THE UTILIZATION OF URINE PROCESSING FOR THE ADVANCEMENT OF LIFE SUPPORT TECHNOLOGIES

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The Most Extensive Urinalysis Publication

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The Human Urine Metabolome

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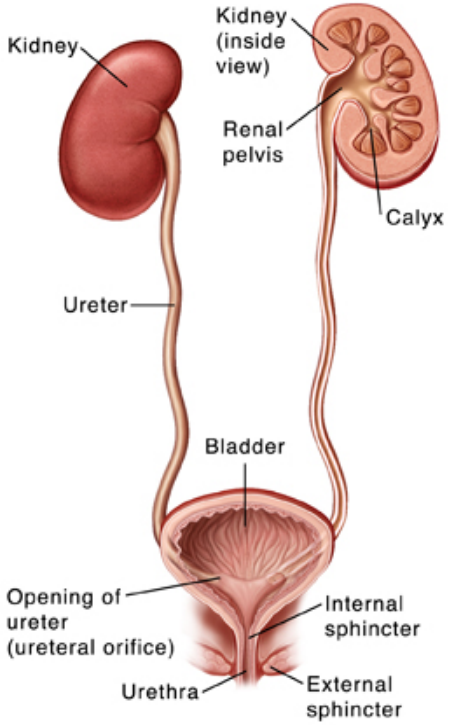
Abstract

Urine has long been a ''favored'' biofluid among metabolomics researchers. It is sterile, easy-to-obtain in large volumes, largely free from interfering proteins or lipids and chemically complex. However, this chemical complexity has also made urine a particularly difficult substrate to fully understand. As a biological waste material, urine typically contains metabolic breakdown products from a wide range of foods, drinks, drugs, environmental contaminants, endogenous waste metabolites and bacterial by-products. Many of these compounds are poorly characterized and poorly understood. In an effort to improve our understanding of this biofluid we have undertaken a comprehensive, quantitative, metabolome-wide characterization of human urine. This involved both computer-aided literature mining and comprehensive, quantitative experimental assessment/validation. The experimental portion employed NMR spectroscopy, gas chromatography mass spectrometry (GC-MS), direct flow injection mass spectrometry (DFI/LC-MS/MS), inductively coupled plasma mass spectrometry (ICP-MS) and high performance liquid chromatography (HPLC) experiments performed on multiple human urine samples. This multi-platform metabolomic analysis allowed us to identify 445 and quantify 378 unique urine metabolites or metabolite species. The different analytical platforms were able to identify (quantify) a total of: 209 (209) by NMR, 179 (85) by GC-MS, 127 (127) by DFI/LC-MS/MS, 40 (40) by ICP-MS and 10 (10) by HPLC. Our use of multiple metabolomics platforms and technologies allowed us to identify several previously unknown urine metabolites and to substantially enhance the level of metabolome coverage. It also allowed us to critically assess the relative strengths and

Physiology of Urine

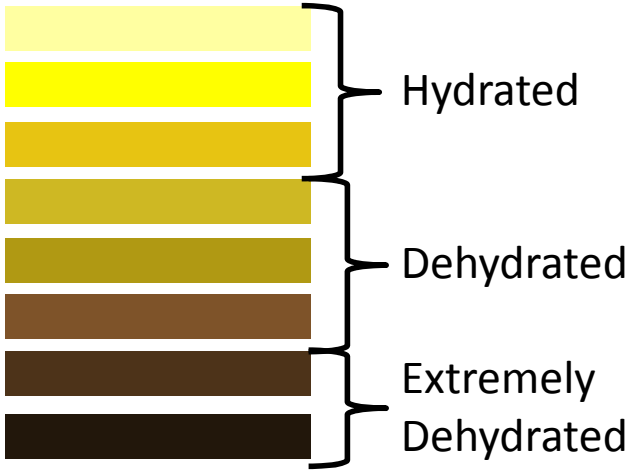
- Urine is used to maintain homeostatic levels within the body at the cellular and system level.
- Water content influences osmotic pressure, blood pressure, digestion, etc.
- Urination is the fastest and most efficient way to remove wastes from the body, including:
  - Metabolic byproducts
  - Unabsorbed nutrients
  - Contaminants
  - Excess water



http://www.uofmchildrenshospital.org/HealthLibrary/Article/40091

Physiology of Urine

- The average person produces 1.5 L of urine/day. 0.6 L – 2.0L is within the acceptable range.
- Urine is used as a diagnostic indicator for hydration level, infection, irritation, or blockage.
- Standard compounds in healthy urine include:
  - Cations
  - Anions
  - Inorganic salts
  - Physiological byproducts



Urine in Space

Bouatra, *et al* for Space

- The most comprehensive study of urine discovered over **3,000 metabolites and compounds**
- **42** were analyzed for potential recovery during spaceflight.
- The review included selecting those with a 100% occurrence rate and an average concentration of 10 µM/mM creatine.
  - Scaling to creatine allowed for normalization/compensation for variation in urine volume.
  - Scaling to creatine also normalizes variability due to donors and average metabolite concentration (which can vary by ± 50 – 350%)

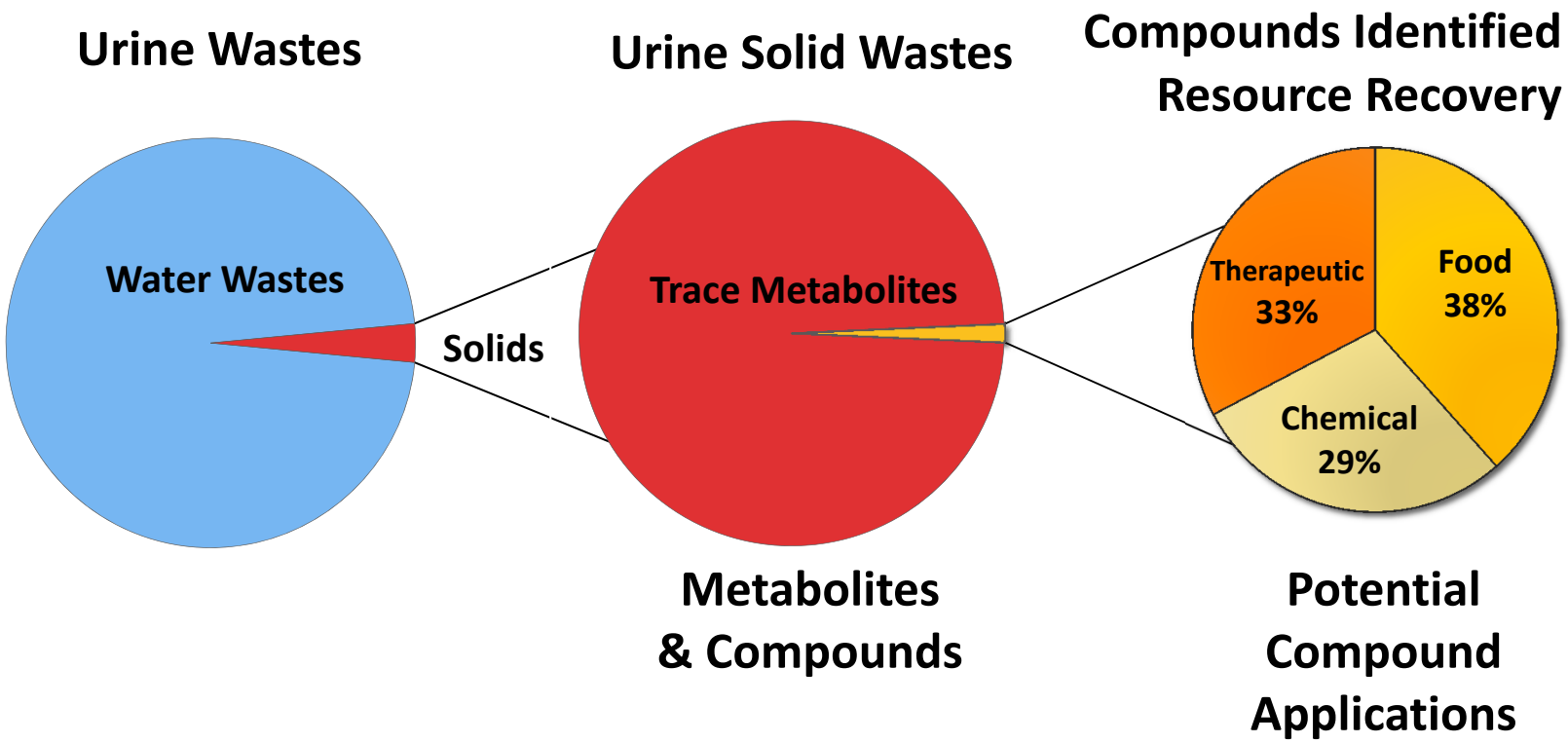
Potential Applications

Food Applications

Therapeutic Applications

Chemical Applications

Urine Contents of Interest



Urine Contents of Interest: Superclasses

- Aliphatic Compounds (9/93)
  - Carbon and hydrogen, non-aromatic.
- Alkaloids & Derivitives (1/45)
  - Organic compounds with rings, can be either aromatic or non-aromatic.
- Amino Acids & Peptones (16/286)
  - Involved in biological synthesis. Some are essential, not made *in vivo*.
- Carbohydrates (9/116)
  - Essential for central metabolism: mono-, di-, oligo-, and polysaccharides.
- Lipids (1/866)
  - Create barriers (lipid bilayers), aid absorption of vitamins and minerals.
- Organic Acids (9/108)

Aliphatic Compounds

Dimethylamine (30.8 µM/mM creatine)

- Concentration in urine based on dietary intake
  - plant and animal-based foods, specifically fish and seafood
- Common component in soaps, cleaning supplies, agricultural fungicides

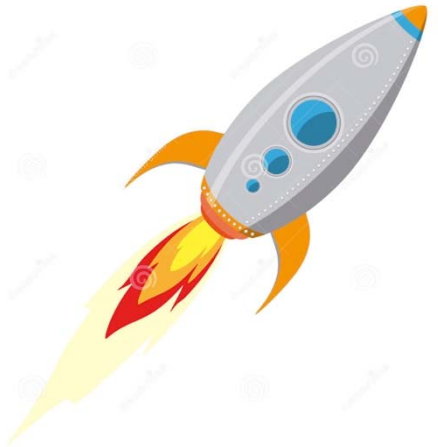
Dimethylamine

↓ sodiun

dimethylnitrosamine

↓ reduction

1,1-dimethylhydrazine



Alkaloids & Derivatives

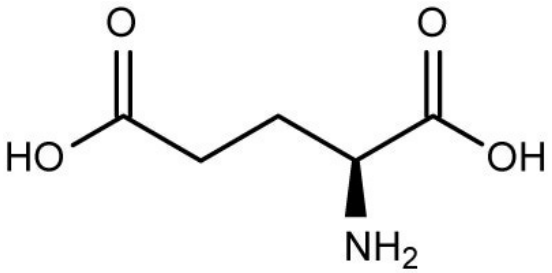
Trigonelline (31.1 µM/mM creatine)

- Concentration in urine based on dietary intake.
  - Fenugreek, oats, peas, hemp seeds, potatoes, coffee, and soybeans
- Prominent in Traditional Chinese Medicine
  - Neuroprotectant, antimigrane, and memory-improving effects
  - Antibacterial, antiviral, and antitumorogenesis effects
- Studies have shown positive effects of trigonelline on:
  - Diabetic complications and CNS diseases
  - Insulin secretion, glucose metabolism, neuron activation, β cell regeneration, and oxidative stress

Amino Acids and Peptides

L-Glutamine (37.2 µM/mM creatine)

- Abundant free amino acid found in high-protein foods
  - Fish, red meat, dairy, beans
- Acts as a metabolic energy source for many cell types
  - Enterocytes, lymphocytes, macrophages, and fibroblasts
- Reduces muscle cramps and recovery time, maintains barrier functions within the digestive tract
  - Common supplement for athletes and bodybuilders
- Prevents bacterial translocation post-surgery, reduces sepsis and organ failure
- Suppression of tumor growth



Carbohydrates

Gluconic Acid (21.5 µM/mM creatine)

- Used as an acidity regulator and food additive
- Used in many “eco-friendly” cleaning products due to its ability to break up mineral deposits

D-Galactose (25.2 µM/mM creatine)

- Food additive, gelling agent (alternative to gelatin)
- Used as an emulsifier and thickening agent in cosmetic lotions and creams

Organic Acids

Formic Acid (26.8 µM/mM creatine)

- Simplest form of carboxylic acid, intermediate in transmethylation
- Commonly associated with bee and ant stings
- Strong antimicrobial properties and food preservation
- Used in common house cleaners
  - Laundry detergent, fabric softeners
- Has the potential for spaceflight applications for the preservation of synthesized foods, or as a cleaning agent

Organic Acids

Taurine (81.0 µM/mM creatine)

- Sulfur-containing AA that is essential for development
  - Produced in the body, dependent on vitamin B6
  - Found in the brain, heart, gall bladder, kidneys
  - Serves as a neurotransmitter, cell membrane stabilizer, and a facilitator for the transport of ions
- Improves athletic and cardiovascular performance
- Intake may be important for maintaining proper physiological function, avoiding depression and other neurological disorders

Potential Applications for Space

Food	Therapeutic	Chemical
Building Block/Synthesis	Building Block/Synthesis	Building Block/Synthesis
Sweeteners/tart flavors	Transport/Uptake	Cleaners
Gelling agents	Emergency Med	Gas scrubbing
Preservatives	Diuretics	Chelators
Dietary Supplement	Pain relief	Surfactants
Reduction of Fatigue	Antimicrobial, antiviral	Solvents
Athletic improvements	Vasodilators	Fuel production
Stamina/Focus	Preventative Agents	Hygiene products
Acidity regulator	Specific Targets	Firefighting materials
	Antihistamines	Lacquers
	Immune Support/Boost	Sealants

Potential Applications for Space

Metabolite and Compound Availability

59 g of urine solids per crewmember per day 

X 5 crew members 

X 1095 days (3 year mission)

323,025 g of urine solids

Approximately 1.5% of total urine solids is 4.5kg of biologically synthesized valuable products.

Thank you!

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