

**Table S1.** NGS cluster and MLST of the 773 study strains.

	NGS cluster	no. of strains	MLST <sup>a</sup> (no. of strains)	<i>vanA</i> gene no. (%)	<i>vanB</i> gene no. (%)
<b>Complete Analysis</b>	all	773	ST80 (346), ST117 (200), ST17 (54), ST192 (38), ST203 (26), ST18 (18), ST202 (17), ST925 (7), ST132 (5), ST78 (4), ST64 (2), ST125 (2), ST280 (2), ST 323 (2), ST104 (1), ST262 (1), ST412 (1), ST612 (1), ST721 (1), ST780 (1), ST1201 (1), ST1299 (1), NT (42)	166 (21,5)	607 (78,5)
<b>Cluster Analysis</b>	1	21	ST203 (20), NT (1)	21 (100)	0 (0)
	2	29	ST192 (26), NT (3)	1(3.4)	28 (96.6)
	3	19	ST17 (19)	0 (0)	19 (100)
	4	18	ST17 (17), NT (1)	0 (0)	18 (100)
	5	275	ST80 (273), NT (2)	0 (0)	275 (100)
	6	106	ST117 (103), ST1201 (1), NT (2)	0 (0)	106 (100)
	7	54	ST117 (54)	19 (35.2)	35 (64.8)
	8	31	ST80 (31)	0 (0)	31 (100)
	9	15	ST202 (15)	15 (100)	0 (0)
	10	3	ST925 (3)	0 (0)	3 (100)
	11	3	ST925 (3)	0 (0)	3 (100)
	12	2	ST203 (2)	0 (0)	2 (100)
	13	3	ST192 (3)	3 (100)	0 (0)
	14	3	ST192 (3)	3 (100)	0 (0)
	15	2	ST17 (2)	0 (0)	2 (100)
	16	2	ST17 (2)	0 (0)	2 (100)
	17	2	NT (2)	0 (0)	2 (100)
	18	5	ST117 (5)	0 (0)	5 (100)
	19	2	ST80 (2)	0 (0)	2 (100)
	20	3	ST80 (2)	0 (0)	3 (100)
	21	7	ST117 (7)	0 (0)	7 (100)
	22	6	ST117 (6)	0 (0)	6 (100)
	23	2	ST132 (2)	0 (0)	2 (100)
	24	3	ST117 (3)	3 (100)	0 (0)
	25	3	ST117 (3)	3 (100)	0 (0)
	26	2	ST203 (2)	2 (100)	0 (0)
	27	4	NT (4)	0 (0)	4 (100)
	28	6	ST80 (6)	1 (17)	5 (83)
	29	6	ST80 (6)	5 (83)	1 (17)
	30	2	ST80 (2)	2 (100)	0 (0)
	31	3	ST18 (3)	3 (100)	0 (0)
	32	2	ST18 (2)	2 (100)	0 (0)
	33	2	ST117 (2)	0 (0)	2 (100)
	34	3	ST17 (3)	0 (0)	3 (100)
	35	7	NT (7)	0 (0)	7 (100)
	36	2	ST80 (2)	2 (100)	0 (0)
	37	3	ST132 (3)	3 (100)	0 (0)
	38	2	ST64 (2)	2 (100)	0 (0)
	Single	110	ST80 (27), NT (20), ST117 (17), ST 17 (11), ST18 (7), ST192 (6), ST78 (4), ST125 (2), ST202 (2), ST203 (2), ST280 (2), ST323 (2), ST104 (1), ST262 (1), ST412 (1), ST612 (1), ST721 (1), ST780 (1), ST925 (1), ST1299 (1)	56 (51)	54 (49)

a, MLST: Multi locus sequences type.

**Supplementary Table S2.** Characteristics of NGS clusters and NGS subclusters.

	NGS cluster	no. of strains	core genome (bp)	no. of sub-clusters	max. SNP within cluster <sup>a</sup>
<b>Complete Analysis</b>	all	773	1791934	-	-
<b>Cluster Analysis</b>	1	21	2821540	7	31
	2	29	2554476	18	55
	3	19	2994142	1	13
	4	18	2714833	7	120
	5	275	2523841	13	69
	6	106	2601519	12	62
	7	54	2424419	5	79
	8	31	2766300	7	20
	9	15	2663379	2	16
	Cluster (n<8), singletons	205	n.d.	n.d.	n.d.

a, In the first lane the maximal SNP difference within a cluster is shown for all strains. For the detailed cluster analysis, the maximal SNP difference with the corresponding NGS cluster is shown.

**Table S3. Overview of VREfm screening, colonization and infection in hematooncology patients.**

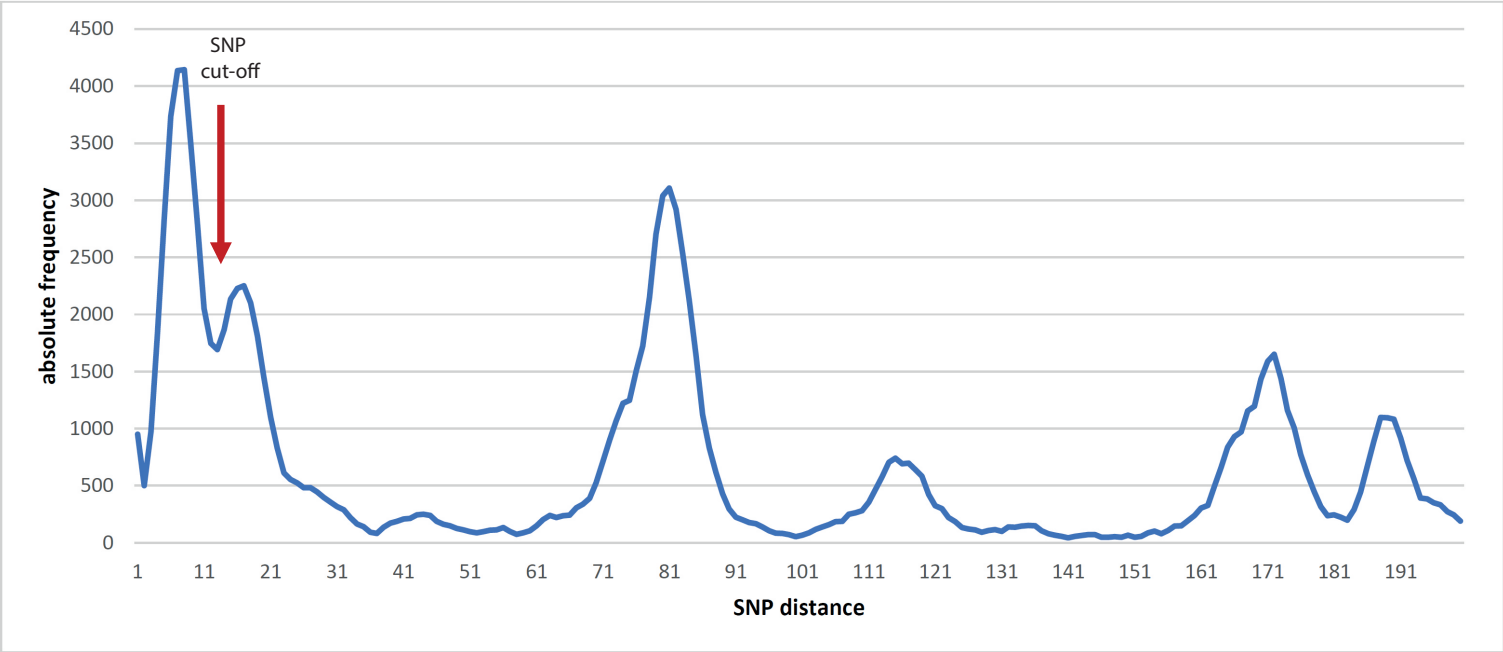
<b>Number of</b>	<b>Q1/15</b>	<b>Q2/15</b>	<b>Q3/15</b>	<b>Q4/15</b>	<b>Q1/16</b>	<b>Q2/16</b>	<b>Q3/16</b>	<b>Q4/16</b>
patients screened for VRE	327	360	341	344	373	410	453	452
patients positive for VRE (first isolate) (%) <sup>a</sup>	1 (0.3)	5 (1.4)	11 (3.2)	11 (3.2)	49 (13.1)	41 (10.0)	33 (7.3)	47 (10.4)
patients from whom clinical specimens were obtained <sup>b</sup>	168	165	166	167	153	160	185	178
patients with VREs isolated from clinical specimens <sup>b</sup> (%)	0 (0.0)	1 (0.6)	1 (0.6)	1 (0.6)	3 (2.0)	3 (1.9)	6 (3.2)	4 (2.2)
patients from whom blood culture were obtained <sup>c</sup>	90	86	90	94	99	101	100	97
patients with VREs isolated from blood culture <sup>c</sup> (%)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	1 (1.0)	0 (0.0)	1 (1.0)	1 (1.0)

a, percentage referred to patients screened for VRE. Q: quarter

b, clinical specimens included urine, wound, abscesses and blood cultures

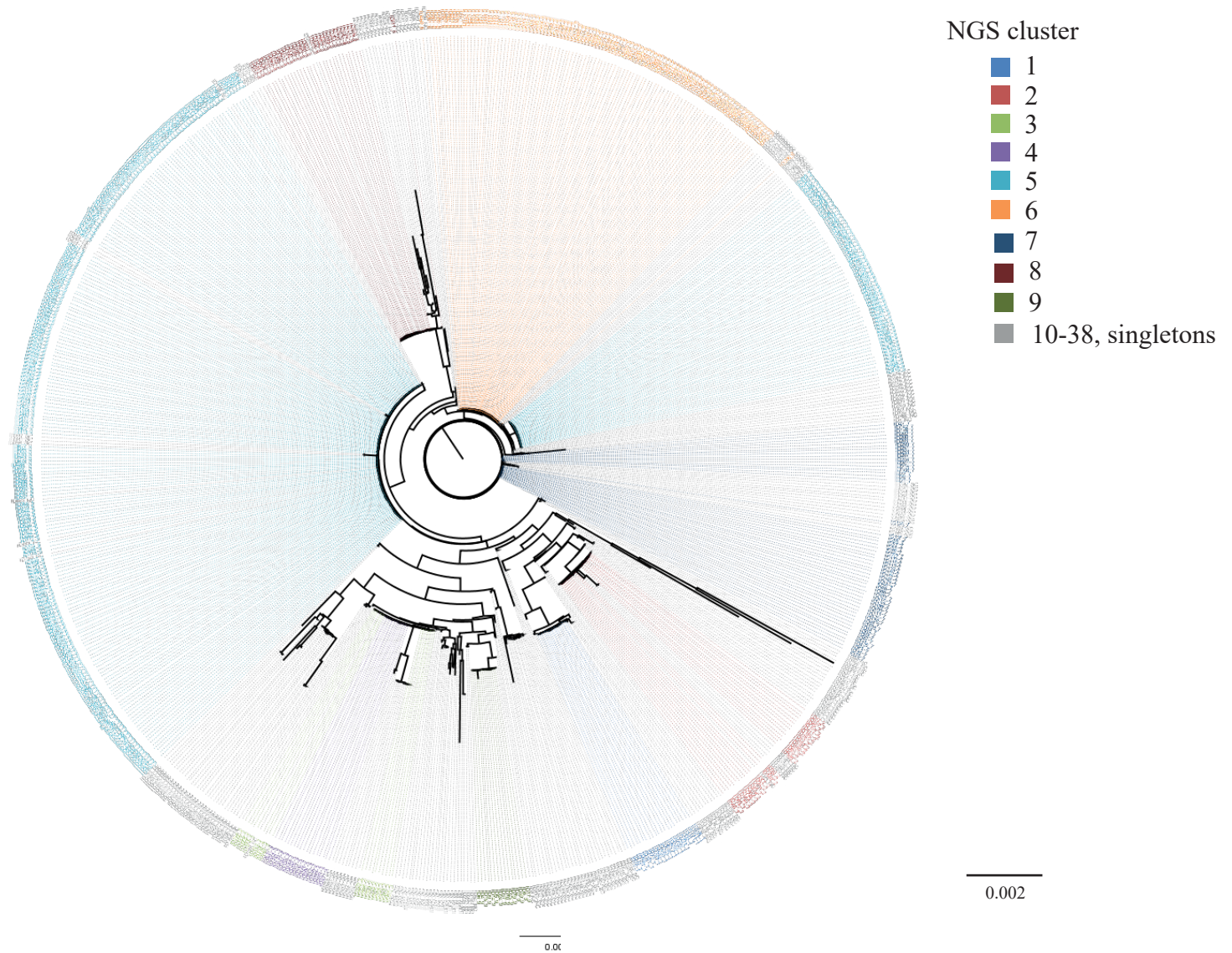
c, blood cultures isolates only

**Figure S1**



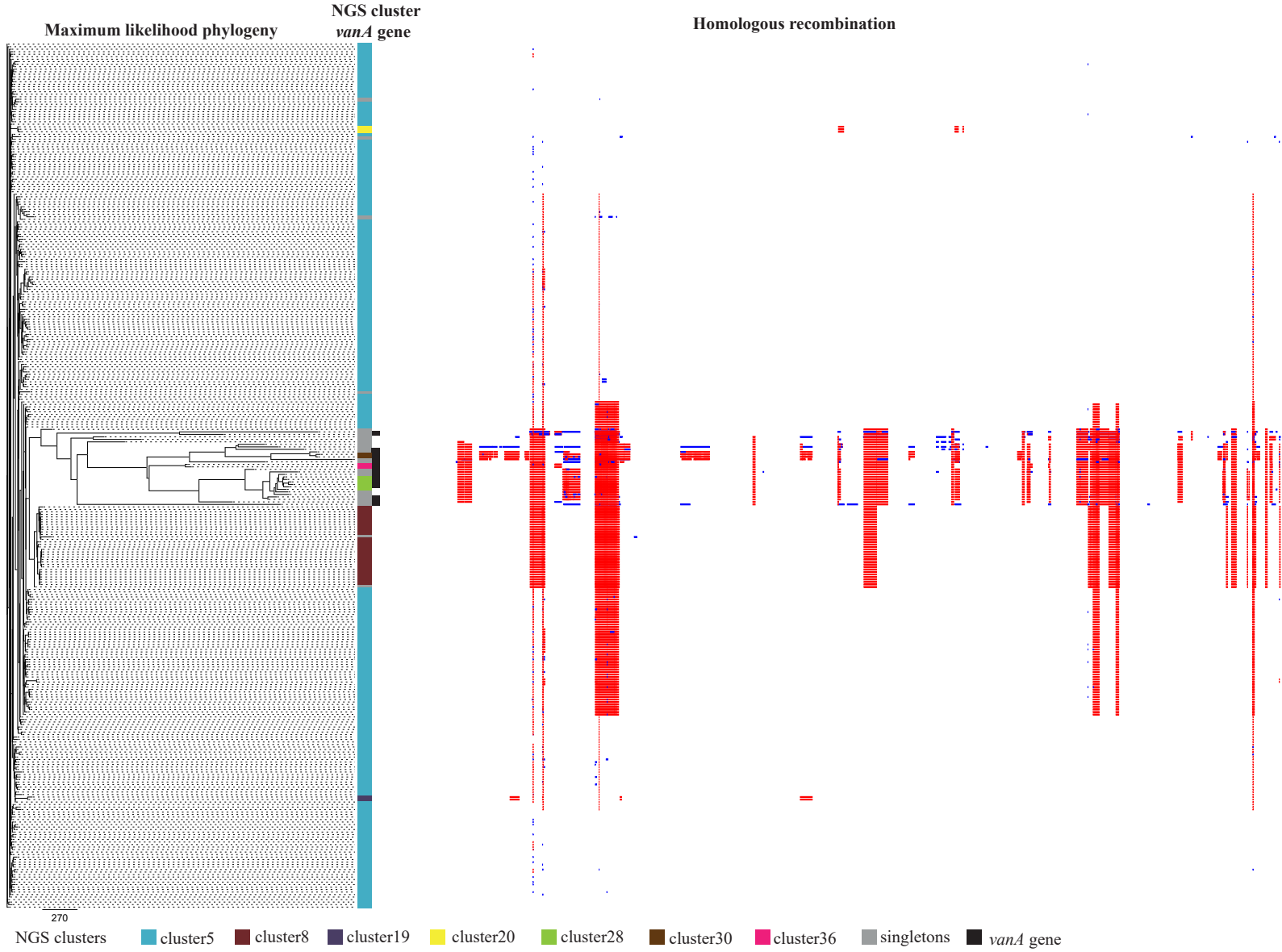
**Figure S1. SNP distance of VRE*fm* isolates.** The SNP distance from each VRE*fm* isolate to each other isolate was calculated and plotted in the diagram. Assuming the SNP distance distribution peaks reflect the distance within the NGS cluster (first peak) and the distance to the closest cluster (second peak), the SNP distance with the lowest frequency between those peaks was chosen as the SNP cut-off for NGS cluster assignment.

Figure S2



**Figure S2.** Maximum likelihood phylogeny based on core genome SNPs. The different NGS clusters defined by SNP differences of the UPGMA pairwise comparison (Figure 1) are colored. The scale bar represents the expected number of changes per site.

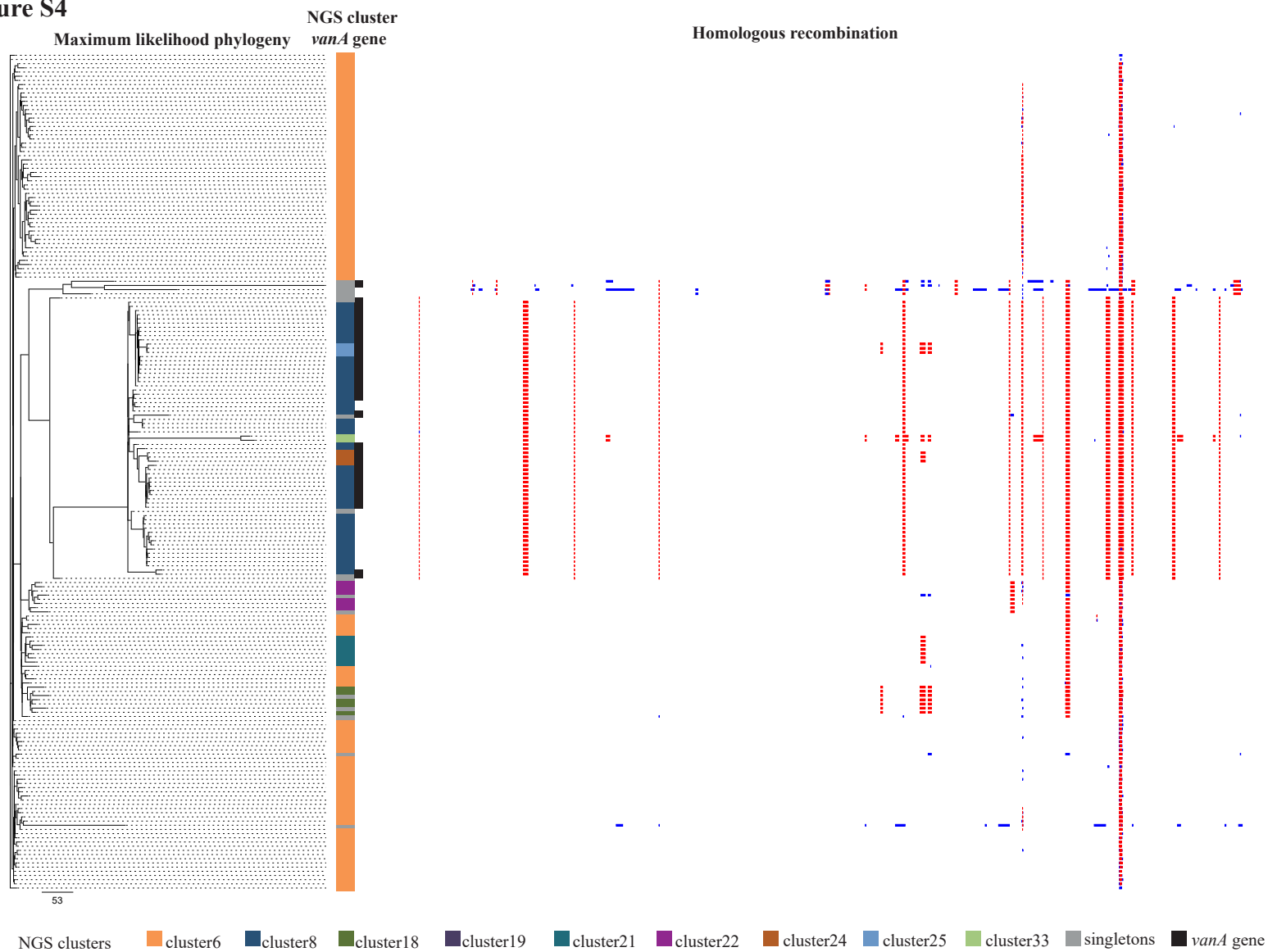
Figure S3





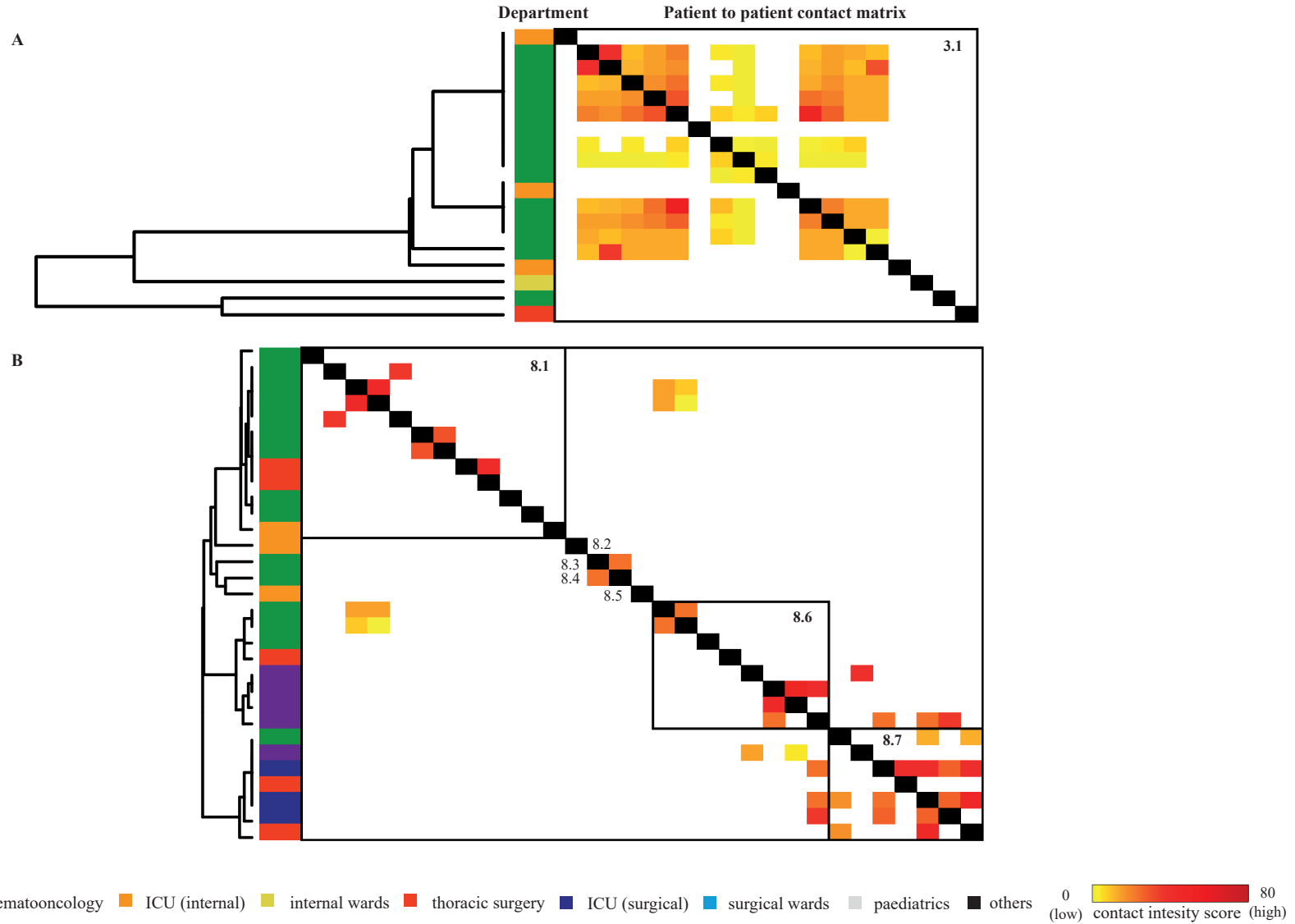
**Figure S3.** Maximum likelihood phylogeny and the recombination of the core genome of isolates belonging to ST80. In the group of ST80 strains (n=346), high diversity with various recombinations predicted in the core genome. Recombination events that are shared among several strains are colored in red, whereas recombination present in a single isolate is colored in blue. Both strains from NGS cluster 5 (n=273), cluster 8 (n=31), cluster 19 (n=2), cluster 20 (n=3), cluster 28 (n=6), cluster 30 (n=2) and cluster 36 (n=2), and 27 singletons, are present within the ST80 group. Strains harboring a *vanA* gene (n=23) are marked in black; the remaining isolates harboured a *vanB* gene (n=323). The scale bar shows a distance of 270 SNPs.

Figure S4



**Figure S4.** Maximum likelihood phylogeny and the recombination of the core genome of isolates belonging to ST117. In the group of ST117 strains (n=200), several clusters are formed, reflecting a high diversity within the ST117 strains. Recombination events that are shared among several strains are colored in red, whereas recombination present in a single isolate is colored in blue. Both strains from NGS cluster 6 (n=103), cluster 7 (n=54), cluster18 (n=5), cluster 21 (n=7), cluster 22 (n=6), cluster 24 (n=3), cluster 25 (n=3), cluster 33 (n=2), and 17 singletons, are present within the ST117 group. Strains harboring a *vanA* gene (n=35) are marked in black; the remaining isolates harbored a *vanB* (n=165) gene. The scale bar shows a distance of 53 SNPs.

Figure S5



**Figure S5. Phylogeny of NGS cluster 3 and cluster 8 in combination with the epidemiological links of the corresponding patients.** A phylogeny based on the specific cluster core genome, the VRE isolation site and the intensity score of patient-topatient contact is shown. Patients are shown on the x and y axis in the same order. For orientation, the dark gray middle line represents the symmetry axis of the matrix. Based on the SNP differences within the cluster (Tab. S2), the strains of cluster 8 were divided in seven subclusters. In cluster 3.1., a strong epidemiological and geographic link could be established, suggesting intra-hospital transmission. For cluster 8, less epidemiological links were observed; however, several genetically closely related strains along with epidemiological data also suggested transmission (e.g., department purple, subcluster 8.1).