

1 **Supplementary Text:**

2 A number of the singletons also appear to be rather closely related to isolates in other STs  
3 within this collection based on the cladogram in Fig. 1 including ST<sup>T</sup>-T517 (n=1), 99.8%  
4 identical (SLV differing by four nt in *salA*) to ST<sup>T</sup>-T525 (n=2); ST<sup>T</sup>-T505 (n=1), 99.9%  
5 identical (DLV differing by one nt in *ace* and one nt in *salA*) to ST<sup>T</sup>-T509; ST<sup>T</sup>-T514  
6 (n=1), 99.9% identical (SLV differing by a 6 nt deletion in *lsa*) to ST<sup>T</sup>-144 (n=1); ST<sup>T</sup>-  
7 T507 (n=1), 99.7% identical (SLV differing by 8 nt in *salA*) to ST<sup>T</sup>-107 (n=2); ST<sup>T</sup>-T504  
8 (n=1), 99.7% identical (SLV differing by 6 nt in *salA*) to ST<sup>T</sup>-T506 (n=1); and ST<sup>T</sup>-T512  
9 (n=1), 99.9% identical (SLV differing by one nt in *lsa*) to ST<sup>T</sup>-20 (n=4).

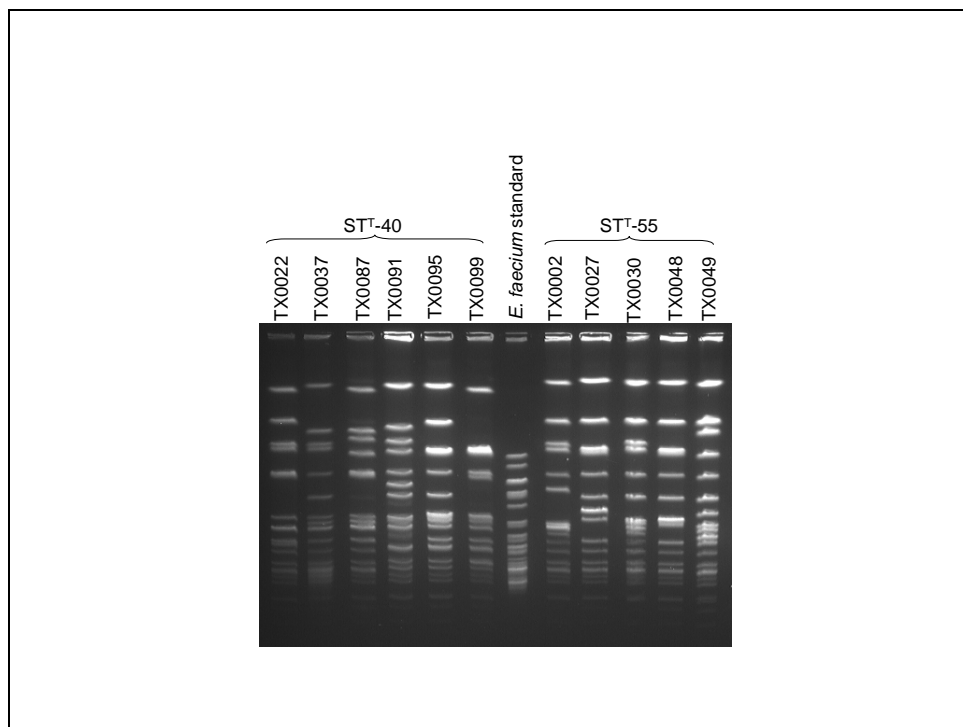
10

11

12 **Supplementary Figures:**

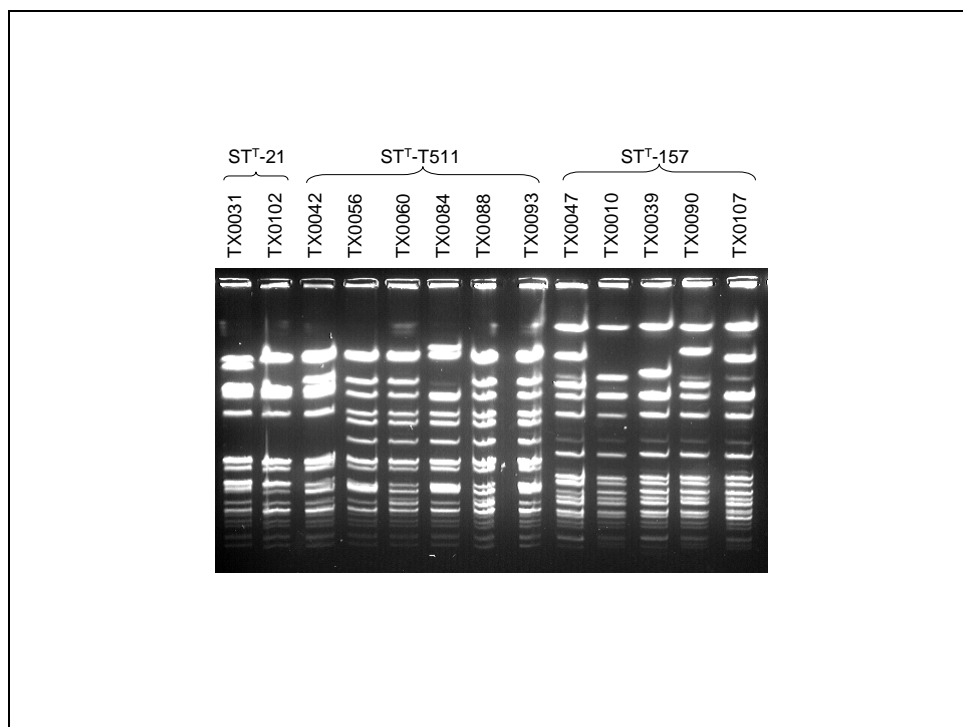
13 Fig. S1a:

14



15  
16

17 Fig. S1b:



18  
19

20 **Supplementary Tables:**21 Table S1: Infective endocarditis *E. faecalis* isolates from the United States collected between 1974-2004

Strain <sup>a</sup>	Isolation Location	Year	ST <sup>†</sup> ( <i>ace, salA, lsa</i> )	ST <sup>Mb</sup>	CPS type <sup>c</sup>	Reference
TX0104	CT	2002	2 (8, 2, 9)	(2)	5	(1)
TX0011	Houston, TX	1991	6 (3, 8, 13)	6	2	
TX0053	Houston, TX	1991	6 (3, 8, 13)	(6)	2	
TX0052	Springfield, MO	1993	6 (3, 8, 13)	(6)	2	(2,3,4,5)
TX0026	WI	1975	20 (17, 20, 3)	(19)	1	(6)
TX0023	Rochester, MN	1976	20 (17, 20, 3)	(19)	1	
TX0043	ND	1983	20 (17, 20, 3)	19	1	(1)
TX0029	Rochester, MN	1992	20 (17, 20, 3)	(19)	1	
TX0031	Rochester, MN	1992	21 (17, 1, 25)	21	1	(1)
TX0102	Rochester, MN	>1990	21 (17, 1, 25)	21	1	(1)
TX0083	Rochester, MN	>1990	30 (17, 1, 21)	-	5	
TX0109	Houston, TX	2002	33 (6, 25, 23)	59	1	(1)
TX0037	IL	1974	40 (19, 25, 19)	40	1	
TX0022	Rochester, MN	1992	40 (19, 25, 19)	40	1	
TX0087	Rochester, MN	>1990	40 (19, 25, 19)	(40)	1	
TX0091	Rochester, MN	>1990	40 (19, 25, 19)	(40)	1	
TX0095	Rochester, MN	>1990	40 (19, 25, 19)	(40)	1	
TX0099	Rochester, MN	>1990	40 (19, 25, 19)	(40)	1	
TX0007	Houston, TX	1992	44 (28, 14, 18)	(44)	1	
TX0008	Reno, NV	1992	44 (28, 14, 18)	(44)	1	
TX0009	Reno, NV	1992	44 (28, 14, 18)	(44)	1	
TX0101	Rochester, MN	2001	44 (28, 14, 18)	44	1	
TX0027	IA	1975	55 (19, 25, 21)	55	1	(1)
TX0030	IA	1983	55 (19, 25, 21)	(55)	1	
TX0002	New Haven, CT	1993	55 (19, 25, 21)	55	1	
TX0048	Boston, MA	<1980	55 (19, 25, 21)	(55)	1	
TX0049	Boston, MA	<1980	55 (19, 25, 21)	(55)	1	
TX0044	Rochester, MN	1975	62 (6, 1, 30)	62	1	
TX0103	Rochester, MN	>1990	62 (6, 1, 30)	(62)	1	
TX0125	Houston, TX	2004	107 (6, 18, 5)	107	1	
TX0086	Rochester, MN	>1990	107 (6, 18, 5)	(107)	1	
TX0017	Houston, TX	1992	144 (19, 31, 29)	144	1	(1)
TX0010	Columbia, OH	1992	157 (17, 34, 25)	157	1	
TX0039	IL	1975	157 (17, 34, 25)	(157 or 21)	1	
TX0047	Boston, MA	<1980	157 (17, 34, 25)	(157 or 21)	1	(6)
TX0090	Rochester, MN	>1990	157 (17, 34, 25)	(157 or 21)	1	
TX0107	Houston, TX	2002	157 (17, 34, 25)	21	1	
TX0001	New Haven, CT	1993	283 (17, 10, 30)	283	1	(6)
TX0073	Houston, TX	1995	283 (17, 10, 30)	(283)	1	

TX0041	Rochester, MN	1985	287 (30, 1 40)	(287)	2	
TX0055	Rochester, MN	1994	287 (30, 1, 40)	(287)	2	
TX0092	Rochester, MN	>1990	287 (30, 1, 40)	287	2	
TX0034	Rochester, MN	1978	317 (12, 33, 34)	(317)	5	(6)
TX0045	Boston, MA	<1980	317 (12, 33, 34)	317	5	(2,3,7)
TX0046	Boston, MA	<1980	317 (12, 33, 34)	(317)	5	(2,3,7)
TX0066	Houston, TX	1994	T501 (3, 1, 21)	-	5	
TX0079	Houston, TX	1996	T502 (4, 7, 35)	-	1	
TX0085	Rochester, MN	>1990	T503 (4, 32, 20)	-	2	
TX0004	New Haven, CT	1993	T504 (6, 18, 42)	-	5	
TX0014	Houston, TX	1992	T505 (6, 34, 21)	-	1	
TX0025	Rochester, MN	1984	T506 (6, 34, 42)	-	2	(6)
TX0036	Rochester, MN	1977	T507 (6, 35, 5)	-	2	
TX0065	Rochester, MN	1994	T508 (6, 36, 44)	-	5	
TX0006	New Haven, CT	1993	T509 (10, 11, 21)	-	1	
TX0003	New Haven, CT	1993	T510 (13, 2, 12)	-	1	
TX0042	Rochester, MN	1992	T511 (17, 1, 36)	21	1	
TX0056	Rochester, MN	1994	T511 (17, 1, 36)	(21)	1	
TX0060	Rochester, MN	1994	T511 (17, 1, 36)	21	1	
TX0084	Rochester, MN	>1990	T511 (17, 1, 36)	21	1	
TX0088	Rochester, MN	>1990	T511 (17, 1, 36)	21	1	
TX0093	Rochester, MN	>1990	T511 (17, 1, 36)	(21)	1	
TX0032	Rochester, MN	1976	T512 (17, 20, 45)	-	5	
TX0012	Houston, TX	1991	T513 (33, 40, 37)	-	1	(1)
TX0063	Rochester, MN	1994	T514 (19, 31, 39)	-	1	
TX0105	TN	2002	T515 (29, 3, 15)	-	5	
TX0013	Houston, TX	1992	T516 (31, 37, 41)	-	1	
TX0098	Rochester, MN	>1990	T517 (34, 42, 32)	-	2	
TX0040	IL	1974	T518 (35, 28, 28)	-	1	
TX0106	TN	2002	T519 (36, 2, 9)	-	5	
TX0020	Houston, TX	1991	T520 (37, 25, 40)	-	5	
TX0035	Rochester, MN	1992	T521 (38, 20, 38)	-	1	
TX0028	Rochester, MN	1992	T522 (39, 38, 47)	-	1	
TX0057	Rochester, MN	1994	T523 (40, 36, 43)	NT	5	
TX0061	Rochester, MN	1994	T523 (40, 36, 43)	(NT)	5	
TX0089	Rochester, MN	>1990	T523 (40, 36, 43)	NT	5	
TX0097	Rochester, MN	>1990	T524 (41, 30, 46)	-	5	
TX0108	Houston, TX	2002	T525 (34, 1, 32)	4	2	
TX0128	Houston, TX	2004	T525 (34, 1, 32)	(4)	2	
TX0018	Houston, TX	1991	T526 (3, 39, 7)		1	
TX0094	Rochester, MN	>1990	T527 (32, 41, 4)		5	
TX0019	Houston, TX	1992	T528 (32, 27, 4)	34	5	

23           <sup>a</sup> Isolates are ordered by ST<sup>T</sup> based on intragenic sequences of *ace*, *sala*, and *lsa*. ST<sup>T</sup>s not  
24 described previously in isolates typed (5) which had MLST data were assigned, when possible, the same  
25 type number as the corresponding ST<sup>M</sup>; isolates which did not have MLST data, the ST<sup>M</sup> was not in the  
26 database, or the ST<sup>M</sup> corresponded with another published ST<sup>T</sup> allelic profile were assigned an arbitrary  
27 number beginning with T500.

28           <sup>b</sup> ST<sup>M</sup>s are per the seven-housekeeping-gene scheme and assigned in accordance with the database  
29 at <http://efaecalis.mlst.net/>. Types contained within ( ) were inferred based on identical ST<sup>T</sup>s.

30 NT – a new ST<sup>M</sup> not assigned yet in the MLST database (allele types as follows: *gdh-9*, *gyd-5*, *pstS-4*, *gki-*  
31 *16*, *aroE-1*, *xpt-11*, *yqil-8*).

32           <sup>c</sup> The *cps* gene locus types were designated as follows: type 1 (corresponding to serotype A & B)  
33 possesses *cpsA* and *cpsB* only; type 2 (corresponding to serotype C) possesses *cpsA-cpsK*; type 5  
34 (corresponding to serotype D) possesses *cpsA-cpsK* except *cpsF* (8, 9,10).

35









42 **Supplementary Figure Legends:**

43 Fig. S1a - PFGE of isolates within ST<sup>T</sup>-40 (n=6) and ST<sup>T</sup>-55 (n=5) clonal lineages.

44 Black brackets group isolates with an identical ST<sup>T</sup>. Strains are listed vertically above

45 their corresponding lanes. *E. faecium* TX0016 (DO) was used as a standard.

46

47 Fig. S1b - PFGE of isolates within CC21 (n = 13). CC21 contains isolates within ST<sup>T</sup>-21

48 (n=2), ST<sup>T</sup>-T511 (n=6), and ST<sup>T</sup>-157 (n=5) clonal lineages. Black brackets group

49 isolates with an identical ST<sup>T</sup>. Strains are listed vertically above their corresponding

50 lanes.

51

52 **References:**

- 53 **1. Nallapareddy SR, Sillanpaa J, Mitchell J, Singh KV, Chowdhury SA, Weinstock**  
54 **GM, Sullam PM, Murray BE.** 2011. Conservation of Ebp-type pilus genes  
55 among enterococci and demonstration of their role in adherence of *Enterococcus*  
56 *faecalis* to human platelets. *Infect. Immun.* **79**:2911-2920.
- 57 **2. Chowdhury SA, Arias CA, Nallapareddy SR, Reyes J, Willems RJ, Murray BE.**  
58 2009. A tri-locus sequence typing scheme for hospital epidemiology and  
59 subspecies differentiation of an important nosocomial pathogen, *Enterococcus*  
60 *faecalis*. *J. Clin. Microbiol.* **47**(9):2713-9.
- 61 **3. Malathum K, Singh KV, Weinstock GM, Murray BE.** 1998. Repetitive sequence-  
62 based PCR versus pulsed-field gel electrophoresis for typing of *Enterococcus*  
63 *faecalis* at the subspecies level. *J. Clin. Microbiol.* **36**:211-215.
- 64 **4. Nallapareddy SR, Wenxiang H, Weinstock GM, Murray BE.** 2005. Molecular  
65 characterization of a widespread, pathogenic, and antibiotic resistance-receptive  
66 *Enterococcus faecalis* lineage and dissemination of its putative pathogenicity  
67 island. *J. Bacteriol.* **187**:5709-18.
- 68 **5. Ruiz-Garbajosa P, Bonten MJ, Robinson DA, Top J, Nallapareddy SR, Torres C,**  
69 **Coque TM, Canton R, Baquero F, Murray BE, del Campo R, Willems RJ.**  
70 2006. Multilocus sequence typing scheme for *Enterococcus faecalis* reveals  
71 hospital-adapted genetic complexes in a background of high rates of  
72 recombination. *J. Clin. Microbiol.* **44**:2220-8.
- 73 **6. Mohamed JA, Huang W, Nallapareddy SR, Teng F, Murray BE.** 2004. Influence  
74 of origin of isolates, especially endocarditis isolates, and various genes on biofilm  
75 formation by *Enterococcus faecalis*. *Infect. Immun.* **72**:3658-3663.

- 76 **7. Tomayko JF, Murray BE.** 1995. Analysis of *Enterococcus faecalis* isolates from  
77 intercontinental sources by multilocus enzyme electrophoresis and pulsed-field  
78 gel electrophoresis. J. Clin. Microbiol. **42**:4879-4881.
- 79 **8. Hancock LE, Gilmore MS.** 2002. The capsular polysaccharide of *Enterococcus*  
80 *faecalis* and its relationship to other polysaccharides in the cell wall. Proc. Natl.  
81 Acad. Sci. U.S.A. **99(3)**:1574-1579.
- 82 **9. Hufnagel M, Hancock LE, Koch S, Theilacker C, Gilmore MS, Huebner J.** 2004.  
83 Serological and genetic diversity of capsular polysaccharides in *Enterococcus*  
84 *faecalis*. Microbiol. **42**:2548-2557.
- 85 **10. McBride SM, Fischetti VA, Leblanc DJ, Moellering RC Jr, Gilmore MS.** 2007.  
86 Genetic diversity among *Enterococcus faecalis*. PLoS One **2(7)**:e582.