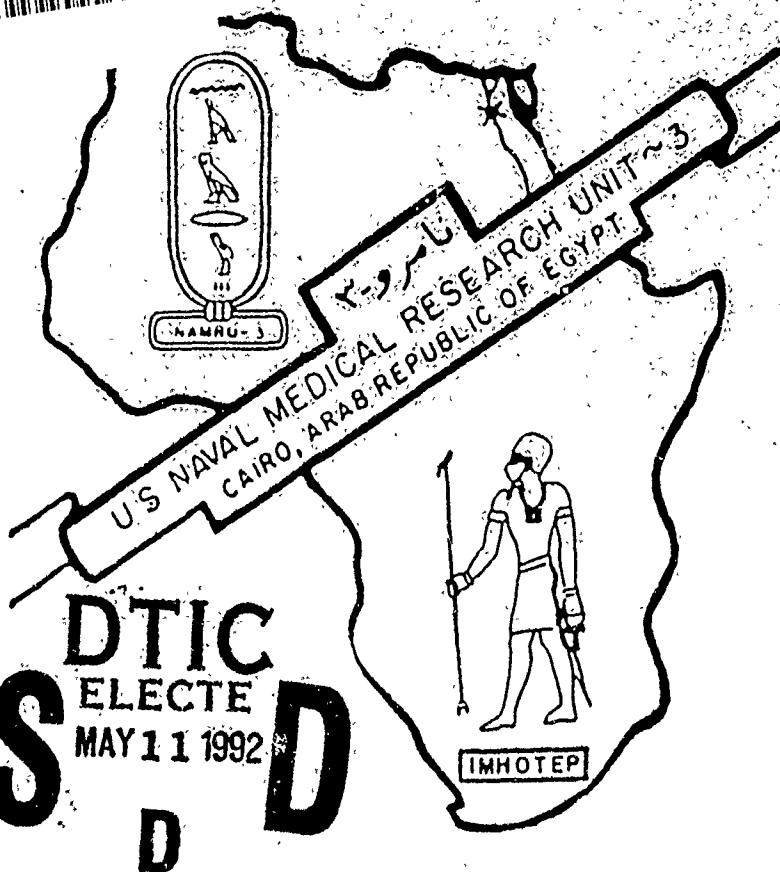


AD-A249 855



2



PUBLICATION REPORT

This document has been approved
for public release and sale; its
distribution is unlimited.

1639

6/91

THROAT CULTURE FROM PATIENTS WITH
MENINGOCOCCAL MENINGITIS

BY

J.E. Sippel and N.I. Girigs

U.S. NAVAL MEDICAL RESEARCH UNIT NO. 3
(CAIRO, ARAB REPUBLIC OF EGYPT)

PSC 452, BOX 5000
FPO AE 09835-0007

92-12362

Selective medium for isolating *Arcanobacterium haemolyticum*

Arcanobacterium haemolyticum is a facultatively anaerobic Gram positive bacillus previously known as *Corynebacterium haemolyticum*. It is most commonly isolated from the upper respiratory tract of patients with pharyngitis,^{1,2} but has also been isolated from skin lesions and occasionally from systemic infections. Isolation of *A haemolyticum* from healthy subjects is rare.³

The only medium previously described for the isolation of *A haemolyticum* is an enriched agar containing human blood or horse blood.³ After 48 hours of incubation on this medium *A haemolyticum* produces colonies which, characteristically, have a central pit and are surrounded by a zone of complete haemolysis. Despite the use of enriched media, isolation of *A haemolyticum* can be difficult as the organism is slow growing and is easily masked by commensal flora. The organism may therefore be a more common cause of pharyngitis than is currently recognised. We have developed a selective medium suitable for its isolation.

In a study of the antimicrobial susceptibilities of *A haemolyticum* we found that all 26 strains examined were resistant to mupirocin (minimum inhibitory concentrations > 128 mg/l). Mupirocin is highly active against commensal staphylococci and streptococci.⁴ Aztreonam and amphotericin B were used to inhibit the growth of Gram negative bacteria and yeasts, respectively. The complete medium consisted of a blood agar base (Oxoid No 2) containing 5% horse blood, 8 mg/l mupirocin (Beecham), 4 mg/l aztreonam (Squibb) and 1 mg/l amphotericin B (Sigma).

Strains from the National Collection of Type Cultures and clinical isolates of *A haemolyticum* grew well on the selective medium and produced characteristic colonies with narrow zones of complete haemolysis and a central pit.

The efficacy of this medium for the isolation of *A haemolyticum* from clinical specimens was investigated during February and March 1989. All throat swabs received by Chelmsford Public Health Laboratory were inoculated on to the selective medium and on to conventional horse blood agar. The inoculated media were incubated for 48 hours at 37°C in an anaerobic atmosphere contain-

ing 10% carbon dioxide. Both media were examined for characteristic colonies of *A haemolyticum*. Identification was confirmed biochemically.⁵

A haemolyticum was isolated from nine of 673 specimens (table). Isolation was much better with the selective medium as only two of the nine isolates were detected on conventional blood agar. The selective medium greatly reduced the growth of commensal organisms, thus permitting easier recognition of *A haemolyticum*. In the group aged 11–20 years the organism was isolated from eight (6.3%) of the 126 specimens. A similar specific age association has been noted by others.² The selective medium would therefore be of most value for the culture of throat swabs from teenagers or young adults. Lancefield group A streptococci were isolated from 18 (14.3%) of the 126 specimens from patients aged 11–20 years, so *A haemolyticum* seems to be a relatively important pathogen in this age group.

Erythromycin is the antibiotic of choice for treatment.¹ With the recent concern over erythromycin resistance in Lancefield group A streptococci,⁶ however, it may not be the initial choice for the empirical treatment of pharyngitis. Precise identification of the infecting organism would seem desirable. Use of a selective medium such as that described would clearly facilitate recognition.

NP BRENWALD
Clinical Microbiology and
Public Health Laboratory,
Addenbrookes Hospital,
Cambridge CB2 2QW

EL TEARE
LK MOUNTFORT
RE TEITMAR
Public Health Laboratory,
New Whittle Street
Chelmsford CM2 0YX

- 1 Selander B, Ljungh A. *Corynebacterium haemolyticum* as a cause of nonstreptococcal pharyngitis. *J Infect Dis* 1986;154:1011.
- 2 Bangk G, Nyman M. Tonsillitis and rash associated with *Corynebacterium haemolyticum*. *J Infect Dis* 1986;154:1037–10.
- 3 Clarridge JB. The recognition and significance of *Arcanobacterium haemolyticum*. *Clin Microbiol* 1989;11:41–5.
- 4 Sutherland R, Boon RJ, Griffin LE, et al. Antibacterial activity of Mupirocin (Pseudomonas acid), a new antibiotic for topical use. *Antimicrob Agents Chemother* 1985;27:195–8.
- 5 Jarvinen H, Nissinen A, Huovinen P, et al. Erythromycin resistance in group A streptococci. *Lancet* 1989;1:1022–3.

Isolation of *Arcanobacterium haemolyticum* from throat swabs of patients in various age ranges

Age range (years)	Number of specimens	Number of isolates	
		Blood agar	Selective medium
0–10	215	0	0
11–20	126	2	8
21–30	92	0	1
> 31	175	0	0
Unknown	65	0	0
Total	673	2	9

Colorimetric determination of human albumin

Many methods, based on a variety of principles, have been described for the measurement of human albumin. Increases in serum albumin are almost exclusively due to dehydration. Decreases are seen in patients

protein intake seen in malnutrition and (iv) gastrointestinal disorders with malabsorption, vomiting, or diarrhoea (before dehydration).

Methods for the quantitative measurement of serum or plasma albumin fall into four categories—namely, salt fractionation, electrophoresis, dye-binding and immuno-

automated, inexpensive, simple and give reproducible results. Data from the United Kingdom External Quality Assessment Scheme for General Clinical Chemistry indicated that 98% of participating laboratories measure serum albumin by either bromocresol green (BCG) or bromocresol purple (BCP) dye-binding methods, of these, BCG methods remain the most widely used. A lack of specificity of BCG for albumin, however, has led the International Federation of Clinical Chemistry Expert Panel on Proteins¹ and other authors^{2,3} to recommend that the method should only be used for screening purposes. It has been reported that not only does BCG overestimate low albumin concentrations, but that it also underestimates concentrations in the high normal range.²

The following assay (patent-pending) uses the specificity of the reduction of BSPT (2-(2'-benzothiazolyl)-5-styryl-3-phthal-hydrazidyl)-tetrazolium chloride to its coloured formazan, in the presence of a reducing agent, electron carrier, and human albumin and benefits from simple colorimetric detection.

To 50 µl serum add 1 ml buffered colour reagent containing BSPT, methoxy-N-methyl phenazinium methyl sulphate, and dithio-threitol. After incubation for two minutes at room temperature the absorbance is read at 590 nm.

The procedure is linear over the albumin range 1–80 mg/ml. No interference was found with transferrin, bilirubin, or heparin and the method compared well with traditional dye-binding and immunological assays.

RJ HINTON
BS MALLON
KD MORRIS
J MILLER
T ATKINSON
PM HAMMOND
Diagnostic Enzymology Group,
Division of Biotechnology
PHLS Centre for Applied Microbiology and
Research,
Porton Down, Salisbury
CP PRICE
Department of Clinical Biochemistry,
London Hospital Medical College,
Turner Street, London E1 2AD

- 1 International Federation of Clinical Chemistry Committee on standards and expert panel news. *Newsletter* 1976;13:5.
- 2 Webster D, Bignell AHC, Attwood EC. An assessment of the suitability of bromocresol green for the determination of serum albumin. *Clin Chim Acta* 1981;53:101–8.
- 3 Ferreira P, Price CP. A comparison of bromocresol green and immunoprecipitation methods for the determination of serum albumin. *Clin Chim Acta* 1971;55:259–62.
- 4 Slater L, Carter PM, Hubbs JR. Measurement of albumin in the sera of patients. *Ann Clin Biochem* 1975;12:33–40.

MATTERS ARISING

Throat culture from patients with meningococcal meningitis

Cartwright and Jones suggest that throat culture can be useful in the diagnosis of meningococcal meningitis when the patient

light of the inability of most of the antibiotics that are used for the treatment of meningococcal infections to serve as prophylactic agents. We performed throat cultures on various populations in Cairo, Egypt,² where group A meningococcal disease is endemic. Most cases occur in school-age children, a population that we found had a 3.8% carrier rate. Only one of the 58 patients positive by culture of cerebrospinal fluid for agents other than *Neisseria* was a group A meningococcal carrier. Group A meningococci, however, were isolated from 55% of 380 patients who were culture positive for this organism and from 30% of 46 patients who were culture negative but shown to have meningococcal meningitis by stain or detection of specific antigen in cerebrospinal fluid.

We therefore concur that culture of patients' throats can contribute to laboratory diagnosis. Jewes *et al* argued that culturing the throats of contacts was not useful for diagnosis due to a lack of correlation in serotype between isolates from contacts and index cases.¹ We found that the rate of group A carriage in the contacts of group A patients (15%)² as four times that in school children, suggesting that monitoring this population could also be helpful in diagnosis of cases.

JE SIPPEL

*Mercer University, School of Medicine,
Macon, Georgia 31207*

NI GIRGIS

*Naval Medical Research Unit No 3,
c/o US Embassy, Cairo, Egypt*

- 1 Cartwright KAV, Jones DM. Investigation of meningococcal disease. *J Clin Pathol* 1989; 42:634-9.
- 2 Sippel JE, Girgis NI. Meningococcal infection in Egypt. Laboratory findings in meningitis patients and their prevalence of pharyngeal infection in patients and contacts. *Am J Trop Med Hyg* 1978; 27:980-5.
- 3 Jewes L, Norman P, McKendrick MW. Value of throat swabs in meningococcal meningitis. *J Clin Pathol* 1989; 42:1229.

Lamina propria mast cells in ulcerative colitis

We were interested to see the response from Dr Crow to our paper on mast cells and eosinophils in Asian and Caucasian patients with ulcerative colitis¹ and would agree that formalin fixed material is not ideally suited to demonstration of mast cells. In our experience, however, carefully controlled use of the Astra blue technique is at least generally acceptable in this context.^{2,3} It should, of course, be remembered that our study was of a comparative rather than absolute enumerative type and the probable lowering of counts for both groups would therefore still clearly show differences between them as there is no reason to presume that staining would differ between the groups.

It might be of interest to run other techniques on our tissues, and if time permits we will consider this.

GFA BENFIELD

KL BRYAN

J CROCKER

*Departments of Respiratory Medicine
and Histopathology,
East Birmingham Hospital
Bordesley Green East,
Birmingham B9 5SS*

Received GFA, Bryan, Crocker, J. Letter on mast cells and mast cells in ulcerative colitis. *Journal of Clinical Pathology*.

- 2 Crocker J, Smith PJ. A quantitative study of mast cells in Hodgkin's disease. *J Clin Pathol* 1984; 37:519-22.
- 3 Crocker J, Smith PJ. Mast cells in non-Hodgkin's lymphomas: a quantitative study. *J Clin Pathol* 1987; 40:470.
- 4 Jackson N, Burt D, Crocker J, Boughon B. Skin mast cells in polycythaemia vera: relationship to the pathogenesis and treatment of pruritus. *Br J Dermatol* 1987; 116:21-9.

Dr Crow comments

Benfield *et al* found that there was no significant difference in the numbers of rectal mucosal mast cells between groups of Asian and Caucasian patients with ulcerative colitis. Unfortunately, the Astra blue technique¹ used to stain the mast cells in this case would seriously underestimate the numbers of such cells in intestinal mucosa fixed in formal-saline² and any differences which might be present would be masked. If there is only formalin fixed material available for study then the long (five to seven day) toluidine blue or trypan toluidine blue techniques³ will, at least partly overcome the blockage to staining induced by formalin and will give a more realistic count. Evidence from other tissues, however,⁴ suggests that fixation in basic lead acetate, isotonic formal acetic acid, or Carnoy's fixative followed by long toluidine blue staining will show up even more mast cells and hence even this staining technique must be regarded as doubtful in formalin-fixed tissue, unless it has been validated against one of the mast cell fixatives mentioned, for the tissue in question.

- 1 Blaes DM, Williams JF. A simplified method for staining mast cells with astra blue. *Stain Technol* 1981; 56:91-4.
- 2 Strobel S, Miller HRP, Ferguson A. Human intestinal mast cells: evaluation of fixation and staining techniques. *J Clin Pathol* 1987; 34:851-8.
- 3 Wingren U, Enerback L. Mucosal mast cells of the rat intestine: re-evaluation of fixation and staining properties, with special reference to protein blocking and solubility of the granular glycosaminoglycan. *Histochem J* 1983; 15:571-82.
- 4 Crow J, More L, Lowe S. The mast cells of the human uterus. *JPAHS* 1988; 96:921-6.

BOOK REVIEWS

Essential Histopathology. PR Millard (Pp 326, £18.50.) Blackwell Scientific Publications 1989 ISBN 0-632-02238-8

Peter Millard's book is eye-catching and has a text clearly laid out and supported by largely excellent photographs with splendid diagrams and line drawings. Many pathology books present the subject in a too detailed and boring manner but this is clearly not Millard's style. The first impression, therefore, is that this book is clearly going to be a hit. Reluctantly, after using it for several

Dr Millard has attempted to present the histopathology an undergraduate requires without unduly overloading him and he has been rightly selective and brief. Sometimes he succeeds in presenting a lucid picture of his target - for example, diabetes mellitus. At other times his brevity fails as in his attempt to unify the malignancies in the gut. He omits any account of bone and joint pathology yet presents two chapters on tissue responses and on tumours. Although these are elegantly illustrated, they are too superficial to be of value to final students and in any case fit better into a general pathology text book.

I hope my students read this book but only as a supplement after buying a larger text which puts more emphasis on mechanisms rather than appearances of disease. The attractive format and the relatively few pages (235) of essential histopathology may well seduce students into buying it at its relatively modest price. Only when they get it home will they find that Dr Millard's publishers have let him down with no less than eight incorrectly printed figures. In the longer term as the examination looms its other deficits will make themselves felt.

"Hit" or "miss", it all depends where you judge the bull to be. My criticisms may reflect not Millard's aim but where he judges the target. With a bit of retargeting, the style and presentation of this book could well make a winner in future editions. At any rate it is a good attempt at presenting pathology in a vital manner which will catch the student's eye, and as such deserves applause.

G SLAVIN

Pathology of the Stomach and Duodenum. H Rotterdam, HT Enterline. (Pp 320; DM 248.) Springer 1989 ISBN 0-510-96823-7.

This book, by two experienced American gastrointestinal pathologists, sets out to offer information on all aspects of gastric and duodenal disease including historical, epidemiological, clinical and pathophysiological data, with the emphasis on diagnostic gross and microscopic pathology. The coupling of stomach with duodenum was decided because of the common pathophysiology of some gastric and duodenal diseases, such as peptic ulcer disease. The exclusion of oesophageal disease seems somewhat arbitrary, therefore, as the principle of common pathophysiology would also seem to apply. The participation by one of the authors in a previously published monograph on this subject is the probable explanation.

The book succeeds in some of its aims and in particular the chapters on anomalies, hyperplasias, and benign epithelial tumours and carcinoid (neuroendocrine) tumours were very good and well referenced. On the debit side there was little current information on *Campylobacter pylori* and the discussion of malignant lymphomas was largely on the basis of the Rappaport classification. There were a number of typographical errors and the quality of many of the illustrations, particularly the photomicrographs, was poor.

In summary, while good in parts, this book does not stand out in a competitive market.

REPORT DOCUMENTATION PAGE

1a REPORT SECURITY CLASSIFICATION UNCLASSIFIED			1b RESTRICTIVE MARKINGS		
2a SECURITY CLASSIFICATION AUTHORITY			3 DISTRIBUTION/AVAILABILITY OF REPORT Approved for public release; Distribution is unlimited.		
2b DECLASSIFICATION/DOWNGRADING SCHEDULE					
4 PERFORMING ORGANIZATION REPORT NUMBER(S) 6/91			5 MONITORING ORGANIZATION REPORT NUMBER(S)		
6a NAME OF PERFORMING ORGANIZATION U.S. Naval Medical Research Unit No. 3		6b OFFICE SYMBOL (If applicable) NAVMEDRSCHU THREE	7a NAME OF MONITORING ORGANIZATION		
6c ADDRESS (City, State, and ZIP Code) PSC 452, Box 5000 FPO, AE 09835-0007			7b ADDRESS (City, State, and ZIP Code)		
8a NAME OF FUNDING SPONSORING ORGANIZATION Naval Medical Research and Development Command		8b OFFICE SYMBOL (If applicable) NAVMEDRSCH DEVCOM	9 PROCUREMENT INSTRUMENT IDENTIFICATION NUMBER		
8c ADDRESS (City, State, and ZIP Code) National Naval Medical Center Building 1, Tower 12 Bethesda, MD 20889-5044			10 SOURCE OF FUNDING NUMBERS PROGRAM ELEMENT NO 64758A		
			PROJECT NO 3M4647- 58D849	TASK NO BH	WORK UNIT ACCESSION NO
11 TITLE (Include Security Classification) Throat Culture from Patients with Meningococcal Meningitis. (UNCLASSIFIED).					
12 PERSONAL AUTHOR(S) Sippel, J E.* and Girgis, N.I. * Mercer University, School of Medicine, Macon, Georgia.					
13a TYPE OF REPORT		13b TIME COVERED FROM TO		14 DATE OF REPORT (Year, Month, Day) 1990	
				15 PAGE COUNT 2	
16 SUPPLEMENTARY NOTATION Published in: J. Clin. Pathol., 43(7):610-611, 1990; Acc. No. 1639.					
17 COSATI CODES FIELD GROUP SUB-GROUP			18 SUBJECT TERMS (Continue on reverse if necessary and identify by block number) meningococcal meningitis; Throat culture; Patients; Cairo, Egypt.		
19 ABSTRACT (Continue on reverse if necessary, and identify by block number) As per attached.					
20 DISTRIBUTION AVAILABILITY STATEMENT ☒ UNCLASSIFIED UNLIMITED ☐ ADVANCED ☐ DTIC USERS			21 ABSTRACT SECURITY CLASSIFICATION UNCLASSIFIED		
22a NAME OF RESPONSIBLE PERSON Research Publications Branch			22b TELEPHONE (Include Area Code) 202-284-1381		22c OFFICE SYMBOL R.P.B