**Figure S6. Selecting the most optimal consensus-path.**

The figures below illustrate and explain how after calculating an individual consensus-path for each genome (see Fig. 6) a more general consensus-path was formulated based on the paths of individual genomes. We show the paths based on the eight moderate- and four more extreme GC% genomes.

**Figure S6A:** We were able to calculate the impact of a certain AGE (combination) using *formula 4* (materials and methods) by taking into account: (1) sensitivity (i.e. the coverage, in other words, the percentage of ORFs predicted of the original known annotation), (2) specificity (i.e. the percentage of correctly predicted ORFs in the fraction of ORFs covered by the sensitivity), and (3) the rank or order in the consensus-path for a genome (as seen in Fig. 6). Dark gray blocks: a certain AGE (combination) was not part of the consensus-path for that genome (see Fig. 3 for these criteria). The average impact shown on the right is calculated by averaging the impact of all eight moderate GC% genomes (see *formula 4*). The table was sorted on the average impact. As a result, the most optimal general consensus-path could be formulated, as shown on the left. This path consists of BASys-ISGA-RAST-xBASE in the first round, ISGA-RAST-xBASE in the second, BASys-ISGA-RAST in the third, ISGA-RAST in the fourth and BASys-RAST-xBASE in the fifth round of annotation prediction comparison. Through this representation, the resemblance of each individual genome to the general consensus-path can also be visually assessed.

**Figure S6B:** A simplified representation of the consensus path across the different genomes. Each iteration (first column) by our consensus-path methodology (as described in Fig. 3) results in an AGE (combination) providing the highest specificity, which is listed for every iteration for each genomes. The consensus-path shown on the right is derived from the calculations shown in Figure S6A. The more
### Moderate GC% Species

<table>
<thead>
<tr>
<th>Path</th>
<th>B. subtilis B168</th>
<th>E. coli K12</th>
<th>L. lactis KF147</th>
<th>L. plantarum WCFS1</th>
<th>Average Impact</th>
</tr>
</thead>
<tbody>
<tr>
<td>ABCD</td>
<td>1 0.427 0.094 0.605</td>
<td>2 0.501 0.503 0.412</td>
<td>3 0.134 0.227 0.012</td>
<td>4 0.231 0.193 0.039</td>
<td>0.449</td>
</tr>
<tr>
<td>BCD</td>
<td>2 0.746 0.203 0.035</td>
<td>3 0.234 0.227 0.012</td>
<td>4 0.134 0.227 0.012</td>
<td>5 0.436 0.328 0.028</td>
<td>0.264</td>
</tr>
<tr>
<td>ABC</td>
<td>4 0.180 0.294 0.053</td>
<td>4 0.504 0.139 0.021</td>
<td>5 0.205 0.257 0.020</td>
<td>5 0.189 0.250 0.433</td>
<td>0.067</td>
</tr>
<tr>
<td>ACD</td>
<td>4 0.180 0.294 0.053</td>
<td>4 0.504 0.139 0.021</td>
<td>5 0.205 0.257 0.020</td>
<td>5 0.189 0.250 0.433</td>
<td>0.067</td>
</tr>
<tr>
<td>AD</td>
<td>9 0.004 0.193 0.000</td>
<td>9 0.004 0.193 0.000</td>
<td>9 0.004 0.193 0.000</td>
<td>9 0.004 0.193 0.000</td>
<td>0.004</td>
</tr>
<tr>
<td>ABD</td>
<td>6 0.030 0.572 0.002</td>
<td>6 0.030 0.572 0.002</td>
<td>6 0.030 0.572 0.002</td>
<td>6 0.030 0.572 0.002</td>
<td>0.003</td>
</tr>
<tr>
<td>AB</td>
<td>5 0.014 0.400 0.001</td>
<td>5 0.014 0.400 0.001</td>
<td>5 0.014 0.400 0.001</td>
<td>5 0.014 0.400 0.001</td>
<td>0.001</td>
</tr>
<tr>
<td>CD</td>
<td>7 0.000 0.634 0.000</td>
<td>7 0.000 0.634 0.000</td>
<td>7 0.000 0.634 0.000</td>
<td>7 0.000 0.634 0.000</td>
<td>0.000</td>
</tr>
<tr>
<td>BD</td>
<td>8 0.036 0.621 0.020</td>
<td>8 0.036 0.621 0.020</td>
<td>8 0.036 0.621 0.020</td>
<td>8 0.036 0.621 0.020</td>
<td>0.000</td>
</tr>
<tr>
<td>B</td>
<td>12 0.009 0.981 0.010</td>
<td>12 0.009 0.981 0.010</td>
<td>12 0.009 0.981 0.010</td>
<td>12 0.009 0.981 0.010</td>
<td>0.000</td>
</tr>
<tr>
<td>C</td>
<td>8 0.009 0.944 0.000</td>
<td>8 0.009 0.944 0.000</td>
<td>8 0.009 0.944 0.000</td>
<td>8 0.009 0.944 0.000</td>
<td>0.000</td>
</tr>
<tr>
<td>A</td>
<td>10 0.004 0.997 0.000</td>
<td>10 0.004 0.997 0.000</td>
<td>10 0.004 0.997 0.000</td>
<td>10 0.004 0.997 0.000</td>
<td>0.000</td>
</tr>
</tbody>
</table>

### Extreme GC% Species

<table>
<thead>
<tr>
<th>Path</th>
<th>M. tuberculosis H37r</th>
<th>M. mobile 163K</th>
<th>S. coelicolor A3</th>
<th>P. putida KT2440</th>
<th>Average Impact</th>
</tr>
</thead>
<tbody>
<tr>
<td>ABCD</td>
<td>1 0.348 0.094 0.315</td>
<td>1 0.239 0.094 0.315</td>
<td>1 0.205 0.094 0.315</td>
<td>1 0.205 0.094 0.315</td>
<td>0.449</td>
</tr>
<tr>
<td>BCD</td>
<td>3 0.254 0.370 0.026</td>
<td>2 0.121 0.250 0.026</td>
<td>2 0.121 0.250 0.026</td>
<td>2 0.121 0.250 0.026</td>
<td>0.258</td>
</tr>
<tr>
<td>ABC</td>
<td>2 0.030 0.193 0.012</td>
<td>2 0.030 0.193 0.012</td>
<td>2 0.030 0.193 0.012</td>
<td>2 0.030 0.193 0.012</td>
<td>0.030</td>
</tr>
<tr>
<td>ACD</td>
<td>4 0.470 0.416 0.067</td>
<td>4 0.212 0.121 0.006</td>
<td>4 0.212 0.121 0.006</td>
<td>4 0.212 0.121 0.006</td>
<td>0.067</td>
</tr>
<tr>
<td>AD</td>
<td>5 0.014 0.944 0.000</td>
<td>5 0.014 0.944 0.000</td>
<td>5 0.014 0.944 0.000</td>
<td>5 0.014 0.944 0.000</td>
<td>0.000</td>
</tr>
<tr>
<td>ABD</td>
<td>7 0.000 0.634 0.000</td>
<td>7 0.000 0.634 0.000</td>
<td>7 0.000 0.634 0.000</td>
<td>7 0.000 0.634 0.000</td>
<td>0.000</td>
</tr>
<tr>
<td>AB</td>
<td>14 0.007 0.981 0.010</td>
<td>14 0.007 0.981 0.010</td>
<td>14 0.007 0.981 0.010</td>
<td>14 0.007 0.981 0.010</td>
<td>0.000</td>
</tr>
<tr>
<td>CD</td>
<td>12 0.004 0.997 0.000</td>
<td>12 0.004 0.997 0.000</td>
<td>12 0.004 0.997 0.000</td>
<td>12 0.004 0.997 0.000</td>
<td>0.000</td>
</tr>
<tr>
<td>BD</td>
<td>10 0.009 0.944 0.000</td>
<td>10 0.009 0.944 0.000</td>
<td>10 0.009 0.944 0.000</td>
<td>10 0.009 0.944 0.000</td>
<td>0.000</td>
</tr>
<tr>
<td>B</td>
<td>13 0.012 0.981 0.000</td>
<td>13 0.012 0.981 0.000</td>
<td>13 0.012 0.981 0.000</td>
<td>13 0.012 0.981 0.000</td>
<td>0.000</td>
</tr>
<tr>
<td>ABD</td>
<td>12 0.004 0.997 0.000</td>
<td>12 0.004 0.997 0.000</td>
<td>12 0.004 0.997 0.000</td>
<td>12 0.004 0.997 0.000</td>
<td>0.000</td>
</tr>
</tbody>
</table>

Figure S6A
Figure S6B