The Forensic Characterization of Bacterial and Fungal Organisms in Traditional Chinese Medicine

Julia Grzymkowski
Virginia Commonwealth University

Christopher J. Ehrhardt
Virginia Commonwealth University

Justin L. Poklis
Virginia Commonwealth University

Michelle R. Peace
Virginia Commonwealth University

Follow this and additional works at: https://scholarscompass.vcu.edu/uresposters

Part of the Bacteriology Commons, Biology Commons, Lipids Commons, Medicinal Chemistry and Pharmaceuticals Commons, Medicinal-Pharmaceutical Chemistry Commons, Pharmacology Commons, and the Toxicology Commons

© The Author(s)
Forensic Characterization of Bacterial and Fungal Organisms in Traditional Chinese Medicine

Julia K. Gryziewski, Christopher J. Ehrhardt, Justin L. Poklis, and Michelle R. Peace
Departments of 1Forensic Science and 2Pharmacology & Toxicology, Virginia Commonwealth University

Purpose
The first aim of this study was to perform a survival experiment to demonstrate the importance of proper herbal brewing technique. The second aim was to conduct a molecular and biochemical survey of microorganisms present on eleven Chinese herbal samples through Fatty Acid Methyl Ester (FAME) analysis with identification of FAMES done using Gas-Chromatography coupled to a Flame Ionization Detector (GC-FID).

Introduction
Traditional Chinese Medicine
• Dates back to 2800 BC
• 6,926 plants, 23,033 chemical substances
• Rx based on Ch. literature and experience
• Dietary Supplements
• Estimated 50% of U.S. population consume
• Dietary Supplement Health and Education Act of 1994
• Recent dangers
  • Improper labeling
  • Toxic contaminants; pathogenic bacteria

Case History
• 43 year old woman with IBS
  • Consulted a traditional Ch. herbal doctor
  • Received a variety of herbs
  • Flu-like symptoms
  • Liver and renal failure
  • Multi-organ failure in surgery
  • Pure growth of Bacillus cereus
    • Herbs prescribed in common
      • Herbs commonly used in herbal medicine
      • Herbs prescribed in traditional Ch. medicine
      • Herbs commonly used in Ch. medicine
      • Herbs prescribed in traditional Ch. medicine
      • Herbs commonly used in Ch. medicine
      • Herbs prescribed in traditional Ch. medicine
      • Herbs commonly used in Ch. medicine

Bacterial Genus of Interest: Bacillus
• Bacillus cereus T-strain
  • Human food-borne pathogen.
  • Produces toxins causing diarrea, nausea, and vomiting.
  • Forms biofilms
  • Bacillus anthracis
    • Releases edema toxin which upsets cell water homeostasis, resulting in massive edema.
    • Skin lesion -> papule -> black eschar
    • Bacillus anthracis
    • Releases edema toxin which upsets cell water homeostasis, resulting in massive edema.
    • Skin lesion -> papule -> black eschar

Samples
Samples were acquired from Tong Ren Tang Herbal Pharmacy in Beijing, China

FAME Method
• Bacteria species have specific lipid profiles
• Herbs cultured on both Blood Agar and Tryptic Soy Agar plates
• Rapid liquid-liquid extraction
• Analysis/identification by GC-FID

Results

Conclusions
• Established a rapid taxonomic survey of biological and fungal contaminants present on suspect herbal samples based on fatty acid biomarkers.
• The survival study demonstrated the potential for pathogenic spores to survive the brewing process.
• The bacterial and fungal identification help to reconstruct toxicological episodes that result in medical emergencies or death.

Funding
UROP 2017 Summer Fellowship:
National Institute of Health
NIH P30DA033934

Method translated from package
Soaked in cold water (30 mins)
Cooking:
Bacillus cereus T-strain
Bacillus anthracis

Traditional American method
Cooking:
Steeped in boiling water (3 mins)

Alternative Ch. method
Cooking:
Bacillus cereus T-strain
Bacillus anthracis

Survival Study
2.80 g of herb mixture + 500 µL liquid Bacillus cereus T-strain spores (2.4 mg dried)
Frozen then lyophilized [n=3]

Anticonvulsive
Herbs prescribed in common
• Astragalus sp.
• Poria cocos

Analgesic
Herbs prescribed in common
• Cinnrao Baccus
• Cinnamomum cassia

Conclusions
Astragalus sp.
Zao by Unknown Author is licensed under

Purpose
The first aim of this study was to perform a survival experiment to demonstrate the importance of proper herbal brewing technique. The second aim was to conduct a molecular and biochemical survey of microorganisms present on eleven Chinese herbal samples through Fatty Acid Methyl Ester (FAME) analysis with identification of FAMES done using Gas-Chromatography coupled to a Flame Ionization Detector (GC-FID).