Electrolytes, Minerals, Trace Metals, and Water Balance

It's What Plants CRAVE!

Electrolytes

* Free ions in body fluids

Cations travel to cathode	Anions travel to anode
Na^+	Cl^-
K^+	HCO_3^-
Ca^{2+}	PO_4^{2-}
Mg^{2+}	SO_4^{2-}
	Organic acids- ketones, lactate
	Proteins (-) charged

Routinely unmeasured

Routinely unmeasured

Electrolytes in the Body

* Electroneutrality
  * Sum of anions = sum of cations
  * If there is a change in one ion, another must change to counteract

Electrolytes in the Body

* Functions:
  * Maintain osmotic pressure
  * Maintain proper pH
  * Regulate heart and skeletal muscles
  * Redox reactions
  * Participate in enzyme catalyzed reactions

Electrolytes in the Body

* Dietary requirements
  * Vary widely some retained when in short supply
  * Some excreted continuously and need to be replaced (Ca^{2+} and K^{+})
  * Excess → Excretion (usually urine)
  * Abnormal loss?
    * Perspiration, vomiting, diarrhea
    * Salt solution administration corrects

Electrolytes in the Lab

* Measurement of all electrolytes simultaneously is inefficient
  * Routine electrolytes: Na^+ K^+ Cl^- HCO_3^-
  * Together they make…
    * ANION GAP!
    * Form of "QC" for patient specimens

Electrolytes in the Lab

* Anion Gap
  * 95% of extracellular: $(Na^+ + K^+)$ – 85% of extracellular: $(Cl^- + HCO3^-)$ = 8-17 mmol/L

How high does it need to be to care?

  * **$Na^+ - (Cl^- + HCO3^-)$ = 0-15 mmol/L (per the clinic)**

    * What makes up the gap? Ca^{2+} Mg^{2+} PO_4^{2-} SO_4^{2-} organic acids proteins

Electrolytes in the Lab

Increased A.G.	Decreased A.G.
Increase in unmeasured anions: organic acids, retention of anions from kidney failure, increased production of anions	Decrease in unmeasured anions: low albumin (nephrotic, liver failure, H20 intox), severe hypophosphatemia, Increased plamsa H2O (IV therapy)
Decrease in unmeasured cations: severe hypocalcemia, severe hypomagnesemia	Increase in unmeasured cations: hypercalcemia and hypermagnesemia, multiple myeloma proteins?
Severe dehydration: hemoconcentration with high albumin so A.G. must balance it	Administration of cationic substances – polymixin B

Let's Get to Know: Sodium

* Major EXTRACELLULAR cation
  * 90% of ECF cations
  * Responsible for almost 50% of osmolality
  * Central roles: hydration, osmotic pressure, neuromuscular excitability

Let's Get to Know: Sodium

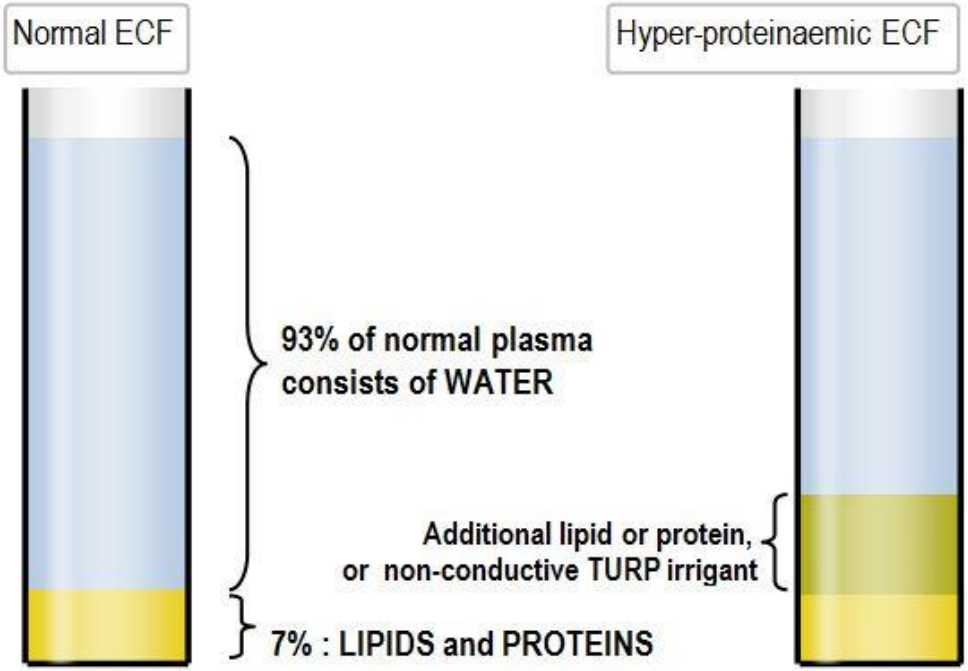
* Homeostasis: Almost all Na^+ is absorbed from GI tract (8-15g of NaCl/day)
* Body needs 58-116 mg/day to extract 1-2 mmol Na^+/day
* Filtered out by glomerulous then 80-85% reabsorbed by distal tubule. Capable of reabsorbing 99% of it!
* Control system?
  * Aldosterone

Let's Get to Know: Sodium

* Aldosterone: produced by adrenal cortex
  * Enhances Na^+ reabsorption from distal tubule
  * Enhances K^+ and H^+ *excretion*
  * Adrenal insufficiency→ Low aldosterone → Na^+ excretion
  * Adrenal hyperproduction → high aldosterone → Na^+ retention
* Normal Na^+ = 136-144 mmol/L
* Renal Threshold = 110-130 mmol/L consequence?

Let's Get to Know: Sodium

* Hyponatremia Na^+<120mmol/L
  * Depletion: prolonged vomiting, sweating, diarrhea, inappropriate diuretic use, 1°or 2° hypoaldosteronism, DKA
    * Alkalosis or alkaline urine (renal tubular acidosis)
  * Dilution: edema, ascites, uncontrolled diabetes, cirrhosis, nephrotic syndrome, malnutrition
  * ***Pseudohyponatremia***
    * glucose >180 mg/dL H^2O shifts to ECF to maintain osmolality
    * high TP or high triglycerides decrease amount of sample that is aqueous solution

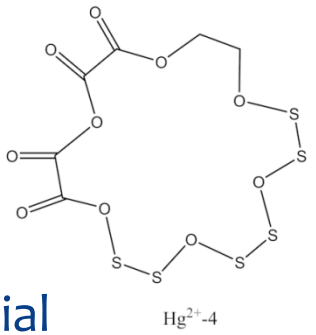

Normal ECF
Hyper-proteinaemic ECF
93% of normal plasma
consists of WATER
Additional lipid or protein,
or non-conductive TURP irrigant
7% : LIPIDS and PROTEINS

Let's Get to Know: Sodium

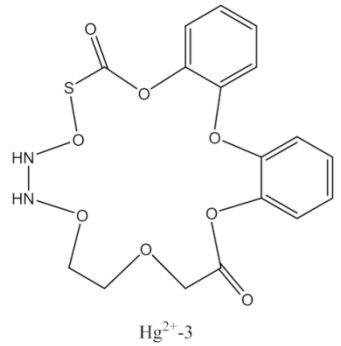
* Hypernatremia: >160 mmol/L
  * Symptoms: thirst, weight loss, flushed skin, decreased BP and oliguria
  * High intake without H_2O, hyperaldosteronism, certain TBIs, treatment of DM, hypercalcemia and hypokalemia

Let's Get to Know: Sodium

* Methods
  * ISE potentiometry – potential difference between Na+ electrode and reference electrode
  * Glass membrane *sensitive* to Na+
  * Crown-ether membrane
    * Na+ passes through ring
    * Attaches to oxygen changes potential
  * Direct vs Indirect (80%)
  * Problems?
    * Protein coating membrane, electrolyte exclusion effect



Hg^{2+}-4



Hg^{2+}-3

Let's Get to Know: Potassium

* Major INTRACELLULAR cation
  * 98% intracellular 23x higher inside RBC
    * Maintained by active ATP pumps (K^+ in RBC Na^+ out)
    * Low cellular permeability
  * Roles: neuromuscular excitability, cardiac contraction, ICF volume, $[H^+]$ regulation
  * Ref: serum 3.5-5.5 mmol/L

    plasma 3.4-4.7 mmol/L

Let's Get to Know: Potassium

* Homeostasis
  * 50-150 mmol/day required
  * Rapidly absorbed, 80-90% of intake excreted
  * Filtered by glomerulous, reabsorbed in the proximal tubule, excreted in the distal tubule (excretion $\propto$ intake)
  * Quick to excrete, slow to conserve
    * Large intracellular stores to supply ECF

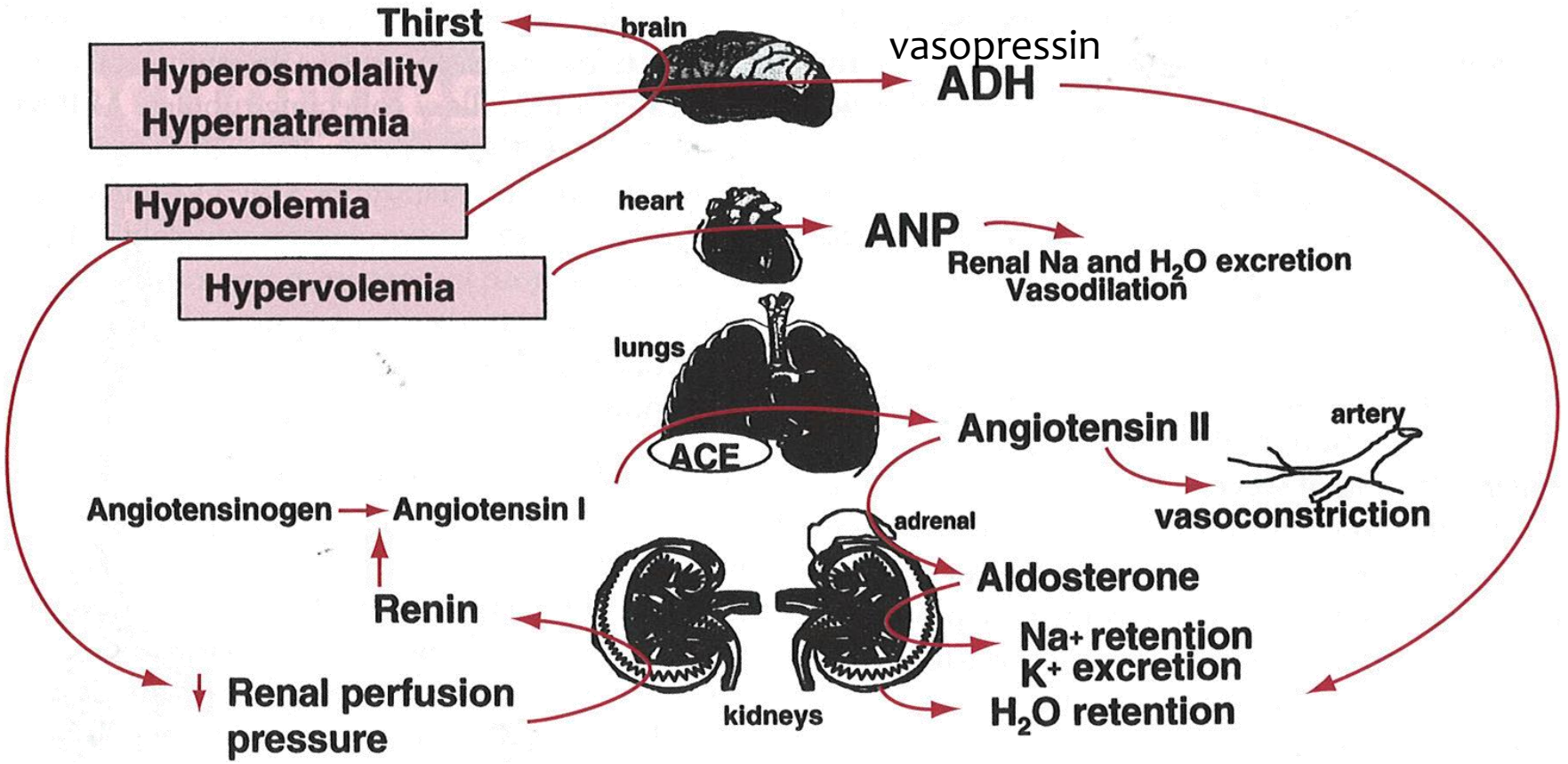
Let's Get to Know: Potassium

Hypokalemia	Hyperkalemia
Decreased intake	High IV infusion rate
Redistribution to ICF (i.e. insulin)	Redistribution into ECF (DKA, MIH, burns, muscle activity)
Loss of K+ rich fluids (vomitting diarrhea	Decreased excretion: renal failure, hypoaldosteronism
Renal loss: hyperaldosteroneism, Cushing's, renal tubular acidosis	

Let's Get to Know: Potassium

* Methods
  * Liquid Ion Exchange Membrane
  * ISE: Valinomycin Membrane similar concept to the crown ether membrane
    * Currently a crown ether using potentiometry with ref. electrode

Water Balance Control



Let's Get to Know Chloride

* Major EXTRACELLULAR anion
* Functions: water distribution, osmotic pressure, hydration, buffering (chloride shift), production of stomach acid

Let's Get to Know: Chloride

* Homeostasis
  * Ingested as salt, almost all absorbed.
  * Glomerulous filters and passively reabsorbs along with Na^+ in proximal tubule
  * Also lost in sweat
  * Aldosterone causes reabsorption of Na^+ Cl^- in sweat glands and kidney
* *Provides least clinical information of "big four"*

Let's Get to Know: Chloride

Ref. Ranges

Blood:96-110mmol/L Urine: 170-250mmol/L Sweat:<40 norm.
40-60 bord.
>60 diag.

Hypochloremia	Hyperchloremia
Salt losing nephritis	Dehydration
Addison's disease crisis low aldo means low Cl-	Renal Tubular Acidosis and Renal failure
Prolonged vomiting	Metabolic Acidosis (low $HCO3^-$)
Metabolic alkalosis (High $HCO3^-$)	High intake
	Diabetes Insipidus (no vasopressin)

Cystic Fibrosis

* Chloride in sweat
  * Diagnostic for cystic fibrosis
    * Patients with CF have Cl 2-5x higher than general pop
    * Hereditary autosomal recessive in caucasian population
      * Chromosome 7 CFTR gene, cells impermeable to Cl^-
    * Characterized by thick secretions that obstruct tissues and organs
      * Lungs, pancreas, sweat glands, GI tract most commonly affected

Cystic Fibrosis Cont'd

* High incidence 1 in 2000 live births among caucasian pop
  * 4-5% of caucasian pop are carriers, most common genetic disorder in North America.
  * DNA analysis possible for high risk families
  * New screening test: Immune Reactive Trypsinogen
  * Aggressive treatment: 1985 27 y.o. 2007 37 y.o.
    * Antibiotics (PSAE), nutritional management, steroids for lungs, mechanical chest percussion

Sweat Chloride

* Method of Collection: Iontophoresis
* Sweat stimulation by pilocarpine (cholinergic)
  * Pilocarpine delivered via (+) electrode
  * Electrolyte solution w/ type I water (-) electrode (completes the circuit)
* Collection onto gauze
* Analysis of sweat chloride
  * Coulometric titration
  * Mercuric nitrate

Sweat Chloride

* Coulometric-amperometric titration (Cotlove)
* Generation of Ag from silver electrode at constant rate
  * Constant generation of Ag^+ from current passed between 2 Ag^+ electrodes
  * Ag^+ combines wil Cl^- in solution
  * AgCl is insoluble precipitate
    * When Cl^- is used up, free Ag^+ causes current to rise, triggers generator shutdown

Let's Get to Know: Chloride

* ISE method for the rest of the specimens
  * Ag+ electrode, PVC membrane
* Mercurimetric titration (Schales and Schales 1953)
  * Not used frequently today
  * Protein free filtrate is titrated w/ mercuric nitrate (known amount) in the presence of diphenylcarbazone (indicator)
    * Free Hg combines with Cl → $HgCl_2$
  * Excess Hg reacts w/ indicator →color change
    * Inversely ∝ [Cl]

Let's Get to Know: Bicarbonate

* **EXTRACELLULAR** anion 21-32 mmol/L
  * We report total CO2 but really measure HCO_3^-
  * Total CO_2: 95-97% HCO_3^-

    3-5% dissolved CO_2

    carbamino compounds, carbonic acid and carbonate ions
  * Function: Buffer, CO_2 transport

Let's Get to Know Bicarbonate

* Increased levels?
  * Metabolic alkalosis or compensation for respiratory acidosis
* Decreased levels?
  * Metabolic acidosis or compensation for respiratory alkalosis

Let's Get to Know: Bicarbonate

* Methodology
  * pCO_2 electrode
    * Silicone rubber membrane permeable to CO_2
    * *Specimen is acidified*, turning all the CO_2 into CO2 gas
    * CO_2 travels across membrane, into buffered solution, is hydrated, and swings the pH which the electrode measures
  * Enzymatic method
    * HCO_3+phosphoenoylpyruvate→oxaloacetate+P_i
    * Oxaloacetate + NADH →malate + NAD^+ 340nm ↓

Let's Get to Know: Ca Mg PO_4

* Ca Mg PO4 all involved in mineral and bone metabolism
* Bone is made of cells and ECM
  * ECM is made of calcium and Type I Collagen
  * 2 types of cells
    * Osteoblasts-synthesize bone matrix
    * Osteoclasts-de-mineralize, digest bone matrix "resorption"
  * 4 processes at work: Mineral deposition, Bone resorption, Intestinal absorption, Renal clearance

Let's Get to Know: Calcium

* 99% exists in bone and teeth
  * Hydroxyapatite $Ca_{10}(PO_4)_6(OH)_2$
* 1% in ECF plasma and soft tissue (8.4-10.2 mg/dL)
* Function: muscle contraction, hormone secretion, coagulation cascade, membrane potentials
* 3 physiological states
  * Free/diffusable Ca^{2+} 50% (1.12-1.30 mmol/L)
  * Protein bound Ca 40%←pH inverse free due to buffering
  * Complexed Ca 10%

Let's Get to Know: Calcium

* Effects of calcium equilibrium
  * Acidosis-decreased pH, decreased binding of Ca^{2+} to protein, increase in free ionized calcium
  * Alkalosis-increased pH, increased biniding of Ca^{2+} to protein, decrease in free ionized calcium
  * pH inversely related to iCa^{2+}

Let's Get to Know: Calcium

* Dietary Sources: Dairy, shellfish, leafy greens
  * Absorption influenced by: gastric acidity, normal fat digestion, adequate supply of Vit. D
    * Net absorption is ~ 25-30%
* Excretion: constantly through urine, sweat, feces
  * Renal threshold 7.0 mg/dL reference range 8.4-10.2 mg/dL
* Hormonal regulation: PTH, calcitonin, 1,25-diOH Vit. D. PTHrP

Calcium Regulation

* PTH parathyroid hormone – controlled by iCa
  * Rapidly cleared by liver and kidneys
    * *N-Terminal fragment*, *Whole molecule*, C-Terminal fragment
  * Functions:
    * stimulates osteoclasts
    * stimulates distal tubules to reabsorb Ca (at expense of PO_4^-)
    * Stimulates vit. D synthesis in kidney
  * Works to increase the total and free Ca, decreases PO_4^- through increased excretion

Calcium Regulation

* Calcitonin-the medullary thyroid answer to PTH
  * Secreted during hypercalcemic states
* Vitamin D
  * 25-hydroxycholecalciferol 23-OH-D_3 inactive form
  * 1,25-$[OH]_2$-D_3 is the active form (kidney production)
    * Increases absorption in gut, enhances osteoclastic effects of PTH, INCREASES PO_4^- retention by kidneys
  * Ergocalciferols D_2 are synthetic

Let's Get to Know: Calcium

* Hypocalcemia
  * Increased neuromuscular excitability
    * Causes cramps, tetany, convulsions, dyspnea, diplopia
  * How? Endocrine (low PTH/pseudo), decreased absorption, increased loss, physiological
* Hypercalcemia
  * Decreased neuromuscular excitability
    * Causes confusion, fatigue, calculi formation
  * How? endocrine (high PTH), malignancy

Let's Get to Know: Calcium

* Methods
  * Spectrophotometric
    * Ca-binding dyes:
      * O-CPC also binds Mg, 8-hydroxyquinoline binds Mg away
      * Aresenao III
        * NM-BAPTA
  * Titration of Fluorescent Ca Complex
    * Calcein
  * ISE
    * Ionized Calciums

Let's Get to Know: Calcium

* Atomic Absorption Spectroscopy
  * Advantages: very few interferences, sensitivity very high
  * Lanthanum Chloride added to prevent $CaPO_4$, TCA precipitates protein
  * Ionization

Let's Get to Know: Magnesium

* INTRACELLULAR cation (1.8-2.5 mg/dL)
* Functions:
  * 300 separate enzymatic reactions require as cofactor
  * Cellular replication and protein synthesis
  * Neuromuscular excitability (inverse relationship)
  * H^+ and K^+ exchange

Let's Get to Know: Magnesium

* Hypomagnesemia
  * Neuromuscular excitability
    * Tremors, hypertension, convulsions, tachycardia, arrythmias
  * How?
    * Malabsorption, alcoholic malnutrition, pancreatitis
    * Increased loss: DKA, diuretics, diarrhea, chronic GN

Let's Get to Know: Magnesium

* Hypermagnesemia
  * Neuromuscular depression
    * Bradycardia, heart block, coma, muscle weakness
  * How?
    * Almost always overadministration Milk of Mg or IV
    * Also dehydration, severe DKA, interference in glomerular filtration of Mg
    * Purposeful critical levels in pregnant women?

Let's Get to Know: Magnesium

* Methods
  * AAS-reference method @ 285nm
    * $LaCl_3$ also needed
  * Spectrophotometric
    * Dye binding color changes EGTA used to chelate Ca^{2+}
      * Calmagite dye 532 nm
      * Methylthymol blue 510 nm
      * Formazen dye & Xylidyl blue 600 nm

Let's Get to Know: Phosphorus

* Intracellular most organic
* Extracellular mostly inorganic
  * 10% protein bound
  * 35% complexed as salt of Na, Ca, Mg
  * 55% free, ionized
* Fuctions: hydroxyapatite, high energy bonds, NADP, phospholipids, nucleic acids, proteins, buffer system

Let's Get to Know: Phosphorus

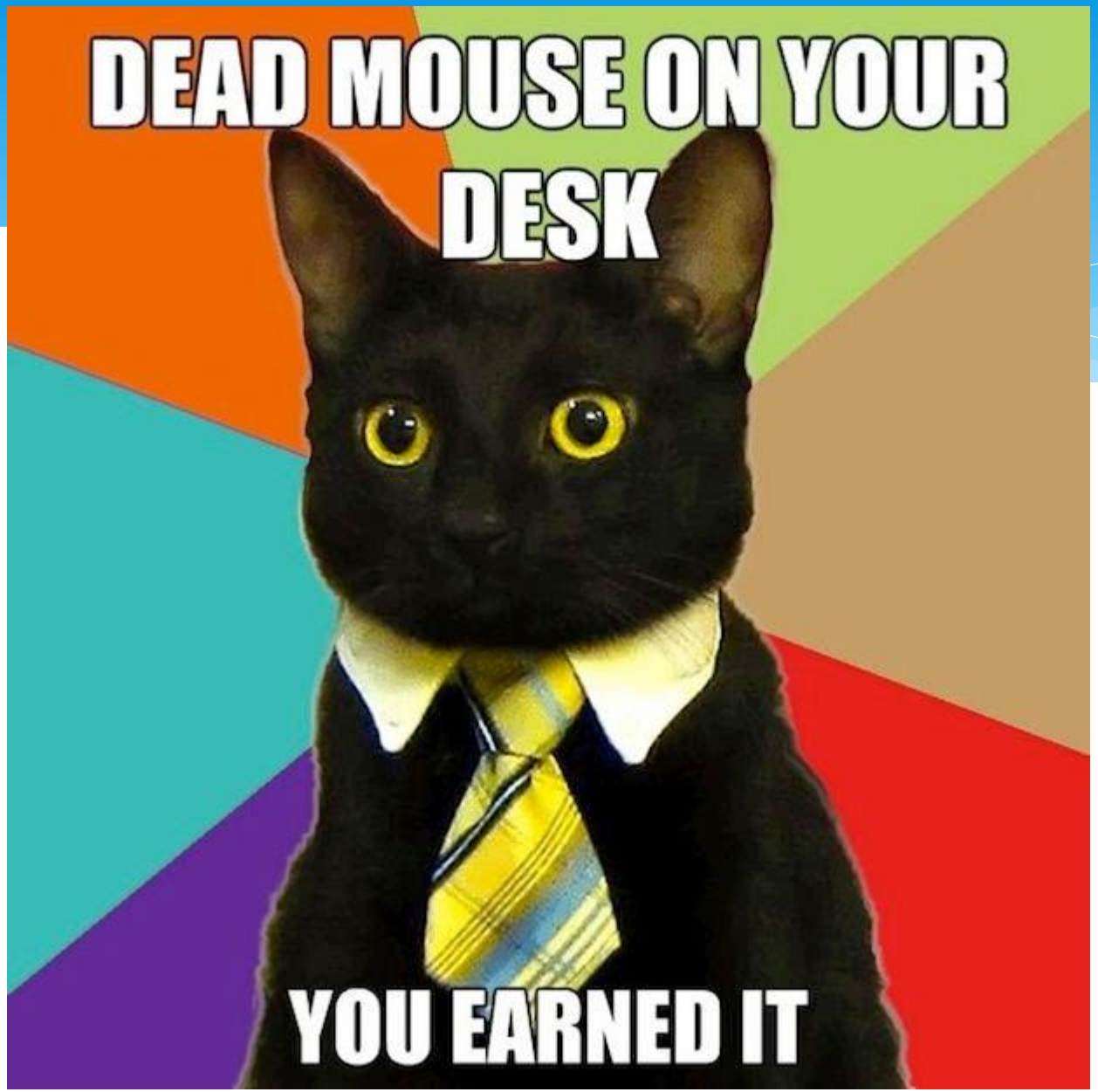
Hypophosphatemia	Hyperphosphatemia
Incidence high for DKA, COPD, TPN, asthma, malignancy	Acute or chronic renal failure, some increased intake
Increased renal excretion, high PTH	Neonates without PTH or vit. D
VERY likely in sepsis	Breakdown of cells

Let's Get to Know: Phosphorus

* Methods
  * Formation of ammonium phosphomolybdate compound
  * Read at 340 nm or turned to molybdenum blue 600-700
  * Hemolysis raises values
* Circadian rhythm, high in morning, low in evening

What Does This Add Up To?

* Serum Osmolality 275-295 mOsm/kg
  * Freezing point depression
    * Supercooled specimen is seeded (stirred, hit)
    * When temperature equilibrium is reached midway between freezing and thawing the freezing temperature can be read
      * Relationship of osmotically active solutes is is direct to freezing point depression
  * $2\mathrm{Na} + \frac{\mathrm{glucose\ (mg/dL)}}{20} + \frac{\mathrm{BUN\ (mg/dL)}}{3}$
  * Osmole? Particle #s (like normality instead of molarity)
  * Osmolal gap? BHB, alcohols, lactic acid


DEAD MOUSE ON YOUR DESK
YOU EARNED IT