

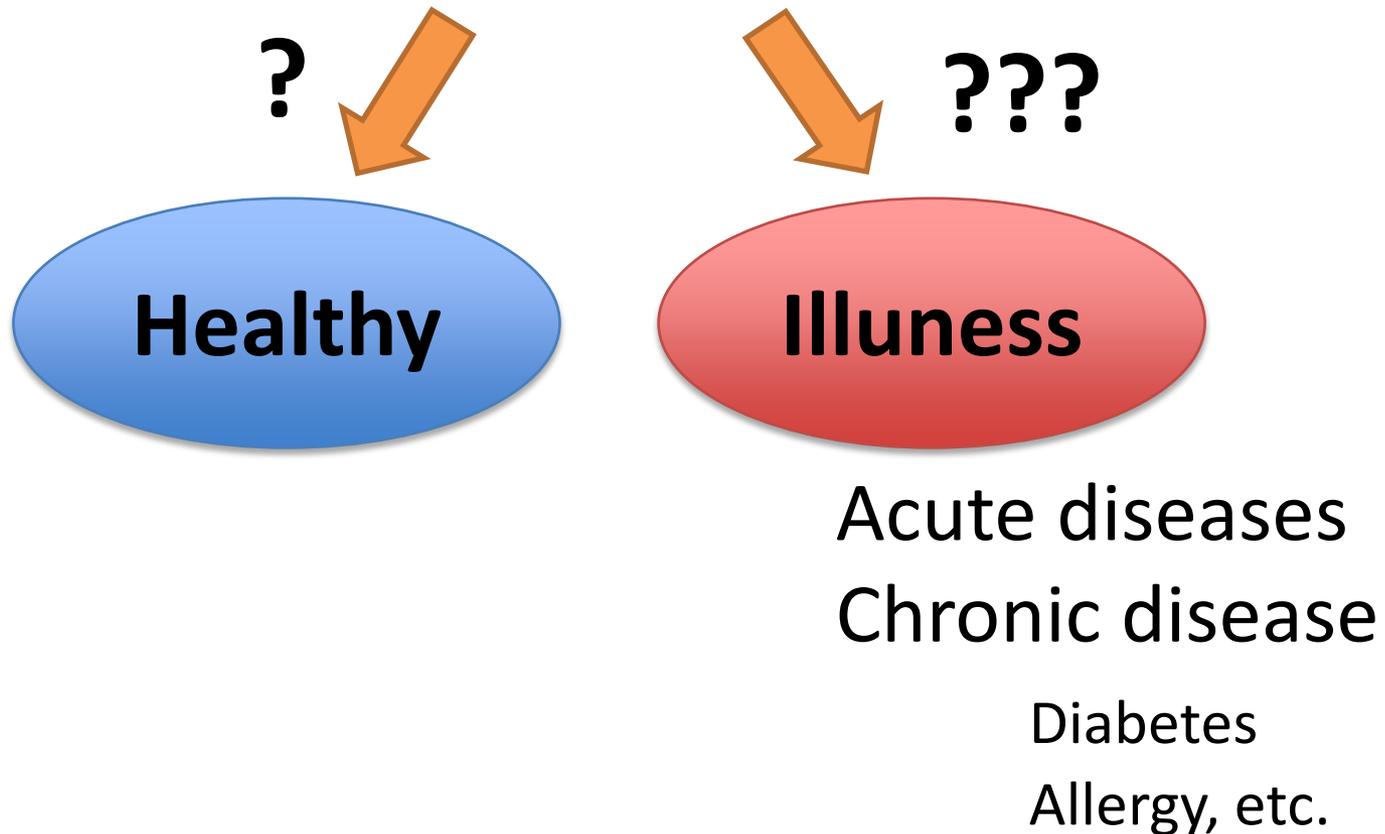
Microbiological Risk Assessment of Bioterrorism Agents and Underlying Diseases

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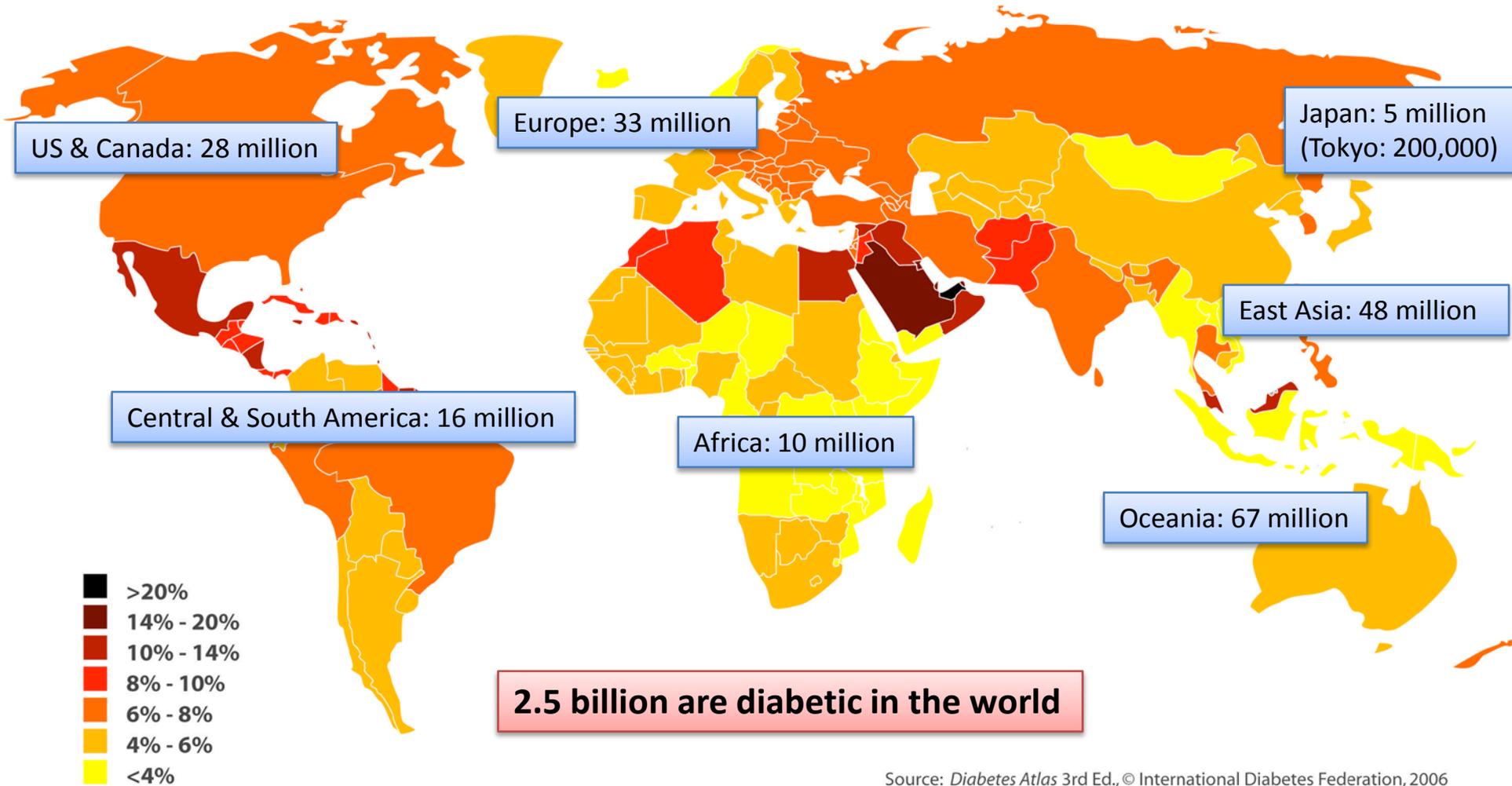
Risk analysis of biological terrorism



Microbiological risk assessment

1. Hazard Identification
2. Exposure assessment
3. Hazard characterization
4. Risk characterization





Source: *Diabetes Atlas* 3rd Ed., © International Diabetes Federation, 2006

Modified from Diabetes Atlas by IDF

Why we use “sick” animal

- Most of risk assessment studies have been done by using healthy animal model.
- Number of underlying diseases (diabetes, obesity, allergy, etc) is increasing especially in big cities (industrial countries), where are possible targets for bioterrorism.
- Some diseases are associated with abnormal immunity.
- Risk factors for increased susceptibility or resistance to biological agents are not known well.



Experimental design

Disease model mice



Diabetic mice



Atopy mice

Inoculate various
number of spores *i.p.*

Bacillus anthracis
Pasteur II strain
pXO1⁺, pXO2⁺, encapsulated

Monitoring clinical
signs, survival rate.

CBC, blood glucose, ALT,
etc. & CT

Bacterial number in
blood, spleens, livers,
lung & brain.

Histology (HE stain)

Cytokine levels



db/db mice

- A mouse model for human type II diabetes mellitus

Left: *misty*
(nondiabetic
control)

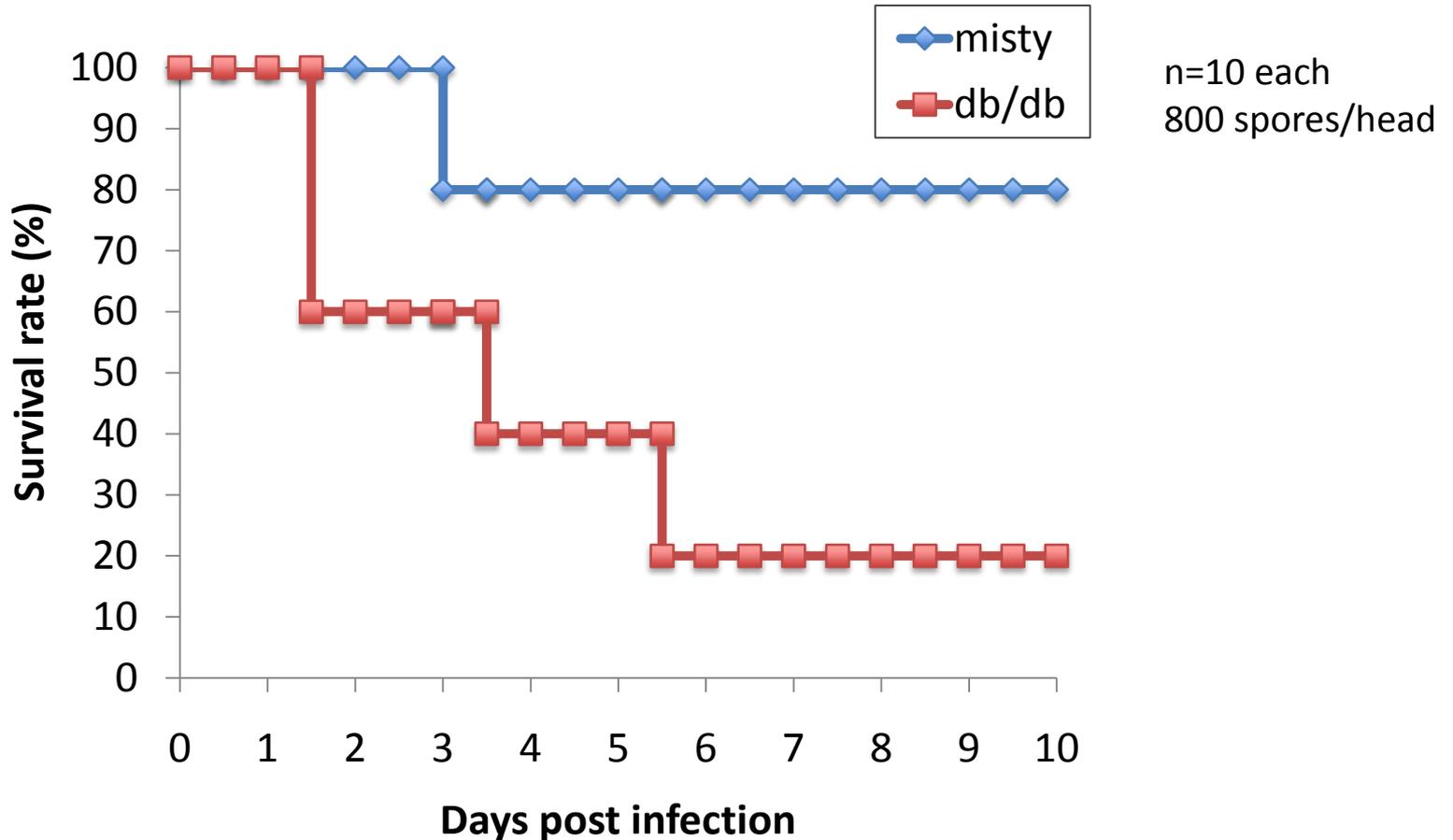


Right: *db/db*
diabetic mice

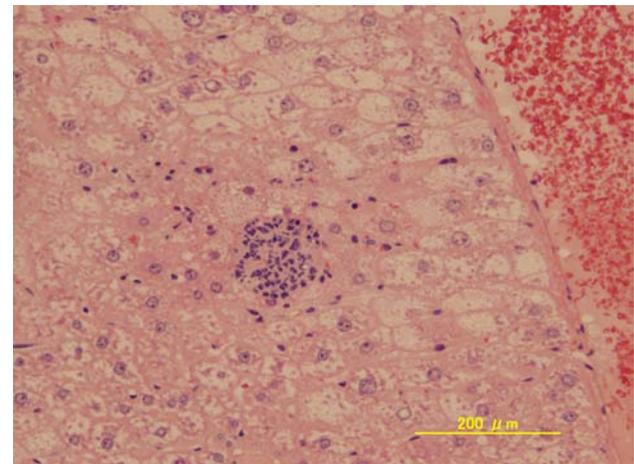
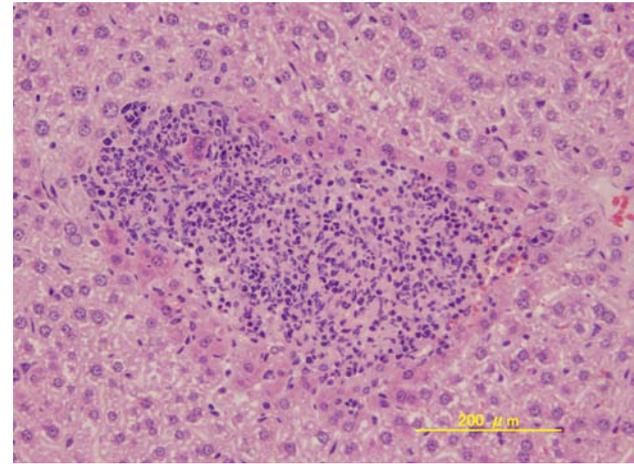
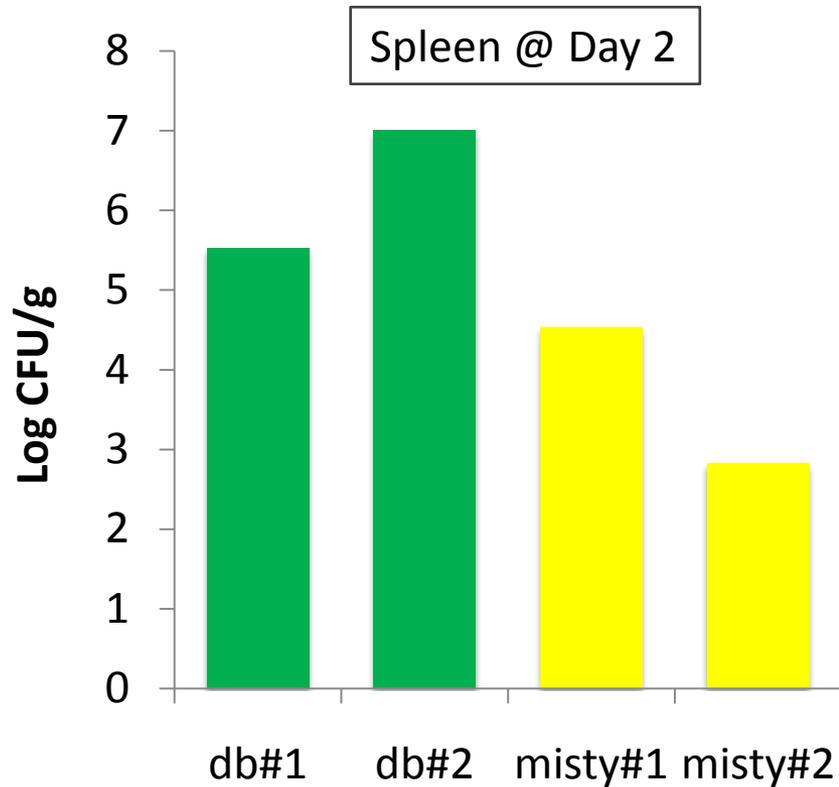
- High blood glucose
- Polyuria, polyposia, bulimia
- Obesity
- Active appetite
- Fatty liver



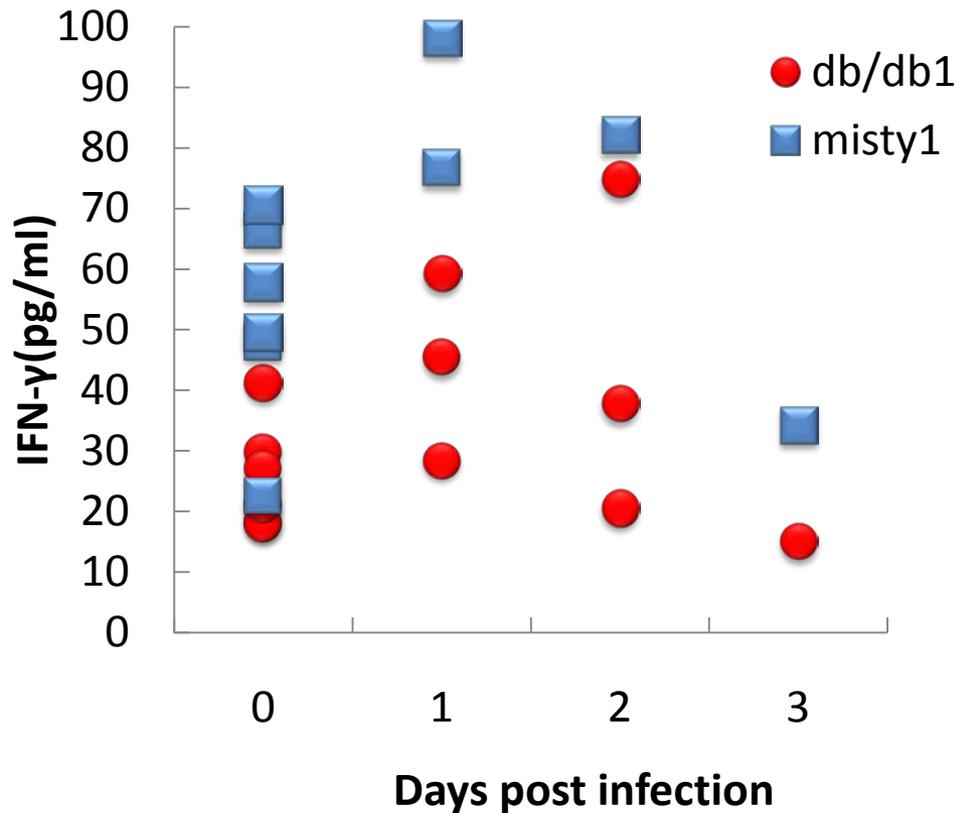
Survival rate in diabetic mice



Impairment of bacterial clearance in diabetic mice



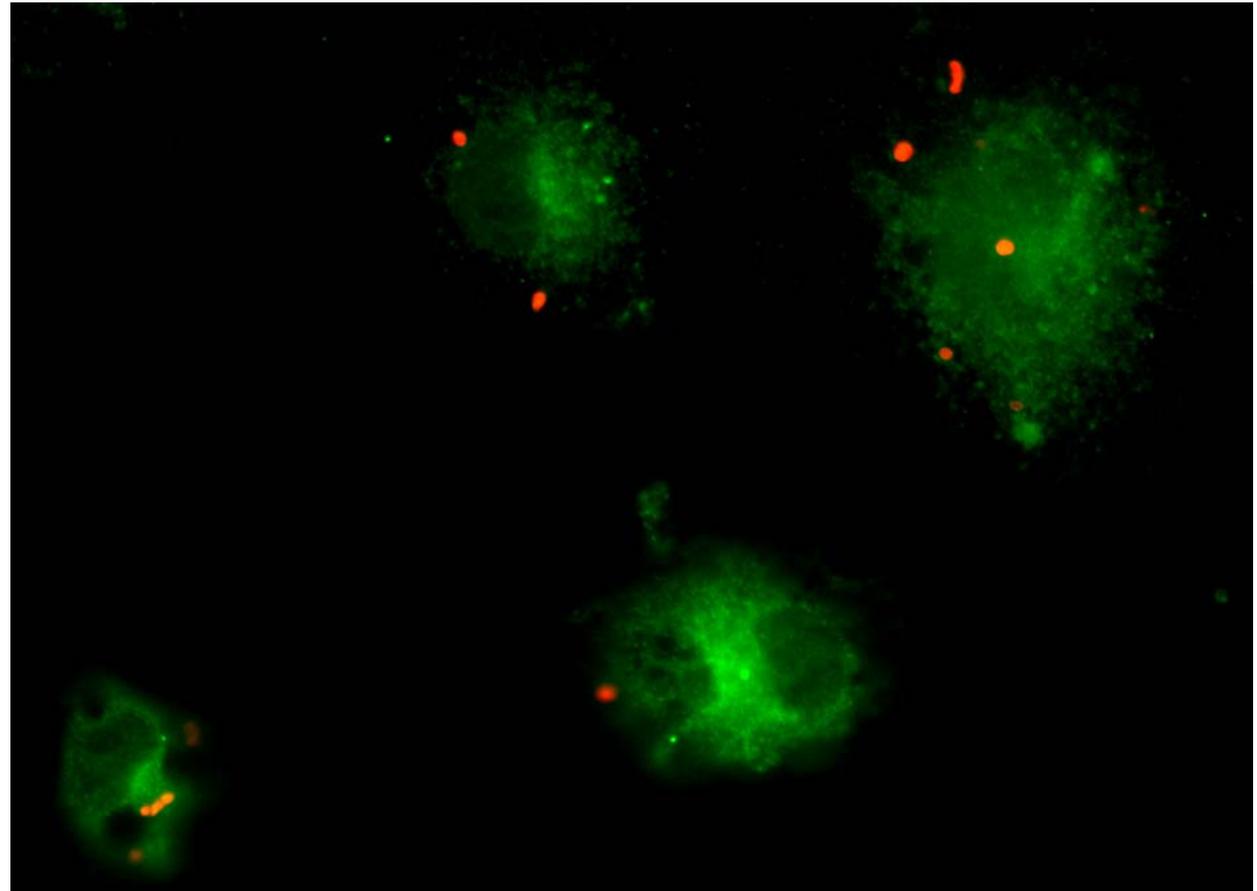
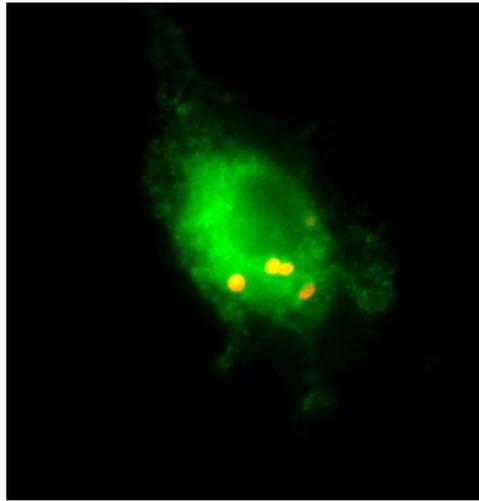
Blood cytokine levels in diabetic mice



- *db/db* > *misty*
 - IL-1 β
- *db/db* < *misty*
 - IFN- γ
 - IL-10
- *db/db* = *misty*
 - TNF- α



Escape of *B. anthracis* spores from *db/db* macrophage lysosomes



Green: LAMP2

Red: *B. anthracis* spore



Summary

- High susceptibility to anthrax in diabetic mice.
- Onset of symptoms was earlier in diabetic mice.
- Retarded clearance of pathogen.
- Impairment of inflammatory reaction.
- Risk ratio for death was 2.03 (diabetic mice vs. non-diabetic)
- Insulin replacement had little protective effect.



Allergic diseases

- is a reaction of hypersensitivity mediated by immunologic mechanisms.
- Asthma, hay fever, atopic dermatitis, allergic conjunctivitis, etc.
- No. of patients is increasing year by year, especially in industrial countries.
- Genetic predisposition, and impairment of barrier function may be attributed.



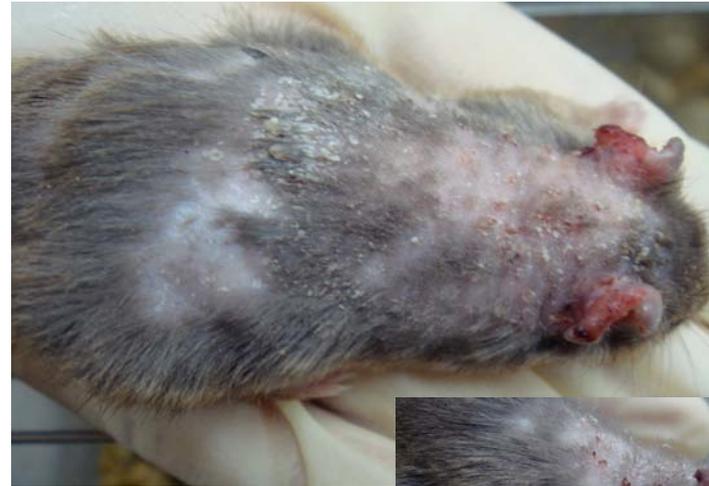
NC/Nga mice

- A mouse model for human atopic dermatitis (AD)

SPF



Conventional



- Scratching behavior due to severe itching
- Skin lesions on face, flexion
- Elevated serum IgE
- Similar symptoms to human AD
- Atopy (Genetically predisposed to allergic diseases)



SPF

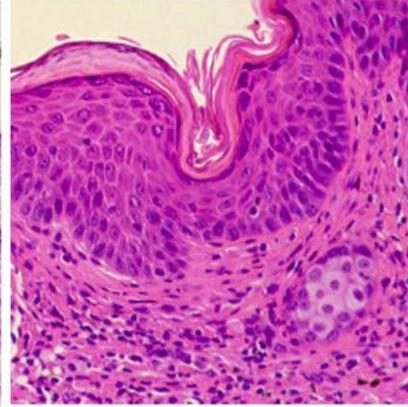
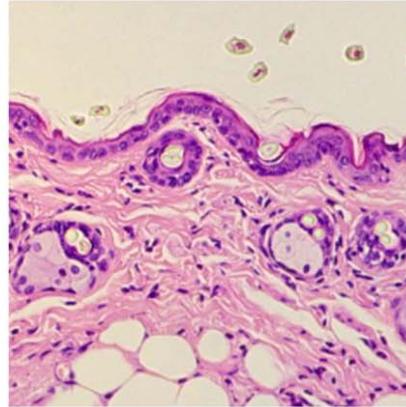
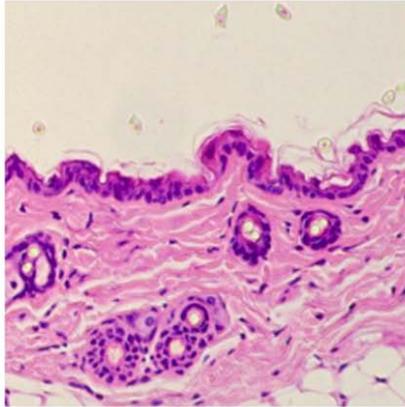
Conventional

Normal

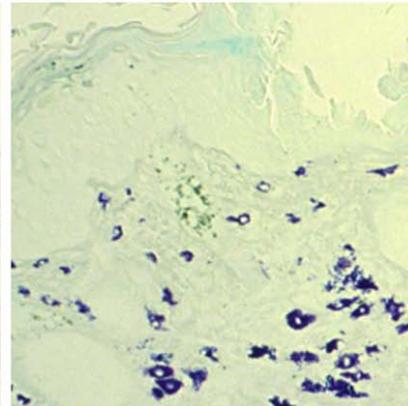
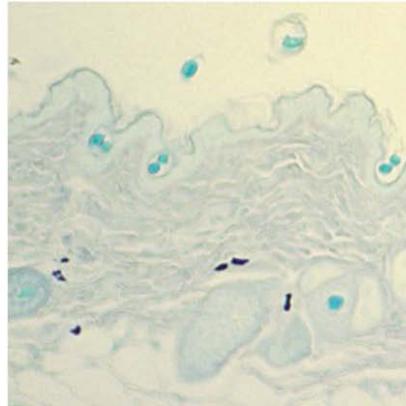
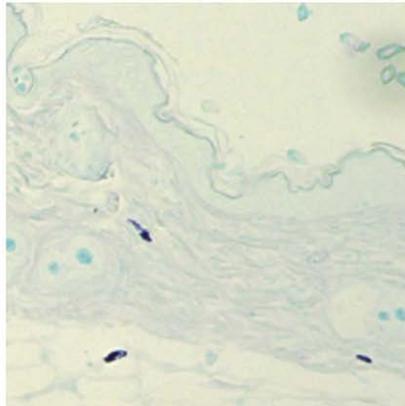
Mild dermatitis

Severe dermatitis

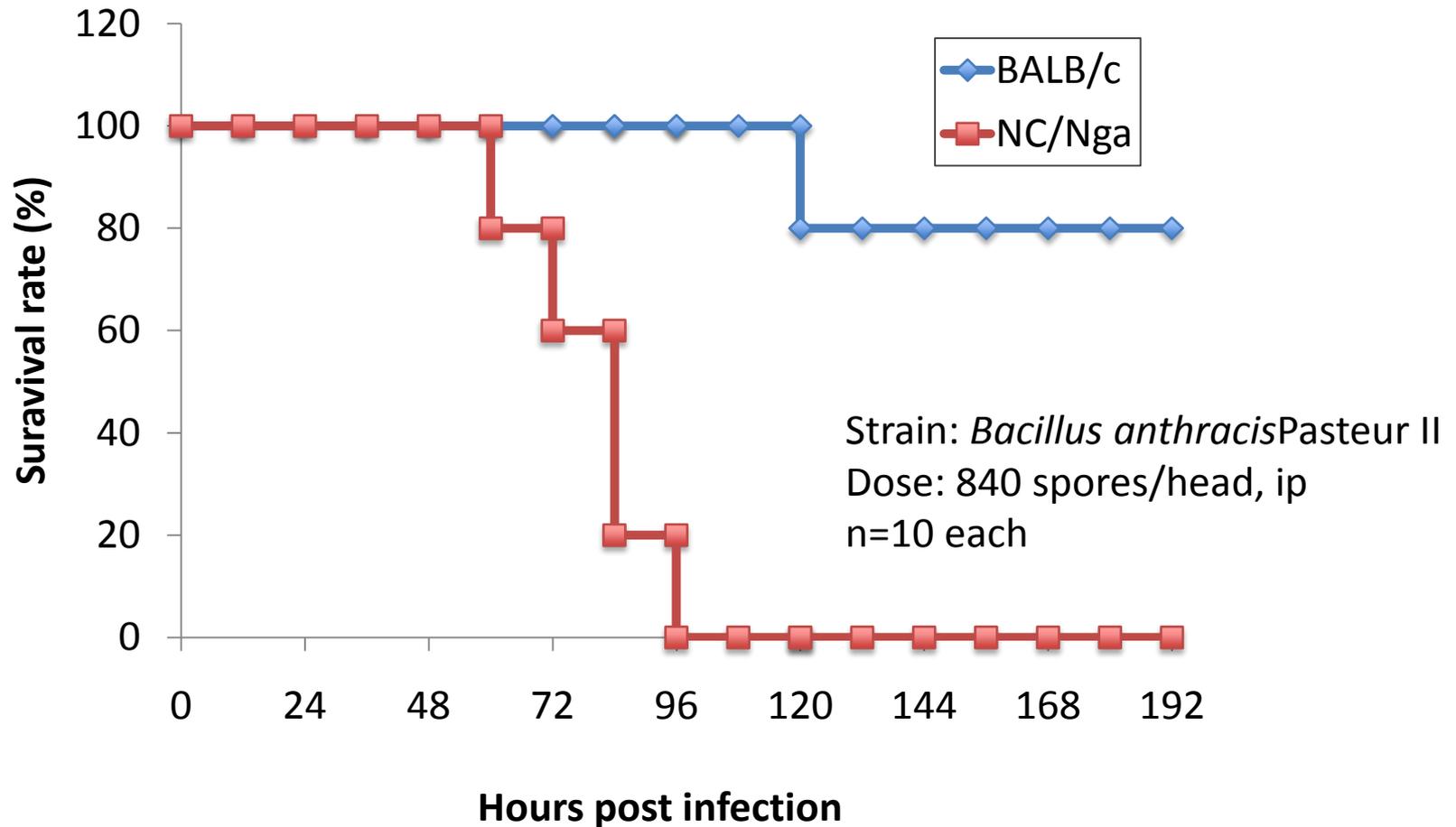
HE stain



Toluidine Blue



NC/Nga is highly susceptible to anthrax



Striking susceptibility in NC/Nga to anthrax

- Unusual immune response
 - Increase in IL-10 production
- Impairment of barrier function
 - Contribute to increase bacterial invasion to tissues



LD₅₀

- *db/db*: 2 to 5 times more susceptible to healthy control
- NC/Nga: 10 to 20 times more susceptible to BALB/c



Conclusion

- Both disease model mice were highly susceptible to anthrax than their healthy control.
- These chronic illnesses lowered resistance to anthrax.
- The unusual immune responses in these mice may attribute to the severity of the disease.
- Atopy should be counted as one of important risk factor to anthrax.

Risk assessment

- Quantitative, qualitative study
- Modeling scenarios
 - Population of risk groups
 - Non-transmittable or transmittable
 - Susceptible hosts release larger amount of pathogen.
- Probability

Other researches

- Tested pathogens
 - *Francisella tularensis*
 - *Brucella abortus*
- Development of rapid & mobile detection system
- Development of safer vaccine for anthrax

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