Consequences of Disturbed Landscapes To Desert Tortoises Populations When Habitats Are Not Restored or Protected

Kristina Drake*
Todd Esque
Ken Nussear
Lesley DeFalco
Phil Medica
Melia Nafus
Wildfire Impacts on Tortoises & Their Habitat

- Loss of perennial plants
- Shifts in vegetation (Native -> Invasive Grasses)
- Emigration of tortoises
- Lower growth and reproductive output
Do Tortoises Use Burned Habitats?
-Habitat Use, Movement, Reproduction, Health, & Survival

10-Year Study (2006-Present)

- \( n = 54 \) resident adult tortoises

**Observation Counts**
- \( N = 19,456 \)
- 51% Unburned, 49% burned

**Home-range**
- Varied (3.4 ha to 314.6 ha)
- 45% in Burned Habitat

Drake/Esque Unpublished Data
Adult Tortoise Movement into Burned Habitat

Reproduction, Health, & Survival in Adult Tortoises

Burned habitat did not influence
- Reproduction ($Z=1.32$, $p = 0.19$)
- Health
- Survival ($Z=-1.33$, $p = 0.18$)

# Eggs/ Clutches per Year
- Animal Size
- Habitat Type (Unburn / Burn)
  - Movement
  - Home-Range
  - Observations
  - Plant Cover
  - Annual Biomass

Drake et al. 2015 JWM 79(4):618-629
Juvenile Tortoises in Burned Habitat

Pilot Study
- Translocated 19 juveniles (unburned / burned habitats)

-Mortality
Unburned (2/9)  22%
Burned (8/10)  80%

Esque et al. Unpublished Data
Impacts of Invasive *Bromus* Grasses

- **Wildfire Regimes**
- **Community Dynamics**
- **Soil Stability**
- **Shrub Establishment**

*Bromus rubens* (red brome)
Mediterranean Grasses Impact Tortoises

1. Dietary / Nutritional Changes
2. Mucosal (mechanical) Damage

Medica & Eckert 2007
Plant Diets

Native Grass
Festuca octoflora

Native Forbs
Mixture

Invasive Grass
Bromus rubens

-K. Drake
Experiment
(April - July)

Annual Plant Diets
1. Brome (B) *Bromus rubens* – Invasive Grass
2. Festuca (Fe) *Festuca octoflora* – Native Grass
3. Forbs (F) – Mix of Native Forbs*
4. Brome & Forbs (BF)
5. Festuca & Forbs (FeF)

*Native Annual Forbs = Brown-eyed evening primrose (*Cammisonia claviformis*), CA poppy (*Eschscholzia californica*), Desert dandelion (*Malacothrix californica*), Indian wheat (*Plantago ovata*)

100 Juvenile Tortoises (20 per Diet)
Plant Nutrients

- Brome
- Festuca
- Forbs

- Ca:P
- Na
- Cl
- ASH
- N
- P
- Cu
- Zn
- Mg
- ADF
- NDF
- Fat
- K
- Mo
- S
- CP
- Ca
- Moisture
- Ca:P
- NSC

June & July

P ≤ 0.05

Drake et al. 2016 Ecosphere
Growth

Cumulative Plastron Growth (mm)

- **Metrics**

Diet

- **(F4,93 = 7.6, p < 0.01)**

Month

- **(F3,275 = 29.3, p < 0.01)**

Size

- **(F1,93 = 1.8, p = 0.18)**

Size = 0.71 ± 0.04 mm per Month

Range = -1.33 to 3.44 mm

Survival

- **Diet**

- **(Z = 9.86, p < 0.01)**

- **Month**

- **(Z = -3.70, p < 0.01)**

Brome 37%

Festuca 32%

Brome & Forbs 20%

Festuca & Forbs 5%

Forbs 0%

Mortality

Drake et al. 2016 Ecosphere
Health and Clinical Condition
Impacts from Grass-Only Diets

1. Body Condition Declined
   - Brome & Festuca Diets ($p = 0.03$)
   - June & July ($p < 0.01$)

2. Muscle Mass & Fat Loss
   - Brome & Festuca Diets ($p < 0.01$)

3. Fecal Staining (Diarrhea)
   - Festuca ($p < 0.01$) 6 (32%)
   - Brome 3 (16%)
   - Brome & Forbs 2 (12%)
   - Forbs 0

4. Shell Thinning & Softness
   - Brome & Festuca ($p = 0.04$)
Health and Clinical Condition
Impacts from Invasive Grass Diets

5. Impacted *Bromus* Seeds
- Inflammation (28%)
  - May
    - # (% in Diet)
      - Brome & Forbs 3 (15%)
      - Brome 4 (21%)
  - June
    - Brome & Forbs 1 (5%)
    - Brome 4 (21%)
  - July
    - Brome & Forbs 8 (44%)
    - Brome 13 (93%)

6. Pale Mucosa
- Grass Diets (p = 0.06)
- Anemia?
Immunosuppressive Effects of Malnutrition

- Overall health declines
- Organ change (morphology & function)
- Limited growth & reproduction
- Defective wound healing
- Increased infection, disease, & cancer
- Shorter life-spans
- Increased mortality

**Immune & Cellular Function**

- Inflammatory Cytokines Interference
  - Cell Signaling & Modulations
  - Humoral & Cell-based Immune Responses (IL-1β, IL-2, IL-6, and TNF-α)

**Energy**  
**Iron**  
**Vitamin A & D**  
**Zinc**  
**Pryiodoxine**
Gene-Based Health Diagnostics

Immune Functions in Reptiles

Gene Transcription

Stressor (Nutrition/Pathogen) Registered by Organism

DNA – cell detects problem, sends message to produce proteins

messenger RNA (Measured using RT PCR)

functional proteins produced to combat problem
Quantitative Real-Time PCR Assay

Gene Transcription
- Immune Function
- Nutritional and Thermal Stress
- Xenobiotic Metabolism
- Calcium Uptake
- Shell Formation
- Tumorigenesis

Identify key genes & immune pathways
When and why are they changing?

Leptin (LEP)
Nutritional/Neuroendocrine/Immune Status

Calmodulin (CaM)
Cellular Activity, Calcium Metabolism, Cell Division

Superoxide Dismutase (SOD)
Antioxidant Defense/Inflammation

Arylhydrocarbon (AHR)
Environmental Toxicants

Cathepsin L (CL)
Antigen Processing/Protein Synthesis/Growth

Mx1
Viral Infection

Heat Shock Protein (HSP70)
Molecular Chaperone/Detox

Serum Amyloid A (SAA)
Proinflammatory

CD9
Molecular Facilitator/Bacterial Infection

ATF
Inflammation Signals/Bacterial Infection

Housekeeping (S18)
Reference Ribosomal Gene

Myeloid Diff. Factor (MyD88)
Thermal/Wide Range Stress/Detox
Blood Sample (0.01 mL)
- March
- July

Genes of Interest
- SAA
- CD9
- Mx1
- HSP
- SOD
- ATF
- AHR
- MyD88
- CL
- CaM
- Lep

Normalized CT Values

Housekeeping (S18) Reference Ribosomal Gene

Total [RNA] = Total Amount of Expressed mRNA
Immune & Physiological Functions

Gene $C_T$ Values

<table>
<thead>
<tr>
<th>Gene</th>
<th>Native Forage</th>
<th>Invasive Grass</th>
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</thead>
<tbody>
<tr>
<td>CaM**</td>
<td>6.78 – 13.92</td>
<td>12.17</td>
</tr>
<tr>
<td>CL*</td>
<td>12.08 – 21.43</td>
<td>18.60</td>
</tr>
<tr>
<td>Lep**</td>
<td>11.85 – 21.69</td>
<td>14.35</td>
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<tr>
<td>RNACon**</td>
<td>5.20 – 54.80</td>
<td>12.98</td>
</tr>
</tbody>
</table>

- ** Indicates $P \leq 0.05$
- * Indicates $P \leq 0.09$

Calcium metabolism, Shell formation, Growth & protein synthesis, Neuroendocrine & Immune functions

Gene expression was assessed by RNAseq analysis. For MANOVA comparisons:
- March ($F_{4,23} = 0.30, p = 0.99$)
- July ($F_{4,22} = 3.09, p = 0.01$)
- Jul - Mar ($\Delta$, $F_{4,22} = 1.97, p = 0.05$)
Similarity Profile Analysis
SIMPROF
(5 Distinct Clusters)
$P \leq 0.05$
Conservation Implications

**B. Rubens reduces**
- Fitness & Survival for Juvenile Tortoises
- Survival & Recruitment Rates Throughout Range

Disease & Health Models

*Drought, Winter Precip, PDO*
Gene-Based Diagnostics

- Improve Assessments
- Identify Latent Problems
- Restoration Efficacy
- Disease Models
- Land-use Patterns
- Climate Change

Habitat Quality
- Quantitative measure
- Conservation & Management priorities
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Drake et al. 2016- Ecosphere (online)

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-K. Drake