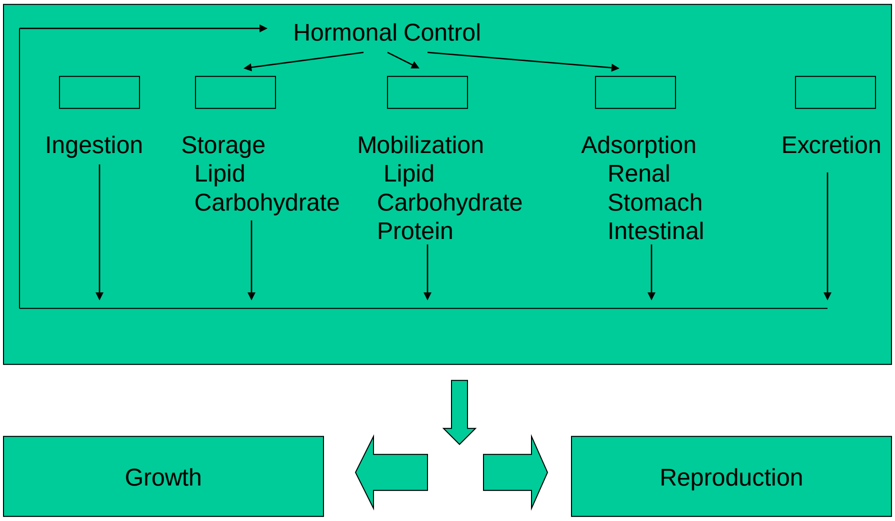
Excretion

- Consumption = Growth + (Metabolism + SDA) + F(egestion) + **U (excretion)**

Energetics – Processes



Hormonal Control

Ingestion

Storage
Lipid
Carbohydrate

Mobilization
Lipid
Carbohydrate
Protein

Adsorption
Renal
Stomach
Intestinal

Excretion

Growth

Reproduction

Assimilated materials must be metabolized and excreted

- Major end products are water, CO_2 and ammonia, with small amounts of urea, creatine, creatinine, and uric acid.
- Lipids and carbohydrates are metabolized directly to water and CO_2.
- Proteins peptides and amino acids are deaminated to yield ammonia and carbon chain oxidized to CO_2 and water.

- Nucleic acids are primarily metabolized to creatine and creatinine.
- Ammonia (NH_3) is major nitrogenous waste product.
- Salmonids fed dry diets produce 25 – 35 g of ammonia per/kg feed consumed

Ammonia Production

- Ammonia excretion is bioenegetically more efficient than urea and uric acid.
- But ammonia is more toxic
- Most ammonia is produced in liver, converted to nontoxic form in blood as glutamine, and transported to gills where it diffuses rapidly into water as NH_3.<25% is synthesized at gills.

$NH_3 + H_2O \leftrightarrow NH_4^+ + OH^-$

- Ammonia is a gas (NH_3) and ion ammonium NH_4, the sum of which is the total ammonia
- The degree to which ammonia forms the ammonium ion increases upon lowering the pH of the solution—
- Temperature and salinity also affect the proportion of NH_4^+.

Dissociation of Ammonia

Henderson-Hasselbalch Equation

- % $NH_3 \Leftrightarrow 100/(1 + \text{antilog}(pKa - pH))$

- $pH = pKa + \log([NH_3/NH^{+4}])$

- The pKa is high (about 9.7 at 10°C)
- Thus little ammonia will be toxic unless water is quite alkaline (pH >> 7)

Summary Relationships

- The activity of aqueous ammonia also is <u>lower</u> at low temperatures and <u>higher</u> at warm temperatures.
- At low temperatures and low pH the activity as NH_3 is even lower, and as NH_4^+ is even higher.
- Therefore, sensitive aquatic organisms can tolerate a higher total “ammonium-N plus ammonia-N” at low temperatures than at high temperatures due to much less aqueous NH_3 being present in the water.

Critical Concentrations

- Un-ionized NH_3 molecules are toxic and can cause gill damage if higher than 0.1- 0.5 mg/L
- Amt toxic NH_3 formed is function of water pH, temperature and salinity or TDS.
- Due to solubility restraining movement is not easy.

$$\% \ NH_3 \leftrightarrow 100/(1 + \text{antilog}(pKa\text{-}pH))$$

- Use charts to get an idea of this outcome
- Ammonia perturbations can affect nerve cell function, ion transport process, metabolism , pH.

	Temperatures			
pH	**5**	**10**	**15**	**20**
6.0	**0.01**	**0.02**	**0.03**	**0.04**
6.4	**0.03**	**0.05**	**0.07**	**0.10**
6.8	**0.08**	**0.12**	**0.17**	**0.25**
7.0	**0.13**	**0.18**	**0.27**	**0.40**
7.2	**0.20**	**0.29**	**0.43**	**0.63**
7.4	**0.32**	**0.47**	**0.69**	**1.0**
7.6	**0.50**	**0.74**	**1.08**	**1.60**
7.8	**0.79**	**1.16**	**1.71**	**2.45**
8.0	**1.24**	**1.83**	**2.68**	**3.83**
8.2	**1.96**	**2.87**	**4.18**	**5.93**
8.4	**3.07**	**4.47**	**6.47**	**9.09**
8.6	**4.78**	**6.90**	**9.88**	**13.68**
8.8	**7.36**	**10.51**	**14.80**	**20.07**
9.0	**11.18**	**15.70**	**21.59**	**28.47**

Freshwater % unionized ammonia by water temperatures

From Wedemeyer

Freshwater

- Copious urine because of continuous osmotic influx of water.
- Serves to dilute small amounts of ammonia excreted by kidneys

Seawater

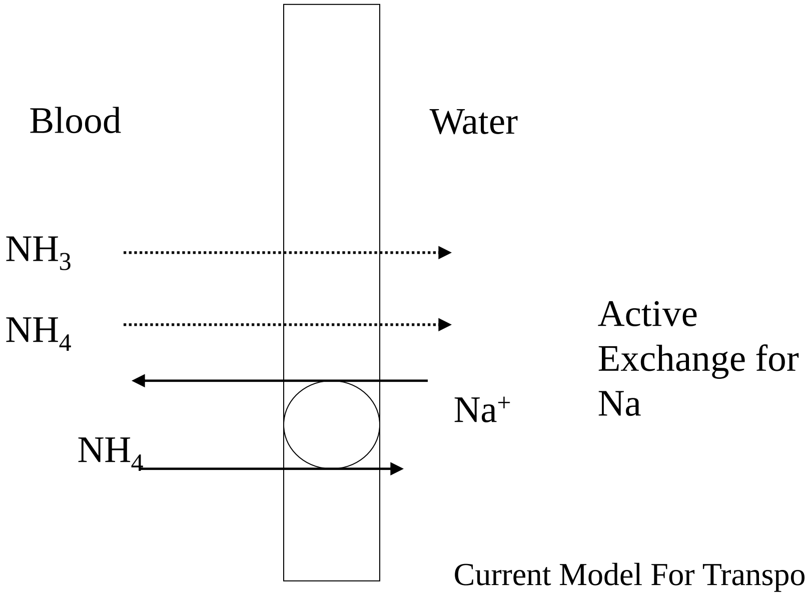
- Less urine produced and more ammonia excreted through the gills
- Sharks and rays produce urea, but as a way of increasing osmolarity of body fluids to approximately concentration seawater.

Water temperature (°C) Seawater relationship % unionized ammonia.

pH	5	10	15	20
7.2	0.17	0.24	0.35	0.51
7.4	0.26	0.38	0.56	0.81
7.6	0.42	0.60	0.88	1.27
7.8	0.66	0.95	1.39	2.00
8.0	1.04	1.49	2.19	3.13
8.2	1.63	2.34	3.43	4.88
8.4	2.56	3.66	5.32	7.52
8.6	4.00	5.68	8.18	11.41
8.8	6.20	8.72	12.38	16.96
9.0	9.48	13.15	18.29	24.45

Transport

- Some blood NH_4 is exchanged by active transport for Na+ in the water. If water pH and ammonia concentration of water are lower than blood, freshwater fish can readily excrete blood ammonia.
- If water pH is more alkaline than blood and dissolved ammonia concentration higher, the outward flow of ammonia is hindered.



Current Model For Transport

Excretion

- Most important factor affecting rate of total nitrogen excretion is feeding
- Gills account for 80- 95% of whole body nitrogen excretion
- Urea sometimes contributes and can be more important in some cases in freshwater

Renal Excretion

- Glomerular recruitment occurs much as lamelar recuritment
- Still, renal nitrogen excretion is not used in high percentage among fishes.

Osmotic and Ionic Regulation

- Water and small non electrolytes like urea and ammonia can move through plasma membrane at internal/external interface by moving though lipid bilayer
- Ions such as sodium, potassium and chloride will not pass easily across membrane without help of membrane transport proteins that span the lipid bilayer.

Extra renal epithelial tissues

- Gills
- Integument
- Gut
- Urinary bladder
- Specific salt glands (rectal gland in elasmobranchs)

Ionic Balance - Seawater

- Oral Ingestion. Gradient may be large. Hyperosmotic external environment withdraws water
- Drinking rates 3-10 X higher than in freshwater adapted fishes.
- Bulk of water and salt uptake is in small intestine, following osmotically the transport of ions.

Oral

- NaCl and water absorbed across intestine and gut absorption is direct function of osmolarity to which fish has adapted.

Gills & Chloride Cells

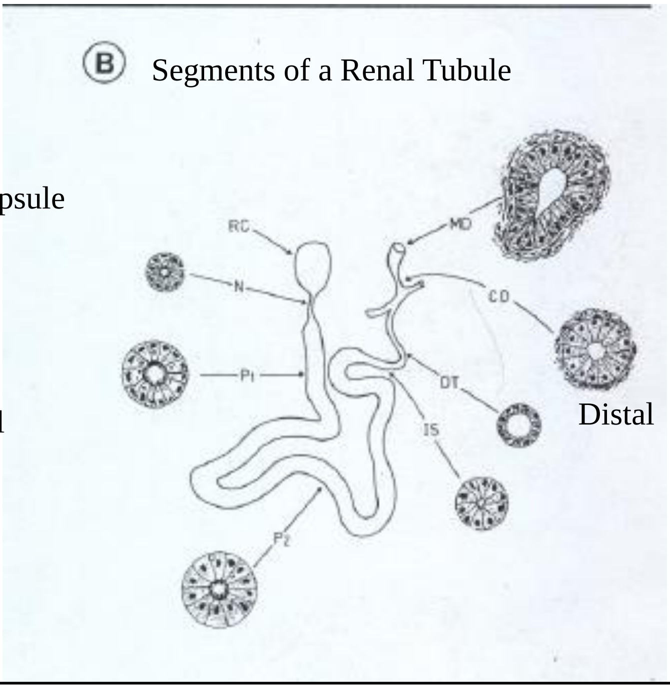
- Chloride- secreting cell (chloride cell)
- Mitochondrial rich
- Multicellular complexes
- Model of operation

Kidney(excretory)

- Renal corpuscle- glomerulus
- Neck
- Proximal segment
- Intermediate segment
- Distal segment
- Collecting tubule

Freshwater Filtration

- Urine less concentrated than blood
- Large glomeruli that filter water from blood
- Reabsorbed from proximal tubule
- Monovalent ions reabsorbed at both proximal and distal tubule segments



Segments of a Renal Tubule

Renal capsule

Proximal

Distal

Seawater Filtration

- Less urine production. Proximal tubules long- higher metabolic cost
- More distal part secretes divalent ions Mg and SO4 into tubules

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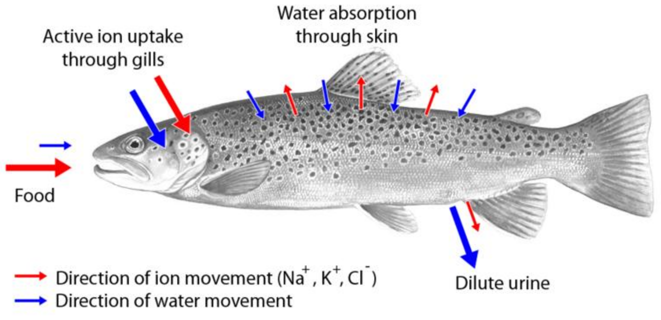
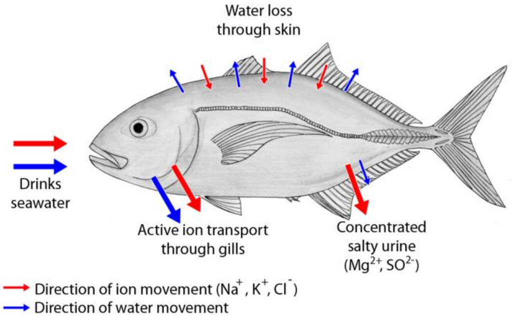
Fresh water

- Bailing out
- Conserving by re-sorption

Sea water

- Reduction of exposure
- Removal of toxics of Mg SO4
- Other wastes that can not pass branchial





Diadromous Fish

- Reproduce Freshwater – Anadromous
  - Examples

- Reproduce Seawater - Catadromous
  - examples

Smoltification- Anadromous

- Parr-smolt transformation
  - – Dynamic and multifactor
    - Size, time, and multifactor endocrine -physiology
  - – Ontogeny of salinity tolerance
    - Different for different species and life history

Term smolt

- First used for juvenile Atlantic Salmon
- Silvery stage
- Annual cycle – mostly in spring but some in autumn
- Photoperiod control
- Induced with short day to long days

Parr smolt transformation

- Developmental process
- Osmoregulatory physiology
- Growth changes
- Energetic and metabolic changes
- Behavioral changes

Physiology Endocrinology - Factors

- Many components
- Not a gold standard except actual migration and survival
- Many studies, lots of data from lab evaluations- not necessarily field
- Factors moving in different directions over time

High Interest

- <1% all fish are diadromous most are of high importance and value – also considered keystone species re nutrient cycles etc…..
- Aquaculture- commercial food fish
- Conservation aquaculture for stock restoration and supplementations
- Management of river and hydrosystems for downstream passage and adequate environmental conditions
- Concern about Xenobiotics and other toxicants in waterways

Pacific salmonids and SH

	Winters in FW as Juv.			Duration	
	0	1	2	in SW	
• Amago	++			4-5 mo	
• Masu		++	+	1 yr	
• Coho		++	+	0.5-1 yr	
• Sockeye	+	++	+	1 – 3 yr	+
• Chinook	++	++	+	1-3 yr	+
• Chum	++			1-4 yr	
• Pink	++			1 yr	
• Steelhead		++	++	not all go	

Review of adaptations FW

- Gills – absorption Na and Cl
- Intestine – reduced fluid uptake
- Kidney – high volume, absorption Na and Cl

Adaptation SW

- Gill – Excretion Na and Cl & Reduced permeability to Water
- Intestine – Absorption fluids, absorption Na and Cl
- Kidney – Low volume, excretion of Mg++ e.g. divalent ions

Bridging the Gap

- Transition into seawater
- Do fish go only when ready?
- What controls migration rates
- What size and time to release smolts?

Na K gill ATPase

- Increases after transfer to seawater
- Ionic gradients generated by enzyme
- Mitochondrial rich chloride cells
- Chloride cells increase in number
- Na flux changes from net influx to net efflux

Intestinal net fluid flux

- Increased absorption of water

Hormonal Controls

- Thyroid - was highly researched
  - Usually a distinct peak in plasma thyroxine
  - Influences behavior, growth, and morphological development

Prolactin and Growth Hormone Adenohypophysis

- Prolactin implicated to increase tolerance to seawater
- Plasma Insulin like growth hormone (IGF) can influence survival and osmoregulation
- Likely growth hormone secreted at greater rate in seawater

Cortisol

- Considered important promoter of adaptation to seawater
- Receptor with affinity for cortisol identified in gill tissues.

Others

- Methyl testosterone can inhibit smolting
- Corpuscles of Stannius
- Calcitonin
- Catecholamines- adrenaline
  - Chloride secretions inhibited

Smolt Development – changes up or down

Migratory behavior

Salinity tolerance

Salinity preference

Body silvering

Growth rate

Condition factor

Oxygen consumption

Ammonia production

Liver glycogen

Smolt Development – changes up or down

Migratory behavior

Salinity tolerance

Salinity preference

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Growth rate

Condition factor

Oxygen consumption

Ammonia production

Liver glycogen

Smolt Development – changes up or down

Blood glucose

Growth hormone

Corticosteroids

Thyroxine

Prolactin

Gill ATPase

Smolt Development – changes up or down

Blood glucose

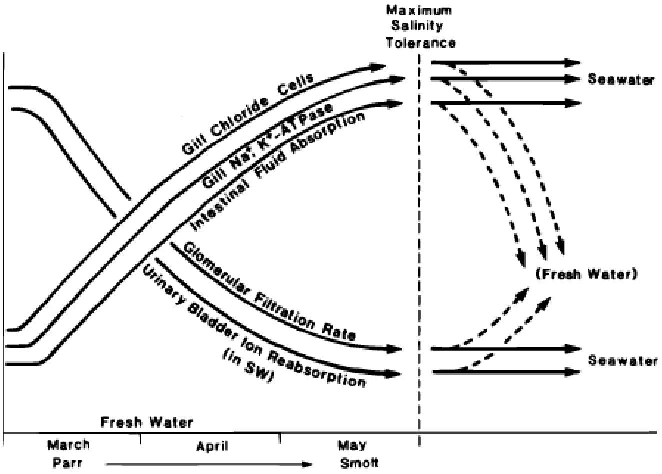
Growth hormone

Corticosteroids

Thyroxine

Prolactin

Gill ATPase



Return Spawning Migrations

what controls that portion of the life history?

- Precociously mature physiology
  - Masu
  - Sockeye
  - Chinook

Blood Plasma Analyses Used to Characterize Steelhead Kelts

Nutritional Factors (Energy Reserves and Fitness)

Cholesterol, Calcium, Alkaline Phosphatase (ALP), Triglycerides, Amylase, Lipase, Glucose, and Phosphorus

Tissue Damage Factors (External / Internal Damage)

Alanine Aminotransferase (ALT), Aspartate Aminotransferase (AST), and Lactate Dehydrogenase (LDH)

Electrolytes (Osmoregulation)

Sodium, Chloride, Potassium, and Magnesium

Hormone Factors

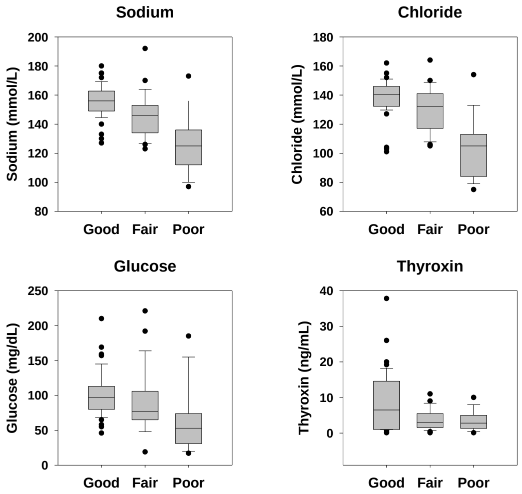
T4 – Thyroid Function
Cortisol - Stress

Is a Kelt a Smolt?

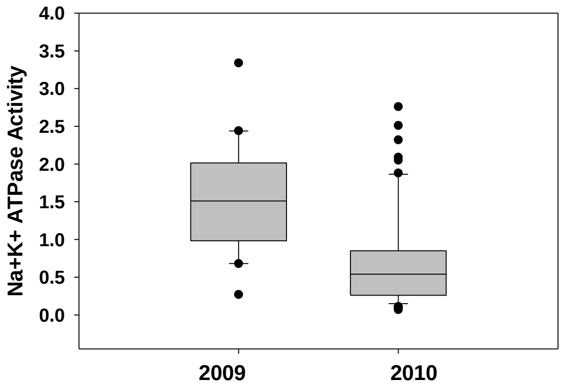
Na^+K^+Gill ATPase is Elevated in Smolts During Migration

Thyroxin Hormone T4 is Elevated in Smolts and Initiates Silvering and Migration

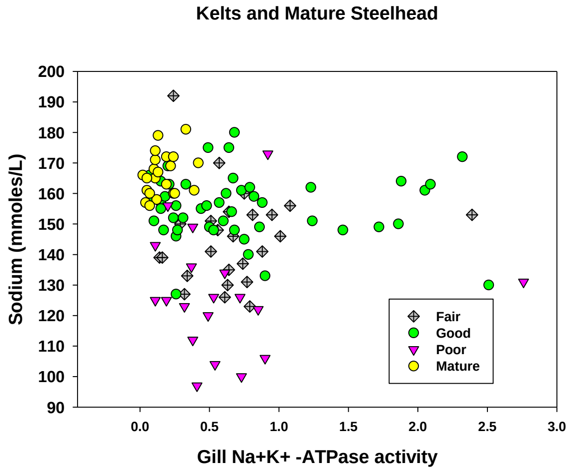
Migrating kelts at LGD- preparation for seawater

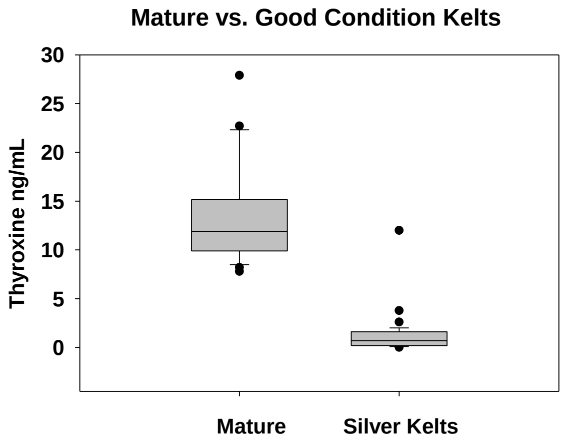


Na K – ATPase of good condition kelts at Lower Granite Dam



2010 Good Condition Fish




Mature vs. Good Condition Kelts
Thyroxine ng/mL
30
25
20
15
10
5
0
Mature
Silver Kelts