

# Explanations for Similarity Query output files

## Upload.txt

Recapitulates the sequences used for query by users. In the example below, sequences are truncated for space economy. For ecological studies, it is important to use 16S rRNA gene sequences as complete as possible to cover all possible variable regions contained in different SRA-derived sequencing projects.

### Example

```
>HE978271
ATGCAAGTCGAGCGGATGAAGGGAGCTTGTCTGGATTAGCGGGGACGGTGAGTAATGCCATAGAATCTGCCTGGTAG ...
>U96927
ATCCAAGTCGAACGGCAGCACGGTGCTGCACCTGGTGGCGAGTGGCAACGGTGAGTAATACATCGAACATGTCTGT ...
>X71837
ATGCAAGTCGAACGAGGGCATTCTTCGGGGATTTCTAGTGGCGACGGTGAGTAACGCGTGGGAATCTGCCTGATAGT ...
>Z29619
ATGCAAGTCGAACGAGGGTTCTTCGGGGGCTTAGTGGCGCACGGTGAGTAACCGTGGAACCTGCCTTCGGTT ...
>Z32635
ATGCAAGTCGAACGGCAGCATACTAGCTGCTAGGTTGATGGCGAGTGGCGAACGGTGAGTAACCGTAGGAATATGCCTT ...
```

## Seq\_id\_map.tab

The reference number assigned to each sequence in the query fasta file. All sequence files and results follow this conversion.

### Example

```
1 HE978271
2 U96927
3 X71837
4 Z29619
5 Z32635
```

## Counts\_overview.tab

The number of hits (sequences) per sample and per query over all selected samples at the different similarity thresholds.

### Example

#SampleID	Size	Description	1.99	2.99	1.97	2.97
SRR536792	476648	human gut metagenome	2452	0	2553	0
SRR389090	359248	human gut metagenome	340	0	342	6
SRR389091	343996	human gut metagenome	61	0	62	19
SRR514790	162501	human gut metagenome	6	0	6	0
SRR505779	3139	human gut metagenome	5	0	5	0
SRR639158	22880	human gut metagenome	3	0	3	0

This example shows the number of query-like sequences in several samples. Size indicates the number of total sequences in the corresponding SRA sample (can be used to calculate relative abundances). Description refers to the taxonomy of the sample in SRA.

It can be seen that sequence 1 (1.99, 1.97) can be found in the selected samples, but not sequence 2 (2.99, 2.97). Moreover, sequences corresponding to query 1 are almost exclusively matches at the strain level, as the number of hits did not increase after relaxing the similarity threshold from 99% to 97% (1.99 vs. 1.97).

## **Selected\_db\_list.txt**

The list of samples that were set for query. The easiest way for third parties to repeat the query is to send this list and then import it in IMNGS.

### **Example**

```
ERR174134
ERR174136
ERR174137
ERR174138
ERR174139
ERR174140
ERR174141
```

## **#.hits.tab**

This file contains the UBLAST output of the hits for each query in tabular format (e.g., 1.hits.fasta). It can be used to trace back OTUs matching the query in all SRA samples analyzed.

### **Example**

Query_name	Target_seq	Identity	Al_length	mismatches	gaps	Query_start	Query_end	Target_start	Target_end	E-value	Bi
HE978271	SRR514792.174404.2;size=1;tax=...	98.5	194	3	0	281	474	21	214	1.1e-94	342.8
HE978271	SRR389090.3588.3;size=340;tax=...	100.0	435	0	0	39	473	435	1	2.4e-233	804.4
HE978271	SRR389090.290961.3;size=2;tax=...	97.5	401	5	5	75	473	398	1	1.6e-175	612.4
HE978271	SRR578501.5520.4;size=1;tax=...	100.0	326	0	0	523	848	326	1	9.3e-175	603.1
HE978271	SRR578403.2695.4;size=1;tax=...	97.5	244	6	0	605	848	244	1	8.1e-120	418.5
HE978271	SRR639307.16357.2;size=3;tax=...	98.8	329	3	1	41	369	6	333	4.9e-164	571.7
HE978271	SRR389091.8838.3;size=61;tax=...	100.0	433	0	0	41	473	433	1	3.3e-232	800.7

## **#.seqs.fasta**

This file contains all fasta sequences of the hits for each query (e.g., 1.seqs.fasta). The sequence description line is semicolon delimited (;) and contains the ID of the centroid sequence representing the OTU cluster, the number of sequences that clustered under that OTU, and the taxonomy assigned to the OTU by RDP classifier.

### **Example**

```
>SRR360670.857.2;size=1;tax=Bacteria;Proteobacteria;Betaproteobacteria;Burkholderiales;Burkholderiaceae;Burkholderia;
TCTCCATCCATCAGAGTTGATCATGGCTCAGATTGAACGCTGGCGCATGCCCTACACATGCAAGTCGAACGGCAGCAC ...
>SRR360615.436.2;size=1;tax=Bacteria;Proteobacteria;Betaproteobacteria;Burkholderiales;Burkholderiaceae;Burkholderia;
TCTAATGCGCTCAGAGTTGATCTGGCTCAGATTGAACGCTGGCGCATGCCCTACACATGCAAGTCGAACGGCAGCAC ...
>SRR360638.1757.2;size=1;tax=Bacteria;Proteobacteria;Betaproteobacteria;Burkholderiales;Burkholderiaceae;Burkholderia;
TCTTGGTTCGTCAGAGTTGATCTGGCTCAGATTGAACGCTGGCGCATGCCCTACACATGCAAGTCGAACGGCAGCAC ...
>SRR360619.1405.2;size=3;tax=Bacteria;Proteobacteria;Betaproteobacteria;Burkholderiales;Burkholderiaceae;Burkholderia;
TCGTTCTTGTCAAGAGTTGATCATGGCTCAGATGCAACGCTGGCGCATGCCCTACACATGCAAGTCGAACGGCAGCAC ...
```

## **Report.#.tab**

The number of samples that were positive for the presence of query-like sequences for each sample category.

### **1. Report.0.tab**

An SRA-derived sample is considered positive if the query-like sequences sum up to more than 0% of the total number of sequences in that sample (i.e. any abundance).

### **2. Report.0.1.tab**

An SRA-derived sample is considered positive if the query-like sequences sum up to more than 0.1% of the total number of sequences in that sample (i.e. excluding rare abundances).

### **3. Report.1.tab**

An SRA-derived sample is considered positive if the query-like sequences sum up to more than 1% of the total number of sequences in that sample (i.e. including only dominant OTUs).

#### **Example**

Environment_source	Samples	number	1.99	2.99	1.97	2.97
human gut metagenome	833	9	0	14	0	
soil metagenome	135	0	19	0	48	

In this example, 968 samples were queried covering two environmental types (human gut and soil) as shown in the first column. The second column shows the exact number of samples in each category followed by the number of samples positive for the presence of query-like sequences. This example is supportive of bacterium 1 and 2 to be specific for human gut and soil, respectively.