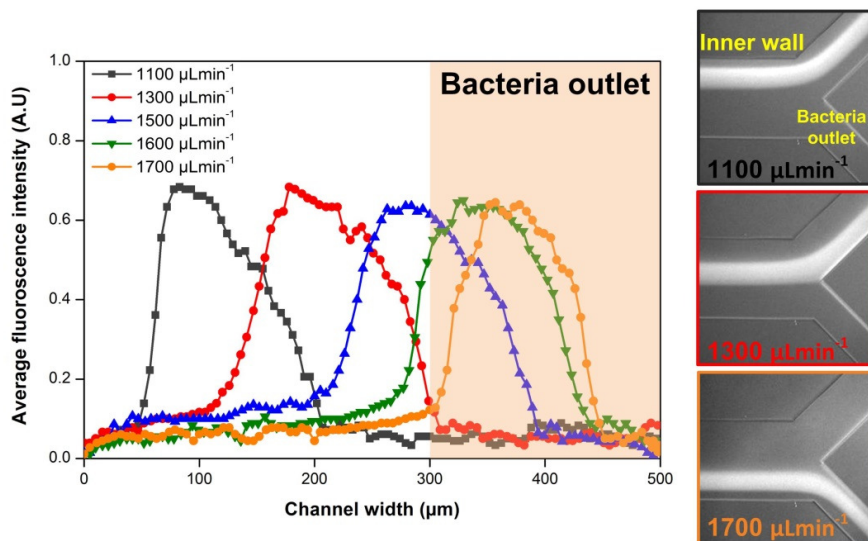


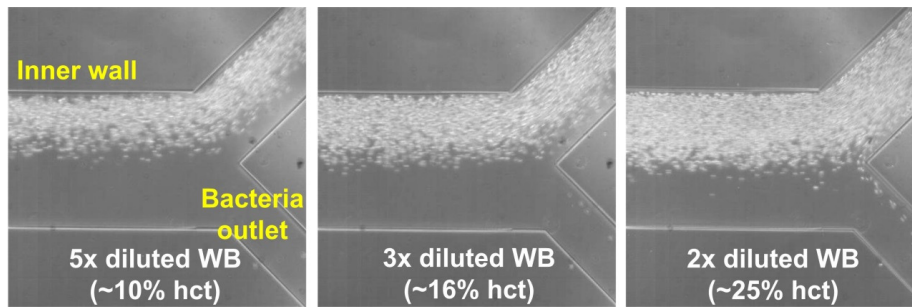
# Direct detection and drug-resistance profiling of bacteremias using inertial microfluidics

Han Wei Hou, Roby P. Bhattacharyya, Deborah T. Hung, Jongyoon Han

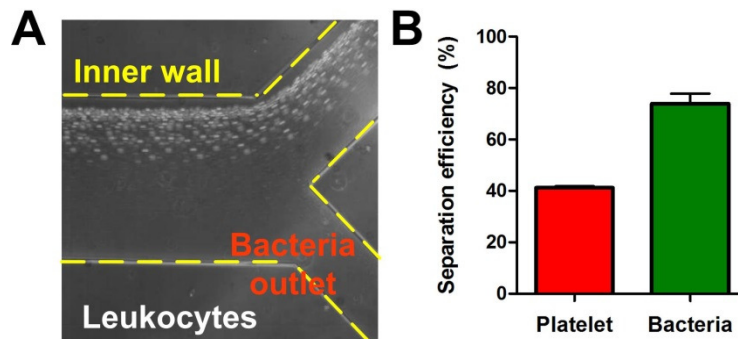
## Supplementary information



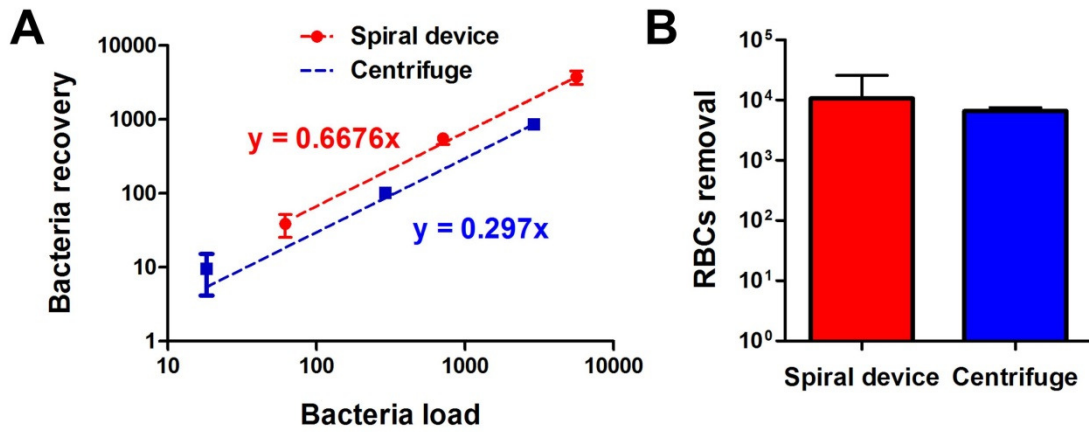
**Figure S1** Average fluorescence intensity linescans indicating normalized distribution of FITC-conjugated *E. coli* bioparticles across the channel width (x-axis origin represents inner wall) prior the outlet bifurcation at different flow rates. Approximate position of the bacteria outlet (outer wall region) is shown on the plot. Corresponding average composite images illustrate efficient collection of bacteria into bottom (bacteria) outlet at a flow rate of 1700  $\mu\text{Lmin}^{-1}$ .



**Figure S2** High speed images indicate broadening of RBCs occupied regions at the channel inner wall for increasing hematocrit (hct) prior outlet bifurcation at a sample flow rate of  $150 \mu\text{Lmin}^{-1}$  ( $1500 \mu\text{Lmin}^{-1}$  for sheath flow (1:10)). 3x diluted whole blood (WB) was chosen for subsequent experiments which resulted in minimal RBCs entering the bacteria outlet (bottom).



**Figure S3** (A) Averaged composite high speed image represents the tight focusing of larger leukocytes at the inner wall which resulted in complete removal at the bacteria outlet. Yellow dashed lines indicate position of channel walls. (B) Histogram plot showing efficient bacteria recovery (>70%) with lower platelet recovery (~40%) in the sorted samples collected from the bacteria outlet.



**Figure S4** Separation performance comparison between spiral device and centrifuge. **(A)** Higher bacteria recovery was achieved with the spiral device over a range of bacterial loads (*E. coli*) spiked in 1 mL of whole blood. **(B)** Histogram plot indicating similar RBCs removal efficiency ( $\sim 10^4$ -fold) for both spiral device and centrifuge.

**Figure S5.** rRNA probes for specific detection of *P. aeruginosa* (PsA), *E. coli* (Ec), and *K. pneumoniae* (Kp). Probe targets are highlighted in multiple sequence alignments of (A) 16S and (B) 23S rRNA subunits from these three species. Green = PsA-specific probe; Blue = Ec-specific probe; Red = Kp-specific probe; Purple = dual-specificity probe for either Ec or Kp but not PsA. Mismatches in other species corresponding to the region recognized by each probe are shown in lowercase letters; asterisks below the alignment correspond to regions of conservation across the three species. Specific probe sequences designed to target these regions are shown in supplementary Table S1.

**A.**

```

PsA_16S -----AACGAGAGGTTGCTGTAGAATGCGCGCCTC
Ec_16S  AGAAAAATCCTGAAATTCAGGGTTGACTCTGAAAGAGGAAAGCGTAATATACGCCACCTC
Kp_16S  -AGAAAAATCCTGAAAAACGGGTTGACTCTGAAAGAGGATAGCGTAATATACGCCACCTC
                *****      ***  **  **  **  **  **

PsA_16S  GGT--GAGACGAAAGCCTTGACCAACTGCTCTTTAACAAGTCGAATCAAGCAATTCGT
Ec_16S  GCGACAGTGCCTAAAGCGCGTCGCAACTGCTCTTTAACAATTTATCA--GACAATCTGT
Kp_16S  GCGACAGAGCGCTAAAGCGCGTCGCAACTGCTCTTTAACAATTTATCA--GACAATCTGT
                *      *  *****      ***** *      ****  **

PsA_16S  GTGGGTGCTTGTGATGTAAGACTGGTGATCGCAAGATTATCAGCAACGCAAGTAACACTC
Ec_16S  GTGGGCACTCGAAG---ATAC-----GGATTCTTAACG-----T-
Kp_16S  GTGGGCACTCAAAGTG--ACAT-----GGATTCTTAACG-----T-
                *****  **      *  *      ***** *  *  *      *

PsA_16S  GTGAATTCGAGAGTTTTATCTCTTTTAAAGAGAATGCGATTGCTGAGCCAAGTTTAGGGT
Ec_16S  CGCAAGACGAAAAATGAATACCAAGTCTCAAGAGTGAACACGTAATTCATTACGAAGCTT
Kp_16S  CCTCGGACGAAAAATGAATACCAAGTCTCAAGAGTGAACACGTAATTCATTACGAAGTTT
                *** *  *  ** *  *      *** **      *      *      ** *

PsA_16S  TTTCTCAAACCCAAGCAGTATTGAACTGAAGAGTTTGATCATGGCTCAGATTGAACGCT
Ec_16S  AATCTTTGAGCATCAAACCTTTAAATTGAAGAGTTTGATCATGGCTCAGATTGAACGCT
Kp_16S  AATCTTTGAGCATCAAACCT--TAAATTGAAGAGTTTGATCATGGCTCAGATTGAACGCT
                *      *  *      *  *  *  *****

PsA_16S  GGCGGCAGGCCTAACACATGCAAGTCGAgCGGatgaAGGgAGCtt-----GCTcCtgga
Ec_16S  GGCGGCAGGCCTAACACATGCAAGTCGAAGCGGTAACAGGAAGCAGCTTGCTGCTTCGCTG
Kp_16S  GGCGGCAGGCCTAACACATGCAAGTCGAgCGGTAgCAcagAGagc--TtgctCTcgGgTG
                ***** *****  ***      *  **      **

PsA_16S  accAGcGGCGGACGGGTGAGTAATGCCTAGGAATCTGCCTGGTAGTGGGGGATAACGTCC
Ec_16S  ACGAGTGGCGGACGGGTGAGTAATGTCTGGGAAACTGCCTGATGGAGGGGGATAACTACT
Kp_16S  ACGAGcGGCGGACGGGTGAGTAATGTCTGGGAAACTGCCTGATGGAGGGGGATAACTACT
                ** ***** ** ***** ***** *  * ***** *

PsA_16S  GGAAACGGGCGCTAATACCGCATACGTCCTGAGGGAGAAAGTGGGGATCTTCGGACCTC
Ec_16S  GGAAACGGTAGCTAATACCGCATAcgcTCgcAAGaccAAAGaGGGGGAcCTTCGGgCCTC
Kp_16S  GGAAACGGTAGCTAATACCGCATAatgTCgcAAGaccAAAGTGGGGGAcCTTCGGgCCTC
                ***** ***** *  *  *      *** ***** ***** ***

PsA_16S  ACGCTATCAGATGAGCCTAGGTCGGATTAGCTAGTTGGTGGGGTAAAGGCCTACCAAGGC
Ec_16S  ttGCcATCGGATGtGCCcAGaTgGGATTAGCTtGTTGGTGGGGTAAcGGctcACCAAGGC
Kp_16S  AtGCcATCAGATGtGCCcAGaTgGGATTAGCTAGTAgGTGGGGTAAcGGctcACctAGGC
                ** *** ** ** ** * ***** ** ***** ** ** ** **

PsA_16S  GACGATCCGTAACCTGGTCTGAGAGGATGATCAGTCACACTGGAAGTGGAGACACGGTCCAG
Ec_16S  GACGATCCCTAGCTGGTCTGAGAGGATGACCAGCCACACTGGAAGTGGAGACACGGTCCAG
Kp_16S  GACGATCCCTAGCTGGTCTGAGAGGATGACCAGCCACACTGGAAGTGGAGACACGGTCCAG
                ***** ** ***** ***** ***** *****

```



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Ec\_16S TCCTTTGTTGCCAGCGGTCCGGCCGGAACTCAAAGGAGACTGCCAGTGATAAACTGGAG  
Kp\_16S TCCTTTGTTGCCAGCGGTCCGGCCGGAACTCAAAGGAGACTGCCAGTGATAAACTGGAG  
\*\*\*\*\* \*\* \* \*\*\*\*\* \*\* \* \*\* \* \*\*\*\*\* \*\*\*\*\* \*\* \* \*\* \* \*\* \*

PsA\_16S GAAGGTGGGGATGACGTCAA**GTATCATGGCCCTTACGGCCAGGGCTACACACGTGCTAC**  
Ec\_16S GAAGGTGGGGATGACGTCAAAGTCATCATGGCCCTTACGaCCAGGGCTACACACGTGCTAC  
Kp\_16S GAAGGTGGGGATGACGTCAAGTCATCATGGCCCTTACGaCCAGGGCTACACACGTGCTAC  
\*\*\*\*\*

PsA\_16S **AATGGTTCGGTACAAAGGGTTGCCAAGCCGCGAGGTGGAGCTAATCCATAAAACCGATCG**  
Ec\_16S AATGGcgcataTACAAAGaGaaGCgAcctCGCGAGagcaAGCggAcCtCATAAAgtgCGTCG  
Kp\_16S AATGGcataTACAAAGaGaaGCgAcctCGCGAGagcaAGCggAcCtCATAAAgtatgTCG  
\*\*\*\*\* \* \* \* \* \*\*\*\*\* \*\* \* \* \*\*\*\*\* \*\* \*

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Ec\_16S TAGTCCGGATTGGAGTCTGCAACTCGACTCCATGAAGTCGGAATCGCTAGTAATCGTGGA  
Kp\_16S TAGTCCGGATTGGAGTCTGCAACTCGACTCCATGAAGTCGGAATCGCTAGTAATCGTAGA  
\*\*\*\*\* \* \*\*\*\*\* \* \*\*\*\*\* \* \*\*\*\*\* \* \*\*\*\*\* \* \*\*\*\*\* \*

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Ec\_16S TCAGAATGCCACG**GTGAATACGTTCCCGGGCCTTGTACACACCGCCCGTCACACCATGGG**  
Kp\_16S TCAGAATGCTACG**GTGAATACGTTCCCGGGCCTTGTACACACCGCCCGTCACACCATGGG**  
\*\*\*\*\* \*\*\*\*\*

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Kp\_16S **AGTGGGTTGCAAAAGAAGTAGTAGCTTAACCTTCGGGAGGGCGCTTACCACT**TTGTGAT  
\*\*\*\*\* \*\*\*\*\* \*\* \* \*\*\*\*\* \*\* \* \* \* \*\*\*\*\* \*\*\*\*\*

PsA\_16S TCATGACTGGGGTGAAGTCGTAACAAGGTAGCCGTAGGGGAACCTGCGGTGGATCACCT  
Ec\_16S TCATGACTGGGGTGAAGTCGTAACAAGGTAAACCGTAGGGGAACCTGCGGTGGATCACCT  
Kp\_16S TCATGACTGGGGTGAAGTCGTAACAAGGTAAACCGTAGGGGAACCTGCGGTGGATCACCT  
\*\*\*\*\* \*\*\*\*\*

PsA\_16S CCTTAATCGAAGATCTCAGCTTCTTCATAAGCTCCACACGAA-----TTGCTTGATTCA  
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Kp\_16S CCTTACCTTAAAGAACTGCCTTTGCAGTGCTCACACAGATTGTCTGATGAAAAACAGCA  
\*\*\*\*\* \*\* \* \* \* \* \* \* \* \* \* \* \* \* \* \* \* \*

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Kp\_16S GTAAAACCTCTACAGGCTTGTAGCTCAGTTGGTTAGAGCGCACCCCTGATAAGGGTGAG  
\* \* \* \* \* \*\*\*\*\*

PsA\_16S GTCGGCAGTTCGAATCTGCCAGACCCACCAATTGTTGGTGTGCTG-----  
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Kp\_16S GTCGGTGGTTCAGTCCACTCAGGCTACCAAATTTGCGAAGCAAATTTGAAGAGTTGC  
\*\*\*\*\* \* \* \* \* \* \* \* \* \* \* \* \* \* \* \*

PsA\_16S CGTGATCCGATACGGGGCCATAGCTCAGCTGGGAGAGCGCCTGCTTTGCACGCAGGAGGT  
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Kp\_16S AAA-----CGATGGGGCTATAGCTCAGCTGGGAGAGCGCCTGCTTTGCACGCAGGAGGT  
\* \*\*\*\*\*

PsA\_16S CAGGAGTTCGATC  
Ec\_16S -----  
Kp\_16S CTGCGG-----



PsA\_23S GACTGGAGGACCGAACCCACTCCCCTTAAAAAGGTAGGGGATGACTTGTGGATCGGAGTG  
Ec\_23S AACTGGAGGACCGAACCCACTAATGTTGAAAAATTAGCGGATGACTTGTGGCTGGGGGTG  
Kp\_23S AACTGGAGGACCGAACCCACTAATGTTGAAAAATTAGCGGATGACTTGTGGCTGGGGGTG  
\*\*\*\*\* \*\* \*\*\*\*\* \*\*

PsA\_23S AAAGGCTAATCAAGCTCGGAGATAGCTGGTTCTCCTCGAAAGCTATTTAGGTAGCGCCTC  
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\*\*\*\*\* \*\* \*\*\*\*\*

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Kp\_23S GTGAATTCATCTCCGGGGGTAGAGCACTGTTTCGGCTAGGGGGTCATCCCGACTTACCAA  
\*\* \*\* \*\* \*\*\*\*\*

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\*\*\*\*\* \*\*\*\*\* \*\* \* \*\* \*\*\*\*\*

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Kp\_23S ACGTCCGTCGTGAAGAGGGAAACAACCCAGACCAGCTAAGGTCCCAAAGTCATGGTT  
\*\*\*\*\* \*\*\*\*\* \*\*\*\*\*

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Kp\_23S AAGTGGGAAACGATGTGGGAAGGCACAGACAGCCAGGATGTTGGCTTAGAAGCAGCCATC  
\*\*\*\*\* \*\*\*\*\* \*\*\*\*\* \*\*\*\*\* \*\*\*\*\*

PsA\_23S CTTTAAAGAAAGCGTAATAGCTCACTAGTCGAGTCGGCCTGCGCGGAAGATGTAACGGGG  
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Kp\_23S ATTTAAAGAAAGCGTAATAGCTCACTGGTCGAGTCGGCCTGCGCGGAAGATGTAACGGGG  
\*\*\*\*\* \*\*\*\*\* \*\*\*\*\*

PsA\_23S CTCAAACCACACCCGAAGC--TGCGGGTGTACGTAAGTGACGCGGTAGAGGAGCGTTC  
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Kp\_23S CTA AACCATGCACCGAAGCTGCGGCAGCGACACTATGTGTTGTTGGGTAGGGAGCGTTC  
\*\* \* \* \* \* \* \*\*\*\*\*

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Kp\_23S TGTAAGCCTGCGAAGGTGTGCTGTGAGGCATGCTGGAGGTATCAGAAGTGC GAATGCTGA  
\*\*\*\*\* \*\*\*\*\* \* \* \* \* \*

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Kp\_23S CATAAGTAACGATAAAGCGGGTGAAAAGCCCGCTCGCCGGAAGACCAAGGGTTCCTGTCC  
\*\*\* \*\*\*\*\* \*\* \* \* \*\*\*\*\* \* \* \*\*\*\*\* \*\*\*\*\*

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\*\*\*\*\* \*\*\*\*\* \*\*\*\*\* \*\*\*\*\* \*\*\*\*\*

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\*\*\*\*\* \*\*\*\*\* \* \* \*\*\*\*\*

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\* \*\* \*\* \*\*\*\*\* \*\*\*\*\* \*\*\*\*\* \*\*\*\*\* \*\*\*\*\*



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Kp\_23S GATAATCAAGGCTGAGGTGTGATGACGAGGCA---CTACGGTGCTGAAGTAACAAATGCC  
\*       \*\*\*\*\*   \*       \*\*\*\*\*   \*       \*   \*   \*       \*\*\*\*\*

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Ec\_23S CTGCTTCCAGGAAAAGCCTCTAAGCATCAGGTAACATCAAATCGTACCCCAACCGACAC  
Kp\_23S CTGCTTCCAGGAAAAGCCTCTAAGCATCAGGTAACATCAAATCGTACCCCAACCGACAC  
\*\*\*\*\*   \*\*\*\*\*   \*\*\*\*\*   \*\*\*\*\*           \*\*   \*\*\*\*\*

PsA\_23S AGGTGGTCCGGTAGAGAATACCAAGGCGCTTGAGAGAACTCGGGTGAAGGAACTAGGCAA  
Ec\_23S AGGTGGTCCGGTAGAGAATACCAAGGCGCTTGAGAGAACTCGGGTGAAGGAACTAGGCAA  
Kp\_23S AGGTGGTCCGGTAGAGAATACCAAGGCGCTTGAGAGAACTCGGGTGAAGGAACTAGGCAA  
\*\*\*\*\*   \*\*\*\*\*

PsA\_23S AATGGCACCGTAACTTCGGGAGAAGGTGCGCCGGCTAGGGTGAAG---GATTTACTCCGT  
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Kp\_23S AATGGTGCCGTAACTTCGGGAGAAGGCACGCTGGTGTGTAGGTGAAGTCCCT-GCGGATG  
\*\*\*\*\*   \*\*\*\*\*       \* \*       \*       \*       \*       \*

PsA\_23S AAGCTCTGGCTGGTTCGAAGATACCAGGCCGTGCGACTGTTTATTA AAAACACAGCACTG  
Ec\_23S GAGCTGAAATCAGTTCGAAGATACCAGCTGGCTGCAACTGTTTATTA AAAACACAGCACTG  
Kp\_23S GAGCTGAGACCAGTTCGAAGATACCAGCTGGCTGCAACTGTTTATTA AAAACACAGCACTG  
\*\*\*               \*\*\*\*\*       \*\*\*\*\*   \*\*\*\*\*

PsA\_23S TGCAAACACGAAAGTGGACGTATAGGGTGTGACGCCTGCCCGGTGCCGGAAGGTTAATTG  
Ec\_23S TGCAAACACGAAAGTGGACGTATAGGGTGTGACGCCTGCCCGGTGCCGGAAGGTTAATTG  
Kp\_23S TGCAAACACGAAAGTGGACGTATAGGGTGTGACGCCTGCCCGGTGCCGGAAGGTTAATTG  
\*\*\*\*\*   \*\*\*\*\*

PsA\_23S ATGGGGTTAGCGC--AAGCGAAGCTCTTGATCGAAGCCCCGGTAAACGGCGGCCGTAAC  
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Kp\_23S ATGGGGTTATCCGTAAGGAGAAGCTCTTGATCGAAGCCCCGGTAAACGGCGGCCGTAAC  
\*\*\*\*\* \* \*       \* \*   \*\*\*\*\*

PsA\_23S ATAACGGTCCTAAGGTAGCGAAATTCCTTGTGCGGGTAAGTTCGACCTGCACGAATGGCG  
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Kp\_23S ATAACGGTCCTAAGGTAGCGAAATTCCTTGTGCGGGTAAGTTCGACCTGCACGAATGGCG  
\*\*\*\*\*

PsA\_23S TAACGATGGCGCGCTGTCTCCACCCGAGACTCAGTGA AATGAAATCGCTGTGAAGATG  
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\*\*\*   \*\*\*\*\*       \*\*\*\*\*

PsA\_23S CAGTGTATCCGCGGCTAGACGGAAGACCCCGTGAACCTTTACTGTAGCTTTGCACTGGA  
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Kp\_23S CAGTGTACCCGCGCAAGACGGAAGACCCCGTGAACCTTTACTATAGCTTGACACTGAA  
\*\*\*\*\*   \*\*\*\*\*   \*\*\*\*\*   \*\*\*\*\*       \*\*\*\*\*   \*\*\*\*\*

PsA\_23S CTTTGAACCTGCTTGTGTAGGATAGGTGGGAGGCTTTGAAGCGTGGACGCCAGTTCGCGT  
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Kp\_23S CATTGAGCCTTGATGTGTAGGATAGGTGGGAGGCTTTGAAGCGTGGACGCCAGTTCGCGT  
\*   \*\*\*\*\*       \*\*\*\*\*   \*\*\*\*\*   \*\*\*\*\*       \* \*   \*

PsA\_23S GGAGCCATCCTTGAAATACCACCCTGGCATGCTTGAGGTTCTAACTCTGGTCCGTAATCC  
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\*\*\*\*\*   \*\*\*\*\*       \* \*   \*\*\*\*\*       \* \*   \* \*   \*

PsA\_23S GGATCGAGGACAGTGTATGGTGGGAGTGTGACTGGGCGGTCTCCTCCTAAAGAGTAAC  
Ec\_23S GGGTTGCGGACAGTGTCTGGTGGGAGTGTGACTGGGCGGTCTCCTCCTAAAGAGTAAC  
Kp\_23S GGGTTGCGGACAGTGTCTGGTGGGAGTGTGACTGGGCGGTCTCCTCCTAAAGAGTAAC  
\* \*   \* \*   \*\*\*\*\*   \*\*\*\*\*   \*\*\*\*\*   \*\*\*\*\*       \*\*\*\*\*   \*\*\*\*\*

PsA\_23S GGAGGAGTACGAAGGTGCGCTCAGACCGGTTCGAAATCGGTTCGAGAGTATAAAGGCAAA  
Ec\_23S GGAGGAGCACGAAGGTTGGCTAATCTGGTTCGACATCAGGAGGTTAGTGCATGGCATA  
Kp\_23S GGAGGAGCACGAAGGTTAGCTAATCTGGTTCGACATCAGGAGGTTAGTGCATGGCATA  
\*\*\*\*\*

PsA\_23S AGCGCGCTTACTGCGAGACAGACACGTCGAGCAGGTACGAAAGTAGGTCTTAGTGATCC  
Ec\_23S AGCCAGCTTACTGCGAGCGTGACGGCGCGAGGTTCGAAAGCAGGTTCATAGTGATCC  
Kp\_23S AGCTAGCTTACTGCGAGCGTGACGGCGCGAGGTTCGAAAGCAGGTTCATAGTGATCC  
\*\*\*\*\*

PsA\_23S GGTGGTTCTGTATGGAAGGGCCATCGCTCAACGGATAAAAGGTAAGTCCGGGGATAACAGG  
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Kp\_23S GGTGGTTCTGAATGGAAGGGCCATCGCTCAACGGATAAAAGGTAAGTCCGGGGATAACAGG  
\*\*\*\*\*

PsA\_23S CTGATACCGCCCAAGAGTTCATATCGACGGCGGTGTTTGGCACCTCGATGTCGGCTCATC  
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\*\*\*\*\*

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Kp\_23S ACATCTGGGGCTGAAGTAGGTCCCAAGGGTATGGCTGTTTCGCCATTTAAAGTGGTACGC  
\*\*\*\*\*

PsA\_23S GAGCTGGGTTTAGAACGTCGTGAGACAGTTCGGTCCCTATCTG**CCGTGGACGTTTGAGAT**  
Ec\_23S GAGCTGGGTTTAGAACGTCGTGAGACAGTTCGGTCCCTATCTGCCGTGGgCGcTgGAGAA  
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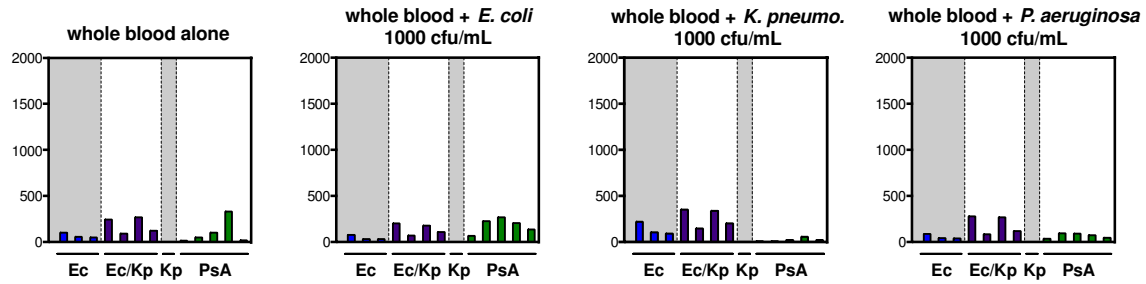
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Kp\_23S TTGAGgGGGGCTGCTCCTAGTACGAGAGGACCGGAGTGGACGcAtCaCTGGTGTTCgGGT  
\*\*\*\*\*

PsA\_23S **TGTCACGCCAGTGGCATTGCCGG**GTAGCTATGTTTCGGAAGATAAACCGCTGAAAGCATC  
Ec\_23S TGTCAtGCCAaTGGCAcTGCCcGGTAGCTAAATGCGGAAGAGATAAGTGCTGAAAGCATC  
Kp\_23S TGTCAtGCCAaTGGCAcTGCCcGGTAGCTAAATGCGGAAGAGATAAGTGCTGAAAGCATC  
\*\*\*\*\*

PsA\_23S TAAGCGGAAACTTGCCCTCAAGATGAGATCTCACTGGGAacTTgAtTCcCCTGAAGGgcC  
Ec\_23S TAAGCACGAAACTTGCCCCGAGATGAGTTCTCCCTGA**ctCcTTgAGagTCCTGAAGGAAC**  
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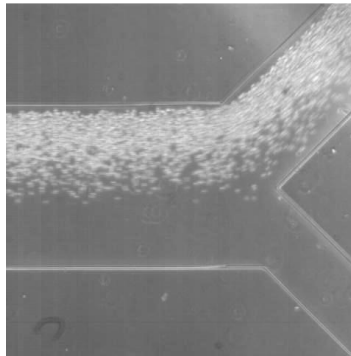
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Kp\_23S **GTTAAAGACGATGACGTTGATAGGCCGGGTGTGTAAGCGCAGCGATGCGTTGAGCTAACC**  
\*\*\*\*\*

PsA\_23S aGTACTAATtgcCCGTGAGGCTTGACCAT--  
Ec\_23S **GGTACTAATGAACCGTGAGGCTTAACCTT--**  
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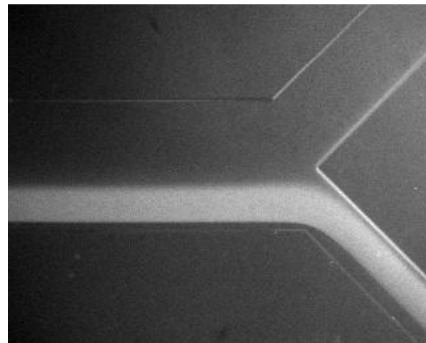
**Figure S6** Similar background transcriptional signals from nCounter probes between whole blood and whole blood spiked with 1000 cfu/mL for different bacteria strains.

## SI Movie Legends



### Movie S1

High speed video (6400 fps) illustrating inertial focusing of RBCs (3x diluted whole blood, ~15% hct) at the inner channel wall at DC 1 and continuous collection into the waste outlet (upper).



### Movie S2

Fluorescent video illustrating focusing of Alexa Fluor® 488 *E. coli* (K-12 strain) BioParticles® conjugates spiked in PBS at the outer wall at DC 1 and collection into the bacterial outlet (bottom).

**Table S1.** Sequences for rRNA probes targeting the 16S or 23S rRNA subunits from *E. coli*, *K. pneumoniae*, or *P. aeruginosa*. Nucleotide positions are relative to the annotated reference genome sequences of *E. coli* strain EcHS, *K. pneumoniae* strain MGH78578, and *P. aeruginosa* strain PAO1, respectively, from the NCBI database.

PROBE	SPECIES RECOGNIZED	GENE	NUCLEOTIDES	PROBE 1 SEQUENCE	PROBE 2 SEQUENCE
<i>E. coli</i>	Ec_1	<i>E. coli</i>	16S rRNA	5-105	TGTTGTTAGGCGCTGCCCGCCAGCGTTCAATCTGAGCCATGATCAAACTCTTCCACTCGTCAGCGAAACACAGCAAGCTGGCTTCCTGTTCACCGTTTCGACTTGCA
	Ec_2	<i>E. coli</i>	16S rRNA	935-1035	GGTAAAGGTTCTTTCGCGTTGCAFCGAATTAACCCACATGCTCCACCGGCTTCCCGAAGGCACATTCTCATCTCTGAAACATCCCGTGGATGTCAAGACCA
	Ec_3	<i>E. coli</i>	23S rRNA	254-354	ACGCTTCCACTAACACACACACTGATTCAGGCTCTGGGCTGCCCGTACTCCATTTTGTGTACGGGGCTGTACCCCTGTATCGCACGCCCTTTCCAG
<i>Klebs. pneumoniae</i>	Ec/Kp_1	<i>E. coli, Klebs. pneumoniae</i>	16S rRNA	1370-1470	ACTCCCATGGTGTGACGGGGCGGTGTGTACAAAGGCCCGGAACTGATTCAGTGGTAAAGCGCCCTCCCGAAGGTTAAGCTACCTACTTCTTTTGCAACCC
	Ec/Kp_2	<i>E. coli, Klebs. pneumoniae</i>	23S rRNA	5-105	CTTTCATCGCCTCTGACTGCCAGGGCATCCACCGTGTACGCTTAGTCGGCTGTAAACGGTTCATACCTTACCGACGCTTTTCGACAGATTAGCAGCT
	Ec/Kp_3	<i>E. coli, Klebs. pneumoniae</i>	23S rRNA	1210-1310	AATGACAGCATTCGGACTTCTGTATACCTCCAGCATGCCCTCACACACACAAACCCCTGGTCTTCCGGGAGCGGGGCTTTTCCACCGCTTTATTCGTTACTTT
	Ec/Kp_4	<i>E. coli, Klebs. pneumoniae</i>	23S rRNA	2790-2890	CGGCCTATCAACGTCGTCGTCCTTCAAGTTCCTTCAGGAGACTTAAAGTACAGGTTCAATAGTACCCGGTTAGCTCAACGGCATCGCTGGCTTACACACC
<i>P. aeruginosa</i>	Kp_1	<i>Klebs. pneumoniae</i>	23S rRNA	260-360	TTCCAGACCGTTCCACTAACACACAAAGCTGATTCAGACTCTGGGCTGCTACAGCCCTGTCCATTTTGGTGTAGGGGACTATCACCCCTGTACCCGTCGGACT
	PsA_1	<i>P. aeruginosa</i>	16S rRNA	165-265	CATCTGATAGCGTGAGGTCGGAAGAATCCCCACATTTCTCCCTCAGGACCGTCCCTTGGTAGGCGCTTTACCCACCAACTAGCTAAATCCGACCTAGGCT
	PsA_2	<i>P. aeruginosa</i>	16S rRNA	1190-1290	ACCGACCAATTGTAGCACGCTGTGTAGCCCTGGCCGTAAGGGCCATGATGACGATCGGTTTTTATGGGATTAGCTCCACCTCGGGGCTTGGCAACCCCTTTGT
	PsA_3	<i>P. aeruginosa</i>	23S rRNA	525-625	CGCTGACCCATTTACAAAAGGTACGCAGTCAACCTAAACAAGTAGGCTCGCTTTCGCTACGCCCTACCCCTATACGGTTAAGCTTGCCTGAAATATAAGT
	PsA_4	<i>P. aeruginosa</i>	23S rRNA	1375-1475	AGCCTTCTCGTCCCTCCATCGCAGTAAACCAAGTACAGGAATATTAAAGATTTACAGCCTAACCCCTTAAACTTGGACAACCAACGGCAAGCTGGCT
PsA_5	<i>P. aeruginosa</i>	23S rRNA	2605-2705	CGGTCCTCTCGTACTAGGAGCAGCCCTCTCAAAATCTCAAAACGTCCACCGGCAATGCCACTGGCTGACAAACGGAAACACCAGAGGTTCTGTCCTC	

**Table S2.** Sequences for mRNA probes targeting ciprofloxacin-responsive ("R\_") and control ("C\_") transcripts from *E. coli*. Nucleotide position is relative to the annotated reference genome sequence of *E. coli* strain EchS from the NCBI database.

PROBE	GENE	NUCLEOTIDES	PROBE 1 SEQUENCE	PROBE 2 SEQUENCE
R_1	<i>phoB</i>	50-150	CTTCCGCTTCGACCCGGGTGAAAGCCATTTTGTTCGAGCACGAAGCAGACAAATTAATCCGGCCAGGGTTCATTCAGTTGATTCACAGCACTGTCAATAAT	AGGGACCTTTCGTGATGATTCACCCGCATGTTTTCACGATCGCTAACCCCTAA
R_2	<i>phoR</i>	1114-1214	TCCGCTACCGCCGGTTTGCCGGGAACGGCTTTATCAACGCGATAAAAAATAGACAACCTTCACTGATAAGCCCGCTGTAGTGTTCCTGTAGAGACA	GTAACAATGGCAACAGTAGAAGTTGCCTCATCATGGGCTGATCTTCGGCGA
R_3	<i>sulA</i>	63-163	GACATTTTCTCTTCGCCCTCACGATGACTACGTATTTAAGCTTCAACCCCGGATCAAGCATTTTGTCAATAGCATCGGCTTCCCTTTTGGTGGTAAAC	CCGATCAAGCATTTTGTCAATAGCATCGGCTTCCCTTTTGGTGGTAAAC
R_4	<i>yebG</i>	3-103	ATGGAACGGTCTTTCACCCAGGCGCATGATGGAGCCTTTACCAAATTTGTCAAGCGGATATCCAGTGAAGCCGAAACCCGGTAGAGATGGTTTCCACATCC	CAAGCGGATATCCAGTGAAGCCGAAACCCGGTAGAGATGGTTTCCACATCC
R_5	<i>recA</i>	57-157	GTTCAGTTCTTCAAATGGTGATTTCTGTAAAAGCCAGTCGGCTCTCCAGCGAATGATCGCGCAGTTTTCCGCAATCTCCATTTCAITGAGCGGTCACCCGTGAC	AGATGATCGCGCAGTTTTCCGCAATCTCCATTTCAITGAGCGGTCACCCGTGAC
R_6	<i>EchS_A3544</i>	34-134	GAAAATGGCGTTGATGTACTTGTATTAAGGTAAATGCCATCGCCCGTACTTAAAGATCGCCGGGTGTTGTTCATGGTCAGAACAAATAATCGACAGGCTTGG	AGAAATCGCCGGGTGTTGTTCATGGTCAGAACAAATAATCGACAGGCTTGG
C_1	<i>rcsB</i>	220-320	GTTCCGGCGCTTCGAAAGAGATAATCCTTGGTAAFAAATAACGTTGGATCTGGGATCAAGTTTTAACTTTCIGGTTGCCAATCTTTCITGGAAAAACGTGCGGGCGC	AGATCAAGTTTTAACTTTCIGGTTGCCAATCTTTCITGGAAAAACGTGCGGGCGC
C_2	<i>secB</i>	31-131	TTTCAACGTCAGGGCGCAATTTTCAGAACCGACACGGTTCAGAGAACTTAAAGATCGGTCACGTA	GCGTCAACGTA
C_3	<i>hflC</i>	361-461		