ESBL-producing *E. coli*, what’s the threat from your pet?

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Pets as a source of AMR?

Top Ten Pets for 2013

1. **Fish kept in tanks**: 20 - 25 million (9% of households)
2. **Fish kept in ponds**: 20 - 25 million (6% of households)
3. **Dogs**: 8.5 million (25% of households)
4. **Cats**: 8.5 million (19% of households)
5. **Rabbits**: 1 million (3% of households)
6. **Caged birds**: 1 million (1.9% of households)
7. **Domestic fowl**: 1 million (0.9% of households)
8. **Guinea Pigs**: Half a million (0.6% of households)
9. **Hamsters**: Half a million (1.5% of households)
10. **Horses and ponies**: 400,000 (1.1% of households)

**Plus**

Reptiles (lizards and snakes): 700,000 (1.5% of households)

http://www.pfma.org.uk/pet-population/
Antimicrobial use in dogs and horses

Total tonnage of antimicrobials used for therapeutic purposes in animals 2006-2011 (VMD)

- 25.9% (n=1179/4559) dogs received antimicrobials
- Penicillins (amoxicillin/clavulanic acid) 48.7%
- Fluoroquinolones 5.6%
- 3rd/4th generation cephalosporins 1.3%
- Only 3.5% of practices had a written antimicrobial use policy document

- 17.0% (216/1273) of all horses attended received antimicrobials
- Trimethoprim sulphonamides 34.4 %
- Penicillins 23.0%
- Fluoroquinolones 5.1%
- 3rd/4th generation cephalosporins 3.1%
- <1% of practices had a written antimicrobial use policy document
Antimicrobial Stewardship

Are you PROTECTing your antibacterials?

**Skin Infections**
- Bacterial: amoxicillin trihydrate, clindamycin, fortified, or fortified + clindamycin + gentamicin. Parenteral therapy is recommended for severe cases.
- Practice Policy: For moderate to severe skin and soft tissue infections, consider parenteral therapy.

**Gastrointestinal Infections**
- Escherichia coli, Klebsiella pneumoniae, and Enterobacter species: amoxicillin trihydrate, clindamycin, fortified, or fortified + clindamycin + gentamicin. Parenteral therapy is recommended for severe cases.
- Practice Policy: For moderate to severe gastrointestinal infections, consider parenteral therapy.

**Ear Infections**
- Bacterial: amoxicillin trihydrate, clindamycin, fortified, or fortified + clindamycin + gentamicin. Parenteral therapy is recommended for severe cases.
- Practice Policy: For moderate to severe ear infections, consider parenteral therapy.

**Antibacterials not indicated unless cytology and/or culture is positive**
- Cerebrospinal:
  - Chronic meningitis: amoxicillin trihydrate, clindamycin, fortified, or fortified + clindamycin + gentamicin.
- Practice Policy: For moderate to severe ear infections, consider parenteral therapy.

**Second and Third Choice Antibacterials**
- These include amoxicillin, clavulanate, and 4th generation cephalosporins.
- Practice Policy: For moderate to severe ear infections, consider parenteral therapy.

**Follow the Cascade**
- Suggested antibacterials for dogs and cats are listed in alphabetical order.
- For further information on individual drugs and dosages, see BSAVA Small Animal Formulary, 7th Edition.
Antimicrobial Stewardship

### Protect ME - Responsible antimicrobial use policy

<table>
<thead>
<tr>
<th>Condition</th>
<th>FIRST LINE</th>
<th>ALTERNATIVES</th>
<th>Notes</th>
</tr>
</thead>
<tbody>
<tr>
<td>Strongly recommended for treatment</td>
<td>Piperacillin</td>
<td>Gentamicin</td>
<td></td>
</tr>
<tr>
<td>Moderate</td>
<td>Piperacillin</td>
<td>Gentamicin</td>
<td></td>
</tr>
<tr>
<td>Not recommended</td>
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**Protect ME**

**Practice name**

**Practice Policy:**

**Dose and routes of administration of common antimicrobials**

<table>
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<tr>
<th>Drug</th>
<th>Dose Per kg</th>
<th>Route</th>
<th>Dosing interval</th>
<th>Spectrum</th>
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<td>PO</td>
<td>12 hours*</td>
<td>+ + + +</td>
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**Notes:**

- **+** Effective against most important pathogens, including staphylococcus for Gram positive and pseudomonas for Gram negative bacteria.
- **+** Effective against many important bacteria.
- **+** Some effect, but many clinically significant bacteria may not be susceptible.
- **+** Indicated a drug, dose, route or dosing frequency that is not listed in the marketing authorization for that product.

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Extended-spectrum β-lactamases

TEM, SHV and CTX-M

- Sensitive to the cephemycins and clavulanic acid
- TEM, SHV
  - Extending spectrum mutations
  - Preferentially hydrolyze ceftazidime
- CTX-M
  - Preferentially hydrolyze cefotaxime and ceftriaxone
  - Predominant ESBL in *E. coli* in UK human hospitals
  - Most common variant CTX-M-15, associated with the pandemic *E. coli* clone ST-131
  - Worldwide distribution
Chromosomosomal β-lactamases of Gram-negatives

- Not inhibited by clavulanic acid.
- Many now plasmid-encoded eg CMY-2
- Can be resistant to 2nd and 3rd generation cephalosporins, cephemycins
- Not resistant to 4th gen cephalosporins
AMR and ESBL-producing *E. coli* in UK horses

- 72.2% (452/650) horses had at least one antimicrobial resistant *E. coli*

Equine ESBL/AmpC positive isolates (n=31)

- $\text{bla}_{\text{CTX-M-1}}$ (n=18)
- $\text{bla}_{\text{CTX-M-14}}$ (n=5)
- $\text{bla}_{\text{CTX-M-9}}$ (n=1)
- $\text{bla}_{\text{CTX-M-20}}$ (n=1)
- $\text{bla}_{\text{CTX-M-65}}$ (n=1)
- $\text{bla}_{\text{CMY}}$ (n=5)

- No $\text{bla}_{\text{CTX-M-15}}$

- Virulence gene microarray
- Low number of virulence genes detected

Spatial variation in equine AMR prevalence

Estimated type-specific probabilities across the mainland United Kingdom (excluding Scotland) for the presence of antimicrobial resistance in a horse’s faecal sample: (a) resistance to any antimicrobial; (b) multidrug resistance

Summary of risk factors for AMR, MDR and ESBL-producing E. coli in horses

- **Being hospitalised within the last 10 days**
  - ampicillin (OR 12.2, P<0.001), gentamicin (OR 28.5, P<0.001), fluoroquinolone (OR 5.2, P=0.02) resistance
  - multidrug (OR 15.5, P=0.02)
  - ESBL-mediated resistance (OR 16.0, P<0.001)

- **Stabled with a horse hospitalised** within the last month
  - ESBL resistance (OR 3.90, P=0.04)

- **Antimicrobial treatment in the previous 10 days**
  - ciprofloxacin resistance (enrofloxacin OR 26.3, P<0.001)

- **Stabled on a farm**
  - ampicillin (OR 5.3, P=0.03), trimethoprim (OR 5.6, P=0.04) and multidrug resistance (OR 5.1, P=0.04)

- **Stabled on racing yard**
  - ESBL-mediated resistance (OR 7.2, P=0.04)

Study Aim

“To determine prevalence and characteristics of antimicrobial resistant E. coli in the faeces of horses in an equine hospital and identify risk factors associated with the presence of resistance”

- Prospective cohort study Phillip Leverhulme Equine Hospital
- 103 horses admitted to the PLEH for a variety of reasons
- Faecal samples obtained every 2 days until discharge
- Data collected via owner questionnaire, referral history, clinical records

**Defined Daily Doses (DDD) = Total amount of antimicrobial on day**

*Average dose required for 500kg horse*

Hospitalised Horses

- 457 faecal samples (694 isolates) collected from 103 horses
- Similar pattern for all antimicrobials with increasing prevalence of resistance from days 0 to 4

**Prevalence of resistance by sample day**

Proportion resistant samples (95% CI)

<table>
<thead>
<tr>
<th>Antimicrobial</th>
<th>Day 0</th>
<th>Day 2</th>
<th>Day 4</th>
<th>Day 6</th>
<th>Day 7 or later</th>
</tr>
</thead>
<tbody>
<tr>
<td>AMP</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>AMX-CL</td>
<td></td>
<td></td>
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<td></td>
<td></td>
</tr>
<tr>
<td>CIP</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>GM</td>
<td></td>
<td></td>
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<td></td>
<td></td>
</tr>
<tr>
<td>NA</td>
<td></td>
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<tr>
<td>TET</td>
<td></td>
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<tr>
<td>TMP</td>
<td></td>
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<td></td>
</tr>
<tr>
<td>MDR</td>
<td></td>
<td></td>
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<td></td>
<td></td>
</tr>
<tr>
<td>ESBL</td>
<td></td>
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</tr>
</tbody>
</table>

Increase in prevalence of ESBL-mediated resistance

- ESBL-mediated resistant *E. coli* - 28.7% of samples
- 53.4% of horses having at least one ESBL-positive *E. coli*
Risk factors for AMR *E. coli* in hospitalised horses

**Summary of Risk Factors**

- **Day of hospitalisation** for all outcomes except trimethoprim resistance
- **Antimicrobial treatment in previous 7 days** for all except ESBL-mediated resistance
- **TMPS doses prescribed in hospital** for all outcomes except tetracycline, ciprofloxacin and ESBL mediated resistance
- **Location** on M yard (mixed purpose) for ESBL-mediated resistance and E yard (orthopaedic) for trimethoprim and ciprofloxacin resistance
- **Admission for surgical gastrointestinal disease** for fluoroquinolone resistance

The Aim of this study: To determine the duration of shedding of antimicrobial resistant *E. coli* in horses in the community for a 5 month period.

Study population: Faecal samples collected from 30 horses, which included 15 horses which had recently been discharged from an equine hospital and 15 which had previously been identified as carrying antimicrobial resistant *E. coli* in our nationwide survey.

Methods: Samples were collected weekly (for four weeks), and then monthly (for four months).

Longitudinal carriage of AMR *E. coli* in horses: In total 225 samples were collected.

<table>
<thead>
<tr>
<th>Resistance outcome</th>
<th>Hospitalised horses</th>
<th>Community horses</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Any resistance</td>
<td>71.2% (63.0-79.4)</td>
<td>79.4% (71.8-87.1)</td>
<td>0.2</td>
</tr>
<tr>
<td>Multidrug resistance</td>
<td>37.3% (28.6-46.0)</td>
<td>35.5% (26.4-44.6)</td>
<td>0.8</td>
</tr>
<tr>
<td>ESBL-mediated</td>
<td>21.2% (13.8-28.6)</td>
<td>9.3% (3.8-14.9)</td>
<td>0.01</td>
</tr>
</tbody>
</table>
Longitudinal shedding of AR, MDR and ESBL-producing *E. coli* in horses

**Duration of shedding of resistant *E. coli***

<table>
<thead>
<tr>
<th>Resistance outcome</th>
<th>Median duration of carriage</th>
</tr>
</thead>
<tbody>
<tr>
<td>Any resistance</td>
<td></td>
</tr>
<tr>
<td>Multidrug resistance</td>
<td></td>
</tr>
<tr>
<td>ESBL-mediated</td>
<td></td>
</tr>
</tbody>
</table>

Shedding of AMR *E. coli* in healthy community dogs (Schmidt 2014, PhD thesis)
AMR and ESBL producing *E. coli* in UK dogs

- 44.8% (260/580) dogs had at least one antimicrobial resistant *E. coli* in faeces
- 18.1% had at least one multidrug resistant *E. coli*
- 4.1% of dogs had at least one ESBL producing *E. coli*

**Canine ESBL/AmpC positive isolates (n=58)**

- $bla_{TEM}$ (n=21)
- $bla_{cmy-2}$ (n=42)
- 10/58 carried a $bla_{CTX-M}$ gene
  - $bla_{CTX-M-1}$ (n=5)
  - $bla_{CTX-M-15}$ (n=2)
  - $bla_{CTX-M-3}$ (n=1)
  - $bla_{CTX-M-14/18}$ (n=1)
  - $bla_{CTX-M-20}$ (n=1)
- MLST
  - 19 sequence types (ST), no ST-131
- Virulence gene microarray
- Large number of virulence genes
- EPEC
AMR and ESBL producing *E. coli* in dogs

Risk factors-

- **MDR *E. coli***
  - Fed raw poultry (OR 3.93, P=0.002)
- **ESBL mediated resistance**
  - Fed raw poultry (OR 17.81, P <0.001)
- **ESBL and AmpC mediated resistance**
  - Antimicrobials in the previous 3 months OR 1.88 P=0.003 and OR 4.46, P <0.001, respectively.
4 hunts kennels were approached for inclusion in the study. Faecal samples were collected from the housing floor over the course of a single day.

**Prevalence of AMR E. coli in hunt kennel dogs**

- β-lactamase and PMQR genes
  - \( \text{bla}_{\text{TEM}}, \text{bla}_{\text{SHV}} \) (all kennels)
  - \( qnrA \) (HKC and HKD), \( qnrB \) and \( qnrS \) (HKC)
  - \( \text{bla}_{\text{CMY-2}} \) (HKB and HKC).

![Graph showing prevalence of antimicrobial resistance in E. coli across different kennels.](image)
ESBL-producing and MDR E. coli in hospitalised dogs

- Sampling of in-patients at 5 small animal referral hospitals in Northwest England
- Faecal samples collected from dogs and cats
- Environmental samples (swabs and boot socks)

- 50.1% samples (167/333) faecal samples from 214 animals had AMR E. coli
- 18.3% (47/254) environmental samples had AMR E. coli

Environmental samples
- ESBL-producing E. coli 5.4%
- AmpC 8.6%
- MDR 8.9%
- FQR 10.5%
ESBL-producing and MDR *E. coli* in hospitalised dogs

- *E. coli* with \( \text{bla}_{\text{CTX-M}} \) identified in 15.6% of faecal samples
  - 6.0% were \( \text{bla}_{\text{CTX-M-15}} \)
  - No isolates were identified as O25/ST131

- \( \text{bla}_{\text{TEM-158}} \) in *E. coli* in 3% faecal samples - IRT
- \( \text{bla}_{\text{SHV-12}} \) in *E. coli* from 0.8% of environmental samples

- The \( \text{bla}_{\text{cmY-2}} \) gene was detected in *E. coli* from 21.6% and 8.0% of faecal and environmental samples, respectively.
Risk factors for ESBL-producing, MDR and FQR *E. coli* in hospitalised dogs

- Length of hospitalisation was a significant risk factor for the isolation of FQR, ESBL and AmpC-producing *E. coli*.
- Every resistance outcome was positively associated with the use of at least one antimicrobial.
  - Fluoroquinolone use was strongly associated with FQR and isolation of ESBL-producing *E. coli*.
  - The use of potentiated amoxicillin was associated with the presence of ESBL and AmpC producing *E. coli*.
  - Cephalexin use was associated with potentiated amoxicillin resistance.
Clinical infection with ESBL-producing *E. coli* in companion animals

**Horses - PLEH, University of Liverpool**

- Very rarely associated with wound infections
- Environmental monitoring indicates circulation of ESBL-producing *Enterobacteriaceae* in the hospital environment.
- Few reports in the literature
- Ability to cause clinical disease????

**Dogs - SATH, University of Liverpool**

- Number of cases of ESBL infections increasing
- Numerous reports in the literature
- ST-131 identified in dogs
**Hazard identification:** This study focused on using microbial risk analysis approaches to investigate the potential risk that ESBL-producing *E. coli* in both dogs and horses might represent to people.

**Exposure assessment:**
- The exposure assessment step seeks to estimate the amount of the hazard to which humans are exposed via the exposure route of interest.
- Exposure pathways identify the routes by which agents might come into contact with people.
- Quantities of interest in the schematic model are the prevalence and the amount of the hazard at each key stage.
- However hazard =

![Diagram showing a plasmid and an ESBL gene.](image)
Assessing the public health risk; risk analysis

Areas where data is lacking:

- Quantity of ESBL-producing *E. coli* shed by dogs
- Survival in faeces
- Transmission by faecal/oral route
- No data on the infective dose and risk factors for human colonisation and/or transfer of resistance determinants

Areas where data is lacking apply equally to horses.
**E. coli** sharing & public health risks

- Sharing within households is common
- Sharing between human and pets may occur — both ways
- May include potentially pathogenic strains
- Correlated with number of individuals

common external reservoir e.g. food?

**3 year surveillance 1 household of 5 humans, 1 dog**

- Persistent strains (1-3 years), including ExPEC
- Human strain causing dog UTI (and vice versa)

Discussion

- Recently hospitalised horses have a significantly higher prevalence of ESBL-producing *E. coli* than horses in the community.
- Similarly, hospitalised dogs have a higher prevalence of ESBL-producing *E. coli* compared to dogs in the community.
  - High prevalence of ESBL and AmpC-producing *E. coli* in some small animal hospitals is worrying
  - Antimicrobial prescribing practices

- Risk to individuals with different levels of contact with different populations of companion animals?
- Different levels of risk for transmission of ESBL-producing *E. coli*?
  - Dog owners, kennel workers, vets and veterinary staff.
  - Horse owners, yard workers (livery, racing and riding), horse riders, vets and veterinary staff.
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Thank you for listening

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