Understanding Spontaneous Regression in Neuroblastoma

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Neuroblastoma Regression–Outline

- Prevalence of neuroblastoma (NB) regression (in situ NB)
- Lessons from mass screening for NB
- Genomic subtypes of NB
- Role of telomere shortening in NB regression
- Role of immune surveillance and NB regression
- Neural crest migration and role of TRK receptors
- Role of TRK receptors in NB regression and differentiation
- Conclusions
“Neuroblastoma In Situ”?  

**IN SITU NEUROBLASTOMAS: A CONTRIBUTION TO THE NATURAL HISTORY OF NEURAL CREST TUMORS**

*J. Bruce Beckwith, M.D., and Eugene V. Perrin, M.D.*

- Beckwith (1963): Adrenal glands of infants <3 mo that died—1/40 had neuroblastic nodules (“NB in situ”).
  - If NB occurs in ~1/8,000 live births, then “NB in situ” is about 200x more common than NB.

  - “NB in situ” is really persistence of a normal phase of adrenal maturation.

Spontaneous Regression of Neuroblastoma

SPECIAL PATTERN OF WIDESPREAD NEUROBLASTOMA WITH A FAVOURABLE PROGNOSIS

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Summary
There is a group of children with disseminated neuroblastoma with a surprisingly good prognosis. Patients who fit the syndrome can have widespread disease in the liver, skin, and bone-marrow, or any combination of these. The primary tumour in some may be relatively small. 21 of 25 such patients (84%) survived for two years or longer. Radiation therapy and chemotherapy may not be necessary in the management of certain children with this syndrome.

- How prevalent is spontaneous regression?
- Danish population-based study suggests 2% detected clinically
- Some estimate as high as 5-10% of NBs may regress
- Stage 4S is special pattern of metastasis with good prognosis
- Spontaneous regression not restricted to stage 4S

D’Angio GJ: Lancet, 1971
Lessons from Neuroblastoma Screening—Rationale

- Infants with NB have a better outcome than older pts., so favorable NB may evolve into unfavorable NB
- NBs detected early may be more curable, and may prevent their genomic evolution into unfavorable NB
- Mass screening for NB can be done by measuring urinary VMA/HVA using spot test and HPLC
- Screening for NB should be done at 3-6 months, while tumors are still favorable
Lessons from Neuroblastoma Screening–Results

- Mass NB screening started in Japan in 1974
- Early results promising (most patients cured), so mass screening started in North America and Europe
- The prevalence of NB in screened populations more than doubled (1/2-3,000 births vs. 1/7-8,000 births)
- Almost all NBs detected by mass screening were favorable biologically and had an excellent outcome
- There was no change in NB mortality or incidence of stage 4 NB in patients over 1 year of age

Lessons from Neuroblastoma Screening—Conclusions

- Screening for NB detected many cases that would not be detected clinically, i.e., they would spontaneously regress.
- NB was 2-3 times more prevalent in screened populations, so at least as many regress as are diagnosed clinically.
- There was no effect on development of NB over 1 year of age, or on overall outcome.
- Favorable NB rarely if ever evolves into unfavorable NB.
- Favorable NBs detected by screening were abnormal genetically, so they are not just unregressed normal cells.
Patterns of Genetic Change Define Risk Groups

(N=82) Mosse YP: GCC, 2007
Genomic Subtypes of Neuroblastoma

Type 1
- 3N
- 3p–, 4p–, +7, 11q–, 17q+

Type 2A
- 2N
- 17q+
- 2N/4N
- 3p–, 4p–, +7, 11q–, 17q+

Type 2B
- 2N
- 17q+
- 2N/4N
- 1p–, 17q+
- CHD5
- MYCN amp.

ALK?

Possible Mechanisms of Spontaneous NB Regression

Immune killing/ADCC  TrkA/NGF signaling pathway

1. TrkA and TrkB receptors
2. Antibodies
3. Telomerase inhibition
4. Methylation changes

Telomere shortening
Epigenetic changes

NB Regression–Loss of Telomerase

- Telomerase activity detected in 94% of NBs (N=100)
- NBs with high telomerase had *MYCN* amplification or other high-risk features, and a poor outcome
- 3 of 8 stage 4S NBs had no telomerase activity
- Loss of telomerase activity would lead to continued telomere shortening and ultimately apoptosis
- This may account for at least some cases of spontaneous regression

NB Regression–Loss of Telomerase

Lu MH, Oncol Rep, 2012
NB Regression–Immune Mechanisms

Cellular Immunity

- Tumor-infiltrating lymphocytes (TILs), NK cells and tumor-associated macrophages (TAMs) found in some NBs
- The number of TILs, TAMs in 4S similar to other stages

Humoral Immunity

- Antineural antibodies found in pts with OMS, but half have NBs, so others may have regressed NBs or (AID)

Most NBs have low/absent Class I HLA expression
NB Regression–Immune Mechanisms

- DNAM/PVR activates NK function
- B7-H3 inhibits NK function
- HLA class 1 downregulated in most NBs
- NB can be upregulated by IFN$\gamma$ NB
- Anti-GD2 induces ADCC and IFN$\gamma$ release, increasing HLA Class I expression
Neural Crest Migration

- NC progenitors migrate to symp. ganglia, adrenal, other sites
- Progenitors express TrkA (±TrkC) but are independent of NGF
- Many more progenitors produced than are needed to form PNS
- Migrating NC cells that connect to a target organ survive
- Other NC cells undergo delayed activation of developmentally programmed apoptosis
- This may be regulated primarily by NGF dependence
# TRK Family Neurotrophin Receptors

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<th>BDNF</th>
<th>NT3</th>
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TRK Signaling–TrkA as Example

- Signaling pathways similar for all three TRK receptors
- Signaling mainly through the RAS/MAPK and PI3K/AKT pathways
- RAS/MAPK more important for neural differentiation
- PI3K/AKT may be most important for survival

Eggert A, Oncogene 2000; Brodeur GM, Cancer Res, 2009
TrkA Isoforms: TrkAI and TrkAIII

- TrkA activation by NGF induces neuronal differentiation
- TrkAIII splices out exons 6 and 7
- TrkA reading frame is maintained, but ligand binding domain lost
- TrkAIII is constitutively active, but cells do not differentiate
NB Regression—Role of TrkA/NGF

- Migrating NC progenitors express TrkA3 and are independent of NGF in their microenvironment

- More mature NC progenitors express TrkA1 and require NGF for survival

- Normal cells die in the absence of NGF, but some cells may survive, accumulate other genetic changes—>NB

- Spontaneous regression may result from delayed activation on developmentally programmed apoptosis

- This could be caused by isoform switch from TrkA3 to TrkA1
Mechanisms of NB Regression

Immune killing/ADCC

TrkA/NGF signaling pathway

1. NGF binds to TrkA and TrkB receptors
2. Antibodies target TrkA and TrkB receptors
3. Telomerase activity is decreased
4. Methylation changes occur

Telomere shortening
Epigenetic changes

NB Regression–Conclusions

- Cause of spontaneous regression may be multifactorial
- TrkA3 to TrkA1 isoform switch may lead to apoptosis and regression for some NBs
- Partial NB apoptosis may elicit immune NK response to dying tumor cells and eliminate remaining tumor cells
- A primary NK cellular immune response is possible, but most NBs have low expression of HLA I
- Telomere shortening may also play a role
- The ability to trigger apoptosis/regression would be an appealing approach to therapy
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