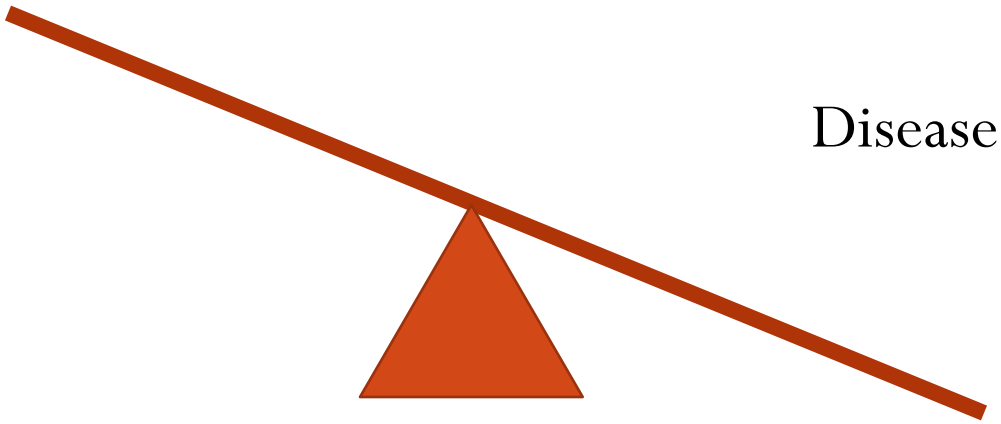
Disease Management in Wild Animal Populations

Nadine Lamberski, DVM, Dipl. ACZM
Corporate Director of Animal Health
San Diego Zoo Global

Health status is a dynamic process

Absolute Health

Relative health



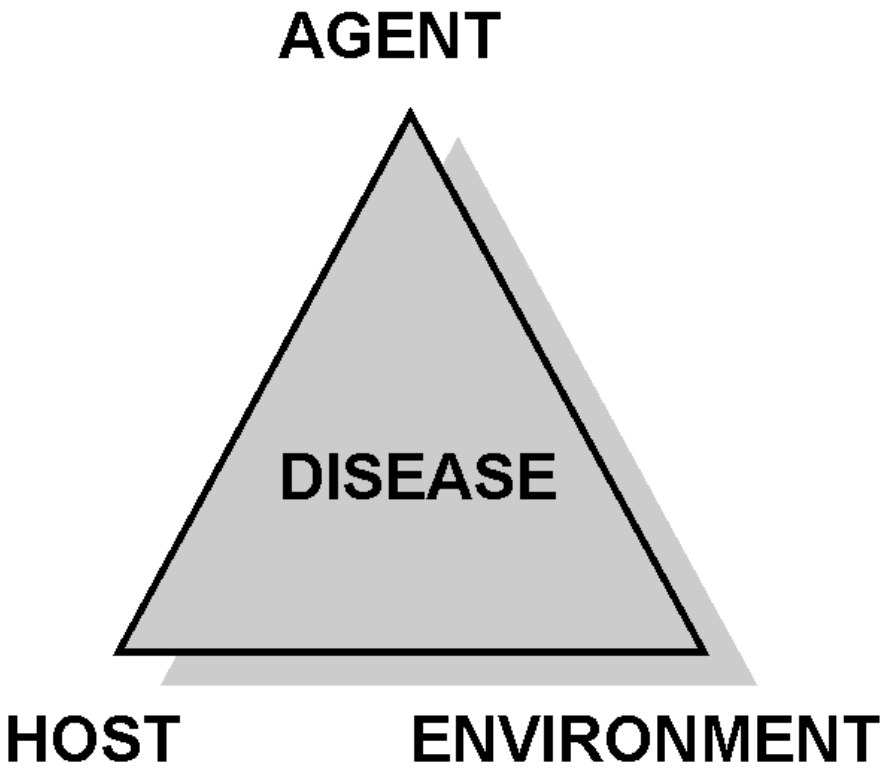
Disease

Death

Epidemiologic Triad

Disease is the result of forces within a dynamic system consisting of:

- agent of infection
- host
- environment



Agent

- Dose
- Pathogenicity
- Virulence
- Mutations
- Strains
- Genome sequence

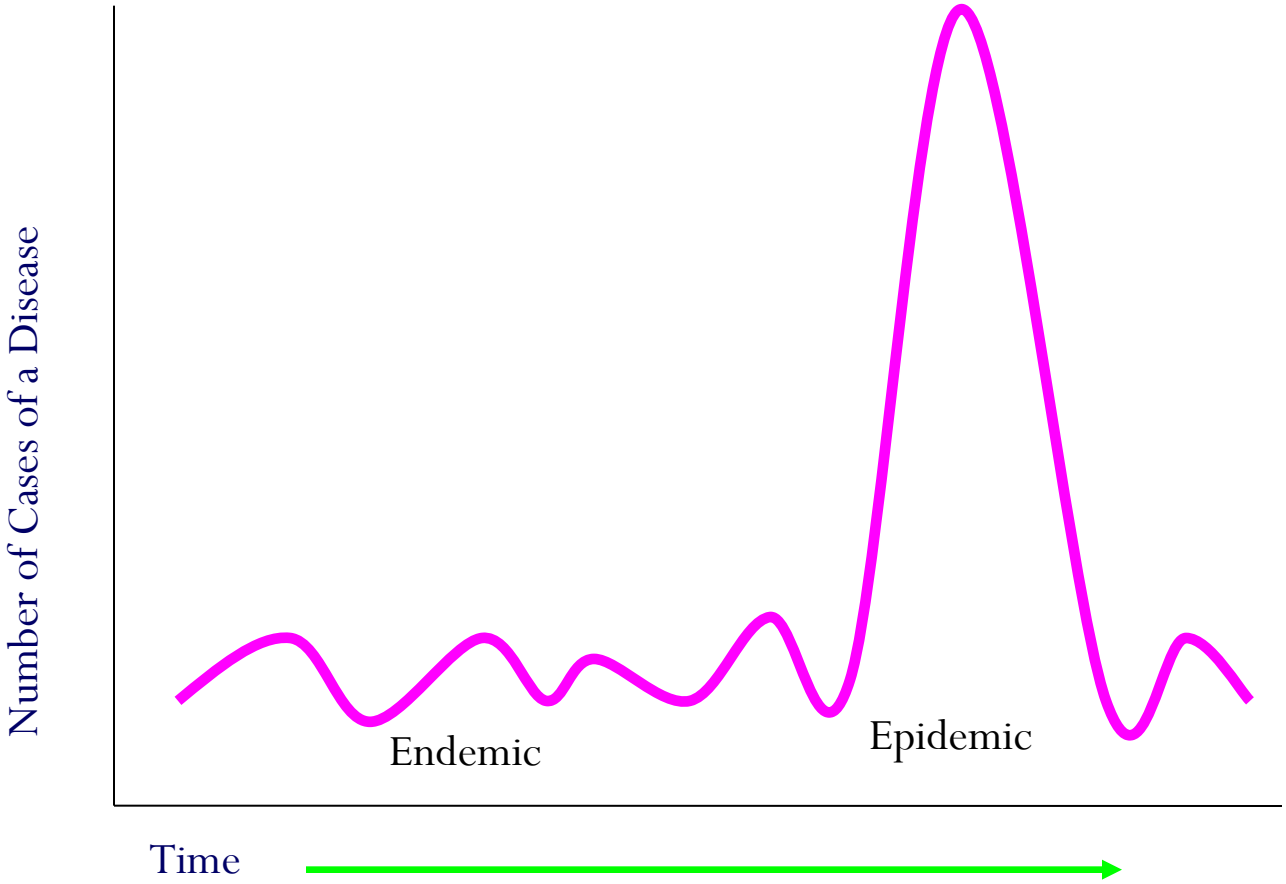
Host

- Genetics
- Immune response
- Immune competence
- Co-infections
- Nutrition
- Other stressors

Environment

- Drought
- Invasive species
- Fragmentation
- Lack of corridors
- Pollution
- Toxins
- Predator prevalence
- Climate change

Endemic vs Epidemic



Mycoplasmosis in desert tortoises

- 1980 – URTD in captive desert tortosies described
- 1988 – URTD seen in wild tortoises in Kern County
- 1989 – detailed survey showed 43% of 468 tortoises showed signs of URTD, 25% of 162 tracked tortoises died within a year
- 1990 – listed as Threatened
- 1991 - Jacobson, et described chronic upper respiratory tract disease
- 1993 – ELISA test developed
- 1994 – *Mycoplasma agassizii* identified as causative agent
- 1997 – clinical signs and antibody production
- 1999 – seroepidemiology
- 2001 – guidelines for field evaluation
- 2002 – management recommendations, diagnostic dilemma, incomplete picture

Mycoplasma spp. are ubiquitous

- *M. alligatoris* – acute, lethal disease in alligators
- *M. crocodyli* – mild disease in crocodiles
- *M. gallisepticum* – outbreaks in wild birds originated from poultry
- *M. ovipneumoniae* – epizootics in bighorn sheep
- *M. agalactiae* – mortality in ibex in French Alps

Key Concepts - Mycoplasmosis

Desert Tortoises	Other species
• Chronic disease	✓ Same
• Progress slowly	✓ Same
• Can have acute outbreaks	✓ Same
• Multi-factorial	✓ Same
• Immune response variable	✓ Same
• Host response influences lesion development	✓ Same
• Predisposing factors	✓ Same
• Asymptomatic carriers	✓ Same

Epizootiology/Epidemiology

- The study of disease patterns within animal populations
- Patterns lead to predictions
- Predictions can drive decision-making while data is being accumulated and can prevent inertia
- Data drives the iterative process
- Data can mislead, epizootiology does not

Areas of research and development

- Agent
  - Genome sequencing
  - Molecular diagnostics
- Host
  - Immune function
  - Gene transcription
  - Post mortem evaluation
  - Clinical assessment
  - Therapeutics
  - Vaccination?
- Environment
  - Ecologic studies
  - Management practices

Health Assessment Training

- 114 students took class
- 9 classes since 2011 (Las Vegas and Phoenix)
- Handling, biosecurity, recognition of clinical signs, BCS, sample collection, sample processing
- Consistent data collection
- 41 (36%) passed 1st attempt
- 27 obtained certification later
- 68 total certified to date
- 8 failed the class on their first attempt

BCS Validation

- Visual body condition appraisal system
- Cheap and easy tool
- Immediate results
- Study complete to compare to relative liver weight (RLW) and Condition Index (CI)
- In process – inter-rater reliability study
- Database will allow for population level monitoring (3 solar sites, approx. 200 tortoises)

Post mortem findings from 408 DTCC tortoises

- Rhinitis (276/373, 74%)
- Pneumonia (182/387, 47.0%)
- Glossitis (161/382, 42.7%)
- Tracheitis (150/363, 41.6%)
- Entero-/colitis(120/379, 31.7%)
- Hepatic vacuolation (110/387, 28.4%)
- Kerato-/conjunctivitis (81/339, 24%)
- Osteopenia (47/262, 17.9%)
- Nephritis (41/380, 10.5%)
- Non-inflammatory renal abnormalities (37/380, 10%)
- Hepatitis (30/387, 7.8%)
- Thinned/softened shell (144/408, 35%)
- Urolithiasis (111/408, 27.2%)

Agents identified

- Mycoplasma agassizii (61/97, 62.9%)
- Mycoplasma testudineum (3/97, 0.03%)
- Herpesviruses (15/408, 3.7%)
- Cryptosporidia (6/369, 1.6%)
- Nematodes (20/367, 5.4%)
- Sarcocysts (36/408, 8.8%)
- Fungi (5/408, 1.2%)

- Chlamydophila – 45% intestinal samples positive – included tortoises with and without enteritis

Treatment Trial

- Enrofloxacin reduced the number of *MyAg* organisms in all treated tortoises and reduced *MyAg* to undetectable in 23-33% of tortoises 1 week post treatment.
- Enrofloxacin reduced the number of days tortoises had nasal discharge 1 week post treatment.
- No weight loss
- Tulathromycin was not effective

Nasal Lavage Vs Oral Swabs

- Oral swabs can substitute nasal flushes for qPCR detection

- Paired nasal flush and oral swab samples (n = 396)
- Group 1: 162 live desert tortoises at the DTCC
- Group 2: 100 necropsied DTCC desert tortoises
- Group 3: 134 wild, live tortoises from a solar site

Oral Swabs recommended

- Procedure much simpler than nasal lavage
- Less handling time/stress
- Less expensive
- Less supplies
- Sample processing simpler
- Less litter in the desert
- Disadvantage – need to open the mouth
- Disadvantage – less recovery if very low prevalence

DOI: 10.7589/2015-09-253

Journal of Wildlife Diseases, 53(1), 2017, pp. 000–000
© Wildlife Disease Association 2017

COMPARISON OF CURRENT METHODS FOR THE DETECTION OF CHRONIC MYCOPLASMAL URTD IN WILD POPULATIONS OF THE MOJAVE DESERT TORTOISE (*GOPHERUS AGASSIZII*)

Franziska C. Sandmeier,[1,2,8] Chava L. Weitzman,[2] K. Nichole Maloney,[3] C. Richard Tracy,[2] Nathan Nieto,[4] Mike B. Teglas,[2] Kenneth W. Hunter,[5] Sally DuPré,[5] C. M. Gienger,[6] and Michael W. Tuma[7]

[1] Department of Biology, Department of Biology, Colorado State University – Pueblo, 2200 Bonforte Blvd., Pueblo, Colorado 81001, USA
[2] Ecology, Evolution, and Conservation Biology, University of Nevada, 1664 North Virginia St., Reno, Nevada 89557, USA
[3] Vanderbilt University, 2201 West End Ave., Nashville, Tennessee 37235, USA
[4] Department of Biological Sciences, Northern Arizona State University, South San Francisco St., Flagstaff, Arizona 86011, USA
[5] Department of Microbiology and Immunology, University of Nevada, 1664 North Virginia St., Reno, Nevada 89557, USA
[6] Department of Biology and Center of Excellence for Field Biology, Austin Peay State University, 601 College St., Clarksville, Tennessee 37044, USA
[7] Department of Biological Sciences, University of Southern California, 3551 Trousdale Pkwy., Los Angeles, California 90089, USA
[8] Corresponding author (email: franziska.sandmeier@csupueblo.edu)

Conclusions

- No single test should be used to diagnose mycoplasmosis in individual tortoises
- Antibody prevalence not significantly associated with the portion of the population that is PCR-positive in captive and wild tortoises
- Antibody response of free-living tortoises may be influenced by various environmental conditions
- qPCR shows most infected tortoises have low-intensity infections not associated with visible signs of disease

USGS Transmission Studies

- *M. agassizii* transmission is time-dependent requiring repeated interactions (5+ days of physical contact)
- Introducing individuals with high infection intensities into populations could increase *M. agassizii* transmission rates.
- Oral swabs analyzed by qPCR combined with visual assessment for clinical signs can be a useful method to estimate infection intensity
- Once infected, some tortoises present few or no clinical signs of disease but have high infection intensities.
- *M. agassizii can be* consistently documented in nasal swabs of tortoises without nasal discharge

- Immune system response and production of antibodies are very slow (>1 year)
- Clinical signs and shedding of *M. agassizii* from the nares was observed prior to seroconversion
- Visual assessments should be combined with both ELISA and qPCR of oral swabs and more than one assessment should be completed per tortoise to adequately assess infection and health status.
- Translocation impacts to tortoise contact patterns that might affect the spread of infections, e.g, by increasing contact rates or connectivity across large areas are being investigated

USGS Gene Expression Research

- Gene transcription panel – Used to identify intrinsic and extrinsic stressors affecting tortoise physiology
  - The mixing of native plant diets with the non-native grass *Bromus rubens* (Drake et al. 2016 in Ecosphere ecs2.1531) negatively influences the physiological ecology, immune-competence, health, growth, and survival of young tortoises
- Transcript profiling can be used to identify ill tortoises
  - Unhealthy tortoises increased molecular reactions for genes responding to environmental toxicants, oxidative stress, microbial infections, and malnutrition.

Will vaccination work?

- Presence of antibody does not appear to be protective in "naturally- vaccinated" tortoises
- Tortoises with positive ELISA titers still develop disease
- ELISA is a non-qualitative assay – detects both neutralizing and non-neutralizing antibody
- Neutralizing antibody defends a cell from an infectious agent by neutralizing any effect it has biologically

- Disease develops in the absence of neutralizing antibody
- Need to develop a Mycoplasma-neutralizing antibody test (complex, expensive)
- Identify strains that are neutralizing and use these for cross-protection
- A subneutralizing antibody response suggests an ineffective immune response

Why attempt management?

1. Wild animals are involved in a disease of humans (zoonotic disease)
   - 80% human infections from animal hosts
2. Wild animals are involved in a disease affecting domestic animals.
3. Disease is having a deleterious effect on one or more wild species

Managing diseases to benefit wild species

- Disease is an important potential risk in translocations
- Disease might imperil small or endangered populations
- Large disease outbreaks cause public concern
- Diseases may spread from domestic animals or humans with serious consequences for wild species

All forms of disease management consist of some combination of

- Reducing exposure of the animals to harmful factors
- And/or increasing the ability of the animals to resist or cope with the harmful effects caused by those factors

Objectives of management

1. PREVENT a disease from occurring in individual animals, in groups of animals in a particular area, or in an entire population
2. CONTROL a disease that is already present by reducing frequency of occurrence, severity of the effects, or to curtail spread
3. ERADICATE or eliminate an existing disease locally or on a larger geographic scale

Matrix of management options

	Agent	Host	Environment
Prevent			
Control			
Eradicate			

Disease Prevention

Agent

- Prevent introduction of novel agents into new areas
- Restrict movement of potentially infected animals by humans
- Test and treat before moving
- Quarantine after moving

Host

- Prevent exposure
- Disperse animals away from or prevent access to sites where disease is occurring
- Reduce population density
- Vaccination

Environment

- Public education and alteration of human activities

Disease Eradication

Agent

- Eradication of any disease is extremely difficult

Host

- Complete elimination of the existing animal population with subsequent restocking with disease-free animals

Environment

- Habitat manipulation

Disease Control

Agent

- Destroy the agent
- Disinfection
- Treatment

Host

- Reduce the level of exposure or increase degree of resistance
- Reduce the density of infected individuals
- Vaccination

Environment

- Sanitation and hygiene (provision of safe drinking water, adequate food, and shelter)
- Prevent or remove situations that artificially concentrate animals and that promote disease exchange
- Vector control

CONTROL

As long as the disease is present in a population, control actions must continue for as long as there is a need to reduce the effect of disease.

Management of disease requires a long-term commitment that may be difficult to sustain, particularly if management is successful and disease becomes less visible

Disease management practices that could help desert tortoises

Increase the ability of the host to handle effects of infection

- Habitat restoration - vegetation
- Corridors to reduce fragmentation
- Reduce exposure to toxic chemicals (petroleum, agrichemicals, heavy metals)
- Prevent habitat degradation – livestock grazing, ATV
- Eliminate invasive species – red brome
- Predator control – ravens, squirrels, carnivores
- Consistent health assessment protocols
- Controlled experimental translocations