Fasciola hepatica

Drug resistant in livestock
and now in humans

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London

- Imported parasitic and infectious diseases
- From any part of the globe
- Tourists, refugees, migrants, business people etc
- 8,600,000 people in London
  - so a window on the world
- Important in sentinel surveillance
Fasciola hepatica – Life cycle
Image from Carlo Denegri Foundation
Global Distribution of Human Fascioliasis

Global Burden of Human Fascioliasis


- 91.1 million people at risk
- Infects 2.5 to 17 million people annually
- Sheep and cattle-rearing communities
  - Up to 21% of children in some parts of Bolivia
  - Rarely greater than 5% in other endemic areas
    - Ecuador
    - Egypt
    - Iran
    - Peru
    - Vietnam
Challenges from *Fasciola hepatica*

- Increase in livestock infection
- Triclabendazole treatment failure
Cumbrian Sheep
Fasciola hepatica in Dairy Herds in England and Wales
Fasciola hepatica in Dairy Herds in Germany
Kuerpick et al (2013) Parasitology 140, 1051-1060
**Fasciola hepatica in Sheep**

Taylor MA (2012) *Vet Parasitol* 189, 2-7

- Can infect all grazing animals
- Mainly sheep and cattle
- Most pathogenic in sheep
- More prevalent with high summer rainfall
- If milder winter, acute fluke disease appears much earlier in the year
Fasciola and UK Climate Change

- Increased rainfall in Autumn and Winter
- Warmer average temperatures throughout the year
- 4 week extension of herbage growing season over past 40 years
# Treatment of *Fasciola hepatica* infection in sheep

http://www.scops.org.uk/content/Liver-Fluke-Fact-Sheet.pdf

<table>
<thead>
<tr>
<th>Flukicide</th>
<th>Age of fluke (weeks)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>1  2  3  4  5  6  7  8  9  10  11  12  13  14</td>
</tr>
<tr>
<td>Albendazole</td>
<td></td>
</tr>
<tr>
<td></td>
<td>50 - 70%</td>
</tr>
<tr>
<td></td>
<td>80 - 99%</td>
</tr>
<tr>
<td>Oxyclozanide</td>
<td></td>
</tr>
<tr>
<td>Nitroxylin</td>
<td></td>
</tr>
<tr>
<td></td>
<td>50 - 90%</td>
</tr>
<tr>
<td></td>
<td>91 - 99%</td>
</tr>
<tr>
<td>Closantel</td>
<td></td>
</tr>
<tr>
<td>Triclabendazole (TCB)</td>
<td>90 - 99%</td>
</tr>
<tr>
<td></td>
<td>99 - 99.9%</td>
</tr>
</tbody>
</table>
Triclabendazole-resistant *Fasciola* in UK Sheep (1)


- Two female sheep
- Live, undamaged flukes at necropsy 7 days after a 3\(^{rd}\) dose of triclabendazole
Triclabendazole-resistant *Fasciola* in UK Sheep (2)

Gordon *et al* (2014) *Veterinary Record* August 11, 159-160

- Two naturally infected ewes with live *Fasciola hepatica* despite 3 Rx with triclabendazole

- Eggs used to infect *Galba truncatula*

- Metacercariae used to infect 6 sheep
Triclabendazole-resistant *Fasciola* in UK Sheep (3)
Gordon *et al* (2014) *Veterinary Record* August 11, 159-160

- Treated with triclabendazole at 12 weeks when infection patent; second dose repeated 3 weeks later

- Post-mortem 14 days after second dose showed between 19 and 70 live adult fluke in each animal
Risk factors for human infection with *Fasciola hepatica*


- Consuming raw vegetables
  - Watercress
  - Radish
  - Washing terrestrial vegetables with untreated water
  - Plant cultures irrigated with natural water
- Chewing Khat
- Eating raw liver
Clinical features in humans: Invasive phase

- Fever
- Abdominal pain
  - Right upper quadrant
  - Epigastric
- Anorexia
- Flatulence
- Nausea
- Diarrhoea
- Cough
- Dyspnoea
- Haemoptysis
- Chest pain
- Urticaria
Clinical features in humans: Chronic phase

- May be none
- Abdominal pain
- Fever
- Fatty food intolerance
- Nausea
- Jaundice
- Pruritis
- Hepatomegaly
- Eosinophilia
Complications in humans

Cholecystitis
Cholangitis
Biliary colic
Biliary obstruction
Liver abscess
Ectopic flukes
Differential Diagnosis in Humans

- Peptic ulceration
- Gall bladder disease
- Suspected malignancy
- Hydatid disease (serological cross-reaction)
Diagnosis – Imaging appearances

Lim JH et al (2007) AJR; 188: 1596-1603

- Clusters of tract-like microabscesses
  - usually subcapsular
- Large cyst-like necrotic lesions
- Non-specific biliary dilatation
  - single or multiple filling defects
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Diagnosis

- Microscopy of faeces or duodenal juice for ova
- Antibody detection
Fasciola hepatica ovum
Online Lecture Library

Slide withheld at request of author
Antibody detection

- IFAT
- ELISA
- Serology is approx. 90% sensitive
- May be positive when ova not detected
  - early cases
  - low worm burden
Fasciola hepatica Removed at ERCP
Online Lecture Library

Slide withheld at request of author
Online Lecture Library

Slide withheld at request of author
Treatment Failure in Humans (2)

- “Lover of wild watercress”
- *Fasciola* eggs in stool
- *Fasciola* IFAT 1 in 1024 (<1 in 28)
- Two doses of triclabendazole given
Treatment Failure in Humans (3)

• Follow-up one year later
  • Much better
  • Eosinophil count 0.34 (normal)
  • Alkaline phosphatase 103 (normal)
  • Fasciola IFAT 1 in 128 (much improved)
  • **BUT** Fasciola eggs in stool
Geographical distribution of triclabendazole-resistant fascioliasis in livestock

- Australia
- Europe:
  - Ireland; the Netherlands; Scotland; Spain
- South America
  - Argentina; Bolivia; Chile; Peru
Reports of triclabendazole-resistant fascioliasis in humans


- Europe
  - The Netherlands
    - TCBZ; nitazoxanide; TCBZ + ketoconazole

- South America
  - Argentina; Bolivia; Chile
  - Peru
    - 7 cases; TCBZ 3 or 4 courses; nitazoxanide
Proposed mechanisms for triclabendazole resistance

- P-glycoprotein linked efflux pump

- Increased metabolism of triclabendazole sulfoxide to sulfone
Triclabendazole vs Albendazole

- Triclabendazole (TCBZ) acts against mature and immature *F. hepatica*
- Albendazole (ABZ) is active against flukes older than 12 weeks
- Some isolates resistant to TCBZ but sensitive to ABZ
- Some isolates resistant to ABZ but sensitive to TCBZ (ABZ drug pressure in the field)
Bithionol

- No longer obtainable in England for human use
- Dosage regimen
  - 30 to 50 mg/kg po alt die in 2 divided doses to a total of 10 to 15 doses
RCT of Artesunate vs Triclabendazole

- Triclabendazole 10 mg/kg
  2 doses, 12 hours apart
- Artesunate 4 mg/kg/d daily for 10 days
## Artesunate vs Triclabendazole


<table>
<thead>
<tr>
<th>Endpoint at 3m, ITT</th>
<th>Triclabendazole</th>
<th>Artesunate</th>
<th>$P$ value</th>
</tr>
</thead>
<tbody>
<tr>
<td>No symptoms</td>
<td>46/50 (92%)</td>
<td>38/50 (76%)</td>
<td>0.05</td>
</tr>
<tr>
<td>Eosinophils &lt;400/µL</td>
<td>21/50 (42%)</td>
<td>8/50 (16%)</td>
<td>0.008</td>
</tr>
<tr>
<td>Improved U/S scan</td>
<td>35/50 (70%)</td>
<td>33/50 (66%)</td>
<td>0.83</td>
</tr>
<tr>
<td>Complete response</td>
<td>18/50 (36%)</td>
<td>5/50 (10%)</td>
<td>0.004</td>
</tr>
</tbody>
</table>
Nitazoxanide vs Placebo

- 50 Adults (40 active Rx)
  Nitazoxanide 500 mg bd for 7 days

- 50 Children (40 active Rx)
  Age 2-3 y 100 mg bd for 7 days
  Age 4-11 y 200 mg bd for 7 days
  Age >12 y 500 mg bd for 7 days
Nitazoxanide vs Placebo

Cure based on egg counts

<table>
<thead>
<tr>
<th>Age group</th>
<th>Nitazoxanide</th>
<th>Placebo</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Adults</td>
<td>18/30 (60%)</td>
<td>1/8 (12.5)</td>
<td>0.042</td>
</tr>
<tr>
<td>Children</td>
<td>14/35 (40%)</td>
<td>0/8</td>
<td>0.038</td>
</tr>
</tbody>
</table>
Experimental Chemotherapy for Fascioliasis 1

- Triclabendazole plus artesunate or artemether
  

- *Fasciola hepatica in vitro* and *in vivo* rat model

- Single agent triclabendazole superior

- Synergistic or antagonistic effects (dose-dependent)
Experimental Chemotherapy for Fascioliasis 2

- Triclabendazole plus ivermectin
  
  - Ivermectin-induced modulation of P-glycoprotein activity decreased triclabendazole efflux from resistant flukes
    
  
  - Clinical study versus triclabendazole-resistant Fasciola in sheep
    
    TCBZ vs TCBZ plus ivermectin plus methimazole
    
    Combination did not reverse triclabendazole resistance
    
    Ceballos et al (2010) BMC Veterinary Research 6:8
Experimental Chemotherapy for Fascioliasis 3

- Triclabendazole plus verapamil
  - In vitro, TCBZ-resistant flukes; combination showed:
    - Greater tegumental disruption
    - Greater damage to mitochondria and granular endoplasmic reticulum


- Greater disruption of vitellogenesis and spermatogenesis

Savage et al (2014) *Parasitology* 141, 1064-1079
Experimental Chemotherapy for Fascioliasis 4

- Triclabendazole plus ketoconazole
  - TCBZ-resistant *F. hepatica* in a laboratory rat model
  - Drug combination showed:
    - Increased swelling of basal infolds and mucopolysaccharide masses
    - Reduced size and number of Golgi complexes
    - Sub-tegumental flooding
    - Sloughing of tegumental covering of spines

Global economic losses in agriculture due to *Fasciola hepatica* 

**US $3 billion per annum**


Therefore, new veterinary drugs will appear

- Which may or may not be suitable for use in humans