Brucella: Science and Challenges

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USDA Animal Health Facilities and Expertise

State-of-the-art:

- ABSL2 and ABSL3 small and large animal facilities
- BSL3 Agriculture large animal facility

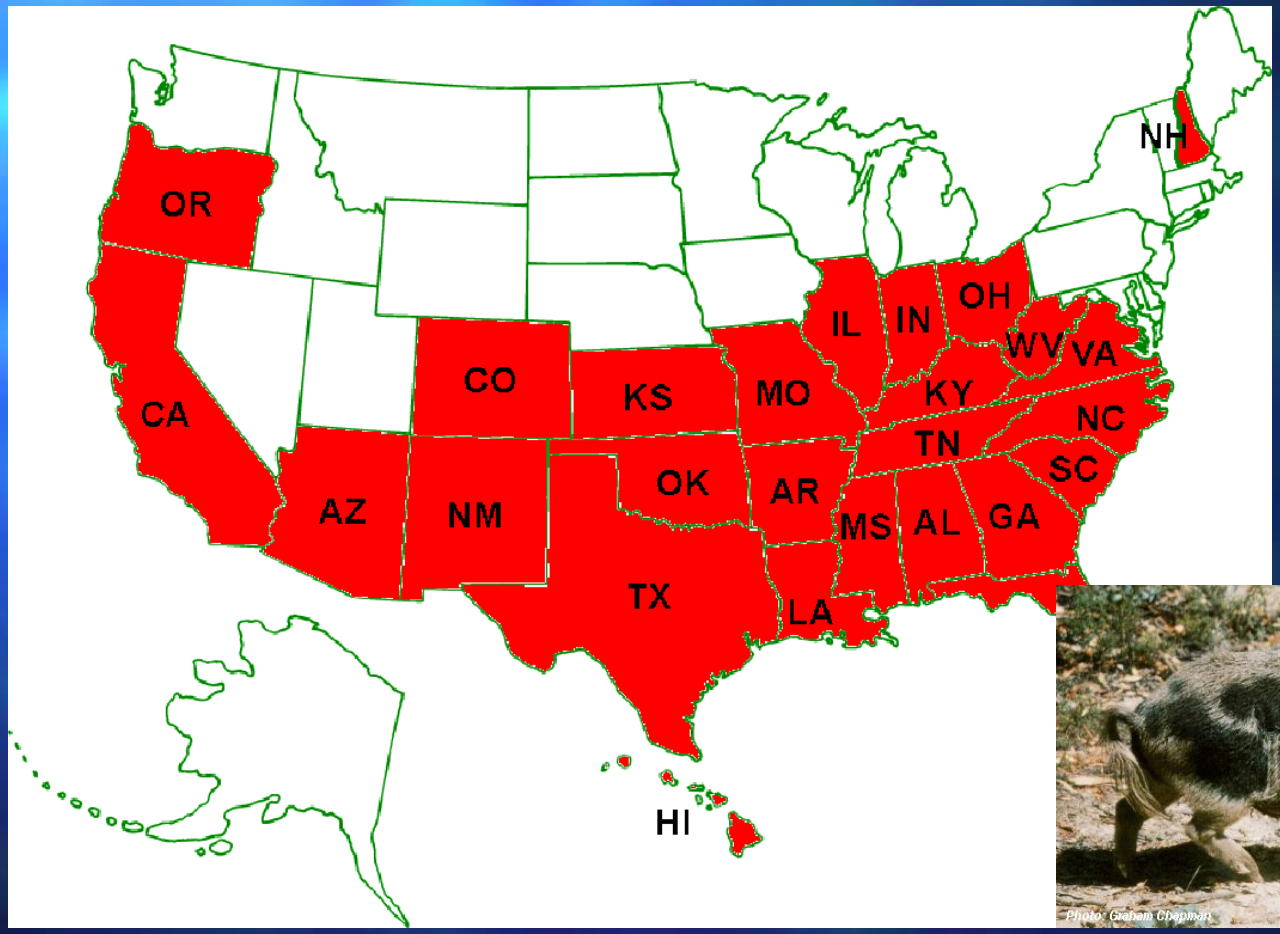
Animal care and facilities operations expertise that supports high-containment research in all major livestock and several wildlife species

Brucella

	Host	Vaccine
B. melitensis *	small ruminants	Rev1
*B. suis**	swine (cattle)	none
B. abortus *	cattle (swine)	RB51 or 19
*B. canis**	dogs	none
B. ovis	sheep	Rev1
B. neotomae	wood rat	none
Marine *Brucella**	marine mammals	none
B. inoptimata	human	none
B. microti	voles	none
Other *Brucella*	Austria foxes, African bullfrogs	

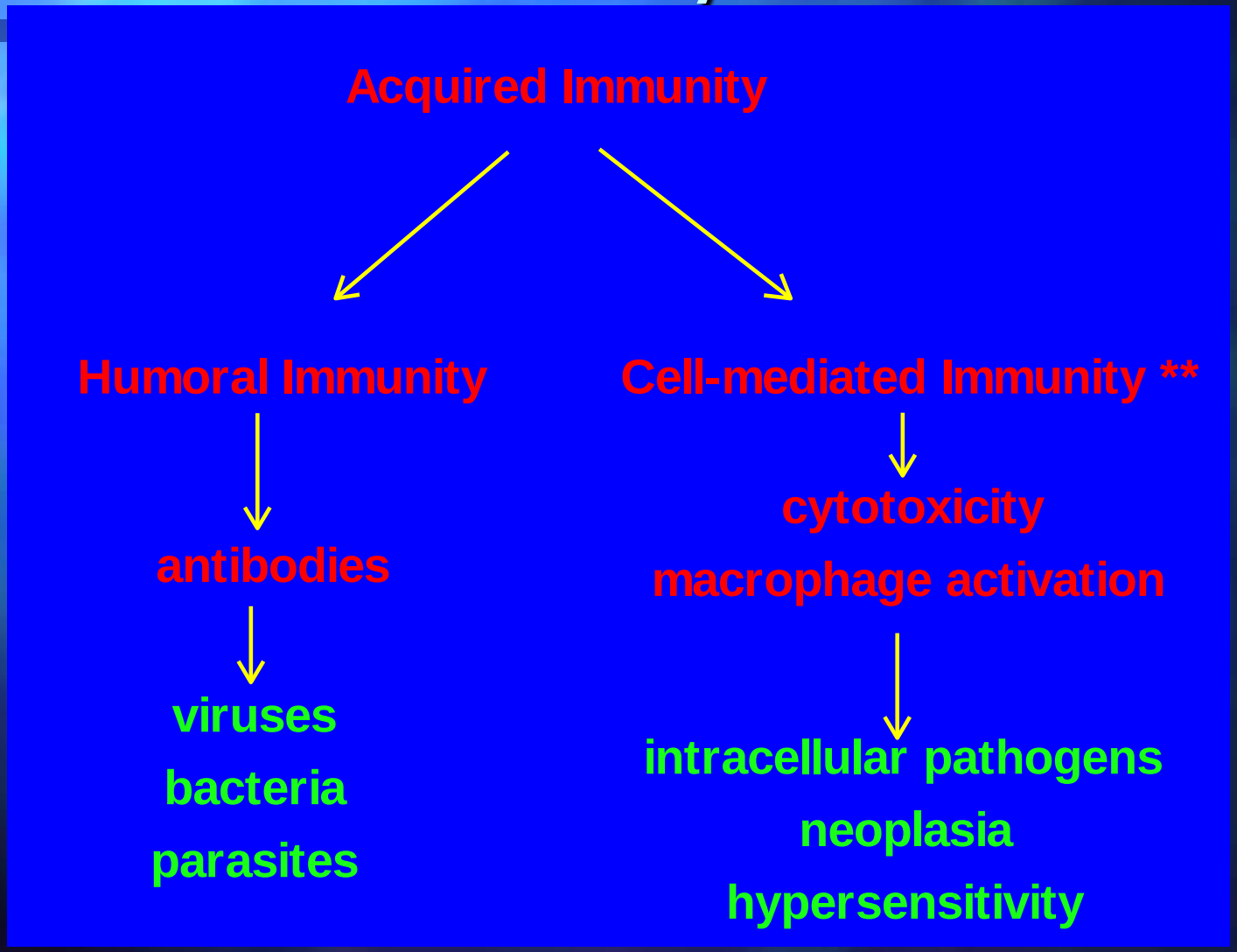
* Zoonotic

Distribution of Feral Swine in the US

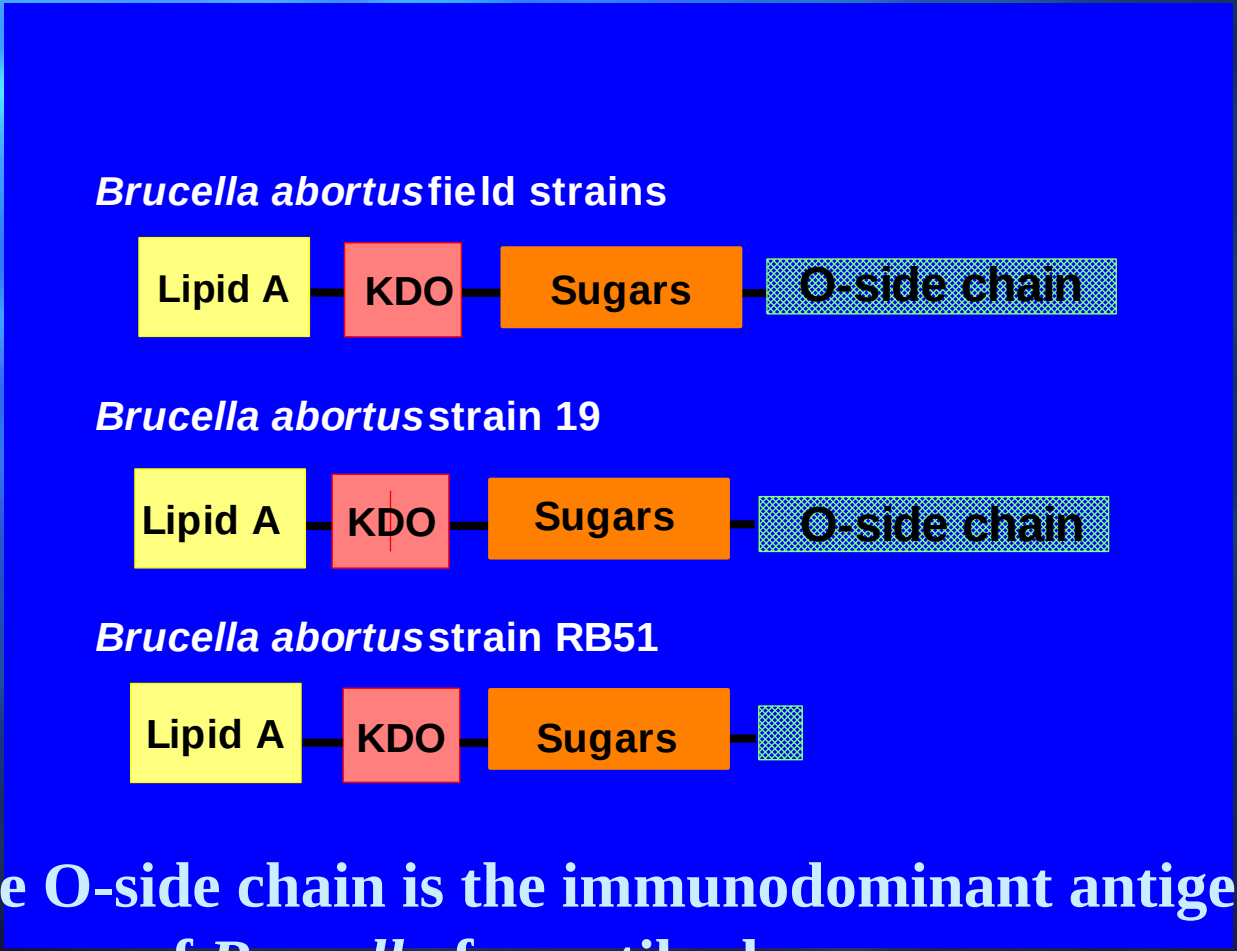


GPS mapping at http://128.192.20.53/infsms/

Protective Immunity against *Brucella*: Primarily Cell-mediated



Lipopolysaccharide structure of virulent and vaccine strains



The O-side chain is the immunodominant antigen of *Brucella* for antibody responses

Variance in Immunologic Responses between Ruminants

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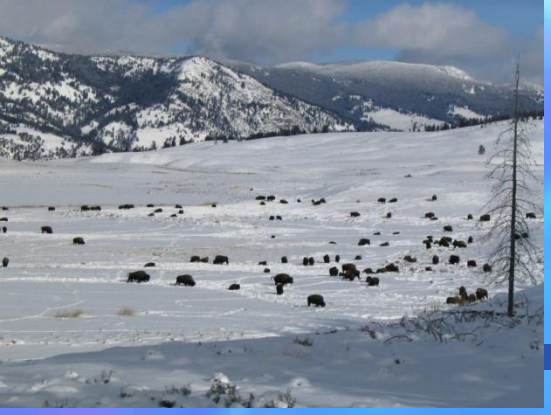

Brucella Vaccines

- **Vaccination alone will not eradicate brucellosis**
- Vaccines are very good at reducing transmission and clinical disease; very poor at preventing seroconversion or transient infection after exposure
- Long-term protection related to cell-mediated immunity
- Antibodies relatively unimportant for efficacy
- Many vaccine strains can be pathogenic in humans or pregnant animals

Comparing susceptibility to *Brucella* challenge

Species (Nonvaccinated)	N	Protection % protected (# aborted/infected / # challenged)		
		Abortion	**Fetal/Mam. Infection**	**Maternal Infection**
Cattle	**46**	**54% (21/46)**	**54% (21/46)**	39% **(28/47)***
Bison	**50**	**16% (42/50)**	**12% (44/50)**	**0% (50/50)**

Assessment of Vaccine Efficacy by Experimental Challenge

- Standardized method of vaccine evaluation
- Cattle challenge model developed in 1940's
- Evaluates all animals at most susceptible time (pregnant, end of second trimester) and receiving known infectious dose of virulent strain
- Field efficacy usually higher (not all pregnant, not all exposed, not all receive infectious dose) but other factors (nutrition, stress) may influence efficacy

Efficacy of RB51 as a Calfhood Vaccine for Cattle

Age at Vaccination	Protection from abortion % protected (# aborted/ #challenged)		
	RB51	Strain 19	Control
10 months	100% (0/20)	100% (0/6)	45% (6/11)
7 months	100% (0/22)	100% (0/5)	63% (4/11)
5-6 months	92% (2/25)	100% (0/4)	57% (6/14)
3 months	87% (2/15)	100% (0/4)	50% (5/10)
Overall	95% (4/82)	100% (0/19)	54% (21/46)

Efficacy of RB51 in Bison Overall Data

Treatment	N	Protection % protected (# aborted/infected / # challenged)		
		Abortion	Fetal/Mam. Infection	Maternal Infection
Control	50	17% (47/56)	11% (50/56)	0% (56/56)
Hand RB51	62	65% (28/80)*	53% (38/80)*	11% (66/74)*
Single Ballistic	30	60% (12/30)*	57% (13/30)*	13% (26/30)*
Ballistic Sx	14	65% (5/14)*	43% (8/14)	14% (12/14)
Hydrogel Bal.	19	32% (13/19)*	21% (15/19)	0% (19/19)

* Significantly different ($P < 0.05$) than Control

Colonization Data

Log CFU/gm

	Parotid LN	Prescap LN	SM LN	Placentome
Abortion				
Cattle (5)	2.4 ± 0.2	1.4 ± 0.6	1.2 ± 0.7	6.3 ± 1.6
Bison (34)	2.7 ± 0.1	2.0 ± 0.2	2.7 ± 0.7	7.4 ± 0.3
Full Term				
Cattle (3)	0 ± 0	0 ± 0	0 ± 0	0 ± 0
Bison (7)	1.7 ± 0.4	1.0 ± 0.4	0.9 ± 0.9	2.5 ± 1.2
Elk (27)	0.8 ± 0.2	0.3 ± 0.2	0.5 ± 0.2	1.7 ± 0.6

IF RB51 A BOOSTER VACCINATION IS GIVEN

Efficacy of RB51 in Bison

Rate of abortion or infection

Rate % (# aborted/infected / total)

Treatment	N	Abortion	Uterine Infect	Mammary Infect	Maternal* Infect
Control	**6**	**83%(5/6)**	**100%(6/6)**	**100%(6/6)**	**100%(6/6)**
Hand RB51	**6**	**67%(2/6)**	**66%(4/6)**	**83%(5/6)**	**83%(5/6)**
Dart RB51	**7**	**57%(4/7)**	**57%(4/7)**	100%(7/7)	**94%(6/7)**
Booster RB51	**5**	**0%(0/5)**	**40%(2/5)**	**80%(4/5)**	**40%(2/5)**

*Not mammary samples

Colonization Data

Treatment	Log CFU/gm (no culture positive/total)			
	Parotid LN	Prescap LN	SM LN	Placentome
Control	2.7 ± 0.3 (6/6)	1.7 ± 0.4 (5/6)	1.9 ± 0.5 (5/6)	7.6 ± 0.3 (6/6)
Hand RB51	0.8 ± 0.4 (3/6)*	0 ± 0 (0/6)*	0.7 ± 0.5 (2/6)	4.0 ± 1.8 (3/6)*
Dart RB51	1.2 ± 0.5 (4/7)	0.3 ± 0.3 (1/7)*	0.9 ± 0.4 (4/7)	4.5 ± 1.6 (4/7)
Booster RB51	0.8 ± 0.6 (2/5)*	0 ± 0 (0/5)*	0 ± 0 (0/5)*	1.7 ± 1.1 (2/5)*

* ($P<0.05$) compared to control

Eradication of Brucellosis from the GYA

- Species: *Brucella abortus*
- Hosts: Bison, elk and cattle
- Current status: Good vaccine and coverage for cattle; Moderately effective vaccine for bison; No vaccine currently for elk; Delivery issues
- Would need to combine vaccination with test and removal

Thoughts on "Natural Immunity"

- Intracellular environment and immunologic responses to *Brucella* complex
- Many redundancies and feed-back loops
- *Brucella* a excellent pathogen and stealthy
- I don't believe a single gene of the host regulates susceptibility/resistance

Thoughts on Seropositives

- No easy way to determine if "exposed" or infected
- We're evaluating new technology for detecting infection, but high risk approach (Aperio)
- How seropositives are handled should be based on control program objectives
- Contribution to herd immunity can be argued both pro and con

Opportunities and Constraints for Development of New Vaccine

- Select Agent Act
- Challenges in Developing a New Vaccine
  - "low hanging fruit" has been picked
  - laboratory animal models do not replicate responses in natural hosts
  - Cost
- Solve problems, not just study *Brucella*

Opportunities for New Vaccines

- Nanoparticles
- DNA Vaccines
- Recombinants in which "stealthiness" has been diminished
- New Adjuvants

- Need good scientists/laboratories to collaborate as possible

Other Related Research

- Sequencing Bison Genome with Texas A&M, ISU, and Univ. of Maryland
- Initiating transcriptomics studies
- Exploring Immunogenicity of a Nanoparticle Vaccine
- Evaluating effect of synthetic adjuvants on immune responses by bison and elk
- Collaboration with University of Wyoming scientists on efficacy of adult Vx in 2014

Some Final Thoughts

- Vaccines and/or Delivery program may have to be engineered by species
- Addressing Select Agent concerns must be based on science and facts
- Development of new vaccines is a challenge but new technology may help
- Developing vaccine that prevents seroconversion will be very hard