

Blood Culture and Conventional Media for Vitreous Culture in Infectious Endophthalmitis

Yosanan Yospaiboon, MD*,
Sarawuth Saree, MD*, Sirichai Pasadhika, MD**

* Department of Ophthalmology, Faculty of Medicine, Khon Kaen University, Khon Kaen

** Department of Ophthalmology, Faculty of Medicine, Chulalongkorn University

Objective: To study culture of vitreous fluid specimens in patients with infectious endophthalmitis, using blood culture bottles compared with conventional culture media.

Material and Method: Patients with infectious endophthalmitis, occurred within 6 weeks after ocular trauma or intraocular surgery, were prospectively studied. Vitreous fluid specimens were cultured in both blood culture bottles and conventional culture media. The measured outcome is the yield of positive culture and time to positive culture.

Results: The vitreous fluid culture was positive in 14 of 27 eyes (51.85%). Blood culture bottle was positive in 14 of 14 eyes (100%), whereas conventional culture media was positive in 7 of 14 eyes (50%). Most specimens in both techniques showed positive culture within 24 hours.

Conclusion: Vitreous fluid culture with blood culture bottles is superior to conventional media with statistically significant difference. There was no significant difference in time to positive culture.

Keywords: Infectious endophthalmitis, Blood culture bottle, Conventional culture media

J Med Assoc Thai 2005; 88(5): 639-42

Full text. e-Journal: <http://www.medassocthai.org/journal>

Infectious endophthalmitis is a rare, but severe complication occurring after intraocular surgery or penetrating ocular trauma. Occasionally it may result from hematogenous spread of organisms to the eyes. Successful management of this condition depends on prompt diagnosis and treatment with appropriate antimicrobial therapy. Identification of the responsible microorganisms is, therefore, essential for successful management. Microbiological investigation of vitreous fluid and aqueous humor specimens is the only method that permits reliable identification of the causative microorganisms. In the literature, positive cultures have been reported in 35-85% of the cases^(1,2). The low yield rate may be attributed to the limited amount of the fluid specimen obtained. There were many reports of increasing positive culture results in a variety of fluid specimens such as joint fluid^(3,4), pleural fluid⁽⁵⁾, pancreatic pseudocyst⁽⁶⁾ and ascitic

fluid⁽⁷⁻⁹⁾ by using blood culture media. To the best of the authors' knowledge, there has been no report on comparison between blood culture media and conventional media for culture of vitreous fluid specimens from patients with infectious endophthalmitis. The authors hypothesize that blood culture media yields a more positive result than conventional media and the present study is, therefore, conducted to compare blood culture bottles with conventional media for vitreous culture in such patients.

Material and Method

The present study complied with the International Conference for Harmonization Guideline for Good Clinical Practice (ICH-GCP) and was approved by the Khon Kaen University Ethics Committee. All patients with infectious endophthalmitis at Srinagarind Hospital, Faculty of Medicine, Khon Kaen University were eligible for the study if they had best-corrected visual acuity of 20/50 or worse, symptoms and signs of endophthalmitis no longer than three weeks, history of intraocular injury or surgery no longer than six weeks

Correspondence to : Yospaiboon Y. Department of Ophthalmology, Faculty of Medicine, Khon Kaen University, Khon Kaen 40002, Thailand. Phone & Fax: 0-4334-8383, E-mail: yosanan@kku.ac.th

and signed the written informed consent form to the study. The patients who had a history of uveitis or using eyedrops which might interfere with blood ocular barrier within one week were excluded from the present study.

Microbiological investigation of vitreous fluid was done in all patients. The specimens from vitreous tapping were inoculated into both conventional media and blood culture media. For conventional culture, vitreous fluid specimens were inoculated onto blood agar, chocolate agar, Sabouraud dextrose agar and thioglycolate broth. Within 30 minutes, these media were sent to the Clinical Microbiology Laboratory where plates were inoculated and examined for identification of the causative microorganisms. For blood culture system, the specimens were inoculated into blood culture bottles containing brain-heart infusion media (Bac T/ALERT, Organon Teknika) and sent to the Clinical Microbiology Laboratory within 30 minutes. Growth indices changed in color at the bottom of the blood culture bottles. Positive cultures yielded in both blood culture system and conventional culture were compared. McNemar Chi Square was used for statistical analysis. The significant difference was considered when P value < 0.05.

Results

Twenty seven patients were recruited in the present study. There were 18 males and 9 females. Age ranged from 6-78 years of age. Symptoms prior to admission were between 2 and 21 days. Sixteen of 27 patients (59.26%) had a history of ocular injury, and 11 of 27 patients (40.74%) had a history of ocular surgery within 6 weeks prior to admission.

Significant micro-organisms were recovered from 14 of 27 eyes (51.85%) (Table 1). Blood culture bottles obtained positive culture in all fourteen eyes (100%), whereas only seven eyes (50%) yielded a positive result in conventional media (p = 0.008)

Most causative microorganisms were detected by blood culture system within 24 to 48 hours, and 8 of 14 specimens showed a positive result within 24 hours. Conventional cultures also yielded visible growth within 24 hours.

Discussion

Successful management of infectious endophthalmitis depends on timely diagnosis and institution of appropriate antimicrobial therapy. Microbiological investigation of intraocular fluid is the only method that permits reliable identification of

Table 1. Microorganisms recovered in vitreous fluid cultures

No.	Microorganisms	Blood culture	Conventional culture
1	<i>Staphylococcus spp.</i>	+	NG
2	<i>Staphylococcus spp.</i>	+	+
3	<i>Staphylococcus coagulase neg</i>	+	+
	<i>Enterobacter spp.</i>	+	+
4	<i>Streptococcus pneumoniae</i>	+	NG
5	<i>Streptococcus pneumoniae</i>	+	+
6	<i>Streptococcus non A, B, D</i>	+	+
7	<i>Pseudomonas aeruginosa</i>	+	NG
8	<i>Pseudomonas aeruginosa</i>	+	+
9	<i>Klebsiella pneumoniae</i>	+	NG
10	<i>Klebsiella pneumoniae</i>	+	NG
	<i>Escherichia coli</i>	+	NG
11	<i>Escherichia coli</i>	+	+
12	<i>Stenotrophomonas maltiphilia</i>	+	+
13	<i>Alcaligenase spp.</i>	+	NG
14	<i>Fusarium spp.</i>	+	NG

+ = positive culture NG = no growth

the causative microorganisms. Initial smear examination and cytology have limited roles in the diagnosis since they are consistent with the culture results in only half of the cases^(10,11).

Some authors advocate the use of polymerase chain reaction in the microbiological determination of the bacteria and fungus in intraocular fluid and reported the yield of 91-100%⁽¹²⁻¹⁵⁾. Although it is a specific and sensitive method in the diagnosis, it has high cost, sophisticated process and may result in false positive reaction in some cases⁽¹⁶⁾. Use of electron microscopy and immunocytochemistry also offers an alternative means of identification, but these approaches have drawbacks principally associated with interpretation⁽¹⁷⁾. Some authors propose the use of fluorescent Gram stain in the microbiological diagnosis of infectious endophthalmitis⁽¹⁸⁾. However, the low sensitivity, quenching of fluorescence and change in Gram reaction presently preclude the use of this method as a diagnostic tool.

Even when these modern microbiological techniques are used, intraocular fluid culture is still used as a gold standard for the diagnosis. Previous studies reported culture to yield positive results in 35-85%^(1,2). In an attempt to improve the recovery of microorganisms from fluid specimens, the blood culture system has been reported to have increasing positive culture results in a variety of fluid specimens⁽³⁻⁹⁾. For intraocular fluid, blood culture bottles also yielded a high incidence of positive culture⁽¹⁹⁾. In the present study, blood culture bottles became posi-

tive for all microorganisms obtained and were found to be superior to the conventional media. However, the yield for positive culture in the present study was 51.85% which is rather low when compared to the previous studies. This may be attributed to the limited amount of vitreous fluid specimens. Only 0.1-0.2 ml of vitreous fluid was obtained from vitreous tapping. Furthermore, some patients may receive antimicrobial therapy before cultures of vitreous fluid are obtained, or some cases may be caused by microorganisms that are not detected by currently available techniques.

In the present study, only undiluted vitreous fluid was obtained for microbiological culture because previous studies revealed that vitreous fluid was more sensitive than aqueous humor in making a diagnosis^(11,17,20), and the use of a membrane filter to concentrate the vitreous samples obtained at vitrectomy increased further diagnostic yield⁽¹¹⁾. However, obtaining culture of both an undiluted vitreous samples and the vitrectomy membrane filter had a significant advantage compared with culture of only one sample⁽¹⁰⁾.

The reasons for the superiority shown by the blood culture system over conventional cultures for the recovery of microorganisms are not clear. It may be attributed to a volume-related effect and dilutional effect⁽⁴⁾. Only a small volume of fluid can be plated on solid media while more volume can be inoculated into a blood culture bottle. However, a limited amount of intraocular fluid cannot explain the superiority by this volume-related effect. In addition, it is postulated that purulent fluid exerts an inhibitory effect upon the organism⁽⁴⁾. Dilution of this detrimental factor in a large volume of blood culture media below their inhibitory concentration substantially enhances the chances of recovery.

In conclusion, the blood culture system is superior to conventional culture of vitreous fluid specimens from patients with infectious endophthalmitis. Direct inoculation of vitreous fluid into a blood culture bottle should be recommended as an acceptable adjunct or alternative to conventional solid plates and broth for microbiological diagnosis.

Acknowledgement

This study was supported by invitation research grant (I-43205) from the Faculty of Medicine, Khon Kaen University.

References

1. Ness T, Pelz K. Endophthalmitis: improvement of culture results. *Ophthalmology* 2000; 97: 33-7.
2. Forster PK, Abbot RL, Gelender H. Management of infectious endophthalmitis. *Ophthalmology* 1980; 87: 313-9.
3. von Essen R. Culture of joint specimens in bacterial arthritis: impact of blood culture bottle utilization. *Scand J Rheumatol* 1997; 26: 293-300.
4. Yagupsky P, Dagan R, Howard CW, Einhorn M, Kassis I, Simu A. High prevalence of *Kingella Kingae* in joint fluid from children with septic arthritis revealed by the BACTEC blood culture system. *J Clin Microbiol* 1992; 30: 1278-81.
5. Xioli X, Castellvi JM, Guardiola J, Sese E, Castellote J, Perello A, et al. Spontaneous bacterial empyema in cirrhotic patient: a prospective study. *Hepatology* 1996; 23: 719-23.
6. Ljubicic N, Bilic A. Inflamed pancreatic pseudocyst: optimization of pseudocyst fluid culture technique. *J Gastroenterol* 1993; 31: 198-200.
7. Blondeau JM, Pylypchuk GB, Kappel JE, Pilkey B, Lawler C. Comparison of bedside- and laboratory-inoculated BACTEC high-and low-volume resin bottle for the recovery of microorganism causing peritonitis in CAPD patients. *Diagn Microbiol Infect Dis* 1998; 31: 281-7.
8. Singh N, Rihs JD, Gayowski T, Miele L, Yu VL. Improved detection of spontaneous bacterial peritonitis with BACTEC as compared with conventional culture methods. A prospective study. *Diagn Microbiol Infect Dis* 1994; 19: 1-4.
9. Rayner BL, Williams DS, Oliver S. Inoculation of peritoneal dialysate fluid into BCB improves culture rates. *S Afr Med J* 1993; 81: 42-3.
10. Sharma S, Jalali S, Adiraju MV, Gopinathan U, Das T. Sensitivity and predictability of vitreous cytology, biopsy, and membrane filter culture in endophthalmitis. *Retina* 1996; 16: 525-9.
11. Forster PK. Etiology and diagnosis of bacterial post-operative endophthalmitis. *Ophthalmology* 1978; 85: 320-6.
12. Anand AR, Madhavan HN, Therese KL. Use of polymerase chain reaction (PCR) and DNA probe hybridization to determine the Gram reaction of the infecting bacterium in the intraocular fluids of patients with endophthalmitis. *J Infect* 2000; 41: 221-6.
13. Hidalgo JA, Alangaden GJ, Elliott D, Akins RA, Puklin J, Abrams G, et al. Fungal endophthalmitis diagnosis by detection of *Candida albicans* DNA in intraocular fluid by use of a species-specific polymerase chain reaction assay. *J Infect Dis* 2000; 181: 1198-201.
14. Therese KL, Anand AR, Madhavan HN. Polymerase chain reaction in the diagnosis of bacterial endophthalmitis. *Br J Ophthalmol* 1998; 82: 1078-82.
15. Lohmann CP, Heeb M, Linde HJ, Gabel VP, Reischl U. Diagnosis of infectious endophthalmitis after cataract surgery by polymerase chain reaction. *J Cataract Refract Surg* 1998; 24: 821-6.

16. Ginesu F, Pirina P, Sechi LA, Mollicotti P, Santoru L, Porcu L, et al. Microbiological diagnosis of tuberculosis: a comparison of old and new methods. *J Chemother* 1998; 10: 295-300.
17. Kinnear FB, Kirkness CM. Advances in rapid laboratory diagnosis of infectious endophthalmitis. *J Hosp Infect* 1995; 30: 253-61.
18. Roychoudhury B, Sharma S, Reddy MK, Das T. Fluorescent Gram stain in the microbiological diagnosis of infectious endophthalmitis. *Curr Eye Res* 1997; 16: 620-3.
19. Joondeph BC, Flynn HW Jr, Miller D, Joondeph HC. A new culture method for infectious endophthalmitis. *Arch Ophthalmol* 1989; 107: 1334-7.
20. Koul S, Philipson A, Arvidson S. Role of aqueous and vitreous cultures in diagnosing infectious endophthalmitis in rabbits. *Acta ophthalmol* 1990; 68: 466-9.

ขวดเพาะเชื้อในเลือดและวุ้นเพาะเชื้อแบบเดิมสำหรับการเพาะเชื้อจากวุ้นตาในผู้ป่วยตาติดเชื้อ

ยศอนันต์ ยศไพบูลย์, สรวุฑ สารีย์, สิริชัย ปาสาทิก

วัตถุประสงค์: เพื่อศึกษาการเพาะเชื้อจากน้ำวุ้นตาในผู้ป่วยติดเชื้อในลูกตา (*Infectious endophthalmitis*) โดยใช้ขวดเพาะเชื้อในเลือดเปรียบเทียบกับกรเพาะเชื้อด้วยวุ้นเพาะเชื้อแบบเดิม

วัสดุและวิธีการ: ศึกษาในผู้ป่วยตาติดเชื้อที่เกิดภายใน 6 สัปดาห์ หลังจากเกิดอุบัติเหตุที่ตา หรือหลังการผ่าตัดตา เก็บตัวอย่างส่งตรวจจากน้ำวุ้นตา แยกเพาะเชื้อทั้งในขวดเพาะเชื้อในเลือด และวุ้นเชื้อแบบเดิม การวัดผลดูจากจำนวนตัวอย่างส่งตรวจที่ให้ผลบวก และระยะเวลาที่ใช้ในการเพาะเชื้อจนให้ผลบวก

ผลการศึกษา: ศึกษาในผู้ป่วยตาติดเชื้อ จำนวน 27 ตา พบว่าเพาะเชื้อจากตัวอย่างที่ส่งตรวจให้ผลบวก 14 ตา (ร้อยละ 51.85) เมื่อเปรียบเทียบระหว่างการเพาะเชื้อด้วยขวดเพาะเชื้อในเลือด กับการเพาะเชื้อด้วยวุ้นเพาะเชื้อแบบเดิม พบว่าขวดเพาะเชื้อในเลือดให้ผลบวก 14 ใน 14 ตา (ร้อยละ 100) และวุ้นเพาะเชื้อแบบเดิมให้ผลบวก 7 ใน 14 ตา (ร้อยละ 50) ระยะเวลาที่ใช้ในการเพาะเชื้อจนให้ผลบวกของทั้ง 2 วิธีไม่แตกต่างกัน

สรุป: การเพาะเชื้อโดยใช้ขวดเพาะเชื้อในเลือด ให้ผลบวกดีกว่าการเพาะเชื้อโดยใช้วุ้นเพาะเชื้อแบบเดิม อย่างมีนัยสำคัญทางสถิติ เมื่อผลการเพาะเชื้อให้ผลบวกทั้ง 2 วิธี ระยะเวลาที่ใช้ในการเพาะเชื้อจนให้ผลบวก ไม่แตกต่างกัน
