

1 ***In planta* colonization and role of T6SS in two rice *Kosakonia* endophytes**

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21 **ABSTRACT**

22 Endophytes live inside plants and are often beneficial. *Kosakonia* is a novel
23 bacterial genus including many diazotrophic plant-associated isolates. Plant-
24 bacteria studies on two rice endophytic *Kosakonia* beneficial strains were
25 performed including comparative genomics, secretome profiling, *in-planta* tests
26 and a field release trial. The strains are efficient rhizoplane and root
27 endosphere colonizers and localized in the root cortex. Secretomics revealed
28 144 putative secreted proteins including type VI secretory system (T6SS)
29 proteins. A *Kosakonia* T6SS genomic knock-out mutant showed a significant
30 decrease in rhizoplane and endosphere colonization ability. A field trial using
31 rice seeds inoculated with *Kosakonia* sp. showed no effect on plant growth
32 promotion upon nitrogen stress and microbiome studies revealed that
33 *Kosakonia* was significantly more present in the inoculated rice. Comparative
34 genomics evidenced that several protein domains were enriched in plant-
35 associated *Kosakonia*. This study highlights that *Kosakonia* is an important
36 recently classified genus involved in plant-bacteria interaction.

37 Rice is the most important food crop in the developing world, being a
38 staple food for over two billion people in Asia and for many millions in Africa and
39 Latin America (Khush 2003; Zeigler and Barclay 2008). The challenge in the
40 future will be to increase rice yields for a growing world population and to
41 decrease the use of chemical pesticides and fertilizers for more sustainable
42 approaches (Schütz et al. 2018; Mano and Morisaki 2008). The use of
43 microbially-based biopesticides and biofertilizers is currently believed to be a
44 promising way to render agriculture more sustainable by reducing the chemical
45 input (Berg 2009; Gupta and Dikshit 2010; Mahanty et al. 2017; Schütz et al.
46 2018).

47 Plant-associated microbiota constitutes the plant microbiome playing a
48 fundamental role in plant growth promotion (PGP) and health (Okubo et al.
49 2014; Turner et al. 2013; Schlaeppi and Bulgarelli 2015). PGP activities by
50 plant-associated microbes include induction of plant immunity, acquisition of
51 nutrients and resistance to biotic or abiotic stresses (Compant et al. 2010;
52 Lugtenberg and Kamilova 2009; Glick 2014). The plant microbiome represents
53 many diverse microorganisms that interact and colonize different plant-
54 associated niches (Müller et al. 2016; van der Heijden and Hartmann 2016).
55 One of these compartments is the rhizoplane, which consists of the root-
56 surrounding soil being influenced by root exudates and has a high diversity and
57 distribution of microbial life (Berg et al. 2005; Hartmann et al. 2008; Lundberg et
58 al. 2012). Some microbiome members do not only colonize the rhizoplane but
59 also thrive as endophytes inside plant tissues (Berg et al. 2014; Reinhold-Hurek
60 and Hurek 2011). Endophytes mostly enter via the roots and have evolved an
61 intimate relationship with the plant host many of which do not elicit a plant

62 immune response and some display PGP properties (Glick 2014; Hayat et al.
63 2010; Reinhold-Hurek and Hurek 1998; Sessitsch et al. 2012; Garrido-Oter et
64 al. 2018). Endophytes constitute therefore an important class of beneficial
65 bacteria now considered to be a potentially important group that can be used as
66 microbial inoculants for a more sustainable agriculture. However, more
67 information is needed on the endophytic life style and mechanisms of plant
68 entry and colonization.

69 General features of the rice microbiome, and plant microbiomes in
70 general, include less species richness in the plant endosphere than on the
71 rhizoplane (surface of the root) and in the rhizoplane (Edwards et al. 2015;
72 Lundberg et al. 2012; Bulgarelli et al. 2012). Microbial communities of the rice
73 rhizoplane and root endosphere stabilize after seven to eight weeks from
74 germination due to the plant life cycle (Edwards et al. 2017). The rice
75 endophytic bacteriome has a prevalence of Proteobacteria representing more
76 than 50% of the bacterial community with Gammaproteobacteria being the most
77 abundant class. Many rice endophytes possess nitrogen fixation genes as well
78 as genes related to nitrification and denitrification processes which suggest they
79 are involved in the entire nitrogen cycle (Sessitsch et al. 2012). Examples of
80 PGP rice endophytes include *Pantoea agglomerans* YS19 (Feng et al. 2006;
81 Yang et al. 1999; Jiang et al. 2015) and *Pseudomonas stutzeri* A15, a
82 rhizospheric and endospheric diazotrophic root colonizer (Pham et al. 2017).
83 Other rice endophytes with potential use as nitrogen biofertilizers include
84 *Gluconacetobacter diazotrophicus* LMG7603, *Herbaspirillum seropedicae*
85 LMG6513, *Azospirillum lipoferum* 4B (LMG4348), and *Burkholderia*
86 *vietnamiensis* LMG10929 (Govindarajan et al. 2008; Trần Van et al. 1996;

87 Baldani et al. 1986; Rouws et al. 2010).

88 We previously reported the isolation and characterization of bacterial
89 endophytes from rice grown in Italy that resulted in a collection of over 1300
90 putative isolates (Bertani et al. 2016). Several *in vitro* and *in planta* selection
91 steps resulted in a smaller set of putative endophytes, which displayed efficient
92 *in planta* colonization levels as well as having PGP traits. Among these, were
93 two strains that belong to the recently described *Kosakonia* genus (Brady et al.
94 2013; Alnajar and Gupta 2017), which consists mostly of plant-associated
95 diazotrophs (Kämpfer et al. 2016; Y. Li et al. 2017). Some *Kosakonia* strains,
96 as for example *K. radicincitans* DSM 16656, are promiscuous endophytes and
97 promote plant growth in different plants including wheat, maize, tomato, pea,
98 and cruciferous vegetables (Berger et al. 2013; Höflich and Ruppel 1994;
99 Schreiner et al. 2009). In recent years, members of this genus have gained
100 attention and several genome sequences have been reported (Chen et al.
101 2014; Shinjo et al. 2016; Kämpfer et al. 2016; Bergottini et al. 2015; Li et al.
102 2017; Mohd Suhaimi et al. 2014; Becker et al. 2018).

103 In this study, we characterized two *Kosakonia* strains that we previously
104 isolated as PGP endophytes of rice (Bertani et al. 2016). In order to begin to
105 study features that make them efficient endophytic colonizers, we performed
106 plant colonization, genomic and protein secretome studies. We also report a
107 rice field release study of one *Kosakonia* strain and its effect on plant yield and
108 on the rice root endophytic microbiome.

109

110 RESULTS

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112 Genome sequence and analysis of the two *Kosakonia* strains

113 It was of interest to determine the genome sequences of the two rice
114 beneficial *Kosakonia* sp. KO348 and KO774 endophytic strains which we
115 previously identified (Bertani et al. 2016). We previously reported the genome
116 sequence of strain KO348 (Meng et al. 2015) and in this study we re-
117 sequenced it; the assembly yielded a higher quality genome compared to the
118 previous version. The new sequence gave 26 scaffolds of average size
119 192.7Kbp vs. 56 scaffolds of average scaffold size 89.3 Kbp in the new vs. the
120 old genome version, respectively. In the case of strain KO774, we report here
121 for the first time the genome sequence which was performed on an Illumina
122 MiSeq platform (2x300 bp) by *de novo* assembly using Velvet 1.2.09. The
123 assembly using IMG/M (*Kosakonia* sp. KO774) yielded 12 scaffolds giving a
124 total of 4,875,574 bp, including 4,530 putative protein coding genes and 153
125 RNA genes; the assembly also revealed a putative plasmid (74 Kbp). The
126 contig containing the putative plasmid included loci for plasmid replication,
127 conjugation, segregation genes, few toxin-antitoxin systems and a large
128 cellobiose phosphorylase gene (**Supplementary Figure S1**).

129 The bacterial genomes, KO348 and KO774, had 3,853 orthologous
130 genes identified as bidirectional best hits (BBH). Namely, 82.5% of the genes in
131 each genome display at least 70% sequence identity over at least 70% of the
132 length of the shorter sequence in each BBH pair. The genomic average
133 nucleotide identity (gANI) between the BBH pairs is 83.69% (Varghese NJ NAR
134 2015). Both strains share some PGP related genes involved in siderophore

135 production (enterobactins), phosphate solubilization (phytase), flagellar motility,
136 plant tissue degrading enzymes (cellulase) and the *nif* gene cluster for nitrogen
137 fixation. The genome size of KO774 is approximately 100 kb smaller compared
138 to the one of KO348. Interestingly the KO348 strain has extra phage-related
139 proteins while KO774 has a higher copy number of flagellin-related proteins
140 **(Supplementary Table S1).**

141 **Comparative genomics in the *Kosakonia* genus**

142 A phylogenetic tree was constructed comprising all the *Kosakonia*
143 complete genomes publicly available in the Integrate Microbial Genomes &
144 Microbiomes database (IMG/M) of the Joint Genome Institute (Nordberg et al.
145 2014), including strains KO348 and KO774 (n=15), the analysis was performed
146 having *Escherichia coli* K12 MG1655 as an outgroup. The phylogenetic
147 analysis showed that the strain *Kosakonia* sp. KO348 is most closely related to
148 the strain *K. sacchari* CGMCC.1.12101, while *Kosakonia* sp. KO774 is an
149 outgroup distantly related to the phytophyla group **(Figure 1)**. The hierarchical
150 clustering in the phylogenetic tree showed a clear separation between two
151 groups, the *Kosakonia* strains isolated from plants (phytophyla group) and the
152 *Kosakonia* isolated from human/animal samples **(Figure 1)**. Interestingly, when
153 analyzing the enriched protein domains between the genomes of the
154 phytophyla group and the human/animal associated group, we found that in the
155 *Kosakonia* phytophyla group, nitrogen fixation, cobalamin biosynthesis,
156 ethanolamine and phosphonate metabolism domains were enriched, while in
157 the human/animal associated strains the domains for host adaptation and
158 virulence such as immunoglobulin A1 protease and Haem utilization were
159 enriched **(Figure 1) (Supplementary Table S2).**

160 **Plant colonization assays on rhizoplane and endosphere**

161 To determine the colonization ability of the rice rhizoplane and
162 endosphere of both *Kosakonia* strains and also determine if they out-competed
163 each other, we conducted colonization studies. Single-inoculation and co-
164 inoculation using both *Kosakonia* strains were performed (**Figure 2**). Both
165 *Kosakonia* sp. KO348 and KO774 strains were able to colonize the rice
166 rhizoplane and endosphere efficiently and at very similar levels when inoculated
167 independently, no statistically significant differences were found between the
168 colonization abilities of both strains (**Figure 2**). When both strains were co-
169 inoculated on rice, both were able to equally colonize the two plant
170 compartments, no statistically significant differences were found between the
171 *Kosakonia* strains, without out-competing each other and likely forming stable
172 and mixed communities.

173 Bacterial CFU of strain KO348 attached to the surface of the seedling
174 root after one hour of inoculation was 1.2×10^7 CFU/g of root and the number of
175 bacterial cells recovered from the root endosphere after 30 dpi was on average
176 1.8×10^4 CFU/g of root (**Figure 3**) which indicates that KO348 is a good
177 endophytic root colonizer. We also determined the root endosphere colonization
178 at three other time points, 5, 10 and 50 dpi showing 1.8×10^4 CFU/g, 8.3×10^4
179 CFU/g and 1.1×10^4 CFU/g, of root respectively. These latter experiments are
180 the results at each time point of 5 plants handled and processed independently.
181 It was concluded that *Kosakonia* sp. KO348 is a good and stable rice root
182 endosphere colonizer under the tested conditions.

183 **Visualization by confocal microscopy in the rice rhizoplane and** 184 **endosphere of strain *Kosakonia* sp. KO348**

185 In order to unequivocally determine the internal plant colonization by
186 *Kosakonia* sp. KO348, confocal microscopy localisation was performed.
187 Location of the strain on the rhizoplane (including root hair zone, secondary root
188 emergence and grain surface) and inside the root endosphere (transversal
189 sections of the root) of rice roots were determined within rice plants inoculated
190 with *Kosakonia* strain KO348(pBBRgfp) at different time-points of 5, 10, 30 and
191 50 dpi.

192 Strain KO348 presented higher densities in the rhizoplane than in the
193 root endosphere at the four time points analyzed (**Figures 4-5**). At 5 and 10 dpi
194 it was mainly found at the root hair zone, secondary root emergence, and on
195 grain surface, presenting at 10 dpi clear bacterial aggregation, especially at the
196 root hair zone (**Figure 4**). At 30 dpi we observed also high densities of bacterial
197 aggregation on the rhizoplane that was less evident at 50 dpi (**Figure 5**).
198 *Kosakonia* sp. KO348 was also observed as endophyte in the transversal root
199 sections, up to the arenchyma, in all the four time points determining that the
200 strain was able to colonize the root endosphere (**Figure 4-5**). The un-inoculated
201 plants did not present fluorescence at any time point analyzed neither in the
202 rhizoplane nor in the root endosphere (**Figure 5 and Supplementary Figure**
203 **S2**). It was therefore concluded that KO348 was able to attach and colonize the
204 rhizoplane of rice plants and as endophyte forming communities observable
205 until 50 dpi.

206 **Secretome profile determination of *Kosakonia* KO348**

207 It was of interest to determine which proteins *Kosakonia* sp. KO348
208 produced and secreted in the extracellular medium since these could play a role
209 in the endophytic colonization process. A total of 144 putative secreted proteins

210 were detected when strain KO348 was grown in a minimal plant-mimicking
211 medium (**Table 1, Supplementary Tables S3 and S4**). Among the ones found
212 were ten flagella-related proteins (FliD, FliK, FlgK, FlgL, FlgE, FlgJ, FlhA and 3
213 different proteins of flagellin) and five proteins belonging to the type VI secretory
214 system (Hcp, a protein with a FHA domain and three different proteins of VgrG)
215 (**Table 1**).

216 **Role of Type VI secretory system in rhizoplane and root endosphere** 217 **colonization in *Kosakonia* sp. KO348**

218 The secretome profile of strain KO348 determined in this study
219 demonstrated that several proteins of the Type VI secretion system were
220 present (see above). A database search for type VI secretion systems (T6SS)
221 domains among the *Kosakonia* available genomes at the IMG/M showed that
222 the T6SS was present among all genomes, including the two strains of this
223 study (**Supplementary Table S5**). We searched for T6SS known annotations,
224 found two loci and identified in one of them a complete gene cluster of T6SS of
225 strain KO348 (**Supplementary Figure S3**). We further searched for putative
226 T6SS effectors by aligning the 70 proteins found in the T6SS loci
227 (**Supplementary Table S6**) against the proteins identified in the secretome and
228 it was then evidenced that three proteins matched; two were T6SS tip proteins
229 VrgG (gi|780193605 and gi|780193664) and one secretion system-associated
230 FHA domain protein TagH (gi|780193691).

231 Since T6SS component proteins and candidate effectors were expressed
232 in plant-mimicking medium, it was of interest to determine the possible role of
233 T6SS in rhizoplane and root endosphere colonization. A knock-out mutant of
234 the T6SS *hcp* gene (responsible for the formation of the needle-like structure for

235 the passage of the effectors) called KO348hcp and its complement
236 KO348hcp(pBBRhcp) (carrying a plasmid with the complete *hcp* gene) were
237 generated. Growth curves of the three strains, KO348, KO348hcp and
238 KO348hcp(pBBRhcp) were performed in triplicate in LB media; the three strains
239 had comparable growing rates/curves (**Supplementary Figure S4**). In order to
240 assess rice colonization ability of the *hcp* knock-out mutant of *Kosakonia* sp.
241 KO348, the four following inoculation groups were performed; the KO348 wild-
242 type (WT), the KO348hcp mutant, the complemented *hcp* mutant
243 KO348hcp(pBBRhcp) and the KO348 WT and mutant KO348hcp together in a
244 competition experiment. For all the four groups the same amount of total
245 bacteria 1.3×10^7 CFU/ml was used for plant inoculation, in the case of the
246 competition experiment we used 0.65×10^7 CFU/ml of each strain.

247 *In planta* experiments determined that at 14 dpi the colonization of the
248 rhizoplane by WT strain KO348 was near 70-fold higher than the colonization of
249 the *hcp* mutant KO348hcp (2.3×10^6 CFU/g of root vs. 3.3×10^4 CFU/g of root
250 respectively, **Figure 6**). A significant difference was also observed between the
251 WT KO348 and the *hcp* mutant when they were co-inoculated (7.2×10^5 CFU/g
252 of root vs. 3.6×10^4 CFU/g of root respectively). Complementing the mutant with
253 the *hcp* gene harbored in a plasmid, resulted in restoration of its ability to
254 colonize the rhizoplane (2.3×10^5 CFU/g) (**Figure 6a**). In this complementation
255 experiment the percentage of bacterial strains that retained the plasmid was
256 78% indicating a low incidence of plasmid loss. Rhizoplane colonization was not
257 affected by the presence of the empty plasmid vector in the mutant strain
258 KO348hcp(pBBRMCS-1), the plasmid retention was 69% (**Supplementary**
259 **Figure S5**).

260 We also performed studies of root endosphere colonization and results
261 showed a significant difference between the colonization ability of WT and the
262 *hcp* mutant in plants inoculated independently (6.4×10^4 CFU/g of root vs.
263 2.2×10^3 CFU/g of root respectively). This significant difference between WT and
264 mutant was maintained when plants were co-inoculated (2.7×10^4 CFU/g of root
265 vs. 1.4×10^4 CFU/g of root respectively) (**Figure 6b**). However, unlike in the
266 rhizoplane experiment, the complemented mutant did not result in the
267 restoration of endophytic colonization to WT levels (4.9×10^2 CFU/g). The low
268 plasmid retention (22%) at the root endosphere colonization level was also
269 observed in the mutant strain harboring the empty plasmid vector
270 KO348*hcp*(pBBRMCS-1). The *hcp* mutant strain not carrying the plasmid was
271 significantly more recovered in the root endosphere than the mutant harboring
272 the empty vector KO348*hcp*(pBBRMCS-1), indicating plasmid loss
273 (**Supplementary Figure S5**). In this case the percentage of bacterial cells
274 which retained the plasmid harboring the *hcp* gene was only 23%; this is the
275 most likely the reason for the lack of complementation. In summary, these
276 results suggest a significant role for T6SS in the rhizoplane colonization and to
277 a much lesser extent in the root endosphere colonization by *Kosakonia* sp.
278 KO348.

279 **Field rice inoculation with *Kosakonia* sp. KO774**

280 As it was previously determined that the diazotrophic *Kosakonia* strains
281 studied here displayed plant growth promoting properties (Bertani et al. 2016), it
282 was of interest to perform a field rice experiment in order to assess if it could
283 compensate a reduction in nitrogen fertilization. Between May and October
284 2016 in Valencia, Spain, we performed a rice field trial with diazotrophic

285 *Kosakonia* sp. KO774 with the aim of testing if rice seeds inoculated with the
286 bacterial strain can compensate a 50% reduction in nitrogen fertilization. A total
287 of 16 growth plots were grown which were divided in four groups, (i) eight plots
288 receiving 100% nitrogen/urea fertilization, four planted with seeds inoculated
289 with strain KO774 and the other four plots with the seeds not inoculated and (ii)
290 eight plots receiving only 50% of urea/nitrogen fertilization, four of these planted
291 with seeds inoculated with strain KO774 and the other four plots with the seeds
292 not inoculated (**Supplementary Figure S6**).

293 All the plots were harvested 100 days post sowing and different growth
294 parameters such as germination/plot, weight of 1,000 grains/plot and 25
295 panicles/plot and yield (kg/ha) were then assessed/measured (**Table 2**). No
296 statistically significant differences in any of the measured parameters were
297 found between the inoculated and uninoculated plots. This indicated that
298 inoculation with *Kosakonia* did not result in any plant growth promotion and/or
299 nitrogen biofertilization under the conditions tested (**Table 2**).

300 **Rice microbiome analysis of the rice field trial using seeds inoculated with** 301 ***Kosakonia* sp. KO774**

302 Following the rice seed inoculation with *Kosakonia* sp. KO774 in the field
303 trial (see above), it was also of interest to determine the colonization of strain
304 KO774 and the effect on the total endospheric microbial community of rice
305 plants. We determined the root endomicrobiome at three different time points
306 (30, 60 and 90 post-sowing) in plants which have been fertilized with 50% of the
307 recommended amount of nitrogen and have been inoculated with KO774 as
308 well as with ones that were not.

309 In the endomicrobiome we first determined the presence of bacterial

310 sequences with 100% identity to the 16S rDNA gene fragment of *Kosakonia* sp.
311 KO774. This sequence was present in all three samples in the *Kosakonia*-seed
312 inoculated group at 30 dpi, however at 60 dpi we only observed it at much lower
313 levels in one of the three samples. At 90 dpi this 16S rRNA gene sequence of
314 *Kosakonia* sp. KO774 was not detected (**Supplementary Figure S7**). In the
315 group which was not seed inoculated with strain KO774, only in one sample at
316 90 dpi and low abundance was observed the 100% identity *Kosakonia* DNA
317 sequence (**Supplementary Figure S7**).

318 A total root endomicrobiome analysis was also performed analyzing the
319 main bacterial genera (>1% abundance) at 30 dpi, when *Kosakonia* sp. KO774
320 was still present. It was observed that the hierarchical clustering positioned
321 closer the samples within each treatment group, indicating that all seed-
322 inoculated samples were clustered together and all the un-inoculated samples
323 were closer among them (**Figure 7**). However, when observing the Z-scores
324 based on distribution and relative abundance of each genus, we observed a
325 significant difference between the two group-sets (inoculated vs. un-inoculated)
326 in only a few genera like *Kosakonia* (as expected), *Rhodospirillum*,
327 *Asticcacaulis* and *Enterobacteriaceae* NA (**Figure 7**).

328 When analyzing the clustering and patterns of distribution of all samples
329 by treatment and time point by non-multidimensional scaling analysis (NMDS), it
330 was observed that all samples were mainly clustered by time point, with seed
331 inoculation of *Kosakonia* sp. KO774 not being a major factor for clustering. One
332 sample of 30 dpi belonging to the untreated group can be clearly identified as
333 an outlier (**Figure 8**).

334 These findings suggested that *Kosakonia* sp. KO774 was able to
335 colonize the rice root endosphere at the given conditions only in the first time
336 point analyzed (30 dpi) after seed sowing and that the endosphere microbial
337 community was not significantly affected by the inoculation.

338 DISCUSSION

339

340 Different strains of the recently described *Kosakonia* genus have been
341 isolated from crops and vegetables (Shinjo et al. 2016; Kämpfer et al. 2016;
342 Bergottini et al. 2015; Witzel et al. 2012; Berger et al. 2018) and many possess
343 plant beneficial phenotypes such as nitrogen fixation and phosphate
344 solubilization. In this study, we report the characterization of two endophytic
345 diazotrophic *Kosakonia* strains.

346 Genome analysis of some members of the *Kosakonia* genus, including
347 strain KO774 and KO348 studied here, has revealed that they share some
348 common genetic loci such as enzymes which can facilitate endophytic
349 colonization by degrading plant cell-wall polysaccharides or by removing
350 reactive oxygen species (Li et al. 2017; Reinhold-Hurek and Hurek 2011). In the
351 case of KO774, its genomic analysis revealed a plasmid containing a putative
352 large cellobiose gene, enzyme possibly involved in cellulose degradation; a
353 similar enzyme has been previously reported in other endophyte *Enterobacter*
354 sp. 638 (Taghavi et al. 2015). A recent comparative genomics study based in
355 *Kosakonia radincincitans* DSM 16656 described multiple flagellar and secretion
356 systems contributing to high motility and high competitiveness thus increasing
357 bacterial fitness (Becker et al. 2018). Comparative genomics revealed that
358 enriched protein domains include the nitrogen fixing cluster (*nif* regulon) which
359 has been reported in diverse *Kosakonia* plant- associated strains (Becker et al.
360 2018). Other enriched protein domains include functions related to cobalamin
361 biosynthesis; this is an enzyme cofactor synthesized only by prokaryotes which
362 in *Sinorhizobium meliloti* involved in symbiosis and nodule formation (Taga and

363 Walker 2010). Phosphonate metabolism is also enriched in plant- associated
364 *Kosakonia* strains; phosphonate is a rich source of soil phosphate which plays a
365 role in plant-bacteria interaction (Kamat and Raushel 2013). Phosphonate
366 utilization strains of *Stenotrophomonas rhizophila*, *Cupriavidus basilensis*,
367 *Caulobacter segnis*, among others have been isolated from the rhizoplane of
368 *Lolium perenne* (Fox et al. 2014). Finally, some ethanolamine utilization protein
369 domains were also enriched; ethanolamine can be used in some bacteria as a
370 valuable source of carbon and nitrogen (Kaval and Garsin 2018). Ethanolamine
371 utilization is important for bacterial pathogens of animals and plants as for
372 example in the plant pathogen *Erwinia chrysanthemi* (Kaval and Garsin 2018).
373 Interestingly, recently ethanolamine and derivatives have been linked to plant
374 bacterial interkingdom signaling (Coutinho et al. 2018).

375 Previous studies have shown that good endosphere colonizers are
376 recovered *in vitro* in the order of 10^4 - 10^6 cfu/g after more than one week post-
377 inoculation (Schmidt et al. 2011; Luna et al. 2010). Colonization studies
378 performed here, indicate that the two *Kosakonia* strains are very efficient root
379 endosphere and rhizoplane colonizers. Fluorescence microscopy visualization
380 also confirmed the KO348 ability to colonize the rice roots endosphere. *K.*
381 *radicincitans* DSM 16656 has also been recently observed in the root cortex of
382 cucumber by confocal microscopy (Sun et al. 2018). The weak GFP signal
383 observed in the endosphere could be due to plasmid loss, as we have also
384 observed in this study. Other studies have shown that transmission electron
385 microscopy (TEM) allowed the localization of bacterial diazotrophs in rice in the
386 apoplasm (Gyaneshwar et al. 2001; Hurek et al. 1994; Egner et al. 1999).

387 Endophytes are likely to have evolved an intimate relationship with their
388 plant host probably involving interkingdom signaling (Reinhold-Hurek and Hurek
389 2011; Lòpez-Fernàndez et al. 2017). We detected approximately 144 putative
390 secreted proteins of *Kosakonia* sp. KO438 and many of these can be involved
391 in plant-bacteria interactions; flagellar and T6SS proteins were among the most
392 abundant found. The secretome profile contained several membrane-
393 associated proteins; these are not necessarily secreted since some proteins
394 can end up in the supernatant since they are loosely associated with membrane
395 or due to cell lysis. A similar secretome analysis has been performed in the
396 endophyte *H. seropedicae* SmR1 and 41 secreted proteins have been reported
397 including 19 flagella-related proteins (Chaves et al. 2009). No proteins
398 belonging to the T6SS have been found in the secretome of *H. seropedicae*
399 however, the presence of T6SS in genomes is very common among plant-
400 associated bacteria (Levy et al. 2018) and in endophytes (Frank 2011).
401 Interestingly, T6SS genes have been found enriched in the rhizoplane of barley
402 (Bulgarelli et al. 2015). The T6SS is a phage-like secretion system found in
403 approximately 25% of Gram-negative bacteria, mainly in Proteobacteria
404 including many plant-associated bacteria (Boyer et al. 2009; Bernal et al. 2018).
405 Interestingly, it is a host-specificity factor in the symbiont *R. leguminosarum*
406 (van Brussel, A. A.Zaat et al. 1986). *K. radicincitans* possesses 3 different types
407 of T6SS, however their function and /or mechanism of action have not been
408 studied (Becker et al. 2018). T6SS in endophytes can be involved in host
409 interaction and/or antagonizing other microbes in the endosphere (Frank 2011).
410 Here, we report that a T6SS mutant of *Kosakonia* sp. KO348 displayed a
411 significant decrease in rice rhizoplane and root endosphere colonization thus

412 suggesting a role in the host-bacteria colonization/interaction. The *hcp* mutant
413 was not affected in bacterial growth under the conditions tested hence the
414 T6SS system does not play a role in bacterial growth. A limitation in the T6SS
415 root endosphere colonization experiment was that the many *Kosakonia* cells *in*
416 *planta* lose the pBBR1MCS-5 plasmid vector, regardless whether it harbors or
417 not the *hcp* gene. This vector is stable in *Kosakonia* KO348 in laboratory media
418 thus it is lost during *Kosakonia in planta* colonization due to the lack of selection
419 hence affecting the studies involving complementation of genomic mutants.

420 Nitrogen is one of the mostly used fertilizers worldwide for all cereals
421 including rice with an annual utilization growing rate of 1.9 and it is expected
422 that 201.66 million tons will be used in 2020 (FAO 2017). Biofertilizers are
423 considered an alternative to decrease the use of chemicals, however only few
424 reports of greenhouse and field trials using rhizospheric or endospheric
425 diazotrophic strains have been performed thus far. Most of these are in wheat
426 or maize and reporting an increase between 6 and 33% in total yield (Santi et
427 al. 2013). Furthermore, plant-associated microorganisms applied in agriculture
428 as biofertilizers or biopesticides are usually subject to a rigorous risk
429 assessment which requires a better understanding of the mechanisms involved
430 in the mutualism to facilitate and promote the development and application of
431 sustainable microbial solutions in crop production (Brader et al. 2017). The
432 associative microbial nitrogen fixation supplied by microbes in rice is predicted
433 to be between 20-25% of the total nitrogen needed by the plant (Ladha et al.
434 1987; Saikia and Jain 2007). Inoculation experiments using *H. seropedicae* or
435 *Burkholderia* sp. revealed that 11-20% of the total nitrogen accumulated in rice
436 plants can be attributed to the bacterial strains (Divan Baldani et al. 2000).

437 Similarly, inoculation studies using *K. radicincitans* (DSM 16656) increased
438 plant root or shoot dry weight by 150% under high nitrogen conditions (350 mg
439 per plant) and 130% in low nitrogen conditions (150 mg per plant) (Berger et al.
440 2013). This latter study also showed that plants with low nitrogen
441 supplementation increased the pathogen defense-related markers and
442 suggested this plant response could negatively affect/inhibit the PGP effect of
443 *Kosakonia*. A recent report using the AbiVital product (67% *K. radicincitans*
444 DSM 16656^T and 37% cryopreservation additives) in maize resulted in an
445 increase in yield of around 30% in field trials including organic and conventional
446 cultivation systems (Berger et al. 2018). In our field trial, we decreased the
447 nitrogen fertilisation by 50% hoping that by supplementation via seed-
448 inoculation of the *Kosakonia* strain could at least in part overcome nitrogen
449 deficiency; this was not the case in any of the measurements performed. This
450 experiment could have benefited from knowing the nitrogen concentration in the
451 soil used for the field trial. We observed that under the used conditions the
452 colonization of the inoculated strain was rather inefficient over a longer period of
453 time (more than 30 days) and that it did not affect the root endosphere
454 microbiome; thus a possible limiting factor was likely to be the establishment of
455 the strain in the plant endosphere. This could have been due to inoculation
456 method, soil microbial community or abiotic factors which are not favorable for
457 the *Kosakonia* strain that we used. Interestingly, Becker et al. 2018 reported a
458 significant impact on the bacterial community composition of tomato following
459 inoculation of *K. radicincitans* DSM 16656^T. This field trial was performed in
460 Spain on a different rice variety used in the laboratory experiment hence
461 colonization efficiencies of *Kosakonia* could be different from the ones reported

462 in this study. However other members of *Kosakonia* have been shown not to
463 display host specificity and in general endophytes are generalists being able to
464 colonize any different hosts (Ma et al. 2011; Compant et al. 2005).

465 Due to the recent description of the genus *Kosakonia* (Brady et al. 2013),
466 there are only a few reports on the presence of *Kosakonia* sp. in rice
467 microbiome. *Kosakonia* sp. is a dominant colonizer in seeds of three salt-
468 tolerant rice varieties (Walitang et al. 2017, 2018b). In addition, when
469 inbreeding seed varieties containing *Kosakonia*, it was then present with a
470 similar abundance or even at higher levels within the offspring, suggesting that
471 is part of the core microbiota of some rice varieties (Walitang et al. 2018a).
472 *Kosakonia* sp. has been isolated from different rice varieties, largely
473 representing approximately one third of the total isolates (Hardoim 2015). This
474 indicates that members of *Kosakonia* are common endophytes of rice.

475 This work has characterized two *Kosakonia* strains giving some
476 highlights of their interaction with the plant host and its colonization. Further
477 studies on the genus *Kosakonia* are important for understanding the
478 mechanisms which allow members of this genus to be successful endophytic
479 colonizers and be part of the microbiome of economically important crops.

480

481 MATERIALS AND METHODS

482 Bacterial strains and growth conditions

483 The *Kosakonia* strains used in this study KO348 and KO774 were
484 previously isolated from the root endosphere from rice grown in Italy (Bertani et
485 a 2016). Strains KO774, KO348, KO348(pBBRgfp), KO348hcp and
486 KO348hcp(pBBRhcp) were routinely grown in Luria-Bertani (LB) broth at 30 °C.
487 In order to obtain spontaneous rifampicin resistant KO348 and streptomycin
488 resistant KO774, strains were grown in 1/6 TSB (Tryptic Soy Broth) medium
489 supplemented with gradually increasing amounts of rifampicin (Rif) or
490 streptomycin (Sm) respectively, ranging from 15 to 100 µg ml⁻¹. Finally, cultures
491 were plated on TSA (Tryptic Soy Agar) and single colonies were re-inoculated
492 in TSB containing Rif 100 µgml⁻¹ or Sm 100 µg ml⁻¹. When required, antibiotics
493 for *Kosakonia* strain growth were added at the following concentrations:
494 rifampicin, 50 µg ml⁻¹, gentamicin, 25 µg ml⁻¹ and kanamycin, 100 µg ml⁻¹.
495 *Escherichia coli* DH5α and S17 were grown at 37 °C in LB broth and when
496 appropriate antibiotics were added at the following concentrations: ampicillin,
497 100 µg ml⁻¹, and gentamicin, 15 µg ml⁻¹.

498 Genome sequencing of *Kosakonia* strains

499 The genome of *Kosakonia* sp. KO348 has been previously sequenced
500 and was deposited at DDBJ/EMBL/GenBank under the accession
501 no. JZLI000000000 (Meng et al. 2015) and in IMG/M (U.S. Department of
502 Energy- Joint Genome Institute) as genome ID 2651869662. We re-sequenced
503 KO348 using Illumina HiSeq technology at 230x sequencing depth. The
504 genome was assembled using spades v. 3.10.1. The genome was annotated
505 via IMG Annotation Pipeline v.4.16.0. The draft genome sequence of *Kosakonia*

506 sp. KO774 was also determined in this study. For this, the genomic DNA was
507 obtained by Sarkosyl-Pronase lysis protocol as described by Better et al. (1983)
508 and then used to prepare a sequencing-ready library. Sequencing was
509 performed on Illumina MiSeq platform using 150-bp paired-end reads. The
510 genome of *Kosakonia* sp. KO774 was deposited in IMG/M as genome ID
511 2758568389. Automated annotation of *Kosakonia* sp. KO774 draft genome
512 sequence was performed using IMG/M (U.S. Department of Energy- Joint
513 Genome Institute).

514 ***Kosakonia* genome analysis**

515 In order to analyze the genomes for a phylogenetic analysis, 15
516 *Kosakonia* genome sequences were retrieved from the Integrated Microbial
517 Genomes database IMG/M (U.S. Department of Energy- Joint Genome
518 Institute).

519 *Escherichia coli* K12 MG1655 served as an outgroup. The list of single
520 copy marker genes was retrieved for all genomes and consisted of mainly
521 ribosomal proteins. Only genes that were present in all 15 genomes were used
522 and these included the following Clusters of Orthologous Groups (COGs):
523 COG0012, COG0016, COG0052, COG0087, COG0090, COG0091, COG0092,
524 COG0094, COG0096, COG0097, COG0098, COG0099, COG0102, COG0103,
525 COG0124, COG0186, COG0197, COG0200, COG0201, COG0522, COG0525,
526 COG0533, COG0541. The genes of each COG in all 15 genomes were aligned
527 separately using MAFFT multiple aligner version 7.221 (Kazutaka Katoh and
528 Daron M. Standley) using default parameters. The multiple sequence alignment
529 was trimmed with trimAl v1.3 using default parameters. Next, the different COG
530 alignments were concatenated together using a custom script to yield 15

531 sequences of all 23 single copy genes. RAxML version 7.6.3 (The Exelixis Lab -
532 Heidelberg Institute for Theoretical Studies) was used to construct the tree
533 using the following parameters: raxmlHPC-PTHREADS-SSE3 -f a -p 12345 -x
534 12345 -# 1000 -m PROTGAMMALG -T 8 with the outgroup being *E. coli*. The
535 Best-scoring ML tree with support value was visualized using iTOL (Letunic I
536 and Bork P).

537 We retrieved all proteins (n=70) located within and adjacent to the T6SS
538 operons in the KO348 genome and we blasted them with the proteins found in
539 the secretome (see below) searching for possible hits which might be T6SS
540 effectors.

541 **Plant colonization experiments**

542 For all the rice endosphere colonization experiments, we followed the
543 inoculation protocol described previously by Bertani et al. (2016), with a few
544 modifications. *Kosakonia* strains Rif or Sm resistant were grown on LB media
545 to an OD₆₀₀ of 0.8 and 7 days-old germinated rice plantlets cv. “Baldo” were
546 then submerged in this bacterial suspension for 1 hour and transferred
547 independently to a tube containing Hoagland’s semi-solid solution (Steindler et
548 al. 2009). Plantlets were then watered and grown for a number of days;
549 *Kosakonia* strains were then re-isolated from roots and/or the green aerial part
550 of the plant after surface sterilization and sterility controls were performed as
551 previously reported (Bertani et al. 2016). Plant material was finally macerated in
552 PBS solution and serial dilutions of this macerate were plated in TSA containing
553 the appropriate antibiotics, then incubated at 30 °C for 24 hours and counted for
554 CFU/g calculation.

555 In the case of rhizospheric colonization: roots were rinsed with sterile
556 water removing all remaining Hoagland's semi-solid solution and then vortexed
557 in 5ml of PBS solution for 1 min. Serial dilutions of this PBS solution were then
558 plated on the appropriate selection media for CFU/g calculation.

559 Plasmid-loss calculation of *Kosakonia* plant colonization strains was
560 performed by plating complemented *Kosakonia* cells KO348hcp(pBBRhcp)
561 isolated from the rhizoplane and root endosphere in the following selective
562 media: LB supplemented with kanamycin 100 $\mu\text{g ml}^{-1}$ plus gentamicin 25 $\mu\text{g ml}^{-1}$
563 for plasmid complemented cells and LB supplemented with only kanamycin
564 100 $\mu\text{g ml}^{-1}$ for cells which lost the plasmid. CFU/g and percentage of plasmid
565 loss was calculated.

566 For comparing the rhizoplane and endosphere colonization ability
567 (**Figure 2**) between KO348 and KO774 strains, Kruskal-Wallis test was used
568 for specific pairs of data (KO348 vs. KO774) in single and co-inoculation by
569 Prism 7 (Graphpad Software, Inc.). In the analysis of the rhizospheric and
570 endophytic colonization of KO348 (**Figure 3**) and, on the effect of T6SS in
571 colonization ability of KO348 (**Figure 6**) Kruskal-Wallis test was also used for
572 corresponding specific pairs of data. All statistical analyses were performed with
573 Prism 7 (Graphpad Software, Inc).

574 **Visualization of *Kosakonia* sp. KO348 in rice roots by confocal** 575 **microscopy**

576 To further describe colonization process by *Kosakonia* strains, rice
577 plantlets were inoculated with strain KO348(pBBRgfp) harboring plasmid
578 pBBR2GFP, which constitutively expressed the autofluorescent GFP protein (da
579 Silva et al. 2014), as described above. Colonization assessment of rice by strain

580 KO348 harboring the pBBR2GFP was performed at several time points (5, 10,
581 30 and 50 days post inoculation). For surface visualization, samples (roots and
582 shoots) taken from 10 plants at different time points were rinsed with distilled
583 water and directly observed under a confocal microscope (Olympus Fluoview
584 FV1000 with multiline laser FV5-LAMAR-2 HeNe(G) and laser FV10-
585 LAHEG230-2). For internal colonization, samples were surface sterilized, after
586 being rinsed, with ethanol 75% for 2 minutes and rinsed thrice with distilled
587 water. Then samples were treated with sodium hypochloride (7%) solution for 2
588 minutes and then rinsed followed by two 75% ethanol treatments for 1 minute
589 and finally rinsed thrice with distilled water. Samples were then cut with a razor
590 transversally or longitudinally and observed under the confocal microscope. X,
591 Y, Z pictures were taken at 405, 488, 633 nm and with 10X, 20X or 40X
592 objectives. Z stacks were observed using Imaris software or with Image J
593 (National Institute of Health, U.S.A.). Pictures were cropped and due to the
594 convolution process in the microscope, whole pictures were sharpened and the
595 light/contrast balance improved to better observe the image details, as seen
596 when samples are observed in the dark under the microscope (as described in
597 Glassner et al. 2015).

598 **Determination of the *Kosakonia* sp. KO348 protein secretome**

599 In order to determine the proteins which were secreted by *Kosakonia* sp.
600 KO348, the strain was grown in 200 ml plant mimicking AGF liquid media (Ryan
601 et al. 2007) at 30 °C for 16 hrs. The culture was then centrifuged at 3,800 ×g at
602 4 °C for 15 min and the spent supernatant filtered through a 0.45 µm membrane
603 in order to remove any residual bacterial cells. Trichloric acetic acid (TCA) was
604 then added to a final concentration of 10% w/v and incubated 16 hours at 4 °C.

605 Samples were then centrifuged for 60 min at 15,000 ×g at 4 °C. Pellets were
606 washed with acetone and air dried. Protein pellets were then resuspended in
607 NuPage LDS buffer 1x (ThermoFisher Scientific Inc., Waltham, MA, USA),
608 boiled for 5 min and then run 3 cm in a precast NuPAGE 12% Bis-Tris gel
609 (ThermoFisher Scientific Inc., Waltham, MA, USA). The gels were stained with
610 colloidal commasie brilliant blue (Sigma-Aldrich Inc., San Louis, MS, USA). The
611 stained area of the gel was cut into five bands and processed for in-gel
612 digestion with trypsin using standard procedures (Wysocka et al. 2003). LC-
613 MS/MS of the digests was performed using an Easy-nLC II coupled to an
614 Amazon ETD mass spectrometer (Bruker Daltonics, Hamburg, Germany). The
615 resulting spectra were searched using the X!tandem (The Global Protein
616 Machine Organization) search engine and the Uniprot *Kosakonia* sp. KO348
617 published proteome (UP000253187) and filtered at a 2% false discovery rate.
618 Table 1 and Supplementary tables S3 and S4 show the unique peptides and
619 the statistical confidence, the log(e), of the protein matches.

620 **Construction of the *Kosakonia* strain KO348 *hcp* genomic knock-out**
621 **mutant and its genetic complementation**

622 A genomic knockout mutation of the *hcp* gene was constructed, using
623 genomic DNA as template, by amplifying the 5' DNA flanking regions with
624 primers pEXhcp1Fw 5'-AGGATCCTTTAATTTCTACCCGCCTGG3-' and
625 pEXhcp1Rv 5'-ACTCGAGTTTGCAGACAGACAGCTCAAC-3') and 3' DNA
626 flanking regions with primers pEXhcp2Fw 5'-
627 AGAATTCAGGTGTGACCTATGCATTCCA-3'and pEXhcp2Rv 5'-
628 AGGTACCTTGTTTGCAGCCATTTCCG-3'). The 5' and 3' fragments were
629 then ligated on either side of a kanamycin resistance gene and the final

630 fragment cloned in gene replacement vector pEX19Gm (Hoang et al. 1998)
631 generating pEX19Kmhcp. This latter plasmid was then electroporated
632 into strain *KO348* and following selection (Km^R Gm^S) resulted on the generation
633 of an *hcp* knock-out *mutant* strain which was named *Kosakonia* *KO348hcp*.

634 The *hcp* full-length gene (including its gene promoter) was amplified with
635 the primers prom+hcpFW 5'-AGGTACCTGTTTCTGAAGGTCGATGGAG-3'
636 and prom+hcpRv 5'-AGGATCCTGTTTGACAGCCATTTTCGGT-3', the
637 sequence was verified via DNA sequencing and the 802 bp fragment was
638 cloned in the gentamicin resistant pBBR1MCS-5 vector (Kovach et al. 1995).
639 This plasmid was electroporated in the mutant strain *KO348hcp*, and selected
640 for Km^R and Gm^R , and the resulting *KO348hcp* complemented strain was
641 named *KO348hcp(pBBRhcp)*. The pBBR1MCS-5 plasmid vector (Kovach et al.
642 1995) was also electroporated in *KO348hcp*, resulting in *KO348hcp(pBBRMCS-*
643 *1)*.

644 **Bacterial growth curves**

645 Three biological replicates of cultures of strains *KO348*, *KO348hcp* and
646 *KO348hcp(pBBRhcp)* were grown in LB broth supplemented with appropriate
647 antibiotics and grown at 30°C with shaking at 200 rpm. Optical density values at
648 OD600 were measured every hour until reaching the stationary phase.

649 Three biological replicates of cultures of strains *KO348*, *KO348hcp* and
650 *KO348hcp(pBBRhcp)* were grown in LB broth supplemented with appropriate
651 antibiotics and grown at 30°C with shaking at 200 rpm. Optical density values at
652 OD600 were measured every hour until reaching the stationary phase.

653 **Rice field trial using seeds inoculated with a *Kosakonia* strain**

654 A rice field trial using seeds inoculated with *Kosakonia* sp. *KO774* was

655 carried out between May and October 2016 at Catarroja, Valencia-Spain
656 (39,3859292°N 0.376225411°W). It consisted in 16 experimental plots of wet-
657 seeded paddy rice cv. "J. Sendra" divided in four groups of treatment as follows;
658 (i) eight plots received 100% nitrogen/urea recommended fertilization with four
659 of these planted with seeds inoculated with *Kosakonia* sp. KO774 and the other
660 four plots with the seeds not inoculated and (ii) eight plots received 50% of
661 urea/nitrogen recommended fertilization with four of these planted with seeds
662 inoculated with *Kosakonia* sp. KO774 and the other four plots with the seeds
663 not inoculated. The seed inoculation with *Kosakonia* sp. KO774 was performed
664 by soaking rice seeds in a solution containing 10^8 CFU/ml of the strain for 24
665 hrs. Rice plants were harvested at day 100 post rice-sowing and
666 measurements including germination/plot, weight (1000 grains/plot), (25
667 panicles/plot), yield (m^2) and yield ($kg\ ha^{-1}$) were performed. Statistical analysis
668 of variance (ANOVA) was performed for analyzing the phenotypic differences
669 between groups using Prism 7 (Graphpad Software, Inc).

670 **Microbiome studies**

671 Microbiome analysis was performed on rice roots grown in two plots of
672 the following two treatment groups of the field trials, (i) rice seeds soaked in
673 *Kosakonia* and the soil fertilized with 50% of the recommended nitrogen
674 (treated group) and (ii) rice seeds which were not inoculated with *Kosakonia*
675 and the soil fertilized only with 50% of the recommended nitrogen (untreated
676 group). Rice plants were collected at 30, 60 and 90 dpi and rice roots were
677 washed and surface sterilized. In order to maintain the variability but decreasing
678 the number of samples, one sample was considered as the sterilized roots of

679 three different plants of rice derived from the same plot and collected at the
680 same time point (**Supplementary Figure S7**).

681 DNA from sterilized roots was extracted using PowerMax Soil DNA
682 isolation kit (MO BIO Laboratories, Carlsbad, CA, USA) following the
683 manufacturer's protocol and using 0.5 grams of each sample. The 16S rRNA
684 gene amplicon library was prepared following the manufacturer's protocol
685 (15044223 B) (Illumina Inc., San Diego, CA, USA). Briefly, samples were
686 amplified in the V3 and V4 regions using denaturated primers (Klindworth et al.
687 2013) in a limited cycle PCR, followed by an AMPure XP bead clean-up
688 (A63880I; Beckman Coulter Inc., Brea, CA, USA). A second PCR reaction was
689 then performed to attach dual index and Illumina sequencing adapters using the
690 Nextera XT Index Kit; followed by a final AMPure XP bead clean-up. 16S rRNA
691 gene concentration was measured by fluorimetric quantification using Qubit 2
692 (Invitrogen Inc., Carlsbad, CA, USA). Sequencing was performed using the
693 Illumina Miseq technology. The sequences of raw data were filtered out and the
694 reads were trimmed to a consistent length. Then the data was denoised,
695 chimera filtered, and taxonomically assigned using DADA2 v1.1.5 (Callahan et
696 al. 2016). For the taxonomic analysis, the sequencing reads were clustered into
697 operational taxonomic units (OTUs) defined as groups of sequencing reads that
698 differ by less than a fixed dissimilarity threshold (97%) generated in DADA2
699 using the Greengenes database v13.5 (The Greengenes Database
700 Consortium) modified for including sequence
701 "CTACGGGTGGCAGCAGTGGGGAATTTCCGCAATGGGCGAAAGCCTGAC
702 GGAGCAATGCCGCGTGGAGGTGGAAGGCCACGGGTCGTCAACTTCTTTT
703 CTCGGAGAAGAAACAATGACGGTATCTGAGGAATAAGCATCGGCTAACTC

704 TGTGCCAGCAGCCGCGGTAAGACAGAGGATGCAAGCGTTATCCGGAATGA
705 TTGGGCGTAAAGCGTCTGTAGGTGGCTTTTCAAGTCCGCCGTCAAATCCC
706 AGGGCTCAACCTGGGAACTGCATTGAACTGGCAGGCTGGAGTCTCGTA
707 GAGGGAGGTAGAATTCCAGGTGTAGCGGTGAAATGCGTAGAGATCTGGAG
708 GAATACCGGTGGCGAAGGCGGCCTCCTGGACGAAGACTGACGCTCAGGT
709 GCGAAAGCGTGGGGAGCAAACAGGATTAGATACCCCTGTAGT” as Bacteria
710 Proteobacteria Gammaproteobacteria Enterobacterales Enterobacteriaceae
711 Kosakonia_S (belonging to our strain KO774).

712

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714

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723 LITERATURE CITED

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1042 **Table 1. Flagella-related proteins and Type VI-secretion system related**
 1043 **proteins in the *Kosakonia* KO348 secretome**

Uniprot ID	Protein name	unique peptides	log(e)
Flagella- related proteins			
A0A369A2B9	Flagellin	73	-820,4
A0A368ZZS7	Flagellar hook-associated protein 2	64	-743,7
A0A369ACC2	Flagellar hook-associated protein 1 (FlgK)	47	-556,8
A0A368ZXF8	Flagellin-like protein (Fragment)	4	-471,4
A0A369ADA8	Flagellin	5	-261
A0A368ZX89	Flagellar hook-length control protein FliK	15	-113,9
A0A369A9W2	Flagellar hook-associated protein 3 FlgL	11	-100,6
A0A369A9A4	Flagellar hook protein FlgE	2	-11,6
A0A368ZXA5	Flagellar biosynthesis protein FlhA	3	-11,5
A0A369ACY1	Flagellar protein FlgJ	2	-6,8
Type-VI secretion system proteins			
A0A368ZWR4	Type VI secretion system secreted protein Hcp	7	-61,9
A0A368ZZA3	Type VI secretion system secreted protein VgrG	8	-47
A0A368ZRA6	Type VI secretion system secreted protein VgrG	3	-42,1
A0A368ZYZ5	Rhs element Vgr protein (Fragment)	2	-28,3
A0A368ZZB3	FHA domain protein	3	-11,1

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1047 **Table 2. Field trial biological parameters by group**

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	Weight (1,000 grains/plot)	Weight (25 panicles/plot)	Yield (m ²)	Yield (kg/ha)
Nitrogen 100%	35	96,8	958,8	9587,5
Nitrogen 100% + KO774	35	90	943,8	9437,5
Nitrogen 50%	34,3	98,8	853,5	8585
Nitrogen 50% + KO774	35,3	101,3	828,5	8285

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1051 **FIGURE CAPTIONS**

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1053 **Figure 1. *Kosakonia* phylogenetic tree showing enriched protein domains**1054 **by source of isolation (plants vs. human/animals)** Phylogenetic tree showing1055 the phylogenetic position of the *Kosakonia* sp. strains KO348 and KO7741056 based on 23 single copy genes found among the 14 complete *Kosakonia*1057 genomes from IMG dataset. *Escherichia coli* K-12 MG1655 was used as an1058 outgroup. Enriched protein domains in the plant-associated *Kosakonia* vs.1059 human/animal isolated *Kosakonia* strains are shown by genome. KO774 is

1060 indicated as AG774 and KO348 as AG348.

1061

1062 **Figure 2. Rhizoplane and root endosphere colonization by the two**1063 ***Kosakonia* strains.**1064 Rhizoplane and root endosphere colonization by *Kosakonia* strains was1065 evaluated in rice plants at 14 dpi by antibiotic selection (KO348 Rif^R and KO7741066 Sm^R). Three treatment groups were evaluated, KO348 and KO774 in single1067 inoculation (1×10^8 CFU/ml) and a third group of plants co-inoculated with1068 0.5×10^8 CFU/ml of each strain. Three biological replicates were performed at

1069 different times. Each handled sample consisted of roots of 4 rice plants. The

1070 first biological replicate was performed with 12 plants per group (three handled

1071 samples) while the second and the third with 8 plants (two handled samples

1072 each). Kruskal-Wallis test was performed between strains in single and co-

1073 inoculation, no significant differences were found.

1074

1075 **Figure 3. Endophytic colonization by *Kosakonia* strain KO348.** The

1076 endosphere colonization of the strain KO348 was evaluated in roots of rice

1077 plants at 30 dpi by plating serial dilutions from previously sterilized plant tissues.
1078 The endosphere colonization was evaluated in roots of three different biological
1079 replicates performed at different times, each replicate consisted of 10 plants, all
1080 plants were analyzed for CFU/g independently for confirming colonization
1081 ability.

1082

1083 **Figure 4. Microscopic visualization of *Kosakonia* strain KO348(pBBRgfp)**
1084 **in rice roots at rhizoplane and root endosphere level at 5 and 10 dpi.**
1085 KO348(pBBRgfp) was visualized by fluorescence microscopy at 5 and 10 dpi in
1086 the root hair zone (rhizoplane) and in the root aerenchyma (endosphere). A
1087 total of 10 plants were analyzed for each time point.

1088

1089 **Figure 5. Microscopic visualization of *Kosakonia* strain KO348(pBBRgfp)**
1090 **in rice roots at rhizoplane and root endosphere level at 30 and 50 dpi.**
1091 KO348(pBBRgfp) was visualized by fluorescence microscopy at 30 and 50 dpi
1092 in the root hair zone (rhizoplane) and in the root aerenchyma (endosphere) and
1093 compared to control plants. A total of 10 plants were analyzed for each time
1094 point.

1095

1096 **Figure 6. Role of the Type VI secretion system of *Kosakonia* KO348 in**
1097 **rhizoplane and endosphere rice root colonization.** The effect of type VI
1098 secretion system was tested in the rhizoplane (6a) and in the endosphere (6b)
1099 colonization of rice root plants at 10 dpi. KO348h_{hcp} mutant was inoculated
1100 independently and in competition assays (in the same rice plant) with KO348
1101 WT. Three different biological replicates were performed at different times. In

1102 part A each replicate had 3 plants analyzed individually; in part B each replicate
1103 had 4 plants analyzed individually. For the calculation of plasmid loss during
1104 rhizoplane and root endosphere colonization, the complemented mutant
1105 KO348hcp(pBBRhcp) was plated in Km for KO348hcp and in Km Gm of
1106 KO348hcp(pBBRhcp).

1107

1108 **Figure 7. Heatmap of the most abundant genus (>1%) by sample and**
1109 **treatment at 30 days post inoculation in the field trial.** The clustering of the
1110 most abundant bacterial genera and OTUs present in rice root endosphere at
1111 30 dpi among different samples is showed in the heatmap. The heat map scale
1112 displays the row Z score. Where the Z score is calculated as $(\text{relative abundances of a genus in one sample} - \text{mean relative abundance of the same}$
1113 $\text{genus among total samples}) / \text{standard deviation}$.

1115

1116 **Figure 8. Distribution patterns analysis by NMDS of the microbiomes by**
1117 **time and treatment.** Non-multidimensional scaling analysis plot showing
1118 clustering (NMDS) plot showing clustering of samples of rice endophytic
1119 microbial communities by time and treatment based on Bray-Curtis
1120 dissimilarity.

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1123 **e-XTRA FIGURE CAPTIONS**

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1125 **Supplementary Figure S1. Sequence of a putative plasmid present in**

1126 ***Kosakonia* KO348** Strain *Kosakonia* KO348 genome assembly revealed a
1127 putative plasmid of 74Kbp. The figure shows the contig containing the putative
1128 plasmid included loci for plasmid replication, few toxin-antitoxin systems and a
1129 large cellobiose phosphorylase gene among others.

1130

1131 **Supplementary Figure S2. Visualization of control un-inoculated rice**

1132 **plants at 5 and 10 dpi by confocal microscopy.** Fluorescent colonies
1133 containing *Kosakonia* KO348(pBBRgfp) were not detected in the control plants.

1134

1135 **Supplementary Figure S3. Cluster of Type VI Secretion System of KO348**

1136 T6SS cluster was identified using the online database SecreT6 (Microbial
1137 Bioinformatics Group, SKMML, SJTU), the genes are named according to the
1138 most used nomenclature of Shalom et al. 2007, where *hcp* is named *tssD* and
1139 *vgrG* as *tssl*. The 3 copies of *vgrG* that belong to this cluster are the ones
1140 found on the secretome profile of KO348.

1141

1142 **Supplementary Figure S4. Growth curves of WT (KO348), mutant**

1143 **(KO348hcp) and for KO348hcp(pBBRhcp).** Growth curves of the strains was
1144 performed in biological triplicates.

1145

1146 **Supplementary Figure S5. Rhizoplane and root endosphere colonization**

1147 **by KO348hcp(pBBRMCS-1)** Rhizoplane and root endosphere colonization by

1148 the T6SS KO348hcp mutant harboring the empty plasmid vector
1149 KO348hcp(pBBR). For each experimental group 4 different rice plants were
1150 used. For the calculation of plasmid loss during rhizoplane and root endosphere
1151 colonization, KO348hcp was plated in Km containing plates and in Km Gm for
1152 KO348hcp(pBBR)). Statistically significant endosphere colonization differences
1153 are indicated.

1154

1155 **Supplementary Figure S6. Rice field trial layout** A total of 16 parcels of
1156 paddy rice were followed during 100 days divided in 4 groups of treatment.
1157 Urea 100% fertilized (225 kg/ha) with and without the addition of seed-
1158 inoculated *Kosakonia* strain KO774, and urea 50% fertilized (125 kg/ha) with
1159 and without the addition of strain KO774. For the microbiome analysis, 2 groups
1160 were tested; both urea fertilized 50% with (Treated) and without (Untreated) the
1161 addition of strain KO774. 3 samples were taken from 3 different parcels at 3
1162 time points (30, 60 and 90 days after rice seeds were sowed).

1163

1164 **Supplementary Figure S7. Abundance of 16S rDNA gene sequence 100%**
1165 **identical to the KO774 strain**

1166 Using a modified version of the data base greengenes gg_13_8_99 that
1167 includes the sequence belonging to the V3-V4 16S rDNA fragment of
1168 *Kosakonia* strain KO774 we determined the abundance by sample by treatment
1169 and time point (T= inoculated with *Kosakonia*, U=control).

1170

1171

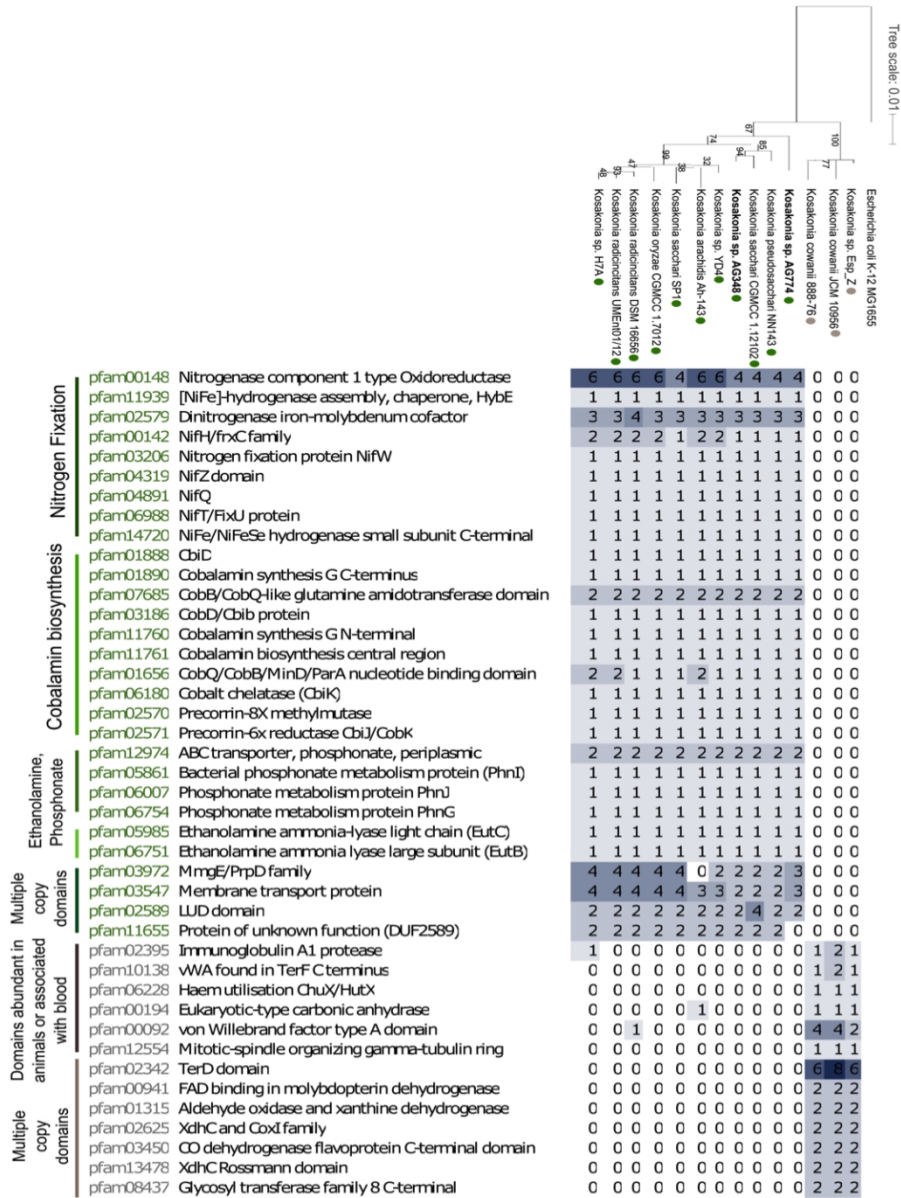


Figure 1. *Kosakonia* phylogenetic tree showing enriched protein domains by source of isolation (plants vs. human/animals) Phylogenetic tree showing the phylogenetic position of the *Kosakonia* sp. strains KO348 and KO774 based on 23 single single copy genes found among the 14 complete *Kosakonia* genomes from IMG dataset. *Escherichia coli* K-12 MG1655 was used as an outgroup. Enriched protein domains in the plant-associated *Kosakonia* vs. human/animal isolated *Kosakonia* strains are shown by genome. KO774 is indicated as AG774 and KO348 as AG348.

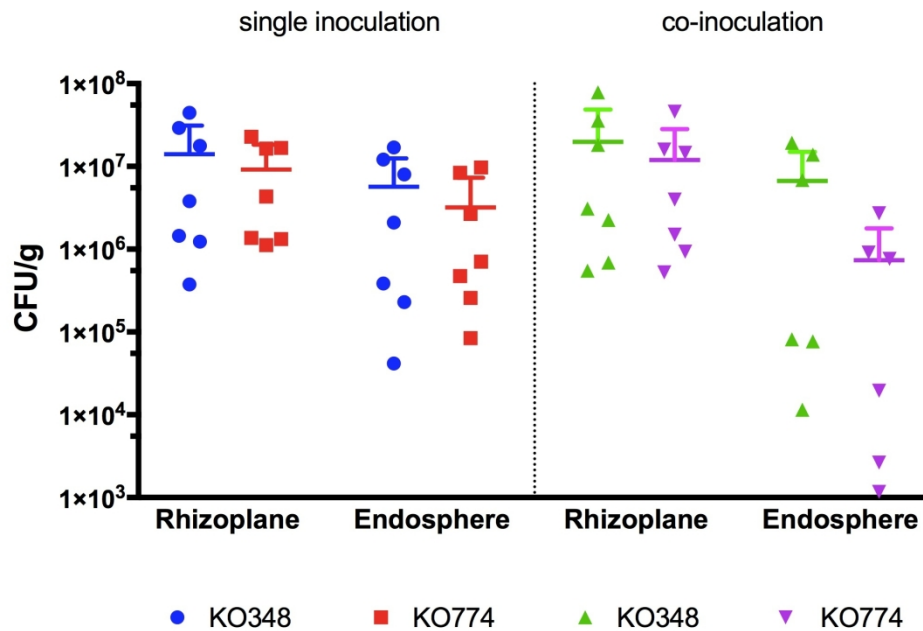


Figure 2. Rhizoplane and root endosphere colonization by the two *Kosakonia* strains. Rhizoplane and root endosphere colonization by *Kosakonia* strains was evaluated in rice plants at 14 dpi by antibiotic selection (KO348 Rif^R and KO774 Sm^R). Three treatment groups were evaluated, KO348 and KO774 in single inoculation (1×10^8 CFU/ml) and a third group of plants co-inoculated with 0.5×10^8 CFU/ml of each strain. Three biological replicates were performed at different times. Each handled sample consisted of roots of 4 rice plants. The first biological replicate was performed with 12 plants per group (three handled samples) while the second and the third with 8 plants (two handled samples each). Kruskal-Wallis test was performed between strains in single and co-inoculation, no significant differences were found.

182x117mm (300 x 300 DPI)

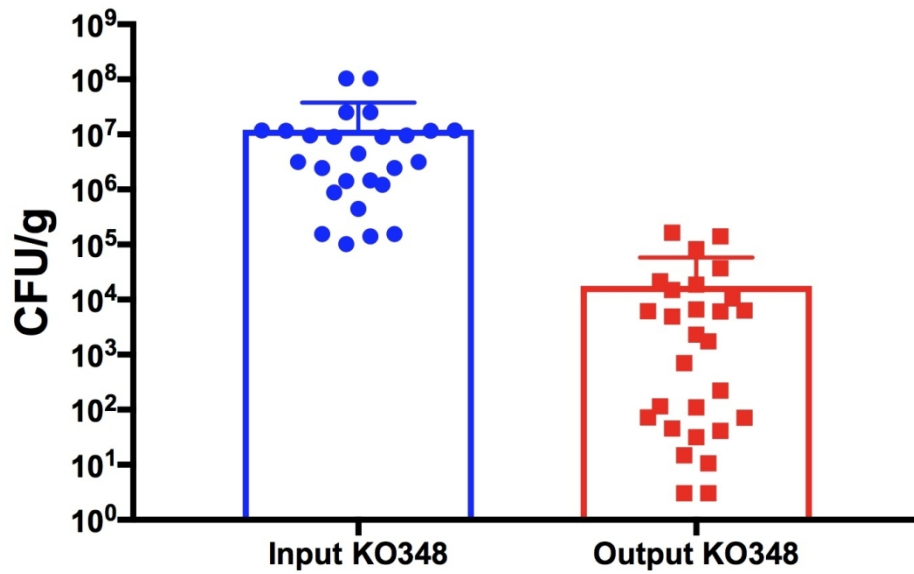
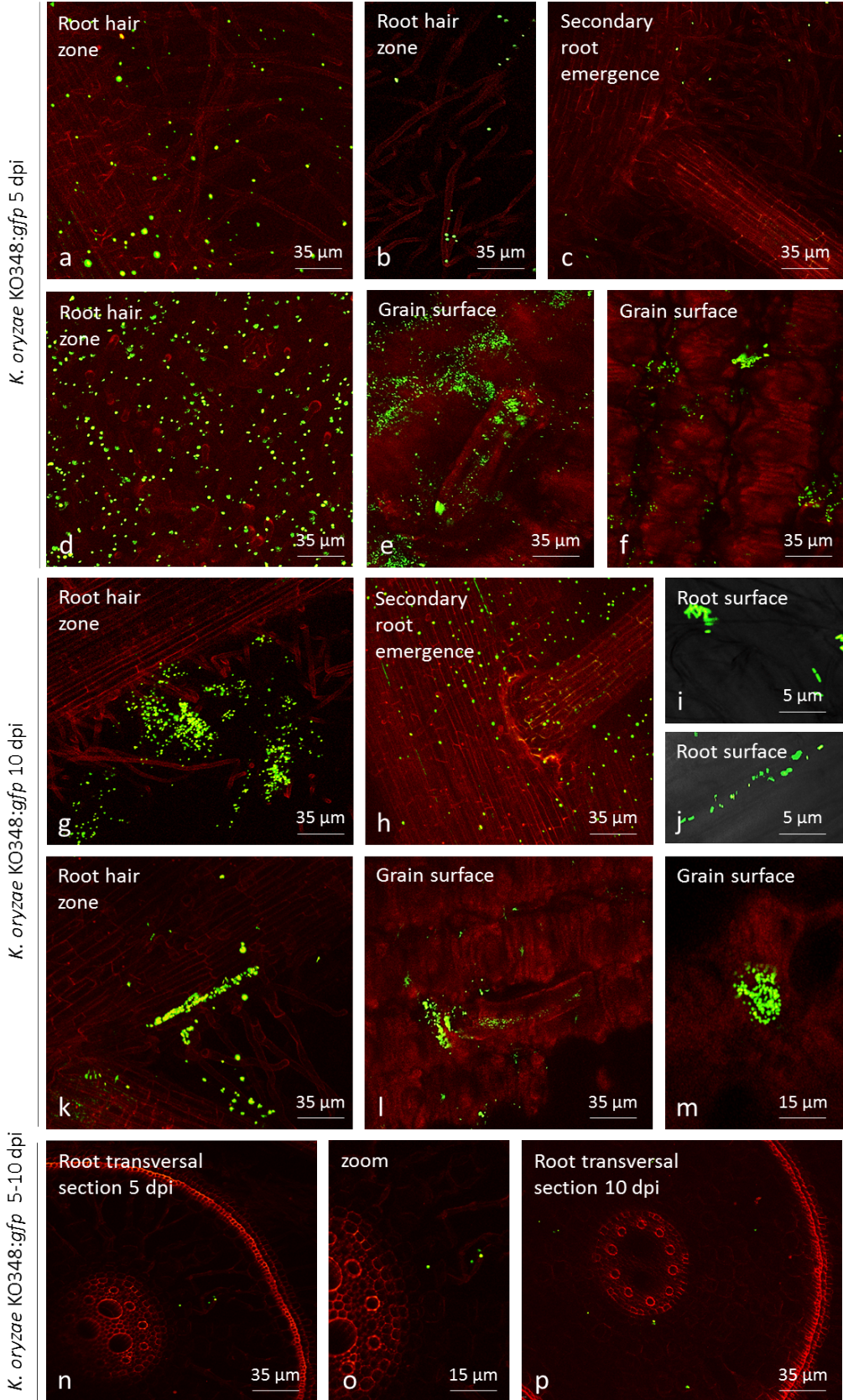
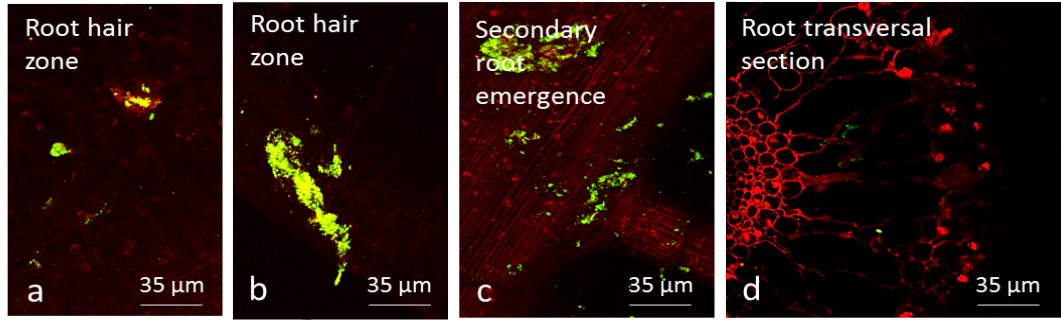
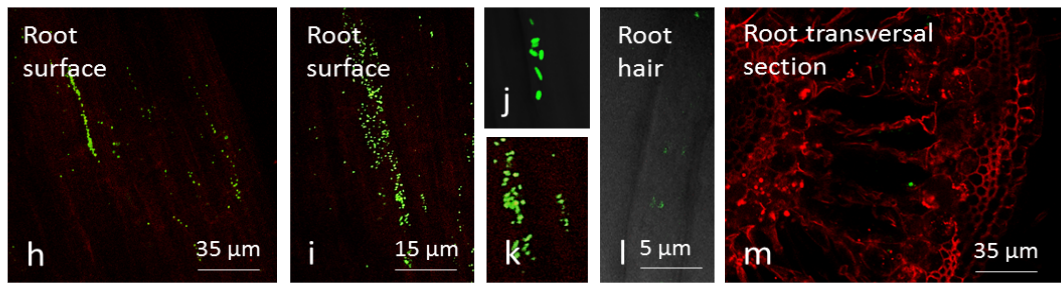
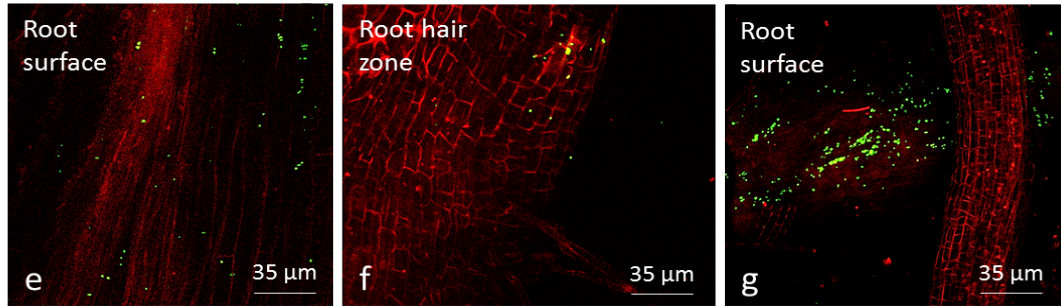


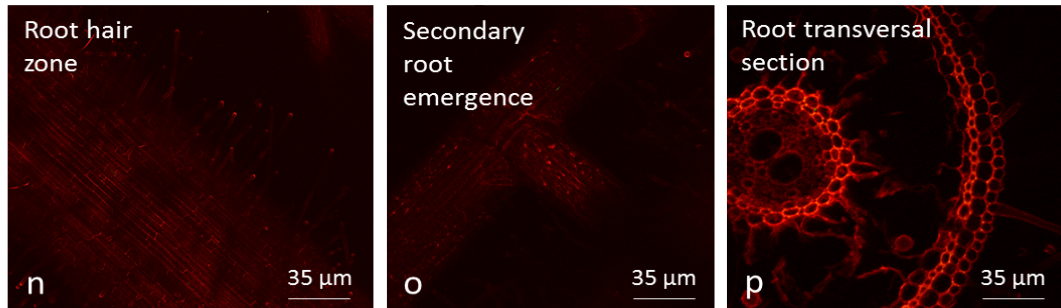
Figure 3. Endophytic colonization by *Kosakonia* strain KO348. The endosphere colonization of the strain KO348 was evaluated in roots of rice plants at 30 dpi by plating serial dilutions from previously sterilized plant tissues. The endosphere colonization was evaluated in roots of three different biological replicates performed at different times, each replicate consisted of 10 plants, all plants were analyzed for CFU/g independently for confirming colonization ability.

126x73mm (300 x 300 DPI)

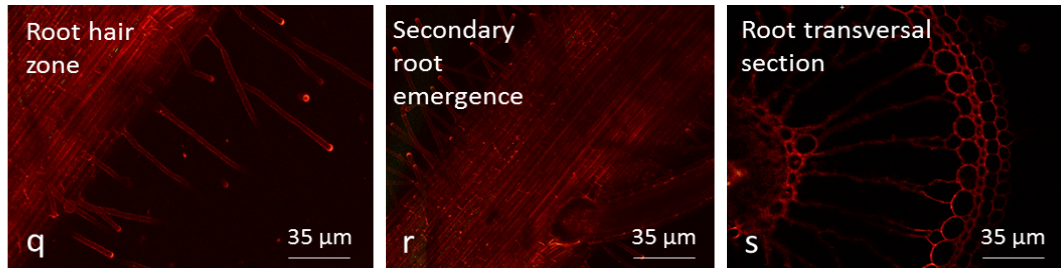


K. oryzae KO348:*gfp* 30 dpi*K. oryzae* KO348:*gfp* 50 dpi

Control 30 dpi



Control 50 dpi



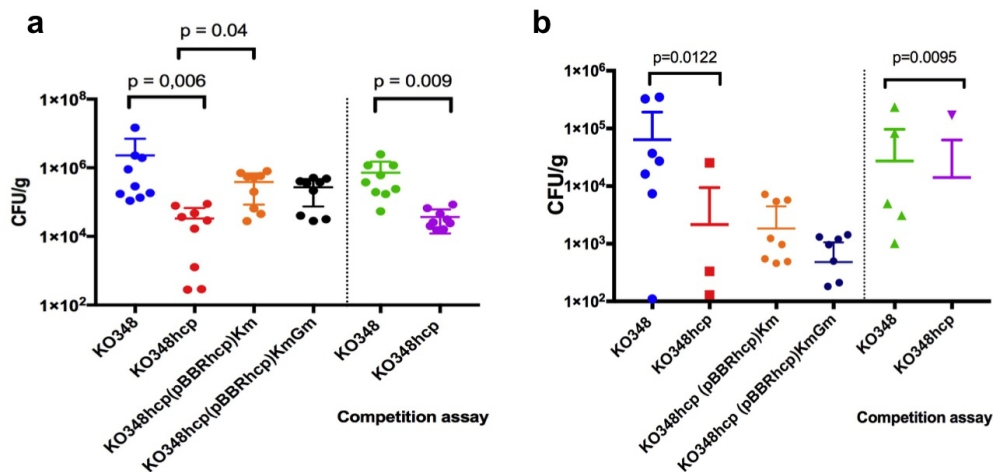


Figure 6. Role of the Type VI secretion system of *Kosakonia* KO348 in rhizoplane and endosphere rice root colonization. The effect of type VI secretion system was tested in the rhizoplane (6a) and in the endosphere (6b) colonization of rice root plants at 10 dpi. KO348hcp mutant was inoculated independently and in competition assays (in the same rice plant) with KO348 WT. Three different biological replicates were performed at different times. In part A each replicate had 3 plants analyzed individually; in part B each replicate had 4 plants analyzed individually. For the calculation of plasmid loss during rhizoplane and root endosphere colonization, the complemented mutant KO348hcp(pBBRhcp) was plated in Km for KO348hcp and in Km Gm of KO348hcp(pBBRhcp).

217x104mm (150 x 150 DPI)

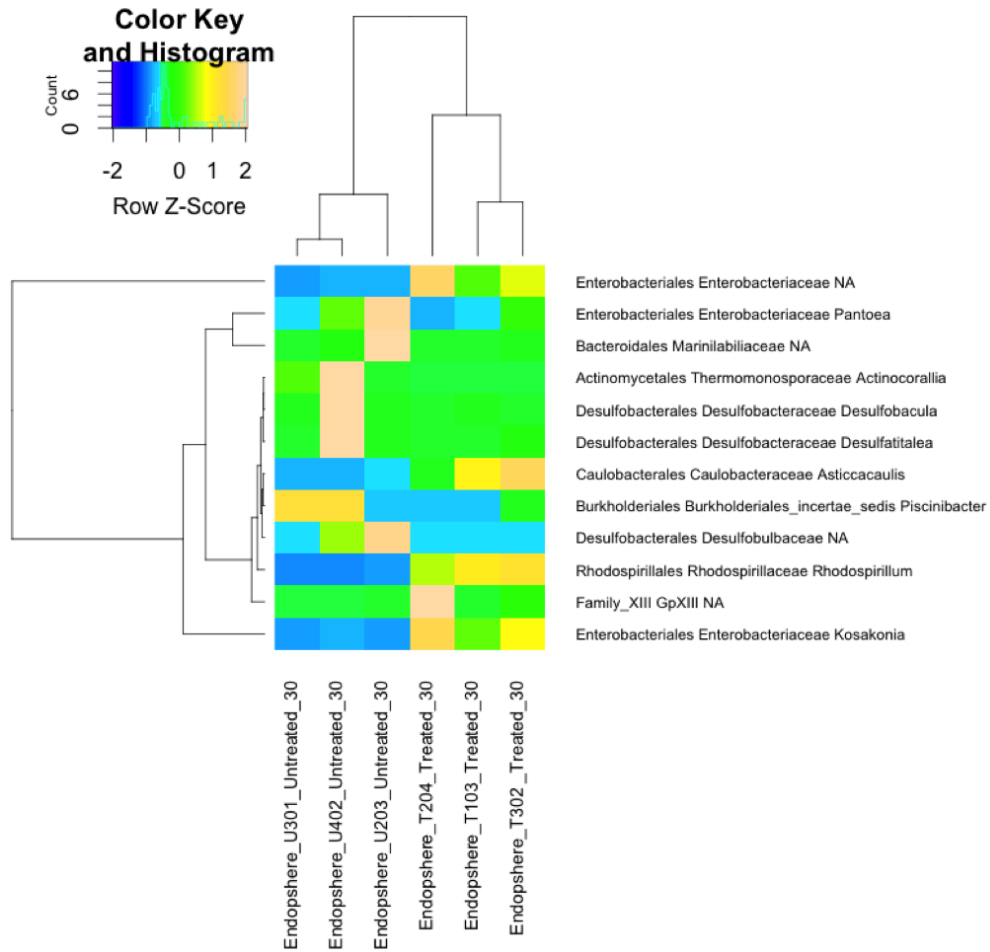


Figure 7. Heatmap of the most abundant genus (>1%) by sample and treatment at 30 days post inoculation in the field trial. The clustering of the most abundant bacterial genera and OTUs present in rice root endosphere at 30 dpi among different samples is showed in the heatmap. The heat map scale displays the row Z score. Where the Z score is calculated as $(\text{relative abundances of a genus in one sample} - \text{mean relative abundance of the same genus among total samples}) / \text{standard deviation}$.

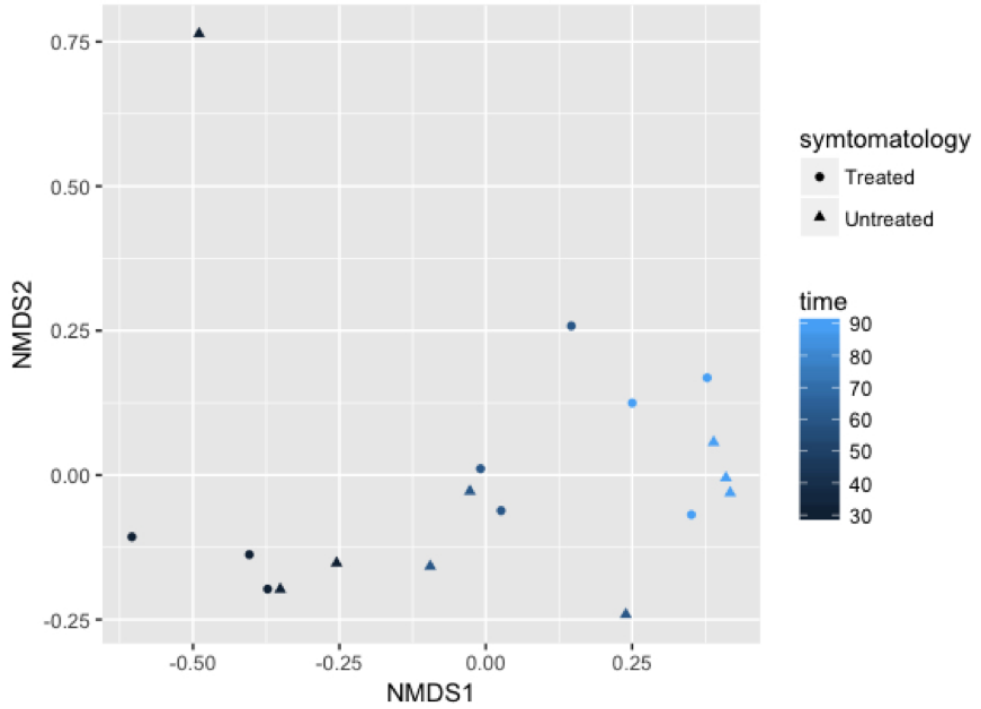
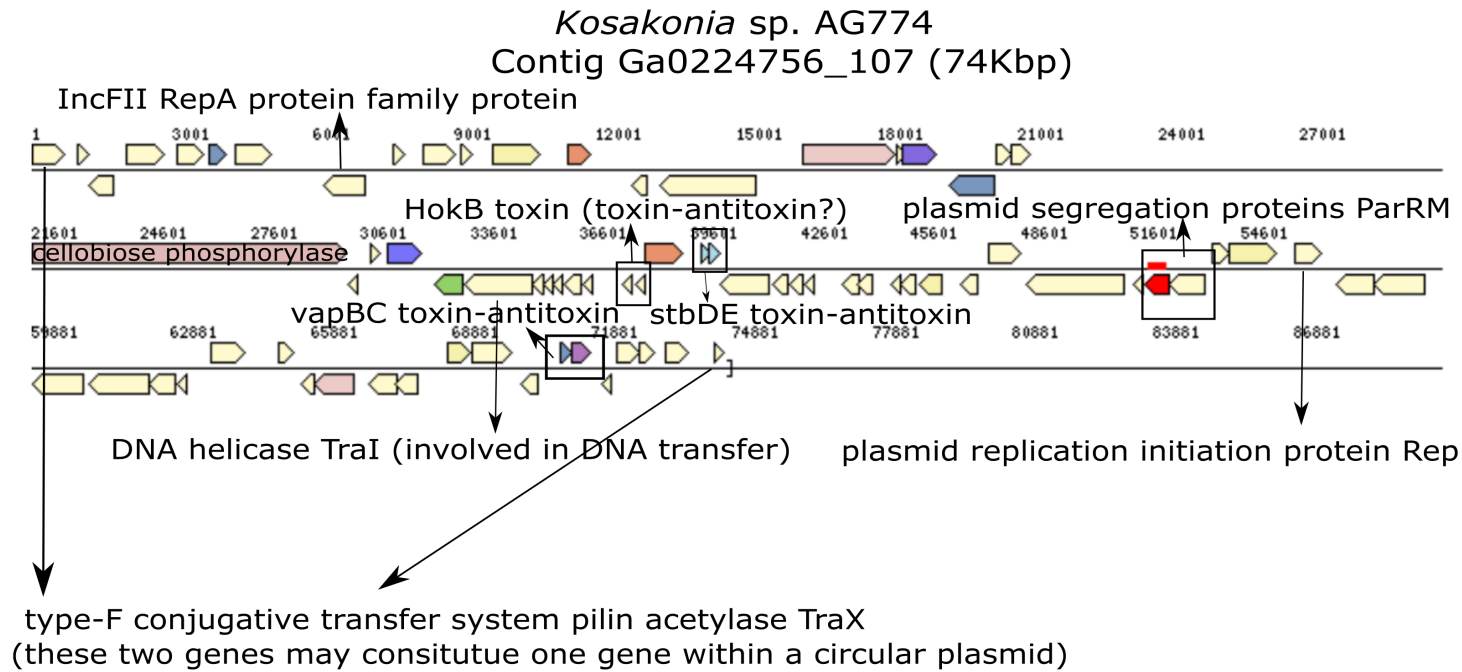
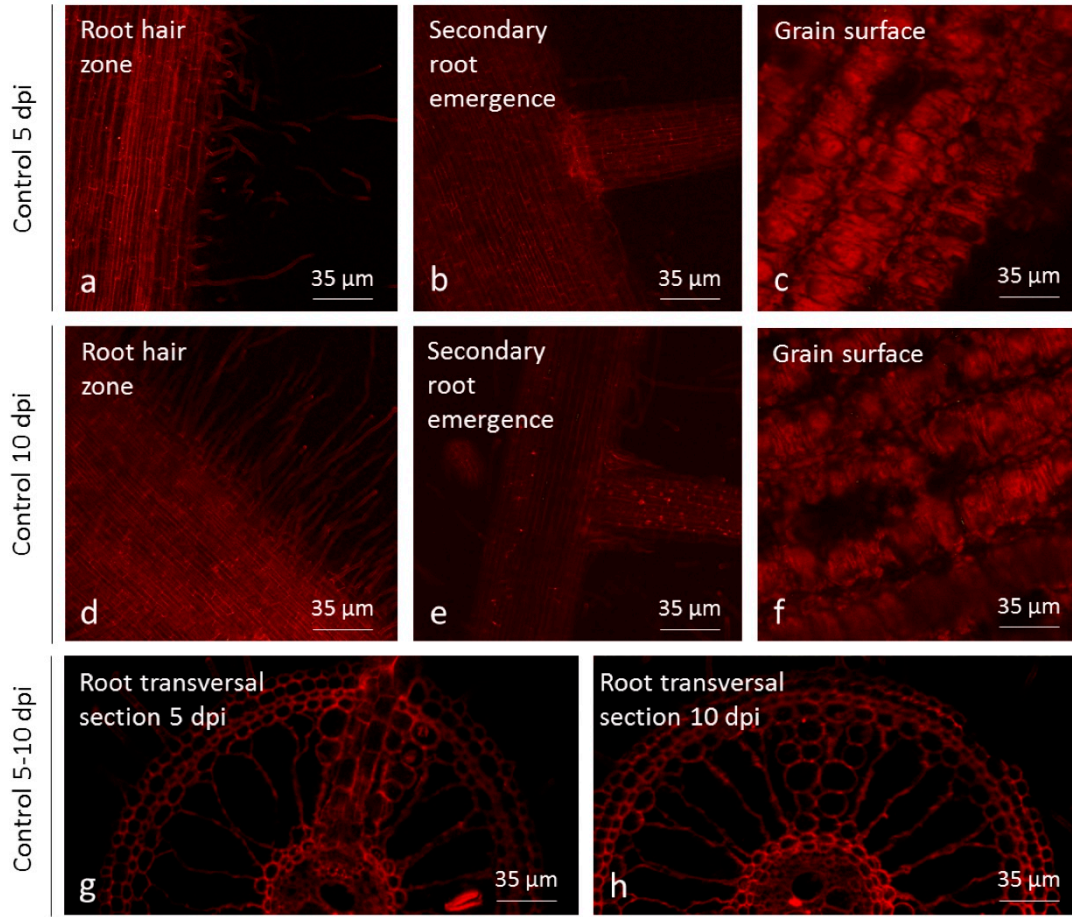


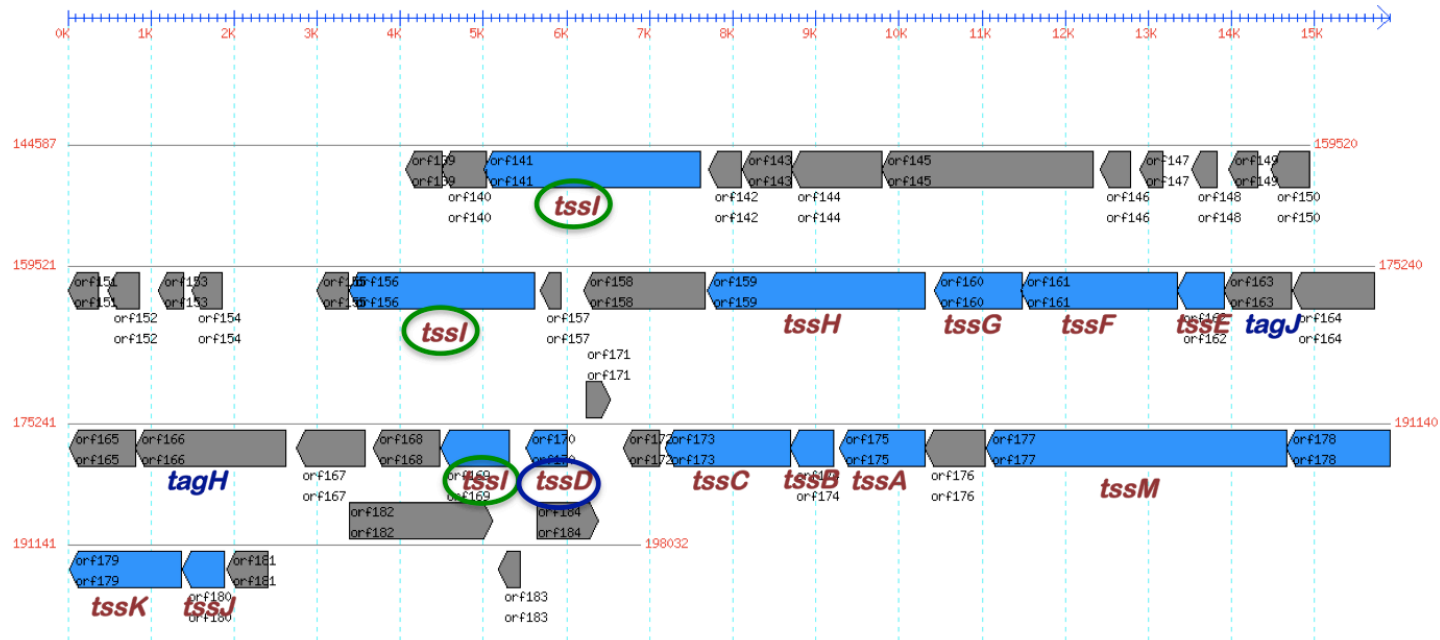
Figure 8. Distribution patterns analysis by NMDS of the microbiomes by time and treatment. Non-multidimensional scaling analysis plot showing clustering (NMDS) plot showing clustering of samples of rice endophytic microbial communities by time and treatment based on Bray-Curtis dissimilarity.



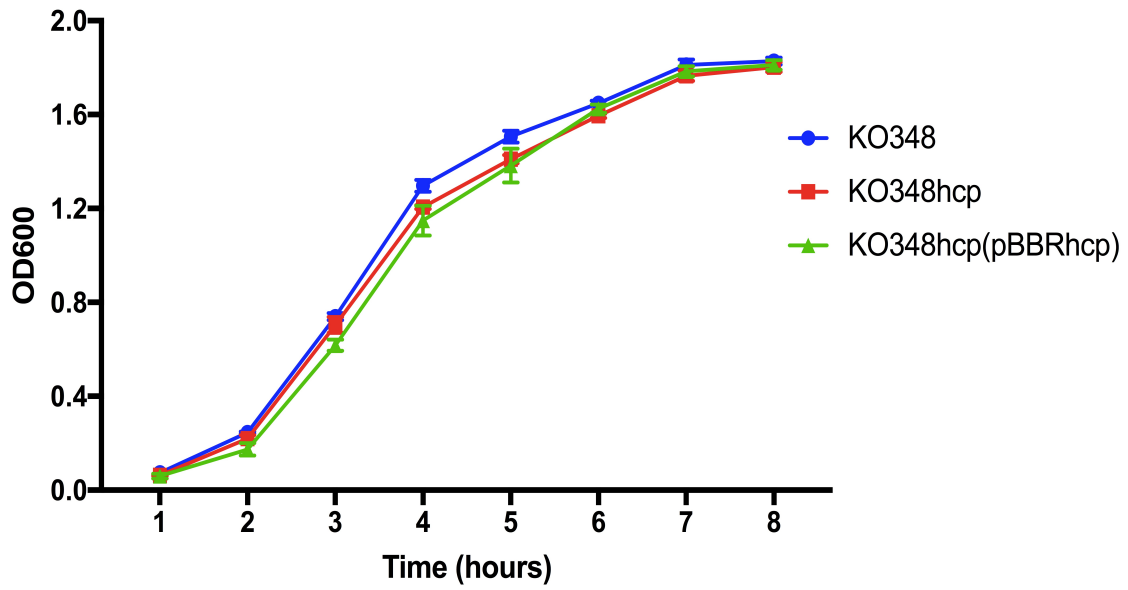
Supplementary Figure S1. Sequence of a putative plasmid present in *Kosakonia* KO348. Strain *Kosakonia* KO348 genome assembly revealed a putative plasmid of 74Kbp. The figure shows the contig containing the putative plasmid included loci for plasmid replication, few toxin-antitoxin systems and a large cellobiose phosphorylase gene among others.



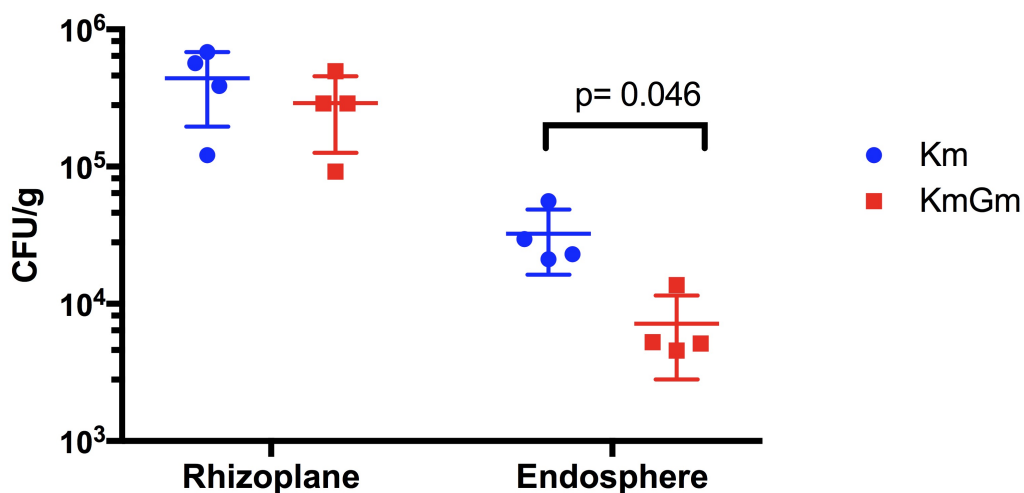
Supplementary Figure S2. Visualization of control un-inoculated rice plants at 5 and 10 dpi by confocal microscopy. Fluorescent colonies containing *Kosakonia* KO348(pBBRgfp) were not detected in the control plants.



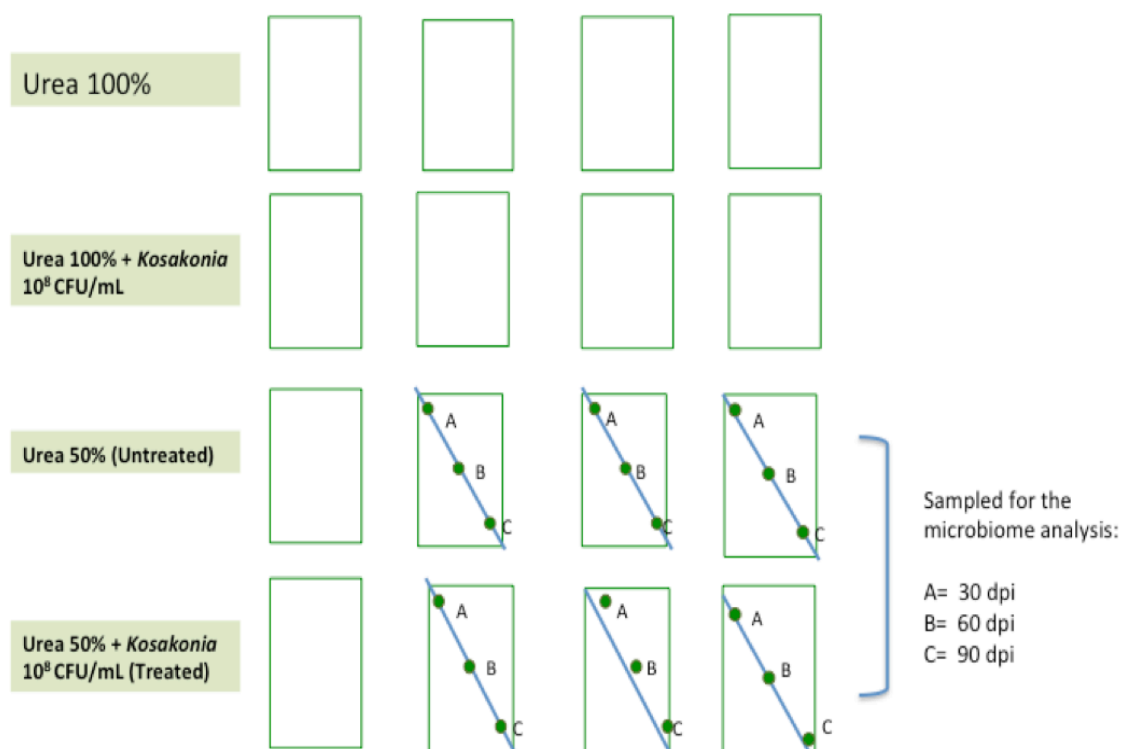
Supplementary Figure S3. Cluster of Type VI Secretion System of KO348. T6SS cluster was identified using the online database SecreT6 (Microbial Bioinformatics Group, SKMML, SJTU), the genes are named according to the most used nomenclature of Shalom et al. 2007, where *hcp* is named *tssD* and *vgrG* as *tssI*. The 3 copies of *vgrG* that belong to this cluster are the ones found on the secretome profile of KO348.



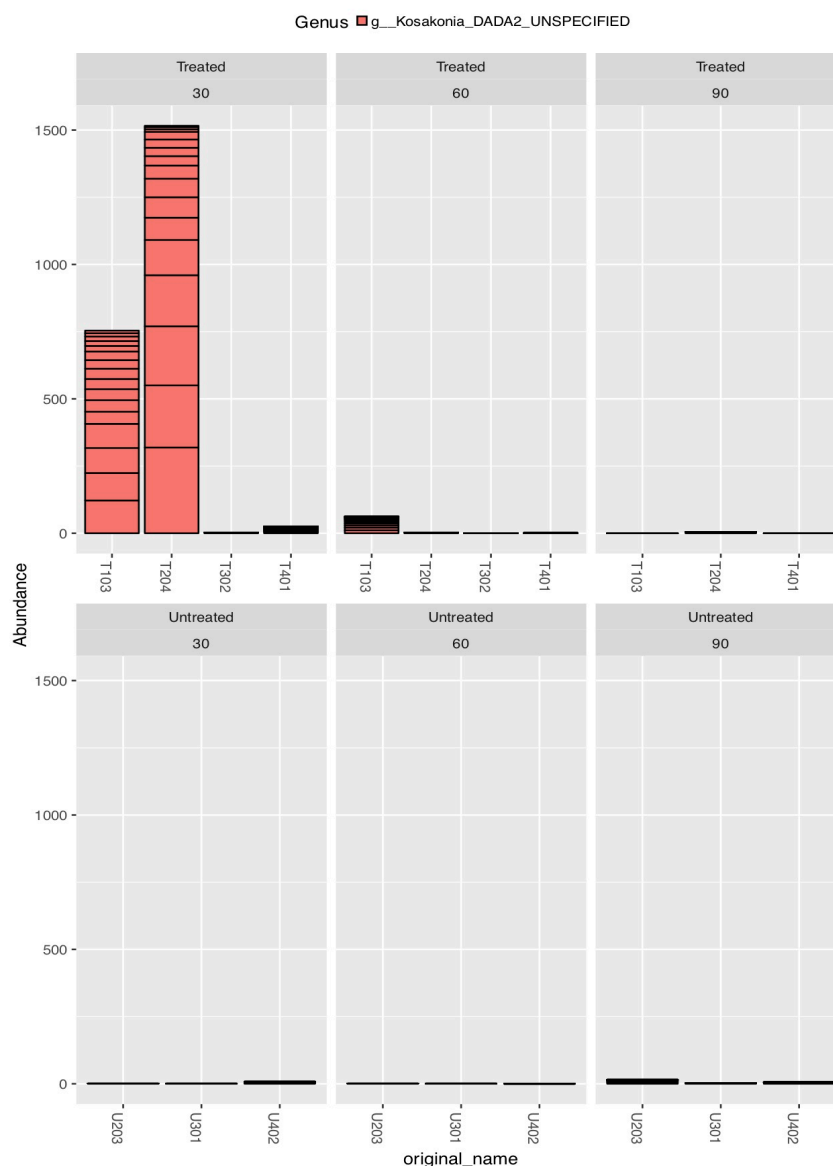
Supplementary Figure S4. Growth curves of WT (KO348), mutant (KO348hcp) and for KO348hcp(pBBRhcp). Growth curves of the strains was performed in biological triplicates.



Supplementary Figure S5. Rhizoplane and root endosphere colonization by KO348hcp(pBBRMCS-1) Rhizoplane and root endosphere colonization by the T6SS KO348hcp mutant harboring the empty plasmid vector KO348hcp(pBBR). For each experimental group 4 different rice plants were used. For the calculation of plasmid loss during rhizoplane and root endosphere colonization, KO348hcp was plated in Km containing plates and in Km Gm for KO348hcp(pBBR)). Statistically significant endosphere colonization differences are indicated.



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Supplementary Figure S7. Abundance of 16S rDNA gene sequence 100% identical to the KO774 strain. Using a modified version of the data base greengenes gg_13_8_99 that includes the sequence belonging to the V3-V4 16S rDNA fragment of *Kosakonia* strain KO774 we determined the abundance by sample by treatment and time point (T= inoculated with *Kosakonia*, U=control).

Supplementary Table S3. Secretome profile of KO348 in plant minimal mimical media

rank	log(e)	% (measured)	unique	total	Mr	Uniprot ID	Protein name
Flagella- related proteins							
1	-820,4	80	73	1041	31,6	A0A369A2B9	Flagellin
2	-743,7	94	64	515	49,4	A0A368Z57	Flagellar hook-associated protein 2
3	-556,8	85	47	276	57,8	A0A369ACC2	Flagellar hook-associated protein 1 (FlgK)
4	-471,4	18	4	65	14,6	A0A368ZF8	Flagellin-like protein (Fragment)
6	-261	28	5	13	31,7	A0A369ADA8	Flagellin
7	-113,9	33	15	25	43,3	A0A368ZX89	Flagellar hook-length control protein FlhK
8	-100,6	48	11	25	34,4	A0A369A9W2	Flagellar hook-associated protein 3 FlgL
61	-11,6	11	2	2	44,4	A0A369A9A4	Flagellar hook protein FlgE
62	-11,5	11	3	3	74,7	A0A368ZA5	Flagellar biosynthesis protein FlhA
105	-6,8	14	2	2	35	A0A369ACY1	Flagellar protein FlgJ
Type-VI secretion system proteins							
9	-61,9	33	7	19	16,9	A0A368ZWR4	Type VI secretion system secreted protein Hcp
13	-47	24	8	8	82,2	A0A368ZZA3	Type VI secretion system secreted protein VgrG
14	-42,1	7,5	3	3	70,7	A0A368ZRA6	Type VI secretion system secreted protein VgrG
22	-28,3	12	2	2	62,6	A0A368ZY25	Rhs element Vgr protein (Fragment)
68	-11,1	11	3	3	65,3	A0A368ZB3	FHA domain protein
Uncharacterized proteins							
5	-444,6	82	43	189	50,4	A0A369A179	Uncharacterized protein
11	-52,3	34	8	8	35,1	A0A369AEY4	Uncharacterized protein
24	-28,3	25	6	6	66,5	A0A368ZUT2	Uncharacterized protein YjcR
48	-12,8	19	3	3	40,9	A0A368ZSW4	Male sterility protein
49	-12,8	35	3	5	17,5	A0A368ZTY2	Uncharacterized protein
84	-7,2	11	2	2	37	A0A368ZUB5	Uncharacterized protein DUF968
95	-6,9	48	2	2	8,7	A0A369A9X6	Uncharacterized protein DUF2526
102	-6,8	20	2	2	26,1	A0A369A2W0	Uncharacterized protein (TIGR03581 family)
121	-6,5	27	2	2	12,8	A0A369A2Q2	Uncharacterized protein
133	-6,2	5	2	2	61,1	A0A368ZPY5	Uncharacterized protein
136	-6,2	6,5	2	2	43,8	A0A368ZRC2	Uncharacterized protein
Transport/ Membrane-related							
10	-57	16	11	11	143,2	A0A368ZRY8	Tetratricopeptide repeat protein
15	-38,6	14	6	6	83,3	A0A369AGH3	Ferric enterobactin receptor
20	-30,1	14	6	6	99	A0A368ZWW1	Fumarate reductase flavoprotein subunit
21	-29,5	20	6	6	76	A0A368ZT10	Alpha-amylase
23	-28,3	27	6	6	70,2	A0A369AAQ1	Macrolide export ATP-binding/permease protein MacB
26	-23,5	7,5	4	4	111	A0A369A602	Efflux pump membrane transporter
29	-19,7	8,7	4	4	84,5	A0A369ACV8	ATP-dependent Clp protease ATP-binding subunit ClpA
32	-18,3	34	4	4	31,6	A0A368ZX64	Putative copper resistance protein D
33	-18,2	19	4	4	55,1	A0A368ZS20	Peptide/nickel transport system substrate-binding protein
35	-18,2	12	4	4	59,6	A0A369ADD5	ATPase subunit of ABC transporter with duplicated ATPase domains
38	-17,5	10	4	4	61,8	A0A369A3X0	Methyl-accepting chemotaxis sensory transducer with Cache sensor
39	-15,4	11	3	3	55,2	A0A368ZLK6	ATP synthase subunit alpha (AtpA)
40	-14,5	15	3	4	60,3	A0A369A497	Methyl-accepting chemotaxis sensory transducer with TarH sensor
41	-14,1	25	3	3	35,1	A0A368ZS22	Peptide/nickel transport system ATP-binding protein
45	-12,9	24	3	3	36	A0A368ZTK4	Peptide/nickel transport system ATP-binding protein
46	-12,9	13	3	3	60,5	A0A369AAC9	Oligopeptide transport system substrate-binding protein
51	-12,6	15	3	3	40,8	A0A369ADH8	Outer membrane pore protein F
52	-12,5	7,3	3	3	111,8	A0A369A125	Multidrug resistance protein MdtB
58	-11,8	36	3	3	17,9	A0A369A8C4	Spy/CpxP family protein refolding chaperone
65	-11,4	13	3	3	42	A0A368ZT49	Carbohydrate ABC transporter ATP-binding protein (CUT1 family)
69	-9,3	14	2	2	48,7	A0A369A5A8	NAD(P) transhydrogenase subunit beta
71	-8	12	2	2	58,1	A0A369A5F6	Methyl-accepting chemotaxis sensory transducer with TarH sensor
73	-7,7	14	2	2	39,8	A0A369A585	DHA1 family inner membrane transport protein
75	-7,5	23	2	2	35,9	A0A369A9K3	Cationic peptide transport system permease protein
77	-7,5	19	2	2	30,8	A0A369AAM8	Formate transporter
80	-7,4	10	2	2	47	A0A368ZT82	High-affinity gluconate transporter
82	-7,3	20	2	2	42,3	A0A369A0A1	Putative MFS family arabinose efflux permease
86	-7,2	17	2	3	35	A0A368ZPY7	ATP-dependent 6-phosphofructokinase (PfkA)
87	-7,1	6,4	2	2	62,3	A0A368ZR51	Cellulose synthase operon protein YhjU
91	-7,1	5,6	2	2	43,5	A0A369A079	Multidrug resistance protein MdtA
97	-6,9	6	2	2	133,9	A0A368ZT65	Tfp pilus assembly protein PilF
100	-6,8	4,2	2	2	106,7	A0A368ZWD1	Sensor histidine kinase RcsC
106	-6,8	12	2	2	46,1	A0A368ZQA9	Gluconate permease GntT
107	-6,8	6,5	2	2	85,1	A0A368ZV34	Penicillin-binding protein 1C
108	-6,8	13	2	2	35,4	A0A369A9P6	Monosaccharide ABC transporter substrate-binding protein (CUT2 family)
115	-6,6	4,8	2	2	63,8	A0A369AAF4	ABC-2 type transport system ATP-binding protein
116	-6,6	5,1	2	2	42,7	A0A368ZW18	Membrane fusion protein (Multidrug efflux system)
124	-6,5	14	2	2	44,8	A0A368ZQZ4	Protein transport protein HofQ
126	-6,5	14	2	2	37,3	A0A369A6U0	Oligopeptide transport system ATP-binding protein
128	-6,4	5,1	2	2	99,4	A0A368ZUK1	Cellulose synthase catalytic subunit [UDP-forming]
132	-6,3	19	2	2	29,2	A0A369A6N7	Phosphatidylglycerophosphatase
135	-6,2	5,8	2	2	98,7	A0A368ZWX2	Phosphotransferase RcsD
138	-6,2	4,1	2	2	133,8	A0A368ZUP3	YD repeat-containing protein (Fragment)
140	-6,1	15	2	2	37,2	A0A368ZS21	Fe(3+) ions import ATP-binding protein FbpC
142	-6,1	6,7	2	2	81,5	A0A368ZTN1	ATP-dependent Lhr-like helicase
143	-6	5	2	2	69	A0A369A1L2	Methyl-accepting chemotaxis protein-2 (Aspartate sensor receptor)
144	-6	18	2	2	30	A0A369A363	Iron complex transport system ATP-binding protein
Cellular processes/metabolism							
12	-49,3	35	9	9	57,3	A0A369A0E1	60 kDa chaperonin (GroL)
16	-37,8	28	7	7	69,2	A0A369A6A6	Chaperone protein DnaK
17	-34,6	16	6	6	104,4	A0A368ZRN8	Glycine dehydrogenase (decarboxylating) (GcvP)
18	-31,3	8,8	5	8	136,9	A0A369ADW8	Filamentous hemagglutinin family protein
25	-23,8	13	5	6	49,1	A0A368ZSV5	D-glucarate dehydratase
27	-23,4	20	5	5	28,5	A0A369A880	Ankyrin repeat protein
28	-20,6	30	4	4	35,6	A0A369A2Y1	Glucose-binding protein /galactose-binding protein
30	-18,6	32	4	4	23,5	A0A368ZV41	Phosphoribosylglycinamide formyltransferase
31	-18,4	15	4	4	66,1	A0A369A099	DNA repair protein RadD
34	-18,2	31	4	4	35,8	A0A368ZR26	tRNA-dihydrouridine synthase B (DusB)

37	-17,7	9	4	4	129,5	A0A368ZQG7	RecBCD enzyme subunit RecC
42	-13,7	33	2	2	12,3	A0A368ZM57	50S ribosomal protein L7/L12 (RpL)
43	-13,5	24	3	3	44	A0A368ZRP9	2-octaprenyl-3-methyl-6-methoxy-1,4-benzoquinol hydroxylase
44	-13	25	3	3	32	A0A369A6J4	Polyamine aminopropyltransferase
47	-12,8	6	3	3	97,3	A0A369A4I2	DNA topoisomerase 1 (TopA)
50	-12,7	9,8	3	3	102,5	A0A369A1L4	Bifunctional uridylyltransferase/uridylyl-removing enzyme (GlnD)
53	-12,3	14	3	3	36,4	A0A369AAR4	L-threonine aldolase
54	-12,1	44	3	3	24,4	A0A368ZND5	Ribulose-phosphate 3-epimerase
55	-12,1	16	3	3	58,2	A0A369A8D4	Nitrogenase molybdenum-iron protein beta chain
57	-11,9	25	3	3	32,6	A0A368ZS40	Carboxymethylenebutenolidase
59	-11,7	13	3	3	45,9	A0A368ZVD5	(S)-ureidoglycine-glyoxylate aminotransferase
60	-11,7	24	3	3	35,2	A0A368ZQ49	Thioesterase domain-containing protein
63	-11,5	5,4	3	3	163,6	A0A368ZQ29	Glutamate synthase (NADPH) large subunit
64	-11,4	11	3	3	62,1	A0A369AAV0	2-isopropylmalate synthase (LeuA)
66	-11,3	14	3	3	42,4	A0A369A395	Benzoyl-CoA reductase/2-hydroxyglutaryl-CoA dehydratase subunit BcrC/BadD/HgdB
67	-11,2	7,9	3	3	95	A0A369A557	Ion-translocating oxidoreductase complex subunit C (RnfC)
70	-8,2	10	2	3	47,3	A0A369A596	EAL and modified HD-GYP domain-containing signal transduction protein
72	-8	11	2	2	40,4	A0A368ZFE5	Glycerophosphoryl diester phosphodiesterase
74	-7,6	12	2	2	51,6	A0A368ZVD9	Exodeoxyribonuclease 7 large subunit
76	-7,5	10	2	3	57,4	A0A369ADR7	Apolipoprotein N-acyltransferase
78	-7,4	5,7	2	2	85,1	A0A369A9Y4	Formate C-acetyltransferase
79	-7,4	10	2	4	55,8	A0A369A4L1	Diguanylate cyclase (GGDEF)-like protein
83	-7,2	15	2	2	24,7	A0A369A181	GTP cyclohydrolase 1
85	-7,2	7,1	2	2	55,3	A0A368ZP1	Histidine ammonia-lyase
88	-7,1	9,8	2	2	49,4	A0A368ZT58	Alkaline phosphatase
90	-7,1	8,1	2	2	61,2	A0A368ZPU8	DNA repair protein RecN
92	-7	4,5	2	2	84,8	A0A369AAN4	Molybdopterin-dependent oxidoreductase alpha subunit
93	-7	19	2	2	40,6	A0A368ZRV2	Beta sliding clamp
94	-6,9	25	2	2	25,7	A0A369ABS0	NAD(P)-dependent dehydrogenase (Short-subunit alcohol dehydrogenase family)
96	-6,9	9,4	2	3	36	A0A369A1M6	UDP-3-O-(3-hydroxymyristoyl)glucosamine N-acyltransferase (LpxD)
98	-6,9	7,9	2	2	77,5	A0A368ZXD9	Elongation factor G (FusA)
99	-6,9	20	2	2	32,9	A0A368ZTC5	Fructokinase
101	-6,8	20	2	2	31	A0A369A5E5	Exodeoxyribonuclease III
103	-6,8	11	2	2	41,6	A0A368ZM59	Site-specific recombinase XerD
104	-6,8	13	2	2	34	A0A369AAK7	Biotin-dependent carboxylase-like uncharacterized protein
109	-6,7	5,3	2	2	67,4	A0A368ZTX0	1-deoxy-D-xylulose-5-phosphate synthase (Dxs)
110	-6,7	12	2	2	38,7	A0A368ZQJ9	Phospho-2-dehydro-3-deoxyheptonate aldolase
111	-6,7	6,4	2	2	87,1	A0A369A5B5	Phenylalanine-tRNA ligase beta subunit (PheT)
112	-6,7	16	2	3	32,6	A0A369A5C7	Phosphofructokinase
113	-6,7	9,3	2	2	44,9	A0A369ACS9	Peptidase T (PepT)
114	-6,7	26	2	2	17,7	A0A369A3Z0	2-amino-4-hydroxy-6-hydroxymethylidihydropteridine diphosphokinase
117	-6,6	16	2	2	17,1	A0A368ZTI6	Cys-tRNA(Pro)/Cys-tRNA(Cys) deacylase
120	-6,5	10	2	2	73,7	A0A368ZFF1	CheA signal transduction histidine kinase
122	-6,5	7	2	2	47	A0A369A3Q3	Chaperone SurA
123	-6,5	6,5	2	2	45,5	A0A369A4B1	Integrase
125	-6,5	16	2	2	23,6	A0A368ZRS4	CRP/FNR family cyclic AMP-dependent transcriptional regulator
127	-6,4	7,4	2	2	56,3	A0A368ZMX1	L-threonine dehydratase
129	-6,4	9,4	2	2	45	A0A368ZW89	Gamma-glutamyl phosphate reductase (ProA)
130	-6,3	15	2	2	42,2	A0A369A090	L-alanine-DL-glutamate epimerase-like enolase superfamily enzyme
131	-6,3	6,1	2	2	88,7	A0A369A174	Bifunctional aspartokinase/homoserine dehydrogenase
134	-6,2	8,4	2	2	77,3	A0A368ZUP5	Fatty acid oxidation complex subunit alpha (FadJ)
137	-6,2	16	2	2	32,5	A0A369A3X5	Carbamate kinase
Regulators							
36	-18,1	29	4	4	37,7	A0A368ZN54	LacI family transcriptional regulator
56	-12	15	3	3	59,8	A0A368ZT76	Transcriptional regulatory protein RtcR
81	-7,3	9,4	2	2	45,1	A0A369A599	Sigma-B regulation protein RsbU (Phosphoserine phosphatase)
118	-6,6	6,8	2	2	36,1	A0A369A9I3	LysR family cys regulon transcriptional activator
119	-6,6	26	2	2	20,8	A0A369A006	Glycine cleavage system transcriptional repressor
139	-6,1	17	2	2	24,4	A0A368ZXL2	TetR family transcriptional regulator
Non-bacterial proteins							
19	-30,9	19	6	6	62,1	K1C9_HUMAN	
89	-7,1	6,4	2	2	85,6	GELS_HUMAN	

Supplementary Table S5. Type VI secretion system-related genes among *Kosakonia* genus

Function ID	pfam12790	pfam06744	pfam05943	pfam05638	pfam04717	pfam05947	pfam05591	pfam06996	pfam06812	pfam05936
<i>Kosakonia</i> sp. KO774	1	1	1	1	1	1	1	1	1	1
<i>Kosakonia oryzae</i> KO348	1	1	2	1	3	1	2	1	2	2
<i>Kosakonia arachidis</i> Ah-143	2	1	3	4	3	2	3	2	2	2
<i>Kosakonia cowanii</i> Esp_Z	1	1	2	1	1	1	1	1	1	1
<i>Kosakonia cowanii</i> JCM 10956	1	1	1	1	1	2	1	1	2	1
<i>Kosakonia diazotrophica</i> S29	1	1	3	2	0	1	3	1	3	3
<i>Kosakonia oryzae</i> CGMCC 1.7012	3	2	3	2	2	3	3	3	4	3
<i>Kosakonia oryzae</i> D4	3	1	2	2	2	2	3	2	3	2
<i>Kosakonia oryzendophytica</i> REICA_082	1	1	1	1	2	1	1	1	1	1
<i>Kosakonia oryziphila</i> REICA_142	3	3	4	5	3	3	3	3	4	3
<i>Kosakonia radicinicans</i> DSM 16656	3	2	3	2	3	3	3	3	4	3
<i>Kosakonia radicinicans</i> UMEnt01/12	3	2	3	2	2	3	3	3	4	3
<i>Kosakonia radicinicans</i> YD4	3	2	3	2	1	3	3	3	4	3
<i>Kosakonia sacchari</i> CGMCC 1.12102	2	2	2	2	2	2	2	2	3	2
<i>Kosakonia sacchari</i> SP1	3	2	3	2	2	3	3	3	4	3
<i>Kosakonia sacchari</i> SP1	2	2	2	2	2	2	2	2	3	2

PFAM

pfam12790: Type VI secretion lipoprotein, VasD, EvfM, TssJ, VC_A0113

pfam06744: Type VI secretion protein lcmF C-terminal

pfam05943: Type VI secretion protein, EvpB/VC_A0108, tail sheath

pfam05638: Type VI secretion system effector, Hcp

pfam04717: Type VI secretion system, phage-baseplate injector

pfam05947: Type VI secretion system, TssF

pfam05591: Type VI secretion system, VipA, VC_A0107 or Hcp2

pfam06996: Type VI secretion, TssG

pfam06812: ImpA, N-terminal, type VI secretion system

pfam05936: Bacterial Type VI secretion, VC_A0110, EvfL, ImpJ, VasE

pfam12790: Type VI secretion lipoprotein, VasD, EvfM, TssJ, VC_A0113

pfam06744: Type VI secretion protein lcmF C-terminal

pfam05943: Type VI secretion protein, EvpB/VC_A0108, tail sheath

pfam05638: Type VI secretion system effector, Hcp

pfam04717: Type VI secretion system, phage-baseplate injector

pfam05947: Type VI secretion system, TssF

pfam05591: Type VI secretion system, VipA, VC_A0107 or Hcp2

pfam06996: Type VI secretion, TssG

pfam06812: ImpA, N-terminal, type VI secretion system

pfam05936: Bacterial Type VI secretion, VC_A0110, EvfL, ImpJ, VasE

Supplementary Table S6. Type VI secretion system-annotated proteins present in KO348

>2652308089 Ga0077644_106131 hypothetical protein [Kosakonia oryzae KO348 : Ga0077644_106]

MINKAWREELQELKRDGSACQFHDAEWGVIRLKLRYRGEFLFFQLNERAL
ICEVSARYSTLDKTSLKRWDDGSGVIGADEREALAKIARYRCLCWKDDLRL
IN

>2652308090 Ga0077644_106132 hypothetical protein [Kosakonia oryzae KO348 : Ga0077644_106]

MKIDKLIKRVMLHDGVETRFNQPVELICPRCQKPIEPDLYQSGDFARL
PEDIQAAVAARIKIVTFNPETFTRYSAPEGSLLCSAHSCEGGQKTLVIF
SYKEQQPARYIATLYALLVVSDDQ

>2652308091 Ga0077644_106133 hypothetical protein [Kosakonia oryzae KO348 : Ga0077644_106]

MRRIFPVFALLFAFSAQAAQTPLTENDFTVEINKQAITLGQDWRNLLTV
LGKQTRDFVGEVFPFGEENYKYRHFAGFDIYSANIDWQQRKSVDSYV
IGQITLHAPTLHTARGVAPSDAKQRVIEHYGAGETDNSDGEWIMYSLAH
KNIAFDVTGGKVRAINISTGND

>2652308092 Ga0077644_106134 hypothetical protein [Kosakonia oryzae KO348 : Ga0077644_106]

MHAIHSTLQRLYVVASFLSCCACAASATDFSGQWHGSESNSTLTKL
SQQNNKLTGSYCFITQRGNRIDCPDEQDNLGVEKNDTAVTFDSSFQ
KNGKATLVINGDKLAWHLTQPPEHGDYAPENYALVKETVHAGATTKIIR
TDNFMLAIRNCGAFTTPCDDLTYTGARNRDSQQISLHGKTRQDNENRVI
GAEFRNGDVLYLVLDYNPPKLVVTQAGKTLVNQAGSWLK

>2652308093 Ga0077644_106135 hypothetical protein [Kosakonia oryzae KO348 : Ga0077644_106]

MDLLTREEGEALLKFLSRALKNPSDIETLMTMAREHPSTIPMKGIYQY
DRMEKNTLSKAELDDLSTLMFFYGP

>2652308094 Ga0077644_106136 hypothetical protein [Kosakonia oryzae KO348 : Ga0077644_106]

MRNLFTGNVRLSILFICLFSFFSFDAAVIDINHDTLETVENITVDKQV
QASLKNVLGADYAAFAGNFDVYGEPRHTADGGFLVEGWLKDLYLENASAF
VIYPDGRLSAAVWVPAASVAHYKSNTGEKRIPDALQQWVSRFQDVSNTP
AITQTAETFVDFETPKFKIKVTVCGNGAHCDEATYYGVRKNDRAEVNL
HGFAVRKSCEQLICPVITYTFKNGTTTYLLSKIDNSLTVIQNNKILLDEK
GIWKAHE

>2652308095 Ga0077644_106137 hypothetical protein [Kosakonia oryzae KO348 : Ga0077644_106]

MAEVNVIHKEKGDFFILQSDDKYLICMIWPYNSMWVQKCFILDHAELSR
YSDFHEMVSALAKMRDNYDKYKHREVPVPEFKVR

>2652308096 Ga0077644_106138 hypothetical protein [Kosakonia oryzae KO348 : Ga0077644_106]

MSNALCDHIPFLNVNHYTIIPSFIFLLNIASRTAIVQISAWSFN

>2652308097 Ga0077644_106139 hypothetical protein [Kosakonia oryzae KO348 : Ga0077644_106]

VFSFCPIHSVARDATVSKKIHEALERNIIVDDIVELTIVQPDENMAKQHV
LVEQQLNELGVNNVIVLYDRDRVNKKMNDRVVINKLSSHLCNVCPICLS
QKLGLKISK

>2652308098 Ga0077644_106140 RHS repeat-associated core domain-containing protein [Kosakonia oryzae KO348 : Ga0077644_106]

MSDKHAARQGDIIHSSVFADITSIVAEGVAYAAIGSAVAFAAATAAPLL
GAGAAAASVAAIGSSCLLSGIVGGILANVAGITDDISNMANS LGDALFPP
SPAGKIVTGSANVLTNNKLAARAAGTLTPADTPPAEPQSPASFADYAGML
LAGAKHFGSEMWPQTVGSADAGTSPLEQDKVACKKHSGPQYLAQGSKSVF
INGQPAVRAKDKTTCCEGTVSDNVSPNVIIGGETLTVRDIKSGKLPGLAVA
MIASLIRGRPGKILKNMPCALASAGGMLADMAVNAIFGSPHPVHAATG
VKVLNDEQELDFSLPGRFPLRLQRSYNSLTSRAGLFGAGWSTVFD SYLVL
TGDEACWFDETGRELRFTLPSVDQAMYSISSEGVIIRNDNGDVAIADDDG
AVWRLF KPTRANPAILRLASLSDEYGNALETGWDEHGRLVRLHDAPCAID
VTFAYDDARFSQRVTSASHFDGEHHWPLMRWHYDARGQLATVTDASGIVT
REYRYNDGLMVWHRAAGGLESEYRWAMFDHWRVIENRTNTGDGCRFAYD
LDAGLTTVTHYDQTRQHYWNTQGLIVRFVDERGENWRFWVNDNEQLTRR

IDPLGNAMTFVYDEMGNRVQEIDADGNERATQWLENRALPAVITEPGGST
 TRFFYDPHFGLARTVDALGQSTVWHRDEFQVIEEVDAAAGNSRRMEYNDA
 GQVIRETDCSGLHTRYHYHPLGWLVAVQTADGEETRYHYDAAGRVPQLER
 AEGWLETLRWNEQGLPTEHEAADGSRSAFRYDNTGRLVATRNLHGEEIRR
 SWDSRGRLLVALHNENGEAYQFRWGADSLLEEGLDGVVSRYTYDACGRT
 LSRTFAAGHPEATHRFWSAAGQLLARSTPEGQTRWRWSAAGFPERISL
 HPALDENSWSAEAEQELNFTFDALGRVIGEQGENTLWGYDALGNRTSL
 QLPDRELKQFYYSGLHLSIALDNLPISEDFGRDTHREISRTQGLLTAR
 SDYDRLGRLHRRDVFSGNAQRPAPRRWSRRWDYDHRNNLVREERDDNPFN
 WYRWKYDDAGRLTQDGLTPGQEQWRWDAASNPDVTTVQHAVRHNRVTQL
 HGIRWQYDIHGRTVEKDNQTRWRYRYDGEHRLTEVISQPRDRNKPQVQV
 SFRYDPLGRRISKTRRQMRAGQPVGQSVTTHFVWDGFRLLQEIHDDVPLT
 YVYSEQGSFDPLARIDGITDPDVYWFHNQPNGTPERLTAEGELRWEGQN
 SAWGKLLHETPLRAPEYAQNLRMQGQYLDRETGLHYNLFRYDPCGRFT
 QQDPIGLAGGLNLYQYAPNAQGWDPWGLSNRKCSTNSKQGHSTSAIRN
 QSGTAVIHWHDNRSTTNRFGHYSVEIKLNGTSLHTHQAGAPGEHTMVTTR
 GFEWLPAAKKAEPVLPKADAEAIKYLGGKLEKDGPLYDLKTQSCVTHVCD
 VLRSGVDVPTPEGAQMKYLLKLLR

>2652308099 Ga0077644_106141 hypothetical protein [Kosakonia oryzae KO348 : Ga0077644_106]

MQYTLQEGSFLFPAGWQDTSMTMLRDEESGLSLIVSRGPIPDGSDFEKE
 FYRQWDVLRQMGDIAQSEFARILVGRDNKTRAVEVETVFRNGQQIWK
 QFAVQAPGAAVMIFTLRALRAFTDEDGERWDAIKHSLTLHE

>2652308100 Ga0077644_106142 hypothetical protein [Kosakonia oryzae KO348 : Ga0077644_106]

MTICKVRHLTKIIVMTFALFSSFYSCAEENMKVQSLWQFATQLKNTIGQ
 DVEELDAIIPGRFVRENPNIAERLKAEPFTIDGGIEIRNMEVRLDLHNP
 KVYIISYDVANADILLEDVRKTYPLKLDIVPRGRSKDETFSWITPLDEK
 GNGIGFGFPYAQPSYLNMTLRNFQD

>2652308101 Ga0077644_106143 type VI secretion system secreted protein VgrG [Kosakonia oryzae KO348 : Ga0077644_106]

MANRITATLPVGDLLFWKLSGREALSESFTLALTVLGTDARADRSKLLGQ
 SATINIPTQGTGTRYINGKITRVAVSAVELSGTRYAVYQLTVEPDLWPMK
 RDRNLRIFQGGTAPQIVKTLLEGEYQVNVEDKLTGSYRTWDYCVQYQESSL
 DFISRLMELEGIAYHFRHEADRHVLVLTDAATEHQPFSGYETIPYHQTPS
 GGSTDEEGISQWALEDSVTPGIYSLDDYDFRKPNAWLFQARQNPASPQPG
 SIDVYDWPGRFVEHGHGEYARIRQERWQVEHQIQGTATAVGVAPGNTF
 ALYNAPFFSDNGEYLTTEANYFFEENRYASGSDGETVHRIDFTVIPSSVV
 FRPAAVTAWPKTYGPQTAKVVGPGQGESIWDKYGRVKVKFHWDRLAKGDD
 TSSCWVRVSSAWAGQGGVQIPRVGDEVVIDFINGDPDRPIVTRGVYNE
 ASMPPWALPAAATQMGFLSRSKDGSVDNANALRFEDKAGEEQVWQAERN
 MDVHVKNDAASRSIGNSHYVRKNELYRVETNQTQAVKGGTEILTGGKGL
 DAVEQFILASGTQLRLVSGHSAIELNANGKINLIGKSFNFFVEEDGHIT
 TGGKHLNAPGKTAPTTAPGADHKGNINSAVQAKFSPQGNVQHAAPVAAA
 PAGAAPVTKYKAPPPLKGDYVFSNEKSKSQFMPFSDGVVKKINSSPKMQ
 SDLKKLMDDQWNISPNVPGGGSWTDTKNKVMVLDPEMADDNEAVMTLAH
 EVGHATSPYKNDFFSSKSNFVNGMLKDEGQATLNEIQVRREIYHNSGIDIG
 SMTDSSNEMKYIAFKDMDSGKITRDEASKAIGEYRRGEVAGSSTTNEV
 YEDHYGNMYDDYMQGKAPH

>2652308102 Ga0077644_106144 protein of unknown function (DUF4150) [Kosakonia oryzae KO348 : Ga0077644_106]

MFANCQLMGVDLAFPDVCLTPMPAPTPIPYPDIALGPTAIPNALNILFMG
 MPAHNMATITPLTNGDNPVATGVASGTVMGSPSRHLTGAFTVLLKGT PAT
 RLTSVSLQNSTNAIGMRIVPSQFKVLM LAP

>2652308103 Ga0077644_106145 Protein of unknown function (DUF3540) [Kosakonia oryzae KO348 : Ga0077644_106]

MNNLNQPLTLATLPGGQFSARVTHCFDDGSLMVECDGRGWHCRAVSCVI
 APQAGDVTLISAVDNQMWLLAVLERGNEDATELSVPGDLRITSQGALILS
 SDALNVSAAGDCHISEMNYS GDKISAWVTL SRIVGKRAESVWQTVTQMS
 QHLFRTRQTEHVVRAGQLDKAEDYLRMHAQNTVITSKAITKVDSEIHM
 G

>2652308104 Ga0077644_106146 Uncharacterized protein Yjbl, contains pentapeptide repeats [Kosakonia oryzae KO348 : Ga0077644_106]

MSQLSAAELQKKVKSGEAIMELNLDGCDLRGCDLSSGGIFQEVSFEGASLQ
GCNLQESVFTTECQLAGAVLTAHLEETVFNQCDDVAANFSNTSLLRCVFN
ECTLNGCDFTQSADFSTQFMRSPLNKSFLTGARLERSTLFECPDLAGAKLN
HCHNLLTTYGIDLRDLDLSSGSQFERAVFFNCDQRGKNYAQHQTGCQFT
DNQLDGDADFSGAQLTQC�FKGASLKQARLNNVNATQALFMQADLSGAHAH
GSLFDQAIFVGATLQQASFQSRFFQSILQHVAARQVDFTLCDFTYADFS
GAEVCEADFRGAIFSRFRHRARQEGARFADRKGILEYDEELLAEEAWTA
ERHSRIYGDLDQ

>2652308105 Ga0077644_106147 Uncharacterized protein Yjbl, contains pentapeptide repeats [Kosakonia oryzae KO348 : Ga0077644_106]

MKIIKPLRLSVLNRPFLRQGNHLGVSVLALLDMSAQPKLRPEVELWQLA
ASELQTSGGVLDMAMPKARAEFLAIGFAYSHHHQDKTACAVRIEIGELSK
TLAVTGDRYWAGSRPTAAKPFQMRDLWSRAFGGEGFEENPHGIGAVEEN
HNGTRFRRLPNIELLNQRVTSAPRAKPEPASFGPLDLLWPRRFRRMGKNYD
ARWLQHDFPGFAPDIDWRVFNAAASPDQWWEERDALPPQAAWRIWNMHPEQ
HLQEGTLPPWQARCFMQRQRGDEILFEEIALRATTWFFPHLEQMVLWQ
GNQRINEDDAADVQLMPALEKIGAPRSVNHYRKVLHQRLDKEKGALFAF
REKDLIPEEVIGPWIDSEVQENHSPMRDNQQNRAMQLREQHRARLEAQA
DTADLLQEMEEPALPKLEDLPEFIEEMERKARQMQAQAE­TRKAEMEAREFP
QMNAQENQPRGPESMMRMQD­LLERNADSMSEK­K­K­K­SREALH­K­L­Y­L­M­S­A­A
EQPPAIKLTGDIALIIRQRAERTMAQGGDFSGLDLTGADFSGMDLRGANF
RNALLECANLSQCQLDGADFSNAMLARTDLQGASLCECNFTEASLALAQC
HQTNFTGAQFTESEMTDALFDACDFSHARLEKLLLRKTGFSCVFRHATL
DNCVFMELTLPQPDFSAATMNKSSFIQCDLQSASFAGAHLEGCSWVESRL
ERALFRDATLITCAVASGGTLCGADFSGAQLKQSNLRQAVLTDARFVRAK
LDNSDLSEAQCEGADFSGATLTGSLFMRTDFRRVRF­TDANLMGAMM­Q­K­S­R
LEGADLSFSNLF­RADLSQSIADSTTKFEGAYN­K­R­V­K­T­L­P­K­R­D­G­E­V­I

>2652308106 Ga0077644_106148 hypothetical protein [Kosakonia oryzae KO348 : Ga0077644_106]

MDYFKYLAYLFP­SL­V­I­V­V­F­I­K­I­A­I­R­D­I­K­N­N­R­I­K­K­R­I­A­S­D­P­V­H­V­N­A­R­I­T­Q­A
VAGTPAPNGIVN­V­T­L­D­Y­E­F­N­D­H­T­G­K­V­F­T­Q­Q­N­V­V­T­V­V­K­T­M­E­M­L­N­Y­K­A­G­E­T­V
PVIYLRSDPSLN­K­V­N­L­P­R­V­F

>2652308107 Ga0077644_106149 hypothetical protein [Kosakonia oryzae KO348 : Ga0077644_106]

MTDLIAKNL­T­L­F­S­W­S­I­M­A­I­F­I­Y­L­F­I­K­L­R­E­P­T­D­D­A­K­L­I­Q­Y­R­A­T­L­R­D­W­Y­G­S
EK­E­G­V­L­E­T­W­A­P­L­K­S­T­P­K­G­E­Y­F­L­F­H­F­I­I­V­I­D­R­K­T­Q­K­K­S­S­G­F­N

>2652308108 Ga0077644_106150 hypothetical protein [Kosakonia oryzae KO348 : Ga0077644_106]

MMGILLAIGIIAV­V­V­I­M­C­F­I­S­V­N­K­T­E­K­I­R­E­G­R­P­I­M­A­V­I­E­N­I­R­P­V­S­T­D­D­S
GNTTVSYV­L­N­V­E­G­R­K­I­E­G­R­E­K­I­D­T­F­Y­A­P­Q­M­Q­P­G­M­H­I­K­I­M­Y­V­D­D­K­H­F­V­I­F
EK

>2652308109 Ga0077644_106151 hypothetical protein [Kosakonia oryzae KO348 : Ga0077644_106]

MFTGDNVGL­L­I­S­I­V­A­P­C­F­L­I­Y­L­V­F­H­T­G­M­V­H­D­D­F­K­K­N­G­I­R­T­V­A­K­I­N­N­I­K­I
STSGTGSP­K­C­V­F­T­L­S­F­T­T­Q­D­G­H­D­I­S­L­E­K­T­Q­V­V­T­V­L­D­M­M­P­L­E­R­E­R­K­V­D­I­Y­Y
KKENPKKI­W­L­I­L­E­S­R­I­K

>2652308110 Ga0077644_106152 hypothetical protein [Kosakonia oryzae KO348 : Ga0077644_106]

VNLYCHHTI­H­E­S­K­F­L­L­L­V­Q­F­F­I­Y­I­T­V­N­M­S­V­F­T­E­T­P­L­I­G­V­I­V­S­I­L­G­I­L­A
FSTLKPN­K­D­E­V­T­T­L­R­D­W­S­S­R­E­K­W­S­G­Q­I­I­E­T­S­L­E­S­W­H­Q­T­D­T­K­Y­G­N­D­F­L­Y­D­F
TFTATIND­T­R­K­K­Y­V­A­K­G­L­V­R­P­N­E­I­H­K­I­Q­K­G­L­T­L­I­I­K­N­A­D­N­P­P­R­I­A­V­M­A­I
NYK

>2652308111 Ga0077644_106153 hypothetical protein [Kosakonia oryzae KO348 : Ga0077644_106]

MDYLKVL­H­I­I­L­V­G­L­V­A­G­L­I­Y­L­I­C­R­D­I­V­G­V­M­K­I­L­R­N­P­V­Y­T­H­A­T­I­S­A­V
MPGTPSS­N­G­L­V­N­L­T­I­D­Y­E­F­K­D­L­T­G­A­N­Y­S­R­K­N­V­L­T­I­I­K­T­I­N­L­V­E­Y­Q­I­G­S­A­V
PVVYLR­T­N­P­E­K­H­F­L­N­E­G­K­D­G­V­L­I­R

>2652308112 Ga0077644_106154 hypothetical protein [Kosakonia oryzae KO348 : Ga0077644_106]

MSYLSY­I­M­P­K­L­A­W­I­P­L­C­I­L­L­F­V­G­Y­V­V­Y­K­N­R­Q­R­D­A­Q­D­A­Y­I­K­E­H­G­V­S­L­D­A­E
ISDVVY­D­K­I­Q­R­I­N­N­Y­F­V­V­A­S­V­K­Y­N­Y­S­D­K­I­F­V­S­K­R­G­F­S­F­L­I­T­E­K­E­K­I­I­P­G

QIIKIRVNRNMPEQFYEDYQNY

>2652308113 Ga0077644_106155 hypothetical protein [Kosakonia oryzae KO348 : Ga0077644_106]

MDYLLLIAMAAVFLIIYKGTARHDRLMRDGRPVMATIENVTPVSSDDAG
NTTIVYTLNIEGRHVKGKEKIDTFFSPQFQPMQIKIMYINDEDYMFVFK
K

>2652308114 Ga0077644_106156 hypothetical protein [Kosakonia oryzae KO348 : Ga0077644_106]

MAISYFVTYPLLVFFGMFVFFVYNIIMAGRGAVKIDPTDAGQAQGEIVN
IRSSSGENSFAFINVIIQVRFITADHKIVNTEGKAVIDVVKIPEYQKGVKV
PVTYSKKEPENIKINIPSPLDK

>2652308115 Ga0077644_106157 Protein of unknown function (DUF3592) [Kosakonia oryzae KO348 : Ga0077644_106]

MAAIVFDSPVFIISLIFIVCIGILLYCYTGVVHDKFKKEGVRTEAKVLS
KEKIGASGTGNTRFRMVVEFTTKNETVTVTTKRYFTPEDLIKIMRNNTVV
LYYLPQDPQQVLLMPGEMK

>2652308116 Ga0077644_106158 hypothetical protein [Kosakonia oryzae KO348 : Ga0077644_106]

MSDSTMWSIVIVVLIIAWLIYGIMKAGYDDDRFRAKGIKVEAKILDKKNI
GVSGTGNVFKVKVEFETKDGVVRAQAKRYFTPEDLIKVMRKNTVQLFYL
PENPQQIYLVPDME

>2652308117 Ga0077644_106159 hypothetical protein [Kosakonia oryzae KO348 : Ga0077644_106]

MGIEYYSKYFLYGGIAFSLLSVVVFYFMFRGDKTDVAGMKNYLSSSQWQG
EFIDSRIESWQQTNARYGNDFYDISFRLNDEKLYTAQTLVVRPSQMHLMK
EGLKIKVKKGSKNRLAVVEVNFEDN

>2652308118 Ga0077644_106160 type VI secretion system secreted protein VgrG [Kosakonia oryzae KO348 : Ga0077644_106]

MQNRITATLPVGGLLFWKLAGREALSESFTLALTVLGTDARADRSKLLGQ
SATINIPTQGTGTRYINGKITRVAVSAVELSGTRYAVYQLTVEPDLWPMK
RDRNLRIFQGGTAPQIVKTLLEGEYQVNVEDKLTGSYRTWDYCVQYQESSL
DFISRLMELEGIAYHFRHEADRHVLVLTDAATEHQPFSGYETIPYHQTPS
GGSTDEEGISQWALEDSVTPGIYSLDDYDFRKPNAWLFQARQNPASPQPG
SIDVYDWPGRFVEHGHGEYARIRQERWQVEHQIQGTATAVGVAPGNTF
ALYNAPFFSDNGEYLTTEANYFFEENRYASGSDGETVHRIDFTVIPSSVV
FRPAAVTAWPKTYGPQTAKVVGPGGESIWDKYGRVKVFWHWRDLAKGDD
TSSCWVRVSSAWAGQGFGGVQIPRVGDEVVIDFINGDPDRPIVTGRVYNE
SSMPPWTLPPDDSTRMGFMTRSKDGSKDNASYLFFEDRAGSEAVELHSEKD
MKVSVENDKTVNIDGNRTTILKEQKDDVTGDASFYRAKRTTTVDEAET
TTFNNSQTETIKNGRTLNITSGGDVVTVKEGRITEVEGTESHHTVGLVTE
KFDGSGQMTTIEGGLTVDVNSGNWTQNVNGGTITISSPNMIRISSKEQIVM
DAPEAVFKPKFHTLSVTAFFSESVGHASGTGMTSSATGIALSANGMSIG
FKLKDLSKTLFKKEANGVVINYEVNINISNRTLNITNANALYIFT

>2652308119 Ga0077644_106161 hypothetical protein [Kosakonia oryzae KO348 : Ga0077644_106]

MNLLTHSNQGGGEFTFVVSRSNAKADDKVHALAARKTRELEMTLIDFHLE
SSEMLEVDGYPVELFYQFKNDNHVIFQRQTLILLDDPAGKMMVSYVGTG
PGEFSEEHQKQYQQIIQSIKFHRAK

>2652308120 Ga0077644_106162 Serine/threonine protein kinase [Kosakonia oryzae KO348 : Ga0077644_106]

MTDHDNNRSVPNALPPGYRFNEFEILEVIGGGGFGIVYRAKDHQLERTIA
IKEFMPSSLAVRNDMMTLVLRSERFSKAFAGLNSFIQEARLLARFNHPN
LLHVLRFVWQNDTAYMGTVFYSGTTLRSLREKNPTLINEAWIRRMPLMLL
GAIKTIHDEGYLHRDISLDNIQIQDNGLPVLLDFGSARRSIGSVSDETET
MLRPGYAPIEQYTDNNESEQGPWTDIYALGAVLHHTLIIGSPPPVSVVRSI
QDTYVPLTQRGLPGYSHTLLQAVDRALSLEKMDRPTVDEFAALIEPVA
GIDDMVSVKQPGTMLVPEETTEKTDALSSVKRYKVPGLVAAAGVIVGVIA
GAVLFGGNSSSDAAATQNASNSDQQQTQQAAEPPRQSEQETTAPAATP
SQTQTQAQSQTTAPAVQEPVAQIFVRMNEGEKLTNGEAQSVSPATNGF
ASLKLPGRYNLVLQGNQTRSQTITIGQPGTWLINPQAQ

>2652308121 Ga0077644_106163 type VI secretion system protein VasG [Kosakonia oryzae KO348 : Ga0077644_106]

MSEISRAVLFGKLDTLFSLSESATAFCKLRGNPYVELVHWHQLMQQTD
 GDLLQVIRHFSLDEEALTRDIVAALDKLPRGASSVSDLSEHIDTAVERAW
 VYGS�KYGVTRIRGGHLLAGILKTWSLANVLKGISPQFERISADALLDNF
 DAIFANSKESQQVTVLND DAGAGVPQQQGT LAQYGGDLTARARDGKIDP
 VVGRDEEIRQMVDILMRRRQNNPLLTGEAGVGKTAVVEGLALRIAAGDVP
 EPLVDVQLWLLDIGMLQAGAGMKGEFEARLQSLINEVQSSPTPIILFIDE
 IHTLIGAGGQGTGDAANLLKPALARGQLRTIGATTWAEYKKYIEKDPAL
 TRRFQTVQVAEPDEEKAVLMLRSTVSALEKHHRVLLLDEAVVAVKLSHR
 YIPARQLPDKAVALLDTACARVAVSQSSPPPQLEDCLHRIAALDVEVEIA
 NREAKMATGESDRVEKLQVELEKLAQQRDELTAWEQEALVDIAIALRA
 QLHTSPEEAQADIRATLTQQQAE LRALQGDAPLLFTSVDANVVAAVVSDW
 TGIPLGRMVKNEIDAVLKLADTLNERVIGQRHGLELIAKRVQTSRRLDD
 PNKPVGVMFCGSPGVGKTETALALAESLYGGEQNVITINMSEFQEAHTV
 STLKGAPPYGVYGEVVTEAVRRRYPYVLLDEIEKAHPDVHEIFFQV
 FDKGWMEDEGRHIDFRNTIIILTSNVTGTELITGMCADPELMPEPDALRD
 ALRPPLLQVFPALLGRLLVVPYPLSDEMLAMIVRLQLKRIQRRLADNH
 GIVSEVDDSVVEQIVARCTEVESGGRMVDAILTNTLLPLMSQLLLDASAR
 DEQYKRLRVTFEQGEFHCQFAA

>2652308122 Ga0077644_106164 type VI secretion system protein ImpH [Kosakonia oryzae KO348 : Ga0077644_106]

MTETLSSAPVITRASALPEAFWQNMATPWRYDLFTLLRRVDARGGERYP
 LGRAPLPRFEPLRIGQTPSLGFAPSTLSSVRQRENSLYDVSILSFGFLG
 PNGPLPVHLTEYASERIAHHQDDSLSAFADLFHHRLLALLFYRAWADAQPT
 VSLDRQDNKRFEYIASLIGMGQPGQLEKGSLSPHARFALAGHLTRNGRD
 PEGLAKILRSYFNVPTIVENVPQWMPLESERERARLQGGRHAPRLGQSAF
 LGEAVRDVCHKFRIEIGPLNVDYRRFMPGEKVVWALRDWVRQYLGIEYQ
 WAVKVLRSQVAVGATLGGAGRLGYSAWLGTQPRPQARGDLVFSPEG

>2652308123 Ga0077644_106165 type VI secretion system protein ImpG [Kosakonia oryzae KO348 : Ga0077644_106]

MDSKLLYYNRELAWLREMGQEFYAARYPKVAGRLGMRGMDVSDPYVERLM
 EGFAFLTSRVQLKMDAEFFPRFSQRLLMVAPNYLAPTSPMAIAELQPDSA
 KGDLSNGFVVPRGTMMDSQVMKNGVTCSYTTAHDVTLPLKISQVELGG
 VPADLPLAQVGLSQRGAQSALRIRLSCDGPVNLSHLDFDRLEFFLSGPD
 QALKLLELVMEHQVILCQANGQKTPPLVLADDALRQEGFSADQALLPDD
 LRNFDGYRLLQEYFAPSRFLFISLHGLRMLAQSGEAKSFDIILLDKA
 DAQLERVVDKSHLALHCTPVINLFPKVAERQKLSDSLHEYHLVVDNIRPL
 DYEIYAVTKIHASIDGQRDEETFRPFWSSWSQDEGNYGAYFSLRREQRAL
 SEHAQRYGTRTYIGSEVFASLVDEQHAPWREELRYITAIEVLCTSRDLPL
 MLQQEIGQFVLPDLSLVKTLQLRKGPTPPRPAEGLSTWRLISQLQMN
 YLSLMDGEDGEGAAALRQLGLYTRLAEPVARQIEGVRHCVLEPVHRRVP
 EPGPIVFARGIGITLVDEQAFSGFSPYLFVGLERVVFAVLVGMNSFTEF
 TLKSQQRGEVGYWPPRMGKRALI

>2652308124 Ga0077644_106166 type VI secretion system protein ImpF [Kosakonia oryzae KO348 : Ga0077644_106]

MSNSAHDEESDLLRSGWRSRRGKDTV GARDKMQPSLLDRLTDDAPDKVQE
 PVNNNLVSHSALRRHVLRLDLQWLFNTINNEAQQDLSGFDQVRRSVNFGV
 SPLAGKRMSDIEWQDIQRKLTDAILHFEPRIPLQGLQVRCISDTKSLDLH
 NVLSIEIKGRLWCVPYPLEFLFRDLDLENGHFELKDAG

>2652308125 Ga0077644_106167 type VI secretion system protein ImpE [Kosakonia oryzae KO348 : Ga0077644_106]

MNTLFQQLAGESLRESLAQLESRIQTQPGDADLRAAFAQLLCDGNWSRA
 LAQLKSWQALKPQAQPTVTLLEQAIEGERQRADVMAGRARPVTPDQQWPW
 LASMVSALEPEANASADREAALEMADANPGQLTTQDQTLNFDWLMGDG
 CRFGPVCEAIVNGRYFWLFPFSAISAMQFPASVTDLVWRHTLVRLQDGS
 EQVCQIPARYPLDANADDRFKLRCRVTEWQPLPGDAPHYIGQGQKVVWLNDS
 AEYSLDLATVSFNVEAADE

>2652308126 Ga0077644_106168 hypothetical protein [Kosakonia oryzae KO348 : Ga0077644_106]

MKLWPLGLALLAVCGSVQAENYRIVQSPSQKLDVWIDNVADNTPKSWCAK
 TLPLRIVASGDKKPSVLSFLPRLGALLENQCGTLTQVRWKLTDPPQGTTL
 AEGTADKAKAWDPVVTSTGATTAPAATPSAGLVAPTGRAEDLSPPASRAP
 WQEFTLQDGCRLRTFWQGGAGTALFIPAKEDGKCEKGGWLNDRSVVTQL

SNGVEKKITMTFVHGFPVSGLNASVDADRLLITTVNNERMVVSEGTLAQS
 WMILPYIDSLNGWQANGTVAVEISRVDANDPARLQARIEEVRKAWMPWFE
 PGTHLNILLIDSLHPQLRNPVAVGTYKTVN

>2652308127 Ga0077644_106169 Serine/threonine protein phosphatase PrpC [Kosakonia oryzae KO348 : Ga0077644_106]

MNISTASISRQGERASNQDQTGETIGERAACFVVCDCIAGLPGGDVAAKL
 ARNAIITRFDGDEHLNAQYIRQYVNGANRAIREEQKAVQDYHRMGTTMVS
 LFIDRDYQLAYWAHAGDSRLYLFRRGWLYHVTTDHSLVQQMKDAGHQTEG
 INGNLLYFALGLGEDEREPSYSDVVPIDGDAFLCTDGFWHGVTEEQMQ
 QSLHMVNTPDDEWLTLMNQILLKNGVGAQTQQDNYSAAVWVMGTPQETLL
 HTLSEAAQFFPLRD

>2652308128 Ga0077644_106170 FHA domain protein [Kosakonia oryzae KO348 : Ga0077644_106]

MRFTIITSKPGHQPPQSSCDFYPPGGTIGRGTDNNLVLPDNDRSISRLQA
 IVHIANGECRVTNRGNVTRVVLNDIPLERGRQVELQDGDILGIDEYRIE
 VTDLIQDTRPVTRMAEEMYPKPAQPQVAKPAPAPAKNAPEGAPASVPTE
 IWDSLMEQFSISDSISSRAKPQEAQNLNPFAPKGPERNPEDPLSLLNN
 NEPLVTPKSLASDQLFNDEQLFKNDSIFNDSTPSALVPPVEPTQKSHAPD
 SDELPLALFGGSGSTKQTRSDDPLGLLSGAVPLAHADELAAQKPAEPQP
 IITDLNTPPEPPTAQPIITELRPEDLSSTPLFADDAPLSAPEEVQEQQDY
 AGITLPTPQAVQRSAAQTPKGRLRIDPVQSNGTKSTPSVSTGDSGDVLKG
 ELLDALLEGMLSDMQPVPQFDKENMRQFGQMLSMFSQGTVALSSRSIL
 KRGVKADMTMVLDDANNPFKLLPSGKTVLMQMFQTRMPGFMPKKSVRDA
 LIDLQAHQLGMISGIRAIIAAMLQQFNPEQLEDDAKRDGATARLGLVLSNR
 KAALWDYVVRTYAQTAGEIDDDFHTLFGEAFLHAYDMEVNQYKDSQSGSE
 E

>2652308129 Ga0077644_106171 Protein of unknown function (DUF1311) [Kosakonia oryzae KO348 : Ga0077644_106]

MTGSIIIRFTPARVKVALAALVSLSHCAASAAEAPASISGPWRIIANSTD
 TQATISFYTKVNDPSYVGRVRFDNHAVSGDAALNIDCQQPAYLQQSPMT
 LNEAIVKTSGERFFPKIPVAEDFGLNGQGMQKITPIVLQCCQGHLPDG
 ESIGNWVALLSADKLLMYGNDNSYFVLKRVTADEKITPRFSCNAKLSATE
 QAICGDNELAAWDRSVTDAYTIQLQQQEQEIDPADKATLAGMKAQRDWLK
 KRNQCQTDAACLTKSMQERTFELVSKIQ

>2652308130 Ga0077644_106172 Protein of unknown function (DUF1311) [Kosakonia oryzae KO348 : Ga0077644_106]

MTTHTKQIVAATMTCLALFSSAVQAGNSPVQGEWQVEKAFINTETERTLN
 YQFNDDRLLVGRFLSVTPQGISTTLPGGSNQCQSPAMKESSTLDAWVAATQ
 SIPEKDAAKTYELGLDGSQAKTQVENITCASGHFANGDAGSDASLAFVNQR
 LLLNWTQGTILLKLPVDKNSKQASFDCAKAASAPEKAICGDRELAALDN
 SVARSYKAFRKEAASLGNDLENKLSQSQKAWLSQRNSCNGDVQCLKKS
 MNRLETLAHSLDGV

>2652308131 Ga0077644_106173 Phage lysozyme [Kosakonia oryzae KO348 : Ga0077644_106]

MSVMRKGDRGTNVKELQQLLNKSGAKVGEDGIFGLKTEMAVKQFQAKQNL
 HVDGIVGRRTLAALGKPVTTTRAPQPPISGSAGRQAVGAMDISASGMTFIF
 HREAWANKSCYLHWPGGASGVTLGPGYDMKERSEQSIKAKMIEIGIDAIT
 AEKISKAARLHDDQASQFVADNAKLVRLTADQETNLLRATVPPYVNAVRN
 GIFVPLKQYEFDALVSFAYNPGGRLNNVFGFINRGQISDAMTEIKRANTS
 KHKVMKGLINRRNFEVNLFLNGDYGTN

>2652308132 Ga0077644_106174 type VI secretion system secreted protein Hcp [Kosakonia oryzae KO348 : Ga0077644_106]

MAIDMFLKVDGVTGESKDSNHTGWTDITSFSWASQPGNMSVGGGGGAGK
 VNFNDLHVNALIDKSTTAILKHKASGKHLTKVELSVCKAGGQVEYARIT
 LEDVLVTSVQYTGADNGDVTGVTYAFQAAKVKQYWEQSSSGGKAETTA
 GWNIKENKEA

>2652308133 Ga0077644_106175 Type VI secretion system (T6SS), amidase effector protein 4 [Kosakonia oryzae KO348 : Ga0077644_106]

MRPLYEQLKRYHRSSLYQPGFLSPEELFKEIGYDYQALKASNPNYENTC
 GVRMSLALLKNNIDFTGRFIKIDGPKGKIEPGAKLLADQLYKDSVFGK
 AEVYTDIHEAGRKLNRNRKGVVYFHRIAGYGGGHIDLEPLSNMFMQCN
 SNGCHTDSKEVWFWEK

>2652308134 Ga0077644_106176 hypothetical protein [Kosakonia oryzae KO348 : Ga0077644_106]

MKMRIGLAGFLVMFSTMISAETKTYSPQASPLPEDSVLNLMRDPALLNG
FDVNMSAQRFADAWFSKTNERERIKADMYLLGVLDTTTEGKTWCGYNRLLP
SSIHENLYSYFENLTAEKGLRASKVISDAMTELMPCCKGNNK

>2652308135 Ga0077644_106177 type VI secretion system protein ImpC [Kosakonia oryzae KO348 : Ga0077644_106]

MSNPSQQQELQQGAQAFSQDEFSALLSKEFRPKTDQARSAVESAVKTLAQ
QALENTVTFSSDYRTIQNLIAGIDEKLSQQINQIIHHEDFQKLESAWRG
LSHLVNNNTETDEMLKIRFMSISKQELGRNLKRYKGVGWDQSPFLFKIYEE
EYGQFGGEPFGCLVGDYDFHSPQDVELLGEMARIGAAAHCPFITGTAPS
VMQMESWQELANPRDLTKIFQNTTEYAAWRSLRESEDARYLGLVMPRFSLR
LPYGIRTNPVDSDFEETDGANHNNSWANAAAYAMATNINRSFKEYGWC
TSIRGVESGGAVENLPCHTFPSDDGGVDMKCPTTEIAISDRREAELAKNGF
MPLIHRKNSDFAAFIGAQLKQKPMEYHDADATANARLASRLPYLFACCR
AHYLCIVRDKIGSFRRERDEMERWLNWVMMNYVDGDPANSSQETKARKPL
AAAEVQVQIEIDNPGYAAKFFLRPHYQLEGLTVSLRLVSKLPSLKTKEA

>2652308136 Ga0077644_106178 type VI secretion system protein ImpB [Kosakonia oryzae KO348 : Ga0077644_106]

MAISNSGQKFIARNRAPRVQIEYDVEVYGAERKIQLPFVMGMADLVGKP
VENLPSIEDRKFLEIDVDFDERMKALKPRVAFNVDNLTGEGRLNVDLT
FDSMDDFLPDAVARKVEPLNKLLEARTQLSNLLTYMDGKNGAEELIAKVL
QDPTLLKSLSQLPNSEDSAQGKEE

>2652308137 Ga0077644_106179 type VI secretion system protein ImpA [Kosakonia oryzae KO348 : Ga0077644_106]

MTIESLLAPVSPEQPCGENLEYDADFQAMEQASLGKAEQQFGSTIIPAEP
ADWTRVEKLATGLLARTKDIRVMMALTHAWTRRRRGLEGYADGLMMLGQAL
ALYWDQLWPSLTDGGEFDPFYRINALAGLSDKSSLTTLRQSTLLRSNGD
ELNVRDAQALLDGSKTECAGYPGGRVRLIDELTRGGQPGIEAICQIEGRL
QTIRTWLLQLGESGVPMEQLLKTVGLIAGVSRANRTEEQQATEQTAPA
DSAPQPVVATPLAAHTDWRTAQVTRADAQLMLEKVKQYFTQHEPSPHAP
LMIDRVQRILIEIDFMEIIRDLPDGVNQLQNIIFGRQD

>2652308138 Ga0077644_106180 type VI secretion system protein ImpM [Kosakonia oryzae KO348 : Ga0077644_106]

MTTSAISWYGLPSAGDFLQRRFPDALQRQWSHWFQVGLMNNWQKEEQRT
NDRQFSNAPIWNFVPPMLGGQQVQMGCLLPGRDSVGRQYPLCALMAINP
LEWSPRHLAKAGDWYEQLGRTMLHAVRNNGFSPEQLDQALLSIPPVQLVEP
ETRSEILDVIGYDGDGESTIGWRQAECFDPLRQISFWWTNRSDGYPLYT
HVHSGNFTGQLFTLLFEPAGGARPGRHGLYPPMFEE

>2652308139 Ga0077644_106181 type VI secretion system protein ImpL [Kosakonia oryzae KO348 : Ga0077644_106]

MLLSLLTNRILWGFLGVTGLAAVIWMIGPLLSVVDSPLESEQNRMITIA
VMYLIWVHSHIVPRLYNLNRKLMNDNLKSEEAKDPVERKRLNNDQVLN
ERFEEAAQVLKKAHFNQPGQRGQWAQRFSTQYLYQLPWYVVIIGAPGSGKT
TALVNSGLQFPLADRFKGTALRGIGGTRNCDWWTNEAVLLDTAGRYTTQ
ESEQVQDASEWLKFLGLLQKYRRRQPINGVIVTVSIADLLTQSAEASREQ
ALNLRQRTELHEQLGIRFPVYVLTAKDLLKGFRAFYAGFDKAQRDQIW
GFTFPWEKAKLADFDLQGSFIQEFALLQQRDAALPDLLRESDAQARAE
CFLFPQEFAALRPLADYLNIFARSNFETEFSPRGIYFASGTQEGLPFD
RVMGELNRLSLPQKSGDNWDSVSKEEPVPGGKQGSFFIKHLLQNVIFQ
EAGIAGQNRWWELRSRAVIWVSGYAALLALLIIISALWFTSYGNRRDYLDE
VQTKVPALDQKIKALRNRQQGDLFALLPLNGLSALPESEKFDVNNPPIT
RRMGLYRGNDVADASQTLYQKALQMLLPEVAMRITTVLNRNDNGSDVEYS
YEALKAYQMLYQPKHYDGKFLHSWVMLNLQRNLQNVTKAQLSQLEWHLT
QLLEPQIQSSPYAKDEALISREQALINQQPLSTRVYGRKRLLERDENLK
QVSLASLGGPQSELVFSRKSQKPVGEGLPGLYTPDGYWNSFNAQIAPVTA
SLHEDDVWVLGSTTQAEDKQQTDAVNRQLYARDFIAIWDNFLSDIQLNNS
ADLNQRINTARLLSGNNSPLRRLVINLSQQLKLSRDDVADGKEKAPESSN
RGTQMLETLFSNHGASATANAASQTPEQRVTEHFAPIIELAQPLEKGG
KTIVFDDFLKQVDELYRYLTAVQDAANSQMPGPPGGEAISRLQASAGRLPG
GLQTMFSNMAVGASDQRRDMENVRKRISVEVGSFCRQAIAGRYPLVRS
ASSEVTPDDLARMFAPGTGLMDVFFRDNLTKVDTTQATWRFMPGIDGKT

LPGSEGLVLPFQQAQSIRDAFFANGSATPSFRTTVRTVRMDNTILNMTLD
 VDGQILRYSHGPQAVQVSWPGSGGTNQVRMQLGLANGTTATLVTNGAWA
 LNRFFDKARVSPGSSSLSRQATFTVDGHQVTFLEFAPNSIRNPFQLPRFAC
 P

>2652308140 Ga0077644_106182 type VI secretion system protein ImpK [Kosakonia oryzae KO348 : Ga0077644_106]

MQEQQASGSDAALAGASGNNPLVAAANPLLNAIPQIRYSVSHDDQSGLRQ
 HLIDEIRRFVRCQQSGLAYEVIVGARYCLCTALDEAAAALTPWGSRGVWS
 GSGLLVTFHNETWGGEKFFQLLARLSQNPRESHIALLELINFCLLLGFEGR
 YRVMDNGRTQLETIKQRLWQMIRGVGRGNYPPLSPHPEDQPVMRKLWRPV
 IPLWACVGLAGFLACLFIYVLNWRLLGDSTNPVLAKIYQTLPEAAIEQPV
 QNVQPVNLRAFLRPEIQAGLVAVRDEADRSVVTLKGDGLFASGSTVARE
 SYEPVIDRVAQAMNNVSGKILVGFSDNPIRSARFASNYELSLERARSV
 QSLQKHLSPARVKAEGRGEMNPIAPNNSAENRARNRRVEITLLVSPGN
 TAAELNGLPQGN

>2652308141 Ga0077644_106183 type VI secretion system protein ImpJ [Kosakonia oryzae KO348 : Ga0077644_106]

MNKAKEVWVTEGMFLRPHHFQRAESYLQHHIREWGTLQRPYLWGYLDIEL
 DDAMLRQGCIALSYASGLLPDGTFFSFHDARQAPTPLAIPDNINNERVVL
 ALPARRGGRDEVIFSEEKDSLARYVTWESEVDDDNAMSVGPAAVQFGRRL
 LKLMLEKDLSAEWTAIGVAHVVEKRNDNHVRIDSRYIPMLNAVNNPAIY
 AIINDLQSLMQRSQQIGQRLRQPGRFNTSEMVEFTLLALINHVGQVSH
 LKTLPMIHPEELWRSWLAFATELTTWTASRSPEETLPVYDHDLAGCFGK
 LQLMLRNLGLSLVMEEHAIQLPLTERTHGLNIATLPTTTMAREFGFVLAVK
 ASVPGELQTHFPAQMKVAPVTKIRDVLQQLPGMKLRTMPVAPPQIPWH
 AGYNYFELEKGGELWNEMEKSGAFALHLAGFPGLDMEFWAIRSPT

>2652308142 Ga0077644_106184 type VI secretion system protein VasD [Kosakonia oryzae KO348 : Ga0077644_106]

MNNNSVVRQISLVFFFVLATLASGCGSSSHSVPTSYNLQFRAHPQINESA
 PLKVRVLLLKSDATFMSADFWSLQNNADSVLGANLLNSDEFFLMPGQLSK
 TLSGKSAPDARYIGVMAEYQALDGGKWRMSLPLPVQGETHFYEFWKSSSD
 ELEANIFLDVNGIRVVSK

>2652309532 Ga0077644_11595 type VI secretion system protein ImpB [Kosakonia oryzae KO348 : Ga0077644_115]

MDSDFQREIPKARINLKLDLHTGGAQKTELPLKLLVTGDFSNGQETASL
 SERVKNVNNKNNFNSVLSDYSPKVNLIVENTLAGNASEENISLTFRDMKD
 FTPEEVARQIPQLKAMLAMRNLLRDLKANLLDNQTFRKELEKILLNPSLS
 AELRDELSSLAPKQP

>2652309533 Ga0077644_11596 type VI secretion system protein ImpC [Kosakonia oryzae KO348 : Ga0077644_115]

MSVQNESVATGESVVLQGTQAGGVYASLFKINLNPVTTLSALDIWQDAQ
 AMSDATADERLTAGMQVFLECLTKSDSKVEKLDNRNLIDHHIAELDYQISR
 QLDAVMHHEAFQAVESLWCGLSLVDKTDFFRQNVKIELLDLSKDDLQDF
 EDSPEIISGLYKHTYIDEYDTPGGEPIAALISAYEFDASAQDVALLRNI
 SKVSAAAHMPFIGSAGPKFFLKDAMADVAAIKDIGNYFDRAEYIKWKSFR
 ETDDSRYIGLVMPRVLGRLPYGPDTPVRSFNYYVEEVKGPDHDKYLWTNA
 SFFAANMVRSFINNGWCQIRGPQAGGAVQDLPIHLYDLGTGNQVKIPS
 EVMIPETREFEFANLGFIPLSYYKNRDYACFFSANSTQKPALYDTADATA
 NSRINARLPYIFLLSRIAHYKLIQRENIGTTKDRRLELELNTWVRSV
 TEMTDPGDELQASHPLRDAKVVVEDIDDPGFFRVKLYAIPHFQVEGMDV
 NLSLVSQMPKAKS

>2652309534 Ga0077644_11597 type VI secretion system protein ImpJ [Kosakonia oryzae KO348 : Ga0077644_115]

MKIYRPLWNEGALLAPQQFQQSGWESFSRAGLSRLYSPFPWGVVERVEFN
 EALLASDRVQVQTLRLWLPDGTLDVDTQNSDLPEPEPREVALPDSGQVESVT
 VLIALPVMQPGIVNVQMETVSAERPLRYREEWVAVQDLFGQEEEEPMAVAR
 FNLAFRFDHESNDAWQTCAVARLLRDGQGGWRQDPDFVPPMAMFASLL
 RERLVLLNRQLRSRRQRLMAMRRESNDRMADFAVADVSLFWLLNALNTHA
 RVLTEFERFPAHPEQVWAEARLAGSLLTFSLDRDLDAIPSYDHHAPEN
 TFPPLFELISELLEASLPSRVAVNMTRLDEQTKPRRMKQTVAGR

>2652309535 Ga0077644_11598 hypothetical protein [Kosakonia oryzae KO348 : Ga0077644_115]

MNQYNYNRLLKQIDSSNDGIYSEQEYLQAVHNPSYRDHLHRIIVMHPSEW
YYGKADIYF

>2652309536 Ga0077644_11599 hypothetical protein [Kosakonia oryzae KO348 : Ga0077644_115]

MCNRIYLVYLLISLLSGIAHSDDDIKETRWWFDKISERNLFGKHNTYTV
NYLNMIIYNKSILNIGNKRLIITNDFLENKNICSMYEVKLLKKSALSYFMST
NIVNMYSMFLKYENIQFPEDVYEITSLFPGKECPVPYDAIVKAGSNLFTV
DQDYVVFYKQSNVITPQDNALYLNNNWKGKYCHNRNVESQFDGTSEYICIF
DNMGIKESYQEVISFDESASGKLSKILPNDNNSYKANGFSVDYKWVDNDR
LKILVAMDSETTSYFYKNNTGTNLHVLVEAQY

>2652309537 Ga0077644_115100 PAAR motif-containing protein [Kosakonia oryzae KO348 : Ga0077644_115]

MKFGGIGVARKGDKVSCPKEGHGPTTIVEGNPDYLDQGVPAFHGHKCGC
GCTLISSFSAGKVA

>2652309538 Ga0077644_115101 type VI secretion system secreted protein VgrG [Kosakonia oryzae KO348 : Ga0077644_115]

MVRVIGVSADCRCLCAVFFILDWYESGAFYARLHHERELNKSARLHLFSNA
AGLMPGMVLEADGSNLNDLKDGLMLTLITYRAARDTRLHMSVWGMPLYSEQ
FCFRPQEEPRPHIHGTLPARIESREQGDIYAWLDNQGRYRVKMDFSREDA
EPGYNLWIRQAKPCSGDITYGWHPLTDGTEVGIAYEDGDIDRPIAHAF
HDSEHQDIVTRDNRSQNILRTAAGNELRMEDLRGQEHIALTTPFGGTQLN
QGHISDEQSKQRSGFELRTDEYGVIRVAKGLFITADGQQKAVGEVLDME
TALKEIDVCQQQLKALAAAAEAQAQALEADIASQKAMFDQRLKPLNGMIHS
HGPQGVAFSTSGEHLQLAAGQNVAVNAGGDFSSGAMGNAAIAGEGVGLFA
RTGSLTLNASEGVPVQIQAQNGAMHLSAEQKLSLISASDILFAGKKVTLI
GGGSYLVIDNGKVEYGTDMTYTRRIKRTYLTAPASLSGMMPSFSQSGICL
SCLLEAAENGAPILIKGE

>2652309539 Ga0077644_115102 protein of unknown function (DUF4123) [Kosakonia oryzae KO348 : Ga0077644_115]

MKHSIVDRLKEIQNESDKRKIYALVDGAQYDRFCTIELFKRNGVMPLFDS
WEDRCLAFAGPWLLALESIDNDLFSILNNLELKYPVSWILSSSSFDLDF
FHLKNRLEVIMPNNQVAMLRYYDPRVLIFLPEVLTDKQLDVFMSIISWG
CKYNGEDYFIK

>2652309540 Ga0077644_115103 type VI secretion system secreted protein VgrG [Kosakonia oryzae KO348 : Ga0077644_115]

MGVKVRVQSPSEVARCEDPAISATLPCASCWSKDYAKEIKVNPVKRYSDQ
LNAEGKSYEYNFGARQYKLTICYCKTLKKVSEIRLKIIEPKGVDETVIEK
SKKSLIKIGIKENWKNKFLKTTDPKCGVKIFIEFKVEFVASNEHYVFR
HKQYNREGVTGKFLDVSTDTGSWVYAHEFGHCFGLPDEYGYKAGVQKDKQ
VVYYKPDGKLDAPFSVPYNGGNAPEPSSTIMAAYGNTTILKRHWLIAIE
ARDLLNEPGLGRKIECDII

>2652309541 Ga0077644_115104 hypothetical protein [Kosakonia oryzae KO348 : Ga0077644_115]

MIENNYFIISTNCERQEECVYNNEREIPVVVTIKNISSKGFYIPLKFIK
TGPVELIDRRRTKKKHALETKYC

>2652309542 Ga0077644_115105 hypothetical protein [Kosakonia oryzae KO348 : Ga0077644_115]

MKISTDQLVLLDFYSKNAFANELSEHYKEIFPIYNELGAKKLIKISLAKL
INKAKELNITQRGPLQLYIDMAIVLGYGFDTPMYSCFSSVNEQYSSGTE
LERSLQIYDKFNGYLKDVMGENNLHILGFKNLANEKFDGFSNINFSQDI
FDLLNDLYPQKCRYLGCDKVKELICLLENPDYTYLNLGSRAVFLVMFV
VGYKQSDCFHLYEGFFSEETGGDDQFLIYKAKEFLTKYIDAMVT

>2652309543 Ga0077644_115106 type VI secretion system secreted protein VgrG [Kosakonia oryzae KO348 : Ga0077644_115]

MIHFHDPQGVAFSTSEYIQLAAGHNVAVNVCGYFSSGAMGNMAILGGEGV
GLFARTGSLLNASEGVPVQLQAQK

>2652309544 Ga0077644_115107 Protein of unknown function (DUF3304) [Kosakonia oryzae KO348 : Ga0077644_115]

MGLFKLFSKVDNAVNRGYARWGKIWAALITPFIYGACIVWASIWGPPV
GPVTLIIHSEIDRPILGFSVNGVGGGNASSYKRNPNYSGGGGAATCCGSIS
GKTAEVIWTLDITHEQYLKGMRLKRRVVMPLPERKWGENDLHVHFLPGD
KVLLGWSDNAWSPYEKRPDIPGKTVKQEHN

>2652309545 Ga0077644_115108 hypothetical protein [Kosakonia oryzae KO348 : Ga0077644_115]

MDSESIIAANRAQQAEDAGLGNCSTWVHVGFFFDGIHRNIDQDASEQRL
SNVAPSSGYNCKSTRK

>2652309546 Ga0077644_115109 type VI secretion system protein [Kosakonia oryzae KO348 : Ga0077644_115]

MENSSPSLYETLYGNFTGGLDLHQVNEQNQAILSVLDNMQRILNCRAGTL
KHLDPDYGLPDMTKILQGMPGTAHQLLQVFSVDVLLKYEPRLKKITVVLLEQ
EIPGELRYAIDAEANGIGLVRYGTVFMPEGRVLLRHLKQQQYLDKTL

>2652309547 Ga0077644_115110 type VI secretion system protein VasL [Kosakonia oryzae KO348 : Ga0077644_115]

MNDISPRKIKTGCDPRTLADYAILRDELSKLTHPARPDVNWRVVEKLCLS
LFEQNGVELQTAAWYTLARTQLAGLFGLEGLAILEALISHQWGVLPWPQP
VHARMEILSNLSQRLQQRMRTPPLNYSDSLQYRAEQLLTSLGAVLQRLE
LKHLSQLDTLRSMIHNSAVRENSDGDSDGATIQAGIVLPAPVMNSAGI
LSDALPGIPVTEKREPANTVNWVYVAHPEHQPNVDVLTAMPVQVKKWKPF
AAGMCTMLIISAATVWGWHALHRLDPLQTQLAASLAPLPAILTPAQLDTL
RQHAPIPQTMFTATQQQLVRLGQLPPDWNINYSRQLVEQAQSLWPEQAKP
LAQRWFRQLNAASVPTENLSGWHQGMTKLQQLSNRLSGLDGHKGYMTVS
ELKSSVFGMMTSFQQTEPTEEQLRQINLLPVASPLRQQLIRQLEQHLRAQ
IYTLGQMKNSAPVTKLLREQAVNPSPLND