Practical Avian Micribiology

by
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Muller Hinton plates are used to determine antibiotic sensitivities. The green pigment of Pseudomonas is evident compared to the yellowish-white or cream colored growth of most other bacteria.

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The irridescent green on the EMB side of this Blood/EMB Bi-plate is an indicator of E. coli.

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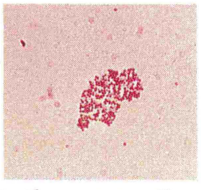

Most avian pathogens are Gram negative though Staph. aureus is Gram positive bacteria that can cause disease.

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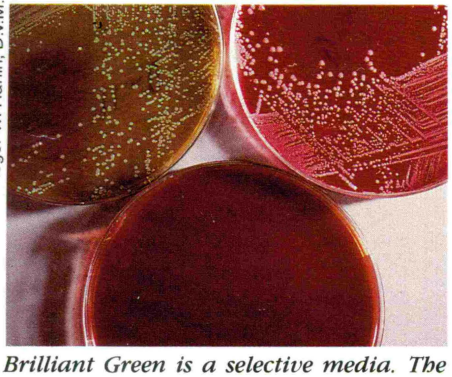

Brilliant Green is a selective media. The lower plate is uninnoculated but the yellow green growth of E. coli can be compared to the pinkish white growth of Salmonella on the other two plates.

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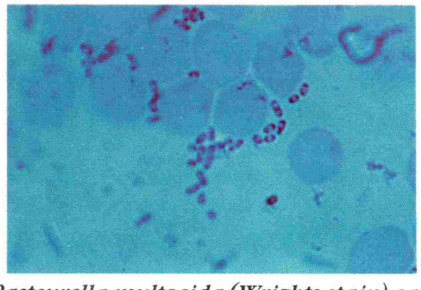

Pasteurella multocida (Wrights stain) can cause a rapidly fatal septicemic disease following animal bite wounds. A bacteriocidal antibiotic such as amoxicillin is often the drug of choice.

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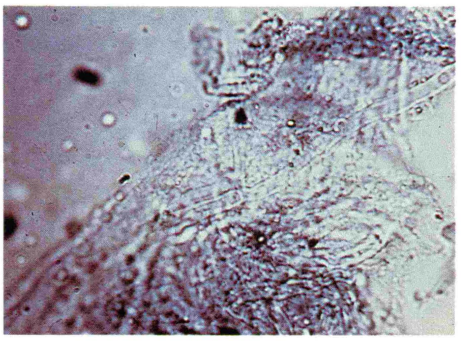

Aspergillus (exudate from an infected eye) is often the organism causing chronic illness following antibiotic therapy.

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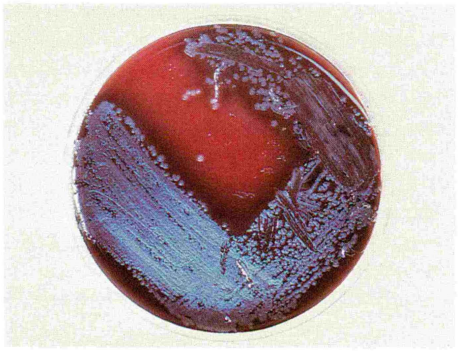

Growth of Pseudomonas on blood agar. This gram negative bacteria causes diseases that are often very difficult to treat.

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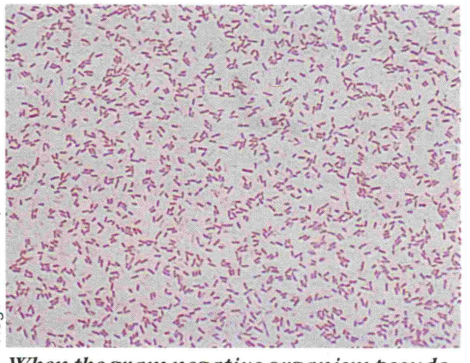

When the gram negative organism pseudomonas aeruginosa is isolated, antibiotic sensitivities should include gentomycin, carbenicillin (ticarcillin), spectionmycin, Tobramycin, Cephotaxime, and Amikacin as well as the more routinely used antibiotics.

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Proteus, a gram negative bacteria, is a potential pathogen. This photo shows its spreading swarming growth on EMB Agar.

A number of diagnostic procedures are available to practitioners that can greatly enhance their effectiveness with avian patients. One of these involves identifying organisms causing disease (pathogens) and their sensitivities to antibiotics and other theraputic agents by culture and microscopy. These techniques may also be of value to an aviculturist with a knowledge of basic microbiological technique and an understanding of normal flora and pathogenic organisms of birds.

Samples must be taken by sterile technique to avoid contaminate organisms. Their introduction into culture media may obscure pathogens giving misleading results.

Body discharges and lesions at the site of infection will often contain sufficient numbers of causitive organisms to yield a diagnosis of infectious disease. When cultures are taken on autopsy, the results may be valuable in preventing disease in flocks or other exposed birds. When taken in a clinically ill bird, for example a nasal discharge from a bird with upper respiratory disease, careful consideration must be taken of the various organisms that may be isolated. A sterile flush and/or a needle aspiration of a sinus would give a better indication of the organism involved in the disease than just rubbing a swab onto the discharge from the external nares.

Choanal swabs are valuable in respiratory illnesses and for routine screening of patients as are cloacal swabs in screening for potential intestinal pathogens. Sufficient research has been done concerning the normal flora of these two culture sites so the potential pathogenicity of the organisms isolated can be evaluated.

Difficulty in isolating bacteria from blood samples has been reported by avian practitioners. Special media are available for blood culture and may be of use in finding bacteria involved in

Continued on page 30

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septicemic disorders.

Samples are streaked onto Blood Agar, Levine EMB (Eosin Methylene Blue), and/or MacKonkeys Agar. Bi-Plates are available and are a little more economical but full plates better facilitate isolation of organisms. The samples are also innoculated into thioglycollate broth and placed on a microscope slide and gram stained. Actually visualizing the organisms may give the clinician valuable information, such as, the gram reaction of the organisms, the morphology and percentage of normal flora to abnormal, or even if non-bacterial agents may be involved.

The Thioglycollate Broth (Brain Heart infusion works well too) will propagate most organisms collected while the EMB and MacKonkeys Agar are selective media allowing only gram negative bacteria and only a certain few gram positive bacteria to grow. Group D Strep (Enterococcus) are exceptions but most gram positive organisms' growth is inhibited by MacKonkeys Agar and EMB. Gram positive bacteria as well as some yeasts and fungi will grow on Blood Agar. The ability of the organism to utilize blood (hemolysis) may be determined on Blood Agar plates.

After incubating overnight, the samples are ready to be examined. The vast majority of avian diseases are caused by gram negative organisms so the clinician will be most concerned with those, though hemolytic gram positive organisms and too many yeasts and fungi are usually not normal. If growth is insufficient, what was grown overnight in the Thioglycollate Tube may be streaked onto selective media and incubated another night. Some of the "more difficult to culture" organisms may be isolated by taking this extra step.

With little experience, a majority of the significant bacteria will be easily recognizable by the clinician. Most have characteristic colony morphology on different agars. E. coli causes a distinct iridescent green sheen on EMB while Klebsiella has a pink muciod colony. On EMB, lactose fermenters cause a purple coloration and on MacKonkeys they are dark pink. Non lactose fermenters grow in colorless to tan colored colonies on MacKonkeys and the purple color will be absent on EMB. Proteus may exhibit a swarming growth pattern over a plate while Pseudomonas has a characteristic appearance and smell. These are just a few examples as every species of microorganism will have its own characteristics. The most commonly isolated avian pathogens recovered in our clinic are: *Escerichia Coli., Klebsiella, Pseudomonas aeruginosa, Salmonella* sp., *Pasteurella multocida, Staphylococcus, Streptococcus, Candida albicans* and *Aspergillus fumigatus.*

Not all organisms will be easily identified. Advances in microbiological testing have made identification of those inobvious ones quite convenient. Our practice uses API strips (Analytical Products Index). This is a system that allows the microbiologist to do over 20 biochemical tests very simply to accurately identify organisms. Since the most common bacterial pathogens affecting avian patients are gram negative, the most frequently used strips are series 20E for *Enterobacteriacae,* though test strips are available for other families of organisms including Staphs, Streps, yeasts, and anerobes (bacteria that grow without the presence of air).

Once an organism has been determined to be a pathogen, its sensitivity to antimicrobial drugs should be determined. This information should be known before any antibiotics are administered, but clinical situations often force veterinarians to institute therapy before drug sensitivities are known. Disease processes in birds often progress at such a rate that waiting for time consuming lab work may be impractical when the condition of the patient is rapidly deteriorating.

In situations of acute bacterial disease it is imperative that the avian patient receive antibiotic therapy quickly. The clinician must evalute the patient and consider a number of factors in selecting a drug. Consideration must be given to: the toxicity of the drug, the route of administration, the mechanism and rate of action (bacterial or static), whether or not effective blood level and tissue distribution can be maintained, and undesirable side effects and federal regulations associated with the drug.

Not all diseases are infectious and antibiotics are not always indicated. Resistance to antibiotics is an ever increasing problem, and many antibiotics are becoming very ineffective. Though often helpful in arresting growth of pathogens, antibiotics have an effect on the normal flora of birds. Antibiotics may reduce the normal bacteria, thus increasing the chances of a pathogen to establish itself. Remember that it is difficult for a potential pathogen to become opportunistic and cause disease when the normal bacteria are flourishing, never giving the opportunist a chance. ●

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