Supplement Table of Contents

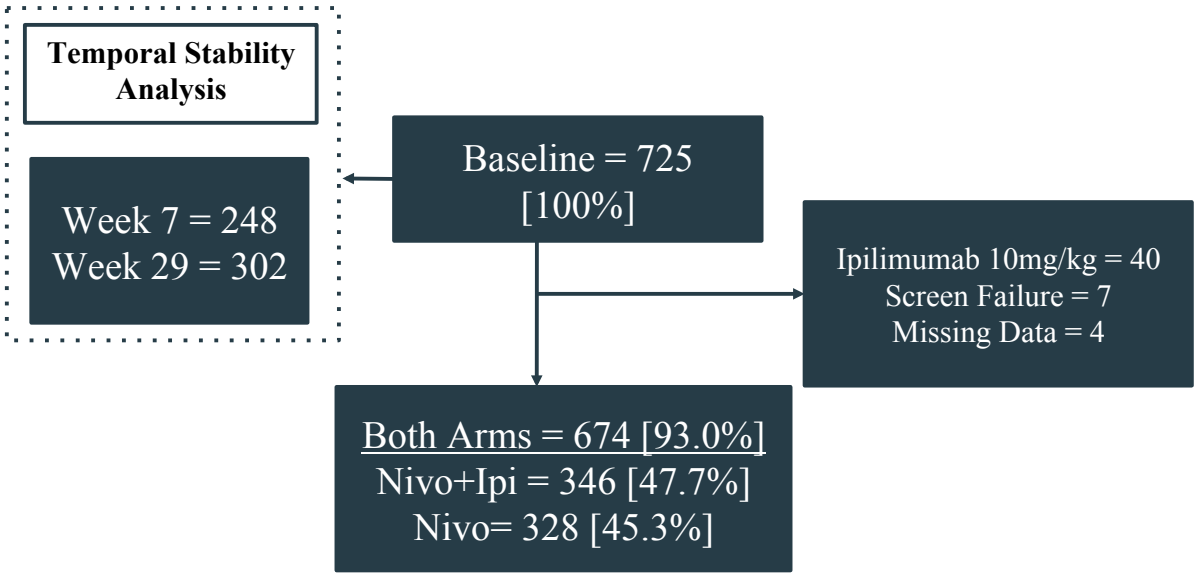
Supplemental Table 1. Region Stratified ANCOM-BC Results

Region	Recurrence Associated Strain	OR	95% CI, low	95% CI, high	p-val	q-val
North America	Firmicutes bacterium CAG:137	0.17	0.25	0.11	1.64E-05	0.028
North America	Firmicutes bacterium CAG:884	0.40	0.49	0.33	8.43E-06	0.015
North America	Clostridium sp. CAG:780	0.21	0.30	0.15	9.26E-06	0.016
North America	Eubacterium sp. CAG:115	0.054	0.10	0.030	9.76E-07	0.0017
North America	Eubacterium sp. CAG:786	0.09	0.13	0.058	1.67E-10	2.89E-07
North America	Peptostreptococcus anaerobius	0.49	0.58	0.42	1.13E-05	0.020
North America	Eubacterium siraeum	0.088	0.15	0.052	3.39E-06	0.006
North America	Ruminococcus sp. CAG:177	0.059	0.11	0.033	9.74E-07	0.0017
North America	Aeromonas salmonicida	0.42	0.51	0.34	2.37E-05	0.041
Western Europe	Bariatricus massiliensis	1.49	1.64	1.36	1.50E-05	0.028
Western Europe	Blautia schinkii	1.45	1.58	1.33	1.13E-05	0.021
Eastern Europe	Lawsonia intracellularis	2.50	3.09	2.02	1.62E-05	0.030
Rest of World	Clostridiales bacterium 1_7_47FAA	0.22	0.31	0.15	1.60E-05	0.032

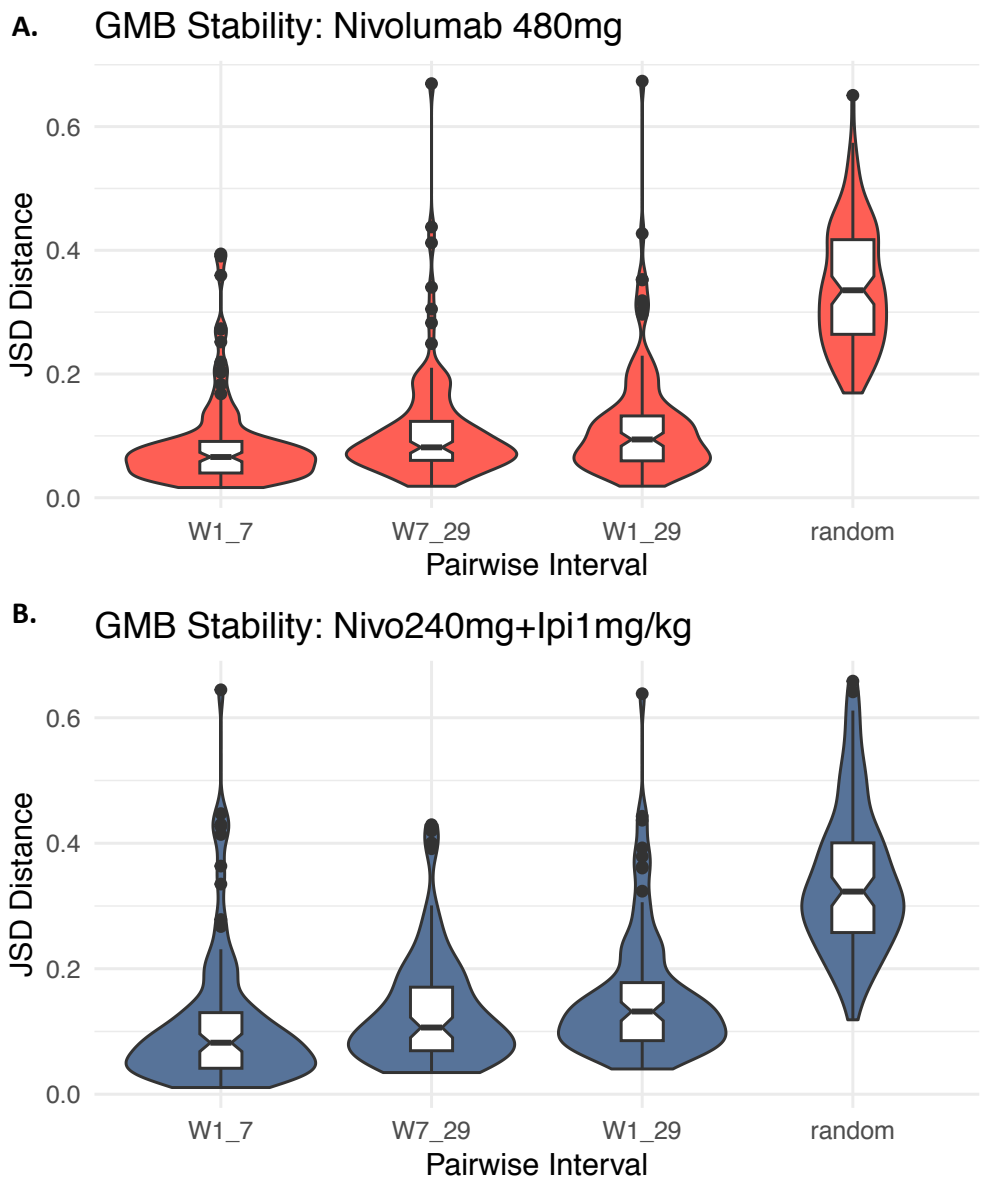
Table shows the results of ANCOM-BC analysis of recurrence as the outcome with GMB as the core predictor with adjustment for participant age, sex, tumor stage, BRAF mutation and study arm. Odds ratios (ORs) are shown with associated p-values as well as adjusted values using the “holm” method.

Supplemental Table 2: Patients with Longitudinal Sampling

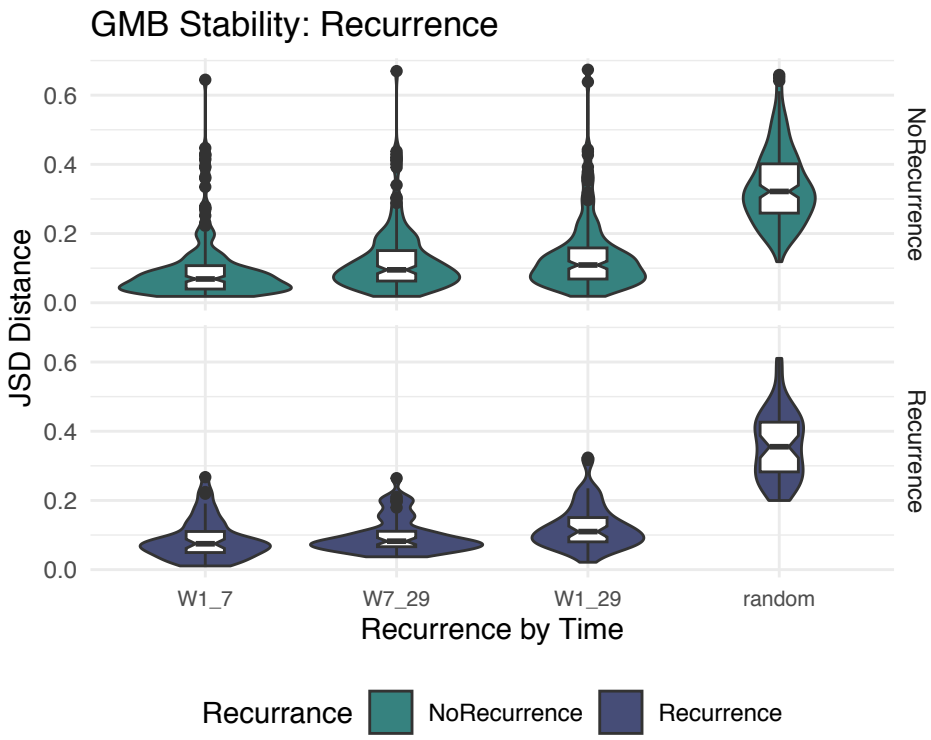
Variable	No	Yes	OR	pval
N	424	301		
Recurrence				
No	228 (55.2%)	219 (72.8%)	0.46 (0.33 - 0.64)	1.68E-06
Yes	185 (44.8%)	82 (27.2%)	-	-
Missing (n = 11)				
Age Mean± SD	54.54± 14.32	55.4± 13.3	-	0.685
Gender				
Female	178 (42.4%)	120 (39.9%)	1.11 (0.81 - 1.52)	0.54
Male	242 (57.6%)	181 (60.1%)	-	-
Missing (n = 4)				
Region				
Australia	77 (18.2%)	64 (21.3%)	-	0.13
Eastern Europe	24 (5.7%)	21 (7%)	-	0.125
ROW	29 (6.8%)	19 (6.3%)	-	0.284
North America	66 (15.6%)	39 (13%)	-	0.295
Western Europe	224 (52.8%)	158 (52.5%)	-	0.148
Stage at entry				
Not Reported	7 (1.7%)	0 (0%)	-	1
Stage IIIB	113 (26.7%)	102 (33.9%)	-	0.125
Stage IIIC	234 (55.2%)	152 (50.5%)	-	0.16
Stage IIID	10 (2.4%)	5 (1.7%)	-	0.53
Stage IV	56 (13.2%)	42 (14%)	-	0.141
B.Raf.Mut				
Invalid/Not Reported	104 (24.5%)	75 (24.9%)	-	0.145
Mutant	119 (28.1%)	79 (26.2%)	-	0.157
Wildtype	197 (46.5%)	147 (48.8%)	-	0.141
Melanoma Subtypes				
Acral	14 (3.3%)	9 (3%)	-	0.268
Cutaneous	357 (84.2%)	264 (87.7%)	-	0.142
Mucosal	4 (0.9%)	1 (0.3%)	-	1
Not Reported	8 (1.9%)	0 (0%)	-	1
Other	37 (8.7%)	27 (9%)	-	0.146
LD_baseline Mean± SD	218.92± 85.25	213.48± 88.16	-	0.207



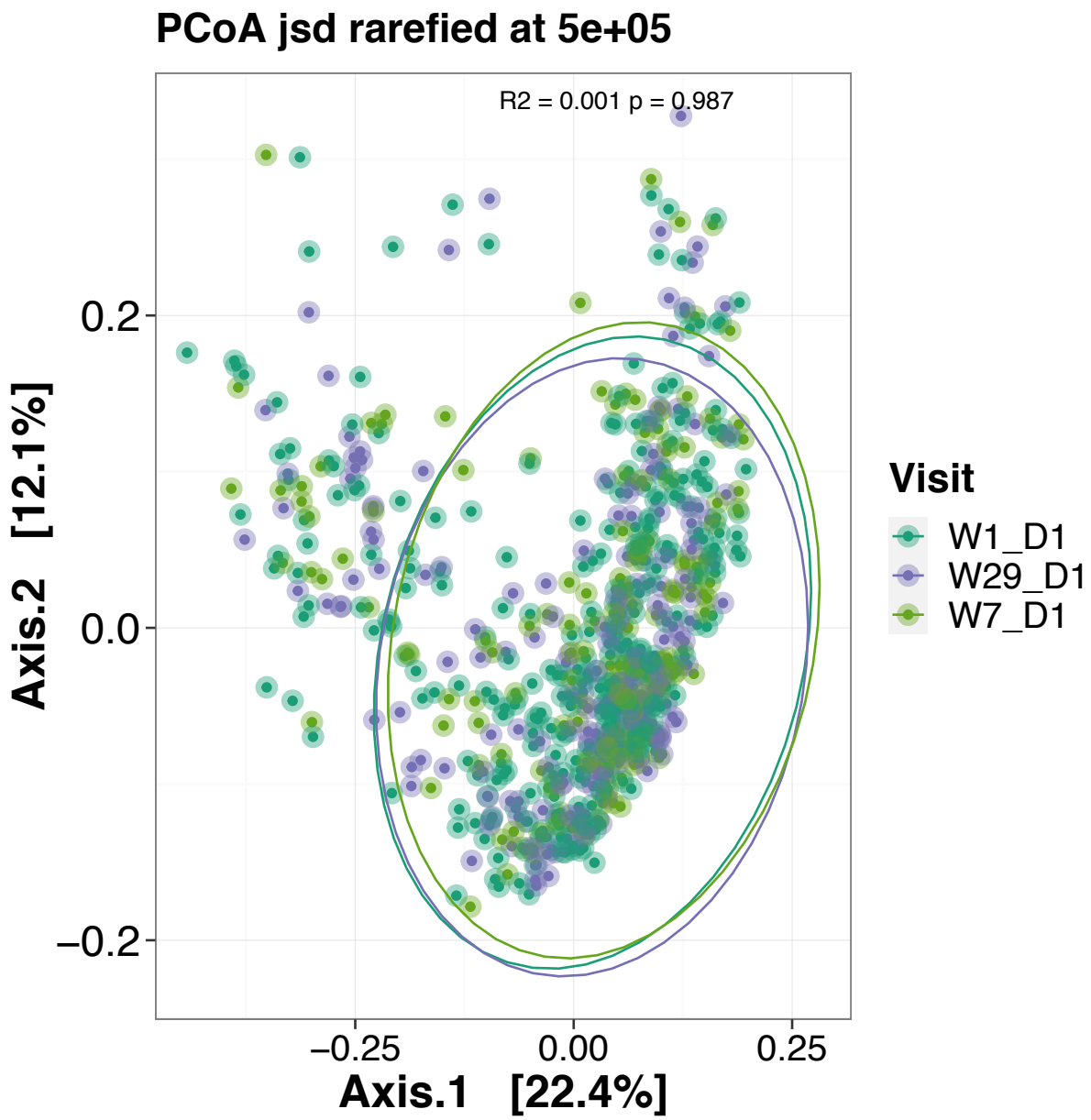
Supplemental Figure 1. Study Consort Chart
725 represented baseline. Of these , approximately half of the patients had follow-up sampling at weeks 7 and 29. From the baseline samples, 51 individuals were excluded due to coming from a supplementary arm of the original trial (n = 40), being screen failures (n = 7) or having missing randomization data (n = 4). Overall we utilized 674/725 (93.0%) of the available shotgun metagenomic samples for our core analysis.



Supplemental Figure 2. JSD Distances Across Time, Stratified by Trial Arm
The figure shows the bacterial β-diversity measured using Jensen Shannon divergence between measured visits (intra-patient variation) as well as between all unpaired samples for reference (inter-patient variation), stratified by the treatment arm (red, panel A is mono treatment and blue panel B is combination treatment). Overall GMB was largely unchanged across baseline, week 7 and week 29 measurements in both arms.



Supplemental Figure 3. JSD Distances Across Time, Stratified by Recurrence Status
The figure shows the bacterial β-diversity measured using Jensen Shannon divergence between measured visits (intra-patient variation) as well as between all unpaired samples for reference (inter-patient variation), stratified by recurrence status (green, panel A: no recurrence group and navy, panel B, recurrence group). Overall GMB was largely unchanged across baseline, week 7 and week 29 measurements in both panels.



Supplemental Figure 3. PCOA plot by Time Points
PCOA plat for three-time points as the outcome for the BMS patients using JSD distances.