| Analytic factor | Ref | Affected Metabolites |
|-----------------|-----|----------------------|
| **Blood Samples** |     |                      |
| Serum vs. plasma |     |                      |
| [1] | Higher in serum than plasma: charged peptides, peptide fragments; higher in plasma than serum: lysoPI. |
| [2] | Levels varied between serum and plasma: carbohydrates, organic acids, amino acids and lipids. Details can be found in Nishiumi et al., Figure 2. |
| [3] | Higher in serum than plasma: glutamine, arginine, taurine; higher in EDTA plasma vs serum: sarcosine. Details can be found in Paglia et al., Table 1 and Figures 3 and 4. |
| [4] | Found in serum not plasma: sulfate, dipropylacetic acid, leucine, serine, methionine, arachidonic acid, α-glucopyranosiduronic acid, citrate, cellobiose, eicosanoic acid; found in plasma not serum: 2-methylsuccinic acid, α-hydroxybutyric acid, aspartic acid, 4-hydroxyproline, 3-phenylpropanoic acid, tyrosine, α-linolenic acid, urea, citrulline. |
| [5] | Higher in serum than plasma: arginine, PC (C38:1), lysoPC (C16:0), lysoPC (C17:0), lysoPC (C18:0), lysoPC (C18:1), serine, phenylalanine, and glycine. |
| [6] | Methionine sulfoxide higher in serum from a gel-barrier tube than for tube with clot activator. |
| [7] | Variability greater for bile acids and carbohydrates in heparin plasma than in EDTA plasma. Details can be found in Townsend et al., Table 3. |
| [8] | Higher in citrate plasma than EDTA plasma: glutamic acid; higher in EDTA plasma than citrate plasma: sarcosine. Details can be found in Paglia et al., Table 1 and Figures 3 and 4. |
| **Tube additives** |     |                      |
| [8] | Fasting status affected taurine, acylcarnitines, PCs and sphingolipids. Details can be found in Carayol et al., Supplementary Table S1. |
| [7] | Fasting status affected bile acids, purines, pyrimidines, and vitamins. |
| [9] | Fasting status affected bile acids and vitamins. |
| **Fasting status** |     |                      |
| [10] | Metabolites affected include signaling metabolites, amino acids, lipids, carbohydrates, energy metabolites, hormones, vitamins, cofactors, nucleobases, and miscellaneous. Details can be found in Kamlage et al., Table S1. |
| [11] | Hemolysis increased levels of hemoglobin and lysoPCs. |
| **Hemolysis** |     |                      |
| [12] | Time of day of blood collection affected acylcarnitines, lysophospholipids, bilirubin, corticosteroids, and amino acids. Details can be found in Ang et al., Table 1. |
| [9] | Time of day of blood collection affected bile acids and vitamins. |
| [9] | Season of blood collection affected bile acids, purines, pyrimidines and organic acids. |
| [13] | Delayed processing for up to 24 h at either 4°C or 25°C affected glucose, pyruvate, and lactate. |
| [14] | Delayed for up to 8 h at 4°C for up to 8 h affected lactate, pyruvate, and glucose. |
| [6] | Delays for up to 24 h on cold packs affected arginine, methionine, serotonin, hexose and other metabolites. Details can be found in Breier et al., Figures S3-S12. |
| **Collection time of day and season** |     |                      |
| [15] | Delays of up to 36 h at either 4°C or 22°C affected pyruvate, lactate and ornithine. Details can be found in Brunius et al., Supplementary Table S1 and Figure S3. |
| [16] | Delays of up to 24 h at either 4°C or room temperature affected lactate and glucose. |
| [17] | Delays of up to 6 h at room temperature affected aspartate, glutamate, and methionine. |
| [18] | Delays of up to 20 h at room temperature affected metabolites involved in glycolysis/gluconeogenesis/pyruvate metabolism, phospholipid and purine metabolism, the TCA cycle, fatty acids, amino acids and other metabolites. Details can be found in Jain et al., Figures 1-3 and Table 2. |
| [19] | Delays of up to 6 h at 4°C or 22°C affected lactate, glucose, fatty acids, choline and acetone. Details can be found in Jobard et al., Table S1. |
| **Pre-centrifugation time delay and temperature** |     |                      |
| Centrifugation conditions | [26] Centrifugation speed affected glycerophosphocholines and sphingomyelins. |
|---------------------------|---------------------------------------------------------------------------|
| Post-centrifugation time delay and temperature | Delays up to 24 h at either 4°C or 23°C before separation of plasma affected amino acids, carboxylic acids, fatty acids, fatty alcohols, monosaccharides, sugar acids, inositol, palmitic acid, lactic acid, uric acid, α-ketoglutaric acid, creatinine, adenosine-5-monophosphate, cholesterol, taurine, and urea. Details can be found in Malm et al., Figure 2. |
| Buffy-coat contamination | [10] Contamination with Buffy coat affected inositol and lipids. Details can be found in Kamlage et al. 2014, Table S1. |
| Post-processing time delay and temperature | Post-processing delays of up to 36 h at room temperature affected acylcarnitines, amino acids, lysoPCs and PCs. Details can be found in Anton et al., Table 1. |
| | Post-processing delays of up to 36 h at 4°C affected trimethylamine-N-oxide (TMAO). |
| | Post-processing delays of up to 24 h at room temperature affected albumin, triglycerides, LDL/VLDL, proline, citrate and histidine. |
| | Post-processing delays of up to 60 minutes at room temperature affected tyrosine, serine, phenylalanine, aspartic acid, isoleucine, glutamate, methionine, and cysteine. Details can be found in Hirayama et al., Supplementary Table S3. |
| | Post-processing delays of up to 16 h at 4°C, 12°C, or room temperature affected signaling metabolites and amino acids. Details can be found in Kamlage et al. 2014, Table 2. |
| | Post-processing delays of up to 24 h at room temperature affected ribose, aspartate, and phenylalanine. |
| Storage time | [30] Storage for up to 5 years at -80°C affected amino acids, hexoses, acylcarnitines and lipids. Details can be found in Haid et al., Figures S1-2. |
### Freeze/thaw cycles

| Freeze/thaw cycles | Description |
|--------------------|-------------|
| [11]                | Up to four freeze/thaw cycles affected carnitine. |
| [17]               | Up to 10 freeze/thaw cycles affected amino acids. Details can be found in Hirayama et al., Supplemental Figure 6. |
| [25]               | Up to three freeze/thaw cycles affected arachidonic acid. |
| [16]               | Up to five freeze/thaw cycles affected choline, glycerol, methanol, and ethanol. Details can be found in Fliniaux et al., Tables 2 and 3B. |
| [32]               | Up to five freeze/thaw cycles affected lipids, alanine, glucose, acetone, and pyruvate. |
| [17]               | Up to four freeze/thaw cycles affected glycine, methionine, phenylalanine, tryptophan and tyrosine. Details can be found in Anton et al., Supplemental Tables 1 and 2. |
| [6]               | Up to two freeze/thaw cycles affected methionine sulfoxide, amino acids, PCs, and acetylornithine. Details can be found in Breier et al., Table S7. |
| [31]               | Storage for up to 5 years at -80°C affected lysoPCs, acylcarnitines, hypoxanthine, and serotonin. Details can be found in Yang et al., Figure 3. |

### Urine Samples

#### Centrifugation conditions

| Condition | Description |
|-----------|-------------|
| [13]      | Glutamate increases in samples not centrifuged prior to storage and/or analysis. |
| [33]      | Centrifugation affected changes in levels observed after four weeks storage at room temperature of acetate, citrate, formate, succinate, trimethylamine, uracil, urea, creatine, creatinine, and phenylacetylglycine. Details can be found in Saude and Sykes, Tables 2 and 3. |

#### Filtration and additives

| Addition | Description |
|----------|-------------|
| [33]     | Filteration affected changes in levels observed after four weeks storage at room temperature of acetate, benzoate, formate, glycine, Hippurate, lactate, succinate, trimethylamine, uracil, and urea. Details can be found in Saude and Sykes, Tables 2 and 3. |
| [13]     | Addition of sodium azide affected urea, succinate, acetate, lactate, and glutamate/glutamine. Details can be found in Bernini et al., Figure S3. |

#### Time delay and temperature

| Delay | Description |
|-------|-------------|
| [34]  | A delay of one h at room temperature affected succinate. |
| [35]  | Delays up to 24 h at 9°C or 20°C affected arginine, valine, leucine/isoleucine, serine, methionine and hexose H1. Details can be found in Rotter et al., Table 1. |
| [36]  | Delays up to 72 h at room temperature affected creatine, cholic acid, valine, aniline isomer, threonolactone, orotic, trimethylamine oxide, 2-hydroxy-3-methylbutyric acid, methylglycinosine, methininosine, glutamine, dimethylguanosine, 3-methyl-2-oxovaleric acid, N-acetylcytidine, urobininogen, uroblin, ketotrienoic acid glucuronide isomer, hydroxyretinoic acid glucuronide isomer, and ascorbic acid. Details can be found in Roux et al., Table 2. |

#### Storage time and temperature

| Storage | Description |
|---------|-------------|
| [33]    | Storage for four weeks at room temperature affected acetate, benzoate, citrate, creatinine, formate, glycine, Hippurate, lactate, malonate, succinate, trimethylamine, and urea. Details can be found in Saude and Sykes, Table 4. |
| [37]    | Storage for seven days at 4°C affected acetate. |

#### Freeze/thaw cycles

| Cycle | Description |
|-------|-------------|
| [33]  | Two freeze/thaw cycles affected urea. |
| [35]  | Three freeze/thaw cycles affected acylcarnitines and hexose. Details can be found in Rotter et al., Table 4. |

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