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Genome Sequence of the Cat Pathogen, *Chlamydophila felis*

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Abstract

Chlamydophila felis (*Chlamydia psittaci* feline pneumonitis agent) is a worldwide spread pathogen for pneumonia and conjunctivitis in cats. Herein, we determined the entire genomic DNA sequence of the Japanese *C. felis* strain Fe/C-56 to understand the mechanism of diseases caused by this pathogen. The *C. felis* genome is composed of a circular 1 166 239 bp chromosome encoding 1005 protein-coding genes and a 7552 bp circular plasmid. Comparison of *C. felis* gene contents with other *Chlamydia* species shows that 795 genes are common in the family *Chlamydiaceae* species and 47 genes are specific to *C. felis*. Phylogenetic analysis of the common genes reveals that most of the orthologue sets exhibit a similar divergent pattern but 14 *C. felis* genes accumulate more mutations, implicating that these genes may be involved in the evolutional adaptation to the *C. felis*-specific niche. Gene distribution and orthologue analyses reveal that two distinctive regions, i.e. the plasticity zone and frequently gene-translocated regions (FGRs), may play important but different roles for chlamydial genome evolution. The genomic DNA sequence of *C. felis* provides information for comprehension of diseases and elucidation of the chlamydial evolution.

Key words: comparative genomics; genome inversion; obligate intercellular bacteria; chlamydia; infectious disease

1. Introduction

Chlamydiae are obligate intracellular eubacterial pathogens, including two genera and nine species based on ribosomal RNA gene sequences.[1] The genus *Chlamydia* includes a human conjunctivitis and sexually transmitted disease agent, *C. trachomatis*, a mouse pneumonia agent, *Chlamydia muridarum* and a pig pneumonia agent, *Chlamydia suis*. The other genus, *Chlamydophila*, includes a human pneumonia agent, *C. pneumoniae*

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Data deposition: The sequences reported in this paper have been deposited in the DDBJ/EMBL/GenBank database (accession chromosome, AP006861, plasmid, AP006862).

and the animal disease or zoonosis agents, *Chlamydophila psittaci*, *Chlamydophila caviae*, *Chlamydophila pecorum*, *Chlamydophila abortus* and *Chlamydophila felis*, which infect birds, hamsters, cows, sheep and cats, respectively. The genomic DNA sequences of *Chlamydiaceae* have been published for five species, *C. trachomatis*,[2] *C. muridarum*,[3] *C. pneumoniae*,[3–5] *C. caviae*,[6] and *C. abortus*.[7]

Chlamydophila felis strain Fe/C-56, of which the whole genome DNA sequence has been determined in this study, is a frequent cause of infection in cats, largely causing conjunctivitis[8] as well as pneumonitis.[9] Seroepidemiological studies have shown that *C. felis* is widely spread among cats with prevalence rates of anti-*C. felis* Fe/Pn1 antibody in ≈50% of street cats and ≈20% of domestic cats and in 1.7% of the general human population and 8.8% of veterinarians in small animal clinics in Japan.[10] Although *C. felis* is a common pathogen in both humans and animals, the infection is rarely identified in humans.[11,12] Seroepidemiological data reveals, however, that the infection in humans may be more frequent and are most likely acquired from infected cats.[10,13] This suggests that the feline chlamydiosis is widely transmitted to humans when in close contact with infected cats, but it rarely causes any serious illness in humans. In contrast, infection of a taxonomically close chlamydial species, *C. pneumoniae*, is very frequent in human and is sometimes associated with a serious illness. Moreover, at extremely high frequencies of detection and culturing from atherosclerotic plaques, *C. pneumoniae* is suspected to involve the development of atherosclerosis.[14] The *C. felis* genome sequence and the comparative analysis with other chlamydial species will be of importance to investigate the genes relating to their pathogenicity in humans, tropisms in hosts and disease prevention as well as the evolution and intracellular parasitism.

Herein, we determined the complete sequence of *C. felis* genome and present the comparative analyses with other *Chlamydia* genome DNA sequences, especially to that of *C. pneumoniae*.

2. Materials and Methods

2.1. Strain and culturing

Chlamydophila felis Japanese strain Fe/C-56 was originally isolated from conjunctival mucus of an infected 5-year-old female cat.[8] The genomic DNA for sequencing was prepared from *C. felis* elementary bodies in the culture medium of the fourth passage with McCoy cells, following five passages with fertile hens' eggs. Chlamydial culturing and detection were performed as described previously.[15] The nomenclature *Chlamydophila felis* is in dispute, but it is used here instead of the formerly used 'a cat pneumonitis agent of *Chlamydia psittaci*'.[1]

2.2. DNA sequencing, assembling and gene prediction

The DNA sequencing was performed by a method using the whole genome shotgun.[16] The total 24 194 sequence reads gave an 11-fold coverage as average. The sequences were assembled and edited using the Phred/Phrap/Consed package software (University of Washington).[17] Protein-coding genes were first predicted using the combination of three programs, GenomeGambler,[18] GeneHacker plus[19] and Glimmer2.0,[20] and at last manually determined. Annotation for each genes was carried out by using programs BLASTP[21] and FASTA3[22] against the non-redundant protein database. Transmembrane protein and tRNA genes were predicted using SOSUI[23] and tRNAscan-SE,[24] respectively. Putative *inc* genes were predicted by informatics analysis using hydropathy.[25,26]

2.3. Data analysis

C. pneumoniae J138 genomic DNA sequence[4] was used for intra-genus comparative analyses. Genomic DNA sequences of other *C. pneumoniae* strains,[3,5] *C. trachomatis*[2] and *C. muridarum*[3] were used for inter-genera comparisons. *C. caviae*[6] and *C. abortus*[7] were used for analysis within evolutionarily closed species. The gene annotation was performed by homology search using FASTA3,[22] in which the similarity with the expectation values $<10^{-4}$ was defined significant. Two genes conserved most reciprocally in two different organisms were assigned as orthologous genes using FASTA3. Classification of *C. felis* genes into *Bacteria*, *Eucarya* or *Archaea* is carried out based on no chlamydial orthologues, described previously.[27] The genes which were categorized into *Eucarya* and *Archaea* were confirmed by phylogenetic analyses using CLUSTALW.[28] The detail results for gene annotation, classification, gene divergence patterns and gene content comparisons are available in the Supplementary Tables 1–5 are available at www.dnares.oxfordjournals.org. The sequences in this paper have been deposited in the DDBJ/EMBL/GenBank database (accession; chromosome, AP006861, plasmid, AP006862).

4. Results and Discussion

4.1. Genome structure

The *Chlamydophila felis* Fe/C-56 contains a circular chromosome consisting of 1 166 239 bp and 1005 protein-coding genes (Table 1). The *C. felis* also harbors a 7552 bp plasmid, pCfe1, with eight genes and the copy number of the plasmid per chromosome is approximately 4.5 (Table 1). The overall G+C contents are 39.4 and 33.9% for the chromosome and plasmid, respectively. The G+C content and gene density appear to be uniform in the genome (Fig. 1). The putative replication origin

(Ori) of the chromosome was determined based on comparisons with other chlamydial predicted Ori sequences, the GC skew and the assumed DnaA binding sequences. The replication terminal was determined at a maximum point, 583 kb from the Ori, in a curve of the cumulative GC skew.[29] There is no strong correlation between the directions of transcription and replication, except from the 880 kb position to the Ori observed slightly positive association. Many of the essential genes for transcription, translation and replication are located in a large region around the Ori.[5]

Table 1. General features of *C. felis* Fe/C-56 chromosome and plasmid.

	Chromosome	pCfe1
Size	1 166 239	7 552
GC content (%)	39.4	33.9
Relative copy number	1	4.5
Protein-coding genes		
Total number	1 005	8
Function assigned	682	4
Conserved hypothetical	281	4
C. felis specific in the *Chlamydiaceae*	47	0
Stable RNA genes		
rRNA	1	0
tRNA	38	0

4.2. Plasticity zone

To figure out *C. felis* Fe/C-56 genome characteristics, genome structure and gene contents of *C. felis* were compared with those of *C. pneumoniae* J138.[4] Although *C. felis* and *C. pneumoniae* relatively diverged far from each other, the two species share 879 orthologous genes and overall high synteny is exhibited, except a region surrounding the Ter, termed the plasticity zone.[3,30] Synteny of the orthologous genes between *C. felis* and *C. pneumoniae* are displayed in a concentric presentation (Fig. 2A). In this report, *C. felis* plasticity

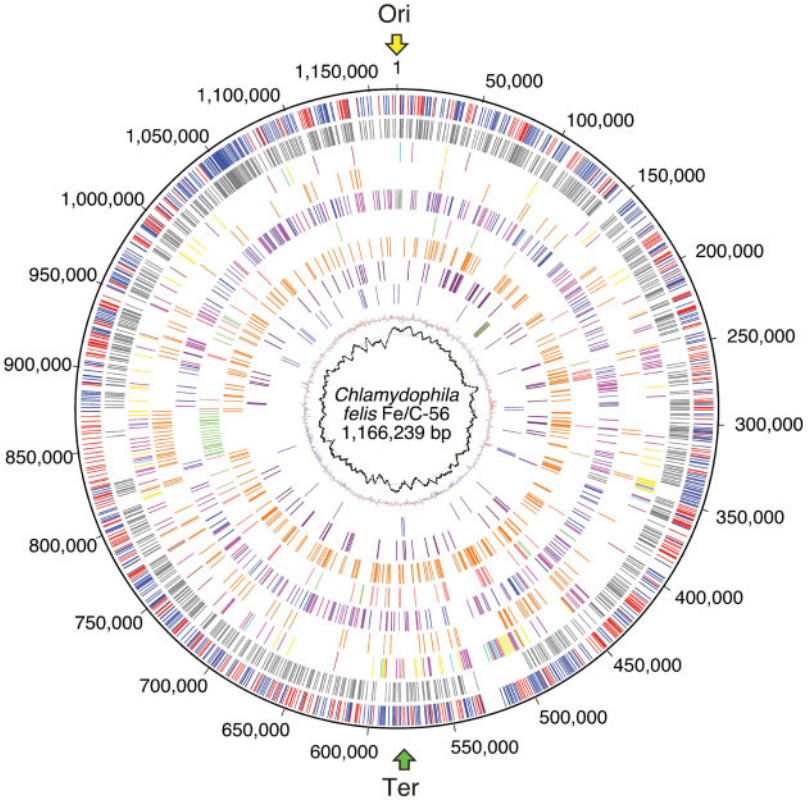

Figure 1. Circular exhibition of the *Chlamydophila felis* Fe/C-56 genome. The outermost scale is marked for nucleic acid position in base pair. The track 1, gene positions and directions, clockwise in red and anti-clockwise in blue; track 2, common genes in three chlamydiae, *C. felis*, *C. pneumoniae* J138[4] and *C. trachomatis*[2] in gray; track 3, common genes in only two chlamydiae *C. felis* and *C. pneumoniae* in yellow, *C. felis* and *C. trachomatis* in light blue, *C. felis* specific in violet; track 4, paralogous genes in orange; track 5, gene origin classification, gram positive in dark blue, gram negative in violet, Archaea in green, Eukaryote in light purple; track 6, transmembrane proteins, *inc* family in pink, *omp/pmp* (outer membrane proteins or polymorphic outer membrane proteins) family in light green; track 7, total transmembrane proteins in orange; track 8, virulent factors; track 9, tRNA in light purple, rRNA in dark green; track 10, GC content, a radius is the average of genome GC content. Red and blue bars illustrate for plus and minus from the average (radius: 100%); innermost, respectively, GC skew, outside and inside of green circle indicate values>0 and values<0 calculated by (G–C/G+C).[29] Based on cumulus of the GC skew values and putative DnaA binding sites, a hypothetical origin, shown as Ori, is determined and the base numbers are counted from the origin in the direction settled arbitrarily.

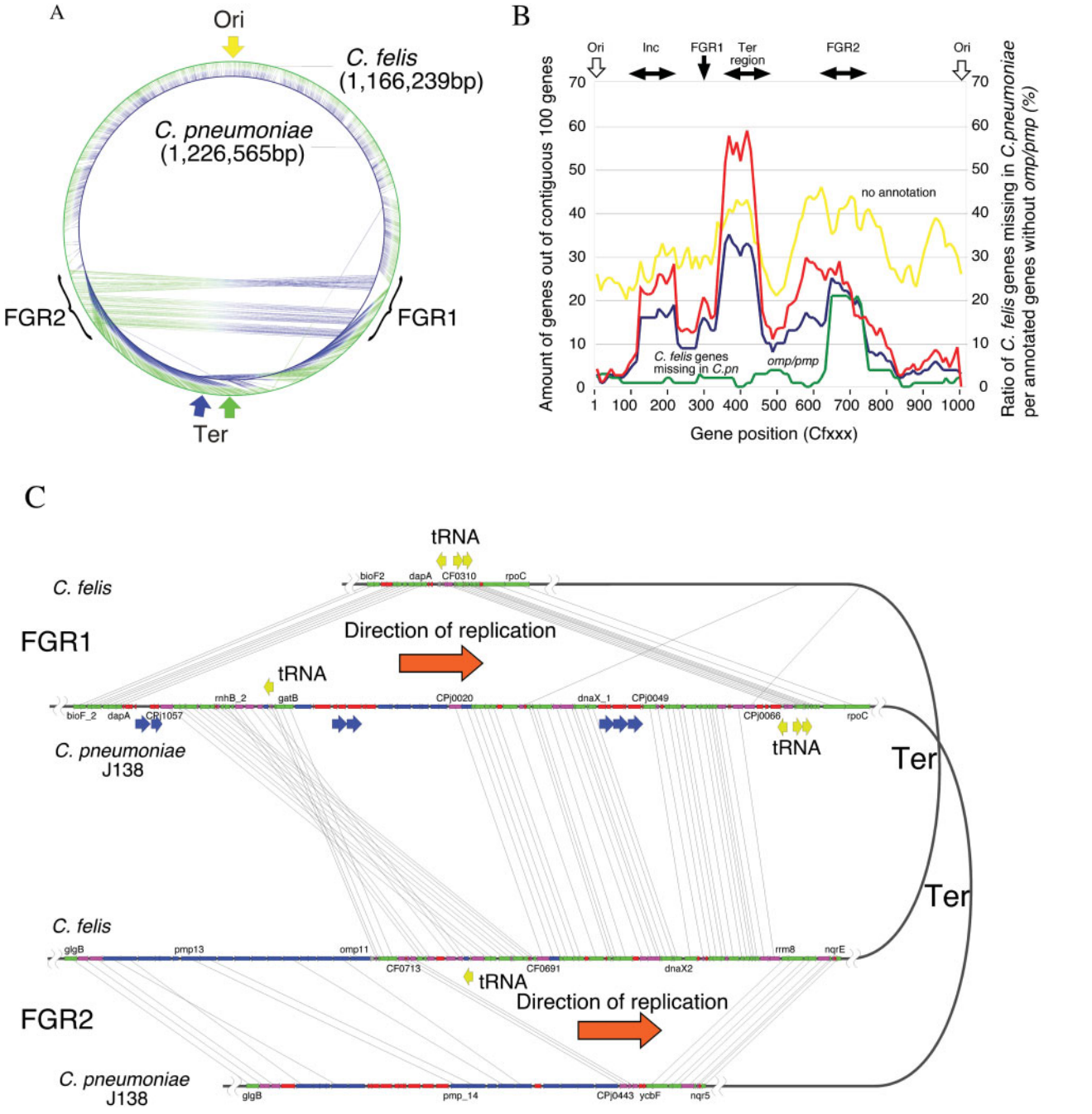

Figure 2. Analyses of the orthologous genes between *Chlamydophila felis* and *Chlamydophila pneumoniae* J138. (A), Circular presentation of locations of orthologous genes on whole chromosomes. Two orthologues are linked between *C. felis* (outer in green) and *C. pneumoniae* J138[4] (inner in blue). FGR1 and FGR2 indicate the 'frequently gene-translocated regions'. (B), Gene distributions for *C. felis* genes without *C. pneumoniae* orthologues (*C. felis* no-*C.pn*-genes: blue), hypothetical genes (yellow) and *omp/pmp* genes (green) on the genome are calculated at window: 100 genes and step: 10 genes. The red line indicates a ratio of annotated *C. felis* no-*C.pn*-genes per annotated genes excepting *omp/pmp*. (C), Schematic orthologous linkages in the FGRs. Arrows in each chromosome exhibit *omp/pmp* genes in blue, annotated and conserved genes in green, annotated but specific to each species in gray, not-annotated and conserved genes in purple, not-annotated and specific to each species in red. Arrows outside the chromosomes show tRNA genes, directions of replication and *C. pneumoniae* seven repetitive sequences are indicated in yellow, orange and blue, respectively.

zone was assigned from 491 to 596 kb (CF0398-0484: 87 genes) including the Ter region.[6] In the *C. felis* plasticity zone, *C. felis* genes without *C. pneumoniae* J138 orthologues (*C. felis* no-*C.pn*-genes) are enriched with up to 30 genes out of the total 126 *C. felis* no-*C.pn*-genes on the chromosome (Fig. 2B). While 48 genes are functionally annotated out of 87 genes in the plasticity zone, *C. felis* no-*C.pn*-genes are 14 out of the 48 genes. The ratio (14:48) is much higher than the one in the *C. felis* chromosome (46:684). Most of the annotated

genes in *C. felis* are conserved in some other organisms and thus the annotated no-*C.pn*-genes are thought to have dropped out of *C. pneumoniae* genome.[3,6]

4.3. Frequently gene-translocated regions (FGRs)

When two orthologous genes between *C. felis* and *C. pneumoniae* J138 are linked on whole chromosomes shown with two concentric circles, segmental genome translocations are observed as sheaves of parallel lines horizontally crossing the origin-terminal axis between *C. felis* and *C. pneumoniae* (Fig. 2A). The regions with translocated genes in the *C. felis* genome are from 355 to 359 kb (CP0307-311) and from 759 to 878 kb (CP0654-0739, 86 genes), ones in *C. pneumoniae* J138 genome are from 1200 to 87 kb (Cpj1051-0072, connected at the termini of the genomic sequence) and from 485 to 550 kb (Cpj439-474) (Fig. 2C). The regions, termed frequently gene-translocated regions (FGRs) here, seems to have been formed by the genomic inversion events across the whole genome with the replication terminal region. When *Chlamydophila felis* genome is compared with *Chlamydia trachomatis* one, the same phenomenon for the gene translocation is observed in similar regions but the translocation between *C. felis* and *C. trachomatis* is more frequent than one between *C. felis* and *C. pneumoniae* (data not shown). The FGRs, which include the region reported as a hyper variable region[31] or tmh/Inc locus,[7] exhibit distinctive characteristics of the plasticity zone or the other regions of genome; (i) Out of 49 polymorphic outer membrane proteins genes *omp/pmp* (described below) in the *C. felis* chromosome, 22 *omp/pmp* genes are in the FGRs, whereas interestingly the plasticity zone contains no *omp/pmp* (Fig. 2B). (ii) Total 22 *omp/pmp* genes are conserved in *C. pneumoniae* J138, but in the FGRs only eight out of the 22 *omp/pmp* are conserved ($P < 0.01$). (iii) The *omp/pmp* and hypothetical genes are tandemly localized in the FGRs and seven repetitive sequences in *C. pneumoniae* genome contain *omp/pmp* genes, however *C. felis* genome lacks such repetitive sequences[31] (Fig. 2C). (iv) *C. felis* no-*C.pn*-genes are enriched (27 genes) in the FGRs similarly to the plasticity zone. But ratio of annotated *C. felis* no-*C.pn*-genes per annotated genes in the FGRs (2:57) is almost equal to the one in the whole chromosome (30:684), when the *omp/pmp* genes are excluded from the annotated genes. (v) Of the 66 total *inc* genes (described below), 16 *inc* genes are enriched in the plasticity zone ($P < 0.01$) but only 6 in the FGRs ($P = 0.5$).

Those characteristics implicate that the FGRs may assume a role for the multiplication of the *omp/pmp* genes rather than for genome reduction and *inc* genes may be on the verge of gene loss in the plasticity zone. Relationship between the gene translocation (or genome inversion) and gene multiplication are unknown, however it should be noted here that chlamydial genomes conserve an almost complete set of DNA recombination, repair genes and tRNA genes located in or near FGRs may be involved in the inversion fashion[32] (Fig. 2C). The characteristics observed in the FGRs of *C. felis* and *C. pneumoniae* are detected as well as in the FGRs of *C. trachomatis*.

4.4. Comparison within the family Chlamydiaceae

Four *Chlamydiaceae* species, i.e., *C. felis*, *C. pneumoniae* (three strains),[3–5] *C. trachomatis*[2] and *C. muridarum*[3] share 795 common genes (Fig. 3A), 104 genes are specific to *C. felis*. Adding, *C. caviae*[6] and *C. abortus*,[7] formerly belonging to *C. psittaci* as strains, to this analysis, 47 genes are specific to *C. felis* including an extra adenylate kinase gene (CF265) and 8 polymorphic outer membrane protein genes (CF379, 380, 718, 719, 723, 724, 728, 735).

All of the orthologue sets, except two, surprisingly illustrate the similar phylogenetic patterns, in which the genes from the *C. felis* and *C. pneumoniae* and those from *C. trachomatis* and *C. muridarum,* are closest (Fig. 3B). The two exceptional genes, CF0973 and CF0599, are too highly conserved to statistically draw relevant phylogenetic trees. Thus, no inter-genus gene

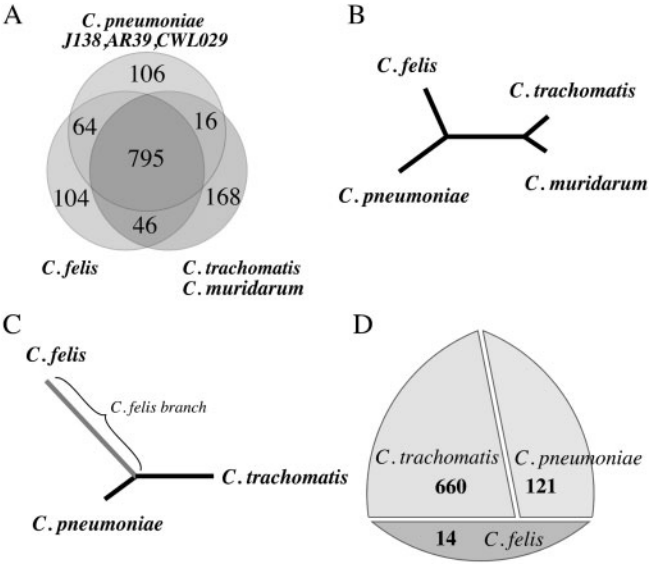

Figure 3. Orthologue comparison within the family *Chlamydiaceae*. (A) Numbers of orthologous and species specific genes represented in Venn diagrams, using *Chlamydophila felis* (this work), *C. pneumoniae* J138, AR39 and CWL029,[3,5] *Chlamydia trachomatis*[2] and *C. muridarum*.[3] (B) Divergence pattern for all orthologous gene sets, except two, CF0973 and CF0599. (C) A schematic branching pattern for an orthologous gene set showing *C. felis*-specific evolution. Based on the multiple alignment and phylogenic analysis of orthologues using CLUSTALW,[28] divergent distance is calculated as a branch length for an orthologous gene of each species. Genes are chosen for *C. felis* when the length of *C. felis* branch is longer than one of any other branch. (D) Numbers of genes exhibiting species specific evolution for *C. felis*, *C. pneumoniae* and *C. trachomatis* within the common 795 genes in Fig 3A.

exchanges are detected during the chlamydial species establishment in this method.

Regardless of the same phylogenetic pattern illustrated with orthologue sets, the lengths of *C. felis* divergence branches among the phylogenetic trees varied. To figure out *C. felis* genes which accumulate mutations more than the orthologous genes, the gene categorization was carried out using the definition that in the phylogenetic tree with *C. felis*, *C. pneumoniae* and *C. trachomatis*, a branch of *C. felis* gene from the divergent point is longest amongst three branches (Fig. 3C). Similar categorization was used for *C. pneumoniae* and C. trachomatis (Fig. 3D). Fourteen *C. felis* genes, such as four hypothetical genes (CF0008, CF0256, CF0470, CF0615), six genes encoding nucleic acid associating proteins (CF0085, CF0313, CF0899, CF0909, CF0925, CF0947) and four genes encoding membrane proteins including IncB (CF0032, CF0329, CF0415, CF0516) are categorized here as genes on faster evolution, implicating that these genes may be devoted for adaptation to the niche of *C. felis*.

Gene variation of tryptophan biosynthesis is one of the most remarkable features in the comparison of chlamydial gene repertories. *C. trachomatis* and *C. pneumoniae*, worldwide common parasites to humans but not to animals, contain partial and no tryptophan biosynthesis genes in the genomes, respectively. On the other hand, the genomes of the animal pathogens, *C. felis* and *C. caviae*, surprisingly not *C. abortus*, conserve the tryptophan operon consisting of *trpA*, *B*, *F*, *C*, *D* and *R* and related genes, *kynU* (CF0435-CF0440 and CF0434, respectively) are conserved.[6,7,33] Tryptophan depletion by IFN-γ is thought as a crucial host defense mechanism against chlamydiae. On the contrary the chlamydial tryptophan biosynthesis gene is thought to be directly involved in chlamydial resistance against IFN-γ.[3,5,34] IFN-γ treatment of host cells was performed to show how susceptible *C. felis* is against IFN-γ comparing with other chlamydial species without a complete tryptophan synthesis pathway. Neither inhibition of inclusion body formation nor reduction in the size of inclusion bodies on average were observed for *C. felis* in the tests with concentrations of up to 10 ng/ml of human IFN-γ, while both the infection and growth of *C. pneumoniae* J38 and *C. trachomatis* were reduced under the same experimental conditions.[34] Tryptophan might play a key role for the molecular basis of host–parasite interaction in terms of IFN-γ resistance, the gene repertories of tryptophan biosynthesis cannot explain all about host tropisms of individual chlamydial species. However, tryptophan utilization evokes all evolutional relationships between host defense and parasite infection, leading to chlamydial host and tissue tropisms.

4.5. Horizontal gene transfer

One of most interesting characteristics of chlamydial genome constituents is that a few percentages of the total genes are similar to genes of taxonomical distinctive organisms.[2,35] To figure out such genes in the *C. felis* genome, sequence-based gene classification was performed as a screening of the horizontal gene transfer from non-chlamydial organisms to the *C. felis* genome. Of the total of 1005 protein-coding genes of *C. felis*, 537, 59 and 5 genes may be derived from bacterial, eukaryotic and archaeal origins, respectively (Fig. 1). The numbers of chlamydial specific and ambiguous genes are 323 and 81, respectively. All the eukaryotic and archaeal origin genes in *C. felis*, except CF0874, are conserved and exhibit synteny in *C. pneumoniae*, *C. trachomatis* and *C. muridarum*. Based on phylogenetic analysis, the CF0874 coding a pyrimidine metabolism enzyme, orotate phosphoribosyl transferase, is seemed to be lost from *C. trachomatis* and *C. muridarum*. It is possible that the eukaryotic and archaeal genes in *C. felis* were laterally transferred before the genera divergence.[2] However, no further proof such as different G+C contents or genome structures is observed to show that the genes have been derived from taxonomical distinctive organisms.

4.6. Plasmid

Chlamydophila felis and many other chlamydial species carry a plasmid, pCf01, similar to other chlamydial plasmids.[36] Interestingly, two genes, pCf07 and pCf08, on the plasmid exhibit high similarities to CF0055 and CF0056 on *C. felis* chromosome, respectively. The CF0056 and pCf08 gene products show significant similarities to *parA* and *minD* gene products conserved widely among bacterial genomes. The phylogenetic analysis of the *parA/minD* family has illustrated that the chlamydial chromosome and plasmid genes form independent groups, both of which are separated from the other bacterial genes. Moreover, the divergent patterns in each chlamydial group are analogous (Fig. 4). It implies that the chlamydial plasmid had already been presented in an ancient chlamydia before the two genera divergence and that no intra-genus plasmid exchanges and no *parA/minD* gene exchanges between the chromosome and plasmid occurred after the divergence.

4.7. Inclusion membrane proteins (Incs)

Two hundred and sixty five genes in the *C. felis* genome contain transmembrane domain(s). The ratio of the number of transmembrane genes per the total number of genes in *C. felis* agrees with the finding in most bacteria,[37] but the distribution of the number of transmembrane domains is different from non-chlamydial bacteria. Noticeably, *C. felis* contains 75 genes coding two transmembrane domains, which is twice as many as some non-chlamydial bacteria. Informatics analysis[25] has revealed a total of 63 putative *inc* genes in the 75 genes. In addition, three more genes similar to eukaryotic myosin heavy chain genes have been classified as *inc* because four

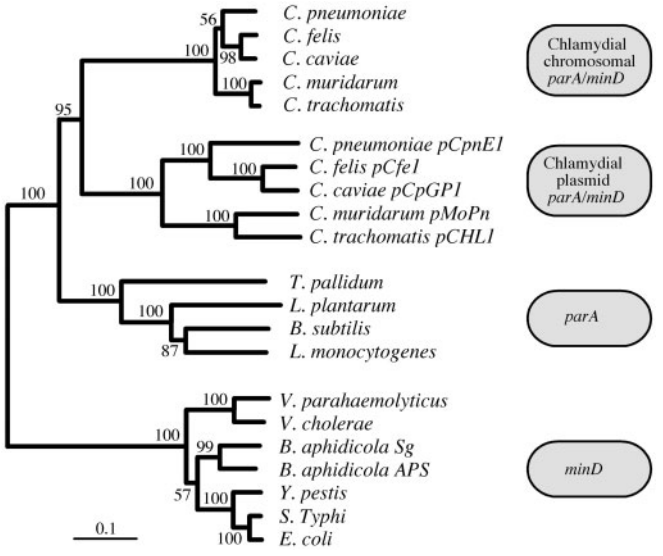

Figure 4. Phylogenetic tree of *parA/minD* genes. The *parA/minD* genes used here are for chlamydial chromosome; *Chlamydophila pneumoniae* J138 (Accession: AP002548-9), *Chlamydophila felis* Fe/C-56 (CF0056), *Chlamydophila caviae* GPIC (AE016997-167), *Chlamydia muridarum* (AE002353-7) and *Chlamydia trachomatis* D/UW-3/CX (AE001329-1) and plasmid; *Chlamydophila pneumoniae* N16 pCpnE1 (X82078-9), *Chlamydophila felis* Fe/C-56 pCfe1 (pCf08), *Chlamydophila caviae* GPIC pCpA1 (AE016997-168), *Chlamydia muridarum* pMoPn (AE002162-6) and *Chlamydia trachomatis* pCHL1 (J03321-7) and for other bacterial genes; *Lactobacillus plantarum* WCFS1 (AL935261-129), *Treponema pallidum* Nichols (AE001208-8), *Bacillus subtilis*168 (Z99124-191), *Listeria monocytogenes* EGD-e (AL591984-146), *Vibrio parahaemolyticus* O3:K6 (AP005076-8), *Vibrio cholerae* N16961 (AE004271-9), *Buchnera aphidicola* Sg (Schizaphisgraminum) (AE014108-5), *Buchnera aphidicola* APS (Acyrthosiphon pisum) (AP001119-13), *Yersinia pestis* CO92 (AJ414151-83), *Salmonella enterica* Typhi (AL627272-29) and *Escherichia coli* K12 (AE000216-6).

other genes partially similar to myosin heavy chain genes are categorized into the putative *inc* genes and two myosin heavy chain like protein of the *C. pneumoniae* J138 were experimentally shown to localize on inclusion bodies (data not shown). However, the functions of Inc proteins are still largely unknown. Forty-six putative *inc* genes are identified by the same method for *C. felis inc* identification in the genome of *Parachlamydia amoebophila* UWE25, which is diverged at about 700 million years ago from the last common ancestor with the family *Chlamydiaceae*[38] (data not shown). It indicates that before the divergence of the two chlamydial families, *inc* gene family has already expanded in the common ancestor genome.

4.8. Polymorphism membrane proteins (PMPs)

The *omp/pmp* genes, which encode outer membrane proteins or polymorphic membrane proteins, compose the most important and characteristic gene family in chlamydia. OMP/PMPs are reported to conserve N-terminal repeat motives, GGAI (or variants) and FXXN and terminate in a phenylanine residue.[4,7,39] In chlamydial genomes such as *C. pneumoniae* strains, *C. trachomatis*, *C. muridarum*, *C. caviae* and *C. abortus*, 18, 21, 9, 9, 17 and 18 *omp/pmp* genes are identified, respectively.[2–7] FASTA analysis with the all chlamydial *omp/pmp* genes reveals that in the *C. felis* genome 39 genes are similar to *omp/pmp* genes. Of the 39 putative *omp/pmp* genes, 18 genes (CF0209, CF0379, CF0721, CF0722, CF0725-CF0732, CF0735-CF0737, CF0801, CF0802, CF0992) conserve the N-terminal repeat motives and phenylanine at C-termini and 4 (CF0380, CF0719, CF0723, CF0733) and 6 (CF0101, CF0525, CF0720, CF0724, CF0734, CF0958) genes conserve only the N-terminal repeat motives or phenylanine at C-termini, respectively. No-*C. felis* genes are identified as new *omp/pmp* genes by the motif analysis. Gene expression of those genes and utilization of the gene products should be tested further.

5. Conclusions

Here we exhibit genome analyses of a worldwide spread pathogen for pneumonia[9] and conjunctivitis[8] in cats, *Chlamydophila felis* (*Chlamydia psittaci* feline pneumonitis agent). *C. felis* infection is rarely identified in humans while a few were reported,[11,12] but seroepidemiological study revealed that the prevalence rates of anti-*C. felis* Fe/Pn1 antibodies (1.7% and 8.8% for the general human population and veterinarians in small animal clinics in Japan, respectively) are rather higher than expected.[10] Thus the genome data of *C. felis* must be a useful tool to understand chlamydial tropism and pathogenicities, or to aid in the detection and prevention of *C. felis* causing diseases in both human and animals.

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