

Grand Rounds

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Department of Ophthalmology
October 24, 2013



Case Presentation

Ophthalmology was consulted on a neonate shortly after delivery.

NICU team reported partial fusion of eyelids and inability to visualize the globes.

History

- Primigravid mother (29 y/o, G1P0) and father reportedly healthy, no known familial disorders or social history
- Full-term uncomplicated pregnancy
 - 2640 g, 40 week gestation
 - Quad screen positive for increased risk of Down Syndrome 1:74, refused aminocentesis
 - Hx of spontaneous abortion in first trimester and ectopic pregnancy
 - APGAR 8/8

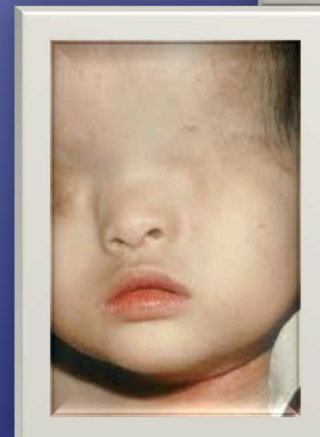
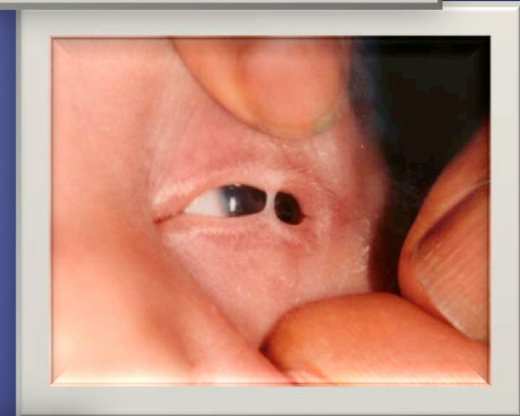
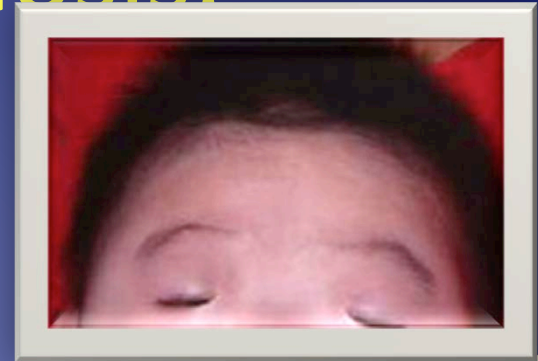
Case Presentation



Differential Diagnosis?

Differential Diagnosis:

- Microphthalmia
- Anophthalmia
- *Blepharophimosis Syndrome*
- *Ankyloblepharon*
- *Cryptophthalmos*



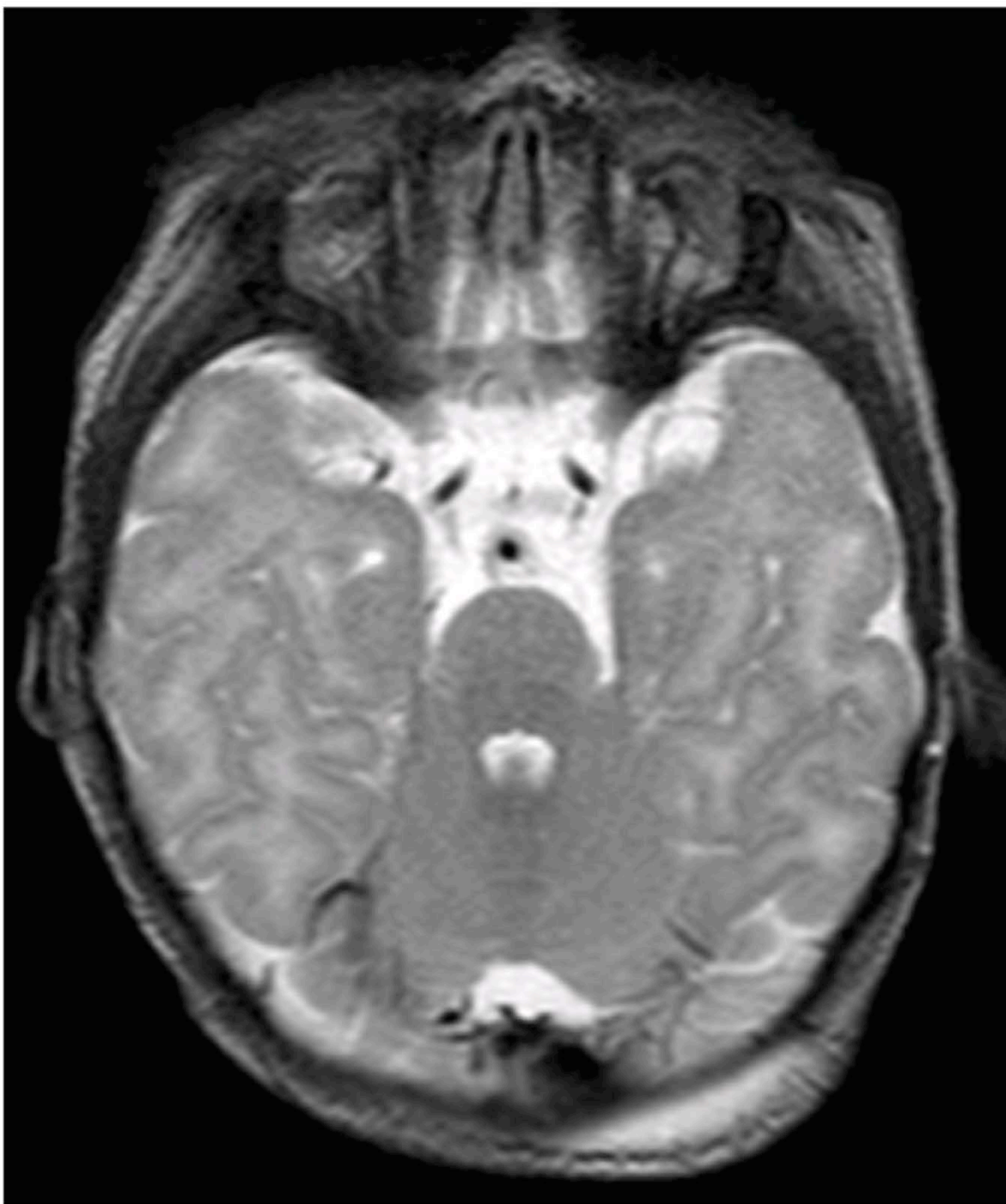
NEXT STEP?

Imaging? Anything else... urgent?



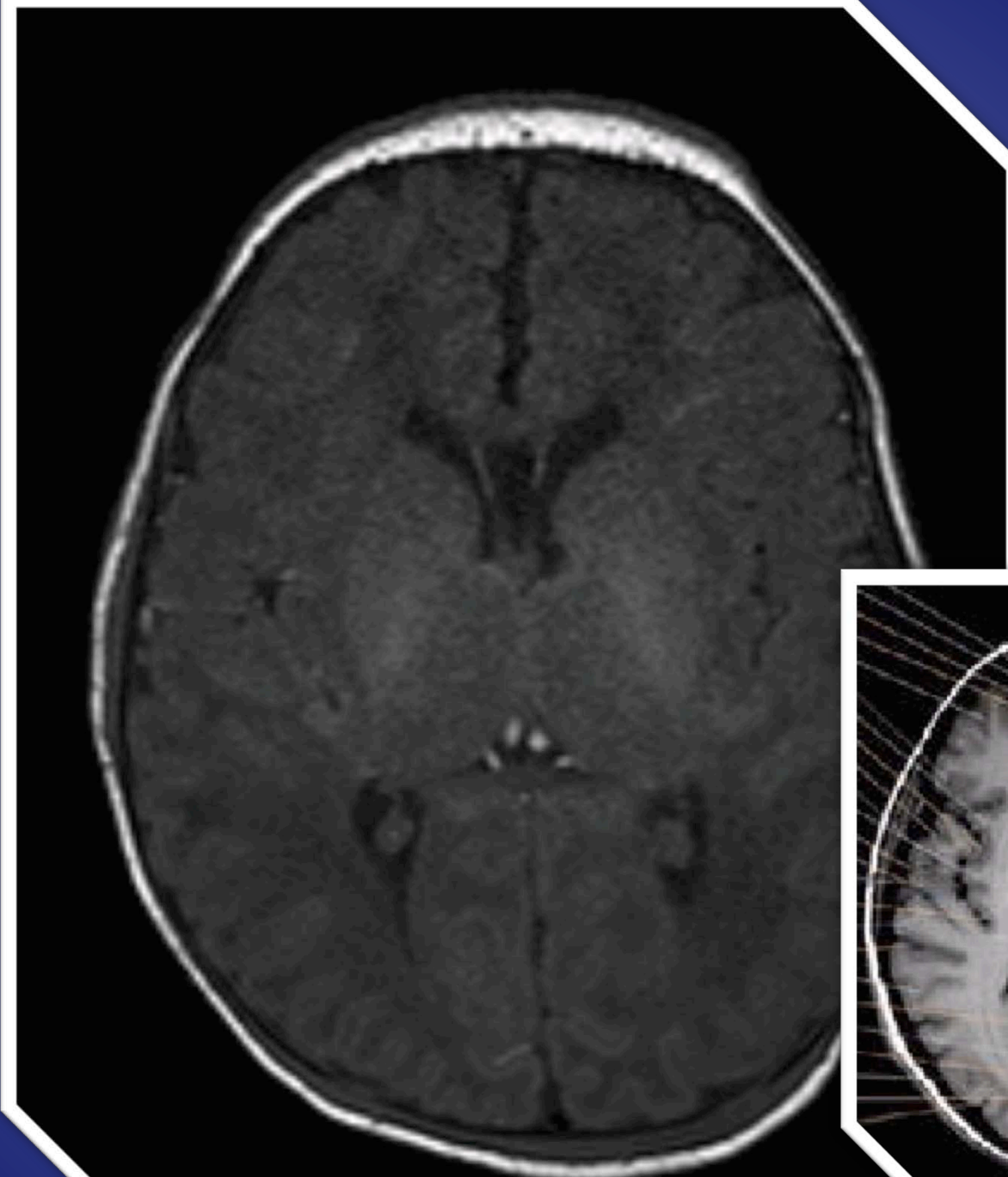
Initial Workup

- B-Scan: No globes, organized soft tissue, or cystic structures visualized
- Urgent:
 - Full metabolic and endocrine workup
 - Cardiac, gastrointestinal, and genitourinary evaluation
 - TORCH titers: negative
- Genetics: 895 kb deletion, includes *SOX2* gene
- Imaging:
 - Ultrasonography vs MRI
- Consultations:
 - Endocrinology and Pediatric Geneticist
 - +/- ENT









Pituitary Function Testing

- Pediatric Endocrinology was consulted
- LH/FSH thought to be low.
- Cortisol low-normal:
 - ACTH Stimulation test with normal response- may be physiologic.

Infant Hearing Screen

ep 13 1118

Event Status

supplemental

#

Results of several evaluations suggest the auditory system in the LEFT ear to be functioning normally through the level of the brainstem (approximately the inferior colliculus) Furthermore review of right ear cochlear microphonics suggests that auditory neuropathy/auditory dysynchrony (AN/AD) is not present . RIGHT EAR data - the ABR data again demonstrated poor morphology, waves were not as replicable as 7/17/13 and wave V was not readily identifiable at 60 dB nHL. This may be a result of child's state of arousal, this audiologists interpretation of the data or a true change in responses. DPOAE screening was passed but TEOAE screening was not passed. This suggests that the auditory system has no more than a moderate impairment through the level of the outer hair cells of the cochlea but retro-cochlear problems such as AN/AD could not be ruled out. All OAE data is pre-neural/pre-synaptic.

IMPRESSION: Peripheral hearing is grossly adequate in at least one ear. Peripheral hearing is grossly adequate in at least one for the development of speech and language ear. A delayed onset hearing loss cannot be ruled out for either ear RECOMMENDATIONS: 1. Per pediatrician. 2. Per team at Spence-Chapin Agency 3. Child should have further audiological evaluation to determin hearing acuity in right ear. Furthermore, child must have on-going audiological evaluation due to chromosomal anomalies (see genetics note dated 8/16/13) and vision problems. No follow-up appointment given because child is to be placed with adoptive family next week (per social worker) Social worker informed of need for on-going audiological care and she expressed understanding.

Anophthalmia

- Anophthalmia: Congenital absence of optic tissue
 - True Anophthalmia: Histological absence of neuroectodermal tissue
 - Clinical Anophthalmia: Absence of globe clinically and radiologically
- Microphthalmia:
 - Axial length less than 2.5 SD below mean
 - With or without cyst

Prevalence

- Anophthalmia: 3/100,000 births
- 1/8 chance in siblings
- 2/3 are due to genetic abnormality

Classification

- *Primary Anophthalmia:*
 - Primary optic vesicle does not develop from cerebral vesicle (weeks 0-4)
 - Rare, bilateral and sporadic
- *Secondary Anophthalmia:*
 - Failure of development of anterior neural tube
 - Rare, may be fatal
- *Consecutive/Degenerative Anophthalmia:*
 - Optical vesicles form, but degenerate
 - e.g., lack of blood supply (unilateral)

Anophthalmia vs Microphthalmia

- Considered by many as part of the same spectrum
- Distinctions may not matter
- Often overlap



Causes

- Genetic:

- *SOX2* mutation:

- found in 10% of anophthalmic/
microphthalmic cases

- Other: OTX2, PTCH, CHD7
(CHARGE Syndrome), PAX6,
RAX, CHX10, BCOR, BCL

CHARGE syndrome
Goltz Syndrome
Branchio-oculo-facial
syndrome

Other causes:

Gestational infections,
typically viral
Toxoplasmosis, Rubella,
Influenza virus
Vitamin A deficiency,
Thalidomide, Radiation

- Chromosomal:

- Trisomy 13, Mosaic trisomy 9

- Syndromes (molecular):

- Lenz microphthalmic
syndrome
 - Matthew-Wood Syndrome

Associated Syndromes

- More than 50% of patients with A/M have extraocular findings: Musculoskeletal abnormalities, limb malformations, anomalies of the face, ear, and neck.
- 25-30% with chromosomal abnormalities
- 20-40% with an associated syndrome

SOX2 gene mutations

- 4-20% of cases of A/M
- The most common known cause for A/M
- Usually a severe, bilateral A/M
- SOX2 is a transcription factor, codes a protein with a high mobility group DNA binding domain, interacts with PAX6 and OTX2 to effect gene regulation, , coregulates RAX.
- Majority *de novo*, but may be inherited as autosomal dominant pattern.

SOX2 Mutations

- Ocular findings: iris hypoplasia, cataracts, colobomas, pupillary defects, hypermetropia, retinal dystrophy, retinal detachments
- Neurological findings: mesial-temporal hamartomas, gray matter heterotopias, mesial temporal malformations, agenesis of the corpus callosum, disordered muscle tone, ataxia, seizures

SOX2-associated syndromes

- Endocrine abnormalities:
 - Pituitary hypoplasia → profound gonadotropin deficiency → hypogonadotropic hypogonadism.
 - Growth retardation
 - Dolichocephaly, facial asymmetry, tall forehead, short and narrow palpebral fissures, dysplastic ears, hearing loss
- Anophthalmia-Esophageal-Genital syndrome
 - Tracheo-esophageal fistula, esophageal atresia, cryptorchidism, micropenis, hypospadias, horseshoe kidney

Lee Langer, Kathleen Sulik, Larysa Pevny, (2013) Cleft Palate in a mouse model of SOX2 haploinsufficiency. *The Cleft Palate-Craniofacial Journal* In-Press.

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ONLINE AHEAD OF PRINT

Cleft Palate in a Mouse Model of SOX2 Haploinsufficiency

Lee Langer, Ph.D., Kathleen Sulik, Ph.D., Larysa Pevny, Ph.D.

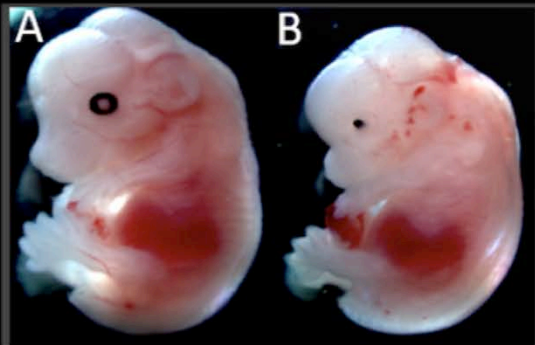


Figure 1: Reduced levels of SOX2 result in anophthalmia in mice. A) E14.5 control embryo with well-developed eye. **B)** Age matched Sox2 hypomorphic embryo displaying a degenerated eye.

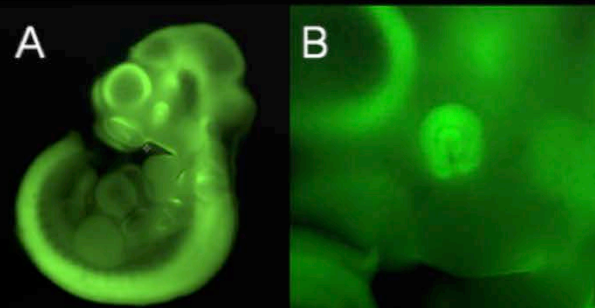


Figure 2: GFP reporter expression in an E10.5 embryo. A) Whole mount image of a control E10.5 embryo indicating expression of SOX2 along the length of the neural tube. **B)** Magnified image of the eye, with the retina appearing as a ring of fluorescent tissue underlying the surface of the embryo and surrounding the forming lens

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Novel SOX2 Mutations and Genotype-Phenotype Correlation in Anophthalmia and Microphthalmia

Adele Schneider¹, Tanya Bardakjian¹, Linda M. Reis², Rebecca C. Tyler², and Elena V. Semina^{2,3,*}

- Screened 51 A/M cases for SOX2 mutations
- SOX2 mutations found in 10
 - 7 of which were bilateral (21% of bilateral cases)
- The range of SOX2 mutations range from bilateral anophthalmia with severe neurological maldevelopment to normal
- Deletion mutations much more severe than missense mutations (33% bilateral, 33% other ocular malformation, 33% normal)

Mutations within *Sox2/SOX2* are associated with abnormalities in the hypothalamo-pituitary-gonadal axis in mice and humans

Daniel Kelberman,^{1,2} Karine Rizzoti,³ Ariel Avilion,⁴ Maria Bitner-Glindzicz,² Stefano Cianfarani,⁵ Julie Collins,³ W. Kling Chong,⁶ Jeremy M.W. Kirk,⁷ John C. Achermann,¹ Richard Ross,⁸ Danielle Carmignac,⁹ Robin Lovell-Badge,³ Iain C.A.F. Robinson,⁹ and Mehul T. Dattani¹

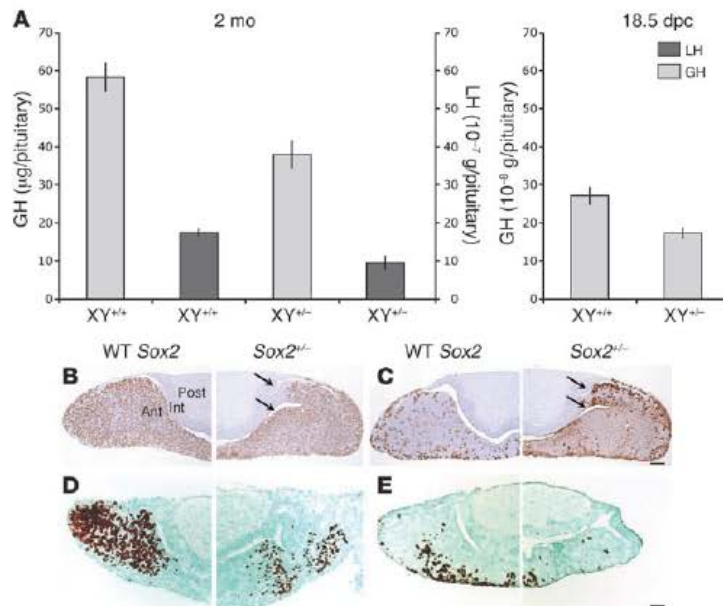


Figure 1

Pituitary hormone levels and immunohistochemistry in *Sox2* heterozygous mice. (A) GH and LH RIAs on pituitary protein extracts from 6 wild-type (XY^{+/+}) and 5 *Sox2* heterozygous (XY^{+/-}) 2-month-old littermates (GH, $P = 0.004$; LH, $P = 0.002$) and 10 wild-type and 9 *Sox2* heterozygous 18.5 dpc littermates ($P = 0.002$). Both GH and LH were affected; GH deficiency appeared before birth. (B–E) Immunohistochemistry for GH (B and D) and LH (C and E) on pituitary sections from 3-month-old (B and C) and 18.5 dpc (D and E) wild-type and *Sox2* mutant littermates. Ant, anterior lobe; Int, intermediate lobe; Post, posterior lobe. Note the presence of extra clefts in the adult sections (arrows in B and C). Staining was clearly reduced in heterozygotes at 18.5 dpc (D and E). Scale bar: 0.3 mm (B and C); 0.05 mm (D and E).

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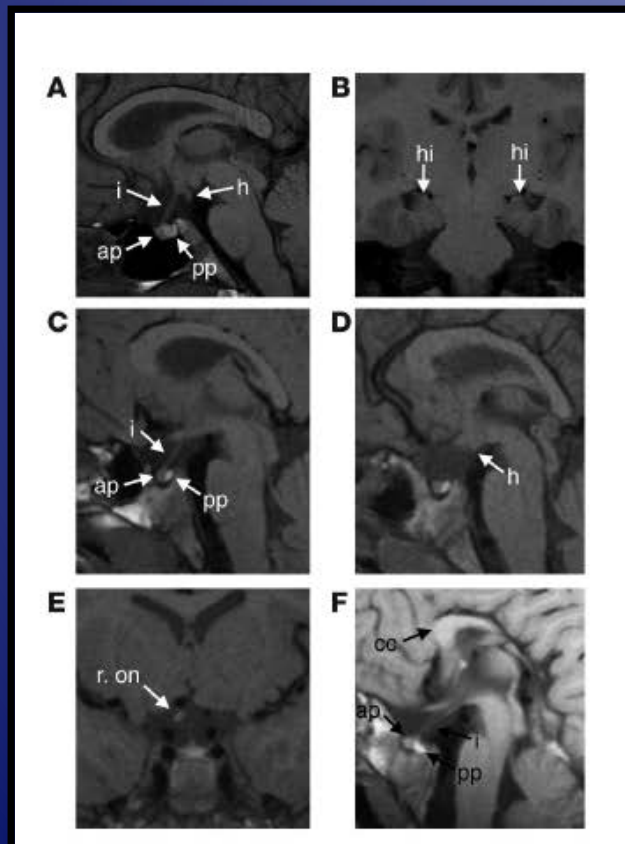


Figure 6

MRI scans on patients with *SOX2* mutations. (A and B) Sagittal and coronal sections from patient 1 showing APH with relatively normal posterior pituitary (pp) and infundibulum (i), a hypothalamic hamartoma (h) and a misshapen abnormal hippocampus (hi). (C and D) Sagittal sections from patient 3 showing APH and a hypothalamic hamartoma. (E) Coronal section in this patient revealed complete absence of the left optic nerve. r.on, right optic nerve. (F) Sagittal section in patient 4 showing hypoplasia of the anterior pituitary (ap) and the splenium of the corpus callosum (cc). The posterior pituitary and infundibulum were normal.

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Clinical phenotype in patients with SOX2 mutations

Pt	Mutation	MRI	Ocular phenotype	Other features
1	c.60insG	Hippocampal abnormalities, small corpus callosum, hypothalamic hamartoma, APH, generalized reduction of white matter bulk, absent optic nerves	Bilateral anophthalmia	HH, learning difficulties, spastic diplegia, esophageal atresia
2	c.70del20	Hippocampal abnormalities, abnormal anterior pituitary, absent left optic nerve	Left anophthalmia, right microphthalmia	HH, learning difficulties
3	c.387delC	Hypoplastic corpus callosum, APH, hypothalamic hamartoma, small left optic nerve and chiasm, generalized lack of white matter bulk, hippocampal abnormalities with small and rotated mesial temporal structures	Left microphthalmia, right coloboma	HH, cryptorchidism, micropenis, learning difficulties, mild spastic diplegia
4	Y160X	Partial agenesis of corpus callosum, small anterior pituitary, hippocampal abnormalities, generalized reduction in white matter	Bilateral microphthalmia	HH, cryptorchidism, micropenis, severe learning difficulties, spastic and dystonic quadriparesis
5	Q177X	Not done	Bilateral anophthalmia	HH, cryptorchidism, micropenis, severe learning difficulties, mild facial dysmorphism
6	c.479delA	APH, small hippocampus, thin corpus callosum, cavum septum pellucidum, absence of optic nerves and chiasm	Bilateral anophthalmia	HH, small testes, micropenis, learning difficulties, sensorineural deafness
7	G130A	Absent septum pellucidum, bilateral optic nerve hypoplasia, bilateral schizencephaly, right porencephalic cyst, normal anterior and posterior pituitary	Roving nystagmus with bilateral optic nerve hypoplasia	Short stature with a normal growth velocity; endocrine status not investigated
8	A191T	Absent septum pellucidum, small optic chiasm, absent infundibulum, severe APH, ectopic-undescended posterior pituitary	Roving nystagmus with bilateral optic nerve hypoplasia	GH, TSH, and ACTH deficiency

Other Mutations

- OTX2: 2-3% of A/M (30 examples)
 - Also associated with anterior segment defects, Leber's congenital amaurosis, hypoplasia/aplasia of the optic nerve and chiasm.
 - Associated with pituitary abnormalities in 19-30%
 - Genital, neurological, and growth retardation defects reported

Molecular links among the causative genes for ocular malformation: *Otx2* and *Sox2* coregulate *Rax* expression

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Edited by Igor B. Dawid, National Institutes of Health, Bethesda, MD, and approved February 20, 2008

“The neural-related genes *Sox2*, *Pax6*, *Otx2*, and *Rax* have been associated with severe ocular malformations such as anophthalmia and microphthalmia, but it remains unclear as to how these genes are linked functionally. **SOX2-missense mutations identified in these ocular disorders. These results demonstrate that the direct interaction and interdependence between the *Otx2* and *Sox2* proteins coordinate *Rax* expression in eye development, providing molecular linkages among the genes responsible for ocular malformation.**”

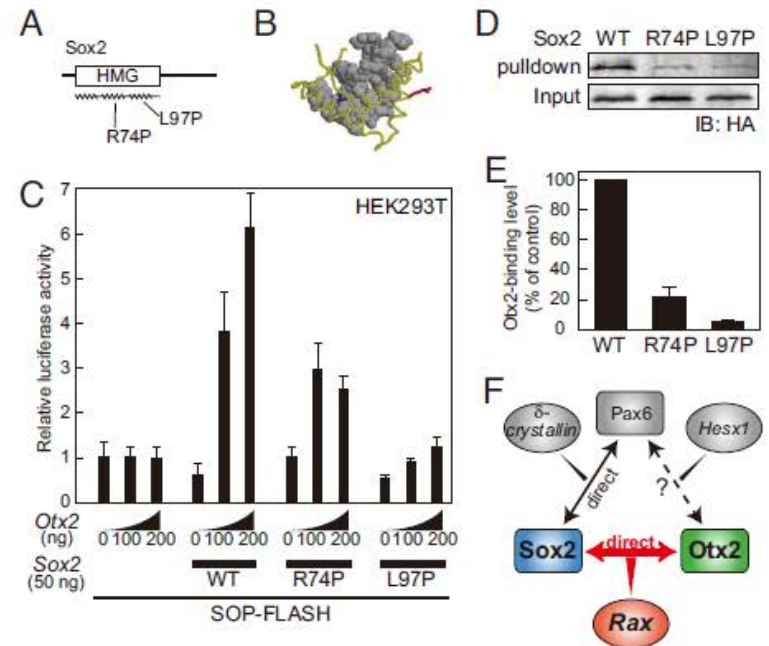


Fig. 5. Missense mutations in the Sox2 HMG domain affect the activities of Sox2 protein associated with Otx2. (A) Missense mutations identified in helices 2 and 3 of the human Sox2 HMG domain (R74P and L97P). (B) The 3D structure of the Sox2 HMG domain (yellow) binding to DNA (gray), based on a previous study (30). The side chains of the 74th arginine (blue) and 97th lysine (red) do not come in direct contact with the DNA. (C) Mutated Sox2 proteins do not induce transcription via CNS1 in cooperation with Otx2. (D) GST pull-down assay showing that the mutated Sox2 proteins lose Otx2-binding activity. HA-tagged Sox2 proteins were *in vitro*-synthesized and pulled down by GST-Otx2, followed by Western blotting with antibody against HA-tag. (E) Quantification of Otx2-binding levels by using the Odyssey infrared imaging system. Error bars indicate SD values. (F) Molecular relationships among ocular malformation-associated genes. The present work demonstrates that a direct interaction between the Otx2 and Sox2 proteins coordinately regulates *Rax* expression (colored). Upstream proteins and target genes are indicated as round rectangles and ovals, respectively. A full scan of the Western blotting data is shown in Fig. S7.

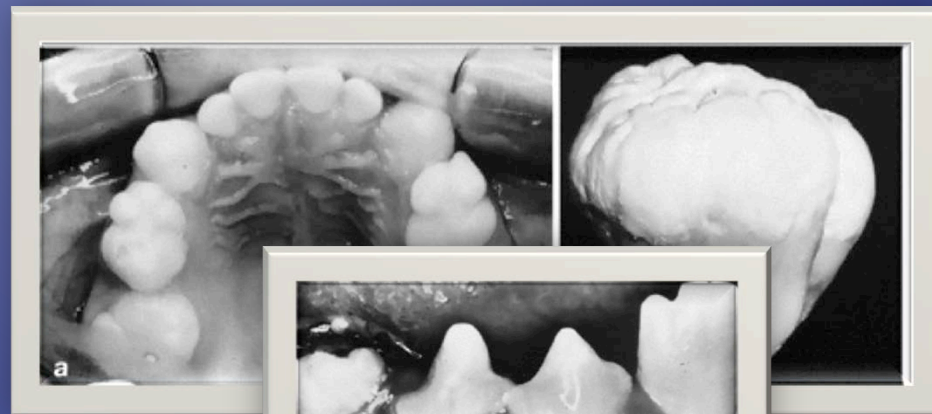
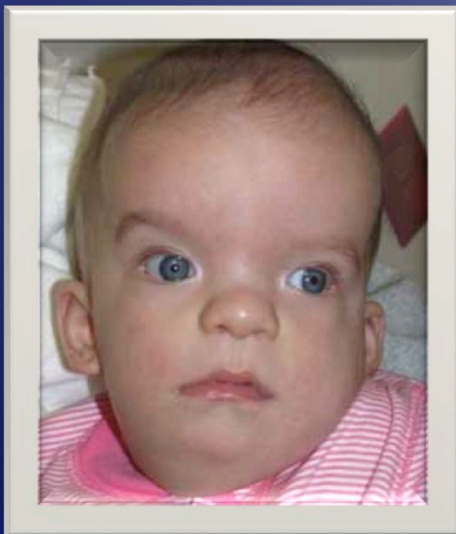
Other Mutations

- Matthew-Wood Syndrome
 - PDAC (pulmonary hypoplasia/agenesis, Diaphragmatic hernia/eventration, Anophthalmia/Microphthalmia, Cardiac Defects



Other Mutations

- Oculofaciocardiodental syndrome
 - Lenz Microphthalmia (Both BCL6 mutations)
 - Long and narrow face, cataracts, atrio-ventricular septal defects, aortic stenosis, Pentalogy of Fallot



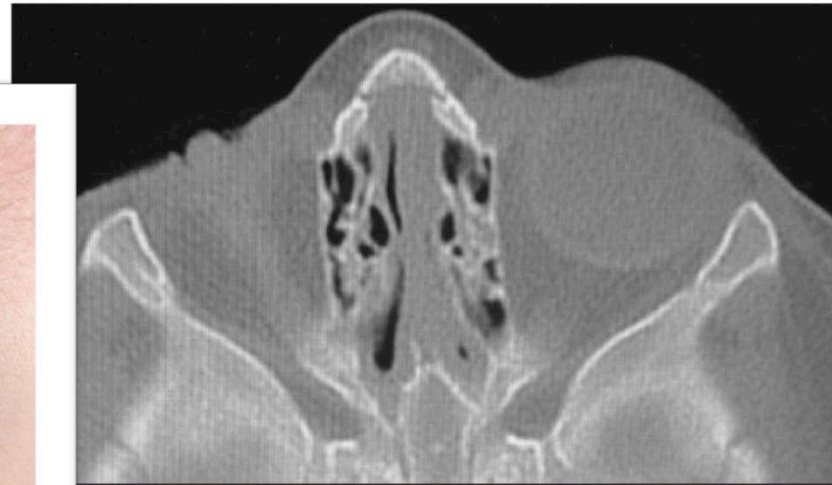
Other Mutations

- Microphthalmia with Linear Skin Defects Syndrome (50 cases)
 - Also known as MIDAS (Microphthalmia, Dermal Aplasia, Sclerocornea)
 - A/M –unilateral/bilateral – PLUS congenital skin defects (linear and patchy erythroderma)



Management of Anophthalmia

- Socket Expansion:
 - The globe volume in a neonate is 70% of an adult's size, the orbit is 40%.
 - Facial development depends in part on the orbit and its expansion in the first 2-4 years of life
 - A shallow orbit will often lead to severe hemifacial malformation.



Socket Expansion

- Unilateral anophthalmia: warrants very aggressive expansion to prevent asymmetry
- Should begin within weeks of birth
- Expansion of conjunctival sac first, then orbit
- Microphthalmos with vision: added complexity of preserving vision in that eye → clear conformer, then painted conformer with clear pupil



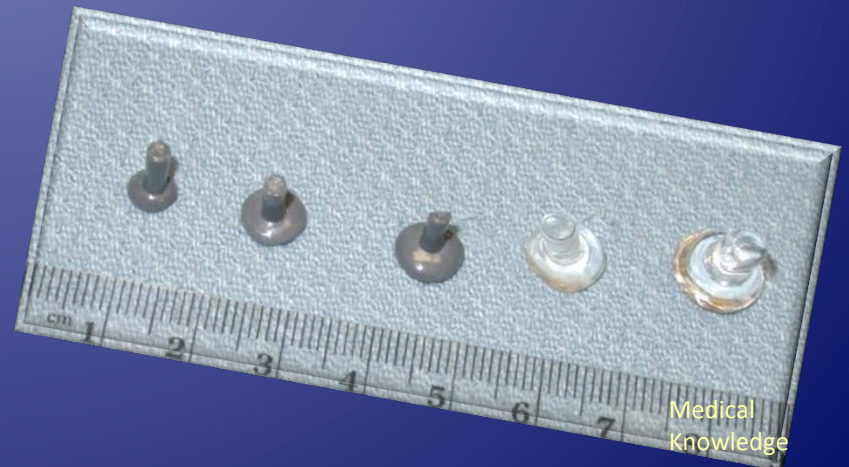
Other considerations...

- Lid expansion, horizontal and vertical
 - Often, patients have microblepharon, with phimotic palpebral fissures
 - Principle: soft tissue resonates with underlying musculoskeletal structures
- Expansion of the conjunctival sac and fornices
- Expansion of the orbit

Modalities of Expansion

Serial conformers:

- In conjunction with an ocularist, conformer made to fit into the orbital space available.
- As the socket grows to accommodate, larger conformers are used
- Lids expanded anteriorly, and conjunctiva/fornix posteriorly.



Socket Expansion

- This used to involve operative management with surgically molding the orbit under anesthesia (repeatedly)
- Hydrophilic expanders: non-invasive, may be done by ophthalmologist.
- Requires parent cooperation: child must have conformer in place at all time
- Versatile: can be used in anophthalmic sockets, over an implant, or over microblepharon

Surgical Implants

- Static vs Dynamic
- Static:
 - Spherical implant, typically acrylic or silicone
 - Progressive increases in size in operating room
 - Mimics orbital development
 - Multiple surgeries
 - Large implant: less surgery, higher chance of extrusion or exposure

Surgical Implants

- Dynamic:
 - Dermis fat grafts
 - Ideal implant: biocompatible, grow with time.
 - Second surgical site, typically gluteal fat
 - Variable: may atrophy (or hypertrophy)
 - Risks: discharge, bleeding, pyogenic granuloma





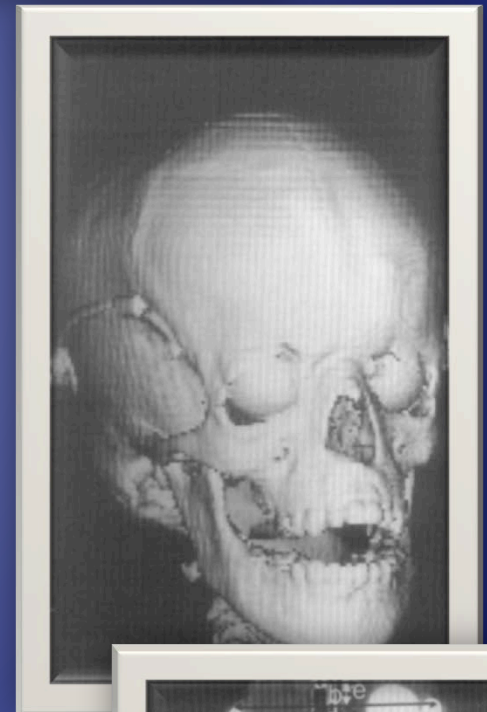
Intraorbital tissue expansion in the management of congenital anophthalmos

D. J. Dunaway and D. J. David

Australian Craniofacial Unit, Women's and Children's Hospital, North Adelaide, South Australia

- **Fluid Chamber:**

- Fluid chamber with progressive saline expansion
- Bladder fixed to bone, subperiosteal. Filling tube leads to the temporalis fossa where an injection port lies.
- Painful, high risk of erosions, extrusion. Requires orbitotomy.



Use of self-expanding, hydrophilic osmotic expanders (hydrogel) in the reconstruction of congenital clinical anophthalmos

Robert A. Mazzoli^{a,b,c}, William R. Raymond, IV^{a,c,d}, Darryl J. Ainbinder^{a,b,c} and Elizabeth A. Hansen^{a,c}

Hydrogel orbital expander: highly hydrophilic polymer

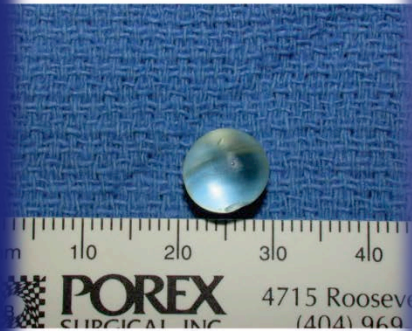
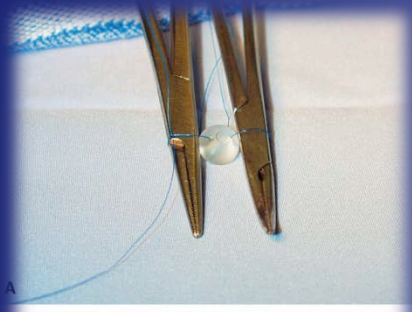
- Expands by osmotically imbibing water, inserted in a dehydrated state

- Expands 10-fold, 20-30 mmHg

- Maximum at 30 days

- Used to make contact lenses, IOLs

- Hemisphere: conjunctival expansion



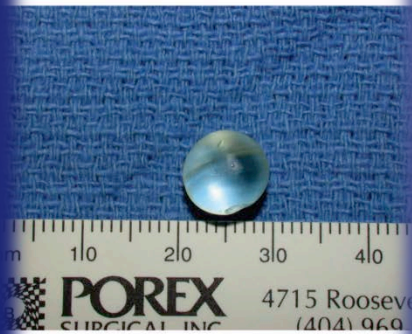
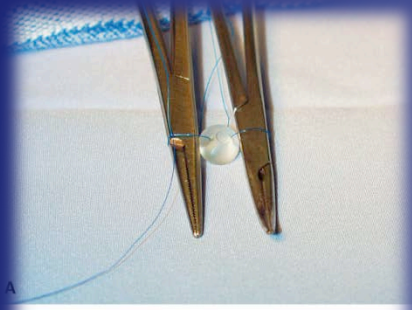
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Once conjunctival socket is expanded, 2/3/4 cc volume implants are then used.

Removed piece-meal due to its consistency

History of MIRAgel: granuloma, IOI, orbital fibrosis



EXTENDED REPORT

Injectable self inflating hydrogel pellet expanders for the treatment of orbital volume deficiency in congenital microphthalmos: preliminary results with a new therapeutic approach

M P Schittkowski, R F Guthoff

- Injectable pellets
- Each pellet
- Can be done
- Injected trans
- rim, directed
- Titrate via op
- Can be used
- the globe (not for use when vision exists)



Question for Audience

- What type of socket expansion devices would you use, initially and later in the course?

Back to our patient...

- Extensively evaluated by Pediatrician, Pediatric Geneticist, Pediatric Endocrinologist, Otolaryngologist, and Ophthalmology
- Found to have SOX2 deletion, low cortisol level s/p supplementation, decreased hearing in the right ear,
- Was sent to ocularist, where four serial conformers were placed
- The baby was ultimately adopted and moved out of state. Full history was sent with patient along with communication with local ocularist and ophthalmologist. Will follow up with local endocrinologist, and have follow up hearing evaluation

Reflective Practice

This case taught me the value of professionalism and patient care in the face of a difficult medical, social, and ethical situation. I learned the value of formulating a good differential diagnosis and careful evaluation for known disorders. I worked together with the attending, pediatrics, pediatric endocrinology, pediatric geneticist, ENT, and the neuroradiology department to carefully manage this patient in need.

Core Competencies

Patient Care- Took care to provide patient care that was compassionate and appropriate, and effective

Medical Knowledge- Recognized the signs and symptoms of Anophthalmia, evaluated for associated defects and medical issues, and treated patients using standardized and a well-thought out plan of care.

Practice-based Learning and Improvement- demonstrate the ability to investigate and evaluate the care of our patients, including improving our methods of management of anophthalmia with regard to literature.

Interpersonal and Communication Skills- demonstrate interpersonal and communication skills with the family, adoptive parents, and interim caretakers, that will result in the effective exchange of information with our patients, teaching and communicating with patient's family in a meaningful way.

Professionalism- demonstrate a commitment to carry out professional responsibilities and an adherence to ethical principles.

Systems-based Practice- demonstrate the ability to call effectively on other resources, such as primary care and ancillary staff in the system to provide optimal health care.

References

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Thank you

- Dr. Elmalem
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- Dr. Pulitzer (Neuroradiology)
- Social Workers, at KCHC
and adoptive agency

