

Nephron Underdosing and Hypertension

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Since Richard Bright (1846), the role of the kidney in the genesis of hypertension of patients with renal disease has been clearly recognised. It is only recently, however, that it has been noted that the kidney also plays a definite role in the genesis of essential hypertension in individuals without renal disease. This concept is based both on studies of Guyton who showed a functional abnormality of the kidney (right-ward shift of pressure natriuresis relationship), but later on also on morphological studies. The latter investigations were stimulated by the concept of "nephron underdosing" proposed by Brenner (AJH 1988;1:335).

He postulated that a diminished number of glomeruli predisposed both to hypertension and to renal injury. This concept has recently been confirmed in patients with essential hypertension (Keller, NEJM 2003;384:101). Experimental studies show that blood pressure goes with the kidney, i.e. transplantation of a hypertension-prone kidney to a normotensive animal provokes hypertension. Conversely, in animals and in humans transplantation of a kidney from a normotensive donor causes persisting normotension (granted that renal function remains normal), as shown by Curtis (NEJM 1983;309:1009). Nephron underdosing may be due to genetic factors (in experimental studies, for instance GDNF $-/+$ heterozygosity). Nephron deficit is associated with hypertension, particularly if this is present perinatally, but low nephron number may be only a surrogate marker for associated changes in tubular function, particularly sodium reabsorption; adult uninephrectomy (as in life kidney donors) increases only slightly the risk of hypertension. Apart from genetic factors the intrauterine environment plays an important role, e.g. placental ischemia or maternal malnutrition and maternal hyperglycemia. Low birth weight (as evidence of intrauterine malnutrition) is associated with less glomeruli both in humans and in animals. Low birth weight is known to be associated with higher blood pressure and a higher risk of renal dysfunction in adult life. This can be provoked experimentally by ligation of the uterine artery as well as by low protein diet, which causes selective apoptosis of precursor cells in the metanephros with subsequent nephron deficit and late hypertension. Nephron underdosing also predisposes to more severe renal injury in renal damage models. One feature of nephron underdosing is salt sensitivity. High salt intake causes not only hypertension, but also target organ damage.

The human body was given 10 organs of which it is the task of the kidney to provide the human being thought

Talmud Brochot



Richard Bright, Tabular view of the morbid appearances in 100 cases connected with albuminous urine, Guy's Hospital Report 1 380, 1846

...the obvious structural changes in the heart have consisted chiefly of hypertrophy with or without valvular disease...

this naturally leads us to look for some less local cause, for the unusual effort to which the heart has been impelled : and the two most ready solutions appear to be, either that the altered quality of the blood affords unwonted stimulus to the organ; or, that it so affects the minute and capillary circulation, as to render greater action necessary to force the blood through the distal subdivisions of the vascular system

“Nephron underdosing”

- ⇒ # hypertension
- # susceptibility to renal damage

Brenner BM et al. Glomeruli and blood pressure. Less of one, more the other ? Am J Hypert (1988) 1, 335

- 10 Caucasian victims of accidents with history of hypertension / LVH

- controls matched for gender, age, height and weight

⇒ kidney unbiased stereological analysis by Gundersen fractionator method

- glomerular number
- glomerular volume

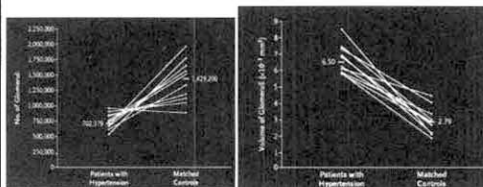
Keller, New Engl J Med (2003) 348: 101

	hypertensive individuals (n=10)	normotensive individuals (n=10)
number of glomeruli	890,869 ± 158,110	1,666,805 ± 411,690 p < 0.001
volume of glomeruli	5.67 ± 0.85	2.41 ± 0.71 p < 0.001

⇒ essentially no evidence of obsolescent or remnant glomeruli

Keller, New Engl J Med (2003) 348: 101

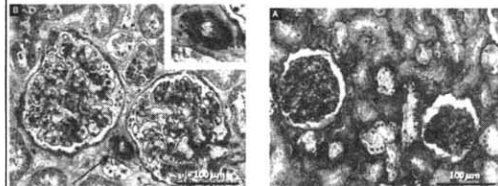
Oligomeganephrony



Keller, New Engl J Med (2003) 348: 101

hypertension

normotension



Keller, New Engl J Med (2003) 348: 101

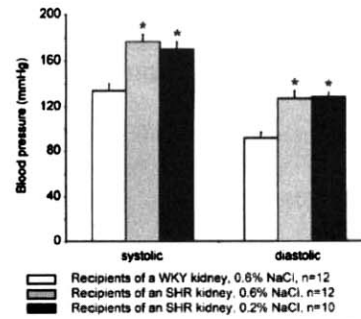
**Animal experiments:
blood pressure "goes with the kidney"**

- kidney genetically programmed for hypertension, transplanted into a normotensive recipient

⇒ persistent hypertension

Rettig R, *Am J Physiol* (1980) 258, F606

Blood pressure goes with the kidney



- cadaveric kidney donors :**
subarachnoid hemorrhage vs head trauma
23 recipients normal graft function/no artery stenosis; 72 months follow-up
in recipients of grafts from donors with subarachnoid hemorrhage :
 ⇒ higher blood pressure
 ⇒ more antihypertensive medication

Strandgaard S and Hansen U
 Hypertension in renal allograft recipients may be conveyed by cadaveric kidneys from donors with subarachnoid hemorrhage
Brit. Med. J. (1986) 292:1041

Hypertension may be transplanted with the kidney in humans

- historical observational study
 85 renal transplant patients no Cyclosporin A
 8 years follow up
 donor and recipient families screened for hypertension (HT)
 evaluation : necessity for antihypertensive treatment

- ⇒ recipient family HT – no impact on start of AHT
 ⇒ kidney from donor with family history HT vs NT
 odds ratio for start of AHT 5.0(1.4-17.89)

Guidi, *J. Am. Soc. Nephrol.* (1996) 8:1131

Persistent normotension after kidney graft from normotensive donor

- 6 black patients "essential hypertension"
- dialysis dependant
- LVH and retinal changes
- nephrosclerosis by histology
- kidney graft from normotensive donor

- ⇒ after 4.5 years follow-up all normotensive
 • normal BP response to Na loading / deprivation

Curtis J. et al.
 Remission of essential hypertension after renal transplantation
New Engl J Med (1983) 309, 1009

Perspectives

Genes in organogenesis(PAX, WT1, GDNF, IGF2 ...) = Genes involved in genesis of hypertension ?

known structural abnormalities in (pre-)hypertension

- # brachydactyly
- # arterioles (luminal narrowing)
- # capillaries (diminished density)

Rarefaction of skin capillaries

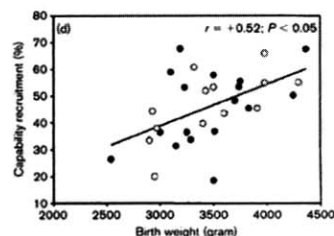
- in patients with essential hypertension

Antonios, *Hypertension* (1999) 33: 998

- in offspring of patients with essential hypertension

Antonios, *Heart* (2003) 89: 175

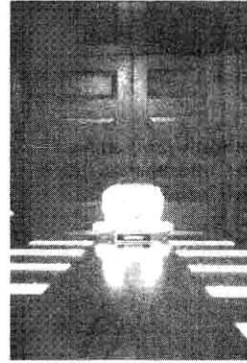
Low birthweight and diminished capillary recruitment



Serne, *J Hypertens.* (2000) 18:1421

특별 강연

- The concept of “nephron underdosing”
- The findings in essential hypertension
- Adult vs congenital “nephron underdosing”



If less nephrons cause hypertension – why not hypertension in adult live kidney donors ?

- reports of no increase in hypertension
- with long follow up hypertension 42 – 51 %

Hakim, Kidn Intern, 1984
Williams, Arch Intern Med, 1986
Najarlan, Lancet, 1992
Saran, NDT, 1997

Obesity - important cofactor in genesis of hypertension and renal failure after uninephrectomy

Praga, Kidn Intern (2000) 58:2111

After neonatal uninephrectomy hypertension (and late glomerular damage)

- neonatal UNX SD rats
- examination at 8 and 20 weeks of age on low and high salt diet
- glomerular volume increased by 40 %
- at week 20 3-fold increase protein excretion and glomerular damage
- greater blood pressure increase on high salt (salt sensitivity)

Woods, Hypertension (2001)38:337

If less nephrons cause hypertension – why not hypertension in adult live kidney donors ?

- ⇒ neonatal uninephrectomy ≠ uninephrectomy in adult life
- ⇒ not low glomerulus number per se, but associated (developmental) functional changes in postglomerular nephron segments (sodium reabsorption ?)

Supplementation of kidney mass by isograft transplantation

in the remnant kidney model

- ⇒ lower systolic bp (-35 mmHg)
- lower urinary protein
- lower glomerulosclerosis index

Ots, Nephrol.Dial.Transplant (2004) 19:337

Potential causes of nephron underdosing

- Genetic (*SHR, Milano rats...*)
- Environmental factors during intrauterine life
 - # placental ischemia
 - # maternal malnutrition
- Programming during organogenesis
 - # hyperglycemia
 - # placental 11βHSD2

Glial cell line derived neurotrophic factor (GDNF)

GDNF – ureteric bud stage branching
 aged GDNF heterozygous mice
 # less nephrons
 # hyperfiltration
 # late hypertension
 ⇒ compensatory mechanism to maintain GFR in presence of reduced nephron endowment

Cullen-Mc Ewen, *Hypertension*(2003) 41:335

Elderly GDNF (-/+) heterozygous mice → reduced nephron numbers, normal GFR, hypertension

Kott, *Hypertension* (2003) 41:335

Idiopathic renal failure – nephron underdosing as result of maternal or fetal “loss of function” mutations ?

maternal neutral endopeptidase (NEP) stop codon; during pregnancy immunisation by NEP expressing fetus
 transplacental transfer of maternal IgG1 alloantibodies into NEP (+) fetus
 ⇒ intrauterine membranous glomerulonephritis ?
 # myelin protein zero (MPZ/P0) in Schwann cells loss of function mutation → demyelinating disease (Charcot Pierre Marie Tooth) in S phase of nephrogenesis MPZ/P0 expressed in podocytes
 renal failure in individuals with mosaicism : kidney (-) ; Schwann cells (+) ⇒ nephron underdosing ?

P.Ronco, 2004

Potential causes of nephron underdosing

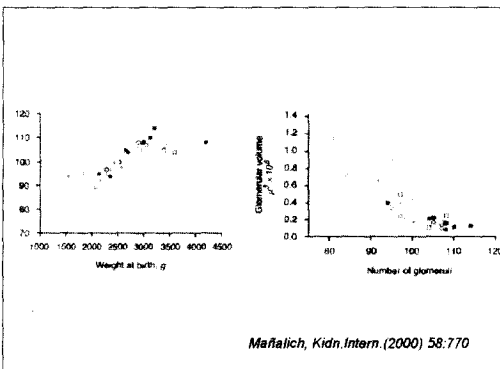
- Genetic (*SHR, Milano rats...*)
- Environmental factors during intrauterine life
 - # placental ischemia
 - # maternal malnutrition
- Programming during organogenesis
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- The findings in essential hypertension
- Adult vs congenital “nephron underdosing
- Nephron underdosing - relation to low birth weight

Less and bigger nephrons in kidneys of low birthweight newborns

- 35 neonates
 - coronal sections of renal cortex
- | | birth weight | |
|---|--------------|------------|
| | low | high |
| glomeruli per 0.6 mm ² | 92.9 ± 4.85 | 106 ± 3.91 |
| glomerular volume (μ ³ x10 ⁻³) | 529 ± 188 | 158 ± 50 |
- correlation between birthweight and :
 glomerular numbers (r=0.87) and volume (r= - 0.821)

Mañalich, *Kid.Intern* (2000) 58:770



Relation between birth weight and glomerular size and number

- autopsy study
- 37 African-Americans, 18 Caucasians
- stereological analysis (disector / fractionator method)
- ⇒ linear relationship glomerular number and birth weight
- ⇒ strong inverse correlation glomerular number and glomerular volume

Hughson, *Kid.Intern* (2003)63: 2113

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- Nephron underdosing – relation to low birth weight / fetal malnutrition
- Hypertension and renal malfunction – relation to low birth weight

The Barker hypothesis :

Initiation of hypertension in utero and amplification throughout life

Brit.Med.J (1993) 306: 24

- 3634 Individuals, birth cohort 1946
- BP at age 36 and 53 years, males :

birthweight (g)	systolic pressure (53 years) (mmHg)
<2500	143
2501-3000	141.5
3001-3500	140.1
3501-4000	140.6
>4000	136.5

Hardy, Lancet (2003) 362: 1178

Exposure to famine during pregnancy : the South-Holland catastrophe

	late	mid	early
n	119	104	65
birth weight (g)	3177	3226	3456
DBP (mm Hg)	86	83	85
Ccr (ml/min)			
female	115	110	128
male	128	121	127
urine albumin/creatinine ratio >2.5 (g/mol)			
percent	8	12	6

→ midgestational famine (phase of rapid nephronogenesis) : odds ratio microalbuminuria 2.1 lower creatinine clearance

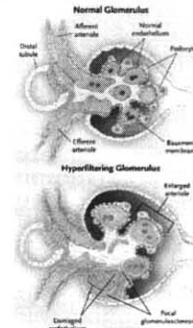
R.Krediet, pers.communication

Low protein diet – nephron underdosing

rat < 13 day of pregnancy
9% protein vs 18 % Isocaloric maternal diet (not starvation !)
⇒ metanephros before formation of glomeruli and tubuli
⇒ apoptosis of precursor cells
less cells on day 15
not recoverable by switch to high protein diet
less nephrons
late hypertension
(# less pancreatic beta cells)

Langley-Evans, Life Sci (1999) 64:965
Woolfe, 2004

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- Hypertension and renal malfunction – relation to low birth weight
- Nephron underdosing – propensity to renal injury



“work kills”

Ingelfinger J.
New Engl.J.Med. (2003) 348: 89

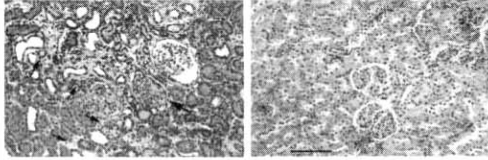


Glomerular hypertrophy sensitizes glomerulus to injury

- uninephrectomy or sham op
- infusion of ANG II (50 ng/min) or saline
- MAP in UNX 102 ± 6 mmHg
 In UNX + 181 ± 13 mmHg
 ANG II
- glomerular capillary pressure
 in UNX 56 ± 3 mmHg
 in UNX + 70 ± 7 mmHg
 ANG II
- ANG II infusion caused albuminuria and glomerulosclerosis in sham op, but dramatically more in UNX

Miller Am.J.Physiol. (1991) 261:F459

**Renal histology after uninephrectomy
in neonatal SHR**



uninephrectomy

control

Woods, Hypertension (2001) 38: 337

**Low nephron number -
more glomerulosclerosis
(neonatal gentamycin model)**

Gibert, Ped Nephrol, 1991

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- Nephron underdosing – propensity to renal injury
- Nephron underdosing – relation to salt sensitivity

**Larger (and less ?) nephrons
in minorities**

- blacks ?

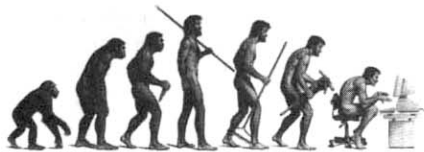
Abdi AJKD (1998) 32:43
Hughson Kidn.Inter.(2003) 63:2113

- Pima Indians

Schmidt JASN (1992) 3:229

- Australian aboriginals

Hoy Kidn.Inter.(2003) 83:S31



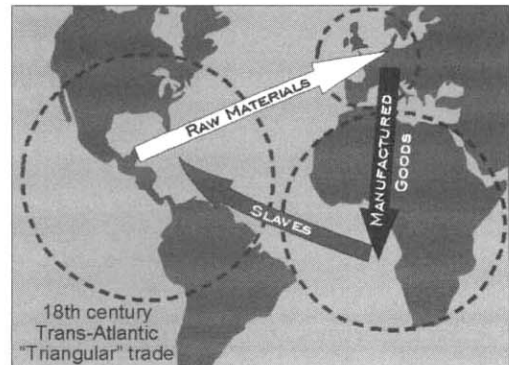
**Salt sensitivity –
selection advantage in evolution ?**

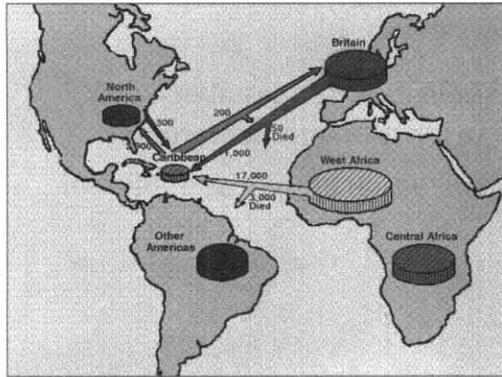
in tropical climates :

→ high infant mortality
diarrhea and fever → salt wasting →
hypovolemia and shock
blacks > whites

genetic bottleneck → slave trade
(transit across Atlantic → death from
diarrhea and fever ~ 6 Mio deaths)

Fleck, Hypertension (1991) 17 (Suppl) 1 115



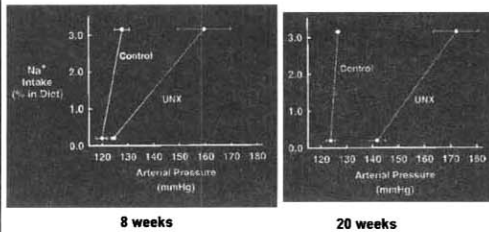


Salt sensitive hypertension after neonatal uninephrectomy and late glomerular damage

- neonatal UNX SD rats
- examination at 8 and 20 weeks of age on low and high salt diet
- glomerular volume increased by 40 %
- at week 20 3-fold increase protein excretion and glomerular damage
- greater blood pressure increase on high salt (salt sensitivity)

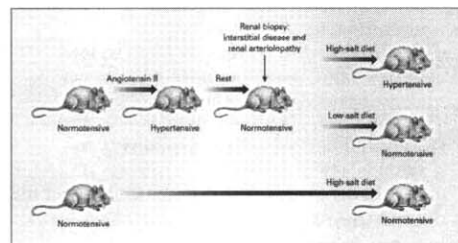
Woods, Hypertension (2001)38:337

BP response to Na⁺ loading after neonatal uninephrectomy



Woods, Hypertension (2001) 38: 337

Development of Salt-sensitive Hypertension in Rats after Exposure to Angiotensin II



N Engl J Med 346, No. 12, March 21, 2002

Sodium intake (mmoles/day)	decrease systolic blood pressure (mmHg)
150	- 2.1
100	- 4.6
50	- 6.7

DASH study New Engl J Med (2001) 344: 3-10

Rats, high salt intake

→ in heart and aorta respectively expression of AT₁ receptor ↑
aldosteron synthesis ↑
ACE expression →
caveolin, PKC, ERK ↑
reactive oxygen species ↑

Takeda, Endocrinol (2001) 141: 1901
Kreutz, J Mol Med (1995) 73: 243

High sodium intake - aggregation of target organ damage

increasing quintiles of sodium intake
→ higher LVMI
albuminuria
pulse pressure
→ steeper slope for relation SBP to LVMI, albuminuria, pulse pressure

Mimran A, 2002, French Population Study

Finnish study sodium intake:

- no sign.correlation to blood pressure
- but**
- per 100 mmol difference sodium intake relative risk coronary heart disease 1.5
- all cause mortality 1.2

Tuomilehto, Lancet (2001) 357: 848

**High sodium intake →
higher: all cause mortality
stroke incidence
coronary mortality
but only in obese**

He, JAMA (1999) 282,2027

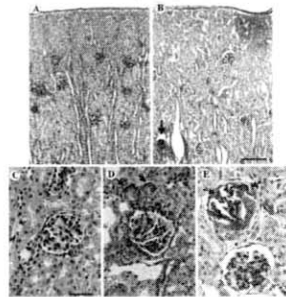
Diminished nephron number

- genetic ?
- maternal factors
 - malnutrition (Barker hypothesis, South Holland)
 - smoking
 - toxins (gentamycin, cyclosporin A)
 - vitamin A deficiency (retinoids)
 - hyperglycemia / diabetes

Merlet-Benichou, Int.J.Devel.Biol.(1999)43:453



Reduced nephron numbers after CyA treatment of pregnant rabbits



Tendron, JASN(2003)14:3188

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- Nephron underdosing – relation to salt sensitivity
- Nephron underdosing – relation to maternal diabetes/hyperglycemia (“metabolic programming”)

Less nephrons in offspring of diabetic rats

	kidney weight	glomerular number
control	210 ± 4	36,000 ± 860
STZ diabetes / low	201 ± 5	30,839 ± 667
STZ diabetes / high	155 ± 5	28,453 ± 840

Amri, Diabetes (1999) 48: 2240

Overexpression of IGF binding protein 1 ⇒ nephron deficit

- IGF binding protein ↑
- ⇒ bioavailability of IGF1
- # pregnant rats overexpressing IGF bp
- ⇒ reduced nephron numbers in pups
- # conditional transgene expression of IGFbp in pup
- ⇒ less nephrons
- # addition of IGF₂ / IGF2-R antisense oligo
- ⇒ stimulation(inhibition) of in vitro nephrogenesis

Doublier Pediatr.Res. (2001) 49:660

Genetic nephron number deficit associated with reduced renal IGF₂/IGF₂receptor ratio during fetal development

- rats with inborn oligonephropathy (MWF) vs Wistar ⇒
- nephron number – 31%
- renal IGF₂mRNA unchanged
- IGF₂receptor expression – 34%
- hepatic IGF₂mRNA and IGF₂r mRNA unchanged

Rothermund,Amer.Heart.Ass. Oct 2003, p714

- **hypertension renal problem (not only, but kidney crucial permissive factor for longterm setting of BP)**
- **structural causes ("nephron underdosing") and functional abnormalities ("salt sensitivity")**
- **apart from undoubted genetic causes, intrauterine environment important codeterminant of blood pressure in adult life (and of CV risk factor profile and diabetes)**