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Clinical Toolkit 8: Laboratory handling and reporting of blood cultures

This clinical toolkit has been developed around existing Public Health England (PHE) Standards for Microbiology Investigation (SMI B37) Investigation of Blood Cultures (for Organisms other than *Mycobacterium* species)¹. It is designed to provide operational solutions to the challenge of rapid reliable processing of blood cultures in order to optimise outcomes for patients, including reducing length of stay, while providing procedural opportunities to improve antimicrobial stewardship and facilitate seven day working. Many of the improvements can be achieved through better utilisation of existing resources.

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Background

Blood culture microbiology forms a parallel pathway to the bedside clinical management of sepsis, the two being intrinsically interlinked. There is commonly inequality between the speed with which blood cultures are handled out of hours, over weekends and bank holidays compared to those processed during “office hours”. This would be obviously unacceptable were it part of the ward-based clinical management of sepsis.

From the patient’s perspective, the clock is already ticking at the time a blood culture is collected. From a systems perspective, its processing should be considered to have started from the moment that the test was considered clinically necessary. All delays in the pathway- bedside and laboratory- should be kept to a minimum.

Results of UK blood culture practice surveys² indicate there is significant room for improvement in turnaround times much of which can be achieved with better use of existing resources.

Professional responsibility and accountability

Clinical Pathology Accreditation (CPA) sets standards for pathology laboratories. The standard for turnaround times is set through local discussions between clinicians and laboratory, allowing standards to vary across the UK. While setting a standard through discussion between clinicians and laboratory is perfectly appropriate for less urgent tests it does not appear logical to permit differing standards for the processing of urgent tests from acutely unwell patients.

In 2013 Public Health England (PHE) published a new standard for blood cultures, introducing target times for the processing of specimens¹. In the Republic of Ireland, following a number of high-profile cases where the pathology sample turnaround was found to be unacceptably slow a standard has been enforced, with the Irish National Accreditation Body (INAB) choosing to adopt the PHE standard. Laboratories were given a three-month period of grace to conform to the standards following which, if they failed to meet the standard, they would lose accreditation for the processing of blood cultures. These changes have yet to be implemented across the UK.

With regard to blood cultures, it is implicit in setting the standard that both laboratory and the clinicians served are aware of how quickly a specimen can be processed. The results of three surveys (local, regional and national)² of blood culture practice suggest that most laboratories are themselves inadequately informed to judge an acceptable standard. The situation is not helped by the biological component of the test; there is a variable time to positivity for blood cultures – the resulting unpredictability and the way positives are dealt with serve to conceal inadequacies of the system and effectively reduce clinician expectations. In contrast, most people are aware that a *Clostridium difficile* toxin result can and should be turned around within twenty-four hours by the laboratory; failure to do so is likely prompt the result being chased by the requesting team. Without national standards and audit of practice against these, neither laboratory nor ward-based clinicians are in a

position to ascertain whether best practice has been attained in managing a patient. Resultant deficiencies in patient care will almost certainly go undetected at both local and national level. The development of clinical effectiveness in pathology should aid in addressing such issues and facilitate incorporation into CPA ISO standards.

Delivering excellent sepsis care:

Recommendations for laboratories

Laboratories should audit the various stages in blood culture processing and where deficiencies are identified utilise the above toolkit to rectify the situation.

We know that improvements in blood culture turnaround time can be provided through better use of existing resources. Depending on the laboratory services available different solutions will be required / appropriate. The changes required are in keeping with many national initiatives such as seven-day working in the NHS, modernisation of pathology, multidisciplinary working and antimicrobial stewardship. In addition this will move to provide equality of laboratory practice regardless of time and place.

Some laboratories audit their blood culture performance by the number of finalised reports issued at the point of the maximum length of time the blood cultures are incubated for. This is inadequate as it provides little information on performance relating to time-sensitive handling of the important positive cultures which make up the minority of specimens. Audit should be broken down into key elements in order to ascertain that the whole pathway is optimised as detailed below.

The impact of future technological advances in blood culture methodology is likely to be negated if the existing blood culture pathway is not already optimised. For example, utilising an expensive PCR based methodology to obtain a four hour result on a positive blood culture when the same result could have been obtained sooner had the specimen been processed with conventional methodology without delay.

Recommended audit standards

By mapping the blood culture process, it is possible to identify critical control points where delays may occur. In targeting these, there is great potential to improve turnaround times leading to more efficient patient care without incurring excess laboratory or other costs.

A1. Sample collection date and time (100%)

This may be difficult initially for some Trusts; increasing use of electronic test requesting will facilitate this by automating capture of these data. Attainment of this

target should be an organisational rather than laboratory responsibility, but consideration should be given to responding to evidence of poor compliance.

- A2.** Date and time of receipt in the laboratory (100%)
- A3.** Date and time of incubation (loading) (100%)
- A4.** Date and time of sample flagging positive (100%)
- A5.** Completion date and time for each ID & sensitivity methodology employed (100%)

These may include:

- Gram stain
- Antigenic ID
- Molecular ID (including MALDI-TOF)
- Biochemical ID
- Sensitivity test resulting

- A6.** Date and time of preliminary positive reporting (100%)
- A7.** Date and time of preliminary negative reporting (where undertaken) (100%)
- A8.** Date and time of reporting of own laboratory's completed findings (100%)
- A9.** Date and time of final reporting of external lab. findings (where undertaken) (100%)

These standards are designed to emphasise the role of the blood culture specimen in sepsis management; they do not assume that the pathology service needs to invest in specific equipment, but encourage the optimal use of the resources already in place.

The above data should be regularly analysed (at least yearly), for all sites served by the lab and for each clinical area within these. The following intervals may be considered to be particularly important, although others may be of local interest:

- Time from blood culture collection to receipt with the laboratory (A2-A1)
- Time from receipt to loading of blood culture machine (A3-A2)
- Time from blood cultures flagging positive to removal and processing (A5-A4)
- Time from preliminary processing to preliminary reporting (A6-A5)

Mechanisms for, and range of actions taken by, the medical microbiologist are beyond the scope of this document. These are currently subject to change as a result of technological advances both within and outside the lab. It is important to note that along with the discussed benefits of rapid turnaround comes the potential for doing patients a disservice if

false positive results are communicated, in some cases even if due diligence in applying caveats have been observed.

The greatest gains in speed of turnaround of blood cultures/impact on patient management are likely to come from:

- Minimising delays from collection to loading on the blood culture machine
- Rapid processing once a blood culture flags positive
- Performing direct antibiotic sensitivity tests (4% of laboratories do not)
- Selective use of rapid identification/sensitivity tests

Linking laboratory performance improvement to clinical management

Effort should be made to identify opportunity to link laboratory audit with evidence of clinical response. An excellent system would provide evidence of communication between the laboratory and the bedside team, and record response to results: escalation of spectrum, de-escalation or cessation of therapy, alteration in duration of therapy and/or new strategies to identify and control source. As part of the Sepsis Team within an organisation, medical microbiologists and antibiotic pharmacists should evaluate what can be done to facilitate bedside decision-making and audit.

Antibiotic policies

Well-written, accessible antibiotic guidelines are essential to managing the infected patient.

Laboratory data are a key component in determining appropriate empirical guidance. Conventional wisdom is that inappropriate initial treatment adversely affects patient outcome. Not all of the literature supports this; furthermore distinction must be drawn between in-vitro resistance and clinical failure.

When revising antimicrobial guidelines, consideration is given to the pragmatic balance between adequate spectrum and risk of nurturing antimicrobial resistance: but what level of failure of empirical antibiotic therapy is acceptable for patients with positive blood cultures? Clinical criteria should also be taken into account, including the collateral implications of the empirical guidance.

For example, if in a hospital approximately 35% of *E. coli* in blood culture are resistant to co-amoxiclav; co-amoxiclav combined with gentamicin for *E.coli* bacteraemia provides successful in-vitro cover in 95% cases. In this instance the bar has been set at an empirical cover failure rate of one in twenty patients but would a failure rate of one in ten be equally palatable? Evidence of the numbers needed to treat and harm as the spectrum of empirical antimicrobial cover in sepsis varies is unavailable.

Antibiotic policies should be based on the best available evidence, but are necessarily limited in their scope and efficacy. With Gram-negative antibiotic resistance increasing, it is inevitable (until new effective agents are produced) that we will see increasing use of broader spectrum agents and rising rates of failure of empiric antibiotic therapy. Timely processing of blood cultures provides an opportunity to administer an effective antibiotic where resistance was not suspected.

Clinical assessment of the patient

Clinically relevant deficiencies in antibiotic therapy are likely to be detected by clinicians noticing a failure to improve or deterioration of the patient and empirical choices adjusted accordingly. A positive blood culture may not only identify patients in whom empiric treatment is ineffective, but allows for the most suitable treatment for each specific patient and scenario to be identified and used: this may include de-escalation. There are limits to clinical acumen and mistakes are made; patients are not infrequently diagnosed as having a chest infection when the source is the urinary tract. It can also be difficult to accurately assess response within the first 24-48 hours of antimicrobial treatment.

It is possible to perform rapid sensitivity testing³ giving same-day extended-spectrum beta-lactamase (ESBL) status and gentamicin sensitivity when Gram-negative rods are identified on Gram staining. Detection of an ESBL-producing organism provides an opportunity to check a patient's condition and recommend a drug with known efficacy.

Example

A patient admitted via the Emergency Department is transferred to the Acute Medical Unit. At the post-take ward round the following morning, the patient is relatively stable clinically and the Gram stain for Gram-negative rods is reported- standard empiric therapy is continued. When the rapid ESBL result is available later the same day, further enquiry of the junior doctor by the medical microbiologist reveals that the patient is deteriorating, prompting an early switch to laboratory-confirmed rescue therapy.

Data collection: proposed solutions

A1. Sample collection date and time / A2. Date and time of receipt in the laboratory

Audit of transport times is key to establishing whether there are undue delays between collection and receipt in the laboratory. Audit points may be obtained from time of request logged by electronic request systems and from date and time stamping of specimens on receipt in reception. Where electronic requesting systems are not in place, we have found that asking junior medical staff to ensure the date and time is recorded on request forms provides sufficient data provided there is an audit system in place.

It is a prerequisite that clinical staff are trained when and how to collect blood cultures correctly (including number of sets, for example where line sepsis is suspected taking a peripheral set and a culture through the line) minimising the risk of contamination. False positives due to contamination cost laboratory time and money, create reduced faith in the system and may have direct and indirect negative effects on patient care.

A3. Date and time of incubation (loading)

Time to positivity on the blood culture machine may be increased by delays in loading after sample collection (A3-A1) (demonstrated in a regional survey of neonatal blood cultures), compounding the turnaround time wasted.

The way blood cultures are handled on receipt during the working day tends to be different compared with out of hours. The aim should be to provide a 24-hour service for the loading of culture samples directly on to the blood culture machine with minimal delay. Placement of blood cultures in a separate incubator or leaving at room temperature overnight incur significant delays and should not be regarded as acceptable alternatives (see Appendix I).

Blood culture machines are becoming easier to load, and technology can flag up in the microbiology laboratory if a blood culture is positive even when the blood culture machine is situated remotely. We suggest that where audit identifies delays in loading, organizations should seek opportunities for improvement considering currently available technology.

Box I. Suggested options for improving times to incubation

Suggested options:

- Blood culture machine placed in a centralised (i.e. not specific to microbiology) specimen reception or blood sciences laboratory. As blood sciences usually operate 24-hour shift systems, blood sciences staff should be trained and expected to load blood cultures at night. This will be something of a culture change and will result in some resistance, but training should reinforce the impact they may have on an individual patient's management.
- Main blood culture machine sited within the microbiology laboratory, with a smaller unit within specimen reception. The newer generation of blood culture machines will communicate with each other and preserve data for each bottle providing the sample is relocated in the other machine within a specified period of time. Thus specimens could be transferred from the smaller machine in specimen reception to the main laboratory at the end and the beginning of each working day.
- Location of smaller 'satellite' blood culture machines at peripheral hospitals without a microbiology laboratory that communicate with the larger blood culture machine in the centralised laboratory. The transport time from the peripheral hospital to the main laboratory machine must be within the timeframe for data to be maintained for the individual bottles on transfer. Such satellite machines may even be placed in relevant high-use clinical areas such as Critical Care.

The model of delivery of pathology is currently at its greatest rate of change. There are now a number of acute hospitals without a microbiology laboratory presence on site. It is commonplace to have 24-hour shift systems in the blood sciences. Many laboratory services have been centralised, and perhaps the greatest impact has been on the delivery of microbiology services. Planning exercises to reduce delays should use process mapping and involve service users and laboratory staff as well as medical microbiologists and budget holders.

A4. Date and time of sample flagging positive

The telephone survey of laboratories outlined in Appendix I highlighted that there is variation in the time when positive blood cultures are removed and processed out of hours. No laboratory routinely processed blood cultures after 10 o'clock at night. In some laboratories, positive blood cultures remain untouched in blood culture machines for up to 24 hours after flagging. This way of processing blood cultures causes delays in patient management but also results in handling challenges in the morning when all the positive blood cultures from overnight have to be unloaded and processed often by one biomedical scientist, thus producing further delays.

A5. Individual completion date and time fields for each ID and sensitivity methodology employed

Gram staining, although a relatively simple test, requires expertise to perform. For trained microbiology biomedical scientists performing a Gram stain should be straightforward. If other staff, outside of microbiology, are to be trained in performing Gram stains then competence can only be maintained if they are performing such a test frequently. It is difficult to envisage how blood science staff would gain enough experience of performing Gram stains solely by performing them on positive blood cultures out of hours. However, technology which is already in existence may allow non-microbiology staff to do the equivalent of, if not better than, a Gram stain. Reliable out-of-hours Gram staining will not be achievable in every laboratory unless they have a microbiology 24-hour shift system in place or multidisciplinary staff who are adequately trained in performing the test.

Empirical plating of blood culture broths for sensitivity and ID is quick and simple. Blood science biomedical scientists can easily be trained to safely set up these tests out of hours, enabling reading and interpretation by specific, trained and experienced staff in routine hours. Performing a blind subculture (i.e. without a Gram stain) will speed up the process of obtaining sensitivities and identification for many positive cultures by at least 24 hours. We would argue that for Gram positive organisms identification of the organism is, on the whole, more important than sensitivities. For common Gram positive organisms once growth is visible, simple tests such as a staphylococcal latex test or a Lancefield grouping are likely to

provide the identity of the organism. For Gram negative organisms the results of antibiotics sensitivity testing are more important. We would recommend as a minimum standard establishing an extensive Gram negative sensitivity testing panel on every blind subculture. Where laboratories have access to Matrix-assisted laser desorption/ionization – time of flight (MALDI-TOF), the identification of organisms once grown on the plate can be established rapidly.

For a Gram negative organism the endpoint may be considered as determination of antibiotic sensitivities, and for a Gram positive it might be identification of the pathogen.

More work is needed concerning the clinical and cost effectiveness of rapid tests for the identification of organisms. MALDI-TOF spectra are increasingly used in clinical laboratories for the rapid identification of bacteria and fungi. Experience using this technology on positive broths, or after a few hours' subculture growth is growing rapidly. Use of MALDI-TOF technology to perform resistance marker profiling is also developing.

A number of rapid antigen tests are available for the identification of organisms (again mainly Gram positive) such as pneumococcal antigen, and for the grouping of haemolytic streptococci and identification of staphylococci. Their use by laboratories at present is highly variable. Rapid identification of staphylococci allows more focused antibiotic treatment of *Staphylococcus aureus* and reduces time spent in dealing with coagulase negative staphylococci where they are contaminants (most instances). Rapid identification of *Streptococcus pyogenes* where the clinical picture is not suggestive of the diagnosis opens up the potential for earlier clinical reassessment and consideration of use of specific agents such as clindamycin and immunoglobulin.

A rapid test for the identification of ESBL-producing organisms in blood culture has been available for some years³. This provides a result within as little as four hours of the blood culture flagging positive. Every laboratory in the country has the potential to perform this test, it only requiring a standard sensitivity agar Petri dish and commonly used antibiotic discs. We recommend that this test become routine.

One of the concerns raised by blood sciences biomedical scientists (BMSs) is that there are occasional periods of time at night where they can be extremely busy (e.g. urgent cross match and product supply in massive haemorrhage) and cannot deal with a positive blood culture immediately. They need to be reassured that in these circumstances a short delay in processing the blood culture is acceptable. However, this must not translate into a situation where it defaults to blood cultures not being processed: hence the requirement for an on-going programme of audit.

Where a blood culture machine is sited at an acute hospital with no microbiology services on-site there will usually be a 24-hour shift system for blood sciences. Arrangements can be made whereby if the blood culture machine flags a positive blood culture out of hours, the

on-site blood science BMS removes a bottle and has it sent by courier to the main laboratory for processing.

A6. Date and time of preliminary positive reporting (100%)

Provided that preliminary pathogen identification is made by a suitably skilled BMS in a CPA-accredited laboratory, there is no reason why the majority of results should not be uploaded to the hospital clinical information system immediately, so the result is available should the patient's condition give cause for concern out of hours. It is straightforward to log and monitor these times, and to automate the process.

A7. Date and time of preliminary negative reporting (where undertaken)

Preliminary negative reporting is particularly relevant in neonatal practice where a 36 hour negative blood culture is used to discontinue antibiotic therapy in neonates not displaying features of infection. The majority of significant positives in all patient groups are positive in less than 36 hours' incubation. Reporting preliminary negatives is an appropriate antimicrobial stewardship measure which can be easily recorded.

A8. Date and time of reporting of own laboratory's completed findings

A9. Date and time of final reporting of external laboratory findings (where undertaken)

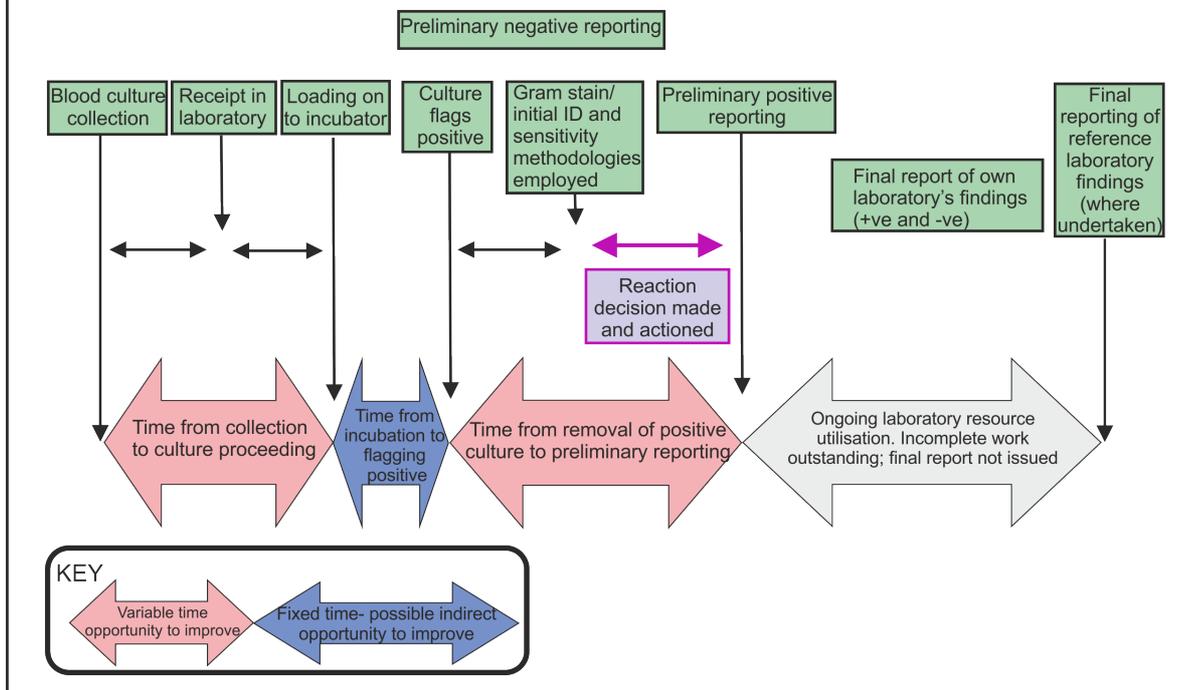
As part of the audit we recommend that a number of patients with positive blood cultures be selected at random (sampled) including those patients clinically identified with sepsis and that a full review be undertaken to ascertain whether the speed and accuracy of delivery of microbiology results, as well as the other components of their treatment and management, was adequate.

Currently, assessments of whether a laboratory's performance is adequate are not routine. Proposed Keogh mortality reviews may be an opportune time to review appropriate use of the laboratory more generally. Given the numbers of patients dying in hospital while or recently following receipt of antibiotics this is expected to make significant demands on infection services.

We would recommend that a senior biomedical scientist should be part of every sepsis group within hospitals. Biomedical scientists are in a position to provide critical audit data, advice on the most appropriate and rapid laboratory techniques and organise the laboratory services to respond to the challenges of sepsis.

An overview of the impact of the differing ways laboratories process blood cultures on turnaround times is illustrated in Appendix 2.

Figure 1. Outlining the critical control points in the blood culture processing pathway and where potential inroads may be made in reducing processing times



Recommended national standards for blood culture processing

Summary Table 1: Pre-Analytical Standards

Inoculated bottles should be incubated as soon as possible, and within a maximum of four hours.

Investigative Stage:	Standard:
Pre-Analytical	Time Period
Collection to Incubation	≤4hr

The four hour turnaround time from collection to incubation for blood culture samples reflects their clinical significance.

Summary Table 2: Analytical Standards

Results of the following identification and sensitivity tests (if performed) should be completed within the following time frames from flagging positive:

Investigative Stage:	Criteria:	Standard:
Analytical		
Flagging Positive to Microscopy, Identification and Sensitivities	Test (if test performed)	Time Period to Result
	Gram Stain	≤2hr
	Rapid Antigen Testing	≤2hr
	Molecular Assays	Same day
	Isolate Identification (Direct/Automated)	≤24hr
	Isolate Identification (Conventional Methods)	24-48hr
	Isolate Sensitivities (Direct/Automated)	≤24hr
	Isolate Sensitivities (Conventional Methods)	24 hours (initial results)
Negative at 36 hours' incubation	Preliminary “negative after 36 hours incubation” report issued. All labs should have this as an automated LIMS facility.	

Summary Table 3: Post-Analytical Standards

Standards have also been set for the laboratory turnaround time (the time between receipt in the laboratory and reporting):

Investigative Stage:	Criteria:	Standard:
Post-Analytical		
Positive Report (from receipt in laboratory to positive reporting)	Preliminary Positive Report (Telephone/Fax/Email)	Immediately, within 2hr of identity/sensitivity availability. (see Summary Table 2 above)
	Final Positive Report	≤5 days (or greater if extended incubation required, or if isolates are sent to a reference laboratory for confirmation)

Appendix I.

Pre-incubation of blood cultures: pros and cons

The gold standard is to load blood cultures once collected onto a continuous monitoring blood culture machine with the minimum delay. This avoids the issues of missing certain organisms (see below), optimises speed of results and provides accurate 36 hour negative reports.

Although placement in a separate incubator might be expected to produce similar results to placement on a blood culture machine this is not the case. Time to positivity for *E. coli* is reduced by 20% by direct placement on the blood culture machine compared to a 37°C incubator. The reasons for the difference include; (a) agitation of the media by the blood culture machine which may result in white cell lysis with release of intracellular organisms and quicker temperature equilibration (b) the blood culture machine does not take readings for approximately 90 minutes after initial placement of the bottle on the machine in order to allow equilibration of hydrogen ion concentration which is temperature dependent. Thus time is wasted when a bottle is pre-incubated as even though the early phase of growth could be detected this will inevitably be delayed by 90 minutes before the machine can take the first reading.

Findings from the national telephone survey² conducted in 2013 were that NO laboratory in the sample (0/43 laboratories) loaded the blood culture machine out of hours.

- National audit data shows that there is approximately a 50-50 split between those laboratories that pre-incubate blood cultures prior to loading on the machine the next day and those that leave blood cultures at room temperature overnight².
- In hospitals where there is no on-site laboratory 75% leave the blood cultures at room temperature overnight.

Problems arising from pre-incubating /or leaving blood cultures at room temperature

- Pre-incubated blood culture bottles, containing primarily non-fermenting organisms such as *Pseudomonas aeruginosa*, *Acinetobacter* spp and some yeasts, that have gone through their growth cycle and entered the stationary or decline phase prior to placement on blood culture machines may fail to register as positive.
- Continuous monitoring blood culture machines use several algorithms for detecting the presence of organisms which are based around change in pH. For organisms that have gone through their growth phase and have entered the stationary or decline phase a threshold algorithm exists which looks at the overall change in pH. The amount of CO₂ produced by non-fermenting organisms and yeasts may be insufficient to activate the threshold algorithm and so they remain undetected (unlike most other organisms which will have produced large amounts of CO₂).
- Many laboratories abandoned pre-incubation due to the risk of missing these organisms (although they only account for a small percentage of positive cultures). Thus the time to detection for all positive blood cultures was significantly delayed– in

retrospect this would appear to be an unbalanced risk assessment in many cases to the impact on patient management.

- There is evidence to suggest that a delay in incubating the blood culture may have a larger effect on time to positivity beyond the period of delay prior to placement on the blood culture machine (Tom Lewis, personal communication). In their audit they found that the time to positivity was significantly longer (more than the delay) in blood cultures where there was a long interval as opposed to positive blood cultures which were placed on the blood culture machine with little delay. One explanation might be that organisms enter a dormant phase if they are not incubated within a specific timeframe.

Appendix 2

Improving performance

As stated, the greatest gains in speed of turnaround of blood cultures/impact on patient management are likely to come from:

- I. Minimising delays from collection to loading on the blood culture machine
- II. Processing blood cultures as they flag positive
- III. Performing direct antibiotic sensitivity tests (4% of laboratories do not)
- IV. Selective use of rapid identification/sensitivity tests

Patterns of laboratory working can impact hugely on speed of results / specimen turnaround time.

Laboratory services and the hospitals served vary immensely requiring different solutions. However progress in a number of areas should make the process easier and this includes;

- A. New developments in blood culture technology allow data on blood culture bottles placed on one blood culture machine to be transferred to another unit if the bottles are moved.
- B. Software developments allow remote signalling one a blood culture flags positive i.e. allowing placement for example of the blood culture machine in a centralised reception whilst notifying the staff in the microbiology laboratory of positive bottles.
- C. Shift systems in the blood sciences permitting multi-disciplinary working of blood cultures.
- D. Medical shift systems proving 24 hour cover, so if a result is available outside of routine hours there is someone to contact.

References

1. SMI B 37: Investigation of blood cultures (for organisms other than Mycobacterium species): Public Health England; 2014.
2. Lewis T, Sturgess I, Kavi J, Weinbren M. The results of three surveys (local, regional and national) of blood culture practice. *The Bulletin of The Royal College of Pathologists* 2015;171: 194-196.
3. Weinbren MJ, Borthwick MA. Rapid detection of extended-spectrum b-lactamase (ESBL)-producing organisms in blood culture: *Journal of Antimicrobial Chemotherapy* 2005; 55: 131–135

