Handling of Upper GI Cancer Specimens

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Pathology of oesophageal cancer

- Epidemiology
- Classification
- Role of Barrett’s oesophagus
- Handling of specimens
  - Resections
  - EMRs
- Newer techniques
Figure 1. Incidence of oesophagus cancer in the male population of the world
Oesophageal Cancer: epidemiology

- Majority are SCC
- Adenocarcinoma on the increase associated with Barrett’s oesophagus
- Variants
- Prognosis related to stage (TNM)
Oesophageal Cancer (SCC)
- Alcohol: spirits, maize-based beer, mate
- Tobacco: smoke or chew
- Diet: Iran – bread and tea, lack of vitamins
- Chemoprevention (Linxian): no benefit
- Infections: fungal, HPV
- Exogenous factors: lye, radiation
- Genetic factors: tylosis (17q25), ALDH2 polymorphism, MTHF reductase
- Associations: achalasia, Plummer-Vinson syndrome, coeliac disease
Precancerous lesions

Tylosis
Oesophageal Cancer: clinical

- Early cancers asymptomatic
- Dysphagia from invasion into wall with narrowing of lumen
- Pain from involvement of adjacent structures
Squamous Cell Carcinoma
Verrucous Carcinoma

- Well differentiated
- Slowly growing
- Rarely metastatic
- Extensive local spread
Superficial Spreading Squamous Carcinoma

- In situ or invasive carcinoma confined to mucosa and submucosa
- > 20mm
- High frequency of lymph node involvement
- Poor prognosis

Soga J et al 1982 Cancer 50: 1641–1645
Basaloid Squamous Carcinoma

- Elderly males
- Solid, discrete nests
- Small, mitotically active cells
- Microcystic spaces or necrosis
- Stromal hyalinisation
  - May be confused with adenoid cystic carcinoma
- Poor prognosis

Spindle Cell Carcinoma

- Polypoid carcinoma, carcinosarcoma, pseudosarcoma
- Biphasic pattern
  - Mesenchymal metaplasia
- Present early
- Better prognosis
Small Cell Carcinoma

- Uncommon
- Identical to lung counterpart
  - TTF-1 positive
- Rarely associated with ectopic hormone syndrome
- May be combined with squamous carcinoma or adenocarcinoma
Adenocarcinoma

- Increasing in frequency
- Males > females
- Smoking, alcohol, obesity
- Barrett’s oesophagus
- p53, c-erbB-2, COX-2 upregulation
- Heterotopic gastric mucosa - inlet patch
Adenocarcinoma arising in CLO
Adenocarcinoma

- Intestinal type
- Tubular or papillary
CK7 and CK20

- Intestinal metaplasia
  - Barrett’s CK7++/CK20+sup
  - Gastric CK7-/CK20++

- Adenocarcinoma
  - Oesophageal CK7+/CK20- 90%
  - Gastric CK7+/CK20- 20%
<table>
<thead>
<tr>
<th>TX</th>
<th>Primary tumor cannot be assessed</th>
</tr>
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<tbody>
<tr>
<td>T0</td>
<td>No evidence of primary tumor</td>
</tr>
<tr>
<td>Tis</td>
<td>High-grade dysplasia</td>
</tr>
<tr>
<td>T1</td>
<td>Tumor invades lamina propria, muscularis mucosae, or submucosa</td>
</tr>
<tr>
<td>T1a</td>
<td>Tumor invades lamina propria or muscularis mucosae</td>
</tr>
<tr>
<td>T1b</td>
<td>Tumor invades submucosa</td>
</tr>
<tr>
<td>T2</td>
<td>Tumor invades muscularis propria</td>
</tr>
<tr>
<td>T3</td>
<td>Tumor invades adventitia</td>
</tr>
<tr>
<td>T4</td>
<td>Tumor invades adjacent structures</td>
</tr>
<tr>
<td>T4a</td>
<td>Resectable tumor invading pleura, pericardium, or diaphragm</td>
</tr>
<tr>
<td>T4b</td>
<td>Unresectable tumor invading other adjacent structures, such as the aorta, vertebral body, and trachea</td>
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</table>

<table>
<thead>
<tr>
<th>GX</th>
<th>Grade cannot be assessed—stage grouping as G1</th>
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<tbody>
<tr>
<td>G1</td>
<td>Well differentiated</td>
</tr>
<tr>
<td>G2</td>
<td>Moderately differentiated</td>
</tr>
<tr>
<td>G3</td>
<td>Poorly differentiated</td>
</tr>
<tr>
<td>G4</td>
<td>Undifferentiated—stage grouping as G3 squamous</td>
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</table>
Oesophageal Cancer TNM

- **NX**: Regional lymph node(s) cannot be assessed
- **N0**: No regional lymph node metastasis
- **N1**: Metastasis in 1-2 regional lymph nodes
- **N2**: Metastasis in 3-6 regional lymph nodes
- **N3**: Metastasis in 7 or more regional lymph nodes
Type I
1-5cm above GOJ
Low oesophageal carcinoma
Type II

1cm above to 2cm below GOJ

True cardia carcinoma
Type III
2-5cm below the endoscopic cardia
Sub-cardiac carcinoma
### Siewert’s Classification

<table>
<thead>
<tr>
<th>Type</th>
<th>Description</th>
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</thead>
<tbody>
<tr>
<td>Type I</td>
<td>Adenocarcinomas of the distal third of the oesophagus (1-5 cm above cardia)</td>
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<tr>
<td></td>
<td>Low oesophageal carcinoma</td>
</tr>
<tr>
<td>Type II</td>
<td>Adenocarcinomas straddling the gastro-oesophageal junction (1 cm above to 2 cm below cardia)</td>
</tr>
<tr>
<td></td>
<td>True cardiac tumour</td>
</tr>
<tr>
<td>Type III</td>
<td>Subcardial gastric adenocarcinomas that grow proximally to involve the GOJ (2-5 cm below cardia)</td>
</tr>
<tr>
<td></td>
<td>Subcardiac carcinoma</td>
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</table>
Siewert’s Classification

Type I  adenocarcinomas of the distal third of the oesophagus (1-5 cm above GOJ)  
        staged by oesophageal rules

Type II adenocarcinomas straddling the gastro-oesophageal junction (1 cm above to 2 cm below cardia) 
        staged by oesophageal rules

Type III subcardial gastric adenocarcinomas that grow proximally to involve the GOJ (2-5 cm below cardia) 
        staged by gastric rules

In TNM7, tumours with an ‘epicentre’ within 5 cm of the OGJ which extend into the oesophagus are classified according to the oesophageal carcinoma scheme
Gastric Cancer TNM

TX  Primary tumor cannot be assessed
T0  No evidence of primary tumor
Tis Carcinoma in situ: intraepithelial tumor without invasion of the lamina propria
T1  Tumor invades lamina propria, muscularis mucosae, or submucosa
T1a Tumor invades lamina propria or muscularis mucosae
T1b Tumor invades submucosa
T2  Tumor invades muscularis propria
T3  Tumor penetrates subserosal connective tissue without invasion of visceral peritoneum or adjacent structures
T4  Tumor invades serosa (visceral peritoneum) or adjacent structures
T4a Tumor invades serosa (visceral peritoneum)
T4b Tumor invades adjacent structures
Gastric Cancer TNM

<table>
<thead>
<tr>
<th>N</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>NX</td>
<td>Regional lymph node(s) cannot be assessed</td>
</tr>
<tr>
<td>N0</td>
<td>No regional lymph node metastasis</td>
</tr>
<tr>
<td>N1</td>
<td>Metastasis in 1-2 regional lymph nodes</td>
</tr>
<tr>
<td>N2</td>
<td>Metastasis in 3-6 regional lymph nodes</td>
</tr>
<tr>
<td>N3</td>
<td>Metastasis in seven or more regional lymph nodes</td>
</tr>
<tr>
<td>N3a</td>
<td>Metastasis in 7-15 regional lymph nodes</td>
</tr>
<tr>
<td>N3b</td>
<td>Metastasis in 16 or more regional lymph nodes</td>
</tr>
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</table>
Gastric Cancer TNM

M0  No distant metastasis
M1  Distant metastasis
Specimen Choices

- Open and ink CRM
- Leave unopened with a foam or paper wick and ink CRM
- Slice transversely first
- Look for lymph nodes first
Effects of Chemoradiotherapy on Epithelial Tumours

- **Tumour**
  - Vacuolation and eosinophilia
  - Nuclear pyknosis, apoptosis
  - Necrosis

- **Stromal**
  - Fibrosis, hyalinisation
  - Inflammation
  - Giant cell granulomatous reaction
  - Calcification, malakoplakia
  - Residual acellular elements
    - keratin
    - mucus pools
Mucin Pools

- Better prognosis
- No effect on outcome when at resection margin

ypTNM staging

- e.g. ypT2 N0
- Categorises the extent of tumour actually present at the time of examination.
- Ignore residual acellular elements eg mucus pools, keratin
- NOT an estimate of disease prior to neoadjuvant therapy
Pathological Response

• Complete or partial
• Quantitation if partial
• Requires widespread sampling
  – Serial slices of site of the previous tumour
Partial Response

- Reduction in tumour volume
- Requires comparison with untreated tumour
  - Pre-treatment biopsy
  - ? representative of the whole lesion
- Microscopic foci only
- Relative proportions of tumour and fibrosis
Mandard Classification

Relative Proportion of Tumour: Fibrosis

**TRG1**  No residual cancer

**TRG2**  Rare residual cancer cells

**TRG3**  Fibrosis ‘outgrowing’ cancer

**TRG4**  Cancer ‘outgrowing’ fibrosis

**TRG5**  Absence of regression

Significant correlation with disease free survival for TRG1-3 vs TRG4-5

Mandard AM *et al* 1994 *Cancer* 73:2680-6
TRG3

TRG4
Pathological Response

• Mandard Classification
  – ? Too complex
  – ? Reproducible

• Simpler System
  – No residual tumour
  – Minimal residual disease
    • only occasional microscopic tumour foci are identified with difficulty
  – No marked regression
Complete response  
disappearance of the primary tumour in the postoperative specimen

Partial response  
microscopic evidence of residual tumour in the postoperative specimen

Stable disease  
less than 50% decrease or less than a 25% increase in tumour volume

Progressive disease  
no significant change in tumour mass or more than a 25% increase in tumour volume

Japanese Society for Esophageal Disease
More than 10 grading systems available (Mandard, Japanese, Dworak, Wheeler, Becker, Junker and Mueller, Rubbia-Brandt, Ryan, Le Sodan, Schneider, Lowy, Mansourd)

This tells us none is entirely reliable

Relies on complete embedding of abnormal area (presumed tumour bed)

‘As there is no national or international consensus, (RCPath) cannot be prescriptive and suggest that regression system to be used should be determined locally by MDT involving pathologist’

RCPath Dataset; Grabsch, Mapstone and Novelli, in preparation
Margin Involvement

- Proximal margin involvement
  - Predicts local recurrence
  - May be discontinuous with the main tissue mass
  - Always sample histologically
- Distal margin involvement
- Circumferential margin involvement
## Margin Involvement

<table>
<thead>
<tr>
<th>Number</th>
<th>median survival</th>
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<tbody>
<tr>
<td>R2</td>
<td>17</td>
</tr>
<tr>
<td>R1</td>
<td>57</td>
</tr>
<tr>
<td>R0</td>
<td>82</td>
</tr>
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</table>

p<0.0001

<table>
<thead>
<tr>
<th>Number</th>
<th>5 year survival</th>
</tr>
</thead>
<tbody>
<tr>
<td>R1/2</td>
<td>46</td>
</tr>
<tr>
<td>R0</td>
<td>137</td>
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</tbody>
</table>

0

48
Circumferential Margins

• 50 patients

• Circumferential margin involved in 20 (40%)

• Median follow up 36 months

• Local recurrence:
  – CRM positive 11/20
  – CRM negative 4/30
Circumferential Margins

- 135 patients
- Circumferential margin involved in 64 (47%)
  - (tumour within 1mm)
- Median survival:
  - CRM positive 21 months
  - CRM negative 39 months
- Effect only seen when “low metastatic burden” (<25% nodes positive)
Circumferential Margins

- 329 patients
- Circumferential margin involved in 20%
- 5 year survival:
  - CRM positive 22%
  - CRM negative 29% difference not significant
Circumferential Margins

- 249 patients
- Circumferential margin involved in 32%
- Median survival:
  - CRM positive  18 months
  - CRM negative  37 months  $p<0.0001$
- CRM status had a greater prognostic effect in T3 tumours with a low metastatic lymph node burden ($p=0.04$).

Griffiths EA et al 2006 Eur J Surg Oncol 32: 413-9
Oesophageal Cancer: clinical

- Surveillance is the application of a test that allows detection at a stage when intervention may improve outcome
- Benefit from surveillance is limited
- Endoscopy with quadrantic biopsies every 2cm + macroscopically abnormal areas
- Patches of dysplasia easily missed
- Annual progression of 0.6% to adenocarcinoma
Oesophageal Cancer: endoscopy

- White light can detect nodules, ulcers or strictures (features of early cancers)
- Often early cancers are macroscopically normal
- Chromoendoscopy: Lugol’s iodine, methylene blue
- Trimodal imaging: white-light, autofluorescence, narrow-band imaging
- Vasculature and mucosal pit patterns
Lugol’s iodine
Oesophageal Cancer: endoscopy
• magnification and high resolution endoscopy
• chromo-endoscopy
• auto-fluorescence endoscopy
• narrow band imaging
• microscopic tools: confocal microscopy; multiphoton microscopy
• in situ molecular analysis: FISH
• spectroscopic analysis: fluorescence, light scattering, optical coherence, Raman (inelastic) spectroscopy
Oesophageal Cancer: endoscopy

- Confocal fluorescence microscopy: high negative predictive value but poor sensitivity
- Elastic scattering spectroscopy: changes in subcellular components during malignant transformation; high sensitivity but poor specificity
- Optical coherence tomography: similar to ultrasound (3mm depth): *in vivo* studies not convincing
Standard endoscopic view of CLO with HGD: no lesion identified
Auto-fluorescence image: purple is abnormal
Narrow band image of same abnormal area
Chromo-endoscopy with indigo carmine dye-spray: HGD on histology
Low grade dysplasia in EMR/ER – makes histological assessment easier
69F. Long history of achalasia. Nodule close to OGJ. Multiple biopsies at three endoscopic sittings had provided equivocal results. EMR.
Oesophageal EMR

- Olympus/Keymed – inject submucosa and form pseudopolyp, direct snare
- Cook Duette band EMR/ER – band ligation and snare
- methodology and subsequent therapy likely influences how important margins are
Oesophageal EMR

Practicalities

• size and number depends on preferred technique of endoscopist
• specimen preparation – band still attached!
• orientation
• adequate fixation
• specimen dissection depends on size
  we are more interested in the deep margin than peripheral margins
• close collaboration with endoscopist and assistant to identify important landmarks
• resect *en bloc* if possible
• keep specimen(s) intact
• pin out on cork
• mark margins
• embed whole specimen(s) for histology
Oesophageal EMRs
Histological assessment of EMR

The rationale for EMR/ER in CLO

- Intramucosal pathology
- Submucosal involvement by carcinoma
- Endoscopic treatment and/or surveillance
- Referral for surgery
The rationale for EMR/ER in CLO: risk of lymph node metastatic disease

<table>
<thead>
<tr>
<th></th>
<th>intramucosal disease</th>
<th>submucosal disease</th>
</tr>
</thead>
<tbody>
<tr>
<td>adenocarcinoma</td>
<td>2.0%</td>
<td>24.6%</td>
</tr>
<tr>
<td>squamous cell carcinoma</td>
<td>3.6%</td>
<td>26%</td>
</tr>
</tbody>
</table>

For mucosal disease, the surgical mortality outweighs the risk of metastasis

*Bergman, 2007*
What are the diagnostic pathological issues in oesophageal EMR?
HGD versus intramucosal carcinoma
Entrapped and submucosal glands mimicking submucosal adenocarcinoma
Reporting oesophageal EMRs

• is it Barrett’s?
• are there treatment effects?

• neoplasia diagnosis
• depth of spread
• lymphovascular spread
• peripheral margins status
• deep margin status
Oesophageal EMR/ER – diagnosis and ‘complete excision’

<table>
<thead>
<tr>
<th>Diagnosis</th>
<th>Count</th>
</tr>
</thead>
<tbody>
<tr>
<td>CLO only</td>
<td>21</td>
</tr>
<tr>
<td>Squamous only</td>
<td>9</td>
</tr>
<tr>
<td>Gastric mucosa only</td>
<td>7</td>
</tr>
<tr>
<td>LGD ‘excised’</td>
<td>5</td>
</tr>
<tr>
<td>LGD at margins</td>
<td>9</td>
</tr>
<tr>
<td>HGD ‘excised’</td>
<td>9</td>
</tr>
<tr>
<td>HGD at margins</td>
<td>36</td>
</tr>
<tr>
<td>IMC ‘excised’</td>
<td>4</td>
</tr>
<tr>
<td>IMC at margins</td>
<td>12</td>
</tr>
<tr>
<td>Ca into SM ‘excised’</td>
<td>8</td>
</tr>
<tr>
<td>Ca into SM at margins</td>
<td>17</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td><strong>137</strong></td>
</tr>
</tbody>
</table>
Low grade dysplasia at margins
Oesophageal EMR/ER in Gloucestershire

- number of ‘normals’ reflects endoscopic difficulties – benign nodules in CLO, hiatus hernia, etc

- LGD and HGD often at margins – reflects endoscopic difficulties – matters less because of subsequent ablative therapy

- IMC often at margins – don’t know the implications of this but one suspects that this, too, will be successfully ablated

- submucosal adenocarcinoma (ironically) more often clear of margins but doesn’t matter much as this is an indication for radical surgery

Shepherd and Barr, Gloucester
CLO and the double muscularis mucosae and entrapment of dysplastic epithelium

Oesophageal EMR

Pathology is important, mainly to confirm or refute:

- The presence of malignancy
- If present, depth of malignancy

Margins matter less (but this does depend on subsequent management strategy)
THANK YOU FOR YOUR ATTENTION
ANY QUESTIONS?