WATER AND ION BALANCE

- Optimal physiological function requires that hydration state and solute concentration within the body be maintained within rather narrow limits.
- Problem = proper internal conditions are almost always very different from environmental conditions

AQUATIC ENVIRONMENTS
- Osmolarity = measure of the solute concentration in a solution (osmoles/liter; 1 Osmole = 1 mole of dissolved solutes per liter); depends on the number of dissolved solutes present. The greater the number of dissolved particles, the greater the osmolarity
- Hyperosmotic = body fluids more concentrated than environment
- Isosmotic = body fluids equal to solute concentration in environment
- Hyposmotic = body fluids less concentrated than environment

- Most invertebrates are isosmotic with seawater (osmoconformers), but they often regulate specific solute concentrations to some extent **(Overhead, Willmer et al., p. 78)**
- FW animals are all osmoregulators (some better than others) that maintain hyperosmotic body fluids **(Overhead, Willmer et al., p. 78)**
- Marine Vertebrates are almost always hyposmotic osmoregulators. There are a couple of examples of isosmotic body fluids, but these organisms still regulate the concentration of specific solutes **(Handout, Composition of Vertebrate Body Fluids)**
- When thinking about water relations in aquatic environments, REMEMBER that water always moves toward a higher salt concentration (by osmosis), while solutes always move down a concentration gradient (by diffusion)

OSMOREGULATION IN FW vs. SW (Vertebrates)
- FW: Animal is hyperosmotic to environment; Problem = taking on water, losing salts
- SW: Animal is hyposmotic to environment; Problem = losing water, taking on salts
- **(Handout, S-N p. 319-320)**

Solutions to Osmoregulatory Problems in Aquatic Environments

1) *Freshwater Fish*
   a. Kidney with large glomeruli that produce high filtration rates → produce large volumes of very dilute urine
   b. Chloride cells in gills capable of active solute uptake (Na^+, Cl^- follows)
2) *Saltwater Teleosts (Advanced Bony Fish)*
   a. Aglomerular kidney or very reduced glomerulus → essentially no filtration (NH_4^+ lost at gills)
   b. Drink SW to gain water. Actively pump Na^+ across digestive tract epithelium, water passively follows
   c. Chloride cells in gills actively pump salts out (Na^+)
   d. Divalent ions (Mg^{2+}, SO_4^{2-}) actively secreted into kidney tubules, excreted with very low volume of urine
3) *Special Saltwater Adaptations*
   A. Hagfish = a primitive jawless fish
      1. glomerular kidney with high filtration rates
      2. isosmotic with SW; tolerates high salt concentration, but regulate concentrations of specific solutes to some extent
      3. regulate divalent ion concentration at skin by actively pumping out Mg^{2+}, Ca^{2+}

   B. Elasmobranchs, Coelacanth
      1. isosmotic with SW, but qualitatively very different. Regulate most ions at normal vertebrate levels

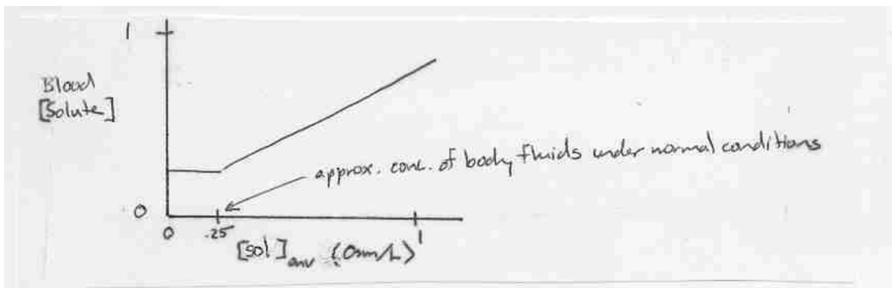
2. isosmotic condition achieved by adding large amounts of urea to blood. Tolerate very high urea levels (100 X that in mammals). Also increased TMAO levels that counteract destabilizing effects of urea on macromolecules (i.e., proteins)
3. rectal glands secrete excess Na^+

(Handout – Vertebrate Body Fluid Composition)

C. Aquatic Amphibians

1. FW amphibians osmoregulate similar to FW fish. Skin serves as site for active ion uptake. Na^+ also actively pumped from the kidney tubule back to blood.
2. Crab-eating Frog (*Rana cancrivora*) – SE Asia. Lives in coastal mangrove swamps and regularly enters SW.

- Adds large amount of urea (plus some Na^+) to body fluids to become isosmotic. Elevates urea by reducing urine volume, rather than pumping urea to blood.



3. Green Toad (Europe) – *Bufo viridis*, tolerates brackish water (up to 80% SW levels).

- Has high plasma osmolarity under normal conditions (high Na^+ and urea), becomes isosmotic by addition of urea, along with some Na^+.

TERRESTRIAL ENVIRONMENTS

- **Major Problem** = dehydration
- Water Loss occurs via 4 avenues:
  a) cutaneous
  b) respiratory
  c) urine
  d) feces
- Cutaneous and Respiratory avenues together constitute *Evaporative Water Loss* (EWL)

1. Cutaneous Route

- Very high in amphibians (a major problem for them) due to their moist skin
- Lower in animals with keratinized skins, which decreases permeability
- CWL tends to be higher percentage-wise for reptiles than for birds and mammals, since RWL is elevated in endotherms because of their high MR and associated high ventilation rates.
- CWL due to sweating can be a problem for a animals that sweat in hot, dry environments to keep cool

2. Respiratory Route

- Major route for EWL in mammals and birds at most temperatures.
- Also important for reptiles (25-50% of EWL), especially at high temperatures.
- Less important for amphibians because of their moist skin (**Overhead, Willmer et al. p. 81)**.

TOTAL EWL → dependent on *Vapor Pressure Deficit (VPD)*

$$VPD_{Ta} = WVP\ (100\%\ RH)_{Tb} - WVP\ (\%RH)_{Ta}$$

↑
Inspired air saturated almost immediately at T_b of organism

EXAMPLES:
Under what conditions would drying be faster? (Refer to Water Vapor Pressure vs. Temp. Handout)

I. 1) Lizard at 35°C, RH = 50%
2) Lizard at 10°C, RH = 10%
- Assume $T_b = T_a$ for lizard

- 35°C, RH = 50% → VPD = 40 – 20 = **20** (40 = 100% RH at 35°C, 20 = 50% RH at 35°C)
- 10°C, RH = 10% → VPD = 10 – 1 = **9**
- Drying is faster under the first set of conditions for the lizard

II. Mammal under the same conditions. Assume T_b = 37°C.

a) VPD = 45 – 20 = **25** (45 = 100% RH at 37°C)
b) VPD = 45 – 1 = **44**

- Drying is faster under the second set of conditions in the mammal

- *In considering water loss, always remember that warm air holds more moisture than cold air.*
- RWL is directly dependent on ventilation volume and VPD.

MECHANISMS FOR DECREASING RWL

1. *Increasing oxygen extraction efficiency* – this allows the animal to use a smaller ventilation volume to extract the same amount of oxygen

- Not used in mammals. Related mammals have similar oxygen extraction efficiencies, regardless of the habitat in which they live.
- Birds with slightly higher oxygen extraction efficiencies than mammals
  - O_2Ex does increase in some birds (but not in others) with decreasing temperatures
  - Altitude-adapted birds sometimes show an elevated O_2Ex, but not in all cases

2. *Reduction of Temperature (and therefore water content) of Exhaled Air by Countercurrent Heat Exchange*

- Inspired air warmed to T_b and humidified
- During inhalation, walls of nasal passages lose heat to air passing over them (both by evaporation and by the lower temperature, usually, of the air passing over them) → cools nasal passages
- During exhalation, warm saturated air from lungs loses heat as it passes over nasal passages → water condenses on walls and is returned to the body water pool
- This mechanism is more effective in mammals and birds than in reptiles because of higher surface areas for exchange.
- <u>Nasal Turbinates</u> = thin, highly convoluted sheets of bone associated with the nasal passages in mammals and birds. These act to increase the efficiency of countercurrent water conservation by increasing the surface area for evaporative cooling. This has the effect of cooling exhaled air to temperatures just slightly above T_a on exhalation. **(Overheads – Willmer et al., p. 583, 106)**
- Reptiles lack turbinates, although some desert forms may show curved (and therefore slightly longer) nasal passages.

OSMOREGULATION IN MARINE AIR-BREATHING VERTEBRATES

Problem = Food with high salt content and often drink seawater, so have a high salt load

Solutions:

1. *Kidney* = excretes hyperosmotic urine (relative to body fluids) in birds and, especially, mammals
2. *Extrarenal Salt Glands* = produce highly concentrated fluid containing mainly Na+ and Cl-. Intermittent function in response to high salt loads.

- Present in several marine reptiles and all marine birds, but not in mammals as their kidneys are sufficient for salt removal.
    a) Marine Iguana – located near anterior nasal cavity
    b) Sea Turtles – open to corner of orbit
    c) Sea Snakes – open to oral cavity
    d) SW Crocodiles – salt glands distributed over surface of tongue
    e) Birds – open near external nares (nasal salt glands)
- Mechanism for concentrating exudate in birds:
    1) Vascularization is countercurrent to secretory tubule → isosmotic secretion
    2) Concentrating power provided by active (ATP-requiring) reabsorption of water by post-secretory tubule. Probably involves uptake of solutes, with water passively following, then active pumping of salts back into tubule.

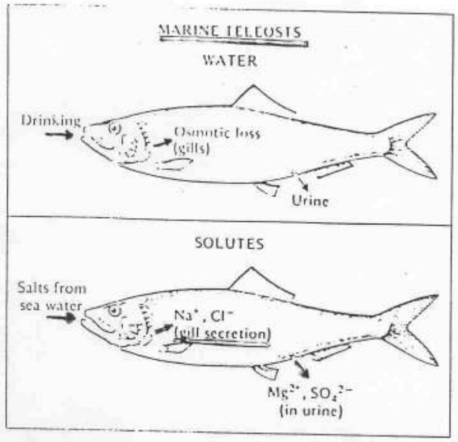
    (See Overhead, p. 436 Willmer et al. and Handout, Phillips et al., p. 96)

REPLACEMENT OF LOST WATER

1. Drinking – most familiar method. For many birds, EWL approximately equals water consumption under standard conditions. Total water consumption is therefore related to body size, and smaller birds have a relatively higher water intake. **(Handout – Phillips et al., p. 284)**
2. Preformed Water in Food – fresh vegetable matter has the highest water content
3. Metabolic Water = water formed from the oxidation of substrates
    a) Very important for animals living under dry conditions. In these animals, metabolic water is the most important source of water gain.
    b) Example: *Kangaroo Rats* – live in dry habitats and eat dry foods, yet never drink → preformed water in food (10%) and metabolic water (90%) are sufficient to meet water demands.
        - Kidney capable of producing very concentrated urine, so WL via urine is very low.
        - Also have highly developed and very effective nasal turbinates to reduce REWL

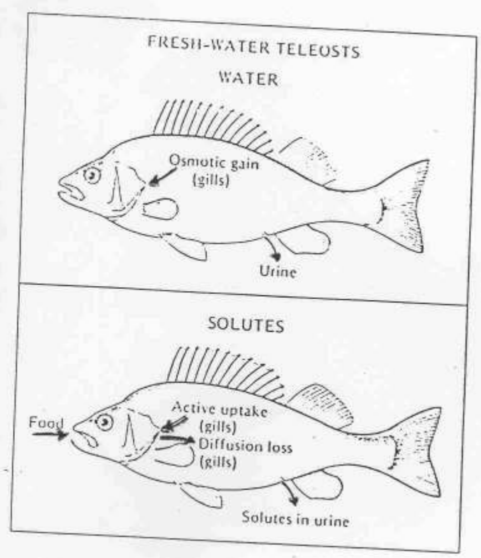
COMPOSITION OF ENVIRONMENTAL AND BODY FLUIDS

	Salt Concentration				
	meq/liter				Δ F.P. (#°C below 0°C)
	Na	K	Ca	Mg	(1 osmolar = - 1.86°C)
Environments					
1) Marine	450-500	8-10	20	110	1.86
2) Fresh water	6-20	5	8-100	0-4	.01
Vertebrate Plasma					
1) Myxini (Hagfish)	570	7	9	24	1.86
2) Elasmobranchs (marine)	150-250	8	10	2	1.85 to 1.92
(F.W.)	150-200				.9
3) Teleosts (all)	150-220	8	10	2	marine- .6 to .8 F.W.- .5 to .55
4) Amphibia	100-110	4	2		.5
5) Reptiles	120-170	4	3		.4 to .7
6) Birds and mammals	140-170	4-10	10	3-5	.5 to .6
Intracellular Fluid					
1) Mammal muscle	27	96		18	.5
2) Frog muscle	10	100	3	15	.5



Salt Water = saltier than blood

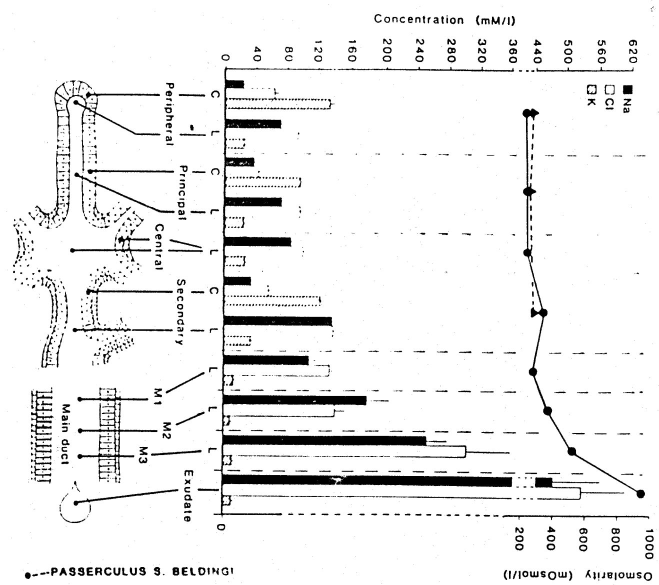
PROBLEM = taking on salts, losing water

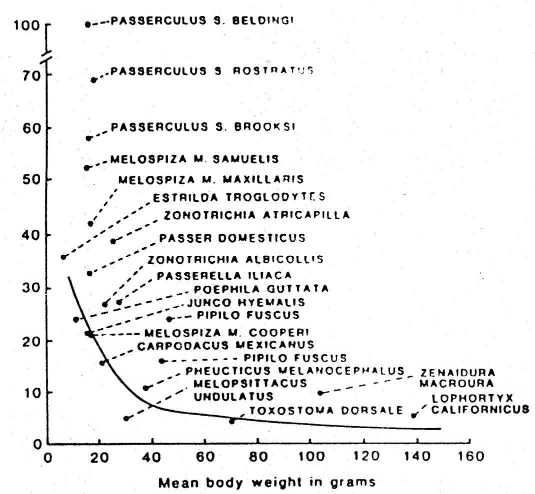


Fresh Water = less salty than blood

PROBLEM = taking on too much water, losing salts

Schmidt-Nielsen p. 319 – 320





NITROGENOUS WASTES AND WATER BALANCE

- An additional osmoregulatory problem for animals is getting rid of nitrogen-containing wastes produced from the removal of amino groups during protein catabolism.
- Removal of amino groups produces ammonia (NH_4^+), which is very toxic, so ammonia must be rapidly excreted or detoxified.

3 Major Nitrogenous Waste Products Exist among Vertebrates:

1) Ammonia – *Ammonotelic* organisms are aquatic. Due to the high solubility of ammonia in water and its small molecular size, ammonia readily diffuses to the aquatic environment at gill or skin surface. Only a small portion is excreted by the kidney. (Occurs in aquatic inverts., teleost fish, cyclostomes)
2) Urea – *Ureotelic* organisms convert ammonia to urea in liver (2N:1C ratio). Energetically more expensive to produce urea, but it is soluble in water and has a low toxicity so it can be tolerated at higher levels than ammonia and excreted as liquid urine. High urea concentration in urine cuts down on excretory water loss. (Occurs in Elasmobranchs, turtles, most amphibians, mammals, and a few invertebrates as well)
3) Uric Acid – *Uricotelic* organisms convert ammonia to uric acid (4N:5C). Energetically the most expensive to produce, but low toxicity and only slightly soluble in water. Removal of water from urine causes uric acid to precipitate, so it is excreted as a semisolid paste with little water loss → serves as an effective water conserving mechanism.

- Some insects deposit uric acid within body (e.g., fat body) so that no water is lost via excretion.

- Occurs in Insects, birds, and reptiles.

(SEE HANDOUT FOR REVIEW)

THE VERTEBRATE KIDNEY

Kidney = major excretory organ in vertebrates, functions by *filtration/reabsorption* mechanism, plus some *secretion*; except for aglomerular kidney, which operates only by secretion.

1) Filtration = occurs at glomerulus where high blood pressure forces fluid from capillaries which then enters kidney tubule (nephron) across wall of Bowman's capsule.
2) Once in the nephron, the filtrate can be modified by tubular *reabsorption* and *secretion* to produce the final urine. In terrestrial vertebrates, often 99% or more of original filtrate volume is reabsorbed.

- Kidneys of all vertebrates capable of producing a urine that is hyposmotic or isosmotic with blood. However, only kidneys in mammals and some birds can produce hyperosmotic urine.

(SEE HANDOUT ON U:P RATIOS IN MAMMALS AND BIRDS)

- The vertebrate kidney posterior to the renal corpuscle is divided into 2 parts:
  1. Proximal Tubule = sodium, glucose, and water are reabsorbed
  2. Distal Tubule = sodium pumped out, water reabsorbed
- These merge into Collecting Ducts that drain to ureters and out of the body through the cloaca (or its derivatives)

Mammalian Kidney (Bird kidney with similar design, but less well developed)
- Structural difference = Loop of Henle positioned between proximal and distal tubules. Allows concentration of urine by countercurrent multiplier system.
- There are actually 2 types of nephrons in the mammalian (and avian) kidney
  1. Cortical Nephrons = relegated to outer cortex, short loops of Henle with only a very minor role in concentrating the urine.
  2. Juxtamedullary Nephrons = proximal and distal tubules in cortex, but L of H extends deep into medulla. These provide concentrating ability, so the higher percentage of these tubules, the greater the concentrating power of the kidney.

Mechanism of Concentration

1) Ability to concentrate is related to the *length of the Loop of Henle* and the *solute concentration gradient* in interstitial fluid of kidney. This concentration gradient increases with depth into the medulla.
2) In the proximal tubule, water flows passively out of tubule as increasing solute concentrations are encountered. This continues in *descending loop of Henle*. Descending loop is permeable to water, impermeable to ions so water moves out and is picked up by *vasa recta* = blood vessels surrounding kidney tubule running in opposite direction (countercurrent) to filtrate flow. Vasa recta are freely permeable to water and ions. Concentration at bottom of L of H is very high, such that approx. 80% of water is removed by this point.
3) *Ascending Loop of Henle* – thin portion permeable to sodium, impermeable to water, so sodium moves out. Thick ascending portion is impermeable to water and ions, but sodium is actively pumped out. By the time filtrate reaches the distal tubule, most sodium has been removed and filtrate is actually hyposmotic to plasma.
4) *Distal Tubule* – permeable to water, impermeable to ions, but active removal of sodium continues. Water moves out since hyposmotic at this point.
5) *Collecting Duct* – impermeable to ions, permeable to water, inner medullary portion permeable to urea. Water continues to move out and sodium continues to be actively pumped out. In inner medullary region, urea diffuses out into interstitium to comprise the main solute in the concentration gradient. Water moves out as the collecting ducts pass deeper into the medulla, so urine increases in concentration (primarily urea).

- ADH (Vasopression) from posterior pituitary regulates permeability of collecting ducts to water. Under dehydrated conditions, increased ADH causes increased permeability and increased reabsorption of water (and decreased urine production). Homologous hormone in non-mammal vertebrates – AVT.

6) Vasa recta carries away reabsorbed solutes and water.

(SEE OVERHEADS)

Adaptation of Concentrating Power

1) Number of juxtamedullary nephrons and length of Loop of Henle correlated with concentrating power. Mostly juxtamedullary nephrons in desert-adapted, with relatively long loops. Mesic mammals (e.g., beaver) with mostly cortical nephrons and little concentrating power.
2) Bird kidneys have nephrons with and without Loops of Henle. Relative numbers determine concentrating power. Desert or marine birds have higher numbers of nephrons with L of H and an increased concentrating power.
3) *Body Size Effects* on Urine Concentration – if concentrating power is dependent on the length of the Loop of Henle, how do small mammals (e.g., rodents) with L of H that is short in absolute terms generate such concentrated urine?

- Because small mammals have high mass-specific metabolic rates, the tubule cells are capable of more intense active transport, so they can generate a higher concentration gradient per unit length than large mammals.
- Additionally, there is a strong correlation of urine concentrating ability with the relative medullary area.

(SEE HANDOUT)

Adaptations to Aridity in Amphibians

1) Distribution of water among body compartments in vertebrates
   Intracellular – 75%
   Intercellular – 15%
   Circulatory – 10%
2) As far as water loss is concerned, amphibian skin (with a few exceptions) behaves as a *free water surface*. This means that the skin presents no significant barrier to evaporation. Consequently, EWL rates are very high in amphibians.

- Exceptions = *Chiromantis* (South Africa) and *Phyllomedusa* (South America) are "waterproof" frogs that have markedly reduced rates of EWL. *Phyllomedusa* has skin glands that secrete a waxy waterproofing substance. *Chiromantis* lacks wax glands so reduction in EWL must occur by some other mechanism – unknown at present.

3) Desert Amphibians – water loss rates are the same as for mesic amphibians.
   a) Tolerance to Dehydration – amphibians in general are more tolerant of dehydration than other vertebrates. Degree of dehydration tolerance is associated with degree of terrestriality.
   (SEE HANDOUT)

- Xeric species tolerate greater dehydration (up to slightly greater than 50% of total body water lost) than mesic species (about 20-30%).
- Increased tolerance accomplished by systematic toleration of hyperosmolarity (mechanism unknown) and by cardiovascular specialization (tolerance to increased solute levels and effective plasma volume regulation).
- Plasma volume maintained by preferential loss of water from extracellular and intracellular compartments before circulatory compartment in *Bufo* and *Scaphiopus*.

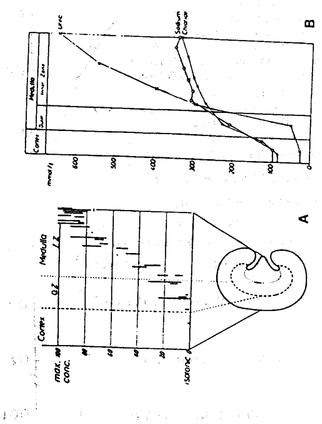
   b) Water Uptake from:
      (1) *Bladder* – large volumes of dilute urine stored in bladder (up to 130% of body mass in Australian Water-holding Frog)

- During dehydration reabsorption of water from bladder is effective in offsetting EWL (AVT regulates bladder permeability to water).

      (2) *Substrate* – can locate and absorb water from moisture at soil surface or on wet or dewy vegetation or rocks. Assume water-absorbing posture with hind legs splayed and ventral surface of legs and abdomen pressed to substrate. Aquaporins (water channels) in skin are involved.

- Burrowing amphibians capable of taking up water from the soil if water potential of the soil (determined by moisture content and the force with which soil particles hold water) is greater than the water potential of the animal (determined by osmotic concentration of body fluids). In many desert environments, for amphibians to pick up moisture from the soil requires burrowing to substantial depths (e.g., spadefoot toads in se Arizona burrow to average depth of 54 cm, 91 cm during the dry season).
- Xeric species can absorb water from soil with lower water potential than mesic species.

c) <u>Cocoon-forming Burrowing Amphibians</u> – several burrowing frogs and an aquatic salamander (*Siren*) estivate in burrows during drought and form a cocoon from multiple layers of shed stratum corneum of epidermis. Cocoon serves to markedly reduce



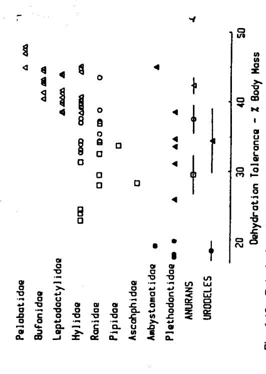


Fig. 6.12 Dehydration tolerance for amphibian species differing in degree of terrestriality. *Squares*, aquatic; *circles*, semiterrestrial; *triangles*, primarily terrestrial. Anurans are represented by *open symbols*, urodeles by *filled symbols*. Horizontal lines are 95% confidence intervals for means of all species of anurans and all species of urodeles of a particular degree of terrestriality. See text for sources.