

UNDERGRADUATE RESEARCH PROGRAMME (UGRP)

Metabolic redundancies, minimal reactomes and interactions  
in gut microbial species.

Under the guidance and supervision of  
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Submitted by:  
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## 1. INTRODUCTION

The enteric nervous system is often considered as the body's second brain and the gut microbiome plays a crucial role in its maintenance and is also responsible for general health and well-being of the human body <sup>1</sup>. The ecosystem composition and the interactions between the gut organisms play important functional roles in aiding digestion, protection against pathogens <sup>2</sup> and has shown to affect the regulation of the immune system and mental health <sup>3</sup>. Dysregulations and imbalance within the gut microbiome have been attributed to diseases such as liver fibrosis <sup>4</sup> and inflammatory bowel disease <sup>5</sup>. Recent studies have also highlighted the involvement of gut organisms in cancer <sup>6</sup> and cardiovascular disease <sup>7</sup>.

Redundancy in metabolic networks is necessary for survival of an organism. In case of sudden perturbations, organisms with higher functional redundancies can survive better <sup>8</sup>. Further, the key metabolic reactions necessary for growth have a much higher redundancy in a particular medium <sup>9</sup>. Along with gene duplications, alternate metabolic pathways contribute to an organism's robustness permitting survival on available resources <sup>10</sup>.

MinReact algorithm <sup>11</sup> can find the minimum reactome of an organism by identifying essential reactions using pFBA (parsimonious Flux Balance Analysis), given an organism's metabolic network as input. In addition, for a few organisms more than one minimal network can be identified. Minimal reactomes give information on the most essential reactions for an organism and may also provide an evolutionary insight into transitions of the minimal reactomes of organisms to their present redundant form. In addition, minimal reactomes can be computed for various diet conditions (specified in AGORA database) to understand the key reactions in resource utilization. Furthermore, the growth rate cut-off and tolerance affecting the minimal reactome can be studied.

This study aims to analyze the redundancy and increase in metabolic reactome size in gut bacterial species by calculating inter-species similarities in minimal and wild type

reactomes and inter-species co-growth interactions. Further, the key question addressed here: Does increase in reactome size and alterations in metabolism lead to better interaction of an organism within its ecological niche?

## 2. METHODS

### 2.1. Finding Minimal Reactomes:

Minimal reactomes were identified using the MinReact algorithm <sup>11</sup> for the human gut organisms in the AGORA database <sup>12</sup>.

Minimal reactomes were computed by setting growth rate cut-off = 5% (Minimum % of growth to be retained) and tolerance = 0 (Minimum flux of a reaction to be considered active).

Diet constraints were imposed according to the AGORA database and minimal reactomes were computed for four conditions -

- i. No diet (Open fluxes)
- ii. High Fiber Diet
- iii. Vegetarian Diet
- iv. Mediterranean Diet

### 2.2. Computing Jaccard distance:

$$J(i,j) = \frac{\text{Reactome}(i) \cap \text{Reactome}(j)}{\text{Reactome}(i) \cup \text{Reactome}(j)}$$

Jaccard distance  $J(i,j)$  was computed as  $1 - \text{Metabolic distance}$  <sup>12</sup>. A total of 809 x 809 Jaccard indices were calculated for both the wild-type and minimal reactomes for gut microorganisms in the AGORA database.

Jaccard similarity scores range between [0-1], with identical species reconstructions having a Jaccard score of 1 and completely dissimilar species having a jaccard score of 0.

### 2.3 Computing pairwise interaction scores:

Constraint-based reconstruction and analysis (COBRA) <sup>13</sup> technique using genome scale metabolic reconstructions (GENREs) of organisms serves as a useful tool to model the gut-microbiome including their interactions, growth rates, metabolism and contribution of each organism to a community.

The COBRA toolbox function 'simulatePairwiseInteractions' predicts the type of interaction for a joint pair of models considering their individual growth rate and growth in the two-pair community. Six types of interactions have been defined:

1. Competition: both organisms grow slower in co-growth than separately.
2. Parasitism: one organism grows faster in co-growth than separately, while the other grows slower in co-growth than separately.
3. Amensalism: one organism's growth is unaffected by co-growth, while the other grows slower in co-growth than separately.
4. Neutralism: both organisms' growths are unaffected by co-growth.
5. Commensalism: one organism's growth is unaffected by co-growth, while the other grows faster in co-growth than separately.
6. Mutualism: both organisms grow faster in co-growth than separately.

Pairwise growth interactions were computed for WT reactomes imposing a High Fiber Diet.

### 3. RESULTS

#### 3.1. Minimal Reactomes on different diets

Minimal reactomes were computed for the AGORA database under 4 conditions.

- No Diet (all reaction fluxes set open [-1000, 1000])
- Vegetarian Diet
- High Fiber Diet
- Mediterranean Diet

The vital minimal number of reactions identified by MinReact varied slightly under the different conditions.

$$f = \frac{\text{No. of reactions in Minimal Reactome of an Organism}}{\text{No. of reactions in WT of an Organism}}$$

Fraction  $f$  was computed for organisms for the four conditions (Figure 1).

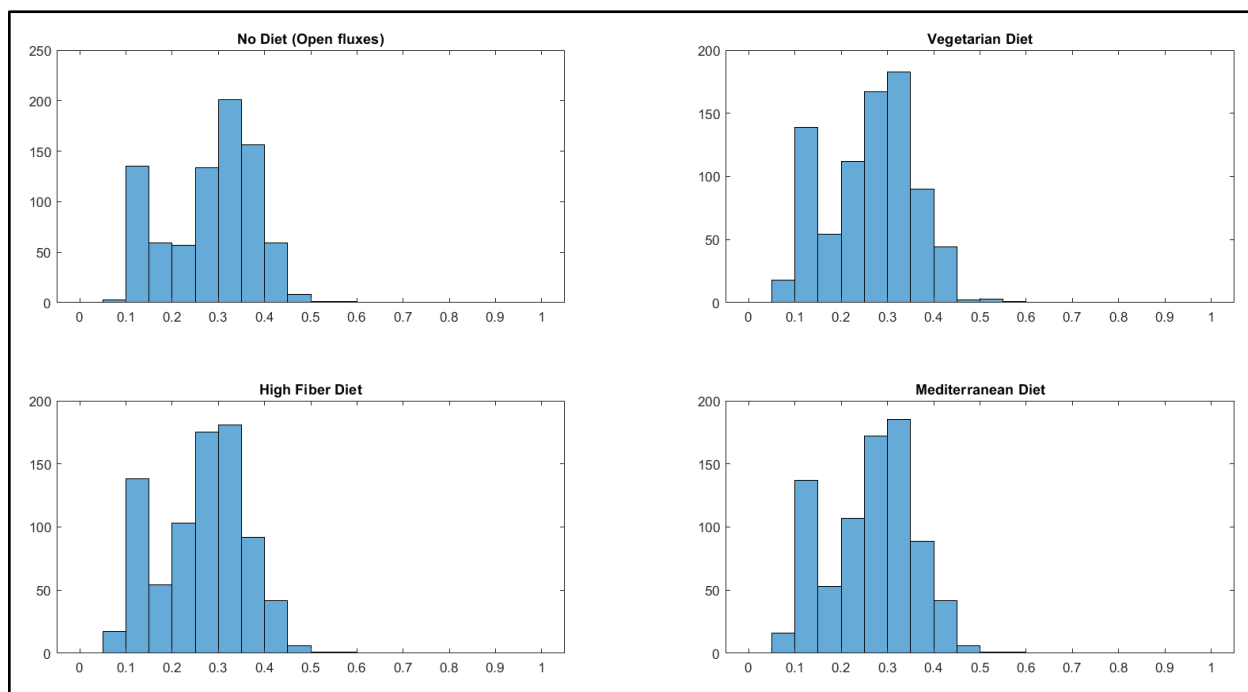


Figure 1:

Histogram of (No. of reactions in Minimal reactome of organism / No. of reactions in WT organism)  
% Only 1 minimal reactome considered, in case multiple minimal reactomes for an organism



Similar trends are observed in the diet conditions: On imposing diets, most gut organisms have only 10–40% reactions absolutely essential for minimal survival, the remaining 60–90% reactions are redundant.

Organisms with larger 'f' have smaller reactomes. Considering a high-fiber diet, the organisms having  $f > 0.43$  are listed in Table 1. A striking feature among these organisms is that they are primitive, and pathogenic (except *Campylobacter hominis*, *M. stadtmanae* and *Melainabacterium*) with a smaller number of genes and reactions. Ureaplasmas and mycoplasmas are the smallest free-living facultative anaerobes without a cell wall<sup>14</sup>. These organisms have small wild type reactomes and are already much more reduced when compared to the other gut bacteria.

It has been observed that many pathogenic bacteria and viruses have reduced genomes. This can be justified since these organisms derive nutrients and rely on host metabolism for survival.<sup>15</sup>

The peak observed for organisms having 10–15% of essential reactions have larger reactome sizes and comprise of species from *Bacteroidetes*, *Bifidobacterium*, *Clostridium*, *Enterococcus*, *E. coli*, *Lactobacillus*, *Ruminococcus*. These species also have high functional redundancy.

Horizontal gene transfer has been shown to contribute significantly to the genetic diversity in microorganisms including gut bacteria and have been shown to change the ecological and pathogenic traits of these species<sup>16</sup>.

It has been found that few species have lost non-essential genes based on its ecological environment. The gut associated *Melainabacteria* have reduced genomes compared to the soil associated *Melainabacteria* species. The gut lineage lacks genes for chemotaxis, production of few amino acids and nitrogen fixation<sup>17</sup>.

Table 1: Organisms with  $f > 0.43$  (Considering a High-Fiber Diet)

Organism	f	No. of reactions in WT Reactome	No. of reactions in Minimal Reactome	Pathogenic Role (Except <i>Campylobacter hominis</i> , <i>M. stadtmanae</i> and <i>Melainabacterium</i> )
<i>Mycoplasma hominis</i> _ATCC_23114	0.589	496	292	<i>M. hominis</i> is responsible for pelvic inflammatory disease and bacterial vaginosis <sup>18</sup> . It can also cause meningitis, pneumonia and abscesses in newborns <sup>19</sup> .
<i>Mycoplasma pneumoniae</i> _309	0.546	535	292	<i>M. pneumoniae</i> is responsible for respiratory tract infections and other non-pulmonary diseases including neurological, hepatic, cardiac diseases, hemolytic anemia, polyarthritis and erythema multiforme <sup>20</sup> .
<i>Alistipes putredinis</i> _DSM_17216	0.498	858	427	<i>A. putredinis</i> has been isolated from faeces, appendiceal tissue of patients with acute appendicitis, abdominal and rectal abscesses <sup>21</sup> .
<i>Methanosphaera stadtmanae</i> _DSM_3091	0.482	823	397	<i>M. stadtmanae</i> has been found to stimulate the development of a healthy intestinal epithelium and immune system and may prevent chronic inflammatory diseases, but their specific role is still unknown <sup>22</sup> .
<i>Campylobacter hominis</i> _ATCC_BAA_381	0.479	941	451	Only known campylobacter species that are not pathogenic to humans <sup>23</sup> .
<i>Bartonella quintana</i> _Toulouse	0.478	894	427	Etiologic agent of trench fever and causes bacillary angiomatosis <sup>24</sup> .
<i>Melainabacterium</i> _MEL_A1	0.451	913	412	Obligate anaerobic fermenters, close relatives of cyanobacteria <sup>25</sup> .
<i>Tropheryma whipplei</i> _str_Twist	0.451	845	381	Responsible for causing Whipple disease and also causes intestinal malabsorption that leads to cachexia <sup>26</sup> .
<i>Prevotella stercoraria</i> _DSM_18206	0.449	878	394	Anaerobe, mesophilic, gram- negative bacterium isolated from human faeces associated with colon dysbiosis in HIV-infected individuals <sup>27</sup> .
<i>Ureaplasma urealyticum</i> _serovar_8_str_ATCC_27618	0.441	664	293	Has pathogenic roles in genital tract infections and infertility <sup>28</sup> .

Campylobacter_cur vus_525_92	0.434	1027	446	<i>Campylobacter curvus</i> has been isolated from blood cultures of a patient with liver abscesses <sup>29</sup> .
Ureaplasma_parvu m_serovar_1_str_ TCC_27813	0.433	665	288	It is a mucosal parasite living within the genito-urinary tracts, causing male urethritis, adverse pregnancy outcomes, chorioamnionitis, surgical wound infections, neonatal meningitis, pelvic inflammatory diseases, pyelonephritis <sup>30</sup> .

### 3.2. Jaccard Distance (WT Reactome vs Minimal Reactome)

Jaccard indices were calculated for 809 x 809 wild type reactome pairs and repeated for the 809 x 809 minimal reactomes pairs of the same species, considering a High Fiber Diet. A positive correlation was observed. The species pairs can be classified into three categories:

- a. Species pairs that are more similar to each other w.r.t their minimal reactomes but diverge out and express alternate genes and less similar w.r.t wild type reactome.
- b. Species pairs that are less similar w.r.t to their minimal reactomes but more similar w.r.t their wild type reactomes.
- c. Species that are equally similar w.r.t to their minimal reactome and wild type reactome.

Studying the addition of reactions to minimal reactomes can analyze the redundancy and increase in metabolic reactome size in gut bacterial species.

Further, it would be interesting to find species that become more or less similar with respect to their reactomes in order to better cooperate/compete with each other.

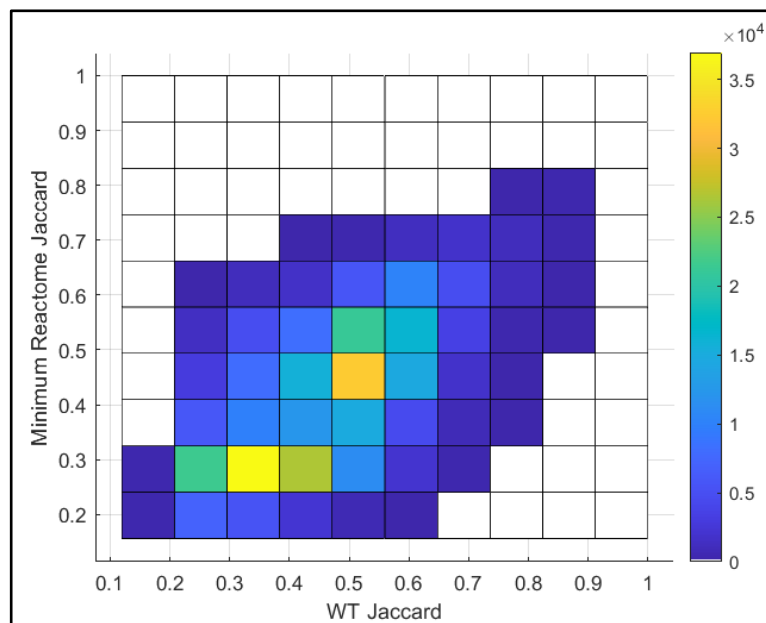
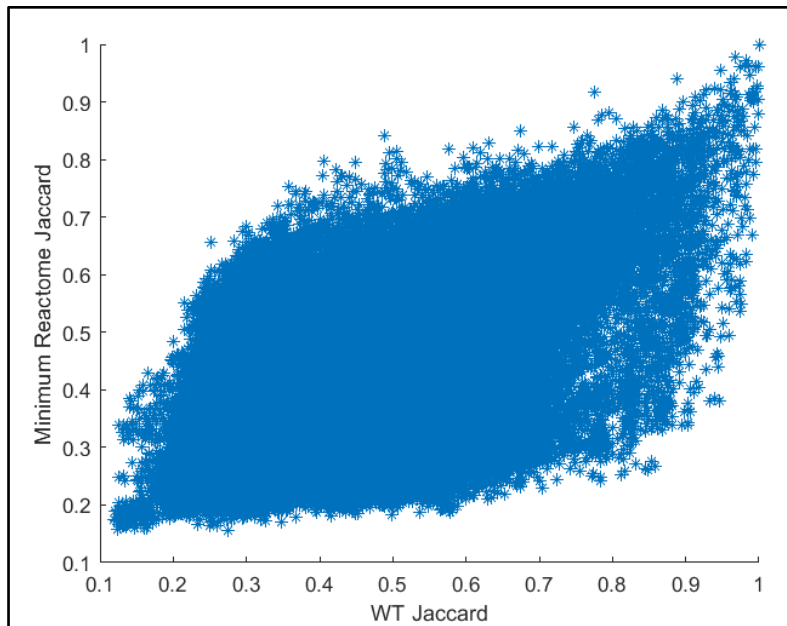


Figure 2:

Minimal Reactome Jaccard vs WT reactome Jaccard

% Only 1 minimal reactome considered, in case multiple minimal reactomes for an organism

% Minimal Reactome computed under High Fiber Diet.

It is expected for minimal reactomes to have higher jaccard indices, since essential vital reactions will remain the same across species.

However, the wild type reactomes appear to have higher jaccard indices on the whole.

(More yellow than blue)

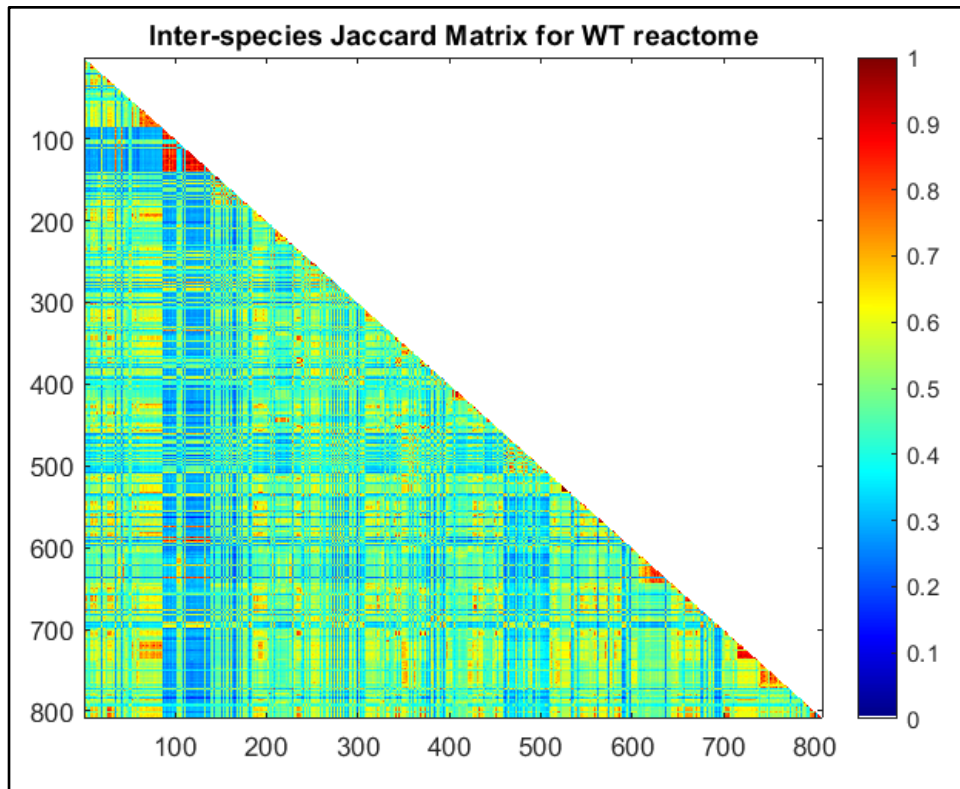


Figure 3: Inter-species Jaccard Matrix for WT reactome

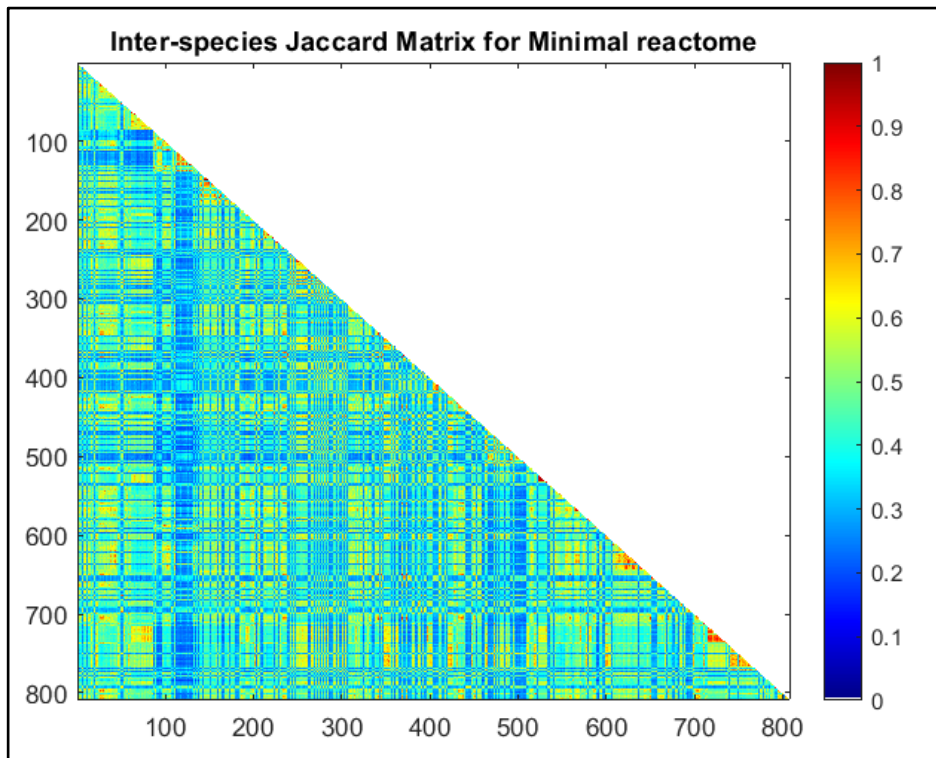


Figure 4: Inter-species Jaccard Matrix for Minimal reactome

% Only 1 minimal reactome considered, in case multiple minimal reactomes for an organism  
 % Minimal Reactome computed under High Fiber Diet.

Computing the difference between the two matrices (WT Jaccard Matrix - Minimal Reactome Jaccard matrix), a gaussian curve with (mean = 0.0476, std = 0.1044) can be fit. Most species tend to remain similar w.r.t their wild type and minimal reactomes. However, few species are observed on either extremes case(a) and case(b) as mentioned above.

Jaccard difference Matrix (JD) = WT Jaccard Matrix (809x809) - MR Jaccard Matrix (809x809).

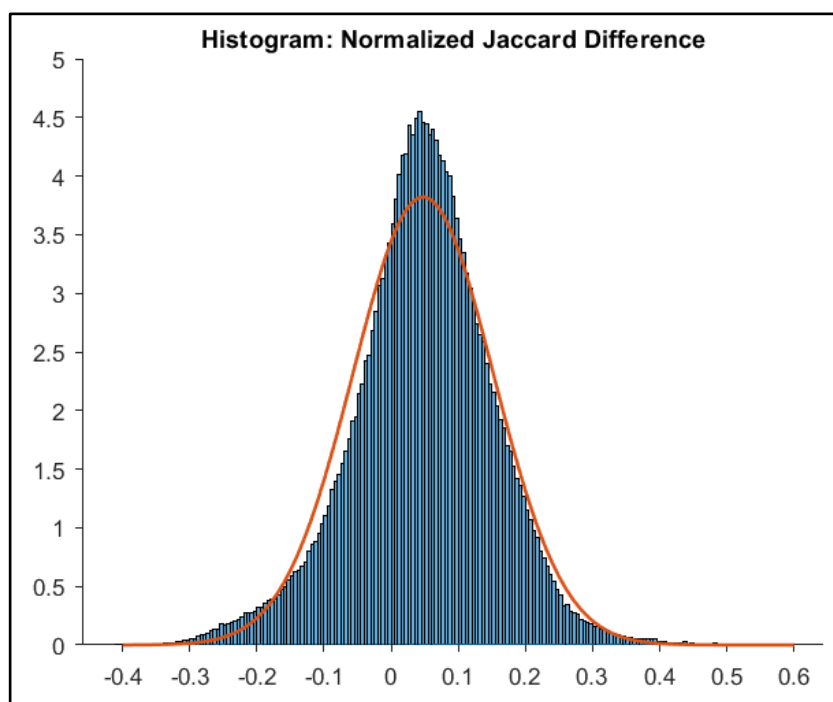


Figure 5: Histogram of Normalized Jaccard Difference Matrix  
% Only 1 minimal reactome considered, in case multiple minimal reactomes for an organism  
% Minimal Reactome computed under High Fiber Diet.

Considering the extremities in the Jaccard differences:

JD > 0.6 = 0 pairs, JD > 0.5 = 76 pairs, JD > 0.4 = 581 pairs.

JD < -0.4 = 1 pair, JD < -0.34 = 65 pairs, JD < -0.2 = 5916 pairs.

The 76 species pairs with JD > 0.5 and 65 species pairs with JD < -0.34 were considered for inter-species interactions.

### 3.3. Inter-species interactions

The 76 species pairs with  $JD > 0.5$  and 65 species pairs with  $JD < -0.34$  were considered for pairwise growth interactions (co-growth) under a High Fiber Diet.

Pairwise interactions were calculated only for the WT reactomes of these organisms and could not be computed for minimal reactomes of these organisms due to coupling constraints.

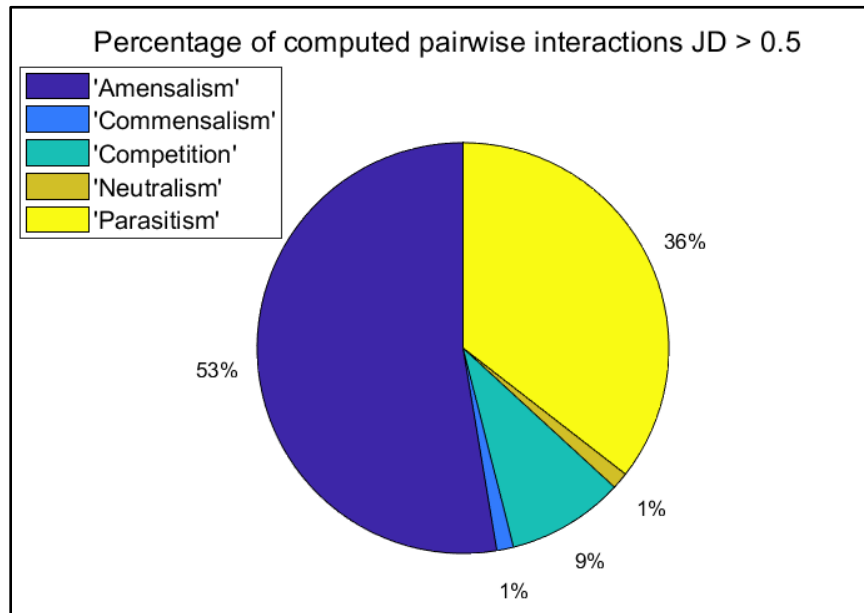


Figure 6: Interactions computed for WT reactomes with  $JD > 0.5$ .

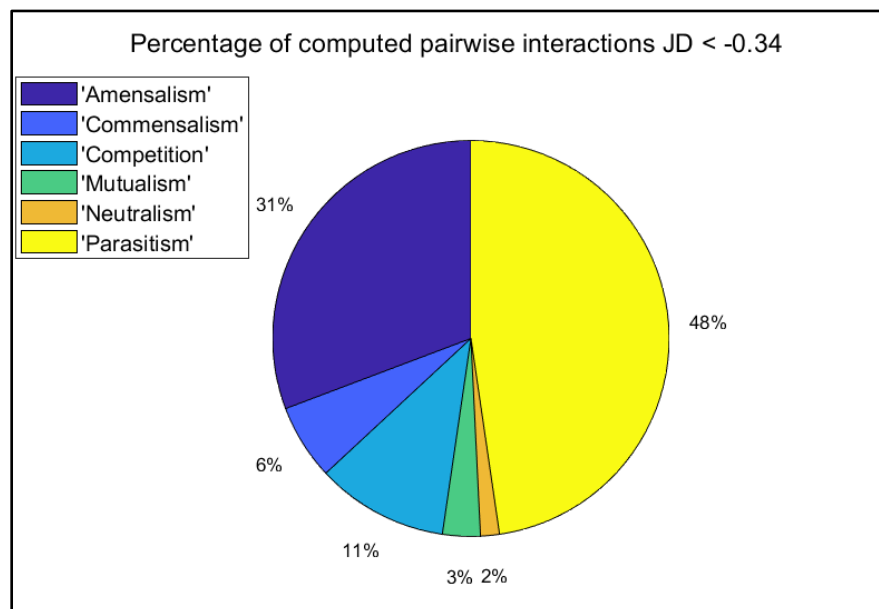


Figure 7: Interactions computed for WT reactomes with  $JD < 0.34$ .

For the 76 reactome pairs with JD > 0.5, following percentage interactions were found: Amensalism: 53%, Parasitism: 36%, Competition: 9%, Neutralism: 1%, Commensalism: 1%.

These pairs having high similarity in their WT reactomes w.r.t their minimal reactomes and do not display mutualism under high-fiber conditions.

Out of the 76 pairs (152 species (with repeats)), 121 of them belong to *Bacteroidetes*, 10 belong to *Proteobacteria*, 10 belong to *Firmicutes*, 10 belong to *Actinobacteria*, and 1 belongs to *Verrucomicrobia*.

The species '*Bacteroides\_sp\_1\_1\_30*' appears in 18 interactions, '*Bacteroides\_fluxus\_YIT\_12057*' appears in 11 interactions. Furthermore, '*Bacteroides\_fluxus\_YIT\_12057*' acts as a parasitism taker in all the 11 interactions.

Table 2: Interactions computed for WT reactomes with JD > 0.5.

S.No.	Organism1	Organism2	Jaccard Distance	Phylum-Organism1	Phylum-Organism2	Outcome_Organism1'	Outcome_Organism2'	Total_Outcome'
1	'Bacteroides_fragilis_NCTC_9343'	'Bacteroides_sp_1_1_30'	0.59158	Bacteroidetes	Bacteroidetes	'Amensalism_Unaffected'	'Amensalism_Affected'	'Amensalism'
2	'Bacteroides_fragilis_YCH46'	'Bacteroides_sp_1_1_30'	0.5875	Bacteroidetes	Bacteroidetes	'Amensalism_Unaffected'	'Amensalism_Affected'	'Amensalism'
3	'Bacteroides_sp_1_1_30'	'Bacteroides_thetaiotaomicron_VPI_5482'	0.58143	Bacteroidetes	Bacteroidetes	'Amensalism_Affected'	'Amensalism_Unaffected'	'Amensalism'
4	'Bacteroides_fragilis_638R'	'Bacteroides_sp_1_1_30'	0.57725	Bacteroidetes	Bacteroidetes	'Amensalism_Unaffected'	'Amensalism_Affected'	'Amensalism'
5	'Escherichia_coli_SE11'	'Escherichia_coli_str_K_12_substr_MG1655'	0.56528	Proteobacteria	Proteobacteria	'Competition'	'Competition'	'Competition'
6	'Clostridium_difficile_CD196'	'Clostridium_difficile_NAPO8'	0.56477	Firmicutes	Firmicutes	'Competition'	'Competition'	'Competition'
7	'Bacteroides_sp_1_1_30'	'Bacteroides_vulgatus_ATCC_8482'	0.56014	Bacteroidetes	Bacteroidetes	'Parasitism_Giver'	'Parasitism_Taker'	'Parasitism'
8	'Bacteroides_clarus_YIT_12056'	'Bacteroides_sp_1_1_30'	0.55995	Bacteroidetes	Bacteroidetes	'Amensalism_Unaffected'	'Amensalism_Affected'	'Amensalism'
9	'Clostridium_difficile_CD196'	'Clostridium_difficile_NAPO7'	0.55947	Firmicutes	Firmicutes	'Competition'	'Competition'	'Competition'
10	'Bacteroides_fluxus_YIT_12057'	'Bacteroides_sp_1_1_30'	0.55817	Bacteroidetes	Bacteroidetes	'Parasitism_Taker'	'Parasitism_Giver'	'Parasitism'
11	'Bacteroides_intestinalis_341_DSM_17393'	'Bacteroides_sp_1_1_30'	0.55556	Bacteroidetes	Bacteroidetes	'Parasitism_Taker'	'Parasitism_Giver'	'Parasitism'
12	'Escherichia_coli_O157_H7_str_Sakai'	'Escherichia_coli_SE11'	0.55525	Proteobacteria	Proteobacteria	'Amensalism_Unaffected'	'Amensalism_Affected'	'Amensalism'
13	'Escherichia_coli_SE11'	'Escherichia_coli_UTI89_UPEC'	0.55116	Proteobacteria	Proteobacteria	'Amensalism_Unaffected'	'Amensalism_Affected'	'Amensalism'
14	'Bacteroides_fragilis_YCH46'	'Bacteroides_sp_3_2_5'	0.54582	Bacteroidetes	Bacteroidetes	'Amensalism_Unaffected'	'Amensalism_Affected'	'Amensalism'
15	'Bacteroides_coprophilus_DSM_18228'	'Bacteroides_sp_1_1_30'	0.5438	Bacteroidetes	Bacteroidetes	'Parasitism_Taker'	'Parasitism_Giver'	'Parasitism'
16	'Akkermansia_muciniphila_ATCC_BAA_835'	'Bacteroides_sp_1_1_30'	0.54123	Verrucomicrobia	Bacteroidetes	'Amensalism_Unaffected'	'Amensalism_Affected'	'Amensalism'
17	'Bifidobacterium_longum_longum_CCUG_52486'	'Bifidobacterium_longum_longum_JCM_1217'	0.53886	Actinobacteria	Actinobacteria	'Parasitism_Taker'	'Parasitism_Giver'	'Parasitism'
18	'Bacteroides_fragilis_NCTC_9343'	'Bacteroides_sp_3_2_5'	0.53751	Bacteroidetes	Bacteroidetes	'Amensalism_Unaffected'	'Amensalism_Affected'	'Amensalism'
19	'Bacteroides_sp_1_1_30'	'Barnesiella_intestinihominis_YIT_11860'	0.5368	Bacteroidetes	Bacteroidetes	'Competition'	'Competition'	'Competition'
20	'Bacteroides_sp_3_1_40A'	'Bacteroides_vulgatus_ATCC_8482'	0.53599	Bacteroidetes	Bacteroidetes	'Amensalism_Affected'	'Amensalism_Unaffected'	'Amensalism'
21	'Bacteroides_sp_2_1_33B'	'Barnesiella_intestinihominis_YIT_11860'	0.53535	Bacteroidetes	Bacteroidetes	'Parasitism_Giver'	'Parasitism_Taker'	'Parasitism'
22	'Bacteroides_fluxus_YIT_12057'	'Bacteroides_sp_3_2_5'	0.53314	Bacteroidetes	Bacteroidetes	'Parasitism_Taker'	'Parasitism_Giver'	'Parasitism'
23	'Citrobacter_youngae_ATCC_29220'	'Escherichia_coli_SE11'	0.5309	Proteobacteria	Proteobacteria	'Amensalism_Affected'	'Amensalism_Unaffected'	'Amensalism'
24	'Bacteroides_fragilis_638R'	'Bacteroides_sp_D20'	0.52985	Bacteroidetes	Bacteroidetes	'Amensalism_Unaffected'	'Amensalism_Affected'	'Amensalism'
25	'Bacteroides_fragilis_NCTC_9343'	'Bacteroides_sp_1_1_14'	0.52873	Bacteroidetes	Bacteroidetes	'Amensalism_Unaffected'	'Amensalism_Affected'	'Amensalism'
26	'Bacteroides_caccae_ATCC_43185'	'Bacteroides_sp_1_1_30'	0.52855	Bacteroidetes	Bacteroidetes	'Amensalism_Unaffected'	'Amensalism_Affected'	'Amensalism'
27	'Bacteroides_sp_3_1_33FAA'	'Bacteroides_vulgatus_ATCC_8482'	0.5263	Bacteroidetes	Bacteroidetes	'Parasitism_Giver'	'Parasitism_Taker'	'Parasitism'
28	'Bacteroides_sp_1_1_14'	'Barnesiella_intestinihominis_YIT_11860'	0.52589	Bacteroidetes	Bacteroidetes	'Amensalism_Unaffected'	'Amensalism_Affected'	'Amensalism'
29	'Bacteroides_sp_3_1_19'	'Barnesiella_intestinihominis_YIT_11860'	0.52396	Bacteroidetes	Bacteroidetes	'Parasitism_Giver'	'Parasitism_Taker'	'Parasitism'
30	'Bacteroides_sp_1_1_30'	'Odoribacter_laneus_YIT_12061'	0.52177	Bacteroidetes	Bacteroidetes	'Parasitism_Giver'	'Parasitism_Taker'	'Parasitism'
31	'Bacteroides_fluxus_YIT_12057'	'Bacteroides_sp_3_1_40A'	0.52138	Bacteroidetes	Bacteroidetes	'Parasitism_Taker'	'Parasitism_Giver'	'Parasitism'
32	'Bacteroides_fluxus_YIT_12057'	'Bacteroides_sp_D2'	0.52136	Bacteroidetes	Bacteroidetes	'Parasitism_Taker'	'Parasitism_Giver'	'Parasitism'
33	'Bacteroides_fragilis_3_1_12'	'Bacteroides_sp_1_1_30'	0.5213	Bacteroidetes	Bacteroidetes	'Amensalism_Unaffected'	'Amensalism_Affected'	'Amensalism'
34	'Bacteroides_fragilis_YCH46'	'Bacteroides_sp_1_1_14'	0.52092	Bacteroidetes	Bacteroidetes	'Amensalism_Unaffected'	'Amensalism_Affected'	'Amensalism'
35	'Alistipes_shahii_WAL_8301'	'Bacteroides_sp_1_1_30'	0.52026	Bacteroidetes	Bacteroidetes	'Commensalism_Taker'	'Commensalism_Giver'	'Commensalism'
36	'Bacteroides_fluxus_YIT_12057'	'Bacteroides_sp_D1'	0.51859	Bacteroidetes	Bacteroidetes	'Parasitism_Taker'	'Parasitism_Giver'	'Parasitism'



37	'Bifidobacterium_longum_E18'	ifidobacterium_longum_longum_CCUG_5248	0.51856	Actinobacteria	Actinobacteria	'Parasitism_Giver'	'Parasitism_Taker'	'Parasitism'
38	'Bacteroides_fluxus_YIT_12057'	'Bacteroides_sp_1_1_14'	0.51852	Bacteroidetes	Bacteroidetes	'Parasitism_Taker'	'Parasitism_Giver'	'Parasitism'
39	'Bacteroides_fragilis_638R'	'Bacteroides_sp_3_1_40A'	0.51837	Bacteroidetes	Bacteroidetes	'Amensalism_Unaffected'	'Amensalism_Affected'	'Amensalism'
40	'Bacteroides_sp_1_1_30'	'Bacteroides_uniformis_ATCC_8492'	0.51772	Bacteroidetes	Bacteroidetes	'Amensalism_Affected'	'Amensalism_Unaffected'	'Amensalism'
41	'Bacteroides_fluxus_YIT_12057'	'Bacteroides_sp_3_1_33FAA'	0.51737	Bacteroidetes	Bacteroidetes	'Parasitism_Taker'	'Parasitism_Giver'	'Parasitism'
42	'Bacteroides_fragilis_638R'	'Bacteroides_sp_1_1_14'	0.51693	Bacteroidetes	Bacteroidetes	'Amensalism_Unaffected'	'Amensalism_Affected'	'Amensalism'
43	'Bifidobacterium_longum_DJO10A'	ifidobacterium_longum_longum_CCUG_5248	0.51671	Actinobacteria	Actinobacteria	'Parasitism_Giver'	'Parasitism_Taker'	'Parasitism'
44	'Bacteroides_sp_2_1_33B'	'Parabacteroides_distasonis_ATCC_8503'	0.51505	Bacteroidetes	Bacteroidetes	'Amensalism_Affected'	'Amensalism_Unaffected'	'Amensalism'
45	'Bacteroides_fragilis_638R'	'Bacteroides_sp_D1'	0.51497	Bacteroidetes	Bacteroidetes	'Amensalism_Unaffected'	'Amensalism_Affected'	'Amensalism'
46	'Bacteroides_fluxus_YIT_12057'	'Bacteroides_sp_3_1_33FAA'	0.51443	Bacteroidetes	Bacteroidetes	'Competition'	'Competition'	'Competition'
47	'Escherichia_coli_SE11'	'Escherichia_sp_3_2_53FAA'	0.51387	Proteobacteria	Proteobacteria	'Amensalism_Unaffected'	'Amensalism_Affected'	'Amensalism'
48	'Bacteroides_sp_3_1_19'	'Parabacteroides_distasonis_ATCC_8503'	0.51386	Bacteroidetes	Bacteroidetes	'Amensalism_Affected'	'Amensalism_Unaffected'	'Amensalism'
49	'Bacteroides_sp_1_1_30'	'Parabacteroides_distasonis_ATCC_8503'	0.51233	Bacteroidetes	Bacteroidetes	'Parasitism_Taker'	'Parasitism_Giver'	'Parasitism'
50	'Bacteroides_sp_D1'	'Barnesiella_intestinihominis_YIT_11860'	0.51215	Bacteroidetes	Bacteroidetes	'Amensalism_Unaffected'	'Amensalism_Affected'	'Amensalism'
51	'Bacteroides_fluxus_YIT_12057'	'Bacteroides_sp_D22'	0.51131	Bacteroidetes	Bacteroidetes	'Parasitism_Taker'	'Parasitism_Giver'	'Parasitism'
52	'Bacteroides_sp_D20'	'Bacteroides_vulgatus_ATCC_8482'	0.5099	Bacteroidetes	Bacteroidetes	'Parasitism_Giver'	'Parasitism_Taker'	'Parasitism'
53	'Bacteroides_sp_3_1_33FAA'	'Odoribacter_laneus_YIT_12061'	0.50981	Bacteroidetes	Bacteroidetes	'Parasitism_Giver'	'Parasitism_Taker'	'Parasitism'
54	'Anaerostipes_hadrus_DSM_3319'	'Clostridium_sp_SS2_1'	0.50956	Firmicutes	Firmicutes	'Neutralism'	'Neutralism'	'Neutralism'
55	'Bacteroides_fragilis_NCTC_9343'	'Bacteroides_sp_D20'	0.50824	Bacteroidetes	Bacteroidetes	'Amensalism_Unaffected'	'Amensalism_Affected'	'Amensalism'
56	'Bacteroides_fragilis_638R'	'Bacteroides_sp_D2'	0.50823	Bacteroidetes	Bacteroidetes	'Amensalism_Unaffected'	'Amensalism_Affected'	'Amensalism'
57	'Bacteroides_sp_1_1_14'	'Bacteroides_thetaiotaomicron_VPI_5482'	0.50732	Bacteroidetes	Bacteroidetes	'Amensalism_Affected'	'Amensalism_Unaffected'	'Amensalism'
58	'Lactobacillus_helveticus_DPC_4571'	'Lactobacillus_helveticus_DSM_20075'	0.50723	Firmicutes	Firmicutes	'Parasitism_Taker'	'Parasitism_Giver'	'Parasitism'
59	'Bacteroides_fluxus_YIT_12057'	'Bacteroides_sp_D20'	0.50722	Bacteroidetes	Bacteroidetes	'Parasitism_Taker'	'Parasitism_Giver'	'Parasitism'
60	'Bifidobacterium_longum_NCC2705'	Bifidobacterium_longum_longum_JCM_1217	0.50712	Actinobacteria	Actinobacteria	'Competition'	'Competition'	'Competition'
61	'Bacteroides_fluxus_YIT_12057'	'Bacteroides_xylanisolvens_SD_CC_1b'	0.50695	Bacteroidetes	Bacteroidetes	'Parasitism_Taker'	'Parasitism_Giver'	'Parasitism'
62	'Bacteroides_fragilis_638R'	'Bacteroides_sp_2_1_22'	0.50671	Bacteroidetes	Bacteroidetes	'Amensalism_Unaffected'	'Amensalism_Affected'	'Amensalism'
63	'Bacteroides_sp_3_1_33FAA'	'Barnesiella_intestinihominis_YIT_11860'	0.5061	Bacteroidetes	Bacteroidetes	'Amensalism_Unaffected'	'Amensalism_Affected'	'Amensalism'
64	'Bacteroides_fluxus_YIT_12057'	'Bacteroides_sp_2_1_22'	0.5051	Bacteroidetes	Bacteroidetes	'Parasitism_Taker'	'Parasitism_Giver'	'Parasitism'
65	'Bacteroides_fragilis_YCH46'	'Bacteroides_sp_D20'	0.50429	Bacteroidetes	Bacteroidetes	'Amensalism_Unaffected'	'Amensalism_Affected'	'Amensalism'
66	'Bacteroides_fragilis_638R'	'Bacteroides_sp_3_2_5'	0.50409	Bacteroidetes	Bacteroidetes	'Amensalism_Unaffected'	'Amensalism_Affected'	'Amensalism'
67	'Bacteroides_sp_1_1_14'	'Bacteroides_sp_1_1_30'	0.50387	Bacteroidetes	Bacteroidetes	'Parasitism_Taker'	'Parasitism_Giver'	'Parasitism'
68	'Lactobacillus_casei_casei_BL23'	'Lactobacillus_rhamnosus_GG_ATCC_53103'	0.50348	Firmicutes	Firmicutes	'Parasitism_Taker'	'Parasitism_Giver'	'Parasitism'
69	'Bacteroides_fragilis_NCTC_9343'	'Bacteroides_sp_2_1_22'	0.50322	Bacteroidetes	Bacteroidetes	'Amensalism_Unaffected'	'Amensalism_Affected'	'Amensalism'
70	'Bacteroides_fragilis_NCTC_9343'	'Bacteroides_sp_3_1_40A'	0.50283	Bacteroidetes	Bacteroidetes	'Amensalism_Unaffected'	'Amensalism_Affected'	'Amensalism'
71	'Bacteroides_fragilis_YCH46'	'Bacteroides_sp_3_1_40A'	0.50186	Bacteroidetes	Bacteroidetes	'Amensalism_Unaffected'	'Amensalism_Affected'	'Amensalism'
72	'Parabacteroides_distasonis_ATCC_8503'	'Parabacteroides_sp_D13'	0.50157	Bacteroidetes	Bacteroidetes	'Amensalism_Unaffected'	'Amensalism_Affected'	'Amensalism'
72	'Parabacteroides_distasonis_ATCC_8503'	'Parabacteroides_sp_D13'	0.50157	Bacteroidetes	Bacteroidetes	'Amensalism_Unaffected'	'Amensalism_Affected'	'Amensalism'
73	'Bacteroides_fragilis_YCH46'	'Bacteroides_sp_3_1_33FAA'	0.50067	Bacteroidetes	Bacteroidetes	'Competition'	'Competition'	'Competition'
74	'Bifidobacterium_longum_DJO10A'	'Bifidobacterium_longum_NCC2705'	0.50052	Actinobacteria	Actinobacteria	'Amensalism_Unaffected'	'Amensalism_Affected'	'Amensalism'
75	'Bacteroides_fragilis_NCTC_9343'	'Bacteroides_sp_3_1_33FAA'	0.50014	Bacteroidetes	Bacteroidetes	'Amensalism_Unaffected'	'Amensalism_Affected'	'Amensalism'
76	'Bacteroides_sp_2_1_22'	'Barnesiella_intestinihominis_YIT_11860'	0.5	Bacteroidetes	Bacteroidetes	'Amensalism_Unaffected'	'Amensalism_Affected'	'Amensalism'

For the 65 reactome pairs with  $JD < -0.34$ , following percentage interactions were found: Parasitism: 48%, Amensalism: 31%, Competition: 11%, Commensalism: 6%, Mutualism: 3%, Neutralism: 2%.

These pairs have low similarity in their WT reactomes w.r.t their minimal reactomes. Out of the 65 pairs (130 species (with repeats)), 53 of them belong to *Bacteroidetes*, 41 belong to *Firmicutes*, 13 belong to *Actinobacteria*, 9 belong to *Proteobacteria*, 8 belong to *Fusobacteria*, 5 belong to *Verrucomicrobia* and 1 belongs to *Euryarchaeota*.

Further the organism '*Alistipes\_shahii\_WAL\_8301*' belonging to *Bacteroidetes* appears in 13 interactions.

Table 3: Interactions computed for WT reactomes with JD < -0.34.

S.No	Organism1	Organism2	Jaccard Distance	Phylum-Organism1	Phylum-Organism2	Outcome_Organism1	Outcome_Organism2	Total_Outcome
1	'Acidaminococcus_sp_D21'	'Holdemania_filiformis_VPI_J1_318_1_DSM_12042'	-0.4058	Firmicutes	Firmicutes	'Amensalism_Affected'	'Amensalism_Unaffected'	'Amensalism'
2	'Lactobacillus_paracasei_subsp_paracasei_ATCC_25305'	'Lactobacillus_reuteri_F275_JCM_11112'	-0.3959	Firmicutes	Firmicutes	'Parasitism_Giver'	'Parasitism_Taker'	'Parasitism'
3	'Bacteroides_fluxus_YIT_12057'	'Prevotella_bryantii_B14'	-0.391	Bacteroidetes	Bacteroidetes	'Competition'	'Competition'	'Competition'
4	'Eubacterium_biforme_DSM_3989'	'Parvimonas_micro_ATCC_33270'	-0.3821	Firmicutes	Firmicutes	'Parasitism_Giver'	'Parasitism_Taker'	'Parasitism'
5	'Eubacterium_biforme_DSM_3989'	'Fusobacterium_periodonticum_2_1_31'	-0.3801	Firmicutes	Fusobacteria	'Parasitism_Taker'	'Parasitism_Giver'	'Parasitism'
6	'Alistipes_shahii_WAL_8301'	'Prevotella_bryantii_B14'	-0.3775	Bacteroidetes	Bacteroidetes	'Amensalism_Unaffected'	'Amensalism_Affected'	'Amensalism'
7	'Bacteroides_thetaiotaomicron_VPI_5482'	'Odoribacter_splanchnicus_1651_6_DSM_20712'	-0.3753	Bacteroidetes	Bacteroidetes	'Parasitism_Giver'	'Parasitism_Taker'	'Parasitism'
8	'Akkermansia_muciniphila_ATCC_BAA_835'	'Prevotella_disiensis_JCM_6334'	-0.3735	errucomicrob	Bacteroidetes	'Parasitism_Giver'	'Parasitism_Taker'	'Parasitism'
9	'Odoribacter_laneus_YIT_12061'	'Prevotella_buccae_ATCC_33574'	-0.3724	Bacteroidetes	Bacteroidetes	'Amensalism_Unaffected'	'Amensalism_Affected'	'Amensalism'
10	'Alistipes_shahii_WAL_8301'	'Mobiluncus_curtisii_ATCC_43063'	-0.3702	Bacteroidetes	Actinobacteria	'Amensalism_Unaffected'	'Amensalism_Affected'	'Amensalism'
11	'Clostridium_sporoforme_DSM_1552'	'Coprobacillus_sp_D7'	-0.3663	Firmicutes	Firmicutes	'Parasitism_Taker'	'Parasitism_Giver'	'Parasitism'
12	'Abiotrophia_defectiva_ATCC_49176'	'Clostridium_sp_SY8519'	-0.3657	Firmicutes	Firmicutes	'Parasitism_Giver'	'Parasitism_Taker'	'Parasitism'
13	'Clostridium_celatum_DSM_1785'	'Solobacterium_moorei_DSM_22971'	-0.3647	Firmicutes	Firmicutes	'Parasitism_Giver'	'Parasitism_Taker'	'Parasitism'
14	'Dysgonomonas_gadei_ATCC_BAA_286'	'Prevotella_corporis_DSM_18810'	-0.3644	Bacteroidetes	Bacteroidetes	'Amensalism_Affected'	'Amensalism_Unaffected'	'Amensalism'
15	'Alistipes_indistinctus_YIT_12060'	'Prevotella_bryantii_C21a'	-0.3629	Bacteroidetes	Bacteroidetes	'Competition'	'Competition'	'Competition'
16	'Clostridium_celatum_DSM_1785'	'Eubacterium_brachy_ATCC_33089'	-0.3625	Firmicutes	Firmicutes	'Parasitism_Giver'	'Parasitism_Taker'	'Parasitism'
17	'Odoribacter_laneus_YIT_12061'	'Prevotella_bryantii_B14'	-0.3613	Bacteroidetes	Bacteroidetes	'Competition'	'Competition'	'Competition'
18	'Bacteroides_fluxus_YIT_12057'	'Prevotella_buccae_ATCC_33574'	-0.3608	Bacteroidetes	Bacteroidetes	'Competition'	'Competition'	'Competition'
19	'Alistipes_shahii_WAL_8301'	'Prevotella_buccae_ATCC_33574'	-0.3605	Bacteroidetes	Bacteroidetes	'Amensalism_Unaffected'	'Amensalism_Affected'	'Amensalism'
20	'Clostridium_celatum_DSM_1785'	'Streptococcus_peroris_ATCC_700780'	-0.3602	Firmicutes	Firmicutes	'Parasitism_Taker'	'Parasitism_Giver'	'Parasitism'
21	'Clostridium_scindens_ATCC_35704'	'Parvimonas_micro_ATCC_33270'	-0.3591	Firmicutes	Firmicutes	'Amensalism_Unaffected'	'Amensalism_Affected'	'Amensalism'
22	'Alistipes_shahii_WAL_8301'	'Campylobacter_guilliermondii_ATCC_35061'	-0.3586	Firmicutes	Proteobacteria	'Amensalism_Unaffected'	'Amensalism_Affected'	'Amensalism'
23	'Eubacterium_biforme_DSM_3989'	'Fusobacterium_gonidiaformans_ATCC_25563'	-0.3574	Firmicutes	Fusobacteria	'Parasitism_Taker'	'Parasitism_Giver'	'Parasitism'
24	'Fusobacterium_nucleatum_subsp_animalis_7_1'	'Holdemania_filiformis_VPI_J1_318_1_DSM_12042'	-0.3558	Fusobacteria	Firmicutes	'Parasitism_Giver'	'Parasitism_Taker'	'Parasitism'
25	'Clostridium_celatum_DSM_1785'	'Peptoniphilus_harei_ACS_146_V_Sch2b'	-0.3551	Firmicutes	Firmicutes	'Parasitism_Giver'	'Parasitism_Taker'	'Parasitism'
26	'Bacteroides_clarus_YIT_12056'	'Neisseria_macacae_ATCC_33926'	-0.3544	Bacteroidetes	Proteobacteria	'Parasitism_Taker'	'Parasitism_Giver'	'Parasitism'
27	'Bifidobacterium_bifidum_517'	'Bifidobacterium_breve_HPH0326'	-0.3539	Actinobacteria	Actinobacteria	'Amensalism_Affected'	'Amensalism_Unaffected'	'Amensalism'
28	'Eubacterium_biforme_DSM_3989'	'Fusobacterium_nucleatum_subsp_animalis_7_1'	-0.3539	Firmicutes	Fusobacteria	'Parasitism_Taker'	'Parasitism_Giver'	'Parasitism'
29	'Holdemania_filiformis_VPI_J1_318_1_DSM_12042'	'Parvimonas_micro_ATCC_33270'	-0.3539	Firmicutes	Firmicutes	'Parasitism_Giver'	'Parasitism_Taker'	'Parasitism'
30	'Abiotrophia_defectiva_ATCC_49176'	'Weissella_cibaria_KACC_11862'	-0.3521	Firmicutes	Firmicutes	'Mutualism'	'Mutualism'	'Mutualism'
31	'Alistipes_shahii_WAL_8301'	'Neisseria_macacae_ATCC_33926'	-0.3516	Bacteroidetes	Proteobacteria	'Competition'	'Competition'	'Competition'
32	'Alloprevotella_tannerae_ATCC_51259'	'Bacteroides_fluxus_YIT_12057'	-0.3514	Bacteroidetes	Bacteroidetes	'Amensalism_Unaffected'	'Amensalism_Affected'	'Amensalism'
33	'Collinsella_stercoris_DSM_13279'	'Parvimonas_micro_ATCC_33270'	-0.3509	Actinobacteria	Firmicutes	'Amensalism_Affected'	'Amensalism_Unaffected'	'Amensalism'
34	'Alistipes_shahii_WAL_8301'	'Fusobacterium_russii_ATCC_25533'	-0.3507	Bacteroidetes	Fusobacteria	'Competition'	'Competition'	'Competition'
35	'Alistipes_shahii_WAL_8301'	'Lautropia_mirabilis_ATCC_51599'	-0.3504	Bacteroidetes	Proteobacteria	'Parasitism_Giver'	'Parasitism_Taker'	'Parasitism'
36	'Abiotrophia_defectiva_ATCC_49176'	'Cryptobacterium_curtum_DSM_15641'	-0.3482	Firmicutes	Actinobacteria	'Commensalism_Giver'	'Commensalism_Taker'	'Commensalism'
37	'Bacteroides_fragilis_NCTC_9343'	'Odoribacter_splanchnicus_1651_6_DSM_20712'	-0.3474	Bacteroidetes	Bacteroidetes	'Parasitism_Giver'	'Parasitism_Taker'	'Parasitism'
38	'Dysgonomonas_gadei_ATCC_BAA_286'	'Prevotella_denticola_DSM20614'	-0.3469	Bacteroidetes	Bacteroidetes	'Amensalism_Affected'	'Amensalism_Unaffected'	'Amensalism'
39	'Alistipes_shahii_WAL_8301'	'Bifidobacterium_coryneforme_DSM_20216'	-0.3467	Bacteroidetes	Actinobacteria	'Parasitism_Giver'	'Parasitism_Taker'	'Parasitism'
40	'Akkermansia_muciniphila_ATCC_BAA_835'	'Neisseria_macacae_ATCC_33926'	-0.3466	errucomicrob	Proteobacteria	'Commensalism_Taker'	'Commensalism_Giver'	'Commensalism'
41	'Bifidobacterium_longum_NCC2705'	'Bifidobacterium_longum_infantis_ATCC_15697'	-0.3464	Actinobacteria	Actinobacteria	'Amensalism_Affected'	'Amensalism_Unaffected'	'Amensalism'
42	'Fusobacterium_nucleatum_subsp_animalis_3_1_33'	'Holdemania_filiformis_VPI_J1_318_1_DSM_12042'	-0.3461	Fusobacteria	Firmicutes	'Competition'	'Competition'	'Competition'
43	'Bifidobacterium_longum_longum_CCUG_52486'	'Fusobacterium_nucleatum_subsp_animalis_3_1_33'	-0.346	Actinobacteria	Fusobacteria	'Parasitism_Taker'	'Parasitism_Giver'	'Parasitism'
44	'Bacteroides_fluxus_YIT_12057'	'Bifidobacterium_coryneforme_DSM_20216'	-0.3458	Bacteroidetes	Actinobacteria	'Parasitism_Giver'	'Parasitism_Taker'	'Parasitism'
45	'Bacteroides_clarus_YIT_12056'	'Neisseria_mucosa_ATCC_25996'	-0.3455	Bacteroidetes	Proteobacteria	'Parasitism_Taker'	'Parasitism_Giver'	'Parasitism'
46	'Actinomyces_odontolyticus_ATCC_17982'	'Helicobacter_pulorum_MIT_98_5489'	-0.3452	Actinobacteria	Proteobacteria	'Mutualism'	'Mutualism'	'Mutualism'
47	'Bacteroides_fragilis_YCH46'	'Odoribacter_splanchnicus_1651_6_DSM_20712'	-0.3438	Bacteroidetes	Bacteroidetes	'Parasitism_Giver'	'Parasitism_Taker'	'Parasitism'
48	'Alistipes_finegoldii_DSM_17242'	'Alistipes_shahii_WAL_8301'	-0.3438	Bacteroidetes	Bacteroidetes	'Amensalism_Unaffected'	'Amensalism_Affected'	'Amensalism'
49	'Bacteroides_thetaiotaomicron_VPI_5482'	'Desulfovibrio_desulfuricans_subsp_desulfuricans_DSM_5213'	-0.3433	Bacteroidetes	Proteobacteria	'Neutralism'	'Neutralism'	'Neutralism'
50	'Alistipes_shahii_WAL_8301'	'Alloprevotella_tannerae_ATCC_51259'	-0.3432	Bacteroidetes	Bacteroidetes	'Amensalism_Unaffected'	'Amensalism_Affected'	'Amensalism'
51	'Alistipes_shahii_WAL_8301'	'Methanococcus_marisnigri_DSM_12130'	-0.3431	Bacteroidetes	Euryarchaeota	'Amensalism_Unaffected'	'Amensalism_Affected'	'Amensalism'
52	'Eubacterium_biforme_DSM_3989'	'Fusobacterium_nucleatum_subsp_animalis_3_1_33'	-0.343	Firmicutes	Fusobacteria	'Parasitism_Taker'	'Parasitism_Giver'	'Parasitism'
53	'Alistipes_shahii_WAL_8301'	'Prevotella_denticola_F0289'	-0.3427	Bacteroidetes	Bacteroidetes	'Amensalism_Affected'	'Amensalism_Unaffected'	'Amensalism'
54	'Mobiluncus_curtisii_ATCC_43063'	'Odoribacter_laneus_YIT_12061'	-0.3423	Actinobacteria	Bacteroidetes	'Amensalism_Affected'	'Amensalism_Unaffected'	'Amensalism'
55	'Akkermansia_muciniphila_ATCC_BAA_835'	'Solanomonas_lenta_DSM_16282'	-0.3422	errucomicrob	Proteobacteria	'Commensalism_Giver'	'Commensalism_Taker'	'Commensalism'
56	'Akkermansia_muciniphila_ATCC_BAA_835'	'Prevotella_intermedia_ATCC_25611'	-0.342	errucomicrob	Bacteroidetes	'Parasitism_Giver'	'Parasitism_Taker'	'Parasitism'
57	'Dysgonomonas_gadei_ATCC_BAA_286'	'Prevotella_timonensis_4401737'	-0.342	Bacteroidetes	Bacteroidetes	'Parasitism_Giver'	'Parasitism_Taker'	'Parasitism'
58	'Alistipes_shahii_WAL_8301'	'Neisseria_mucosa_ATCC_25996'	-0.3416	Bacteroidetes	Proteobacteria	'Amensalism_Unaffected'	'Amensalism_Affected'	'Amensalism'
59	'Bifidobacterium_breve_HPH0326'	'Prevotella_intermedia_17'	-0.3414	Actinobacteria	Bacteroidetes	'Parasitism_Giver'	'Parasitism_Taker'	'Parasitism'
60	'Clostridium_celatum_DSM_1785'	'Enterococcus_durans_ATCC_6056'	-0.3411	Firmicutes	Firmicutes	'Parasitism_Taker'	'Parasitism_Giver'	'Parasitism'
61	'Abiotrophia_defectiva_ATCC_49176'	'Eubacterium_ramulus_ATCC_29099'	-0.341	Firmicutes	Firmicutes	'Parasitism_Taker'	'Parasitism_Giver'	'Parasitism'
62	'Acidaminococcus_sp_D21'	'Lactobacillus_casei_ATCC_334'	-0.3408	Firmicutes	Firmicutes	'Parasitism_Giver'	'Parasitism_Taker'	'Parasitism'
63	'Abiotrophia_defectiva_ATCC_49176'	'Clostridium_bifermentans_ATCC_19299'	-0.3407	Firmicutes	Firmicutes	'Commensalism_Taker'	'Commensalism_Giver'	'Commensalism'
64	'Bacteroides_massiliensis_B846dLKV334'	'Dysgonomonas_gadei_ATCC_BAA_286'	-0.3406	Bacteroidetes	Bacteroidetes	'Parasitism_Giver'	'Parasitism_Taker'	'Parasitism'
65	'Dysgonomonas_gadei_ATCC_BAA_286'	'Prevotella_disiensis_JCM_6334'	-0.3405	Bacteroidetes	Bacteroidetes	'Amensalism_Affected'	'Amensalism_Unaffected'	'Amensalism'

In general, phylogenetically related organisms are likely to share more similarities in their metabolic networks and compete with each other. But there are classes of organisms cooperating in spite of being close relatives.

Furthermore, even distant organisms tend to behave metabolically similar. They may have a greater genetic distance but smaller metabolic distance. The power of micro-organisms to adapt and regulate their metabolomes is puzzling.

It would be interesting to see how minimal reactomes of these organisms interact with each other. If there is high resource overlap within the minimal reactomes, they are more likely to compete within a community. However, in order to cooperate/compete, they might have evolved to their present reactomes.

#### 4. CONCLUSIONS

Even though organisms can evolve minimally, they tend to adapt and express more reactions in order to cooperate or compete better.

Analyzing the organisms with the greatest deviations between their wild type and minimal reactome might give an insight to the organisms with the greatest change in their metabolic status in order to cooperate/compete better within its niche. For a given organism, more than one minimal reactome may be possible, and which among these is more optimal in a given situation is to be understood. The varied phylogeny obtained from minimal reactomes may give an insight to the different paths traversed and the new genes evolved in different organisms and also adaptations to the change in environment.

Furthermore, the scores and interactions for various other diet conditions, growth rate cutoff and tolerance value needs consideration.

## 5. REFERENCES

1. Ochoa-Repáraz, J. & Kasper, L. H. The Second Brain: Is the Gut Microbiota a Link Between Obesity and Central Nervous System Disorders? *Curr. Obes. Rep.* 5, 51–64 (2016).
2. Lynch SV, Pedersen O. The Human Intestinal Microbiome in Health and Disease. *N Engl J Med.* 2016 Dec 15;375(24):2369–2379. doi: 10.1056/NEJMra1600266. PMID: 27974040.
3. Petra, A. I. *et al.* Gut–Microbiota–Brain Axis and Its Effect on Neuropsychiatric Disorders With Suspected Immune Dysregulation. *Clin. Ther.* 37, 984–995 (2015).
4. Ohtani, N. & Kawada, N. Role of the Gut–Liver Axis in Liver Inflammation, Fibrosis, and Cancer: A Special Focus on the Gut Microbiota Relationship. *Hepatol. Commun.* 3, 456–470 (2019).
5. Loh, G. & Blaut, M. Role of commensal gut bacteria in inflammatory bowel diseases. *Gut Microbes* 3, 544–555 (2012).
6. Vivarelli, S. *et al.* Gut Microbiota and Cancer: From Pathogenesis to Therapy. *Cancers* 11, (2019).
7. Yoshida, N., Yamashita, T. & Hirata, K. Gut Microbiome and Cardiovascular Diseases. *Diseases* 6, (2018).
8. Wang, Z. & Zhang, J. Abundant Indispensable Redundancies in Cellular Metabolic Networks. *Genome Biol. Evol.* 1, 23–33 (2009).
9. Sambamoorthy, G. & Raman, K. Understanding the evolution of functional redundancy in metabolic networks. *Bioinformatics* 34, i981–i987 (2018).
10. Mahadevan, R. & Lovley, D. R. The Degree of Redundancy in Metabolic Genes Is Linked to Mode of Metabolism. *Biophys. J.* 94, 1216–1220 (2008).
11. Sambamoorthy, G. & Raman, K. MinReact: a systematic approach for identifying minimal metabolic networks. *Bioinformatics* 36, 4309–4315 (2020).
12. Magnúsdóttir, S. *et al.* Generation of genome-scale metabolic reconstructions for 773 members of the human gut microbiota. *Nat. Biotechnol.* 35, 81–89 (2017).
13. Schellenberger, J. *et al.* Quantitative prediction of cellular metabolism with constraint-based models: the COBRA Toolbox v2.0. *Nat. Protoc.* 6, 1290–1307 (2011).
14. Cell Wall–Deficient Bacteria: Mycoplasma and Ureaplasma. *Clinical Gate* <https://clinicalgate.com/cell-wall-deficient-bacteria-mycoplasma-and-ureaplasma/> (2015).

15. Razin S, Hayflick L. Highlights of mycoplasma research--an historical perspective. *Biologicals*. 2010 Mar;38(2):183-90. doi: 10.1016/j.biologicals.2009.11.008. Epub 2010 Feb 10. PMID: 20149687.
16. Ochman, H., Lawrence, J. & Groisman, E. Lateral gene transfer and the nature of bacterial innovation. *Nature* 405, 299-304 (2000). <https://doi.org/10.1038/35012500>
17. Di Rienzi, S. C. *et al.* The human gut and groundwater harbor non-photosynthetic bacteria belonging to a new candidate phylum sibling to Cyanobacteria. *eLife* 2, (2013).
18. Miettinen, A. Mycoplasma hominis in patients with pelvic inflammatory disease. *Isr. J. Med. Sci.* 23, 713-716 (1987).
19. Pereyre, S. *et al.* Life on Arginine for Mycoplasma hominis: Clues from Its Minimal Genome and Comparison with Other Human Urogenital Mycoplasmas. *PLoS Genet.* 5, (2009).
20. Kashyap, S. & Sarkar, M. Mycoplasma pneumonia: Clinical features and management. *Lung India Off. Organ Indian Chest Soc.* 27, 75-85 (2010).
21. Parker, B. J., Wearsch, P. A., Veloo, A. C. M. & Rodriguez-Palacios, A. The Genus Alistipes: Gut Bacteria With Emerging Implications to Inflammation, Cancer, and Mental Health. *Front. Immunol.* 11, (2020).
22. Methanosphaera stadtmanae - microbewiki.  
[https://microbewiki.kenyon.edu/index.php/Methanosphaera\\_stadtmanae](https://microbewiki.kenyon.edu/index.php/Methanosphaera_stadtmanae).
23. Campylobacter hominis (strain ATCC BAA-381 / LMG 19568 / NCTC 13146 / CH001A).  
<https://www.uniprot.org/proteomes/UP000002407>.
24. Alsmark, C. M. *et al.* The louse-borne human pathogen Bartonella quintana is a genomic derivative of the zoonotic agent Bartonella henselae. *Proc. Natl. Acad. Sci. U. S. A.* 101, 9716-9721 (2004).
25. Noronha, A. *et al.* The Virtual Metabolic Human database: integrating human and gut microbiome metabolism with nutrition and disease. *Nucleic Acids Res.* 47, D614-D624 (2019).
26. Crapoulet, N., Barbry, P., Raoult, D. & Renesto, P. Global Transcriptome Analysis of Tropheryma whipplei in Response to Temperature Stresses. *J. Bacteriol.* 188, 5228-5239 (2006).
27. Dillon, S. *et al.* Gut Dendritic Cell Activation Links an Altered Colonic Microbiome to Mucosal and Systemic T Cell Activation in Untreated HIV-1 infection. *Mucosal Immunol.* 9, 24-37 (2016).
28. Dhawan, B. *et al.* Ureaplasma serovars & their antimicrobial susceptibility in patients of infertility & genital tract infections. *Indian J. Med. Res.* 136, 991-996 (2012).

29. Wetsch, N. M. *et al.* Campylobacter curvus-Associated Hepatic Abscesses: a Case Report. *J. Clin. Microbiol.* 44, 1909–1911 (2006).
30. Ureaplasma parvum - microbewiki.  
[https://microbewiki.kenyon.edu/index.php/Ureaplasma\\_\\_parvum](https://microbewiki.kenyon.edu/index.php/Ureaplasma__parvum).