Blood culture negative endocarditis: analysis of 63 cases presenting over 25 years

C C Lamas, S J Eykyn

Objective: To analyse cases of blood culture negative endocarditis (BCNE) seen at St Thomas’ Hospital, London, between 1975 and 2000.
Methods: Data on all episodes of endocarditis with negative blood cultures seen at St Thomas’ Hospital between 1975 and 2000 were collected prospectively and analysed retrospectively.
Results: Sixty three patients with BCNE were seen during the study period: 48 (76%) with native and 15 (24%) prosthetic valve infection. BCNE accounted for 12.2% of the 516 cases of endocarditis seen at St Thomas’ Hospital. The diagnosis of endocarditis was clinically definite by the Duke criteria in only 21% (7 of 34) of cases of pathologically proven native valve endocarditis but in 62% (21 of 34) of cases by the St Thomas’ modifications of the criteria. Comparable figures for the 11 cases of pathologically proven prosthetic valve endocarditis were 45% and 73%. Despite negative blood cultures a causative organism was identified in 31 (49%) of the 63 cases: in 15 by serology (8 Coxiella burnetii, 6 Bartonella species, and 1 Chlamydia psittaci); in 9 cases by culture of the excised valve; in 3 by microscopy of the excised valve, on which large numbers of Gram positive cocci were seen although the culture was sterile; and in the other 4 by isolation from a site other than the excised valve (2 respiratory specimens, 1 from the pacemaker tip, and 1 from an excised embolus). In addition 5 of the 6 cases of Bartonella infection were confirmed by polymerase chain reaction study of the excised valve. Two thirds of the 32 patients for whom no pathogen was identified had received antibiotics before blood was cultured. Thus truly “negative” endocarditis was very uncommon (6% of the cases).
Conclusion: If blood cultures are negative in definite or suspected endocarditis, serum should be analysed for Bartonella, Coxiella, and Chlamydia species antibodies, and the excised valve or (rarely) embolus should be analysed by microscopy, culture, histology, and relevant polymerase chain reaction. Other specimens may be relevant. The Duke criteria performed poorly in BCNE; St Thomas’ additional minor criteria gave more definite diagnoses.

METHODS

Data on all episodes of endocarditis with negative blood cultures seen at St Thomas’ Hospital between 1975 and 2000 were collected prospectively and analysed retrospectively. The demographic aspects of the series, clinical, microbiological, and echocardiographic data, as well as diagnostic criteria and outcome, were studied. Patients were assessed by the clinical criteria defined by Durack and colleagues’ (box 1) and by proposed modifications (St Thomas) (box 2). Additional modifications by Dukec are summarised and are discussed in the analysis of the 34 pathologically proven cases of native valve endocarditis (NVE). Results are expressed as mean (SD) when appropriate or as percentages. Statistical analyses were performed by use of SPSS for Windows (SPSS Inc, Chicago, Illinois, USA).

In 2000, Li and colleagues6 proposed the following modifications to the Duke criteria:

- Elimination of echocardiographic minor criteria caused by the widespread use of transoesophageal echocardiography (TOE)
- Considering bacteraemia caused by Staphylococcus aureus a major criterion regardless of whether the infection is nosocomial or a removable source of infection is present
- Recommending that Q fever serology be a major criterion.

RESULTS

There were 48 cases of blood culture negative NVE, involving 31 male patients and 17 female patients. Their mean age was 54 (17) years. Twenty nine patients were referred from other hospitals (60%). Eleven male and four female patients had prosthetic valve endocarditis (PVE) with a mean age of 53 (18) years. Eight (53%) were referred from other hospitals. Forty three of the 63 patients presented between 1990 and 2000.

Affected valves

The mitral valve was affected in 13 of 48 patients with NVE (27%), the aortic in 22 (46%), and both in 6 (13%). The tricuspid valve was affected in six patients (13%) and one had aortic, mitral, and tricuspid involvement (2%). In PVE, the mitral

Abbreviations: BCNE, blood culture negative endocarditis; NVE, native valve endocarditis; PCR, polymerase chain reaction; PVE, prosthetic valve endocarditis; TOE, transoesophageal echocardiography; TTE, transthoracic echocardiography
isms and diagnostic studies. All (51%) were no causative organism found. Table 1 shows organisms and both in one (7%). Seven patients had early PVE (less than one year from surgery) and eight had late PVE. PVE was involved in six cases (40%), the aortic in eight (53%) and both in one (7%). Seven patients had early PVE (less than one year from surgery) and eight had late PVE.

Organisms
Blood cultures were negative in all 63 cases but in only 32 (51%) was no causative organism found. Table 1 shows organisms and diagnostic studies. All *Bartonella* species detected by serology were also grown or identified by polymerase chain reaction (PCR) of valve material from 1995 onwards. Twenty two of the 32 (69%) patients for whom no organisms were identified had been given antibiotics before blood samples were drawn.

Predisposing conditions
Table 2 shows cardiac and non-cardiac conditions known to predispose to endocarditis. Six of 36 patients (17%) who had Gram positive cocci seen or grown in sites other than blood (n = 4) or did not have any identifiable organism (n = 32) had poor dentition or recent dental work. Two of the five patients with *Coxiella burnetii* endocarditis lived on farms and one frequently ate goat's milk and soft cheeses. The patient with *Chlamydia psittaci* endocarditis kept racing pigeons. The patient with *Bartonella henselae* endocarditis kept cats that had fleas. The patient with *Bartonella quintana* “lived rough”; the third did not but kept birds.

Clinical features
Table 3 presents the main clinical features. The duration of symptoms was 7 (6) weeks for the 34 cases of known NVE and 7 (4) weeks in the 12 cases of PVE.

Echocardiography
Forty one of the 48 patients (85%) with NVE had transthoracic echocardiograms (TTE) and 8 (17%) had TOE. TTE showed major criteria for endocarditis in 32 patients (78%) and minor

<table>
<thead>
<tr>
<th>Table 1</th>
<th>Causative organism and diagnostic investigations</th>
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<tbody>
<tr>
<td><strong>Diagnostic test</strong></td>
<td><strong>Native valve</strong></td>
</tr>
<tr>
<td>Serology</td>
<td>Bartonella sp</td>
</tr>
<tr>
<td></td>
<td>Chlamydia psittaci</td>
</tr>
<tr>
<td></td>
<td><em>Coxiella burnetii</em></td>
</tr>
<tr>
<td></td>
<td>Serology + PCR (Bartonella sp)</td>
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<tr>
<td></td>
<td>Valve culture</td>
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<tr>
<td></td>
<td>Valve microcopy</td>
</tr>
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<td></td>
<td>No organism</td>
</tr>
</tbody>
</table>

*Valve microscopy was defined as the recovery of a microorganism consistent with infective endocarditis from either a single blood culture drawn more than 12 hours apart, or in the path or regurgitant jets, or on implanted material, in the absence of an alternative anatomic explanation; or a new partial dehiscence of prosthetic valve or new valvar regurgitation.


definite infective endocarditis

- Pathological criteria
  - microorganisms shown by culture or histology in a vegetation, or in a vegetation that has embolised, or in an intracardiac abscess or - pathological lesions: vegetation or intracardiac abscess present, confirmed by histology showing active endocarditis
- Clinical criteria, using specific definitions*: two major criteria, or one major and three minor criteria, or five minor criteria

possible infective endocarditis

- Findings consistent with infective endocarditis that fall short of “definite” but are not “rejected”

Rejected

- Firm alternative diagnosis for manifestations of endocarditis, or
- Resolution of manifestations of endocarditis with antibiotics for four days or less, or
- No pathological evidence of infective endocarditis at surgery or necropsy, after administration of antibiotics for four days or less

*Specific definitions of proposed criteria

Major criteria:

- Positive blood culture for infective endocarditis:
  - typical microorganisms for infective endocarditis from two separate blood cultures (viridans streptococci, *Streptococcus bovis*, *Haemophilus sp*, *Actinobacillus sp*, *Cardiobacterium sp*, *Eikenella sp*, *Kingella sp*, or community acquired *Staphylococcus aureus* or *enterococci*) in the absence or a primary focus, or
  - persistently positive blood culture, defined as recovery of a microorganism consistent with infective endocarditis from (a) blood cultures drawn more than 12 hours apart; or (b) all of three or a majority of four or more separate blood cultures, with first and last drawn at least one hour apart
- Evidence of endocardial involvement
  - positive echocardiogram for infective endocarditis: (a) oscillating intracardiac mass, on valve or supporting structures, or in the path or regurgitant jets, or on implanted material, in the absence of an alternative anatomic explanation; or (b) abscess; or (c) new partial dehiscence of prosthetic valve or new valvar regurgitation

Minor criteria

- Predisposition: predisposing heart condition or intravenous drug use
- Fever ≥ 38°C
- Vascular phenomena: major arterial emboli, septic pulmonary infarcts, mycotic aneurysm, intracranial haemorrhages, conjunctival haemorrhages, or Janeway lesions
- Immunological phenomena: glomerulonephritis, Osler’s nodes, Roth’s spots, or rheumatoid factor
- Microbiological evidence: positive blood culture but not meeting a major criterion or serological evidence of active infection with organism consistent with infective endocarditis
- Echocardiogram consistent with infective endocarditis but not meeting a major criterion

box 1: duke criteria for diagnosis of infective endocarditis

- Newly diagnosed splenomegaly
- Newly diagnosed clubbing
- Splinter haemorrhages
- Petechiae
- High erythrocyte sedimentation rate, defined as more than 1.5 times the upper limit of normal (> 30 mm/h for patients < 60 years of age; > 50 mm/h for those > 60 years of age)
- High C reactive protein concentration, defined as > 100 mg/l
- Microscopic haematuria*
- Central non-feeding venous lines
- Peripheral venous lines

*Haematuria was disregarded for patients with positive urine cultures, menstruating women, patients with end stage renal disease, and patients with urinary catheters.

Box 2: Proposed additional minor criteria for the Duke classification

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- newly diagnosed clubbing
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<tr>
<th>Diagnostic test</th>
<th>Native valve</th>
<th>Prosthetic valve</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>Serology</td>
<td>1</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td>Bartonella sp</td>
<td>5</td>
<td>3</td>
<td>8</td>
</tr>
<tr>
<td>Chlamydia psittaci</td>
<td>1</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td><em>Coxiella burnetii</em></td>
<td>6</td>
<td>3</td>
<td>9</td>
</tr>
<tr>
<td>Serology + PCR (Bartonella sp)</td>
<td>5</td>
<td>5</td>
<td>10</td>
</tr>
<tr>
<td>Valve culture*</td>
<td>2</td>
<td>1</td>
<td>3</td>
</tr>
<tr>
<td>Valve microscopy†</td>
<td>1</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td>No organism</td>
<td>2</td>
<td>8</td>
<td>10</td>
</tr>
</tbody>
</table>

(*S. aureus* (sputum 1, pleural fluid 1), coagulase negative *Staphylococcus* [pacemaker tip], viridans streptococci [embolus]. 
†Gram positive coccus.
§*Streptococcus bovis*, *Streptococcus mitis*, *Enterococcus faecium*, *Histoplasma capsulatum*, *Aspergillus sp*, *Aspergillus fumigatus*. 
¶*Rhodococcus sp*, *Staphylococcus aureus*, *Staphylococcus epidermidis*. 
§*Streptococcus viridans*.*
criteria in 8 (20%) and was non-contributory in just one case. TOE showed major criteria in two cases where TTE was unhelpful. Thirteen of the 15 patients with PVE had TOE (87%); of these, 10 (77%) met major criteria for endocarditis (four had abscess). One TOE showed only a minor criterion and two were non-contributory.

Complications

Table 4 lists the complications.

Surgery and mortality

Thirty six of the 48 patients with NVE had surgery (75%), of whom 33 survived (92%). Three of the six patients who did not have surgery died. Causes of death in these patients were pancreatic carcinoma, sudden arrest on admission, and heart failure. Two of the three patients who had surgery and died had unrelated causes. Both had early PVE (one and two months, respectively) and had emergency valve replacements (one aortic and one mitral valve). Two patients with PVE did not have surgery and survived: one was 43 years old, had had a valve and one mitral valve. The other was 81, had had an aortic prosthesis for nine years, and had C. burnetii endocarditis. He received doxycycline and rifampin for many months and died of unrelated causes. His antibody titre was still positive when last checked. Surgical mortality in the PVE group was 2 of 13 (15%) and overall mortality 2 of 15 (13%).

Analysis of diagnostic criteria in 34 cases of pathologically proven BCNE affecting native valves

Diagnosis of infective endocarditis was pathologically proven in 34 cases: 19 by macroscopic surgical findings, 13 by excised valve histology, and 2 by histopathology of valves at postmortem examination.

The clinical diagnosis of endocarditis was definite according to the Duke criteria in 7 (21%) of the 34 cases, and all were based on one major and three minor criteria. Our modifications of the Duke criteria upgraded probable cases to definite by adding one or more minor criteria to the 14 cases with one major and two minor criteria. In 7 only one criterion was added (erythrocyte sedimentation rate or C reactive protein), in 1 case two criteria were added, in 5 cases three or more criteria were added, and in 1 case four minor criteria were added. These minor criteria were splenomegaly (3), haematuria (4), clubbing (2), splinters (2), rash (1), petechiae (2), and purpura (1) (table 5).

If we consider Duke's modifications of their criteria, 11 of 34 cases would be definite (32%). These 4 new definite cases would be based on considering Coxiella serology a major criterion. All 4 of these cases were upgraded by St Thomas' modifications based on minor criteria.

DISCUSSION

These 63 cases of BCNE constituted 12.2% of all cases of endocarditis seen in St Thomas' Hospital between 1975 and 2000. This accords with reported rates of between 3–23%.

However, in only 6% of our 63 cases did we fail to detect a prosthetic aortic homograft for seven months, and was cured after two weeks of receiving benzylpenicillin plus gentamicin; the other was 81, had had an aortic prosthesis for nine years, and had C. burnetii endocarditis. He received doxycycline and rifampin for many months and died of unrelated causes. His antibody titre was still positive when last checked. Surgical mortality in the PVE group was 2 of 13 (15%) and overall mortality 2 of 15 (13%).

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However, in only 6% of our 63 cases did we fail to detect a
causative organism, which suggests that truly negative infective endocarditis is an uncommon diagnosis. In common with other series 24 25 two thirds of our patients in whom Gram posi- tive cocci were seen or grown from sites other than blood (vessels, sputum, embolus, or pacemaker wire tip) had received antibiotics, suggesting a staphyloccocal or streptococcal cause. Serology to detect antibodies to Gram positive cell walls and broad range PCR on valve material were not performed, but these investigations might have confirmed the identity of the causative pathogen.17 18 PCR amplification of specific gene tar- gets and universal rRNA loci for bacteria and fungi has been done in blood, as well as in valve material from patients with endocarditis, 26 and might have contributed to diagnoses in our series. No studies were done for Tropheryma whippelii, another well described cause of BCNE.17 25

The administration of antibiotics, especially aminoglycosides, also influences isolation of Bartonella species from blood and valve cultures, 27 but our cases of Bartonella endocarditis were diagnosed by serology or PCR of the excised valve, which have higher sensitivity. Nearly one quarter of our cases of BCNE were caused by “atypical” bacteria: C burnetii (8 of 63), Bartonella species (6 of 63) and C psittaci (1 of 3). This is in contrast to the last published series of BCNE where no positive serological results were seen for Coxiella, Legionella or Chlamydia species (Bartonella serology was not done).1 Although cross reaction of Bartonella antibodies with Chlamydia antibodies is described, 28 our patient with Chlamydia antibodies had a strong epidemiological link to Chlamydia organisms, as he kept racing pigeons. C burnetii and Bartonella species are well recognised causes of BCNE, 21 26 and it is estimated that Bartonella species cause 3% of all cases of infective endocarditis. Four of our 63 cases (6%) were caused by fungi (“yeast” 1, Aspergillus species 2, Histoplasma capsulatum 1). This last case has been published.29 The sensitivity of blood cul- ture in fungal endocarditis was 54% in a review of 270 cases of fungal endocarditis, but fungi caused less than 10% of cases of endocarditis overall. 30

Prosthetic valves and bicuspid aortic valves were the most frequently infected in our series (24% and 15%, respectively), which is in contrast to the series reported by Pesanti, 31 with 52 cases of BCNE, in which 50% of the patients had rheumatic heart disease and only 8% had prosthetic valves. Bicuspid aortic valves were not specifically mentioned but they were the most common cardiac locations of NVE in two recent series. 7 11

Seventeen per cent of patients did not have fever, which is a higher incidence than usually reported with positive blood cultures 32 33 but is similar to that of 19% reported by Hoen and colleagues. 2 This may be the result of previous administration of antibiotics. Patients with Coxiella and Bartonella endocarditis are likely to present with low grade or no fever. 27 28 The complications and mortality are similar to recently published series of blood culture positive NVE and PVE.7 11 12 However, when comparing our series with a recently published series of BCNE, 2 our rate of severe complications such as heart failure and valvar regurgitation was less (50% v 78%); the rates of major embolic events (cerebral and pulmonary) were similar (29% v 34.4%). The sensitivity of echocardiograms for the diagnosis of endocarditis is similar to that of positive blood cultures. 6 21 TTE showed a major criterion for endocarditis in 78% of our cases involving native valves; few of these patients had TOE. This is in contrast with our cases of BCNE involving prosthetic valves, where 13 of 15 patients had TOE, which showed major criteria in 78%. TOE is superior to TTE in diagnosing endocarditis in the subgroup of patients with negative blood cultures, as has been shown by Kupferwasser and colleagues’ studying 32 cases of native valve BCNE, and we agree it ideally should be performed in all of these patients.

Analysis of patients with negative blood culture by the Duke criteria shows that these criteria perform poorly in such patients, which is not surprising since blood culture positivity is a major criterion. This was also shown by Habib and colleagues 35 where 22 of 93 patients with pathologically proven infective endocarditis were misclassified as possible by the Duke criteria. Twenty one of the 22 patients in this series had negative blood cultures and 19 had one major and two minor criteria. In our series, the Duke criteria diagnosed only 21% of the pathologically proven native valve cases as definite, whereas the St Thomas’ modifications diagnosed 64% (p < 0.05). This was achieved by the addition of one or more minor diagnostic crite- ria. It has been previously proposed that serology for Coxiella species be considered a major criterion for infective endocarditis. 17 If we use Duke’s modifications of the Duke criteria 36 we would identify only 32% of pathologically proven native valve blood culture negative cases in the present series. It is our experience and that of others (Raoult D, 9th International Congress on Infectious Diseases, Buenos Aires) that rapidly developing clubbing and splenomegaly are common findings in patients with Coxiella and Bartonella deep seated infections such as endocarditis, and these features should be considered in the diagnostic criteria for endocarditis (as they were in the Beth Israel criteria). It may be argued that the Duke criteria would have performed better in the present study if TOE had been performed in all the patients, but as has already been pointed out, TOE was done in most PVE cases (13 of 15). TTE performed well in the NVE subset (78% major criteria detected).

In summary, BCNE may be a diagnostic dilemma but in many cases the causative pathogen can be determined. It is important to elicit a history of any previous antibiotic that the patient has taken; serum should always be analysed for anti- bodies to organisms that cannot be cultured by routine meth- ods such as C burnetii, Bartonella, and Chlamydia species, and if available for antibodies to Gram positive bacterial cell walls; and excised valves or vegetations or other relevant material should undergo microscopy, culture, histopathology, and relevant PCR analysis. Lastly, it should be noted that the Duke criteria and their modifications are unsatisfactory because the yield few definite clinical diagnoses. They require amend- ments to accept serology for organisms besides Coxiella species as a major criterion and they should incorporate additional minor criteria, as has already been suggested.


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REFERENCES
1 Osler W. Chronic infective endocarditis. QJM 1909;2:219
Left atrial thrombus caused by the primary antiphospholipid syndrome causing critical functional mitral stenosis

Intracardiac masses are rare and their exact nature is often difficult to ascertain by conventional imaging methods alone. A 40 year old white woman presented with a six month history of progressive dyspnoea, orthopnoea, and peripheral oedema. Clinically she had signs of biventricular failure. Images from her transthoracic echocardiogram showed a large left atrial mass causing critical functional mitral stenosis (below left and right). A differential diagnosis of vegetation, thrombus, myxoma or other cardiac tumour (for example, lymphoma) was considered. A thrombophilia screen demonstrated raised serum concentrations of both anticardiolipin antibody (aCL) and antibody to its cofactor, β2 glycoprotein I (β2GPI) on two separate occasions. An autoimmune profile (including antinuclear antibody and antibodies to extractable nuclear antigens) was negative, confirming the diagnosis of primary antiphospholipid syndrome.

The patient was initially managed with high intensity anticoagulation, but failed to improve on clinical or echocardiographic criteria, and therefore surgical resection of the mass was performed. Histopathological analysis of the mass confirmed old and recent thrombus undergoing organisation from the base and infiltrated by reactive macrophages. This case highlights the need to screen all patients with intracardiac masses of unknown aetiology for prothrombotic disorders.

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