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Bacterial DNA Translocation in Children with Congenital Heart Disease Undergoing Cardiopulmonary Bypass

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Abstract

Background: Cardiopulmonary Bypass (CPB) is a routine clinical practice in cardiac surgery, however, CPB can result in bacterial DNA (bactDNA) translocation and endotoxemia. We aimed to detect the incidence, risk factors and the effect of the bactDNA translocation on postoperative outcomes in children with Congenital Heart Disease (CHD) after CPB.

Methods: Children with CHD who underwent CPB between March 2017 and May 2017 were identified. General and postoperative outcome-related information was retrieved. Intestinal Fatty Acid Binding Protein (IFABP) level was detected, the data were compared between patients with and without bactDNA translocation.

Results: The median length of CPB was 71.5 (53-102) minutes vs. 53 (45-65) minutes in patients with and without bactDNA in their blood (p=0.004). The median aortic cross-clamp time was 45 (30-61.5) minutes vs. 31 (23-36.5) minutes in patients with or without bactDNA in their blood (p=0.017). A higher postoperative level of IFABP (p=0.038) was identified in patients with bcatDNA translocation. Postoperative Mechanical ventilation time (p=0.004), ICU stay (p=0.008) and hospital stay (p=0.003) were all longer in patients with bactDNA translocation. The incidence of early complication was higher in patients with bactDNA translocation (p=0.000).

Conclusion: Intestinal damage is frequent after pediatric congenital heart surgery. BactDNA translocation into the blood increases increase the incidence of postoperative adverse outcome in children with CHD who undergo CPB.

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Introduction

Cardiopulmonary Bypass (CPB) is a routine clinical practice in cardiac surgery and has allowed for a significant reduction in morbidity in cardiac surgery [1]. However, CPB is known to cause a systemic inflammatory response. The early phase is triggered by blood contact with the synthetic bypass circuit and the late phase by ischemia-reperfusion and endotoxemia [2]. Gastrointestinal mucosal cells are very sensitive to stress response and ischemia, and consequently, there can be gastrointestinal mucosal ischemic damage and intestinal mucosal barrier damage, resulting in bacterial translocation and endotoxemia [3]. These effects synergistically contribute to the development of systemic inflammatory response and multiple organ dysfunction syndrome, which are leading causes of severe complications after cardiac surgery [4]. The presence of intestinal bacteria in sterile tissues and distant organs may cause damage due to chronic inflammation and progression of disorders, including inflammatory bowel diseases, liver cirrhosis, and acute pancreatitis [5]. Bacterial translocation was also reported as one of the nonischemic mechanisms that contribute to gastrointestinal complications after cardiac surgery [6].

DNA is sufficiently stable that it can be amplified by PCR for long periods after bacteria are no longer viable [7]. The clinical significance of diagnosing bactDNA translocation has been addressed in many diseases. Studies demonstrated that the presence of bactDNA is associated with significant hemodynamic changes, even in the absence of clinical infection [8]. Bacterial translocation has been proposed as a trigger for stimulation of the immune system with consequent hemodynamic alteration in patients with liver cirrhosis and it was found in about one-third of patients undergoing

living-donor liver transplant [9]. BactDNA translocation increases the development of postoperative sepsis in patients receiving surgical intervention [10]. It is difficult to assess the respective role of surgical trauma, and CPB technique in the magnitude of systemic inflammatory response to cardiac surgery. While the control of the inflammatory response extent is likely to require a multimodal approach, researches about bactDNA translocation are of great significance to the pathogenesis involved in the outcomes of CPB.

Intestinal Fatty Acid Binding Protein (I-FABP) is suggested to be a reliable serologic marker for early detection of vascular and nonvascular intestinal injury [11]. It has been used as a potential serologic marker for intestinal ischemia-reperfusion injury in patients undergoing elective cardiac surgery with CPB [12]. Herein, we aimed to establish whether elevated I-FABP level was associated with bactDNA translocation and to assess the incidence, risk factors and consequence of bactDNA translocation in children with Congenital Heart Disease (CHD) after CPB.

Materials and Methods

Clinical materials

This trial was conducted between March 2017 and May 2017 in Department of Cardiothoracic Surgery of Shanghai Children's Hospital and the present analysis focused on bactDNA translocation after CPB in the subgroup children with CHD. The study protocol was approved by the Ethics Committee of Shanghai Children's Hospital. Informed consent was obtained from legal guardians of these pooled children. In brief, all children admitted to CPB in the Department of Cardiothoracic Surgery were eligible for the complete study group. Exclusion criteria were as follows: With no parental consent, emergency operations, active or prior history of autoimmune disorders, cardiac function Class III-IV, preoperative infectious disease or fever, other important organ disorders including abnormal coagulation, liver and kidney insufficiency. Types of CHD included Atrial Septal Defect (ASD), Ventricular Septal Defect (VSD), VSD with Pulmonary Hypertension (PH), VSD with ASD, VSD with Patent Ductus Arteriosus (PDA), Fallot Tetralogy (FOT), ASD with FOT, Complete Pulmonary Vein ectopic drainage (TAPVC), Double Outlet of Right Ventricle (DORV), Pulmonary valve stenosis, VSD with aortic valve prolapse, ASD with Pulmonary vein ectopic drainage, Pulmonary atresia, Abnormal origin of the Right Pulmonary Artery in the Aorta (AORPA) with Moderate tricuspid regurgitation.

Measurements

I-FABP measurement: Two (2) ml blood sampling were performed on the first postoperative day professionally. One (1) ml blood was centrifuged at 3000 g for 10 min at 4°C, and plasmas were stored at -80°C for I-FABP measurements, another 1 ml whole blood was for bactDNA extraction. Serum levels of I-FABP were measurement by ELISA (RNA Systems, Minneapolis, MN) according to the manufactures instruction. Assay range was 15.6 pg/ml to 1000 pg/ml. Higher values were determined after dilution with assay diluent. Intra-assay coefficient of variation was 4.1% and inter-assay coefficient of variation was 11.1%.

DNA extraction: Bacterial DNA was extracted using the QIAmp DNA Minikit (Qiagen) according to the protocols in the manufacturer's instructions. The extracted DNA was stored at 4°C until required for PCR. We used the Dream Taq TM PCR Master Mix 2X (Fermentas) (#K1071) containing: Dream Taq TM DNA Polymerase, Dream Taq TM PCR Buffer, 4 mM $MgCl_2$, and dNTPs for the PCRs. Each reaction tube contained: Master Mix 12.5 µl, 0.2 mM of each primer (amount of Primer Mix 2 µl; 1 µl forward & 1 µl reverse each diluted 1:10 from the stock), template DNA 10 µl (approximately 500 ng), and 5 µl 1X PCR buffer. The reaction mixtures were vortexed briefly. Amplification reactions were carried out in a Seegene (SEE AMP) thermocycler. The primer was specific for various gram-negative and gram-positive bacterial species (p16SrRNA+ and P16SrRNA-).

Other data collection: Clinical data collection: The observations including general preoperative data name, gender, age, weight and duration of cardiopulmonary bypass, aortic cross-clamp time, postoperative bactDNA translocation, length of postoperative mechanical ventilation, hospitalization length, ICU length and early postoperative complications. All complications were defined as those occurring within 30 days from the date of surgery.

Statistical analysis

Data are presented as median (M) and inter-Quartile Range (QR) or quantitative variables and as frequencies for qualitative variable. The Mann–Whitney rank-sum test was used for comparison of continuous variables between bactDNA(+) and bactDNA(−) cases. For categorical data, Fisher exact or chi-square tests were used for comparison as appropriate. A P value less than 0.05 was considered to indicate statistical significance. Data processing and analysis were performed with SPSS24.0 software.

Results

I-FABP levels after cardiopulmonary bypass

The median I-FABP levels in bactDNA(-) group is 1.32 (1.28-1.75) ng/ml, 1.82 (1.37-6.48) ng/ml in bactDNA(+) group. The difference between the two groups is statistically significant (p=0.038).

Incidence of bactDNA translocation

Forty-seven children were enrolled in this study, the hospital survival rate was 100%, no readmission and reoperation. Patients were divided into two groups according to the absence (bact-) or presence (bact+) of bacterial DNA. BactDNA was detected in 30 (63.83%) patients, the rest 17(36.17%) were negative. The median age was 24 months (6.5-42) in bact(-) group, 5 months (2.0-13.5) in bact(+) group, the difference is statistically significant (p=0.016). There was no significance difference between the groups for male to female ratio (p=1.00). Patients' characteristics and preoperative data were expressed in Table 1.

Cardiopulmonary bypass-related risk factors and outcome

The patients in bact(+) group had shorter preoperative hospitalization time (4.82 ± 3.0 *vs.* 8.33 ± 5.6 day; p<0.05). Three out of the 47 patients had abnormal preoperative CRP levels, and they showed bactDNA translocation. The CPB time and aortic cross-clamp time were longer in the bactDNA(+) group. When the clinical

Table 1: Baseline patient characteristics.

	Bact(-) (n=17)	Bact(+) (n=30)	P
Male to female ratio	10:07	17:13	1
Age group (no.)			0.016
0-4 weeks	1	4	
1-12 months	6	19	
1-5 years	10	7	
Weight (kg)	10.0 (6.5-15.5)	5.6 (4.7-9.37)	0.022

P value less than 0.05 was considered to indicate statistical significance

Table 2: Clinical data and postoperative outcome.

	bactDNA(-)	bactDNA(+)	P
Preoperative hospital stay (day)	4.82 ± 3.0	8.33 ± 5.6	0.022
Preoperative CRP>5 mg/L	0	3	0.292
Length of CPB (min)	53 (45-65)	71.5 (53-102)	0.004
CPB ≥ 90 min	1	9	0.116
Aortic cross-clamp time (min)	31 (23-36.5)	45 (30-61.5)	0.017
Mechanical ventilation time (day)	1.0 (0.2-1.5)	2.0 (1.0-3.0)	0.004
Stay in intensive care unit (day)	4 (3.5-7.5)	8 (5.0-10.3)	0.008
Length of hospital stay (day)	15 (10.5-2.0)	23 (16.5-30.5)	0.003
Early complication	0	6 (20%)	0

P value less than 0.05 was considered to indicate statistical significance

outcome was compared, we found significant differences in terms of the time of mechanical ventilation (1.0 (0.21-1.5) *vs.* 2.0 (1-3) min; P<0.01, stay in intensive care unit 4 (3.5-7.5) *vs.* 8 (5.0-10.25) day; p<0.01, and length of hospital stay 15 (10.5-2) *vs.* 23 (16.5-30.5) day; p<0.01. The hospital stay was uneventful for all the 17 patients in bact (-) group, 6 out of 30 (20%) patients in the bact(+) group developed early complications including 3 infections and 3 blood transfusion requirements (p<0.01). The clinical data and postoperative outcome of the study group are presented in Table 2.

Discussion

The present study focused on bactDNA translocation of children with CHD after CPB and investigated the relationships between the bactDNA translocation and the postoperative outcomes in children. We demonstrated that the children who underwent CPB had a large risk of bactDNA translocation and the presence of bcatDNA in blood was associated with the increased risk postoperative adverse outcomes in these children with CHD after CPB.

Six out of every 1000 babies have some form of significant congenital heart disease and many will require corrective or palliative cardiac surgery during infancy or childhood [13]. Several studies have documented alterations of the lower gastrointestinal microcirculation and intestinal mucosa damage during CPB [3,14]. Reilly et al. reported that the gastrointestinal system is prone to complications after cardiac surgery because of the interactions between the Gastrointestinal (GI) and cardiovascular systems, complications are largely the result of perfusion abnormalities of the splanchnic circulation that lead to ischemia–reperfusion injury [15]. A study by Ferguson et al. reported that children undergoing congenital heart surgery have a potentially higher risk of GI complication than adults [16]. The main mechanism of intestinal damage following CPB is related to a decrease of tight junction protein expression in the intestinal epithelium, which subsequently leads to changes in structure and function of the intestinal mucosa, increased intestinal permeability, bacterial translocation, and endotoxemia [17]. Since endotoxin originates only from Gram-negative bacteria and the sensitivity of blood bacterial culture is very low in clinical specimens, then the presence of bacterial DNA in blood is recognized as a marker of bacterial translocation [18], we suggest it is necessary and reliable to detect the bactDNA translocation in children who underwent CPB.

In our study, bactDNA translocation occurred in 30 out of 47 children with congenital heart disease undergo CPB. Alternatively, in another clinical study about CPB by Rossi et al. the presence of bactDNA was reported in 10 out of 11 patients scheduled for elective coronary artery bypass graft surgery [19]. For patients with inflammatory bowel disease, it is also a common event. The presence of bactDNA translocation was reported by Gutiérrez et al. as 42.4% in patients with ulcerative colitis and 51.7% in patients with Crohn's disease [20]. Some details might be responsible for the different translocation incidence such as the different time of the blood samples collected and the different group of the enrolled patients. Instead, another study by Klein et al. found only 7.7% of patients subjected to cardiac surgery with CPB had endotoxin translocation, the link between endotoxemia and translocation from the intestines could not be established, because the most possible source of endotoxins was the primary site of infection [21]. The difference in bactDNA translocation incidence of certain pediatric age groups after CPB have never been studied in details. Younger age and lower weight have been associated with increased inflammatory reaction in previous studies [22,23]. In present study, children with younger age and lower weight are at higher risk of bactDNA translocation, the difference in male to female ratio are not significant. Preoperative hospital stay of bactDNA(+) patients was significantly longer, severity of CHD might responsible for this result.

Once the intestinal get injured, intestinal epithelial cells express a family of small cytosolic fatty caid binding proteins [24]. Intestinal Fatty Acid Binding Protein (I-FABP) has been used as a blood biomarker to investigate intestinal damage in CPB and the study reported that the higher level of I-FABP was associated with the longer duration of CPB [25]. Similar results were found in our study, a significant increase of I-FABP was observed in every patient on the first postoperative day in two groups, indicating mucosal intestinal damage. Moreover, the level of I-FABP was strongly associated with bactDNA translocation, it is higher in patients with bactDNA translocation and significantly lower in cases without bactDNA in blood, suggesting bacterial translocation due to impaired intestinal permeability.

There was no difference observed between patients with and without bactDNA translocation for the male to female ratio and preoperative CRP. However, CPB time as well as the aortic cross-clamp time were markedly prolonged in the patients with bactDNA translocation. The gut translocation has traditionally been perceived as the primary source of endotoxemia and the endotoxemia associated with cardiac surgery is thought to be dominantly influenced by the use of CPB [26]. In a study by Tsunooka et al. CPB caused hypoperfusion of small intestinal mucosa and consequently bacterial translocation [27]. These results suggest that CHD patients should receive more attention during CPB to avoid bactDNA translocation. Our data also indicated bactDNA translocation was associated with longer mechanical ventilation time, ICU stay, hospital stay and the increased the incidence of early postoperative complications. Similar results were reported that bactDNA translocation was associated with a longer mean postoperative hospital stay, an increased readmission rate and the postoperative adverse outcomes in patients with Crohn's disease who undergo abdominal surgery [28]. Significant increases in potential microbial translocation, have been identified in many immune-related and inflammatory diseases, such as inflammatory bowel disease, colorectal cancer, rheumatoid arthritis, and liver cirrhosis, for which we currently have no cure or long-term treatment options [29].

The CPB course, postoperative course, and early complications

were comparable between both groups of patients. However, we are aware there are some limitations in our study. First, the population is not big enough to detect all the significant differences between the two groups. Second, we cannot draw any conclusions regarding the effect of bactDNA translocation on the long-term outcomes.

In conclusion, the present study demonstrated that there is a high risk of bactDNA translocation for CPB and the presence of bactDNA in blood is associated with increased adverse outcomes. These results suggest that multiple center and large-scale studies are needed to do and the increased risk should be taken into consideration when planning surgery for children with CHD.

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References

1. Whiting D, Yuki K, DiNardo JA. Cardiopulmonary bypass in the pediatric population. Best Pract Res Clin Anaesthesiol. 2015;29(2):241-56.
2. Durandy Y. Minimizing systemic inflammation during cardiopulmonary bypass in the pediatric population. Artif Organs. 2014;38(1):11-8.
3. Tsunooka N, Hamada Y, Imagawa H, Nakamura Y, Shiozaki T, Suzuki H, et al. Ischemia of the intestinal mucosa during cardiopulmonary bypass. J Artif Organs. 2003;6(2):149-51.
4. Hamilton-Davies C, Barclay GR, Cardigan RA, McDonald SJ, Purdy G, Machin SJ, et al. Relationship between preoperative endotoxin immune status, gut perfusion, and outcome from cardiac valve replacement surgery. Chest. 1997;112(5):1189-96.
5. Hustmyer CM, Landick R. Bacterial chromatin proteins, transcription, and DNA topology: Inseparable partners in the control of gene expression. Mol Microbiol. 2024.
6. Allen SJ. Gastrointestinal complications and cardiac surgery. J Extra Corpor Technol. 2014;46(2):142-9.
7. Sushama S, Maria BC, Ricardo GM, Pedro PVPD, Edward BB. Use of broad range16S rDNA PCR in clinical microbiology. J Microbiol Methods. 2009;76(3):217-25.
8. Pablo B, Juan CG-P, Rubén F, Juan GA, Miguel N, Miguel P-M, et al. Bacterial DNA translocation is associated with systemic circulatory abnormalities and intrahepatic endothelial dysfunction in patients with cirrhosis. Hepatology. 2010;52(6):2044-52.
9. Moharem HA, Fetouh FA, Darwish HM, Ghaith D, Elayashy M, Hussein A, et al. Effects of bacterial translocation on hemodynamic and coagulation parameters during living-donor liver transplant. BMC Anesthesiol. 2018;18(1):46.
10. MacFie J, Reddy BS, Gatt M, Jain PK, Sowdi R, Mitchell CJ. Bacterial translocation studied in 927 patients over 13 years. Br J Surg. 2006;93(1):87-93.
11. Holmes JH, Lieberman JM, Probert CB, Marks WH, Hill ME, Paull DL, et al. Elevated intestinal fatty acid binding protein and gastrointestinal complications following cardiopulmonary bypass: A preliminary analysis. J Surg Res. 2001;100(2):192-6.
12. Struck R, Wittmann M, Muller S, Meybohm P, Müller A, Bagci S. Effect of remote ischemic preconditioning on intestinal ischemia-reperfusion injury in adults undergoing on-pump CABG surgery: A randomized controlled pilot trial. J Cardiothorac Vasc Anesth. 2018;32(3):1243-47.
13. Hoffman JI, Kaplan S. The incidence of congenital heart disease. J Am Coll Cardiol. 2002;39(12):1890-900.
14. Tao W, Zwischenberger JB, Nguyen TT, Vertrees RA, McDaniel LB, Nutt LK, et al. Gut mucosal ischemia during normothermic cardiopulmonary bypass results from blood flow redistribution and increased oxygen demand. J Thorac Cardiovasc Surg. 1995;110(3):819-28.
15. Reilly PM, Wilkins KB, Fuh KC, Haglund U, Bulkley GB. The mesenteric hemodynamic response to circulatory shock: An overview. Shock. 2001;15(5):329-43.
16. Ferguson LP, Gandiya T, Kaselas C, Sheth J, Hasan A, Gabra HOS. Gastrointestinal complications associated with the surgical treatment of heart disease in children. J Pediatr Surg. 2017;52(3):414-9.
17. Sun YJ, Chen WM, Zhang TZ, Cao HJ, Zhou J. Effects of cardiopulmonary bypass on tight junction protein expressions in intestinal mucosa of rats. World J Gastroenterol. 2008;14(38):5868-75.
18. Guarner C, Gonzalez-Navajas JM, Sanchez E, Soriando G, Francés R, Chiva M, et al. The detection of bacterial DNA in blood of rats with CCl4-induced cirrhosis with ascites represents episodes of bacterial translocation. Hepatology. 2006;44(3):633-9.
19. Rossi M, Sganga G, Mazzone M, Valenza V, Guarneri S, Portale G, et al. Cardiopulmonary bypass in man: Role of the intestine in a self-limiting inflammatory response with demonstrable bacterial translocation. Ann Thorac Surg. 2004;77(2):612-18.
20. Gutiérrez A, Francés R, Amorós A, Zapater P, Garmendia M, Ndongo M, et al. Cytokine association with bacterial DNA in serum of patients with inflammatory bowel disease. Inflamm Bowel Dis. 2009;15(4):508-14.
21. Klein D J, Briet F, Nisenbaum R, Romaschin AD, David Mazer C. Endotoxemia related to cardiopulmonary bypass is associated with increased risk of infection after cardiac surgery: A prospective observational study. Crit Care. 2011;15(1):R69.
22. Soares LC, Ribas D, Spring R, Ferreira da Silva JM, Miyague NI. Clinical profile of systemic inflammatory response after pediatric cardiac surgery with cardiopulmonary bypass. Arq Bras Cardiol. 2010;94(1):127-33.
23. Guvener M, Korun O, Demirturk OS. Risk factors for systemic inflammatory response after congenital cardiac surgery. J Card Surg. 2015;30(1):92-6.
24. Smathers RL, Petersen DR. The human fatty acid-binding protein family: evolutionary divergences and functions. Hum Genomics. 2011;5(3):170-91.
25. Adamik B, Kübler A, Gozdzik A, Gozdzik W. Prolonged cardiopulmonary bypass is a risk factor for intestinal ischaemic damage and endotoxaemia. Heart Lung Circ. 2017;26(7):717-23.
26. Aydin NB, Gercekoglu H, Aksu B, Ozkul V, Sener T, Kiygil I, et al. Endotoxemia in coronary artery bypass surgery: A comparison of the off-pump technique and conventional cardiopulmonary bypass. J Thorac Cardiovasc Surg. 2003;125(4):843-48.
27. Tsunooka N, Maeyama K, Hamada Y, Imagawa H, Takano S, Watanabe YJ, et al. Bacterial translocation secondary to small intestinal mucosal ischemia during cardiopulmonary bypass. Measurement by diamine oxidase and peptidoglycan. Eur J Cardiothorac Surg. 2004;25(2):275-80.
28. Li Y, Zuo LG, Zhu WM, Gong JF, Zhang W, Gu LL, et al. The impact of bacterial DNA translocation on early postoperative outcomes in Crohn's patients undergoing abdominal surgery. J Crohn's Colitis. 2015;9(3):259-65.
29. Jin S, Wetzel D, Schirmer M. Deciphering mechanisms and implications of bacterial translocation in human health and disease. Curr Opin Microbiol. 2022;67:102147.