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This presentation will provide answers from a microbiology expert for common questions relayed to microbiologic content, testing, and effect of alternative processing conditions including:

1. What are the acceptable levels of microbes in honey?
2. What methods are recommended for testing microbes?
3. What processing options exist if a customer requests that microorganisms be reduced or eliminated?
4. Is there scientific evidence documenting the microbiologic safety of honey over shelf-life that I can use with customers requesting this information?
5. Is there information I can provide about microbiologic safety of honey made with additions or infusions?

Please note: The perspectives of this expert do not replace regulatory requirements related to honey. These are simply an expert's response to general questions asked.

1. What are acceptable levels of microbes in honey?

Expected levels of microorganisms

Microorganism	Expected Levels	Reference
Standard Plate Count/ Aerobic Plate Count (APC)	10,000 cfu/g	Snowdon and Cliver (1996)
Coliform	100 cfu/g	Snowdon and Cliver (1996)
Non-Osmophilic Yeasts[a]	100 cfu/g	Snowdon and Cliver (1996)
Non-Osmophilic Molds[b]	100 cfu/g	Snowdon and Cliver (1996), Luca (2024)
Osmophilic Yeasts[a, c]	10 to 10^6 cfu/g	Snowdon and Cliver (1996), Luca (2024)
Clostridium and Anaerobic Spores	~ 150 cfu/g	Snowdon and Cliver (1996)
Bacillus and Aerobic Spores	~ 200 cfu/g	Snowdon and Cliver (1996)
Thermophilic Spores[d]	Average of 5 samples ≤125 cfu/10-grams. 0 of 5 samples ≥150 cfu/g.	National Canners Sugar Product Specifications.
Thermophilic Anaerobic Spores[d]	Not present at more than 1 MPN/g in no more than 3 of 5 samples.	National Canners Sugar Product Specifications.
Vegetative bacterial pathogens[e]	Slight risk of presence.	Luca (2024)

Microbiological expectations and references from previous table

[a] Concentration of yeasts is proportional to the amount of moisture in product.

[b] Molds are unavoidable but account for far fewer quality issues than osmophilic yeasts.

[c] Many types of osmophilic yeasts may be present, however genus Zygosaccharomyces, Saccharomyces, and Pichia are examples capable of growth a pH 2.0.

[d] National Canners Sugar Product Specifications.

[e] The presence of vegetative pathogens (Staphylococcus aureus, Salmonella spp.) is possible but not likely and vegetative cells typically die during storage in honey.

2. What methods are recommended for testing microbes?

- Microbiological testing methods may vary but use of AOAC-approved methods such as those found in the FDA Bacteriological Analytical Manual (BAM) are appropriate.

3. What processing options exist if a customer requests that microorganisms be reduced or eliminated?

Comparing options for Eliminating or Removing Microorganisms from Raw and Processed Honey

Microbes	Flash pasteurization	Low temperature heat	Irradiation	
Non-spore-forming bacteria.	176°F for 30 seconds.[a]	140°F to 145°F for 30 minutes.[a]	>10-fold reduction at 1.9 to 12.8 kGy.	Filtration using 0.2-micron pore size removes microbes but is challenging for a viscous product like honey.
Yeast, Mold, Osmophilic Yeast and Osmophilic Mold.	176°F for 30 seconds.[a]	140°F to 145°F for 30 minutes.[a]	>10-fold reduction at 1.9 to 12.8 kGy.	
Spore-forming bacteria including Anaerobic, Anaerobic, and Thermophilic Anaerobic.	Not applicable due to the thermal stability of spores, in particular thermophilic spores[b].	Not applicable due to the thermal stability of spores, in particular thermophilic spores[b].	10-fold reduction at 1.9 to 12.8 kGy.	

[a] Scepankova, H, et al. "Conventional and emergent technologies for honey processing: A perspective on microbiological safety, bioactivity, and quality" *Comprehensive Reviews in Food Science and Food Safety* 2021;20:5393–5420. DOI: 10.1111/1541-4337.12848

[b] Retort (high heat and pressure canning) processes may eliminate spore-forming bacteria. However, thermophilic spore-forming bacteria may still survive these thermal treatments.

Options for control of spoilage by osmophilic yeasts and molds, continued

The main spoilage microorganisms of honey will be osmophilic yeasts. Control of the osmophilic yeasts will result in control of other bacteria present in raw and processed honey. Options include:

- Pasteurization at 176°F for 30 seconds or 140°F to 145°F for 30 minutes will eliminate osmophilic yeasts.
- Alternatively (or additionally) storage < 50°F will drastically slow the growth of osmophilic yeasts.
- Combining storage < 50°F with < 50% relative humidity will drastically slow the growth of osmophilic yeasts.

4. Is there scientific evidence documenting the microbiologic safety of honey over shelf-life that I can use with customers requesting this information?

- At a minimum controlling for of spoilage by osmophilic yeasts and molds should result in a long shelf-life of processed honey, and potentially raw honey.

- A study in 2012 documents stability of honey when correctly processed and/or stored is a publication from Rozanska and Osek (2012). In this study, the authors analyzed 109 samples of honey representing different botanical types over one year of storage. They concluded: "Our results confirmed the opinions that microorganisms are not able to multiply in honey"

Table 1

Comparison of the results of microbiological analysis of honey before and after one year storage at room temperature

Type of honey	Total number of aerobes (CFU/g)		Presence of spore forming anaerobes (number of samples/percentage)		Total number of yeasts and moulds (CFU/g)	
	2010	2011	2010	2011	2010	2011
Multifloral (n = 59)	4.2×10^3	2.5×10^3	11/18.6	12/20.3	2.1×10^2	1.2×10^2
Lime (n = 13)	3.7×10^3	2.4×10^3	2/15.3	4/30.8	1.5×10^2	6.3×10^1
Buckwheat (n = 11)	6.8×10^3	4.5×10^3	1/9.1	1/9.1	2.9×10^2	5.5×10^1
Acacia (n = 11)	6.8×10^3	4.6×10^3	2/18.2	3/27.3	8.2×10^1	5.9×10^1
Forest (n = 8)	3.6×10^3	1.9×10^3	6/75.0	0/0	1.2×10^2	1.5×10^2
Rape (n = 7)	3.4×10^3	8.4×10^2	3/42.9	4/37.1	8.6×10^1	9.3×10^1
Mean	4.6×10^3	2.9×10^3	25/22.9	25/22.9	1.9×10^2	9.8×10^1

Figure reprinted as published in Bull Vet Inst Pulawy 2012, 56: 161-163 Rozanska and Osek "Effect of Storage on Microbiological Quality of Honey."

5. Is there information about microbiologic safety of honey made with additions or infusions?

In addition to the information provided in the NHB brochure on *Infusions 101,* the following advice was provided by the expert.

- Refrigeration and maintenance of a pH <4.6 will control the potential growth spoilage microorganisms and most pathogenic microorganisms.
- However, the addition of spices, seasonings, herbs, etc. to honey may impact the microbiological safety and quality of the resulting infusion. Therefore, any ingredients added to honey should be evaluated for safety according to local, state, and federal regulations.

A good citation for refrigeration and pH control in foods is "Food Microbiology: Fundamentals and Frontiers". M. P. Doyle, L. R. Beuchat and T. J. Montville eds. ASM Press 1997.

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