Can IDEXX Angio Detect™ accurately detect canine angiostrongylosis?

A Knowledge Summary by

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PICO question

In dogs, is IDEXX Angio Detect™ as accurate as Baermann coprology when diagnosing Angiostrongylus vasorum infection?

Clinical bottom line

Category of research question

Diagnosis

The number and type of study designs reviewed

Eight papers were critically reviewed: three diagnostic accuracy studies, two cross-sectional studies (one of which also included a retrospective case series), one cohort study, one case-control study, and one case series

Strength of evidence

Weak

Outcomes reported

Angio Detect™ (IDEXX) was shown to have low-moderate sensitivity and high specificity in comparison to Baermann coprology. Occasionally, false-negative results occurred with Angio Detect™ when compared to Baermann coprology. This was thought to be due to antigen-antibody complex formation. Positive Angio Detect™ assays were obtained in both symptomatic and asymptomatic canine patients. In an experimental setting, Angio Detect™ was shown to obtain a positive result five weeks post-inoculation

Conclusion

There is weak evidence supporting Angio Detect™ as a highly specific and moderately sensitive diagnostic test when compared to Baermann coprology

How to apply this evidence in practice

The application of evidence into practice should take into account multiple factors, not limited to: individual clinical expertise, patient’s circumstances and owners’ values, country, location or clinic where you work, the individual case in front of you, the availability of therapies and resources.

Knowledge Summaries are a resource to help reinforce or inform decision making. They do not override the responsibility or judgement of the practitioner to do what is best for the animal in their care.

Clinical Scenario

A coughing dog with epistaxis presents to your clinic. The patient has thrombocytopenia and prolonged coagulation parameters on in-house blood analysis. The clinical presentation along with the haematological and coagulopathic deficits makes you suspect Angiostrongylus vasorum (A. vasorum) infection as a potential differential diagnosis. It is possible to post a faecal sample to an external laboratory for Baermann coprology (Conboy, 2009), but the results will take at least 2 days to obtain. The patient’s condition is deteriorating. You wonder if the rapid in-house immunochromatography assay (Angio Detect™; IDEXX) could provide you with an accurate diagnosis within a quicker time frame compared to Baermann coprology conducted at an external laboratory.
**The evidence**

Three cross-sectional diagnostic test accuracy studies were found, of which two used case-control selection (Schnyder et al., 2014; and Canonne et al., 2018). The other study used cohort selection for the majority of the study population, but also included a group of known *A. vasorum*-positive cases and a control group of animals with other confirmed nematode infections (Liu et al., 2017). These studies were considered to represent the most appropriate study designs; however, they are subject to biases and limitations that limit the strength of evidence provided.

Two cross-sectional studies included some information regarding the accuracy of Angio Detect™ (Olivieri et al., 2017; and Lempereur et al., 2020), one of which also included a retrospective case series (Olivieri et al., 2017). A small cohort study at a single kennel tested dogs with Angio Detect™ and Baermann coprology on three occasions in a 6 week period (Di Cesare et al., 2014). A case-control study evaluated Angio Detect™ and Baermann coprology in dogs presenting with clinical signs suggestive of *A. vasorum* infection and a control group of dogs presented to participating practices for routine preventive health care or unrelated conditions (Lempereur et al., 2016). One case series utilised Angio Detect™ and Baermann coprology in addition to thoracic ultrasonography and radiography in dogs presenting with respiratory distress to participating practices (Venco et al., 2021).

When not included within the publication, the sensitivity and specificity (with 95% confidence intervals [CI]) of Angio Detect™ in comparison to Baermann coprology were calculated (Sergeant, 2018), where possible, and are presented in italic font within the summary of evidence tables.

**Summary of the evidence**

<table>
<thead>
<tr>
<th>Canonne et al. (2018)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Population:</strong></td>
</tr>
<tr>
<td>· Client-owned dogs presented to the University Veterinary Small Animal Teaching Hospital of Liège, Belgium, between March 2013–April 2017 were retrospectively selected.</td>
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<tr>
<td>· Dogs were of various breeds, aged from 9 months–10 years.</td>
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<tr>
<td>· Inclusion criteria were respiratory clinical signs (cough, respiratory distress or exercise intolerance) and recorded diagnostic results for natural <em>A. vasorum</em> infection (Baermann coprology, Angio Detect™, and enzyme linked immunosorbent assays [ELISA] both for antigens and specific antibodies).</td>
</tr>
<tr>
<td>· <em>A. vasorum</em> infection was suspected based on compatible respiratory signs (as above), radiographic findings, bronchoscopy, and bronchoalveolar lavage (BAL) cytology examination.</td>
</tr>
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<td>· Owner-reported duration of respiratory clinical signs varied from 2 weeks–2 months.</td>
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<td>· Angiostrongylosis was confirmed in all seven dogs by:</td>
</tr>
<tr>
<td>o positive quantitative (q) PCR from BAL samples and clinical recovery after administration of either 50 mg/kg q24h fenbendazole PO for 3 week treatment duration or 2.5 mg/kg moxidectin repeated after 2–4 weeks.</td>
</tr>
<tr>
<td><strong>Sample size:</strong> Seven dogs (two male, five female).</td>
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<tr>
<td><strong>Intervention details:</strong></td>
</tr>
<tr>
<td>· At the time of diagnosis one or two serum samples were obtained and frozen prior to analysis.</td>
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</table>
• One sample per dog was thawed for Angio Detect™ diagnostic testing.
• The second serum sample underwent *A. vasorum* antigen and antibody sandwich-ELISAs; both analysed at the Institute of Parasitology, Vetsuisse Faculty, University of Zurich, Switzerland.
• Faecal samples were obtained from each dog over 3 consecutive days for Baermann coprology. All coprology was undertaken in the Laboratory of Parasitology of the Faculty of Veterinary Medicine, University of Liège, Belgium.

**Study design:** Cross-sectional diagnostic accuracy study.

**Outcome studied:** Objective assessment:

- Positive or negative results from Angio Detect™, *A. vasorum* antigen sandwich-ELISA, *A. vasorum* antibody sandwich-ELISA and Baermann coprology.
  - Positive Baermann coprology result was determined by identifying L1 larvae of *A. vasorum* in the faeces. *A. vasorum* L1 larvae distinguished from *Crenosoma vulpis* (*C. vulpis*) L1 larvae due to their characteristic tail notch.

**Main findings:** (relevant to PICO question):

- Of the seven angiostrongylosis cases, two were positive on Angio Detect™ and three were positive on Baermann coprology.
- In one dog, positive on *A. vasorum* antigen sandwich-ELISA, the Angio Detect™ assay was repeated twice as invalid results were obtained (the control line was not visible).
- **Compared to Baermann coprology, Angio Detect™ had a specificity of 100% (95% CI 39.8–100%; n = 4/4) and sensitivity of 66.7% (95% CI 9.4–99.2%; n = 2/3).** Not all diagnostic test results were available for each dog; however, all PICO relevant data were available.

**Limitations:**

- Small sample size.
- The title is suggestive of angiostrongylosis infection with negative in-house diagnostic results, which was not reflective of all seven cases.
- Results of all diagnostic tests available for each dog were not reported.
- The spectrum of clinical signs and severity of the target disease (angiostrongylosis) found within the university hospital setting may introduce referral bias. The population used may not be representative of all clinical contexts.
- It was unclear whether the indeterminate index result (the invalid Angio Detect™ reading obtained twice for dog 3) was classified as a negative or positive result and therefore could introduce information and interpretive bias to the results.
- No postulated reason for indeterminate Angio Detect™ results was provided, which could aid evaluation of the usefulness of this diagnostic test within a clinical setting.
- No measure of diagnostic accuracy was reported. Evaluation of measures of diagnostic accuracy may have aided the
analysis of superiority or non-inferiority among the index diagnostic tests used.

<table>
<thead>
<tr>
<th><strong>Di Cesare et al. (2014)</strong></th>
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<tr>
<td><strong>Population:</strong> Jack Russell Terrier dogs that resided in a privately owned kennels with a history of <em>A. vasorum</em> infection, in Gallicano Municipality, Tuscany, Italy.</td>
</tr>
<tr>
<td><strong>Sample size:</strong> 15 dogs.</td>
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<tr>
<td><strong>Intervention details:</strong></td>
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<tr>
<td><strong>Day -15</strong></td>
</tr>
<tr>
<td>- Faecal samples were obtained from all dogs for Baermann coprology.</td>
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<td>- Blood samples were taken from all dogs and placed into ethylenediaminetetraacetic acid (EDTA) sample pots. Plasma from each blood sample underwent Angio Detect™ diagnostic testing.</td>
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<tr>
<td><strong>Day 0</strong></td>
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<tr>
<td>- Dogs that had positive test results on day -15 testing (n = 3) were treated with topical Advocate®; Bayer (10% imidacloprid and 2.5% moxidectin).</td>
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<tr>
<td><strong>Days 14 and 28</strong></td>
</tr>
<tr>
<td>- The three treated dogs had faecal samples collected for Baermann coprology and phlebotomy for Angio Detect™ (methodology as above for both diagnostic tests).</td>
</tr>
<tr>
<td><strong>Study design:</strong> Cohort study.</td>
</tr>
<tr>
<td><strong>Outcome studied:</strong> Objective assessment:</td>
</tr>
<tr>
<td>- Positive or negative results from Angio Detect™ and Baermann coprology:</td>
</tr>
<tr>
<td>- Positive Baermann coprology result was determined by identifying L1 larvae of <em>A. vasorum</em>. The dog was considered negative if <em>A. vasorum</em> larvae were not identified.</td>
</tr>
<tr>
<td><strong>Main findings:</strong> No dogs showed clinical signs suggestive of <em>A. vasorum</em> infection. However:</td>
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<td>- Dog 4 tested positive with Baermann coprology but negative with Angio Detect™.</td>
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<td>- Dogs 13 and 14 tested positive with both Baermann coprology and Angio Detect™.</td>
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<tr>
<td>- All three of these dogs were negative with both diagnostic tests on days 14 and 28.</td>
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<tr>
<td>- <em>Compared to Baermann coprology, Angio Detect™ had a specificity of 100% (95% CI 73.5–100%; n = 12/12) and sensitivity of 66.7% (95% CI 9.4–99.2%; n = 2/3).</em></td>
</tr>
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<td><strong>Limitations:</strong></td>
</tr>
<tr>
<td>- Small sample size.</td>
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<tr>
<td>- It was assumed eligibly was limited only to residing in the kennel as eligibility criteria were not reported.</td>
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<tr>
<td>- The population was asymptomatic leading to challenges in comparing data obtained to patients exhibiting clinical signs.</td>
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</table>
- The prophylactic anthelmintic treatment history of each individual dog prior to enrolling in the study was not reported, which could potentially introduce bias in the results.
- It was unclear if the population for this study had been infected with *A. vasorum* before during the kennel’s previous history of *A. vasorum* infection and if this would impact on the results.
- It was not clear if a veterinarian(s) conducted the clinical examinations of the dogs and what aspects of physical examination were being assessed by the individual(s).
- Single, rather than three consecutive day, faecal samples were obtained for Baermann coprology; potentially reducing Baermann sensitivity and introducing measurement bias.

### Lempereur et al. (2016)

**Population:**

- Client-owned dogs were enrolled from one of 17 volunteer small animal practices based in Wallonia, Belgium from November 2014–February 2016.
- The study population included case dogs showing clinical signs suggestive of *A. vasorum* and control dogs presented to their registered clinic for conditions unrelated to *A. vasorum*.
- The clinical signs group contained dogs that presented with clinical signs suggestive of *A. vasorum*, including: dyspnoea, cough, exercise intolerance, bleeding abnormalities and neurological disorders.
- The control group contained dogs that presented to the practices for routine healthcare or unrelated conditions.
- Eligibility for enrolment was not restricted by age, breed or sex.
- Dogs were excluded if travel outside Belgium had occurred within 3 months prior to entering the study.

**Sample size:**

979 dogs were enrolled:

- 757 dogs in a control group;
- 222 dogs in a clinical signs group.

**Intervention details:**

- Angio Detect™ test results were performed following manufacturer’s instructions with all tests undertaken within a strictly adhered 15-minute time frame.
- All staff undertaking the Angio Detect™ test were trained and observed by the first author with positive results recorded by intensity (+, ++, or +++).
- Faecal samples were obtained for Baermann coprology from 41 seropositive dogs (24 controls and 17 symptomatic dogs) and six seronegative dogs with strong suspicion of angiostrongylosis (one control and five symptomatic dogs).
  - Recently passed faeces were collected on 3 consecutive days, or one or two samples per dog where this was not possible.
- Owners completed a questionnaire detailing the patient’s location, breed, sex, age, recent antiparasitic prophylactic protocol, and lifestyle.

### Study design:
Case-control study.

### Outcome studied:
Objective assessment:
- Positive or negative results from Angio Detect™ and Baermann coprology:
  - Positive Baermann coprology result was determined by identifying L1 larvae of *A. vasorum* (using their characteristic tail notch to identify them compared to other parasitic larvae). The dog was considered negative if *A. vasorum* larvae were not identified.
- Prevalence of *A. vasorum* infection in both dogs displaying consistent clinical signs and controls.

### Main findings:
(relevant to PICO question):
- **Control Group**
  - Prevalence of Angio Detect™ positive cases was 3.6% (n = 27/757).
  - 3/27 Angio Detect™ positive cases did not have Baermann coprology performed. 17/24 (70.8%) Angio Detect™ positive dogs had negative Baermann coprology.
- **Clinical Signs Group**
  - Prevalence of Angio Detect™ positive cases was 8.6% (n = 19/222).
  - 2/19 Angio Detect™ positive cases did not have Baermann coprology performed. 8/17 (47.1%) Angio Detect™ positive dogs had negative Baermann coprology.
  - 1/17 positive Angio Detect™ dogs had only *C. vulpis* larvae found on Baermann coprology.
  - 1/5 Angio Detect™ negative dog was positive on Baermann coprology (mixed parasite burden found).
  - Of the 19 Angio Detect™ positive dogs:
    - 6 were coughing;
    - 3 were dyspnoeic;
    - 3 had exercise intolerance;
    - 5 had coughing with exercise intolerance;
    - 1 had a bleeding disorder;
    - 1 had a neurological disorder.

### Limitations:
- Volunteer bias could have been introduced due to the selection process of small animal practices. There was variability in faecal sample collection and the frequency with <3 consecutive daily samples collected was not reported. Both of these factors introduce measurement bias to Baermann coprology.
- It is unclear what time period was considered recent for faecal collection. The method of faecal storage prior to Baermann coprology was also not explained. There may have been inconsistencies and variation, introducing Baermann coprology measurement bias.
Angio Detect™ positive readings were recorded as +, ++, or ++++, however the relevance and results of this classification system were not explained.

Unable to calculate diagnostic accuracy for Angio Detect™ compared to Baermann coprology, as only seropositive dogs were sampled.

**Lempereur et al. (2020)**

**Population:**
- Client-owned dogs were enrolled from one of 26 volunteer small animal practices based throughout Belgium and The Netherlands from February–June 2017.
- The dogs had non-parasitic conditions (examples provided include neutering, trauma, and vaccination).
- Dogs were > 6 months, of either sex and any breed.
- Dogs were excluded if travel outside Belgium or The Netherlands occurred within 3 months or if anthelmintic treatment had been administered within 2 months of entering the study.

**Sample size:** 239 dogs.

**Intervention details:**
- Faeces, collected by the owner, underwent Baermann coprology.
- Dogs underwent phlebotomy at the veterinary clinic. Blood was collected in heparinised or plain tubes, centrifuged and Angio Detect™ conducted using manufacturer’s instructions.

**Study design:** Cross-sectional study.

**Outcome studied:**
- Positive or negative results from Angio Detect™ and Baermann coprology:
  - Positive Baermann coprology was determined by identifying L1 larvae of *A. vasorum*. The dog was considered negative if *A. vasorum* larvae were not identified.
  - Prevalence of *A. vasorum* in dogs not presenting for parasitic conditions.

**Main findings:**
- 3/216 (1.4%) faecal samples were positive for *A. vasorum* on Baermann coprology.
- 1/237 (0.4%) serum samples were positive for *A. vasorum* on Angio Detect™. This dog was also one of the 3 Baermann-positive dogs.
- *Compared to Baermann coprology, Angio Detect™ had a specificity of 100% (95% CI 98.3–100%; n = 213/213) and sensitivity of 33.3% (95% CI 0.84–90.6%; n = 1/3).*
- None of the dogs that tested positive by either test method had signs of gastrointestinal parasitism.

**Limitations:**
- Volunteer bias could have been introduced due to the selection process of small animal practices.
- Recall bias is possible regarding the owner’s recollection of whether and/or when anthelmintics had been administered, which could have affected sample selection since this was an exclusion criterion.
- It is unclear what time period was considered recent for faecal collection. The method of faecal storage prior to Baermann coprology was also not explained. There may have been inconsistencies and variation, introducing measurement bias.
- No explanation for missing diagnostic test results is provided (Baermann 23 dogs and Angio Detect™ two dogs).
- A. vasorum-positive dogs were reported not to have clinical signs of gastrointestinal parasitism, but information regarding other clinical signs was not reported.

### Liu et al. (2017)

#### Population:
- Client-owned dogs that were suspected of clinical A. vasorum infection were selected by their registered veterinarian (based within the United Kingdom [UK]) during June 2012–April 2013.
- Inclusion criteria for suspected clinical A. vasorum infection were at the discretion of the patient’s veterinarian.
- Dogs that had been historically diagnosed (via Baermann coprology performed by two authors based in either Zurich, Switzerland or Copenhagen, Denmark) were also included in the study.
- A control group contained samples from dogs that were A. vasorum negative but positive for other parasite infections.

#### Sample size:
- 214 dogs:
  - 195 dogs suspected of being infected with A. vasorum volunteered from the UK.
  - 19 dogs diagnosed with A. vasorum by Baermann coprology via archive.
An additional 89 negative control samples were obtained:
  - 79 dogs negative for A. vasorum based on antigen ELISA and positive for Dirofilaria immitis (via post-mortem).
  - Three dogs negative for A. vasorum and positive for Crenosoma vulpis based on Baermann coprology.
  - Four dogs experimentally infected with Dirofilaria repens.
  - Three dogs experimentally infected with whipworms, hookworms, and ascarids.

#### Intervention details:
- Faecal samples obtained from the suspected A. vasorum UK cases underwent Baermann coprology at IDEXX Reference commercial Laboratories.
- Serum or plasma obtained from the suspected A. vasorum UK cases were frozen at -20°C (due to time delay between collection and Angio Detect™ testing).
- Serum samples were obtained (from archive) for the historically A. vasorum diagnosed dogs.
- All serum samples (from UK dogs, archive and controlled) underwent randomized blind labelling prior to Angio Detect™ diagnostic testing undertaken by blinded personnel.
- In cases where Baermann coprology results did not match Angio Detect™ results the patient’s sample underwent A. vasorum antigen ELISA testing.

<table>
<thead>
<tr>
<th>Study design:</th>
<th>Cross-sectional diagnostic accuracy study.</th>
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<tbody>
<tr>
<td>Outcome studied:</td>
<td>Objective assessment:</td>
</tr>
<tr>
<td></td>
<td>• Positive or negative results from Angio Detect™ and Baermann coprology, and A. vasorum antigen ELISA (where applicable):</td>
</tr>
<tr>
<td></td>
<td>o Positive Baermann coprology was determined by identifying L1 larvae of A. vasorum (using their characteristic tail notch to identify them compared to other parasitic larvae). The dog was considered negative if A. vasorum larvae were not identified.</td>
</tr>
<tr>
<td>Main findings: (relevant to PICO question):</td>
<td>Dogs with suspected clinical A. vasorum infection from the UK:</td>
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<tr>
<td></td>
<td>• 16/195 of dogs (8.2%) had A. vasorum positive Baermann coprology results.</td>
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<td></td>
<td>• 15/16 dogs had a positive Angio Detect™ test result.</td>
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<td></td>
<td>• The remaining dog of these 16 (i.e. with a positive Baermann coprology result and a negative Angio Detect™ test result) tested negative with A. vasorum antigen ELISA.</td>
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<td></td>
<td>• 2/179 dogs with negative A. vasorum larvae Baermann coprology result tested positive with Angio Detect™. These two samples also tested positive with A. vasorum antigen ELISA.</td>
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<tr>
<td>Archive dog samples:</td>
<td>All 19 samples were positive on Angio Detect™ testing. To be eligible for this group all 19 had previously tested positive for A. vasorum larvae on Baermann coprology.</td>
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<tr>
<td></td>
<td>Compared to Baermann coprology, Angio Detect™ had a reported specificity of 98.9% (95% CI 96.0–99.9%) and sensitivity of 97.1% (95% CI 85.1–99.9%).</td>
</tr>
<tr>
<td>Limitations:</td>
<td>• The process of archive sample selection was not described. This could influence the eligibility of this population group, may introduce selection bias and therefore reduce the strength of the evidence obtained.</td>
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<tr>
<td></td>
<td>• The eligibility criteria for clinical A. vasorum signs were not described and were left to the submitting veterinarian’s opinion, with veterinarians possibly differing in the methods used to identify eligible cases. This could introduce misclassification bias and result in this group not being a true representation of dogs with clinical A. vasorum. It could also affect the accuracy of diagnostic data obtained.</td>
</tr>
</tbody>
</table>
| | • The clinical settings (e.g. first opinion, referral) of UK practices where faecal and serum samples were obtained were not noted in the methods section. This could introduce
spectrum and/or referral bias as the spectrum of clinical signs and disease severity could vary in different clinical settings.

- The method(s) used for faecal sample transport was not described and it is unclear if they were consistent. If faecal storage was not adequate it could have resulted in false-negative results obtained by Baermann coprology.
- The process of thawing the serum/plasma samples and duration of sample freezing was not described and it is therefore uncertain if these could have altered Angio Detect™ testing and introduced measurement bias.
- It is unknown how many faecal samples were obtained over multiple days for a single patient compared to single faecal samples submitted, which could introduce measurement bias for Baermann coprology.

### Olivieri et al. (2017)

<table>
<thead>
<tr>
<th><strong>Population:</strong> Kennel <em>A. vasorum</em> cross-sectional study:</th>
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<tr>
<td>- Dogs that resided at a kennel in Italy.</td>
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<tr>
<td>- Two breeds were present – Belgian Shepherds (n = 3, all males) and Cavalier King Charles Spaniels (CKCS) n = 17; 13 females and four males).</td>
</tr>
<tr>
<td><strong>Retrospective <em>A. vasorum</em> case series:</strong></td>
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<tr>
<td>- These were dogs that had been diagnosed with <em>A. vasorum</em> infection (by post-mortem, Angio Detect™, BAL, and/or Baermann coprology) and were recorded on archive at the Veterinary Pathology Laboratories, Veterinary School of Milan University, Italy, between 1998–2016.</td>
</tr>
<tr>
<td>- Breeds included Jack Russell terrier (n = 1), Cao de Agua Portuguese (n = 1), Maltese (n = 1), Springer Spaniel (n = 1), Italian Hound (n = 1), Lurcher (n = 1), Rhodesian Ridgeback (n = 1) and mixed-breed (n = 4).</td>
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<tr>
<td>- Ages ranged from 8 months to 144 months (but not all age records were complete).</td>
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<tr>
<td>- Both male (n = 5) and female (n = 6) dogs were represented.</td>
</tr>
</tbody>
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<thead>
<tr>
<th><strong>Sample size:</strong> Cross-sectional study: 20 dogs:</th>
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<tbody>
<tr>
<td>- Two clinical angiostrongylosis cases (both CKCS).</td>
</tr>
<tr>
<td>- 18 dogs resident at same kennels.</td>
</tr>
<tr>
<td><strong>Retrospective case series:</strong> 11 dogs.</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th><strong>Intervention details:</strong> Cross-sectional study:</th>
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<tbody>
<tr>
<td>- Faecal samples were obtained from each dog for Baermann coprology.</td>
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<tr>
<td>- Each dog was tested with Angio Detect™.</td>
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<tr>
<td>- All of the above tests were not performed on the dog that was the second clinical case.</td>
</tr>
<tr>
<td><strong>Retrospective case series:</strong></td>
</tr>
<tr>
<td>- Data recorded on cases within the Veterinary Pathology Laboratories archives were examined for positive <em>A. vasorum</em> results.</td>
</tr>
</tbody>
</table>

| **Study design:** Cross-sectional study and retrospective case series. |
| Outcome studied: | Cross-sectional study:  
| | - Positive or negative results from Angio Detect™ and Baermann coprology:  
| | • Positive Baermann coprology was determined by identifying L1 larvae of *A. vasorum*. The dog was considered negative if *A. vasorum* larvae were not identified.  
| Retrospective case series: | - Clinical signs, Angio Detect™ results, results obtained for *A. vasorum* larvae identification, post-mortem findings and clinical outcome of all cases that were recorded positive for *A. vasorum* in the archive records. |

| Main findings: (relevant to PICO question): | Cross-sectional study:  
| | • 1 dog had positive Baermann coprology and was also Angio Detect™ positive.  
| | • Compared to Baermann coprology, Angio Detect™ had a specificity of 100% (95% CI 81.5-100%; n = 18/18) and sensitivity of 100% (95% CI 2.5-100%; n = 1/1).  
| Retrospective case series: | • Four dogs had both Baermann coprology and Angio Detect™ performed.  
| | • Of these, all four were positive on Angio Detect™ and only one was positive on Baermann coprology. |

| Limitations: |  
| | • Small sample size.  
| | • In the cross-sectional study, the origin of each dog was recorded but the relevance of this information was not explained. Additional information such as how long the dog had been in the kennel along with history of when and where any additional boarding may have occurred could have made this information more relevant.  
| | • The search strategy of the retrospective case series was not included and bias may have occurred in retrieving the data due to record keeping potentially not being standardised.  
| | • It is unknown how many different clinicians treated the archive cases and they may not have had standardisation of record keeping. This could introduce selection bias or could have resulted in lower numbers within this study than were truly eligible.  
| | • Single, rather than three consecutive day, faecal samples were obtained for Baermann coprology for the cross-sectional study. Similarly, frequency of faecal collection was not clear in the retrospective case series. Inconsistencies potentially reduced Baermann sensitivity and introduced measurement bias. |

| Schnyder et al. (2014) |  
| Population: | • Naturally infected *A. vasorum*-positive dogs which included:  
|  | • *A. vasorum* Baermann positive dogs from Switzerland. |
O. vasorum-positive dogs (confirmed by larval analysis) from Germany that had been enrolled on previous lungworm studies.
- The population also included experimentally infected A. vasorum-positive dogs, some of which were treated with imidacloprid/moxidectin topical medication.
- There was also a control group consisting of dogs both experimentally and naturally infected with parasitic infections other than A. vasorum as well as negative Baermann coprology. These dogs came from Germany, Switzerland, Czech Republic and Italy.

### Sample size:
- 210 dogs:
  - The group with natural A. vasorum infection included 39 dogs
  - The group of 38 dogs with experimental A. vasorum infection included:
    - 10 dogs who received imidacloprid/moxidectin topical treatment.
  - The control group contained 133 dogs.

### Intervention details:
**Natural A. vasorum infection group:**
- Serum was obtained from all dogs.

**Experimental A. vasorum infected group:**
- For 28 of these dogs that were experimentally infected with a known A. vasorum worm burden, serum samples were obtained before inoculation and also:
  - at days 21, 35, 49, and 62 (±1) after inoculation for all 28 dogs.
  - at days 21, 35, 49, 62, and 76 (±1) after inoculation for 20 of these 28 dogs.
  - at days 21, 35, 49, 62, 76, and 91 (±1) after inoculation for 15 of these 28 dogs.
  - at days 21, 35, 49, 62, 76, 91, and 97 (±1) after inoculation for four of these 28 dogs.
  - two of these 28 dogs received serum samples up until 286 days after inoculation.
  - Baermann coprology results were available for these 28 dogs.
- An additional 10 dogs had serum samples obtained before and after experimental inoculation of A. vasorum. They had Advocate® (10 mg/kg imidacloprid / 2.5 mg/kg moxidectin. This is the equivalent of 0.1 ml/kg Advocate® product application) topically administered between days 81–110 after inoculation.
  - Baermann coprology results were available for these 10 dogs.

**Control group:**
- This group was enrolled for cross-reactivity testing.
- Experienced laboratory personnel analysed all serum samples with Angio Detect™ (following instructions from manufacturer). Serum samples were positive or negative for
<table>
<thead>
<tr>
<th><strong>Study design:</strong></th>
<th>Cross-sectional diagnostic accuracy study.</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Outcome studied:</strong></td>
<td>Objective assessment:</td>
</tr>
<tr>
<td></td>
<td>Positive or negative results from Angio Detect™ and Baermann coprology:</td>
</tr>
<tr>
<td></td>
<td>o Positive Baermann coprology was determined by identifying L1 larvae of <em>A. vasorum</em>. The dog was considered negative if <em>A. vasorum</em> larvae were not identified.</td>
</tr>
<tr>
<td></td>
<td>o <em>A. vasorum</em> antigen ELISA was also used to compare/confirm a positive or negative <em>A. vasorum</em> infection.</td>
</tr>
<tr>
<td><strong>Main findings:</strong></td>
<td>Natural <em>A. vasorum</em> infected group:</td>
</tr>
<tr>
<td>(relevant to PICO question)</td>
<td>• All 39 dogs had positive Baermann coprology.</td>
</tr>
<tr>
<td></td>
<td>• 33/39 dogs tested positive with Angio Detect™.</td>
</tr>
<tr>
<td></td>
<td>• Compared to Baermann coprology, Angio Detect™ had a reported sensitivity of 84.6% (95% CI 69.5–94.1%).</td>
</tr>
<tr>
<td></td>
<td>Experimental <em>A. vasorum</em> infected group:</td>
</tr>
<tr>
<td></td>
<td>• The 28 <em>A. vasorum</em> inoculated dogs:</td>
</tr>
<tr>
<td></td>
<td>o Within 47–55 days post inoculation patency was achieved in all 28 dogs with Baermann coprology.</td>
</tr>
<tr>
<td></td>
<td>o The 10 dogs treated with Advocate®:</td>
</tr>
<tr>
<td></td>
<td>o One dog (which had a worm burden on post-mortem examination) was positive for Angio Detect™ but produced a negative result on Baermann coprology.</td>
</tr>
<tr>
<td></td>
<td>o Baermann coprology was negative 9–20 days after treatment, at 3 weeks after treatment three dogs had negative Angio Detect™ results.</td>
</tr>
<tr>
<td></td>
<td>Control group:</td>
</tr>
<tr>
<td></td>
<td>• Positive Angio Detect™ results were obtained for both dogs that had <em>A. vasorum</em> and <em>C. vulpis</em> combined infections.</td>
</tr>
<tr>
<td></td>
<td>• Compared to Baermann coprology, Angio Detect™ had a reported specificity of 100% (95% CI 74.1–100%).</td>
</tr>
<tr>
<td><strong>Limitations:</strong></td>
<td>The method of selecting the eligible population is not reported and there is the potential for selection bias.</td>
</tr>
</tbody>
</table>
Sensitivity reportedly based on 39 serum samples from naturally infected dogs; however methods state these comprised samples from 28 dogs from Switzerland and 13 serum samples from Germany from previous lungworm studies, with no explanation provided for this discrepancy.

The severity and spectrum of clinical signs of the population are not reported. This makes extrapolation of the tests’ diagnostic accuracy to a clinical setting difficult.

There was no explanation why not all 28 dogs within the experimentally inoculated group were tested for the same duration with both Angio Detect™ and antigen ELISA tests. Flow of experimentally inoculated participants throughout the study was also not clear. As a result, interpretation of the results should be cautious.

### Venco et al. (2021)

**Population:**
- Client-owned dogs presenting with respiratory distress to one of three Italian clinics from November 2016–January 2020.
- Eligible ages were 4 months–2 years (median age 10 months).

**Sample size:** 26 (11 females, 15 males).

**Intervention details:**
- All dogs underwent conscious thoracic ultrasonography (B-Mode and M-Mode) in standing position.
- Faecal samples were obtained from all dogs (except one) for Baermann coprology.
- Faecal sample from thermometer underwent microscopic faecal smear.
- All dogs were tested with Angio Detect™.

**Study design:** Case series.

**Outcome studied:** Positive or negative results from Angio Detect™ and Baermann coprology:
- Positive Baermann coprology was determined by identifying L1 larvae of *A. vasorum*. The dog was considered negative if *A. vasorum* larvae were not identified.

**Main findings:** (relevant to PICO question):
- *A. vasorum*-positive Baermann coprology in 12 of 25 cases.
- Angio Detect™ positive in 11 of 26 cases (9 were also Baermann positive).
- *Compared to Baermann coprology, Angio Detect™ had a specificity of 92.3% (95% CI 64.0–99.8%; n = 12/13) and sensitivity of 75.0% (95% CI 42.8–94.5%; n = 9/12).*

**Limitations:**
- Small sample size.
- High risk of spectrum bias, since the study population comprised solely of young dogs presented for respiratory distress, and as such is not representative of the population of dogs undergoing diagnostic testing for *A. vasorum* infection.
• 25/26 dogs were reported by their owners to be receiving monthly ivermectin-based chemoprophylaxis at the time of presentation, but time since last administration was not reported.
• Single, rather than three consecutive day, faecal samples were obtained for Baermann coprology, potentially reducing Baermann sensitivity and introducing measurement bias.

Appraisal, application and reflection

Canine angiostrongylosis varies from acute presentation of cardiovascular signs, neurological deficits, haematological abnormalities to subclinical infection, making diagnosis challenging to the clinician (Di Cesare et al., 2015). Baermann coprology is the academic reference standard (McGarry & Morgan, 2009) with faecal samples collected over three consecutive days advised due to intermittent shedding of diagnostic L1 larvae (Koch & Willesen, 2009), which can make Baermann coprology sometimes impractical in a clinical setting. Baermann coprology could be considered as a suboptimal reference standard for angiostrongylosis diagnosis. Variations in the pre-patent period of A. vasorum have been reported in experimentally inoculated populations (33–76 days) (Oliveira-Júnior et al., 2006). This poses challenges in a clinical setting, since the duration of inoculation will be unknown and during the pre-patent period larvae will not be found on Baermann coprology (Elsheikha et al., 2014). Reduced, inconsistent larval shedding during low-severity A. vasorum infections has also been reported to reduce Baermann coprology sensitivity (Verzberger-Epshtein et al., 2008). As Baermann coprology is inexact, it is debatable whether bias could be introduced when obtaining sensitivity and specificity results for Angio Detect™ when considering it as reference standard (Naeger et al., 2013). However, it was selected as the comparator for this Knowledge Summary due to the absence of a true gold standard test and because Baermann coprology is commonly considered the reference standard test utilised for angiostrongylosis diagnosis (Conboy, 2009; Traversa et al., 2010; Di Cesare et al., 2014; Schnyder et al., 2014; Liu et al., 2017; Canonne et al., 2018; and Venco et al., 2021). Further alternative diagnostic tests have also been suggested in the literature, such as a faecal smear (Humm and Adamantos, 2010), A. vasorum antigen sandwich-ELISA (Schnyder et al., 2011), or thoracic ultrasonography (Venco et al., 2021). Use of qPCR on BAL could also potentially aid early diagnosis as one A. vasorum-positive dog was diagnosed 16 days prior to Baermann coprology in an experimental study (Barçante et al., 2008). Angio Detect™ may be chosen as a substitute diagnostic test due to its convenience of being a cage-side kit easily stored at room temperature. The versatility of being able to use serum or plasma, and ease of use with a result obtained within 15 minutes with minimal user training required, may also appeal to practitioners.

Literature comparing Angio Detect™ results with Baermann coprology in a canine population was confined to eight papers; all containing limitations. Within the PICO relevant literature, only two publications calculated sensitivity and specificity values with 95% confidence intervals (Schnyder et al., 2014; and Liu et al., 2017). A pre-determined minimum population size is advised in diagnostic accuracy studies to reduce type-1 error affecting the sensitivity and specificity results obtained (Bujang & Adnan, 2016). Small study populations often result in imprecise measures of diagnostic accuracy (Cohen et al., 2016), and inadequate sample sizes may result in misleading estimates of test accuracy. Post-test probabilities and/or likelihoods ratios were not provided, which could influence diagnostic decision making in suspected A. vasorum cases. Although not reported, using the information provided, the sensitivity and specificity of Angio Detect™ compared to Baermann could also be calculated in five included studies (Di Cesare et al., 2014; Olivieri et al., 2017; Canonne et al., 2018; Lempereur et al., 2020; and Venco et al., 2021). Across all seven papers (Di Cesare et al., 2014; Schnyder et al., 2014; Liu et al., 2017; Olivieri et al., 2017; Canonne et al., 2018; Lempereur et al., 2020; and Venco et al., 2021), the sensitivity of Angio Detect™ ranged from 33.3–100%. However, the range of specificity of Angio Detect™ was more consistent between these seven publications; 92.3–100%. The available evidence indicates that Angio Detect™ has high specificity; therefore suggesting a high probability of a negative result indicative of the patient not having angiostrongylosis (Swift et al., 2020). It must also be noted,
due to the small sample size in these studies, the confidence intervals for calculated sensitivity results were

Unanimous results were not obtained using both diagnostic methods with conflicting results occurring within
the literature. Antigen detection inhibited by antigen-antibody complexes were considered the reason for
negative results obtained with Angio Detect™ where Baermann coprology was positive (Schnyder et al., 2014;
and Canonne et al., 2018). Collection of a single faecal sample, reading Angio Detect™ beyond the 15-minute
timeframe and owner recall bias regarding anthelmintic treatment administration were postulated as reasons
for obtaining negative Baermann coprology with a positive Angio Detect™ result (Lemperuer et al., 2016).
Parasites other than A. vasorum cross-reacting on Angio Detect™ serology did not occur in two studies
(Schnyder et al., 2014; and Liu et al., 2017) but happened on one occasion in the case-control study
(Lemperuer et al., 2016). Further research is required on ascertaining Angio Detect™ sensitivity in mixed worm
burdens as an Angio Detect™ negative result was obtained yet Baermann confirmed the presence of A.
vasorum with C. vulpis (Lemperuer et al., 2016).

The three diagnostic test accuracy studies represented the best available design for determining diagnostic
accuracy (Schnyder et al., 2014; Liu et al., 2017; and Canonne et al., 2018). Two of these used case-control
selection (Schnyder et al., 2014; and Canonne et al., 2018), while the other used a cohort selection for part of
the study population (Liu et al., 2017). The retrospective analysis of Canonne et al. (2018) could introduce
selection bias; resulting in a weaker evidence strength compared to that of prospective studies. None of these
diagnostic accuracy studies provided a priori hypothesis (thus data-dredging bias could not be excluded), no
sample size calculations were reported and all lacked a clearly defined intended clinical role for index tests. All
of the naturally infected population in Schnyder et al. (2014) received both Baermann coprology and Angio
Detect™, as did all dogs in both Lui et al. (2017) and Canonne et al. (2018).

Cross-sectional studies are better suited to estimate prevalence rather than evaluating diagnostic test
accuracy, therefore the evidence obtained is of low strength, but still contributes to the PICO question (Olivieri
et al., 2017; and Lemperuer et al., 2020). The population size in Oliveri et al. (2017) was small. Although origin
of the individual was recorded the relevance of this information in relation to interpretation was not
explained. The sample size of the other cross-sectional study was larger overall; however, the prevalence of A.
vasorum within the study population was low (Lemperuer et al., 2020). Details on eligibility criteria were
minimal and further information on why the dog presented to the clinic along with any clinical signs would
have been useful due to the broad spectrum of clinical presentations A. vasorum-positive dogs can present
with. It was uncertain if and when reported administration of anthelmintic treatment was based on owner
declaration (which may be subject to recall bias) or clinical records (misclassification bias is possible if notes
are inaccurate or if the anthelmintic was obtained from another source). Due to the practices entering the
study voluntarily this could introduce volunteer bias. The reason that Baermann coprology and Angio Detect™
tests were not done on all dogs was not explained and could introduce measurement bias.

The retrospective case series is low in the hierarchy of evidence for evaluation of diagnostic accuracy (Olivieri
et al., 2017). Naturally infected case inclusion criteria were limited. The description of methods for archival
retrieval was not particularly detailed, which may introduce misclassification bias if author-led selection
occurred. Missing data could potentially rule additional A. vasorum cases out of inclusion. It is unknown how
many clinicians treated these cases with potentially no standardisation in clinical record keeping. Presumably
differing clinician opinion caused inconsistency in the diagnostic approach to each patient.

The case-control study (Lemperuer et al., 2016) addressed a clear objective. Descriptions of control and clinical
group criteria and size were detailed. Volunteer bias could have existed within population selection. Efforts
were made to reduce measurement bias and maintain interruptive consistency with author monitored Angio
Detect™ interpretation and externally conducted Baermann coprology. Selection bias existed in determining
the eligibility of dogs undergoing Baermann coprology when obtaining a negative Angio Detect™ result due to
subjectively determined allocation. Not all dogs received coprology and the frequency of multiple faecal
collections not occurring were not recorded.
The population size was small for the cohort study (Di Cesare et al., 2014). Population demographic information was not recorded, and the publication also lacked any statistical analysis of obtained results. The aim of the study was not clearly defined nor was the process of kennel selection, with only a single kennel purposively selected. Detailed information of the kennel’s previous *A. vasorum* history was undisclosed.

The population included in the case series was restricted to dogs under 2 years of age presenting to one of three institutions (Venco et al., 2021). Insufficient information was provided to ascertain if referral bias existed as the level(s) of veterinary care provided by these institutions (i.e. first opinion or referral) was not reported. There was inconsistency of faecal collection duration in the PICO-relevant literature. Only one study reported desiccation of faecal samples as a complication (Lempereur et al., 2016), with the other studies not mentioning if this also happened. Two studies did not describe methods for transporting faecal samples for submission (Liu et al., 2017; and Lempereur et al., 2020). The Baermann technique relies on active parasite larvae to obtain results therefore fresh faeces are preferable. If delay in testing is unavoidable, faecal samples should be stored at 2–8°C and transported on ice within a 24-hour period (Broussard, 2003). Delaying Baermann testing and/or the use of inappropriate transport media could result in false-negative results.

All publications, with the exception of one (Schnyder et al., 2014), were on naturally infected populations. This is more reflective of the population encountered clinically. However, details on clinical signs were missing from two studies (Liu et al., 2017; and Lempereur et al., 2020), which makes direct comparison of the sample populations with clinical practice difficult. Additionally, misclassification bias could have occurred, but risk of this bias could not be assessed where defined eligibility criteria were not reported (Liu et al., 2017). Details on disease severity were also lacking in all publications, making extrapolation to the clinical scenario presented difficult. It is unknown if the experimental population in Schnyder et al. (2014) displayed clinical signs.

The available evidence has shown Angio Detect™ can obtain positive *A. vasorum* results in dogs displaying respiratory, haematological, and neurological signs as well as asymptomatic cases (Lempereur et al., 2016). One study obtained positive Angio Detect™ and Baermann coprology in asymptomatic dogs (Di Cesare et al., 2014). Asymptomatic patients will not be presenting in the suggested clinical scenario. However, veterinarians should consider the possibility of encountering an asymptomatic patient; pre-surgical screening with Angio Detect™ was debated in a recent roundtable discussion (Bourne et al., 2020).

Schnyder et al. (2014) highlights the earliest positive *A. vasorum* result could be obtained at 9 weeks post inoculation for Angio Detect™ in an experimental population. Further research in this area is required due to patient drop-out resulting in interpretive bias in this study; however, a larger population size could be an ethical dilemma.

Analysing the available evidence, there is weak evidence indicating that Angio Detect™ is a highly specific and moderately sensitive test for diagnosing angiostrongylosis, in comparison to Baermann coprology. Therefore, in the clinical scenario, the application of the quick, in-house Angio Detect™ test will be an aid for the clinician to rule out angiostrongylosis as a differential diagnosis for the canine patient. However, it should be appreciated that the sample sizes of the studies are small and the comparator test (Baermann coprology) is a debatable reference standard with questionable sensitivity due to intermittent shedding and pre-patent period. As Angio Detect™ has moderate sensitivity, if a negative result were obtained in the clinical scenario (i.e. a patient with high clinical suspicion of angiostrongylosis) additional diagnostic procedures, such as BAL, would be advised.
### Methodology Section

#### Search Strategy

<table>
<thead>
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<th>Databases searched and dates covered:</th>
<th>PubMed on NCBI interface (1920–April 2021)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>CAB Abstracts on the OVID interface (1973–2021 week 15)</td>
</tr>
<tr>
<td>Search terms:</td>
<td>CAB Abstracts:</td>
</tr>
<tr>
<td></td>
<td>1. (dog* or cani* or bitch* or pup*)</td>
</tr>
<tr>
<td></td>
<td>2. (Angiostrongyl* or ‘french heartworm’ or lungworm)</td>
</tr>
<tr>
<td></td>
<td>3. (Baermann* or Baermann-Wetzel or Angio Detect or (Angiostrongyl* and coprol*))</td>
</tr>
<tr>
<td></td>
<td>4. 1 and 2 and 3</td>
</tr>
<tr>
<td></td>
<td>PubMed:</td>
</tr>
<tr>
<td></td>
<td>(dog or canine or bitch or puppy) AND (Angiostrongylosis or ‘french heartworm’ or lungworm) AND (Baermann or Baermann-Wetzel or coprology or Angio Detect)</td>
</tr>
<tr>
<td>Dates searches performed:</td>
<td>22 Apr 2021</td>
</tr>
</tbody>
</table>

#### Exclusion / Inclusion Criteria

**Exclusion:**
- Literature that investigated any species (including human) other than dogs.
- Literature that discussed any parasite other than *Angiostrongylus vasorum*. Exclusion included different species of parasite belonging to the genus *Angiostrongylus*.
- Published review and opinion articles.
- Published research that focused on treatment and did not contain diagnostic results of *A. vasorum* in dogs.
- Published case reports presenting four or less cases.
- Studies on *A. vasorum* prevalence that yielded only one positive case with Baermann and/or Angio Detect™.
- Published research that did not utilise Angio Detect™ as a diagnostic tool.
- Published research that only used Angio Detect™.
- Published research that used diagnostic tests other than Angio Detect™ and Baermann coprology.

**Inclusion:**
- Peer reviewed published literature that focused on *Angiostrongylus vasorum* in dogs (either naturally or experimentally infected) and recorded diagnostic results.
- Diagnosis was using Angio Detect™ in combination with Baermann(-Wetzel) coprology on the same dog.
- The literature had to be fully accessible and could be in any language as long as it could be translated into English.
### Search Outcome

<table>
<thead>
<tr>
<th>Database</th>
<th>Number of results</th>
<th>Excluded – Population not canine and/or the parasite not <em>A. vasorum</em></th>
<th>Excluded – Did not use both Angio Detect™ and Baermann; or used Angio Detect™ and Baermann but had only one positive result</th>
<th>Excluded – Publication had a treatment focus or was a review paper</th>
<th>Excluded – Case study of ≤4 cases</th>
<th>Total relevant papers</th>
</tr>
</thead>
<tbody>
<tr>
<td>CAB Abstracts</td>
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<td><strong>Total relevant papers when duplicates removed</strong></td>
<td><strong>8</strong></td>
<td></td>
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</table>

### CONFLICT OF INTEREST

The authors declare no conflict of interest.

This Knowledge Summary was originally prepared, by the first author, as a graded assessment for the Evidence Based Veterinary Medicine Module as part of the University of Liverpool’s RCVS accredited CertAVP postgraduate qualification. The first author wishes to thank the assessor for their feedback which has been extremely beneficial while preparing the revised Knowledge Summary for publication.

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### REFERENCES


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