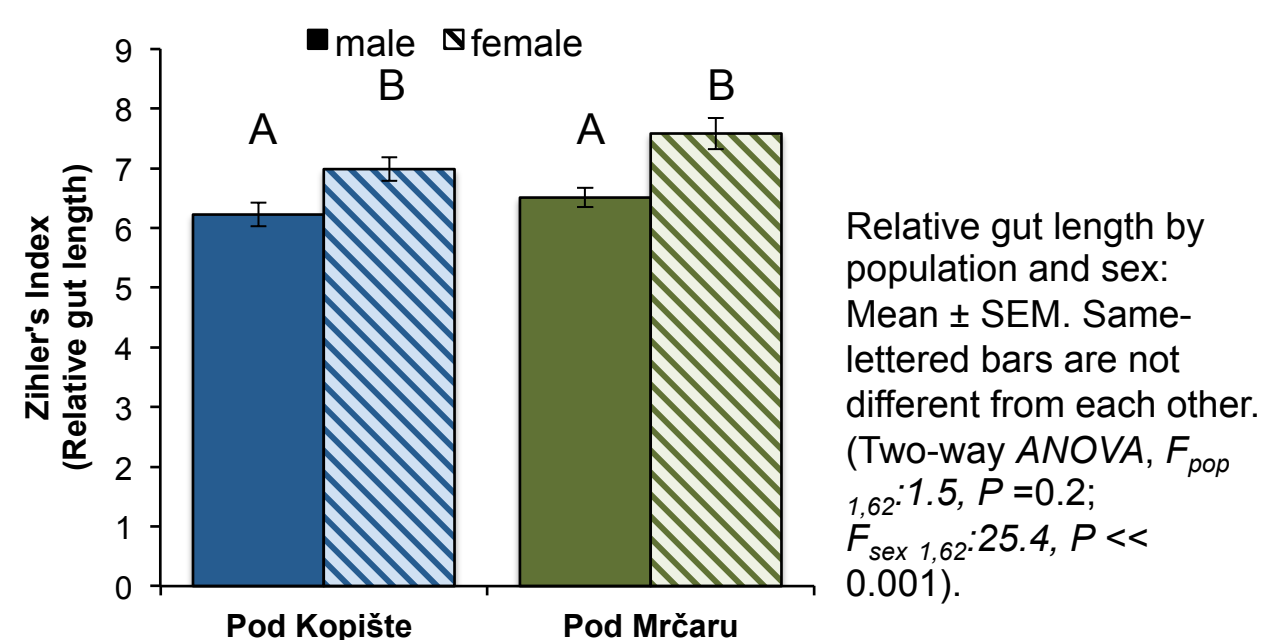
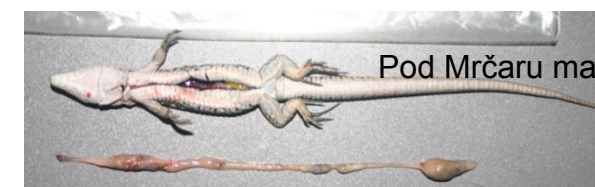
Results

- No population differences
- No seasonal differences
- Females have longer guts ($P=2.4 \times 10^{-6}$)

Methods

We dissected male and female *P. sicula* from both islands in spring and summer and measured:

- total mass
- snout vent length
- gut length



Relative gut length by population and sex: Mean ± SEM. Same-lettered bars are not different from each other. (Two-way ANOVA, F_{pop} : 1.62 ; $P=0.2$; F_{sex} : 1.62 ; $P=0.25$; $P<<0.001$).

Gut Structure

Hypotheses:

Pod Mrčaru lizards will have an increased cross sectional surface area:

- compared to Pod Kopište lizards
- in summer vs. spring
- No differences in surface area by sex

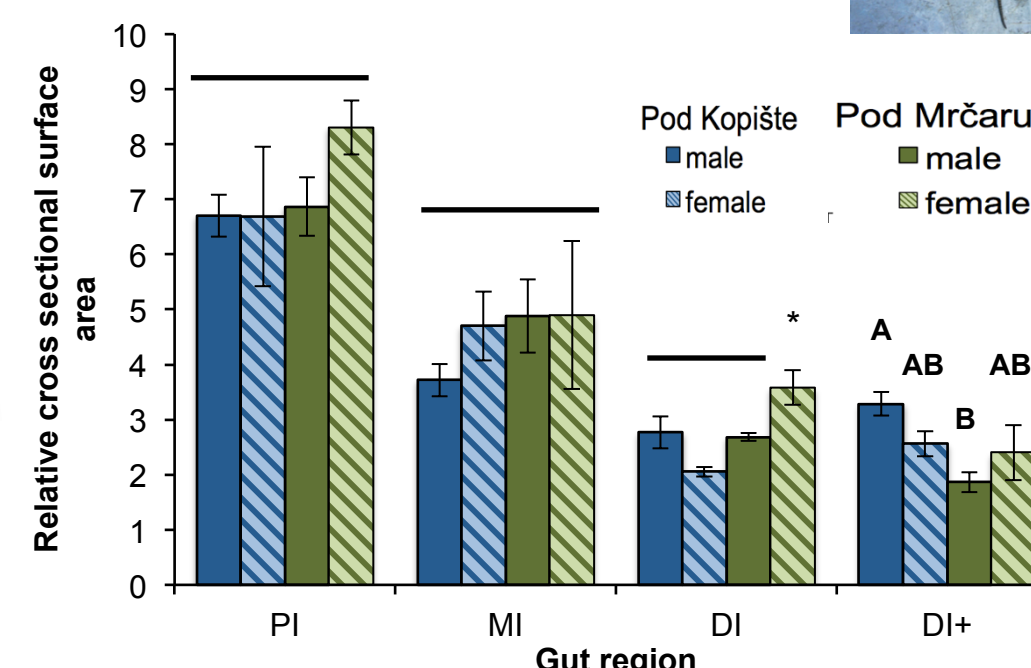
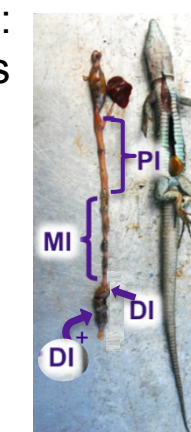
Results

- DI: Pod Mrčaru female
 - > Pod Mrčaru male ($P=0.014$)
 - > Pod Kopište female ($P=0.019$)
- DI+: Pod Kopište male
 - > Pod Mrčaru male ($P=5.98 \times 10^{-4}$)
- No seasonal differences

Methods

Divided guts from males (summer: $N=5$; spring: $N=3$) and females (spring: $N=3$) of both islands into proximal intestine (PI), mid intestine (MI), distal intestine sections (DI, DI+)

Histology: measured ratio of mucosa to serosa perimeters



Perimeter of mucosa × perimeter of serosa⁻¹ in proximal intestine (PI), mid intestine (MI), distal intestine (DI), and deep distal intestine (DI+) by population and sex. Same-lettered bars and bars under the same line are not different from each other. Mean ± SEM. (DI: Two-way ANOVA, $F_{pop;sex}$: 1.14 ; $P=0.002$; DI+: Two-way ANOVA, F_{pop} : 1.16 ; $P<<0.001$, $F_{pop;sex}$: 4.9 , $P<0.05$).

Enzyme Activities

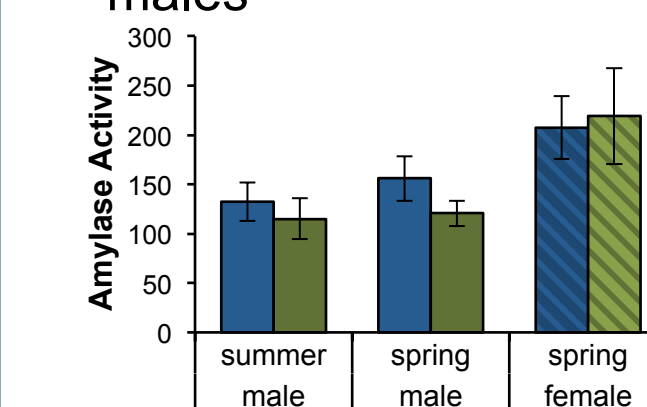
Hypotheses:

Pod Mrčaru lizards will have ↑ amylase activity

- than Pod Kopište lizards
- in summer
- No differences in aminopeptidase activity
- No differences in enzyme activity by sex

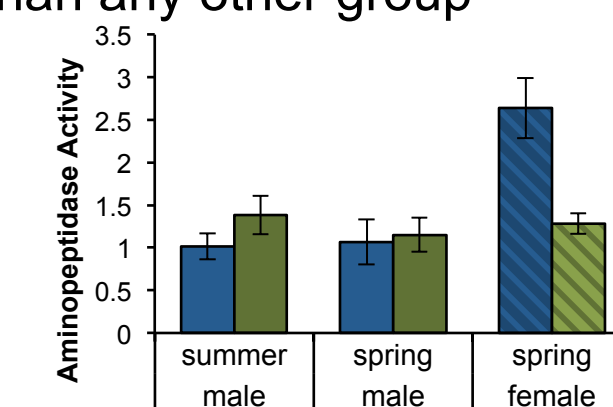
Results

- Amylase activity ↑ in Pod Mrčaru females than in males



Pancreatic amylase activity in nmol of sugar liberated $\text{g}^{-1} \text{min}^{-1}$ by Mean ± SEM. (Pod Mrčaru male:female ANOVA, $F_{1,14}=6.636$, $P=0.002$)

- Pod Kopište female aminopeptidase activity 2x ↑ than any other group



Proximal intestinal aminopeptidase activity in nmol of p-nitroaniline liberated $\text{g}^{-1} \text{min}^{-1}$. Mean ± SEM. (Two-way ANOVA, $F_{pop;sex}$: 17.265 , $P<<0.001$)

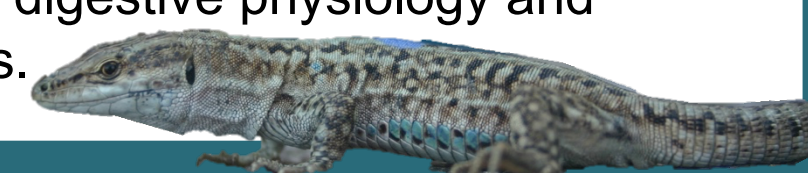
- No seasonal differences in either enzyme

Conclusions

Despite seasonal differences in diet, we found no seasonal differences in digestive physiology.

Although Herrel et al. 2008 found no differences in diet between males and females, we found considerable sex differences in gut form and function. Thus, males and females appear to employ different strategies to meet their nutritional goals. The hindgut difference between Pod Kopište and Pod Mrčaru males suggests microbiome divergence may be key in explaining performance differences in males.

In addition to the hindgut valves discovered by Herrel et al., we have now identified differences in digestive physiology and anatomy throughout the intestines.



†Herrel, A. et al. 2008. Rapid large-scale evolutionary divergence in morphology and performance associated with exploitation of a different dietary resource. PNAS 105:4792-4795.

§Warne, R. et al. 2010. Tissue-carbon incorporation rates in lizards: Implications for ecological studies using stable isotopes in terrestrial ectotherms. PLoS ONE 5: e11717.

Acknowledgements

This project was primarily funded by NSF CNIC grant 1318059. Additional funding was provided by the Company of Biologists, Sigma Xi, and UC Irvine. We thank Park Prirode Lastovsko Otočje and University of Zagreb for their logistical support. Thanks to the following people for field assistance: Nathan Gold, Parth Jhaveri, Martina Ratko, Kyle Chernoff, Iva Salamon, Barbara Horvatović, and Maxime Tavernier. UC Irvine approved IACUC protocol: 2013-3096-0

