Introduction

Nitrite Production of Sporeforming Bacteria and Their Impact on Safety of Powdered Infant Formula

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Nitrite
- Health risk factor of infants
  → Methemoglobinemia, Blue Baby Syndrome

Regulatory limits
- Acceptable Daily Intake (FAO/WHO): 0.06 mg/kg of bw
- In infant formula (China): < 2 ppm
Aim of this study
To isolate & characterize nitrite-producing sporeformer in infant formula processing
Major findings

Isolation of nitrite-producing sporeformer in infant formula process

Sporeformers in infant formula
Both mesophilic and thermophilic bacteria were found

Hypothesis of nitrite production during infant formula process

Experimental Flow

01. Isolation of nitrite-producing sporeformer in infant formula process

Commercial products → Isolates → Nitrile production test

Infant formula processing plants ← Isolates

Nitrite production under processing conditions

02. Identification of high-risk isolates with active nitrite and endospore forming

Nitrite production under processing conditions

Highly heat-resistant spore

Infant formula production process

Mixing raw materials → Heat-sterilization → Condensation → Homogenization → Spray-drying → Final product

Germination, growth, and metabolism

Vegetative cell → Endospore

Mixed milk, Sterilized milk, Condensed milk, Homogenized milk, Semi product, End product

Monophic bacteria total count, Monophic bacteria spore count

Monophic bacteria total count, Monophic bacteria spore count

4th Asia-Pacific International Food Safety Conference & 7th Asian Conference on Food and Nutrition Safety, October 11-13, 2016, Penang, Malaysia
Isolation of nitrite-producing sporeformer in infant formula process

Collection of sporeformers

Nitrite-producing test

Production of nitrite during growth

Griess-test

Total 254 isolates

Mesophilic bacteria n=112
Thermophilic bacteria n=142

Nitrite-producing sporeformers exist in the process

Identification of high-risk isolates with active nitrite and endospore forming

Implication

Active nitrite production

Formation of highly-heat resistant spore

Major high-risk bacteria

New insight

Nitrite production under highly-heat resistant spore

Condition: generally at 55-60°C
Only G. stearothermophilus can produce nitrite above 60°C
G. stearothermophilus strain shows remarkably rapid initiation of production

Highly-heat resistant spore

B. subtilis (n=1)
B. tequilensis (n=1)
G. stearothermophilus (n=11)
All isolates can produce highly-heat resistant spore

Identification of high-risk isolates with active nitrite and endospore forming

Target (Nitrite producer)

Mesophilic isolate

Thermophilic isolate

G. stearothermophilus

G. stearothermophilus

B. subtilis

B. tequilensis

G. stearothermophilus
Sporeforming bacteria in infant formula processing line could determine the risk of nitrite.

**Conclusion**

01. **Risk of nitrite** in infant formula have been underestimated

02. For high-risk isolates obtained in this study, **risk characterization** should be conducted

03. **Intervention methods** for the control of nitrite-producing sporeformers are needed

For me, this research is **duty**

If we can do something for our babies, **We should**.