The Pathologies Associated With GNAS Mutations

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Disclosure Statements

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Every discussion today concerning tumors with GNAS mutations begins with:

FIBROUS DYSPLASIA

Suspected fibrous dysplasia from the rib of a Neanderthal, age 120,000 + years.

Definition
- FD is a neoplastic process involving primarily the intramedullary portion of from one to many bones.
- It is composed of randomly distributed spicules of woven bone, absent prominent osteoblastic rimming set in a background of swirling fibrous connective tissue.

Histopathology
- Fibroblastic spindle cells predominate
- Cells are without hyperchromasia or increased mitosis
- Density highly variable
- Cartilaginous differentiation is common
- Stromal variants common

Epidemiology
- Occurs in children & adults
- Neither favors nor spares any racial or ethnic group
- Equally prevalent in both sexes (monostotic form – slight increase in women)
- Found in antiquity
- Found in many vertebrates (apes, dogs, iguanas, etc.)

Clinical Features
- Congenital forms exist
- New disease may occur in the elderly
- Usually discovered in late childhood (polyostotic earlier than monostotic)
- Monostotic form may stop progressing at puberty
- FD usually spares the epiphysis before puberty
- Extends to ends of bone after maturity

Bones of the Head & Neck
- Temporal Bone
- Tympanic Bone
- Orbit
- Paranasal Sinuses (Including Sphenoid)
- Skull Base

RELATIVELY RARE SITES
- Spine (Cervical to Sacrum)
- Hands & Feet
- Fingers and Toes
FD & Other Genetic/Morphologic Conditions

A. Benign lesion probably secondary to cyst-like change
- Frontal sinus or ethmoid mucoceles
- Simple or empty cysts
- Aneurysmal bone cysts


FD & Other Genetic/Morphologic Conditions

B. Other Benign Conditions
- Osteoid osteoma
- Enchondromata with annular calcification
- Myositis ossificans progressiva
- Osteochondromatosis
- Desmoplastic fibroma


FD & Other Genetic/Morphologic Conditions- Continued

C. Coincidental??
- Gout
- Meningiomatosis
- Peutz-Jeghers Syndrome
- Langerhans cell granulomatosis


FD & Other Genetic/Morphologic Conditions- Continued

D. Multi-organ & Malignant Conditions
- Mazabraud’s Syndrome
- McCune-Albright Syndrome
- Both M-AS & Mazabraud’s Syndrome
- Malignant Transformation


Syndromes Associated with FD

Mazabraud’s syndrome


Another Example of a Patient with Mazabraud’s Syndrome

- A 40-year-old woman presented with a 2-month history of an enlarging mass of her right proximal forearm.
- She had first come to medical attention due to an abnormal gait and a bowing deformity of her forelegs 35-years earlier.
She was of short stature with features suggesting deformities of her maxilla and zygomas bilaterally.

Multiple café-au-lait pigmented macules were present on her neck and back, predominantly left sided.

A scar over her left buttock was secondary to an intra-muscular myxoma removed 10 years prior.

- There was a tender mass palpated over her right proximal radius.
- No epitrochlear or axillary lymph nodes were clinically enlarged.

Radiologic Findings

- Conventional radiographs demonstrated typical features of fibrous dysplasia in the pelvis, femurs, and humeri.

Radiographic of the right proximal radius demonstrated aggressive lytic destruction of bone with a modest periosteal reaction.
Radiologic Findings
MR of the elbow depicted the extent of the large proximal radial mass.

Pathologic Findings
On initial biopsy, a cellular spindle cell lesion in a background of skeletal muscle was seen.

Histopathologic Findings
A second biopsy was performed 4 days later. Pleomorphic spindled cells producing tumor osteoid was noted. There was a high mitotic rate but no necrosis.

Additional History
The patient underwent preoperative radiation therapy, ifosfamide containing four-drug chemotherapy and subsequent surgical resection.
Gross Features

The patient had a surgical resection of her proximal radius and ulna, distal capitulum of the humerus and surrounding soft tissues and skin.

A 5 x 2.5 x 2.5 cm lobular tan tumor mass was identified within the proximal radius. The consistency varied from soft to 'bone hard.'

Microscopic Features

Residual osteoblastic osteosarcoma was identified with a Huvos histologic response grade of III. (< 5% viable tumor)

Microscopic Features

All margins were free of tumor but all 3 bones demonstrated fibrous dysplasia.

Follow-Up

The patient subsequently underwent resection of 2 of the larger gluteal masses. Gross and histologic evaluation revealed intramuscular myxomas. Karyotyping showed a normal 46,XX.

Additional History

2 years later, the patient re-presented with an enlarged mass on the lateral aspect of her elbow.

Radiology

Conventional radiographs of the humerus, radius & ulna showed changes typical of FD. MRI of the distal right humerus demonstrated a heterogenous mass with ↑ T2 weighted signal.

A cell block showed markedly atypical cells with high pleomorphism was noted as were bizarre mitosis.
The patient underwent a right above-elbow amputation and a 10 x 6.5 x 3.7 cm soft tissue, cystic, red-brown mass was removed.

Histologic evaluation verified recurrent osteosarcoma.

Cytogenetic studies using GTG banding techniques showed a normal female karyotype (46,XX) in the FD portion of the spectrum which was confirmed by FISH in 98% of the cells.

FISH carried out by us using probes for CH 5 (D5S23) & CH 7 (ELN, Link 1) showed trisomy 5 & 7 in ~66% of 0GS cells, 2% of FD cells & 0% of "normal" cells.

The 0GS demonstrated a 48,XX,+5,+7 karyotype.

Comparative Genomic Hybridization Findings:

+5q11.2-q22.1, +7p12-p21, +7q21-q25(1q22), -Xq11-q27, +1p12-p31, +1q21-q32, +2p23-p26, +2q11.2-q21, +3q, -3p, +4p, +4q, -5q11-q23, -5p11-q15, +6p11.2-p21.3, -6q27-q28, +7p12-p21, +7q21-q25, +8p11-p12, +8q11.2-q23, +9p, +9q, +11q1, +12p12-p13, +13q, +14q, -15q, -16q, +17q, -18q, -21q, -22.
Syndromes Associated with FD

McCune-Albright Syndrome:

Syndrome characterized by Osteitis Fibrosa Disseminata, areas of pigmentation and endocrine dysfunction with precocious puberty in females


McCune-Albright Syndrome

- Polyostotic FD
- Café-au-Lait skin changes
- Precocious puberty
- Endocrinopathies

McCune-Albright Syndrome Endocrinopathies

- Hyperthyroidism
- Growth hormone excess (pit. gigantism/acromegaly)
- Neonatal Cushing’s syndrome
- Hypogonadism (↓ gonadotrophin hormones)

Collins, Orphanet J Rare Dis: 7 Suppl 1: S4:2012

Malignant Tumors Arising in FD

- Osteosarcoma
- Chondrosarcoma (including dediff & mesenchymal)
- Fibrosarcoma

Rarer Malignant Tumors Associated with FD

- Ewing’s Sarcoma
- Malignant Mesenchymoma
- MFH
- Angiosarcoma
- Leiomyosarcoma

Collins, Orphanet J Rare Dis: 7 Suppl 1: S4:2012

Another Example of a Patient with a Malignant Tumor Arising in Fibrous Dysplasia

- A 55 year old Caucasian woman presented with headache and neck pain of three months duration.
- She was otherwise in excellent health without known major illnesses or surgeries.
- A course of antibiotic therapy did not relieve her pain.
- A subsequent trial of steroids was similarly unsuccessful in alleviating her symptoms.

Clinical History

Three weeks prior to admission to our institution she developed blurred vision and “double vision” with drooping of her left eyelid.
Clinical History

- On physical examination she appeared healthy but with ptosis of her left eyelid with inhibition of both lateral and medial gaze.
- An MRI and CT examination were performed.

MRI Examination

- T2 Weighted Image
  - Homogenous enhancement following intravenous contrast injection
  - Replacement of cavernous sinuses
  - Left wing of sphenoid was enhanced as was the tuberculum sella
  - Brain parenchyma was normal

ENT Evaluation

- Nasal endoscopy demonstrated a mass in the superior portion of the nasopharynx which was smooth and mucosally-covered.
- The neck was free of adenopathy and no lesions were appreciated in the oral cavity.
- Following endoscopic evaluation she underwent biopsy of the mass.
Subsequent Course

- Accepted three courses of chemotherapy (Cisplatin, Adriamycin and Methotrexate)
- Except for modest marrow suppression patient did well
- Regained function of left eye and felt clinically improved
- She refused further preoperative therapy

Subsequent Course

- She refused plan of: resection + post surgical gamma knife
- Sought radical resection at multiple other institutions
- Died nine months following initial diagnosis presumably of her disease or its sequela

Final Diagnosis

Osteosarcoma arising in a background of Fibrous Dysplasia

(involving the sphenoid bone, pterygoids and extending into the right nasal cavity)

Sarcomatous changes are RARE in:

Fibrous Dysplasia
- 0.4 – 0.5% in fibrous dysplasia
- 4% in McCune-Albright syndrome

Mazabraud’s Syndrome
- 3/36 patients (includes current patient) developed sarcoma (8.3%)
- Of these 36 patients, 11 had McCune Albright
- 2 of these 11 patients developed osteosarcoma (18.2%)

### Etiology
- Clonal structural aberrations (CH 3, 8, 10, 12, 15)
- Trisomy 2
- McCune-Albright Syndrome
- Polyostotic FD
- Monostotic FD
- Pituitary adenoma
- Intra-Muscular Myxomas

All have the same genetic abnormality: GNAS 1 mutation in the alpha subunit of stimulatory G protein (CH 20 [20q13]).

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### G Protein and Activation of Adenylyl Cyclase

#### Protein Kinase C (PKC) Pathway
- Extracellular
- Cell Membrane
- Intracellular

#### Activated Adenylyl Cyclase
- Extracellular
- Cell Membrane
- Intracellular

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#### Effect of Gain-of-Function Mutation
- PKC Pathway Hyperactive
  - Extracellular
  - Cell Membrane
  - Intracellular
**Effect of Gain-of-Function Mutation**

- PKC Pathway Hyperactive
- Activating mutations in Gs

**PKC Pathway Hyperactive**

- DAG → PKC → DAG → PI(4,5)P2 → PLC → IP3 → Mobilization of intracellular Ca2+ → PKC

**PKA Pathway Hyperactive**

- cAMP + PPi → PKA → STK

**Activating Missense Mutations in the GNAS gene**

- R201H 57%
- R201C 38%
- Q227L 05%

R = Arginine  Q = Glutamine
H = Histidine  L = Leucine
C = Cysteine

**Etiology**

- Activating mutations in GNAS 1 → Adenylyl cyclase
- Neoplastic progression & transformation → c-Fos
- Activation of PKC & PKA Pathways

**Etiology – Con’t**

- Why do mutations in one gene (GNAS 1) cause different diseases?
- Why are clinical manifestations variable in distribution and appearance?
Etiology – Con’t

- Mutations occur post-zygotically in a somatic cell
- Expression depends on size of the cell mass during embryogenesis
- Where in the cell mass the mutation occurs determines the phenotype


GNAS Related Non-Endocrine Tumors

- Renal (Clear) cell carcinoma
- Pancreatic adenocarcinoma
- Colorectal carcinoma (K233N)
- Choledochal cysts
- Low-grade appendiceal mucinous neoplasms


GNAS Mutations

Tibrabosco et al. Mod Path, 10:1336, 2015.

Cystic Pancreatic Tumors

- GNAS mutations = intraductal papillary mucinous neoplasms (R201)
- β-catenin mutations = solid pseudopapillary neoplasms
- VHL mutations = serous cystadenomas

GNAS Related Entities

- Telangiectatic hepatic adenoma
  - (STAT 3 activation – inflammatory phenotype)
- Hamartomatous polyposis of stomach and upper duodenum (Peutz-Jegher and Carney complex excluded [{-} for STK11 & PRKAR1A mutations])
- Gray platelet syndrome
- Fibrous Dysplasia + primary hyperparathyroidism (without McCune-Albright syndrome.)


GNAS Non-Neoplastic Conditions

- Fibrous Dysplasia + encephalocranio-cutaneous lipomatosis (neurocutaneous syndrome involving ectodermal and mesodermal tissue e.g. eye, skin, brain, fat)

G Protein Abnormalities

- **Heterotrimeric**
  - **A.** \( \uparrow \) (up regulated/activated)
    1. McCune Albright Syndrome
    2. Fibrous Dysplasia
    3. Mazabraud’s Syndrome
    4. Etc.
  - **B.** \( \downarrow \) (down regulated/inactivated)
    1. Pseudohypoparathyroidism
    2. Pseudopseudohypoparathyroidism
    3. Progressive Osteoclastic Heteroplasia
    4. Albright’s Hereditary Osteodystrophy

- **Monomeric**
  - **A.** RAS
    1. HRAS-Costello Syndrome
    2. KRAS-Noonan Syndrome
    3. KRAS-Cardiofaciocutaneous Syndrome
  - **B.** RAB
    1. RAB 7-Chondrodysplasia Punctata
    2. RAB 23-Carpenter Syndrome
    3. RAB 27-Griscelli Syndrome Type 2
  - **C.** RHO
    1. RAC 2-Neutrophil Immunodeficiency Syndrome

Conclusions

- GNAS mutations cause constitutive (activating) GS \( \alpha \) signaling
- Mutations occur somatically & result in mosaicism
- FD is one of a host of neoplasms and other pathologic conditions resulting from G protein abnormalities
- The molecular etiology of this family of abnormalities is beginning to be understood

Thanks for Your Attention
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