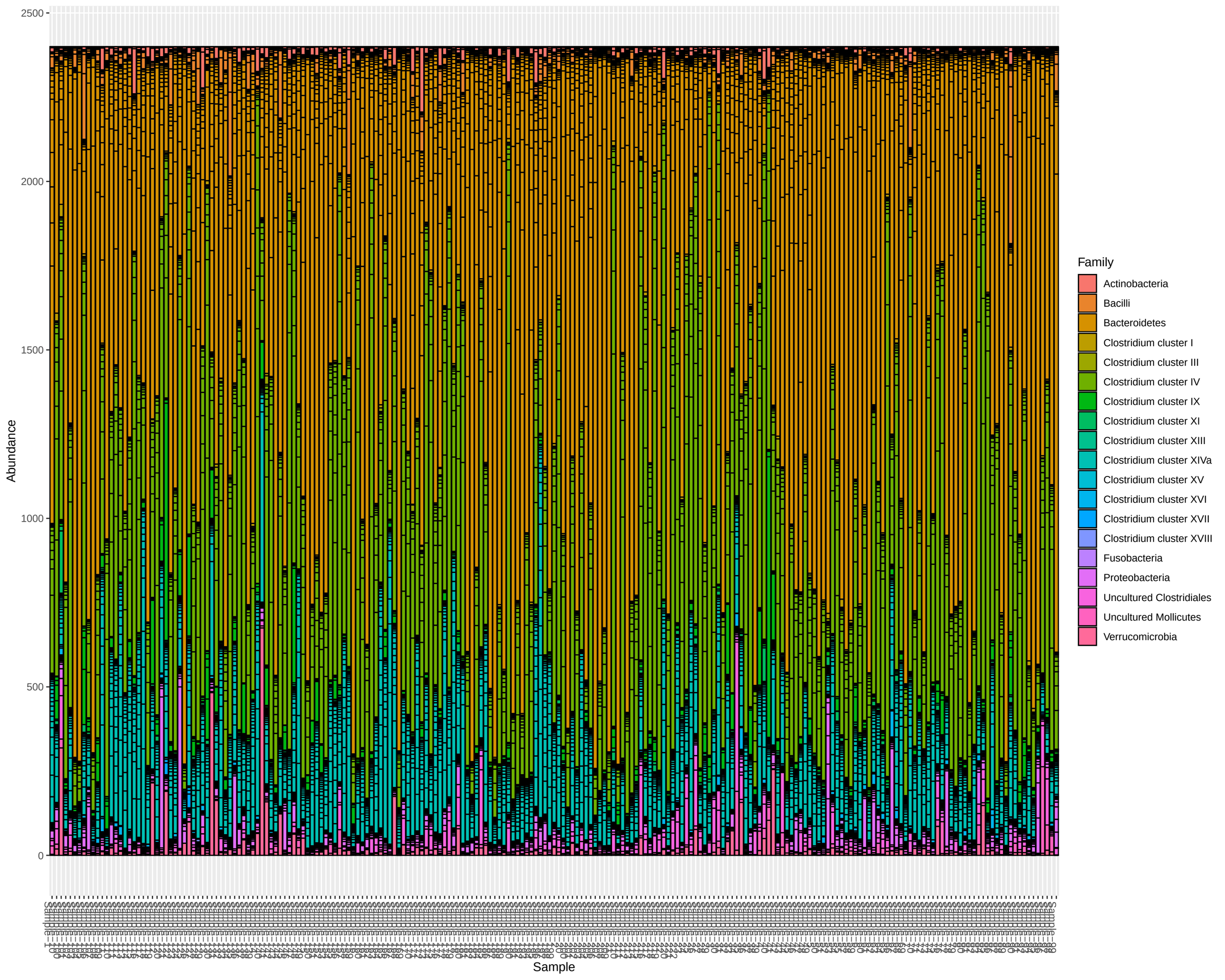
2.Alpha diversity

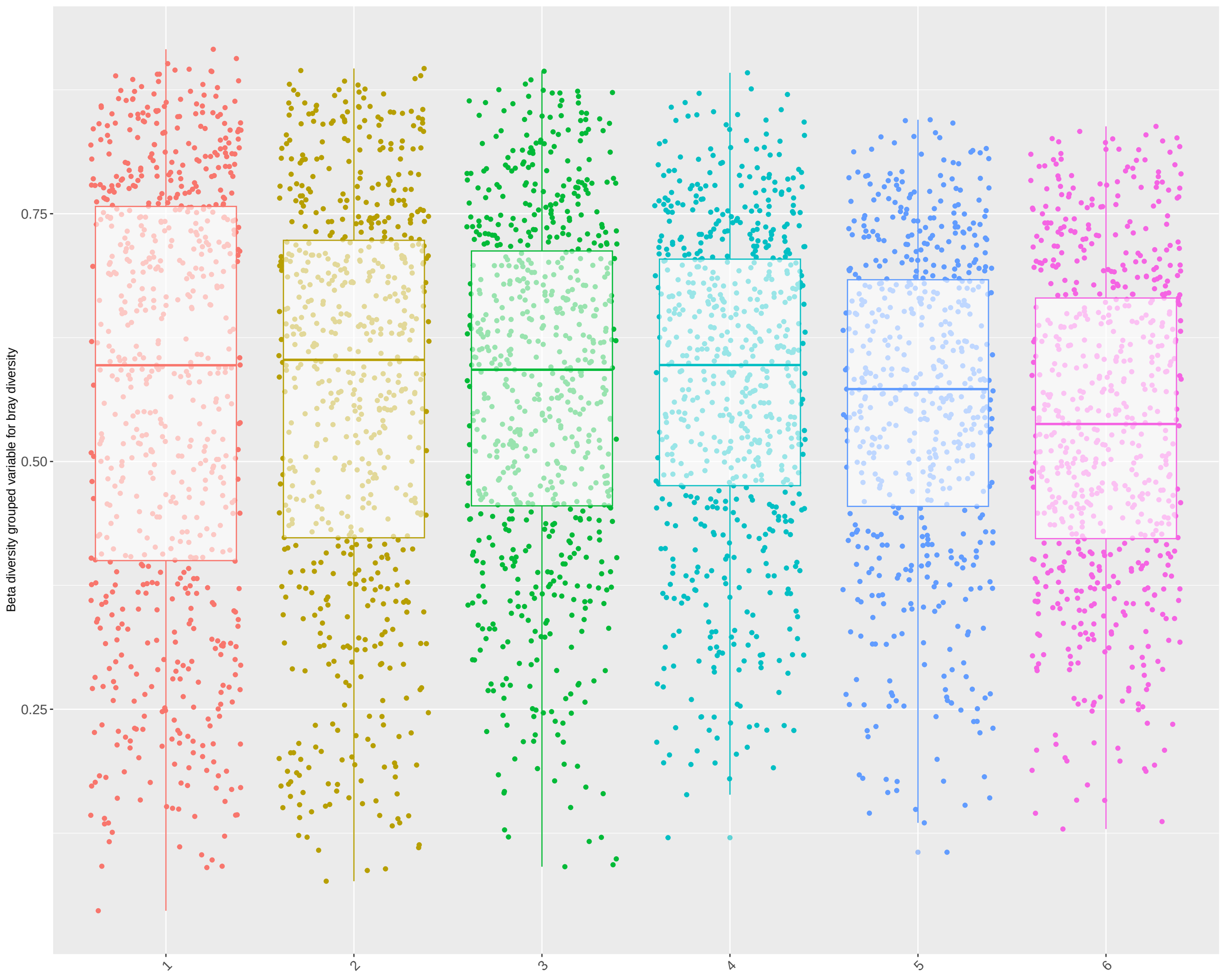

2500
2000
1500
1000
500
0
Abundance
Sample
Family
Actinobacteria
Bacilli
Bacteroidetes
Clostridium cluster I
Clostridium cluster III
Clostridium cluster IV
Clostridium cluster IX
Clostridium cluster XI
Clostridium cluster XIII
Clostridium cluster XIVa
Clostridium cluster XV
Clostridium cluster XVI
Clostridium cluster XVII
Clostridium cluster XVIII
Fusobacteria
Proteobacteria
Uncultured Clostridiales
Uncultured Mollicutes
Verrucomicrobia

Sample.90	81	94.1538461538462	7.99281608090598	96.5374107165347	4.79679421243531	2.41015578791334	0.773992013888889	4.42462240917617	16.1800572404844
Sample.91	77	111.363636363636	17.5151861865188	108.637667485843	5.37136940616098	1.92350914916903	0.691065972222222	3.23693704831803	15.1908553378196
Sample.92	81	123.166666666667	24.7366715159883	100.090877558099	4.82376494030302	2.38399119406156	0.808827777777778	5.23088547266862	16.1800572404844
Sample.93	80	91.4	6.95458630358584	94.3264349735138	4.50572424146085	1.77848857500852	0.569696875	2.32394315054068	15.9313309709258
Sample.94	72	142.2	40.1888758460865	97.9055489964581	4.78133217223914	1.4053853225723	0.485121875	1.94220719709154	13.9760027012311
Sample.95	71	90.25	10.8677492761215	92.2431808341752	5.01601009164139	1.63482299640724	0.562411805555556	2.28525360760609	13.7359652427869
Sample.96	70	97.1428571428571	16.5005915061527	89.1197741423986	4.67204358748537	2.39902876662598	0.807578819444444	5.19693308768201	13.4969171154013
Sample.97	73	101.5	17.9317946977915	89.1224473314607	4.68401264466688	2.49178232743777	0.825075	5.71673574389024	14.217024368527
Sample.98	72	93.8571428571429	13.8797777941804	85.6596358391923	4.46770405310295	2.24410182963486	0.764839236111111	4.25241006817145	13.9760027012311
Sample.99	77	87.4615384615385	6.75236737312226	88.6143911105548	4.37235075537642	1.9889502735934	0.637246875	2.75669575555	15.1908553378196
Sample.100	90	107.1	10.4104915657486	104.205642479214	5.03032314397663	3.24018405628126	0.930360069444444	14.3595777885253	18.460603486945
Sample.101	74	101.083333333333	14.1566586815387	101.995196284364	5.11310897255449	1.84617304171175	0.660489236111111	2.94541471541464	14.4590252584172
Sample.102	82	101	11.2896737186363	98.4343230352769	4.8302297928907	2.46694478255421	0.810669444444444	5.28176763156736	16.4297271894329
Sample.103	68	75.5555555555556	4.98247153772291	80.6757731132831	4.03957891921893	1.32948227245661	0.458407291666667	1.84640595158185	13.0218100540161
Sample.104	72	103.909090909091	16.5104041068042	100.340954888082	5.01637714027634	1.28685078203355	0.439954513888889	1.78556925249747	13.9760027012311
Sample.105	78	89.1428571428571	8.22978385953756	85.8124017957351	4.49287150790469	3.07355439924135	0.919676388888889	12.4496394791901	15.4367258058891
Sample.106	73	90.1	10.4098928658444	87.4619230769231	4.42194085877114	1.97246386190172	0.646677777777778	2.8302776816881	14.217024368527
Sample.107	71	82.0526315789474	6.44784762386685	88.3459611389466	4.49084775738624	1.2389687539107	0.425423611111111	1.740412622826	13.7359652427869
Sample.108	72	81.5454545454545	6.56584199387946	83.3172095761382	4.46717610687544	2.38604572040022	0.757011458333333	4.11542039448019	13.9760027012311
Sample.109	89	123.5	19.2744333180455	113.246971292679	5.15318216537166	3.39838447843337	0.946685069444444	18.7564719597257	18.2035189631603
Sample.110	81	107.25	15.469776401633	104.349375	5.22169607537528	2.84674120191628	0.860159375	7.15099778766006	16.1800572404844
Sample.111	79	106.142857142857	16.5013361628345	101.588300702911	5.07364746869764	3.145034859698	0.919183333333333	12.3736852959373	15.6835524391416
Sample.112	77	83	4.69857113873921	88.7043261749144	4.77671996612468	3.0364962037445	0.875061111111111	8.00391302414514	15.1908553378196
Sample.113	68	88	13.4939193189325	85.4331551628849	4.74274237728147	2.53813751785101	0.795871527777778	4.89887564000068	13.0218100540161
Sample.114	73	84.1428571428571	8.22952658528904	83.9274394160584	4.61331439364544	2.91513815151746	0.870756944444444	7.73735962602762	14.217024368527
Sample.115	76	99.3333333333333	13.6351405964948	96.5829643019515	4.83981285685687	2.81614348552458	0.874343055555556	7.95817536723663	14.9459454111693
Sample.116	78	108.6	20.0084984154179	94.2349796407186	4.86984799268934	3.13192082983615	0.910368402777778	11.1567798744087	15.4367258058891
Sample.117	76	88	7.95955686295069	91.5913128924856	4.88629103442906	3.04987823376226	0.908839583333333	10.9696734236808	14.9459454111693
Sample.118	67	91.4285714285714	15.1641965083449	83.8371313518119	4.49133136855826	1.98031855274714	0.653907291666667	2.8893992156485	12.7857620926352
Sample.119	76	82	4.69855253114465	84.3278612890689	4.5437088267295	3.06497037295816	0.9167375	12.0102086773758	14.9459454111693
Sample.120	82	92.4615384615385	6.75246477403439	94.9379736665425	4.74550925356807	2.72235049447772	0.861513194444444	7.22090451858129	16.4297271894329
Sample.121	81	90.5454545454545	6.56606487850268	91.7443621876732	4.70484967631208	2.84598455150714	0.873002083333333	7.87414491707541	16.1800572404844
Sample.122	82	86.7894736842105	3.60509087799287	92.1389126566978	4.63102434732679	2.7364983402415	0.84858125	6.60420192347381	16.4297271894329
Sample.123	75	85.5	7.20569949853294	85.6106629240974	4.51835086498336	2.53640776289402	0.820305208333333	5.56499156555966	14.702000515631
Sample.124	82	90	5.24857041007177	96.8936541193182	4.9255910983418	2.63935917102687	0.851982291666667	6.75594840144408	16.4297271894329
Sample.125	64	83.125	12.0463579801871	78.5064456696152	4.17652754781938	1.54135428732682	0.550248611111111	2.22345061005926	12.08370965492
Sample.126	85	155.2	40.1909294467522	118.658151134255	5.67485263085648	3.02901115878708	0.89914375	9.91510193964182	17.1843599948278
Sample.127	78	87.4285714285714	7.2557718981011	86.0337719298246	4.57808387538408	3.13654374415627	0.916215972222222	11.9354491127154	15.4367258058891
Sample.128	79	86	5.0560298117196	88.0175709914496	4.46537207129891	2.90478399824335	0.897810416666667	9.78573321644819	15.6835524391416
Sample.129	75	110	25.6088733438118	86.2817944187269	4.56850117787285	3.11270902144616	0.907735069444444	10.8383542260173	14.702000515631
Sample.130	78	101.333333333333	13.6352435629965	98.3128071411944	4.99525329304259	2.93255240382074	0.906525694444444	10.6981270848347	15.4367258058891
Sample.131	75	86.375	8.08289606816159	85.8272390886312	4.60603281046071	2.80322964291892	0.852432986111111	6.77658220253039	14.702000515631
Sample.132	79	96.1	10.4101283627348	100.056899581024	5.14811576423854	2.93649811199503	0.88216875	8.48671298997507	15.6835524391416
Sample.133	76	85.5454545454545	6.565946844549	85.9770425902478	4.40590419321262	2.27157472503843	0.716025347222222	3.5214410519327	14.9459454111693

Test whether the observed number of OTUs differs significantly between the variable using Wilcoxon rank−sum test

	1	2	3	4	5
2	1	NA	NA	NA	NA
3	1	1	NA	NA	NA
4	0.654695752094996	0.167010245153008	0.349530052028851	NA	NA
5	1	1	1	1	NA
6	1	0.22698515648416	0.523084175396183	1	1

3.Beta diversity


Beta diversity grouped variable for bray diversity
0.75
0.50
0.25
1
2
3
4
5
6

Beta diversity heatmap for bray diversity

samples

samples

value

0.75

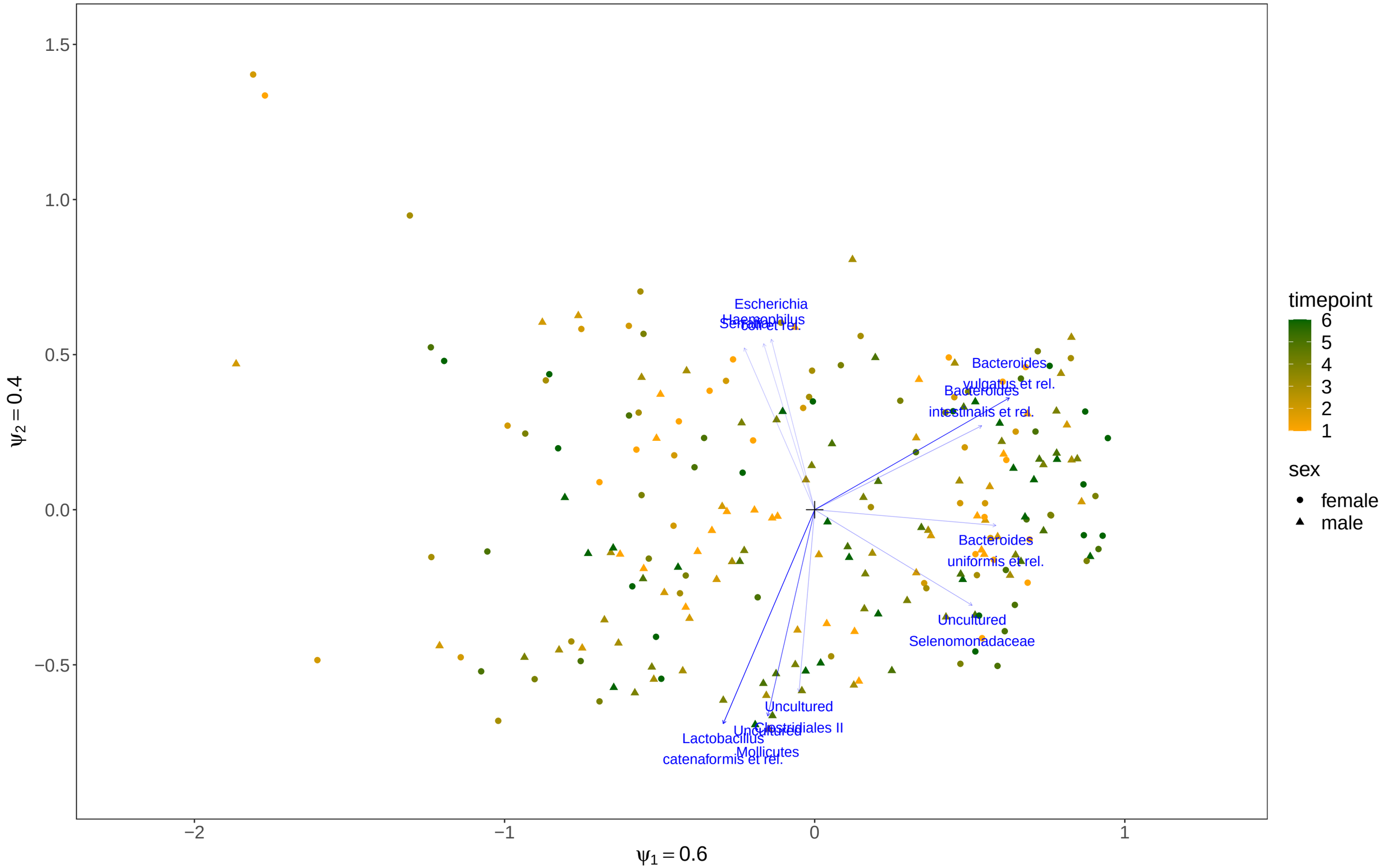
0.50

0.25

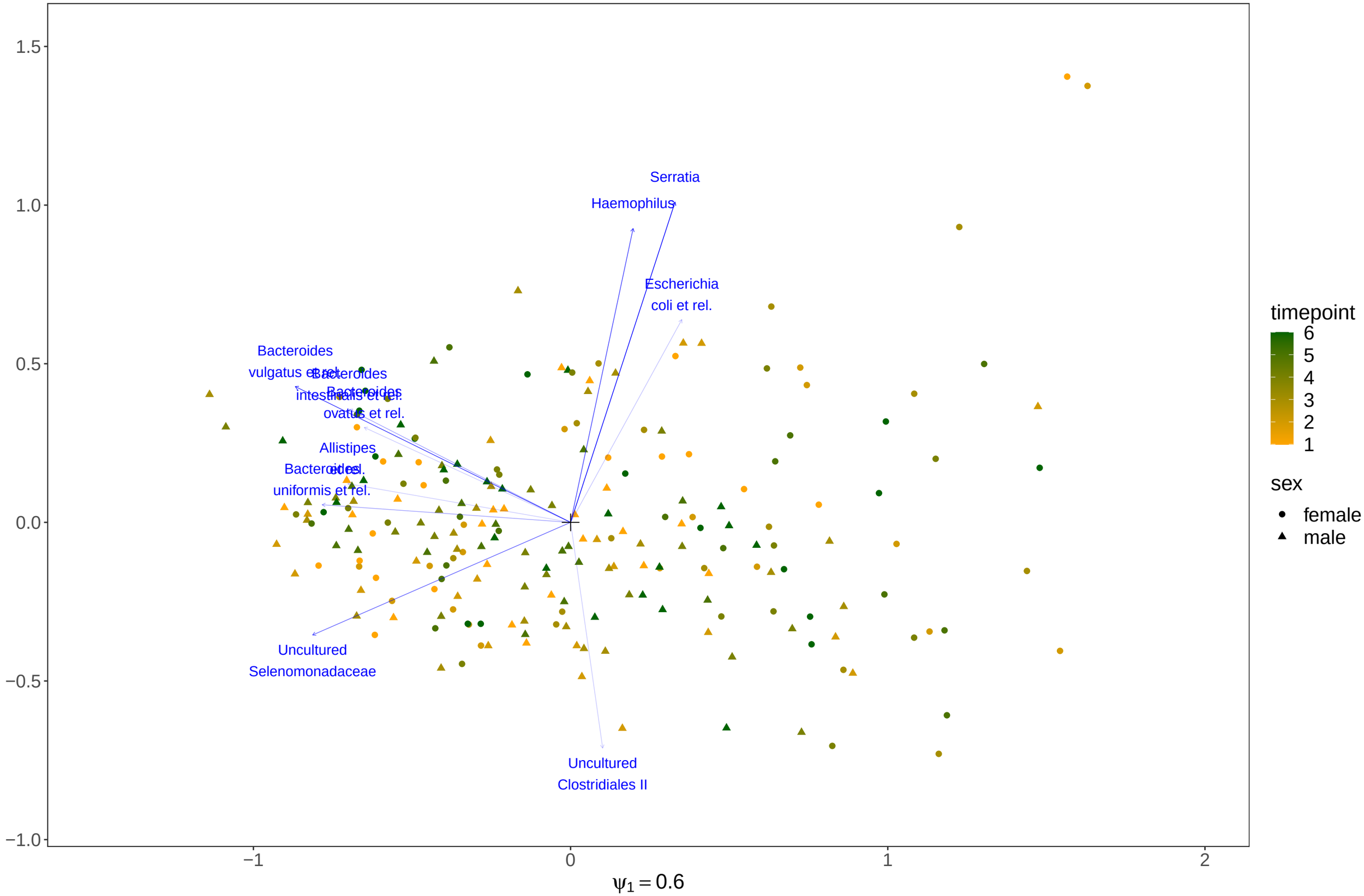
0.00

4.Ordination techniques

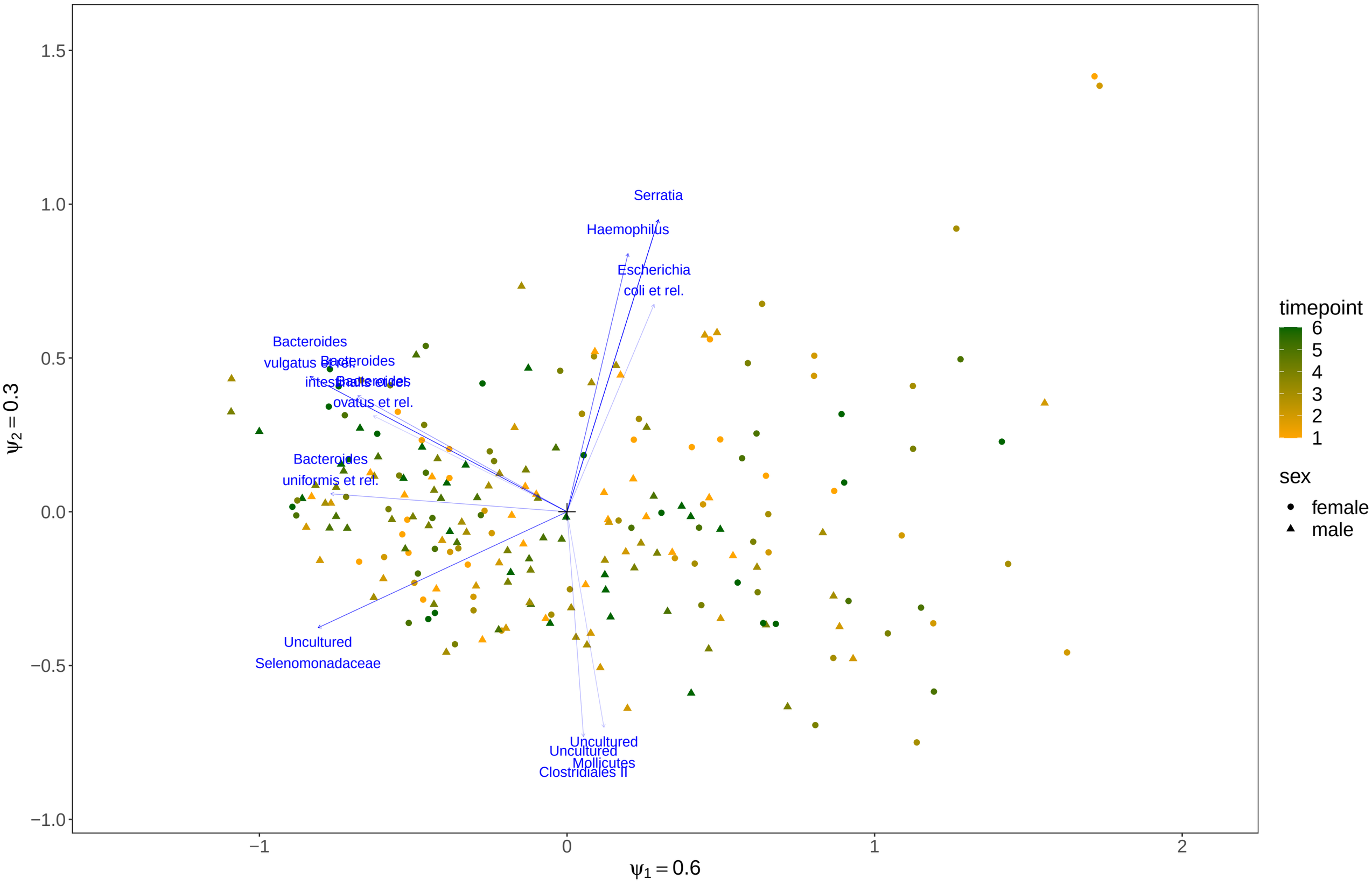
Fit Unconstrained RCM



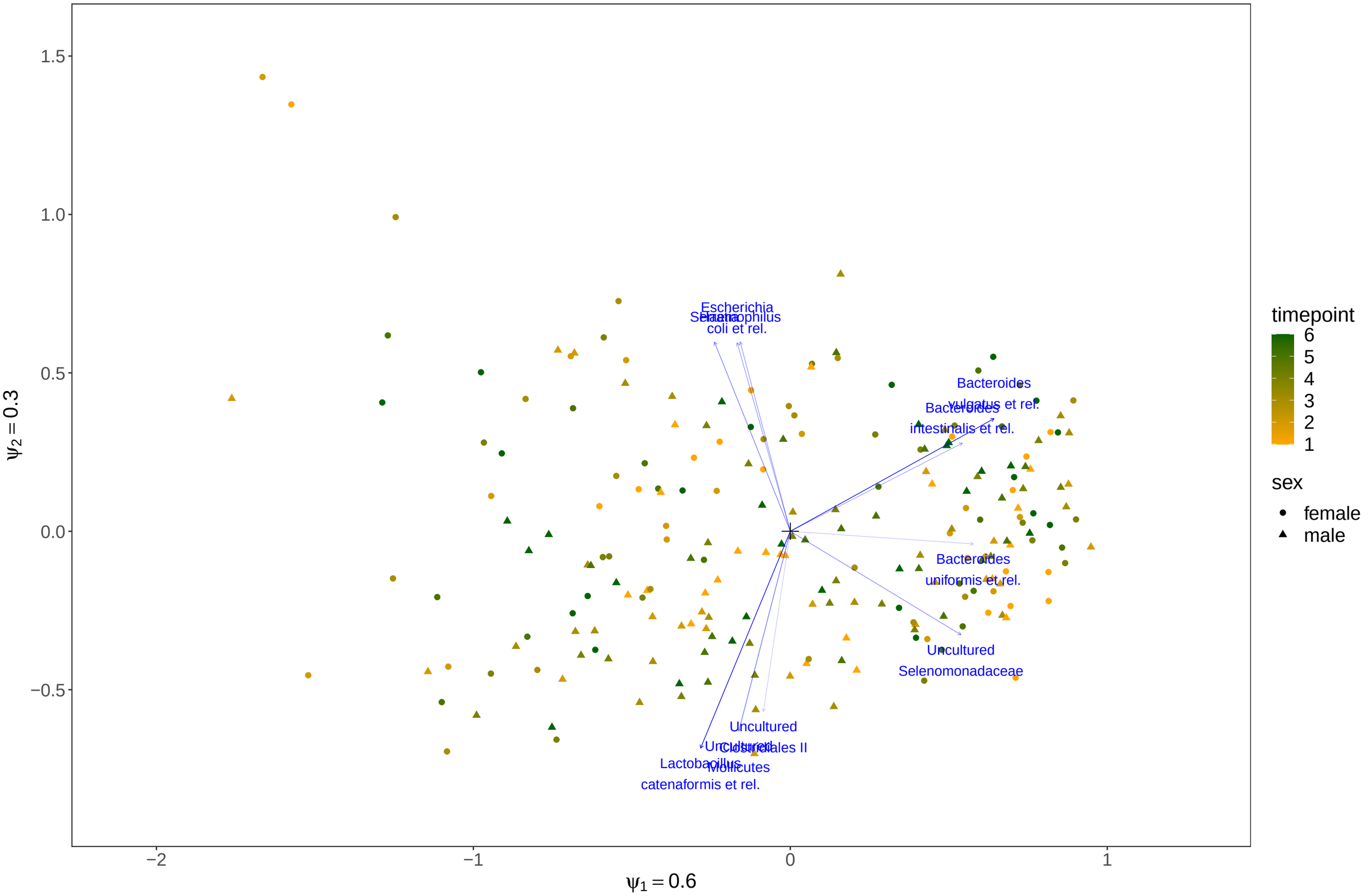
Total confounders RCM



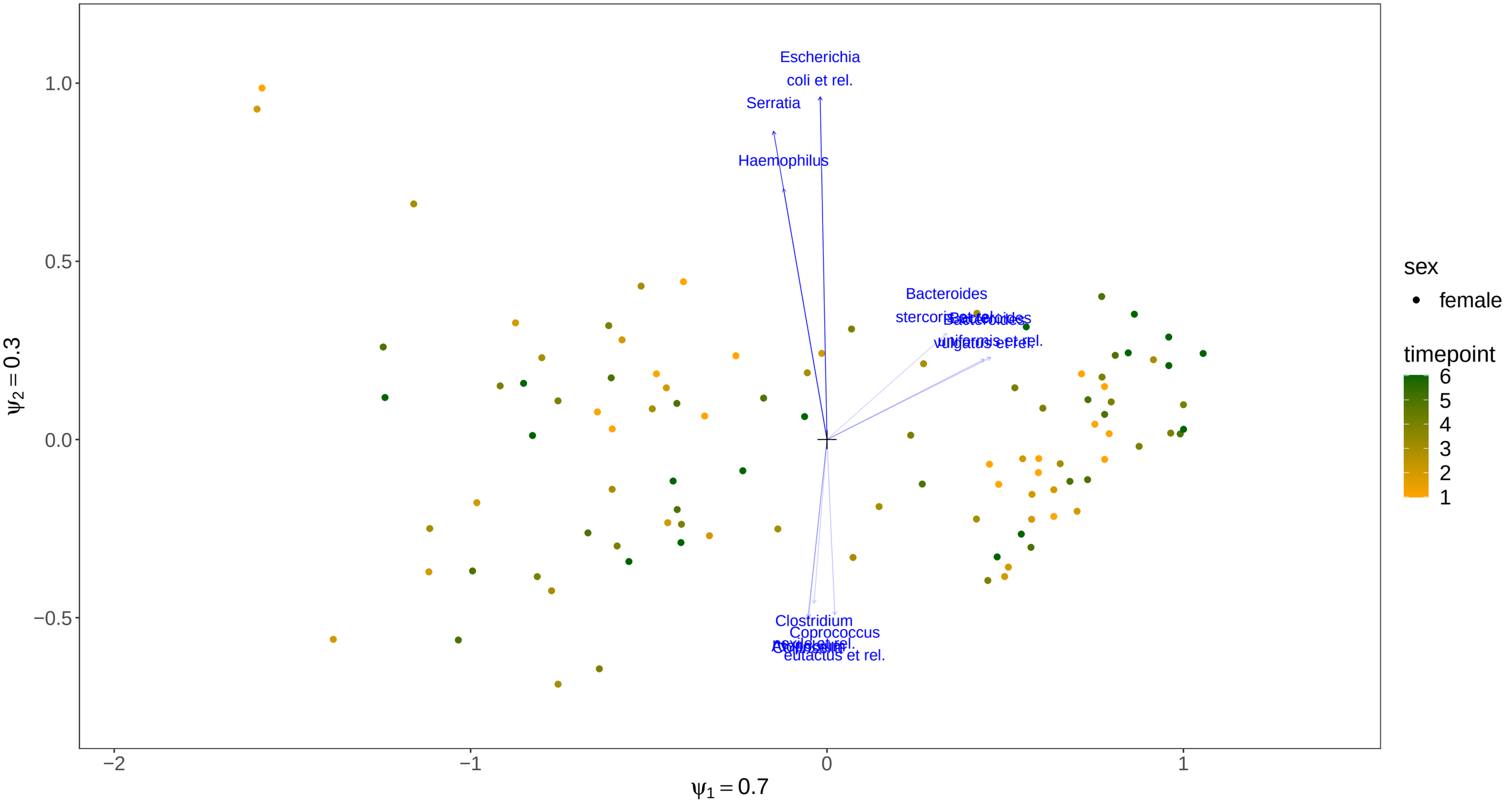
RCM for confounder bmi_group



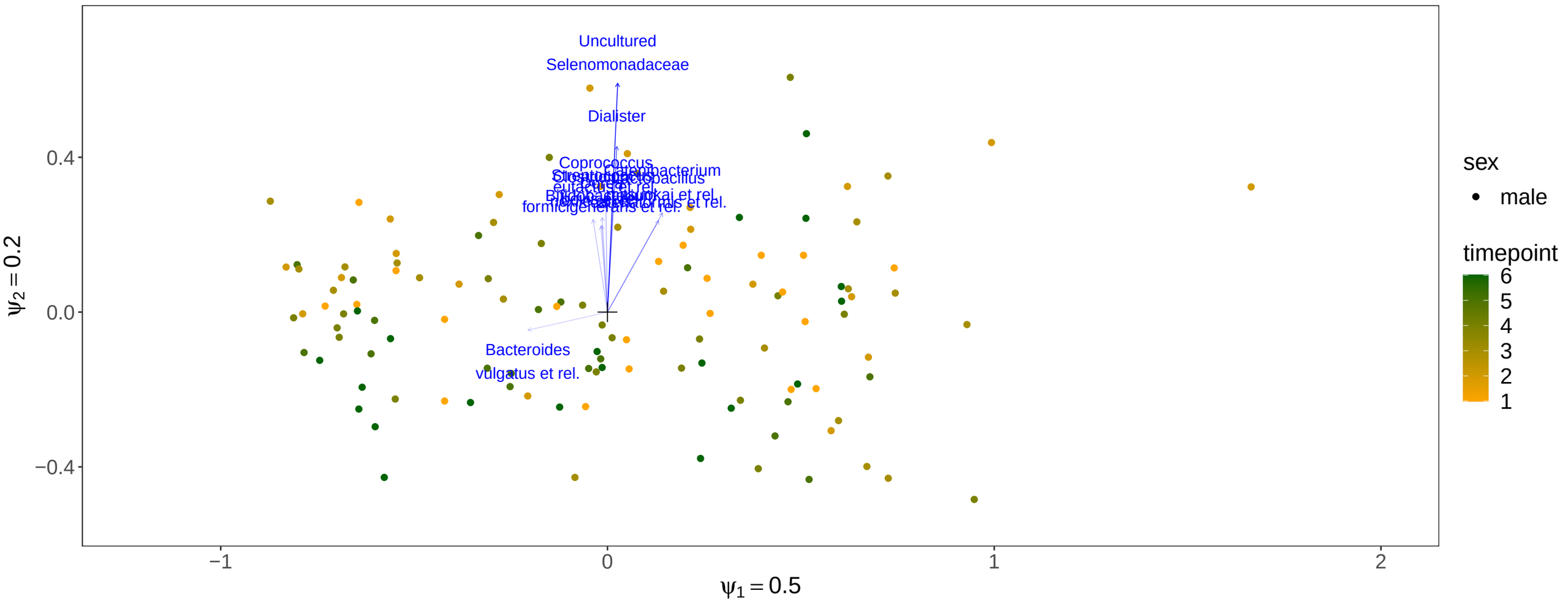
RCM for confounder timepoint



RCM split sex to female



RCM split sex to male



taxa

0.25

0.00

−0.25

−0.50

NMDS2

−0.25

0.00

0.25

NMDS1

Phylum

- Actinobacteria
- Bacteroidetes
- Firmicutes
- Fusobacteria
- Proteobacteria
- Verrucomicrobia

taxa

Actinobacteria

Bacteroidetes

Firmicutes

Fusobacteria

Proteobacteria

Verrucomicrobia

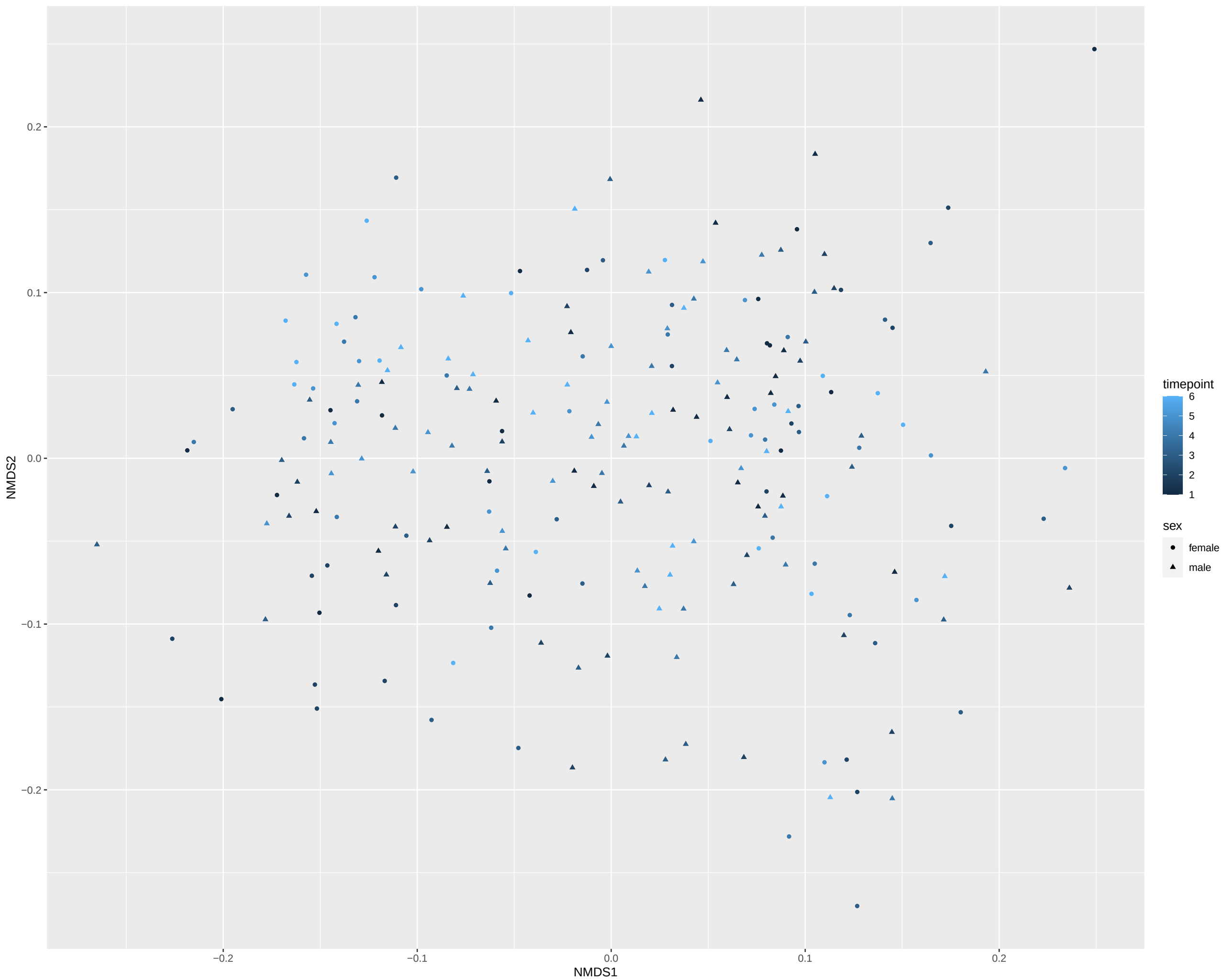
NMDS2

NMDS1

0.25
0.00
−0.25
−0.50

Phylum

- Actinobacteria
- Bacteroidetes
- Firmicutes
- Fusobacteria
- Proteobacteria
- Verrucomicrobia


NMDS2
0.2
0.1
0.0
−0.1
−0.2
−0.2
−0.1
0.0
0.1
0.2
NMDS1
timepoint
6
5
4
3
2
1
sex
female
male

DCA

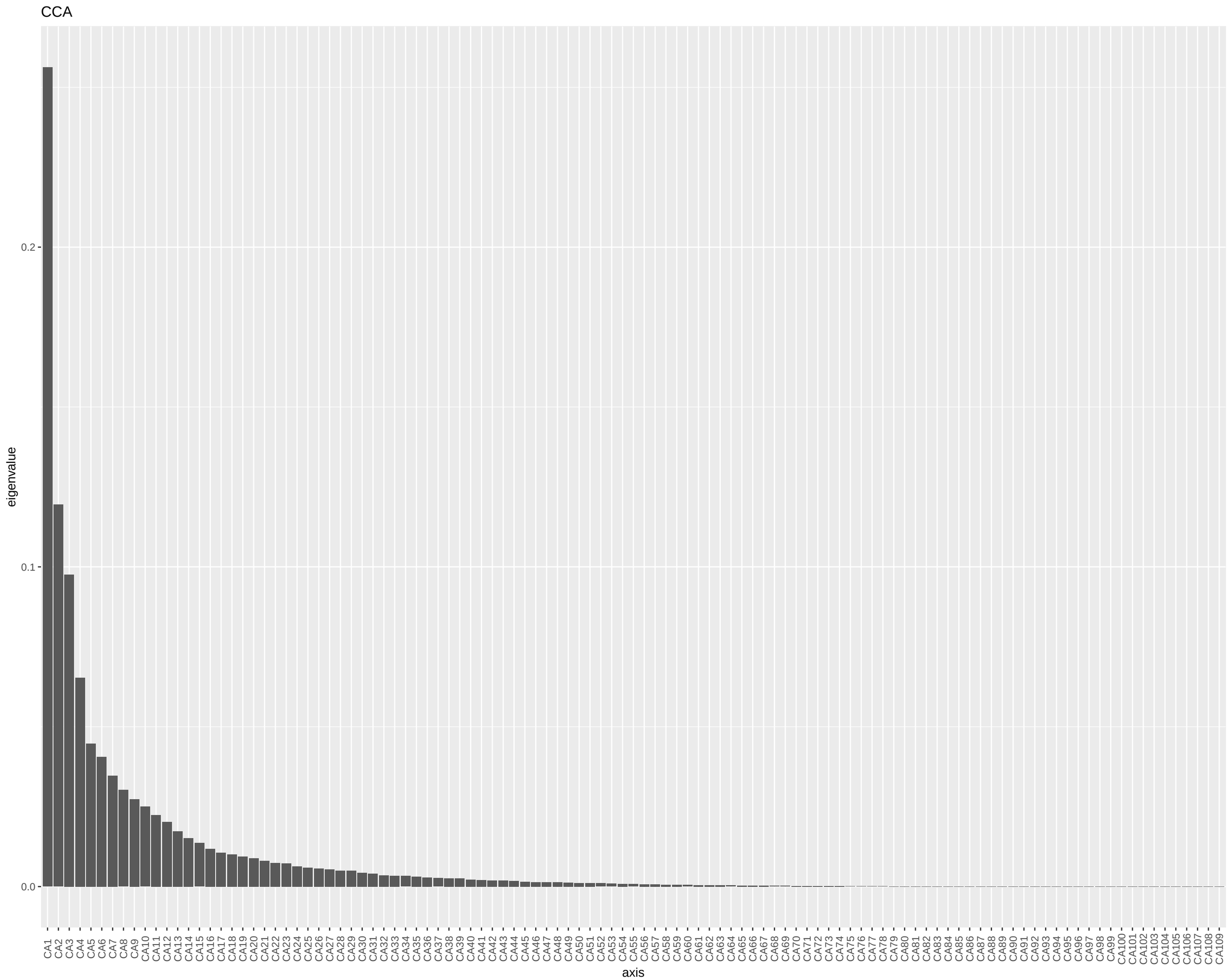
eigenvalue

0.4
0.3
0.2
0.1
0.0

DCA1
DCA2
DCA3
DCA4

axis

CCA


eigenvalue
0.2
0.1
0.0
CA1
CA2
CA3
CA4
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CA7
CA8
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CA11
CA12
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CA105
CA106
CA107
CA108
CA109
axis

RDA

eigenvalue

0.8
0.6
0.4
0.2
0.0

PC1 PC2 PC3 PC4 PC5 PC6 PC7 PC8 PC9 PC10 PC11 PC12 PC13 PC14 PC15 PC16 PC17 PC18 PC19 PC20 PC21 PC22 PC23 PC24 PC25 PC26 PC27 PC28 PC29 PC30 PC31 PC32 PC33 PC34 PC35 PC36 PC37 PC38 PC39 PC40 PC41 PC42 PC43 PC44 PC45 PC46 PC47 PC48 PC49 PC50 PC51 PC52 PC53 PC54 PC55 PC56 PC57 PC58 PC59 PC60 PC61 PC62 PC63 PC64 PC65 PC66 PC67 PC68 PC69 PC70 PC71 PC72 PC73 PC74 PC75 PC76 PC77 PC78 PC79 PC80 PC81 PC82 PC83 PC84 PC85 PC86 PC87 PC88 PC89 PC90 PC91 PC92 PC93 PC94 PC95 PC96 PC97

axis

Fit Unconstrained RCM