

Chapter 30

Reporting Results

SIGNIFICANCE OF ISOLATES

Diagnostic microbiology laboratories attempt to provide data which allow clinicians to diagnose and manage infectious diseases. In order to achieve this, criteria of pathogenicity must be identified for particular organisms and when laboratory reports are issued these criteria must be applied to the particular patient circumstance.

Misleading information concerning pathogenicity may be conveyed because the relationship of particular isolates to disease is not clearly established and because the information conveyed from laboratory to clinician does not always indicate to the clinician the criteria of pathogenicity upon which the report was made.

Much bad medicine is perpetrated because of poor communication between clinician and laboratory. Firstly, the clinician must make available all relevant information to the laboratory. The clinician who sends a specimen to the laboratory requesting 'swab culture' and omitting any other information can scarcely expect useful bacteriology to be performed. It is manifestly impossible to subject every specimen submitted to every possible investigation or to completely speciate and determine antimicrobial susceptibilities of every isolate. It is also completely impossible to devise a routine which will not regularly permit inappropriate procedures to be performed and appropriate ones omitted unless each specimen is capable of being treated in full knowledge of the individual circumstances.

Errors regularly committed by laboratories (other than technical errors) are most commonly of four kinds: those due to lack of knowledge of what to look for when (eg., neglecting to look for acid-fast bacteria in a specimen from an infected breast prosthesis or discarding a *Streptococcus milleri* isolate from an abscess as a probable skin contaminant *Streptococcus viridans*); a too rigid, blind adherence to an established routine without taking sufficient note of individual circumstances (this includes neglecting the abnormal host and his special predilections for infections that may not occur in the normal individual, and ignoring the type and quality of the specimen); reporting virtually anything that grows and leaving it up to the clinician to decide on the significance (all too often the clinician is apt to think the laboratory must have reported an isolate because it was thought significant and treat it accordingly); and disregarding virtually all isolates except those established through long practice as important in disease. These categories do, of course, overlap. Basically, they are mainly due to ignorance, which hopefully this book will help to dispel. Sometimes they are also compounded by considerations of convenience or commercial interests.

It is necessary to distinguish clearly the circumstances under which an organism may be isolated from a specimen. Firstly, it may be a contaminant, ie, not actually present at the sampled site. This may be because of the intrinsic nature of the specimen or a completely extraneous event. Many specimens are by their nature easily contaminated by bacteria present at intervening (eg, sputum, blood cultures) or adjacent (eg, urines in females) sites. Good technique will minimise many of these but they can never be totally eliminated. Intelligent microscopic examination of specimens will identify most of these instances of contamination (and also many instances of extraneous contamination during collection or transport). An obviously contaminated specimen should just not be processed, since any information it provides will be completely misleading. Extraneous contamination within the laboratory can be largely eliminated by good quality control but will always occur from time to time even in the best laboratory. The microbiologist rather quickly learns to recognise the odd colony off the streak that is obviously an aerial contaminant, the plate contaminant that has been picked up and carried on the streak, and the odd organism that is suddenly appearing in cultures from a number of different specimens on a particular type of medium (or in a batch of stains).

It is, however, all too easy to dismiss the odd colony of a significant organism as a contaminant. In most cases of specimens with a normal flora, this may not be of grave importance, though by no means in all. However, in specimens taken from a normally sterile site, it may be extremely important. In most cases, any contamination will (if not completely extraneous) be skin flora; a single colony of, say, *Haemophilus influenzae* or *Streptococcus milleri* can never be dismissed as a contaminant under these circumstances. It needs to be remembered that organisms are frequently present in very small numbers in such specimens. This means also that they may not be seen in a Gram

stain; with a density of 100 organisms/mL (which may often be the case in meningitis), the chances of seeing the organism in a Gram stain are fairly low. On the other hand, if an organism is seen in a Gram stain in such a specimen, it is extremely unlikely to represent contamination.

Another possibility is that the organism is a transient, one that is adventitiously present at the site but is not capable of establishing itself at the site. The individual circumstances will suggest this possibility, a possibility that can best be established by repetitive cultures from the site.

With respect to microorganisms which are actually established at a particular site, it is important to distinguish between three possible conditions: colonisation, infection and disease. It is possible for a microorganism to colonise a biological site without directly affecting the activities of the host in any manner. This is the case with commensals, which make up the bulk of 'normal flora'. Although cases of true symbiosis between man and his resident flora are rare, such commensals frequently perform the very useful function of helping to prevent infection by more deleterious microorganisms. Commensals normally colonise only non-viable (usually terminally differentiated) cells.

An infection may be said to occur when a parasite is modifying the activities of the host in some way, though not necessarily in such a way, or to such an extent, as to cause disease. This usually implies a degree of invasion of viable cells and a greater turnover of involved host cells.

When the activities of a parasite are such that significant damage to host tissue is caused, disease ensues. This may be a direct effect of the parasite, an effect on some other element(s) of the flora which then produce deleterious effects, or mediated by the defence mechanisms of the host.

Which one of these conditions actually occurs depends on the invasive capabilities of the microbe versus the ability of the host to limit such invasion, and the nature and extent of adverse activities produced by the microbe versus the capacity of the host to nullify such activities or repair their effects.

How does all this translate into practice in the laboratory? I do not propose to give a short list of what to report when for all the different kinds of specimens, since it is impossible to include all the possibilities in such a list. The following guidelines should, however, provide at least some of the answers. Instances of obvious extraneous contamination are excluded in these guidelines.

All isolates from blood cultures should be speciated and their biograms and antibiograms recorded. All isolates of Gram negative bacteria and of fungi should be reported, as should all isolates of Gram positive bacteria except single isolates of coagulase negative staphylococci, *Bacillus*, *Corynebacterium* and *Propionibacterium acnes*. In any case, multiple isolates of the same species with the same biogram and antibiogram should be reported.

In the case of specimens from other normally sterile sites, any growth should be reported.

In specimens from sites with a normal flora, only organisms implicated as regularly causing disease at the particular site in the particular patient population represented by the individual should be reported. If it is not possible to obtain information about the patient, organisms potentially significant under certain circumstances should be reported together with an indication of these circumstances. There is a necessary proviso to this: unless there is clear evidence suggestive of an infection caused by this organism. This proviso is necessary because there must obviously occur cases of significant infection due to an organism not previously reported, or only rarely reported, as causing such infections. Our knowledge is not, and never will be, complete on such matters.

What constitutes clear evidence of an infection involving the organism (in a particular case)? Some years ago, the author proposed the following set of postulates of pathogenicity to be used both in the many cases where Koch's postulates are not applicable and in the instance of such 'private pathogens'. The organism must: (1) either be shown to be producing infection at the biological site in question or produce infection in a specific cell system replicating the conditions prevailing at the relevant site; and (2) either be shown to be producing effects which constitute, or can be quantitatively correlated with, the symptoms of the condition, or be shown to be capable, under the conditions prevailing at the site, of producing such effects; (3) evidence of a quantitative relationship between such effects and the activity of the organism must be obtained; (4) it must be demonstrated further that the organism is inhibited in its capacity for producing these effects by agents mitigating the symptoms of the condition; (5) presumed cause and effect through the sequence of events leading to the disease state must be shown to be temporally related.

All this, of course, is a little involved and, despite many years work, is not completely capable of realisation and especially not as a routine laboratory test.

At the present time, evidence of an infection involving a particular organism is usually best established by careful microscopic examination of the specimen. Evidence of infection may be provided by presence of excess numbers of leucocytes, especially non-viable leucocytes. This does not, of course, definitively establish that the suspect organism is responsible for the process, even if it is the only organism present, but it does at least establish an index of suspicion.

If a bacterium is intracellular, it is a pathogen. It may, however, be necessary to establish unequivocally that the organism is in fact intracellular. This can be done for phagocytes by using fluorescence and extracellular quenching, as in the method of Goldner et al, and for tissue by the use of Sowter and McGee's Gram stain.

It is important to realise that organisms which are normal flora at a site may yet be significant under certain conditions. For example, *Streptococcus agalactiae* is normal flora in the female genital tract. However, it can cause problems in post-operative patients and in patients with IUDs. It is also of importance in pregnant women, since it may be transmitted to the baby during birth and cause a potentially fatal infection. Again, *Staphylococcus aureus* is frequently present in low numbers in the female genital tract without causing problems but can be significant in post-operative and postpartum patients. It also represents a potential cause of toxic shock syndrome in females using tampons. Thus, there is some necessity for reporting both these organisms, but the conditions under which they may be significant should be indicated in the report.

URINE CULTURES: Relevant considerations in interpreting urine cultures include: viable bacterial count; whether culture is pure or mixed; cell type in urine microscopy (leucocytes, epithelial cells); presence or absence of bacterial inhibitors; patient's clinical history. The following general guidelines can be given.

A pure growth of an organism at $> 10^5$ organisms/mL represents probable UTI and the organism should be identified and susceptibility tests performed.

A pure growth of an organism at 10^4 - 10^5 organisms/mL indicates possible UTI and the organism should be identified and susceptibility tests performed.

The presence of 2 organisms in equal numbers at $> 10^5$ organisms/mL may indicate either UTI or faulty collection. Both organisms should be identified and susceptibility tests carried out on both.

The presence of 2 or 3 organisms at $> 10^5$ organisms/mL, with one organism clearly predominant indicates probable UTI caused by the predominant organism, which should be identified.

A mixture of 3 or more organisms in equal numbers at $> 10^5$ organisms/mL, or of 2 or more organisms at 10^4 - 10^5 organisms/mL should be reported as a mixed growth with no species predominating.

A pure growth of an organism at 10^3 - 10^4 organisms/mL from a symptomatic female or a male with prostatitis or from any patient with leucocytes $> 100/??L$ represents possible UTI and the organism should be identified and susceptibility tests performed.

A pure growth of an organism at 10^3 - 10^4 in an asymptomatic patient, or in the absence of leucocytes, and with bacterial inhibitors absent indicates no UTI and should be reported as no significant growth.

A pure growth of an organism at $> 10^3$ in the presence of a bacterial inhibitor may indicate treatment failure. Refer to any previous results, identify the organism and perform susceptibility tests.

All isolated organisms from a suprapubic collection, ureteric or in-out catheter (not indwelling catheter) should be identified and reported with susceptibilities and the colony count to the nearest hundred.