Beneficial Microflora in Honey Bee Colonies

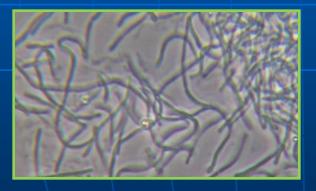


Diana Sammataro, Ph.D.

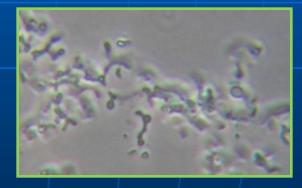


USDA-ARS Carl Hayden Bee Research Center Tucson, AZ

Our Website: http://gears.tucson.ars.ag.gov



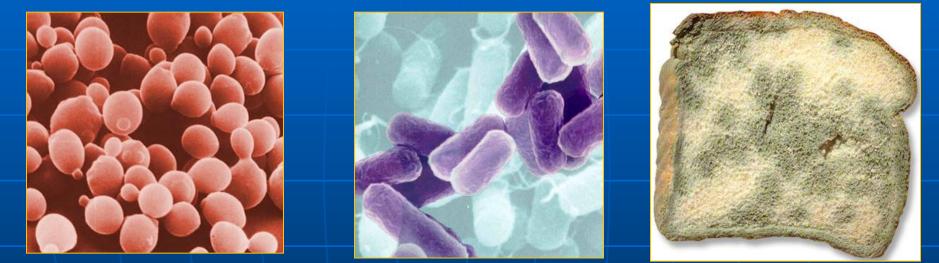
Lactobacillus spp.



Bifidobacterium spp.

Photos by T. Oloffson

What are Microbes?YeastsBacteriaFungus



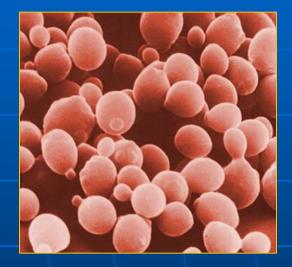
It has been estimated that there are between 10⁴ and 10⁸ microbes per gram of intestinal content typically found in healthy humans

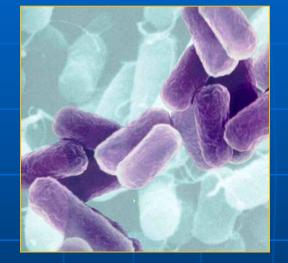
Some are Beneficial

Yeasts

Bacteria

Fungus









Lactobacillus (Yogurt)

Penicillin

Huge Diversity of Microbes

- Original work done by Dr. Martha Gilliam at Tucson Bee Lab
- Found mostly in digestive tract of all stages of bees
 Adult emerging bees inoculated when they emerge
 Active in the conversion of pollen to bee bread
- Keeps uncapped honey from spoiling

Social Insects and Diseases



Social insects evolved mechanisms to fight diseases:

- Microbial: Some microbes fight bad ones
 - Very long association, 25-40 million year old microbes in bees in amber
- Genetic: Hygienic behavior
- Social Behavior: Thermoregulation
- Physical barriers: Cuticle
- Nest components: Propolis
- Humoral : components in hemolymph such as antibodies and enzymes
- Cellular immunity: Hemocytes (phagocytes or white blood cells)
 - and other immune responses

How Microbes Work for Honey Bees

pollen

Lactic acid bacteria in the honey stomach

Lactic acid bacteria from the honey stomach are the first step in preparing pollen (by fermentation and pH) and nectar (protecting against fermenting).

Microbes from flowers



Other fungi, yeasts, bacteria, and molds help to produce enzymes, vitamins, anti-microbial and other substances. Current research is investigating and updating this process.

nectar

Gilliam, M. 1997. Identification and roles of non-pathogenic microflora associated with honey bees. FEMS Microb. Lettrs. 155: 1-10.

Olofsson, T.C. and Vásquez, A. (2008) Detection and identification of a novel lactic acid bacterial flora within the honey stomach of the honeybee *Apis mellifera*. Curr Microbiol. 57:356–363.

Schematic by D. Sammataro

Microflora in Bee Food

- Bacteria Produce:
 - Antibiotics
- Fatty acids and Enzymes that help digest

 Starches, Proteins, Sugars and Cellulose

 Penicillium (a mold)

 Also produce amylases to break down starches

 Yeasts

 Help synthesize B-vitamins

Most Recent Microbes: Honey Stomach Bacteria



Novel lactic acid bacteria (LAB) found IN honey stomachs; belonging to the genus *Lactobacillus* (8), *Bifidobacterium (4)* and the family *Pasteurelaceae* (1)

13 new LAB bacteria now identified

All Apis species have the same LAB as well as some novel phylotypes

***Vásquez ,A., Olofsson, T.C. and D. Sammataro. 2009.** A scientific note on the lactic acid bacterial flora discovered in the honey stomach of Swedish honeybees. Apidologie, 40:26-28.

SEM of Honey Stomach

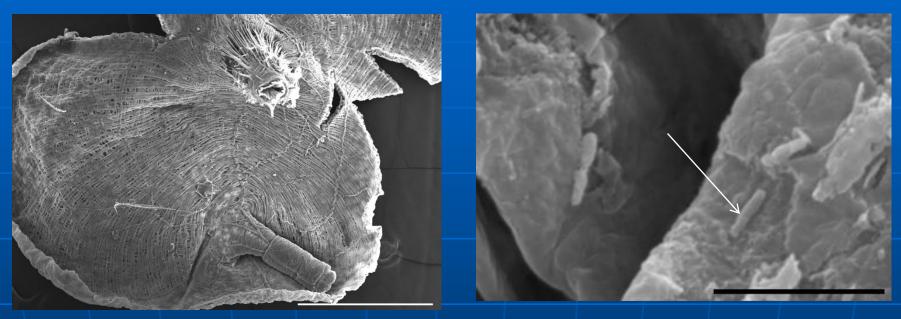
proventriculus



esophagus

Sammataro and Cicero. 2009. Functional morphology of the honey stomach wall of European honey bees (*Apis mellifera* L.); in review Ann. Entomol.

Interior of Honey Stomach



Interior of HS

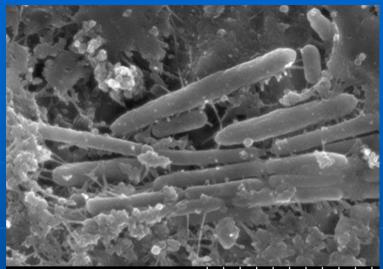
Photos by J. Cicero, UA

Sample Bacteria from Honey Stomach



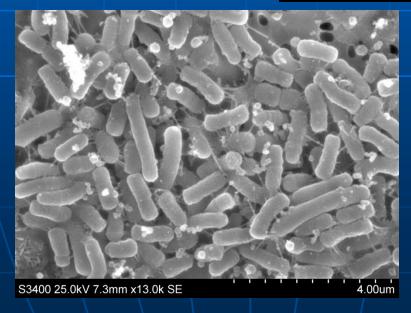
S3400 25.0kV 7.3mm x30.0k SE

1.00um



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S3400 25.0kV 7.3mm x14.0k SE
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4.00um



Photos by J. Cicero, UA

What factors influence microbial populations?



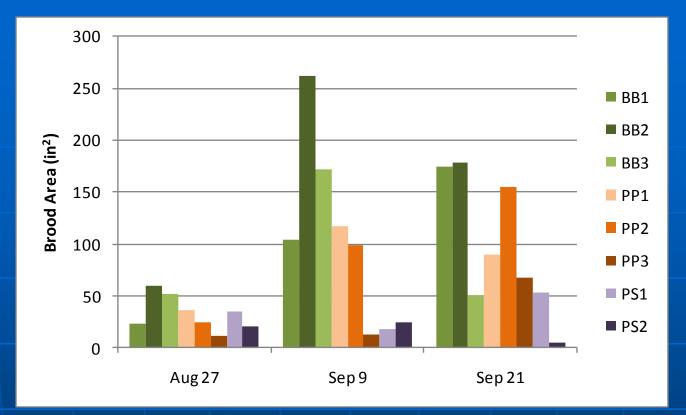
Feeding Experiment: What happens when you limit Nutrition?



Bee Bubble, colonies fed only artificial diet plus syrup.....



After 6 weeks, colony declines: lack of bacteria?

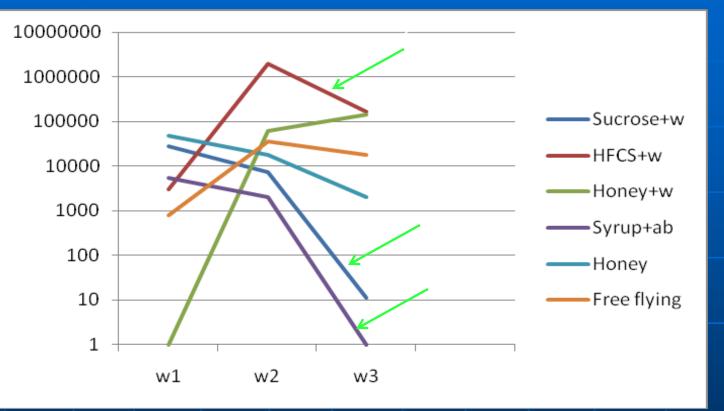


Brood area (in²) of colonies maintained in a closed foraging arena with different in-hive protein sources. On August 27, all colonies were receiving non-pollen protein supplement. On September 9, some colonies were supplied with either frames of bee collected pollen and bee bread or pollen patties.

(Treatments: PS = non-pollen protein supplement; PP= pollen patty; BB= Frames of bee bread). M. Weiss, Master's Thesis.

Preliminary Caged Bee Study on Effect of Diet on LAB





Olofsson, Vasquez, Fries, Sammataro et al. **2010.** Symbiosis between lactic acid bacteria and honey bees. Nature, in review.

How do microbes affect bee health?

- A reduction of beneficial microbes could cause nutritional deficiencies , resulting in:
 - Increased incidence of disease (chalkbrood)
 - Supersedure of queens
 - Reduced colony growth

What good is this information?

- Tells us what microflora are there and when
- Tells us who is doing harm, and who is beneficial
- Gives us insight into how to manage the complex microbial communities found in honey bee colonies

Collaborations:

- Dr. Kirk Anderson (Microbial Ecologist, Tucson)
- Dr. Mark Carroll (Chemical Ecologist, Tucson)
- Drs. Vásquez and Olofsson Lund Un., Sweden
- Dr. Jay Yoder, Wittenberg Un.
 (OH)

