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Regulation of ECF

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Vasopressin

- = function of vasopressin & thirst mechanisms
- total body osmolality ∝ (total Na + total K) / total water
- ↑osmotic pressure in plasma ⇒ ↑vasopressin secretion + ↑thirst
- ↓osmotic plasma ⇒ ↓vasopressin + excretion solute free urine
- ∴ osmolality kept 280-295 mOsm/kg of H20
  ↳max inhibition vasopressin seen at <285; stim at higher values

ADH Molecule

- = 9aa peptide hormone
- aka ADH
- made in magnocellular neurons of supraoptic & paraventricular nuclei of hypothalamus
- transported in their axons to post pituitary where stored
- released via calcium dependant exocytosis

Vasopressin Receptors

- 3 receptors:
  - V1a ⇒ smooth mm sustained vasoC, ↓renin secr, ↓glycogenlysis, platelet aggregation
  - V1b ⇒ ↑ACTH release

  ↳ G_Q-linked → ↑PLC ⇒ ↑ IP3 + DAG → ↑ intracellular [Ca]
  - V2 ⇒ ↑cAMP levels ⇒ ↑protein kinases
    - vasoD, ↑H20 reabsorption, ↑vWF & f8 from endothelial cells
  - V3 = CNS (neurotransmitter)
- All G protein coupled

Effects of Vasopressin

- Vasopressin ⇒ concentrated hypertonic urine & ↓osmolality of plasma
- Without vasopressin ⇒ urine hypotonic & ↑osmolality of plasma

Renal ADH effects

V impt at physiological concentrations

- ↑ Water reabsorption (V_2 ⇒ aquaporin-2 from endosome ⇒ luminal membrane of principle cells).
- Synergism with aldosterone:
  - Na reabsorb/K excretion
  - Principal cells of CCDs
- Mesangial contraction ⇒ ↓GFR (V1a)
- ↑urea reabsorption – inner medullary CDs (aquaporin 3 into basolateral membrane)
- renal afferent vasoC – V1

Non Renal ADH Effects

Less impt at physiological concentrations

- Systemic vasoconstriction (V1a):
  - effect on bp offset by ↓CO via central affect (area postrema)
  - coronary & cerebral vaosD maintained by NO mediated effect
- ↑ACTH release (V1b)
- ↑Cortisol release (V1a)
- Glycogenolysis (V1a) + lipolysis (↑ hormone sensitive lipase)
- neurotransmitter _ neuromodulation (V3)
- Coagulation: - ↑ Factor 8 (V2) and ↑ platelet aggregation (V1)

Metabolism

- circulating vasopressin rapid inactivated in liver & kidney
- half life 18mins

Control of Secretion

- ↑ed secretion:

- ↑osmotic pressure of plasma –
  - osmoreceptors in anterior hypothalamus (OVLT & SFO ie outside BBB)
  - afferents to ADH secreting neurons
  - osmoreceptor system extremely sensitive mechanism for small ie 1-2% changes
    ↳ ie when <2% change osmoreceptors activity > baroreceptors
- ↓ECF volume –
  - sensed by low pressure (more impt) & arterial baroreceptors
  - afferent to hypothalamus neurons
  - when >10% change volume: baroreceptor system activity > osmoreceptors
- pain, stress, exercise
- N&V
- Standing
- ATII
- Drugs eg Carbamazepine, morphine, barbituates, nicotine

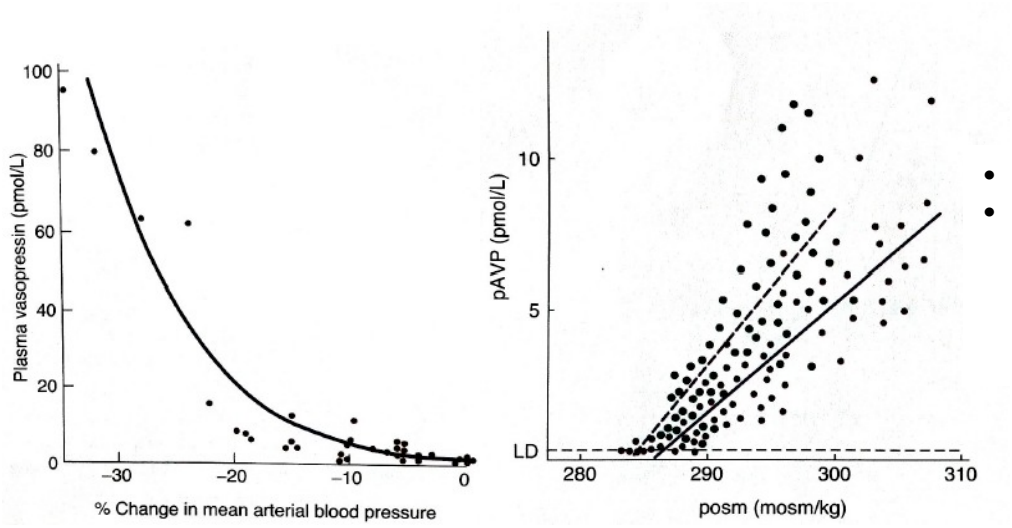
- ↓ed secretion
  - ↓osmotic pressure of plasma
  - ↑ECF volume
  - alcohol
  - local negative feedback: ADH ⇒ intramedullary synthesis of PGs which interfere with ADH induced production of cAMP

Osmotic Stimuli

- >285 rate of discharge of neurons ↑s ⇒ ↑secretion
- osmoreceptors in ant hypothalamus:
  - outside bbb, in OVLT
- thirst ?triggered by osmorecptors also
- delicate feedback system.
- 1% change in osmolality ⇒ big changes in level of vasopressin to keep at 285

Volume Effects

- inverse relationship between rate vasopressin secretion & level of stretch in vascular system causing afferent nerve discharge
  ↳high stretch ⇒ ↓vasopressin
- vascular sensors
  - low pressure – great veins, RA, LA, pulmon vessels
  - high pressure – carotid sinus, aortic arch
- high pressure bp changes ⇒ big change in vasopressin secretion
- low pressure monitor fullness of system
  - mod ↓in blood volume can ↓CVP with no effect on arterial bp
    ↳∴ low pressure sensors primary sensors effecting volume related vasopressin
  - afferents vagi to nucleus tractus solitarius (NTS)
  - NTS via inhibitory connection to CVLM
  - CVLM via excitatory to hypothalamus
- ATII reinforces vasc stretch receptors by causing direct ↑vasopressin secretion



- Vasopressin vs osmolality
- L shift when hypovolaemic & ↓osmolality

- Hypovolaemia and ↓bp ⇒ ↑↑vasopressin
- Hypovolaemia ⇒ L shift with steeper curve

Clinical Implications

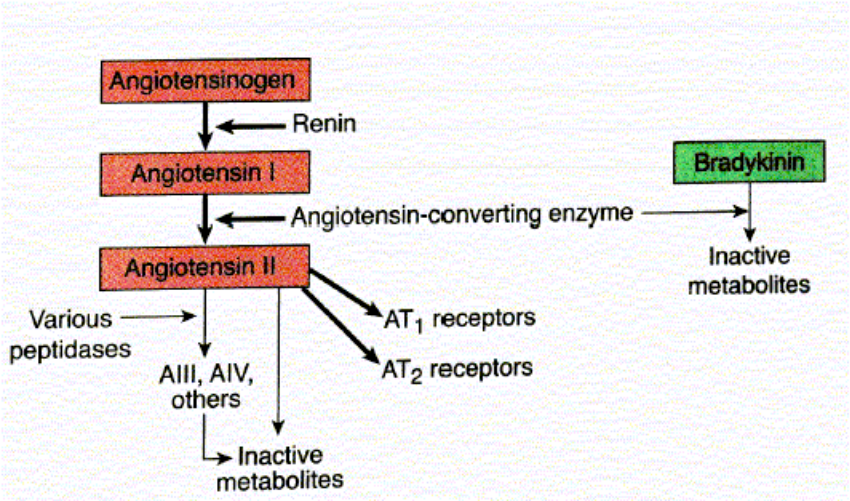
- SIADH –
  - Eg post surgery pain & hypovolaemia both ⇒ ↑vasopressin ⇒ ↓plasma osmolality & dilutional hyponatraemia
  - Also cerebral disease, lung disease, vasopressin secreting tumours
  - Vasopressin escape – prolonged ↑vasopressin ⇒ down reg of aquaporin2 production
  - Rx with Demeclocycline = Abx which ↓s renal response to vasopressin
- DI:
  - Central DI = Vasopressin deficiency
    - Tumour of hypothalamus 30%
    - Post traumatic 30%
    - Idiopathic 30%
    - Rest – sarcoid, vasc lesions, infections
  - Nephrogenic DI = kidneys don't respond to vasopressin
    - V2 receptor X linked recessive mutation
    - Defective gene encoding aquaporin 2 – trapped in intracellular locations
  - Symptoms include polyuria & polydipsia

↳need this to remain hydrated otherwise potentially fatal

Summary Defence of Volume & Normal Osmolality

- Osmolality = no of osmoles/kg water (not influenced by temp)
- ECF [Na] ~ 140mmol/L:
  - Na & Cl most active & abundant solutes present
  - Cl just follows Na ∴ Na is the most impt
  - Na = 85% ECF osmolality
- Obligatory Water Loss =
  - Minimum UO = 500ml/D
  - Need to excrete daily solute load od ~700mosmoles at max conc 1400mmol/L
    ↳ (Na 100-150, K 70-100, Cl 150, urea 400, creatinine 12 mmol)
- Conditions ⇒ ↓↓osmolality ie max ↓ADH secretion ⇒ dilute urine osmolality ~30mosmoles/kg & UO ~22L/day to secrete daily solute load 700msomoles
- Osmolality feedback mechanism:
  - ↑ osmolality (Small change 1-2% change from 280):
    - Ant HT sensing ⇒ ↑ADH ⇒ ↑AT2, ↑thirst, ↑aldosterone ⇒
      - ↑Na & water reabsorb
      - afferent arteriole vasoC ⇒ ↓GFR⇒ restore ECF osmolality
  - ↓osmolality:
    - ↓ADH, ↑ANP (↑GFR) ⇒ ↑Na & water excretion
- max urine conc capable = 1400mmol/L
- Volume ECF is determined by
  - total osmotically active solute in ECF
  - Volume control mechanism –
    - mechanism overrides osmotic regulation of vasopressin secretion
    - sensors less sensitive ie >10% changes
    - effect:
      - ↑volume ⇒
        - ↓vasopressin
        - ↑ANP (atrial) & ↑BNP (brain) ⇒ diuresis
          ↳⇒ ↑Na excretion by kidneys
      - ↓volume ⇒
        - ↑angiotensin II ⇒
          - VC
          - ↑aldosterone
          - ↑thirst
        - ↓bp ⇒ ↓glom cap pressure ⇒ ↓GFR ⇒ ↓Na filtered
        - ↓mean intravascular pressure ⇒ ↑aldosterone ⇒ ↑Na reabsorbed
- in dehydration ⇒ moderate ↓ ECF by loss of water from intravascular & extracellular compartments
- disease states can cause marked loss of Na from ECF ⇒ shock:
  - stool - diarrhoea
  - urine – severe acidosis, adrenal insufficiency
  - sweat – heat+

Renin Angiotension System



Renin

- =acid protease (glycoprotein hormone)
- mw 37326
- secreted by kidney into bloodstream
- synthesised as preprorenin ⇒ prorenin ⇒ rennin
  ↳non active
- active renin half life 80mins
- kidney:
  - active renin secreted by specialized cells (granular cells) = JG cells
  - found in media of afferent arterioles as enter glomerulus
  - renin found in membrane lined secretory granules
  - cells also convert some prorenin to renin
    ↳only place to do this
  - secretes some prorenin – none converted systemically

- Renin also found in lacis cells
  - Found junction between afferent/efferent arterioles
  - Agranular
  - ?significance of renin here
- prorenin also made in other organs eg ovaries
  ↳after nephrectomy prorenin level normal BUT active renin zero

Note:

- Macula densa is close proximity to JG cells
- Juxtaglomerular appartatus =
  - Macula densa
  - Lacis cells
  - JG cells

Regulation of Renin Secretion

- Main regulatory mechanisms:
  - Factors ↑renin secretion (and opposite of factors which ↓renin release):
    - ↑SNS outflow
      - ↑circulating catecholamines
        - β_1 receptors on JG cells ⇒ ↑cAMP ⇒ ↑renin release
      - any cause of ↑post-ganglionic symp activity on kidney via renal nerves

↳ ∴ MOA of BBs on controlling volume status & bp

- Factors ↓renin secretion (opposite of above and…):
  - ↑Na & Cl reabsorb across macula densa
    - renin secretion inv∝ amount of Na & Cl entering distal tubule
    - enter mac densa via Na-K-2Cl transporter in apical membrane
    - ⇒ ↓renin secretion from adjacent JG cells
      ↳?mediated by NO
      ↳ Nb this is diff to TG Feedback (ie↑Na ⇒ vasoC of afferent arteriole)
  - Baroreflex ie ↑afferent arteriolar pressure:
    - ↑MAP at JG cells ⇒ ↓renin release
      ↳ via ↓post ganglionic symp activity via renal nerves
  - AT II – direct action in JG cells ie -ve feedback loop

- Secondary mechanisms:
  - ↑renin release:
    - prostaglandins
    - ↑plasma K level
      - linked to Ks effect on delivery of Na & Cl to macula densa
  - ↓renin release:
    - Vasopressin - ?direct or indirect effect
    - ↑ANP
- Conditons which ↑renin secretion ie anything which ↓ECF, ↓MAP, ↑SNS
  - Na depletion
  - Diuretics
  - ↓bp
  - haemorrhage
  - upright posture
  - dehydration
  - heart failure
  - cirrhosis
  - constriction of renal artery
  - psych stimuli

Angiotensinogen

- synthesized in liver
- alpha glycoprotein (453 aa's) (13% CHO)
- circulating level is ↑ed by:
  - glucocorticoids
  - thyroid hormones
  - oestrogens
  - cytokines
  - AT II

AT1

- Decapeptide (10aas)
- Splite from N-terimal of angiotensinogen by renin's action
- Sole function is as a precursor of AT2

ACE

- ACE:
  - AT I to AT II
  - Inactivates bradykinin
- ↑ tissue bradykinin produced when ACE is inhibited
  ↳this acts on B2 receptors ⇒ cough in 20% of people on ACEI

- most ACE found in endothelial cells
- Most converting in lungs but other parts of body contribute
- ACE exists in 2 forms:
  - Somatic – throughout body
  - Germinal – spermatogenic cells & spermatozoa
- Kidneys do not contain angiotensionogen & ACE
  ↳ ∴ AT2 can be fully produced in kidneys by itself
  ↳ ∴ kidneys influenced by blood borne & intrarenally produced AT2

AT2 & It's Metabolism

- =octapeptide
- Half life 1-2mins
- Removed from circulation by:
  - Metabolised by various peptidases:
    - angiotensinase (Aminopeptidase) – removes aspartic acid residue ⇒ AT III
    - AT III can be converted to AT IV
      ↳both AT III & AT IV have some activity; other peptide fragments inactive
    - occurs in rbcs & many tissues
  - trapping system in vascular beds of non-lung tissue

Actions of AT's

- AT1 – precursor of AT2 ie no action
- ATII – actions (via G_Q-linked receptors→↑ phospholipase C/IP3→ ↑ Ca)
  - Arteriolar VC ⇒ ↑systolic & diastolic bp
    - X4 more powerful vasoC than NA
    - VasoC activity decreased in:
      - Hyponatraemic patients
      - Cirrhosis

      ↳because ↑AT II found circulating ⇒ downregulate ATII receptors on smooth mm
  - ↑aldosterone – direct action on Z Glomerulosa of adrenal cortex
  - ↓renin release: -ve feedback control
  - blockade of NA re-uptake (uptake 1) ⇒ ↑ active NA
    - direct action on postganglionic symp neurons
    - central effect on area postrema
  - ↓GFR – contraction of mesangial cells
  - arteriolar effects:
    - [low concs] ⇒ selective afferent arteriolar constriction ie defend volume by ↓ing GFR
      [higher concs] ⇒ efferent constriction
  - ↑Na & HCO3 reabsorb –
    - direct effect on PCT
    - indirect by ↓interstitial hydrostatic pressure
  - ↓sensitivity of baroreflex ⇒ ↑pressor effect
  - activation of cicumventricular organs on brain (not cross bbb)
    - thirst
    - ↑ADH release
    - ↑ACTH release

    ↳ remember:
    - area postraema ⇒ VC
    - SFO & OVLT ⇒ polydipsia
    - ??organ ⇒ ↑vasopressin & ↑ACTH

- AT III – same actions as AT2 but:
  - VasoC – 40%
  - ↑aldosterone – 100% action ATII

Summary AT2 actions always results in:

- ↓RBF – afferent +/- efferent vasoC

- effect on GFR dependant on conc of AT2:
  - low: same or slightly ↓ed
  - high: ↓↓↓ (via mesangial & afferent arteriolar contraction)

Tissue Renin-Angiotensin Systems

- many tissues contain local indep renin-AT systems which generate ATII for local use
  ↳eg blood vessel walls, uterus, placenta, foetal membranes, eyes, heart, sex organs
- tissue renin contributes little/nothing to circulating renin pool
- local ATII ?role in:
  - growth factor in heart & blood vessels

AT II Receptors

- are 2 classes of ATII receptors:
  - AT_1
  - AT_2

  ↳ ATII has stronger affinity for AT_1

AT_1 Receptors

- = 7 transmembrane domains
- found on chromosome 3
- couple by G protein to phospholipase C
- ATII ⇒ (on AT_1)
  - ↑free cytosolic Ca
  - activates numerous tyrosine kinases
  - in smooth mm: ↑caveolin-1
- regulation of AT_1 receptor depends on location:
  - in arterioles - ↑ATII ⇒ ↓AT_1 receptor predominance
  - in adrenal cortex - ↑ATII ⇒ ↑AT_1 receptor ⇒ ↑sensitivity of gland ⇒ ↑aldosterone release

AT_2 Receptors

- = 7 transmembrane domains
- found on chromosome X
- couple by G protein to various phophatases
- AT II ⇒ (on AT_2)
  - Antagonise growth effects
  - Open K channels
  - ↑production of NO ⇒ ↑ing cGMP

  ↳overall effects unsure but more receptors found in fetal & neonatal life

Hormones of Heart & Other Natriuretic Factors

Structure

- atria & ventricles contain secretory granules
- granules ↑in number when
  - ↑NaCl intake
  - ↑ECF volume

Types

- from heart:
  - ANP – also found in other tissues
  - BNP – brain or beta NP
    - More present in heart (esp ventricles) than brain
  - CNP –
    - Found in brain, kidney, pituitary, vascular endothelial cells

- Very little in heart & circulation
  ↳∴ paracrine regulator

Actions

- ANP & BNP act on kidney ⇒ overall ↑ in Na excretion
- Done by:
  - Dilation afferent arterioles } ⇒ ↑GFR
  - Relax mesangial cells }
  - Act on tubules ⇒ ↓Na reabsorb
  - ↑cap permeability ⇒ extravastion of fluid into ECF ⇒ ↓bp
  - VD of arterioles & vennules
    ↳CNP greater VD effect on veins than ANP/BNP
  - ↓renin secretion
  - counteract pressor effects of catecholamines & ATII
- ANP in brain:
  ↳BNP/CNP prob similar functions but unknown
  - General effects opposite to ATII
  - Found in neurons connecting hypothalamus to brainstem concerned with regulation of CV system
    ↳ANP ⇒ ↓bp & excretion of Na

Receptors

- 3 natriuretic peptide receptors:
  - NPR A
    - Guanylyl cyclase cytoplasmic domain
    - ANP greatest affinity
  - NPR B
    - Guanylyl cyclase cytoplasmic domain
    - CNP greatest affinity
  - NPR C
    - Binds all peptides
    - Truncated Guanylyl cyclase cytoplasmic domain
    - ?function
- All span cell membrane

Secretion & Metabolism

- ANP norm conc in plasma 5fmol/ml
- ANP
  - half life short
  - metabolised by neutral endopeptidase (NEP)

- ↑ANP secretion:
  - ↑ECF volume
  - atrial stretch
    ↳also water immersion up to neck: removes gravity ⇒ ↑CVP ⇒ ↑atrial stretch
- ↓ANP secretion:
  - lie to stand ⇒ ↓CVP
- ↑BNP secretion:
  - ventricle stretch

↳ANP & BNP secretion ∝ to degree of stretch

Na,K ATPase-Inhibiting Factor

- = another natriuretic factor
- it inhibits Na,K ATPase ⇒ ↑bp
- may be ouabain from adrenal glands

Defence of Specific Ionic Composition

- certain ions in ECF under close control:
  - Ca – parathyroids, calcitonin secreting cells
  - Mg – mechanism incompletely understood
- Control of Na & K also depend on:
  - H+ conc
  - pH

Endocrine Functions of Kidney

- Hormones produced by kidney:
  - 1,25 dihydroxycholecalciferol (active Vit D, aka calcitriol)
  - Erythropoietin
  - Prostaglandins
- Hormones produced in circulation as result of enzymes released by kidney:
  - Angiotensin-2 + aldosterone production, initiated by renin released by kidneys
  - Production of bradykinin in circulation, due to kallikrein from kidneys
- Hormones which have site of action **on** kidney:
  - ADH
  - Aldosterone
  - Calcitriol
  - PTH
  - ANP

Calcitriol

- Creation of active form of vit D
- final step of activation
  - 1-alpha hydroxylation occurs in cells of prox tubule
  - reaction ↑ed by:
    - catalysed by 1-alpha hydroxlase (mitochondrial enzyme)
    - ↑ed by PTH
    - ↑gonadal steroids
  - reaction ↓ed by:
    - ↓ s-calcium or phosphate
    - ↑ed calcitriol ie –ve feedback loop
- function:
  - ↑intestinal absorption of calcium & phosphate
  - ↑tubular reabsorption of Ca
  - ↑bone reabsorption of Ca & phosphate ie ↑serum Ca & PO4

EPO

- EPO = circulating glycoprotein
- Half life 5hours
- Takes 2-3days for ↑circulating rbcs

Sources

- mRNA found in liver & kidney
  ↳EPO also found in spleen & salivary glands but no mRNA ∴ not made there
- source from:
  - 85% - interstitial cells in peritubular cap bed of kidney
  - 15% - perivenous hepatocytes in liver
    (↳ in fetus main production is the liver ie not from kidney)
  - trace – brain- protective effect from hypoxia
  - trace – uterus & oviducts – ↑oestrogen ⇒ ↑EPO ⇒ oestrogen dependant angiogenesis
- liver has little compensatory ability if kidneys removed ⇒ anaemia
- recombinant EPO made and can be injected
  - autologous transfusions for surgery

- end stage renal failure

Regulation of Secretion

- ↑EPO:
  - hypoxia:
    - main stimulus
    - O2 sensor in kidney & liver
      - Heme protein:
        - Deoxy form ⇒ ↑EPO
        - Oxy form ⇒ inhibit transcription forming EPO mRNA
  - ↓rbc volume eg anaemia/haemorrhage
  - ↑androgens
  - alkalosis eg high altititude
  - catecholamines via β-adrenergic system
- ↓EPO:
  - ↑red cell volume eg transfusion/polycythaemia

Function

- EPO marrow effects:
  - commited stem cells ⇒ rbc precursors ⇒ mature erythrocytes
- EPO acts via membrane receptor =
  - Linear protein
  - Single transmembrane domain
  - Cytokine receptor family
  - Activates Tyrosine kinase ⇒ activates cytoplasmic transcription factors ⇒ nucleus ⇒ activate new m-RNA synthesis ⇒ inhibited apoptosis of red cells ⇒ ↑growth & development