SC-06: Update in Adnexal Tumors – Sebaceous Tumors

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I have the following financial relationships to disclose:


(mostly scientific advisory boards, clinical trials, research / travel support and business / financial strategy consulting)

These relationships are NOT relevant to the educational content of this lecture.
Roadmap

Sebaceous tumor pathology

Sebaceous tumor biology

Muir-Torre Syndrome (MTS)

MSI / IHC testing & MTS risk stratification
WHO Classification of Skin Tumours

Edited by David E. Elder, Daniela Massi, Richard A. Scolyer, Rein Willemze
**Sebaceous Tumors**

- Sebaceous Hyperplasia
- Ectopic Sebaceous Glands
- Benign Sebaceous Neoplasia
  - Adenoma
  - Sebaceoma
  - Other
    - Sebaceous Epithelioma
    - Superficial Epithelioma with Sebaceous Differentiation
- Sebaceous Carcinoma
  - Ocular
  - Non-ocular
- Other: Nevus Sebaceous, Steatocystoma, Mantleoma, BCC with seb., others
Sebaceous Hyperplasia

- Face
- Older men
- Flesh-colored papule
- Umbilicated
- 1-3 mm
- Do not regress (true hyperplasia)
- Clinical: ? basal cell carcinoma
- No association with MTS
Ectopic Sebaceous Glands

- Unexpected sites
- Open directly to epithelial surface
- No association with hair follicle
- Sites: esophagus, palms & soles, cervix, uterus, tongue, bronchial tree, etc.

- Montgomery tubercles: breast
- Fordyce spots: mucosal lip / oral and genital areas
- Tyson glands: penis
- No association with MTS
Ectopic Sebaceous Glands

Case courtesy of Dr. Huamin Wang, GI Path, UTMDACC
**Sebaceous Adenoma**

- Older adults
- Head & neck region
- Flesh-colored papule; creamy, yellow
- Usually **NOT** umbilicated
- 5+ mm in diameter
- Organoid; increased basal cells
- Associated with MTS
Sebaceous Adenoma
Sebaceoma

- Clinical presentation as adenoma
- Increased basal cell content
  - Greater than 50% of tumor cellularity
  - Lose organoid architecture; just tumor
- Dermis, can involve epidermis
- Often prominent sebaceous ductal structures
- Troy and Ackerman, 1984
- ? Sebaceous epithelioma - ? BCC
- Associated with MTS
Sebaceoma
Nevus Sebaceus-Sebaceoma
Superficial Epithelioma with Sebaceous Differentiation

Case courtesy of Evan Farmer, JHUH & AFIP, Baltimore, MD.
BCC w/ sebaceous dif.

F70, left nasal ala

Case courtesy of Richard Carr, Warwick Hospital, UK.
Ocular Sebaceous Carcinoma

• Up to \( \frac{3}{4} \) of all sebaceous carcinomas

• Ocular sebaceous glands
  – Meibomian, Zeis or other minor glands

• Mild female predominance

• Clinical: chalazion or chronic blepharoconjunctivitis

• Pagetoid upward migration
Ocular Sebaceous Carcinoma

- Difficult diagnosis on small biopsies
- Metastasis and mortality: up to 25%
- Beware: ocular “benign” sebaceous tumors
- Rarely associated with MTS
Sebaceous Immunohistochemistry

- Eosinophilic, bubbly cytoplasm
- Crenylated, indented nuclei
- Fresh tissue: Oil Red O
- EMA(+), CEA(-)
- CK7(+), BerEP4(-)
- New: adipophilin, perilipin, and TIP-47
Adipophilin
Vesicular membranous staining: intracytoplasmic lipid vesicles

Other staining patterns: tumor cells, keratohyaline granules, macrophages
Non-ocular Sebaceous Carcinoma

- \( \frac{1}{4} \) of all sebaceous carcinomas
- Head & Neck region
- Older adults, ? slight male predominance
- Infiltrative border and/or nuclear atypia
- Sebaceous differentiation often focal
- ? Better prognosis (Busam, MSKCC, NY)
- Some association with MTS
Non-ocular Sebaceous Carcinoma
Sebaceous Carcinoma
Human sebaceous tumors harbor inactivating mutations in LEF1


We found that one-third of human sebaceous tumors examined had double-nucleotide substitutions in the same LEF1 allele, irrespective of DNA mismatch repair status. The mutations impaired both LEF1 binding to β-catenin and transcriptional activation, and are the first tumor-associated mutations that inactivate Wnt signaling. Mutant LEF1 not only inhibited expression of β-catenin target genes but also stimulated expression of sebocyte markers, suggesting that it may determine the differentiated characteristics of sebaceous tumors.

of MSH2, or MLH1 and MSH6 (Supplementary Fig. 1 and Supplementary Table 1 online). Therefore, mismatch-repair defects are unlikely to be the primary cause of LEF1 mutations.

Because the S61P and E45K mutations are predicted (Supplementary Note and Supplementary Methods online) to interfere with LEF1–β-catenin binding, we compared transcriptional activation by mutant and wild-type LEF1. We performed TOPFLASH luciferase assays on HEK293 cells, which have endogenous TCF-LEF activity, and on IIA1.6 B cells, which do not express Tcf-Lef genes. We transfected HEK293 cells in the presence or absence of Wnt-3a–conditioned medium (Fig. 1f). We cotransfected IIA1.6 cells with LEF1 and constitutively active (S37A) β-catenin (Fig. 1g).

Transcriptional activation by S61P or E45K single mutants was reduced compared to wild-type LEF1 in both cell types (Fig. 1f,g). Transcriptional activation by the double mutant was further reduced, to a level comparable with ΔN34LEF1 (corresponding to ΔN32Lef1 in KI4ΔNLef1 transgenic mice). In IIA1.6 B cells, the
• Isolate skin stem cells (K15+)
• Transfect mutant *LEF1*
• Drives sebaceous differentiation
• **Tumor promotion**
  • Inhibit ARF/p53/p21 axis
• **Sebaceous fate**
• Isolate “cancer progenitor cells”

Next-generation sequencing identifies high frequency of mutations in potentially clinically actionable genes in sebaceous carcinoma

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Muir-Torre Syndrome (MTS)

- 1968 -- Muir & Torre independently describe patients with both sebaceous tumors and intestinal malignancies

- MTS: (a) sebaceous neoplasm; (b) internal malignancy; [(c) family history]

- Autosomal dominant with variable penetrance for skin tumors
Skin Tumors in MTS

• Sebaceous neoplasms
  – adenoma, carcinoma, epithelioma, sebaceoma (? ocular carcinoma)
  – difficult to classify sebaceous neoplasms (KA-like, cystic)
  – other tumors with sebaceous differentiation reported

• Keratoacanthomomas (Torre does not recognize these w/o sebaceous)
Mechanism of HNPCC/MTS

• Inherit one non-functional MMR gene (usually *MLH1* or *MSH2*)

• Second copy subsequently silenced (often by methylation)

• Clone of mutator cells emerges, markedly increasing rate/chance of procarcinogenic events (loss of TSG)

• Not all HNPCC/MTS patients have MSI (>75 %) -- + other genes
**MLH1 and MSH2**

- Large genes with many exons
- Mutations spread throughout
- Complicated genetic testing
- Easier with NGS
**Microsatellite Repeats in DNA**

- Mononucleotide and dinucleotide repeats
- Spread throughout genome
- Chance of mutation correlates with length
- Seen in both introns > exons
- Can disrupt gene expression
  - Missense (exons)
  - Splicing (introns)
  - Promoters
- MSI leads to different mutational patterns in different tissues (same genetic abnormality)

**Bethesda Criteria for Microsatellite Instability**

**Methods:**

1. Test first five loci:
   - BAT25, BAT26, D2S123, D5S346 and D17S250

2. If only one locus positive, test next five loci
   - BAT40, TGFBRII, D17S787, D18S58 and D18S69

**Interpretation:**

1. 0/5 loci with MSI = microsatellite stable (MSS)
2. >2/5 loci with MSI = high frequency microsatellite instability (MSI-H)
3. 1-3/10 loci with MSI = low frequency microsatellite instability (MSI-L)
Analysis of DNA Mismatch Repair and MSI from Tumors

Immunohistochemistry

H&E x 1

Review and mark slide for dissection

Unstained x 10

Overlay on H&E and scrape tissue from unstained

Extract DNA for MSI testing
Microsatellite Instability

D2S123  D5S318  D5S346  D17S787  TP53  D18S58  D18S69  BAT26  BAT40  BATRII

N   T  N   T  N   T  N   T  N   T  N   T  N   T  N   T  N   T  N   T

Microsatellite Instability

Normal Tissue

Tumor

BAT26 locus

Normal Tissue

Tumor

BAT26 locus
Sebaceous Adenoma: MSH2 loss
Mismatch Repair (MMR) IHC

A

B

MSH2

C

MLH1

D
**Microsatellite Instability**

- PCR and other sequencing based assays are the gold standard
- IHC is > 90% sensitive and ~100% specific
- **MLH1**
  - Sporadic/somatic loss is most common (colon, etc.)
  - Germline loss is relatively infrequent
- **MSH2**
  - Sporadic/somatic loss very, very rare (in most tumors)
  - Germline disruption and loss much more common
  - IHC loss implies inherited/germline alteration (colon)
Muir-Torre Syndrome

• At least 3 of 4 MTS patients show microsatellite instability (MSI)

• Lynch (Hereditary Non-polyposis Colorectal Carcinoma; HNPCC) Syndrome subset

• Majority (> 90 %) of MSI results from loss of MMR proteins MLH1 or MSH2

• MTS: loss of MSH2>>>MLH1

• HNPCC: loss of MSH2=MLH1

• Sporadic colon/endometrial carcinoma: MLH1 (somatic bi-allelic methylation suppression)
Site and Tumor Type Predicts DNAMismatch Repair Status in Cutaneous Sebaceous Neoplasia

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A. Hafeez Diwan, MD, PhD,* Carla L. Warneke, MS,§ Phillip H. McKee, MD, FRCPath,¶
Dina Lev, MD,‖ Stephen Lyle, MD, PhD,¶‖ Eduardo Calonje, MD, Dip RCPATH,‖
and Alexander J. F. Lazar, MD, PhD*
- 94 sebaceous neoplasms
  - n=59, benign
  - n=35, malignant
  - 82% H&N
  - 18% non-H&N

- MMRD associated with:
  - Diagnosis (p=0.0032)
    - Adenoma (45%)
    - Carcinoma (15%)
  - Site (p<0.0001)
    - H&N (27%)
    - Non-H&N (92%)

Cystic Sebaceous Neoplasia

- 11 of 94 cystic
  - 5 benign
  - 6 malignant
- 3 of 11 MMRD (all carcinomas)
- No significant assoc. with MMRD
“Keratoacanthoma”-like architecture

- 10 of 94 KA-like – all benign
- 8 of 10 MMRD (p=0.0321)
• 90 patients with at least one sebaceous tumor (2005-2010)
  – Adenoma, epithelioma, carcinoma, BCC and SCC with seb
  – N=13 (14%) with genetically confirmed MTS

• IHC (MLH1, MSH2, MSH6, PMS2) – 51 patients abnormal

• Sensitivity = 85%
• Specificity = 48%
• PPV = 22%
• NPV = 95%
A clinical scoring system to identify patients with sebaceous neoplasms at risk for the Muir–Torre variant of Lynch syndrome

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Table 2 The Mayo Muir–Torre syndrome risk score algorithm

<table>
<thead>
<tr>
<th>Variable</th>
<th>Score</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age at sebaceous neoplasm(^a) diagnosis (years)</td>
<td></td>
</tr>
<tr>
<td>60 or older</td>
<td>0</td>
</tr>
<tr>
<td>Younger than 60</td>
<td>1</td>
</tr>
<tr>
<td>Total number of sebaceous neoplasms</td>
<td></td>
</tr>
<tr>
<td>1</td>
<td>0</td>
</tr>
<tr>
<td>2 or more</td>
<td>2</td>
</tr>
<tr>
<td>Personal history of any Lynch-related cancer</td>
<td></td>
</tr>
<tr>
<td>No</td>
<td>0</td>
</tr>
<tr>
<td>Yes</td>
<td>1</td>
</tr>
<tr>
<td>Family history of any Lynch-related cancer</td>
<td></td>
</tr>
<tr>
<td>No</td>
<td>0</td>
</tr>
<tr>
<td>Yes</td>
<td>1</td>
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Scores for the four variables are summed to create a total score, the “Mayo MTS risk score,” with a possible range of 0–5. A score of 2 or more has a sensitivity of 100% and specificity of 81% for predicting a germline mutation in a Lynch syndrome mismatch repair gene (Tables 3 and 4).
To determine the origin of the MMR defect in these tumours (somatic versus germline), we assessed the composition of both germline and somatic mutations. Two of the three MSI-high tumours harboured non-synonymous somatic mutations in MMR genes – a somatic mutation in *MLH1* (c.677G>A p.R226Q) in one and a somatic mutation in *MSH2* (c.1885C>T p.Q629*) in the other. Each of these is considered class 5/pathogenic
Take Home Messages

• Sebaceous neoplasia & MTS
  – Benign
  – Malignant

• MTS = Lynch (HNPCC) Syndrome

• Sporadic/somatic MMR loss in sebaceous tumors is very common

• Tumor site and type are important

• IHC and MSI testing can be helpful in context
  – Screening
  – Confirmation

• Germline testing (*MSH2*, *MLH1*)

• Clinical history & Genetic counseling is critical
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