Conclusions. In our study, TMP-SMX was well tolerated; however, only 78% of patients were successfully treated. The majority of treatment failures had prolonged bacteremia due to MRSA perhaps suggesting a higher bacterial burden. The poor outcome in these patients is likely multifactorial, and antibiotic contribution is unknown. TMP-SMX may be a reasonable treatment option for children with OAI when the disease is mild; however, caution should be exercised with severe disease, especially when associated with bacteremia. Prospective, randomized control trials are needed to aid in guideline development and understand the role of TMP-SMX in the treatment of children with OAI.

Table 1. Characteristics and outcomes of patients treated with TMP-SMX

Characteristic	*N* = 21
Sex	
Male (%)	13 (62)
Female (%)	8 (38)
Race	
Non-Hispanic White (%)	17 (81)
Non-Hispanic Black (%)	1 (5)
Other (%)	3 (14)
Age, years (median [IQR])	1.5 (1–3)
Length of hospitalization (days)	5 (4–7)
Infection type	
Osteomyelitis (%)	12 (57)
Septic arthritis	7 (33)
Both	2 (10)
Pathogen	
Staphylococcus aureus	10 (48)
Methicillin-resistant	10
Clindamycin-resistant	3
Salmonella spp.	1 (5)
Enterobacter cloacae	1 (5)
Culture negative[a]	9 (43)
Complications during hospitalization[b]	
Uncomplicated	19 (90)
DVT/Emboli	2(10)
ICU Admission	1(5)
Sepsis	1(5)
TMP-SMX dose (mg/kg/day of TMP, median [IQR])	12.7 (11.3–14.9)
Reason for choosing TMP-SMX	
Physician preference	13(62)
Taste/Tolerated previously	3(14)
Clindamycin-resistant organism[c]	4(19)
Penicillin allergy	1(5)
Adverse drug reaction	2(10)
Acute kidney injury	1
Increased liver enzymes	1
Outcomes	*n* = 18[d]
Successfully completed TMP-SMX treatment	14(78)
Successful treatment; unable to tolerate TMP-SMX	1
Developed Recurrent Infection	3
Soft-tissue infection at the surgical site	1
Chronic osteomyelitis + pathologic fracture	2

[a]One patient had gram-positive cocci on Gram stain, culture negative.
[b]One patient had multiple complications.
[c]One patient had a history of clindamycin-resistant infection.
[d]Three patients lost to follow-up.

#75: Microbial Cell-free DNA Sequencing for Prediction of Culture-Negative Infection Events in Children with Cancer

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Background. New culture-independent diagnostics are being investigated for diagnosis and prediction of infection. One method is microbial cell-free DNA sequencing (mcfDNA-seq), which can detect a wide range of pathogens directly from plasma. Immunocompromised children who develop febrile neutropenia (FN) without documented bloodstream infection (BSI) may have an undiagnosed bacterial infection, but identification of this subset is difficult. The percentage of episodes of FN caused by bacterial pathogens that did not grow in culture is unknown, as is the relative contribution of other specific etiologies such as viruses, fungi, immunotherapies, and immune reconstitution or graft vs. host disease. The value of mcfDNA-seq in FN patients is also unknown. As part of a larger study evaluating the ability of mcfDNA-seq to predict BSI in a cohort of pediatric patients with relapsed or refractory leukemia, we analyzed mcfDNA-seq results in a sample of cases of FN for which the definitive etiology was unknown.

Methods. Eligible participants were <25 years of age and undergoing treatment for cancer. Remnant plasma from clinical testing was prospectively obtained and stored. Samples collected on the day of onset of FN (Day 0) and the day prior (Day –1) underwent mcfDNA-seq by Karius Inc. in a CLIA and CAP-accredited laboratory. Pathogen detection results were reported in molecules per microliter (MPM) of plasma. Negative control samples from study participants without impending or recent fever or infection were also obtained. Testing was batched and blinded, so results were not clinically available.

Results. mcfDNA-seq results were obtained from eight episodes of FN in seven patients. Five episodes occurred in participants awaiting engraftment after HCT and three were receiving chemotherapy only. All participants receiving chemotherapy were receiving antibacterial prophylaxis with vancomycin and ciprofloxacin, antifungal prophylaxis with micafungin or voriconazole, and PJP prophylaxis with TMP-SMX or pentamidine. No HCT recipients were receiving antibacterial prophylaxis, but all received PJP prophylaxis, antifungal prophylaxis or treatment, and antiviral prophylaxis or treatment. Of 8 FN episodes, 4 (50%) had a common bacterial pathogen identified by mcfDNA-seq on Day 0 (Table 1). In 2 (50%) of these cases, the same organism was also identified on Day –1, at a lower concentration. One fungal pathogen was identified prior to and at the onset of FN. A common bacterial pathogen was identified in 3/64 (5%) control samples from the study population. Culture-negative sepsis was the final diagnosis in one episode; in this case, *Streptococcus mitis*, an important cause of sepsis in neutropenic patients, was identified in both Day 0 and Day –1 samples. In another episode where *E. coli* was identified, antibiotics were discontinued after 48 hours, but the patient was re-admitted within 24 hours for recurrent FN.

Conclusions. In this sample of culture-negative FN episodes in pediatric patients with relapsed or refractory leukemia, mcfDNA-seq identified a common bacterial pathogen in 50% of cases. The same organism was identifiable on the day prior to FN in 50% of cases, suggesting that predictive testing might be feasible. More data regarding sensitivity and specificity of mcfDNA-seq to diagnose and predict FN are needed.

Table: Quantitative mcfDNA-seq results for prediction and diagnosis of febrile neutropenia episodes

		Common bacterial pathogens (organism, MPM)		Other organisms (organism, MPM)	
Episode	HCT	Day 0	Day –1	Day 0	Day -1
1	Yes	*Streptococcus mitis*, 657	*S. mitis, 379*	None	None
2	No	*Escherichia coli*, 5728	*E. coli, 98*	None	*Helicobacter pylori, 49*
3	Yes	*S. mitis*, 206	None	*Mucor velutinosus, 559*	*M. velutinosus, 382 HHV5 (CMV), 27*
4	No	*Streptococcus oralis*, 257 *Streptococcus sanguinis*, 131 *Fusobacterium nucleatum*, 708	*Staphylococcus epidermidis*, 113	*Tannerella forsythia*, 136 *Rothia dentocariosa*, 118 *Propionibacterium propionicum*, 131 *Cardiobacterium hominis*, 89 *Prevotella loescheii*, 119 *Gemella morbillorum*, 43 *Campylobacter gracilis*, 49 *Corynebacterium matruchotii*, 431 *Actinomyces oris*, 251 *Campylobacter concisus*, 34 *Neisseria mucosa*, 271 *Actinomyces viscosus*, 750 *Parvimonas micra*, 28 *Campylobacter showae*, 102 *Neisseria elongate*, 112 *Prevotella melaninogenica*, 49 *Neisseria flavescens*, 331 *Veillonella parvula*, 158 *Capnocytophaga granulosa*, 238	*Staphylococcus saprophyticus*, 29 *Prevotella melaninogenica*, 26 *Staphylococcus capitis*, 72
5	Yes	None	None	None	None
6	Yes	None	None	HHV5 (CMV), 68	HHV5 (CMV), 92
7	Yes	None	None	*Bacteroides ovatus, 21 Bacteroides vulgatus, 31*	*B. ovatus, 91*
8	No	None	None	*Bacteroides thetaiotaomicron, 56*	*B. thetaiotaomicron, 34*

MPM, concentration of pathogen cell-free DNA in molecules per microliter of plasma; HCT, history of allogeneic hematopoietic cell transplantation