


Microbial Cell-Free DNA Sequencing for Evaluation of Response to Antibiotic Therapy in Patients with Relapsed or Refractory Leukemia

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Antibiotic Treatment Duration Hinges on Demonstrating Clearance of Organisms

Laboratory results demonstrating microbial clearance in bloodstream infection are crucial to determine optimal length of treatment and line removal/placement decisions

Current treatment guidelines (one size fits all) have to err on longer courses of antibiotics to insure that microbes which are present (below limit of detection) but not cultured on subsequent tests can be covered

Safe antibiotic discontinuation would require a more sensitive test showing microbiological response to treatment

Microbial cell-free DNA sequencing (mcfDNA-seq) is a sensitive predictor of bloodstream infections and might be adequate to measure response to treatment

We hypothesize that daily mcfDNA-seq may provide more precise and detailed clearance dynamics for bloodstream infections than daily blood cultures due to the factors influencing microbial cell-free DNA differing from culture (see fig 1).

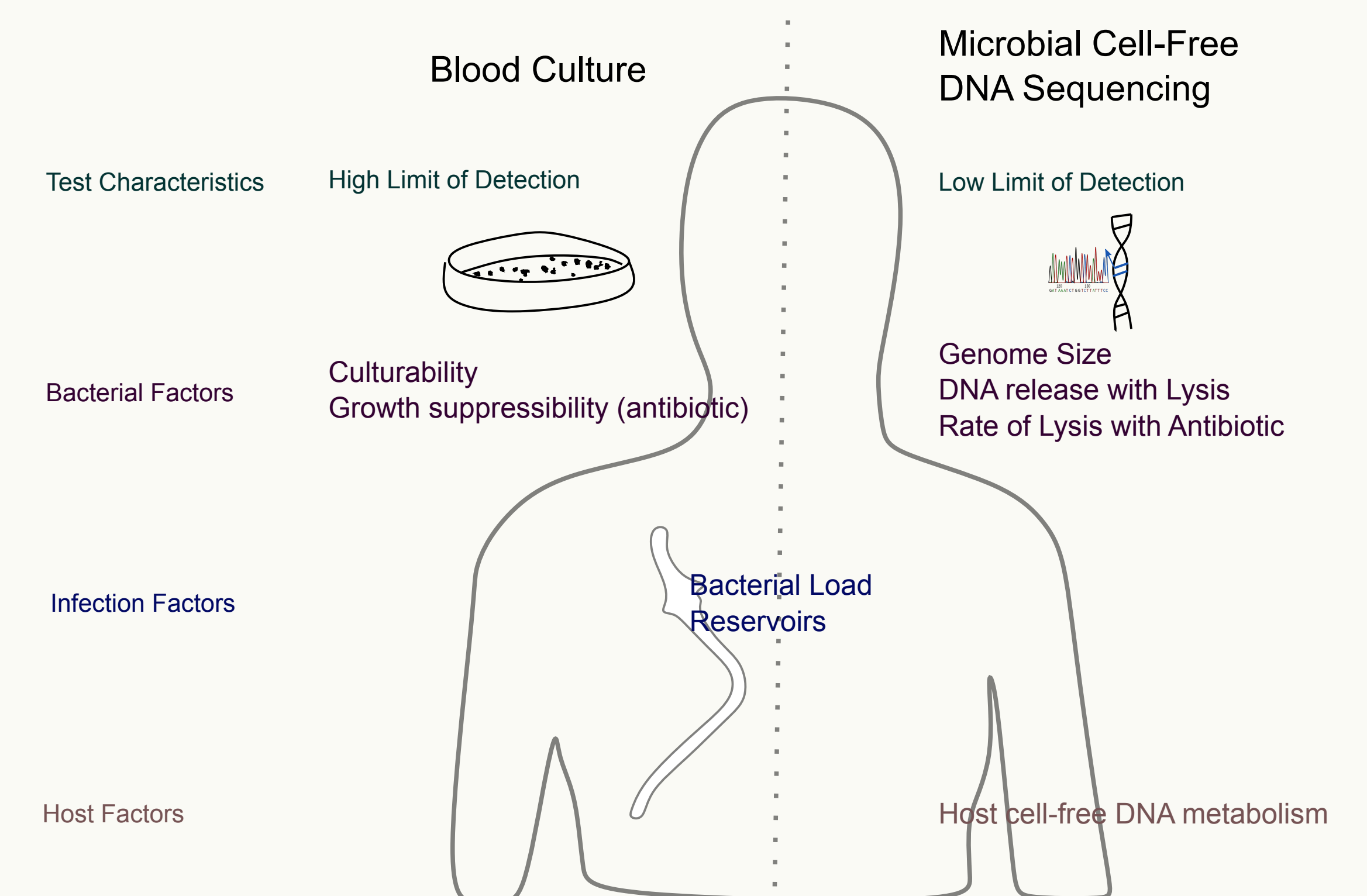
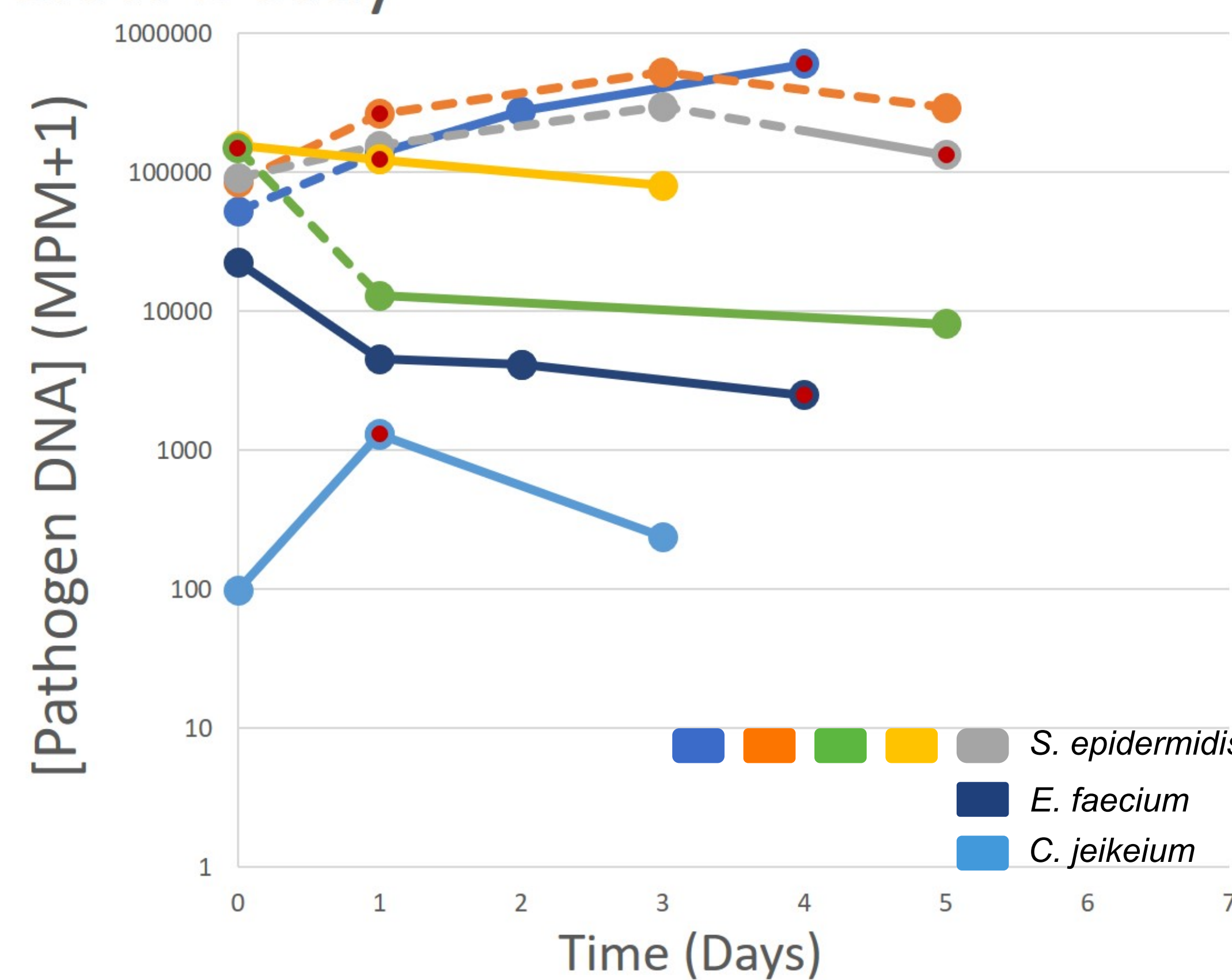


Figure 1: Influences on Microbial cell-free DNA sequencing detection differ from bacterial culture

Slow Decay



Rapid Decay

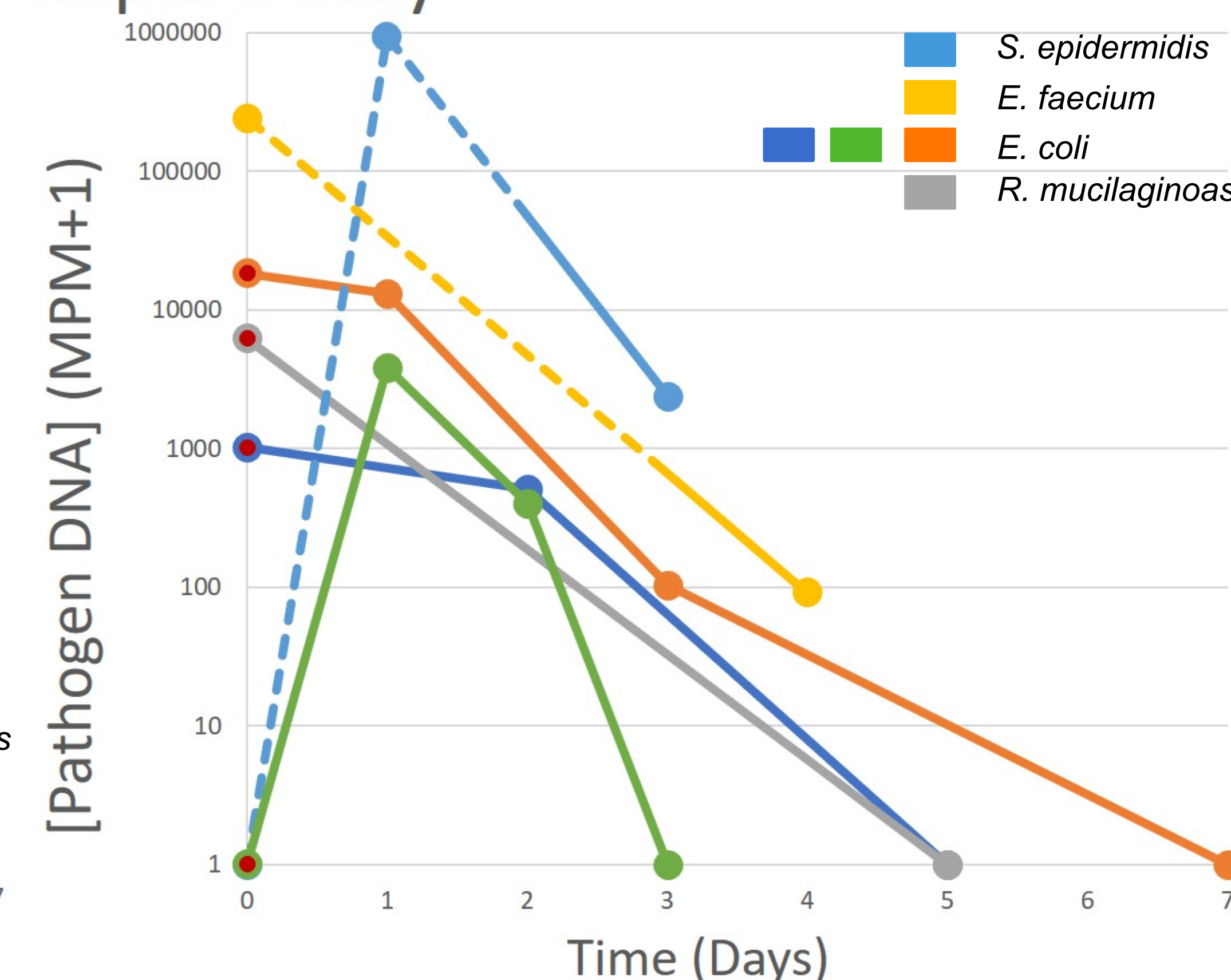


Figure 2: DNA concentration (mcfDNA-seq by Karius reported in molecules per microliter MPM) decay rate of detected bloodstream pathogen after effective therapy in participants (<25yo age during treatment for leukemia) from the peak to the last available sample. Slow DNA decay was defined as <0.5 log₁₀ MPM/day. Dashed lines when blood cultures remained positive. Red circle denotes last fever.

Slow decay of cell-free DNA observed in 50% of bacteremias

There were 13 evaluable BSI episodes in 9 participants, of which 7 had slow DNA decay.

Slow decay = decline of < 0.5 log₁₀ molecules/microliter/day

Slow decay persisted beyond resolution of both bacteremia and fever in 3/7 cases.

Persistence of positive blood cultures or fever ≥1 day after effective antibiotics occurred in 9/13 episodes (7/7 slow decay and 2/6 rapid decay; P = 0.02).

Clinical and antibiotic characteristics differ in slow decay of cell-free DNA episodes

In this convenience sample of pediatric patients with leukemia, slow pathogen DNA decay by mcfDNA-seq correlated with persistent fever or BSI.

Antibiotic killing concentration dependency was an indicator of slow decline, suggesting that specific bactericidal activity for a given organism may influence decay of cell-free DNA.

Use of mcfDNA-seq as a guide for duration of antibiotic therapy in immunocompromised hosts should be investigated, aiming to decrease inappropriate antibiotic therapy while preventing treatment failure.

	Rapid Decline ¹	Slow Decline ²
Organisms Identified count		
C. jeikeium		1
E. coli	3	
E. faecium	1	1
R. mucilaginosa	2	
S. epidermidis	1	5
Characteristics of DNA decay mean (SE)		
Rate of decline (log ₁₀ MPM/day)	0.82 (0.09)	0.14 (0.04)
Maximum Concentration of DNA (log ₁₀ MPM)	4.36 (0.46)	5.00 (0.35)
Genome Size of Organism Identified (Mb)	4.23 (0.82)	2.56 (0.06)
Treatment Characteristics mean(SE)		
Days to Appropriate Antibiotics	0.33 (0.21)	0.14 (0.14)
Clinical Treatment Response Indicators mean (SE)		
Days of Fever after Appropriate Antibiotics	0.17 (0.17)	3.14 (1.16)
Days of Positive Blood Culture after Appropriate Antibiotics	0.5 (0.5)	1.57 (7.8)
Antibiotic Killing Dependence count		
Concentration (e.g. Vancomycin)	1	6
Time (e.g. Cefepime, Meropenem)	4	0
Combination Antibiotics (Concentration and Time)	1	1

Table 1: Clinical and microbiological characteristics of bacteremias in patients with relapsed or refractory leukemia. ¹ Slow decline defined as < 0.5 log₁₀ MPM/day