



The Effects of Changing Needles on Reducing Contamination of Postmortem Hemocultures

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Background: Sepsis is a common cause of death and can be diagnosed through postmortem hemoculture. However, this method carries the risk of contamination. Some experts have argued that using a new needle before inoculation can help reduce the contamination rates.

Objective: To evaluate the contamination rate from postmortem hemoculture in needle-changing and no-needle-changing groups.

Methods: The present study analyzed postmortem hemoculture results from autopsy cases at Ramathibodi Hospital. Forty deceased individuals who had died within 24 hours and were not suspected of having an infection were included in the study. The blood samples were divided into 2 groups: one in which the needle was changed before inoculation and the other in which it was not. Differences in hemoculture results between the 2 methods were examined.

Results: No statistically significant difference was detected in the positive culture rate between the needle-changing (57.5%) and non-needle-changing groups (57.5%) ($P = 1.00$). The Kappa coefficient was 0.795. Viridans group streptococci, *Escherichia coli*, *Klebsiella pneumoniae* complex, *Enterobacter cloacae* complex, and *Acinetobacter baumannii* complex were the most commonly cultured bacteria.

Conclusions: Changing the needles did not reduce the contamination rate from postmortem hemocultures. Microorganisms commonly found in postmortem hemocultures are oral, gastrointestinal, and hospital-associated microorganisms.

Keywords: Postmortem hemoculture, Needle changing, Autopsy, Microbiology, Forensic medicine

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Introduction

Sepsis is a common cause of death in autopsy cases.¹ Typically, in living patients, this condition is diagnosed by blood culture and considered together with various symptoms and signs.² However, the signs and symptoms could not be examined in the case of the deceased. Postmortem hemoculture is the most essential factor for the diagnosis of sepsis.

Postmortem hemoculture is performed by drawing blood from the heart or great vessels after opening the pericardium by using a sterile method. Sealing at the puncture site before drawing blood with a hot, heated scalpel may be used.^{1,3} Blood is then inoculated into a collection tube and sent to a laboratory for culture. All these steps must be performed using a sterile process.²

However, there are some cases where, even though there is no infection in the bloodstream, there are many reasons why the microorganism can be detected in postmortem hemoculture. After death, bacteria from the digestive tract can spread to other body parts, a process known as postmortem translocation. Various types of bacteria are endemic in the human digestive tract. However, many factors can cause these bacteria to spread to other parts of the body during death. The lining of the intestinal wall loses its stability, causing microorganisms to spread between cells. The body's immune system ceases to function, and the tissue deteriorates, losing its integrity.^{1,4}

Another factor is contamination during the collection of postmortem blood samples.^{1,4} Therefore, the results from postmortem hemocultures may not be reliable. One of the factors that can be controlled for a more accurate diagnosis of sepsis is reducing contamination while collecting postmortem blood samples. Some studies have shown that changing the needle before inoculation can reduce the contamination rate.⁵ However, several studies have reported that changing the needle before inoculation

did not reduce the contamination rate.^{6,7} It also increases the risk of needlestick injuries to personnel.⁶ Most studies related to blood culture tests have been conducted in living patients. Therefore, this study aimed to investigate the impact of changing needles to minimize contamination of postmortem hemocultures. This study aimed to provide guidance for the use of blood cultures during autopsies.

There are interesting points regarding the time-to-positive (TTP) of the hemoculture. Some studies have found that it can be used to indicate the tendency of positive cultures, whether caused by contamination during sample collection or actual infection of bacteria in the blood. The TTP may indicate the number of microorganisms detected. If it is caused by contamination, there will be a small number of microorganisms, and the TTP hemoculture will be longer than that in the case of actual infection in the bloodstream.⁸ It can also be used to assess the prognoses of living patients.⁹ However, no studies have been conducted to determine the extent to which TTP is a significant indicator of contamination of deceased blood samples.

Materials and Methods

Study Population

The study was conducted at the Forensic Medicine Division of the Department of Pathology, Faculty of Medicine Ramathibodi Hospital, Mahidol University, Thailand. Data were collected from forensic autopsy cases from July 1, 2023, until December 1, 2023. By design, the research selected cases believed to have had no blood infection before death. The deceased who had died for more than 24 hours or decomposed, had a history of being treated in the hospital for more than 24 hours, suspected infectious disease, intravenous drug use, an open wound penetrating the chest cavity, hemopericardium, and suspected infection through autopsy and microscopic examination were excluded from this study. A total of 40 deaths met the inclusion criteria included in the study.



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Ethics

This study has been reviewed and approved by the Committee on Human Rights Related to Research Involving Human Subjects, Faculty of Medicine Ramathibodi Hospital, Mahidol University, Thailand (MURA2023/401 on May 29, 2023).

Study Design and Setting

Blood samples were collected from each deceased during the autopsy. After opening the pericardium, a scalpel was heated for at least 1 minute to sear the heart, aorta, or nearby locations for 5 seconds. Then, 10 mL of blood was drawn from the heated area, and 5 mL was inoculated into a bacterial culture bottle (non-needle-changing group). The needle was then changed to a new one, and another 5 mL of blood was inoculated into another bacterial culture bottle (needle-changing group) and immediately sent to the laboratory for culture.

Laboratory Analysis

The bacterial culture bottle used was the bioMerieux BACT/ALERT FA PLUS vacuum bottle. When the needle was inserted into the vial, blood was immediately drawn into the vial. After the bottles were transferred to the microbiology laboratory, they were placed in the bioMerieux BACT/ALERT VIRTUO machine, an automated microbial detection system. It uses the principle of a colorimetric sensor array to detect the color of the bottom of a culture bottle, which changes when CO₂ production from bacteria increases. After positive culture results were obtained, the TTP was recorded. The cultured bacteria were identified using conventional MALDI-TOF MS methods. No growth was reported if no organism was detected within 5 days.

Data Collection and Statistical Analysis

Data were collected on the following variables of interest: sex, age, culture results, TTP of hemoculture,

cause of death, and postmortem interval (PMI). The results of postmortem hemoculture from the needle-changing and non-needle-changing methods were compared. The TTP was compared between the 2 groups. The PMI was compared between the positive and negative groups. Microorganisms that are frequently found in postmortem hemocultures were identified. The results were statistically analyzed using McNemar's test and median regression.

Results

Among the 40 deceased persons participating in the study, 32 (80%) were male and 8 (20%) were female. They ranged in age from 14 to 76 years (mean, 49.55 years). The time after death was 3 to 24 hours (median, 13 hours).

The positivity rate for culture was 57.5% in both groups, with no statistically significant difference ($P=1.00$). The Kappa coefficient was 0.795 at 90% agreement, indicating almost perfect agreement, which suggest that the positivity rates of both groups were in the same direction. However, different results were found for cases 19, 31, 39, and 40. In cases 19 and 40, the culture results were positive in the non-needle-changing group but negative in the needle-changing group. In cases 31 and 39, the culture results were positive for the needle-changing group but negative for the non-needle-changing group. The culture results of both needle-changing and non-needle-changing groups showed mixed-growth organisms in 11 cases (27.5%).

The median PMI for which the culture was positive was 12 hours, and the culture was negative for 15 hours in both groups. No statistically significant differences were observed between the groups ($P = .417$).

The TTP culture in the non-needle-changing group was 2.8 to 117.7 hours (median, 9.9 hours) and 3.3 to 43.2 hours (median, 10.1 hours) in the needle-changing group. The results of the median regression analysis revealed that increasing the PMI increased the TTP results. However, this difference was not statistically significant in either group (Table 1).



Hemocultures

Table 1. Summary of Postmortem Hemoculture Results From the Non-Needle-Changing and Needle-Changing Groups

Case	Sex	Age, y	Non-Needle-Changing Group		Needle-Changing Group		Cause of Death	PMI, h
			Result	Microorganism	TTP, h	Result	Microorganism	TTP, h
1	M	44	+	<i>Klebsiella pneumoniae</i> complex	8.6	+	<i>Acinetobacter junii</i>	12.5
				<i>Shewanella</i> spp			<i>Shewanella</i> spp	
				Viridans group streptococci				
2	M	59	+	<i>Acinetobacter baumannii</i> complex	11	+	<i>Acinetobacter junii</i>	9.8
				<i>Enterobacter cloacae</i> complex			<i>Empedobacter</i> spp	
							<i>Weissella confusa</i>	
3	M	48	+	<i>Pseudomonas</i> spp	28	+	<i>Acinetobacter ursingii</i>	43.2
				<i>Staphylococcus hominis</i>				
4	M	27	-	-		-	-	
								Hanging
5	M	45	+	<i>Acinetobacter junii</i>	12.1	+	<i>Klebsiella pneumoniae</i> complex	10
6	M	27	+	<i>Morexella osloensis</i>	26.7	+	<i>Pasteurella multocida</i>	11.3
7	M	58	+	<i>Salmonella</i> spp	6.9	+	<i>Salmonella</i> spp	6.7
8	M	19	+	<i>Staphylococcus hominis</i>	11.4	+	<i>Staphylococcus hominis</i>	11.3
9	F	47	-	-		-	-	
								Coronary atherosclerosis
10	F	50	-	-		-	-	
								Coronary atherosclerosis
11	F	62	+	<i>Gemella haemolysans</i>	9.5	+	<i>Gemella haemolysans</i>	11.7
12	M	48	-	-		-	-	
								Coronary atherosclerosis
13	M	52	+	<i>Enterobacter cloacae</i> complex	10.6	+	<i>Enterobacter cloacae</i> complex	11.1
14	M	14	-	-		-	-	
								Electrocution
15	F	49	+	<i>Escherichia coli</i>	7.1	+	<i>Escherichia coli</i>	7
				Viridans group streptococci (strain 1)			Viridans group streptococci (strain 1)	
				Viridans group streptococci (strain 2)			Viridans group streptococci (strain 2)	
								Coronary atherosclerosis

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Hemocultures

Table 1. Summary of Postmortem Hemoculture Results From the Non-Needle-Changing and Needle-Changing Groups (Continued)

Case	Sex	Age, y	Non-Needle-Changing Group		Needle-Changing Group		Cause of Death	PMI, h	
			Result	Microorganism	TTP, h	Result			Microorganism
16	F	49	+	<i>Acinetobacter baumannii</i> complex	9	+	<i>Escherichia coli</i>	8.2	
				<i>Enterobacter cloacae</i> complex			<i>Lactobacillus lactis</i>		
				<i>Lactococcus lactis</i>			<i>Streptococcus infantarius</i>		
				Viridans group streptococci					
17	M	69	-	-	-	-	Coronary atherosclerosis	16	
18	M	69	+	<i>Bifidobacterium longum</i>	29.9	+	<i>Bifidobacterium longum</i>	32.5	
19	M	74	+	<i>Escherichia coli</i>	14.3	-	-	Intracerebral hemorrhage	11
20	M	56	-	-	-	-	Coronary atherosclerosis	24	
21	M	53	+	<i>Clostridium perfringens</i>	2.8	+	<i>Clostridium perfringens</i>	4.8	
				<i>Streptococcus agalactiae</i>			<i>Streptococcus agalactiae</i>		
22	M	48	-	-	-	-	Coronary atherosclerosis	6	
23	M	64	+	<i>Escherichia coli</i> (strain 1)	3.3	+	<i>Escherichia coli</i> (strain 1)	3.3	
				<i>Escherichia coli</i> (strain 2)			<i>Escherichia coli</i> (strain 2)		
				<i>Klebsiella pneumoniae</i> complex			<i>Klebsiella pneumoniae</i> complex		
				<i>Proteus mirabilis</i>			<i>Proteus mirabilis</i>		
				<i>Streptococcus dysgalactiae</i>			<i>Streptococcus dysgalactiae</i>		
24	F	67	-	-	-	-	Gut obstruction	19	
25	M	61	+	<i>Acinetobacter baumannii</i> complex	6.1	+	<i>Aeromonas</i> spp	5.4	
				<i>Aeromonas</i> spp			<i>Enterobacter cloacae</i> complex		
				<i>Enterobacter cloacae</i> complex			<i>Klebsiella pneumoniae</i> complex		
				<i>Klebsiella pneumoniae</i> complex			<i>Lactococcus garvieae</i>		
				<i>Weissella confusa</i>			<i>Weissella confusa</i>		



Hemocultures

Table 1. Summary of Postmortem Hemoculture Results From the Non-Needle-Changing and Needle-Changing Groups (Continued)

Case	Sex	Age, y	Non-Needle-Changing Group		Needle-Changing Group		Cause of Death	PMI, h
			Result	Microorganism	TTP, h	Result	Microorganism	TTP, h
26	M	59	+	<i>Klebsiella pneumoniae</i> complex	4.1	+	<i>Klebsiella pneumoniae</i> complex	3.8
				<i>Proteus mirabilis</i>			<i>Proteus mirabilis</i>	
				Viridans group streptococci			Viridans group streptococci	
27	M	58	-	-	-	-	-	-
				-	-		Coronary atherosclerosis	14
28	M	24	+	<i>Enterobacter cloacae</i> complex	8.8	+	<i>Enterococcus faecalis</i>	10.1
				<i>Streptococcus pneumoniae</i>			Viridans group streptococci	
29	M	76	-	-	-	-	-	-
				-	-		Ruptured cerebral berry aneurysm	15
30	M	62	+	<i>Acinetobacter baumannii</i> complex	10.3	+	<i>Acinetobacter baumannii</i> complex	10.4
				-	-		Cardiac arrhythmia	19
31	M	28	-	-	-	+	<i>Elizabethkingia meningoseptica</i>	23.8
				-	-		Cardiac arrhythmia	12
32	F	37	-	-	-	-	-	-
				-	-		Smoke inhalation	9
33	M	50	+	Viridans group streptococci	11.6	+	<i>Rothia mucilaginosa</i>	12.6
				-	-		Coronary atherosclerosis	13
				Viridans group streptococci			Viridans group streptococci	
34	M	34	+	Viridans group streptococci (strain1)	8.3	+	Viridans group streptococci (strain1)	9
				Viridans group streptococci (strain2)			Viridans group streptococci (strain2)	
35	M	21	+	<i>Escherichia coli</i>	9.9	+	<i>Escherichia coli</i>	9.7
				-	-		Cardiac arrhythmia	4
36	M	34	-	-	-	-	-	-
				-	-		Drowning	18
37	M	43	-	-	-	-	-	-
				-	-		Coronary atherosclerosis	21
38	M	56	-	-	-	-	-	-
				-	-		Coronary atherosclerosis	5
39	F	72	-	-	-	+	<i>Granulicatella adiacens</i>	14.2
				-	-		Coronary atherosclerosis	8
40	M	69	+	<i>Brachybacterium</i> spp	117.7	-	-	-
				-	-		Coronary atherosclerosis	5

Abbreviations: -, negative hemoculture; +, positive hemoculture, F, female; M, male, PMI, postmortem interval, TTP, time-to-positive.



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The bacteria found with the highest frequency were viridans group streptococci, found in 6 cases (15%) in the non-needle-changing group and 5 cases (12.5%) in the needle-changing group. In addition, common bacteria included *Enterobacter cloacae* complex, 5 cases (12.5%) in the non-needle-changing group, *Escherichia coli*, 4 cases (10%) in both groups, *Klebsiella pneumoniae* complex, 4 cases (10%) in both groups and other microorganisms (Table 2).

Table 2. Frequency of Microorganisms in Each Group

Microorganism	No. of Cases*	
	Non-Needle-Changing Group	Needle-Changing Group**
Viridans group streptococci	6 (8)	5 (7)
<i>Enterobacter cloacae</i> complex	5	2
<i>Escherichia coli</i>	4 (5)	4 (5)
<i>Klebsiella pneumoniae</i> complex	4	4
<i>Acinetobacter baumannii</i> complex	4	1
<i>Staphylococcus hominis</i>	2	1
<i>Proteus mirabilis</i>	2	2
<i>Shewanella</i> spp	1	1
<i>Pseudomonas</i> spp	1	-
<i>Acinetobacter junii</i>	1	2
<i>Moraxella osloensis</i>	1	-
<i>Salmonella</i> spp	1	1
<i>Gemella haemolysans</i>	1	1
<i>Lactococcus lactis</i>	1	1
<i>Bifidobacterium longum</i>	1	1
<i>Clostridium perfringens</i>	1	1
<i>Streptococcus agalactiae</i>	1	1
<i>Streptococcus dysgalactiae</i>	1	1
<i>Aeromonas</i> spp	1	1
<i>Weissella confusa</i>	1	2
<i>Streptococcus pneumoniae</i>	1	-
<i>Brachybacterium</i> spp	1	-
<i>Acinetobacter ursingii</i>	-	1
<i>Empedobacter</i> spp	-	1
<i>Pasteurella multocida</i>	-	1
<i>Streptococcus infantarius</i>	-	1
<i>Lactococcus garvieae</i>	-	1
<i>Enterococcus faecalis</i>	-	1
<i>Elizabethkingia meningoseptica</i>	-	1
<i>Rothia mucilaginosa</i>	-	1
<i>Granulicatella adiacens</i>	-	1

* Numbers in parentheses indicate different strains in the same case.

** No personnel accidents related to needle changes, including needlestick injuries or blood spillage, were observed.



Discussion

Postmortem hemoculture is crucial in determining the cause of death. However, it can produce false-positive results because there are other factors besides postmortem contamination,¹⁰ such as the spread of microorganisms from various organs into the blood after death (postmortem translocation).^{1,4} However, this study was designed to test hemoculture using needle-changing and non-needle-changing techniques and using the same deceased person as the source. Therefore, confounding variables could be eliminated when comparing contamination rates between the 2 groups, and the researchers selected deceased individuals with a low risk of bacteremia to expect only the effect of postmortem contamination.

The positivity rate for culture was 57.5% in both groups, with no statistically significant difference ($P=1.00$). The Kappa coefficient was 0.795 at 90% agreement, indicating almost perfect agreement, which suggested that the positivity rates of both groups were in the same direction. This result indicated that changing the needles does not affect the contamination rate in postmortem hemocultures. Although no data were found for the deceased, the results of this study were consistent with those of most studies on living patients.^{6,11}

Smart et al¹¹ studied the effect of changing needles to reduce the contamination rate of blood culture tests in patients. The rate of contamination from venipuncture tests in the group that did not change needles was 6.4% compared with 4.2% in the group that changed needles, which was not statistically significant ($P>.30$). The study also compared contamination rates from intravenous cannula blood draws and found no statistically significant difference between the contamination rates in the group that did not change needles (4.3%) and the group that did 4.2% ($P>.90$).

Leisure et al⁶ studied healthy medical students and found that blood cultures obtained from needle changes

had a contamination rate of 0.6% compared to 2.2% when the needles were not changed; although, no significant difference was found (95% confidence interval [CI], -0.7% to 4.0%).

However, a meta-analysis conducted by Spitalnic et al⁵ revealed that the group in which the needles were changed had an overall weighted contamination rate of 2.0%, whereas the group in which the needles were not changed had a rate of 3.7%. The difference between the 2 rates was statistically significant (95% CI, 0.75% - 1.75%; $P<.001$). As this study was conducted in living patients, slight differences in rates are significant because of their impact on testing and treatment costs.

The median PMI for those with negative cultures was 15 hours, and 12 hours for those with positive cultures. There was no statistically significant difference between PMI and the rate of positive cultures ($P=.417$). This result corresponded to the study by Sunagawa et al,² who did not find any difference in PMI between groups with positive and negative cultures ($P=.20$). Riedel's study¹ found that the period after death did not affect the positive culture rate if blood samples were collected and sent for testing within 24 hours after death.

From the results, the most common bacteria found in postmortem hemoculture were viridans group streptococci, *Enterobacter cloacae* complex, *Escherichia coli*, *Klebsiella pneumoniae* complex, *Acinetobacter baumannii* complex, *Weissella confusa*, and *Proteus mirabilis*, which are the common bacteria in the mouth, digestive tract, or hospitals, and mainly represent postmortem translocation.¹²⁻¹⁴

Another issue regarding postmortem hemoculture is the interpretation of results. In this study, the deceased had no history of infection before death, and no pathology indicating infection was found during autopsy. However, microorganisms were still detected in the blood cultures. This detection was likely due to postmortem translocation and contamination, which autopsy examiners should carefully interpret when using results to diagnose the cause of death.



Limitations

This study was conducted over a limited period, resulting in a small sample size. The time after death may not be accurate because almost all the patients in the study died outside the hospital. However, the authors used as much data as possible to identify the time of death from police documents, along with an estimate of the time of death from postmortem changes by a forensic pathologist.

individuals. The PMI does not affect the hemoculture positive rate if no signs of decomposition occur. However, the diagnosis of sepsis in postmortem hemocultures should be interpreted with caution because many factors can cause false-positive results. This study recommends studying the detection of bacterial species in corpses to determine the significance of the positive postmortem hemoculture.

Conclusions

This study found that changing the needle before inoculating blood into bacterial culture bottles did not affect the positive hemoculture rate of deceased

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บทนำ: การติดเชื้อในกระแสเลือดเป็นหนึ่งในสาเหตุการตายที่สำคัญในสภที่เสียชีวิตไม่ทราบเหตุ การเจาะเลือดเพื่อตรวจเพาะเชื้อจากสภนั้นมีโอกาสเกิดการปนเปื้อนได้ หนึ่งในวิธีการลดการปนเปื้อนที่ยังมีการถกเถียงกันว่ามีผลลดอัตราการปนเปื้อนหรือไม่คือการเปลี่ยนเข็มก่อนบรรจุลงขวดเพาะเชื้อแบคทีเรีย

วัตถุประสงค์: เพื่อศึกษาเปรียบเทียบอัตราการปนเปื้อนจากการตรวจหาเชื้อในเลือดผู้ป่วยชีวิตระหว่างกลุ่มที่เปลี่ยนเข็มกับกลุ่มที่ไม่ได้เปลี่ยนเข็ม

วิธีการศึกษา: การศึกษาแบบไปข้างหน้าโดยเจาะเลือดจากสภที่ได้รับการผ่าชันสูตรที่โรงพยาบาลรามาธิบดี โดยสภที่นำเข้าร่วมวิจัยเป็นสภที่เสียชีวิตไม่เกิน 24 ชั่วโมง และไม่สงสัยภาวะติดเชื้อ จำนวนทั้งสิ้น 40 ราย จากนั้นนำตัวอย่างเลือดที่ได้มาแบ่งบรรจุลงขวดเพาะเชื้อแบคทีเรียด้วยวิธีการเปลี่ยนเข็มและไม่เปลี่ยนเข็มก่อนบรรจุ และนำผลที่ได้จากการเพาะเชื้อมาวิเคราะห์เพื่อหาความแตกต่างกันระหว่าง 2 วิธี

ผลการศึกษา: อัตราการเพาะเชื้อให้ผลบวกในกลุ่มที่เปลี่ยนเข็ม คิดเป็นร้อยละ 57.5 และกลุ่มที่ไม่เปลี่ยนเข็ม คิดเป็นร้อยละ 57.5 โดยไม่พบความแตกต่าง (P=1.00) ค่า Kappa coefficient เท่ากับ 0.795 โดยเชื้อที่พบการปนเปื้อนบ่อยที่สุด ได้แก่ Viridans group streptococci, *Escherichia coli*, *Klebsiella pneumoniae* complex, *Enterobacter cloacae* complex, และ *Acinetobacter baumannii* complex

สรุป: การเปลี่ยนเข็มไม่มีผลต่อการลดอัตราการปนเปื้อนจากการตรวจเพาะเชื้อในเลือดผู้ป่วยชีวิต และเชื้อที่ตรวจพบปนเปื้อนจากการตรวจเพาะเชื้อจากสภส่วนใหญ่เป็นเชื้อในช่องปาก ทางเดินอาหาร และเชื้อในโรงพยาบาล

คำสำคัญ: การเพาะเชื้อจากเลือดภายหลังการตาย การเปลี่ยนเข็ม การผ่าศพ จุลชีววิทยา นิติเวชศาสตร์

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