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Molecular Biology of *Borrelia burgdorferi*

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1. Introduction

Borrelia may be unique among prokaryote in having a genome that is mainly linear DNA physical and genetic map of linear chromosome of *B. burgdurferi* has been published, it consist of 946 to 952 kb Linear DNA (Sherwood *et al;*1993, Davidson *et al;*1992, Barbour *et al;* 1982).

This bacteria also contains several circular and specially linear plasmids from 5 to 55 kb. Recently analysis of entire *Agrobacterium tumefaciens* C58 genome revealed presence of one 2.1-Mb linear and one 3- Mb circular plasmid (Servent *et al;* 1993) and it has been shown that *rhodococcus fascians* contains 4 Mb linear chromosome (Crespi *et al;* 1992). Presence of several linear plasmids seems the segmentation of *Borrelias* DNA to several linear pieces has led to the suggestion that the relatively small linear chromosome and the linear plasmids actually are minichromosoms. In *B. hermsii* it has been shown that total cellular DNA organized into several complete gnomes (*Kitten et al;* 1992) and it suggests that linear plasmids are like small chromosomes (Ferdows *et al;* 1989). Plasmid profile of B. *burgdorferi* from different geographical area has been revealed significant heterogeneity a feature that can be used for classification of bacteria within given species (Barbour *et al;* 1987, 1989). Another related spirochete *B. hermsii* like *B. burgdorferi* has several linear and circular plasmids and the genes responsible for antigenic variation are located in linear plasmids. In *B. burgdorferi* a 49 kb linear plasmid carries the genes for Outer Surface Protein A and B (OspA and OspB) (Barbour *et al;* 1987, Baril *et al;* 1989). It has been shown that passage of *B. burgdorferi* in BSK medium changes the plasmid profile and loss of plasmids may change the infectivity of organism (Schwan *et al; 1988*, Simpson, *et al;* 1990). Structure of Linear plasmids of *B. burgdorferi* shows similarity to eukaryotic virus such as vaccinia and African swine fever virus in having covalently closed ends like hairpin loops (Hinnebusch *et al;* 1991).

1.1 Taxonomy and classification

Borrelia burgdorferi belongs to the phylum Spirochaetes. The members of this phylum are long, thin, helically coiled bacteria that have flagella (*axial filaments*) running lengthwise between the peptidoglycan layer and the outer membrane. Movement of the flagellum produces a screw-like motion that propels the organism.

The phylum Spirochates contains a single class (Spirochaetes), a single order (Spirochaetales), and three families: Brachyspiraceae, Leptospiraceae, and Spirochaetaceae.

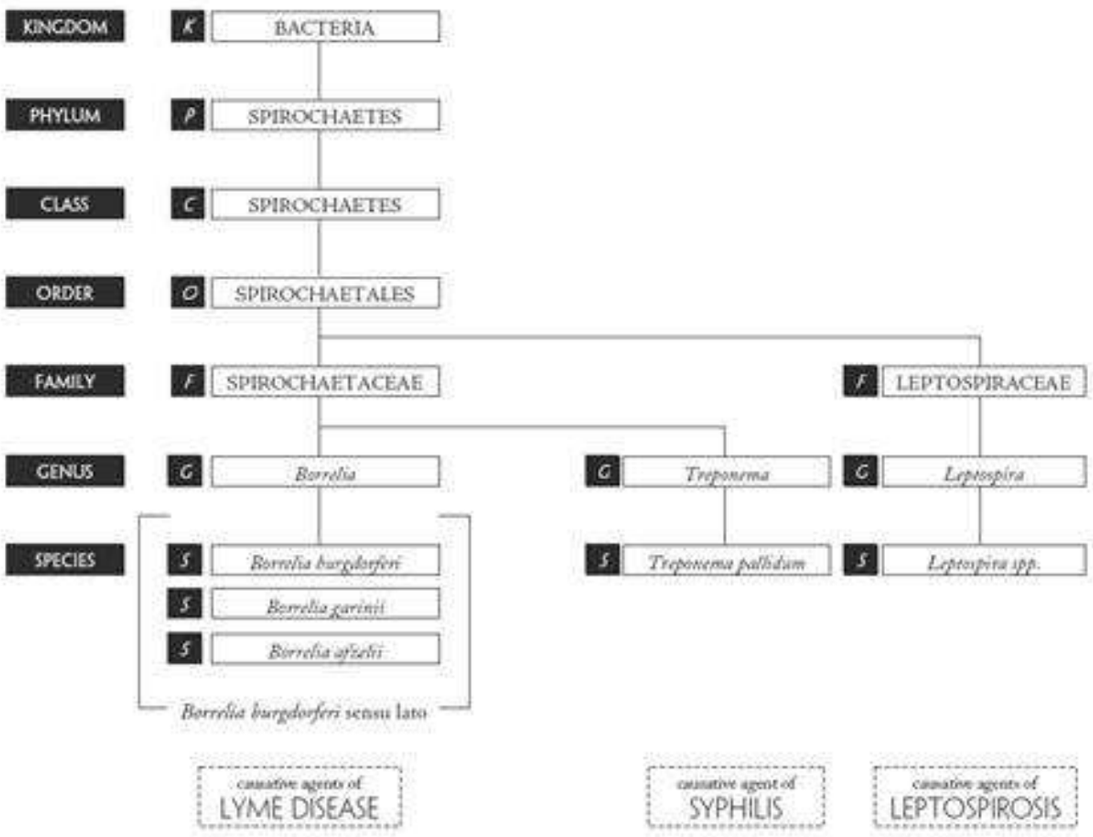


Fig. 1. Spirochaetaceae

The Spirochaetaceae family includes the genus *Treponema* and the genus *Borrelia* . *Treponema pallidum* is the causative agent of the sexually-transmitted disease syphilis.

The three members of the *Borrelia* genus *Borrelia burgdorferi* sensu stricto, *Borrelia garinii* , and *Borrelia afzelii* are collectively known as *Borrelia burgdorferi* sensu lato, and are the causative agents of Lyme disease.

1.2 Structure and morphology

Borrelia cells average 0.2 to 0.5 µm by 4 to 18 µm, and have fewer coils than *Leptospira*. The periplasmic flagella originate from either end of the spirochete (where they are anchored to the cytoplasmic membrane) and wind around the protoplasmic cylinder, imparting both motility *and* shape to the organism—in contrast to other bacteria, in which the peptidoglycan layer determines the shape.

The role of flagella in imparting *Borrelia* 's helical shape was established by inactivation of the *flaB* gene, which encodes the major flagellar filament protein, FlaB. This produced bacteria that lacked periplasmic flagella, were non-motile and rod-shaped.

Whereas the motility of externally-flagellated bacteria is hindered in viscous substances, that of spirochetes is enhanced, and about 6% of the chromosomal genome encodes proteins involved in motility and chemotaxis.

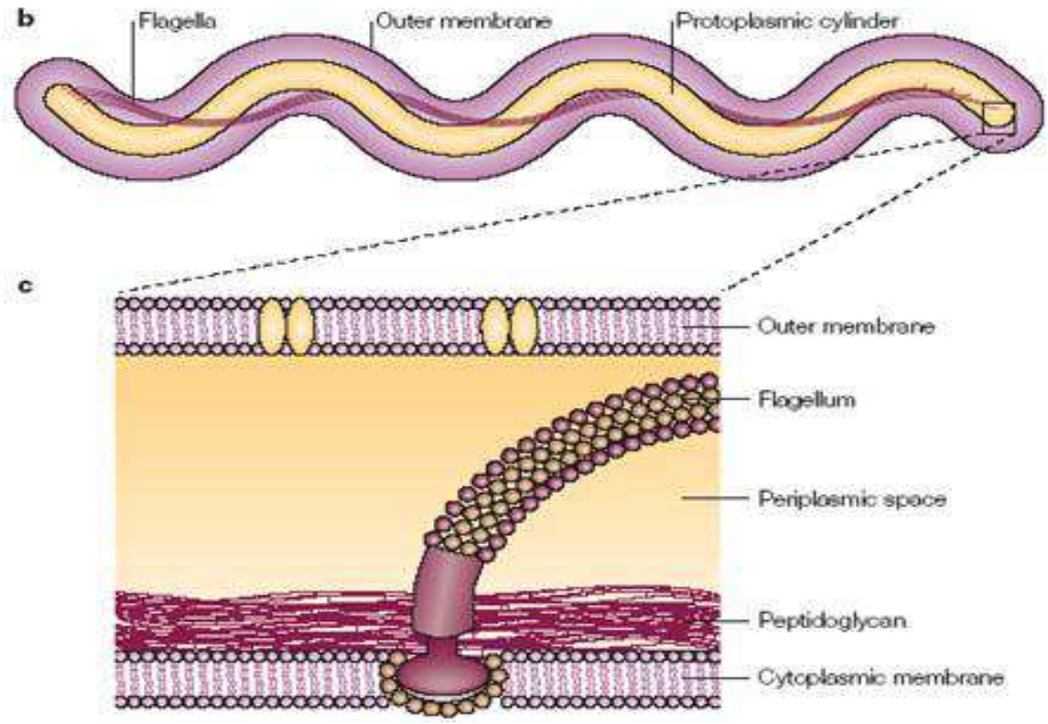

b
Flagella
Outer membrane
Protoplasmic cylinder
c
Outer membrane
Flagellum
Periplasmic space
Peptidoglycan
Cytoplasmic membrane

Fig. 2.

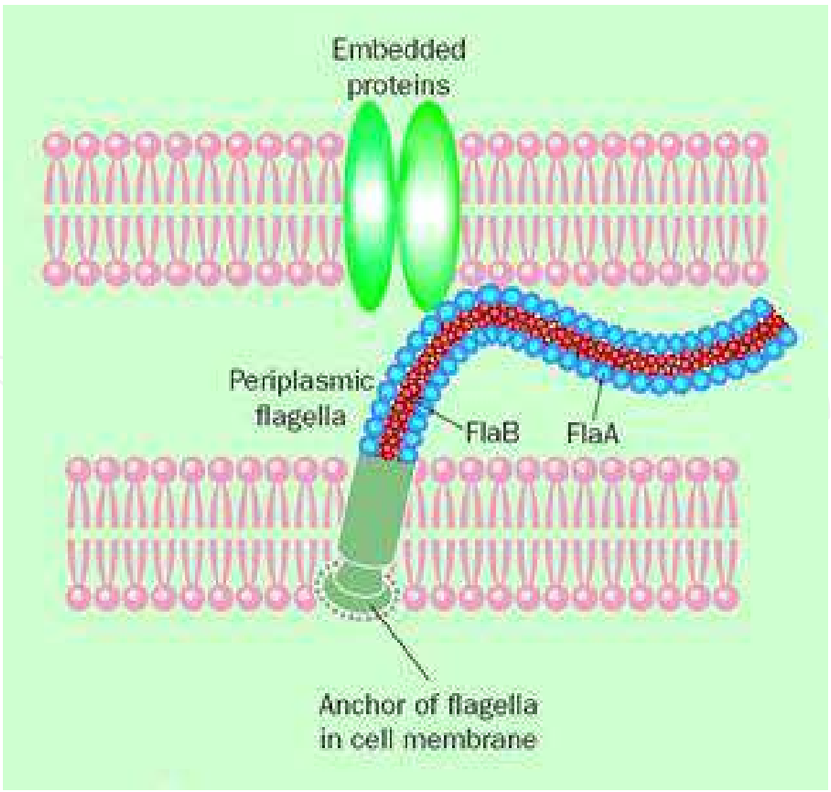

Embedded
proteins
Periplasmic
flagella
FlaB
FlaA
Anchor of flagella
in cell membrane

Fig. 3.

1.3 Genome organization of *Borrelia burgdorferi*

All members of the *Borrelia* genus that have been examined harbor a linear chromosome that is about 900 kbp in length as well as a plethora of both linear and circular plasmids in the 5-220 kbp size range. Genome sequences have been determined for *B. burgdorferi*, *B. garinii*, *B. afzelii*, *B. duttonii* and *B. recurrentis*. The chromosomes, which carry the vast majority of the housekeeping genes, appear to be very constant in gene content and organization across the genus. The content of the plasmids, which carry most of the genes that encode the differentially-expressed surface proteins that interact with *Borrelia's* arthropod and vertebrate hosts, are much more variable. *B. burgdorferi* strain B31, the *B. burgdorferi* type strain, has been studied in the most detail and harbors twelve linear and nine circular plasmids that comprise about 612 kbp. The plasmids are unusual, as compared to most bacterial plasmids, in that they contain many paralogous sequences, a large number of pseudogenes and, in some cases, essential genes. In addition, a number of the plasmids have features suggesting that they are prophages. Some correlations between genome content and pathogenicity have been deduced and comparative whole genome analyses promise future progress in this arena.

The highly unusual segmented genomes of *Borrelia* species can contain over 20 utonomously replicating DNA molecules. Many of the molecules, including the chromosome, are linear with covalently closed hairpin ends.

2. Molecular biology

2.1 The *Borrelia burgdorferi* genome

The genome of *Borrelia burgdorferi* consists of a single linear chromosome and several plasmids, both linear and circular. To date—as of January 2005—only the genome of *Borrelia burgdorferi* sensu stricto B31 strain has been fully sequenced.

Distribution of cellular functions of *E. coli* and *B. burgdorferi* genes		[1]
Category	*B. burgdorferi* genes (%)	
Intermediary metabolism	4.9%	
Biosynthesis of small molecules	3.1%	
Macromolecule metabolism	22.2%	
Cell Structure	37.0%	
Cellular processes	7.4%	
Other functions	5.6%	
Unknown functions	19.8%	

Table 1.

2.2 Chromosomal genome

B. burgdorferi contains a single linear chromosome of approximately 900 kb, and about 90% of it is comprised of coding sequences. Most of the genes encoded by the chromosomal genome are homologous to genes of known function.

2.3 Extra-chromosomal genome

The extra-chromosomal genome of *B. burgdorferi* B31 consists of 12 linear plasmids and nine circular plasmids that total 610 kb in size.

2.3.1 Linear plasmids

There are two linear plasmids in *B. burgdorferi* that are absolutely necessary for persistent infection of a mammalian host. These plasmids, known as lp25 and lp28-1, are relatively unstable in culture, and are commonly lost after a few generations of *in vitro* growth. Bacteria that have lost either of these two plasmids remain capable of *in vitro* growth, but lose their ability to cause persistent infection even in immunocompromised mice. The lp25 plasmid contains a gene, *pncA*, which encodes a nicotinamidase whose function is most likely the biosynthesis of NAD; by all appearances its activity is dispensable growth *in vitro* , but crucial for growth within a host. Transforming the lp25- spirochetes with *pncA* on a shuttle vector replaces the requirement of lp25 *in vivo*. Likewise, reintroduction of the entire lp25 plasmid (by transformation) into lp25- spirochetes successfully rescues infectivity. [2]

2.3.2 Circular plasmids

An unusual feature of *B. burgdorferi* is a series of related 32-kb circular plasmids, termed cp32s. These have been found to be prophage genomes, and it is believed that they play a role in the horizontal transfer of DNA among spirochetes that share a common geographical and ecological niche. [3, 4]

2.3.3 OuterSurface Proteins (Osps)

The Outer Surface Proteins (Osps) of *B. burgdorferi* are lipoproteins that play an important role in interacting with interstitial and cellular components of insect and mammalian hosts. OspA, the most studied of the Osps, is expressed on spirochetes in unfed nymphs and adult ticks, as well as in culture. OspA mediates adherence to the cells of the tick midgut, which presumably allows spirochetes to avoid endocytosis by tick gut cells during digestion of the blood meal. The ability of *Borrelia* to regulate expression of OspA indicates that it also plays a role in detachment from the midgut, which allows the bacteria to enter the mammalian host when the tick takes a second bloodmeal.

During tick feeding, *Borrelia* in the midgut upregulate expression of another outer surface protein, OspC, and begin to move toward the salivary glands. This evident correlation suggests that OspC might play a role in transmission. Once it has entered the mammalian host, *Borrelia* downregulates OspA and exhibits variable OspC upregulation patterns. Although *B. burgdorferi* possesses only one copy of the *ospC* gene, sequences vary significantly from one strain to the next, which accounts for the observed antigenic variation

between OspC proteins. The host immune system plays an important role in selecting for certain strains by eliminating the immunodominant ones.

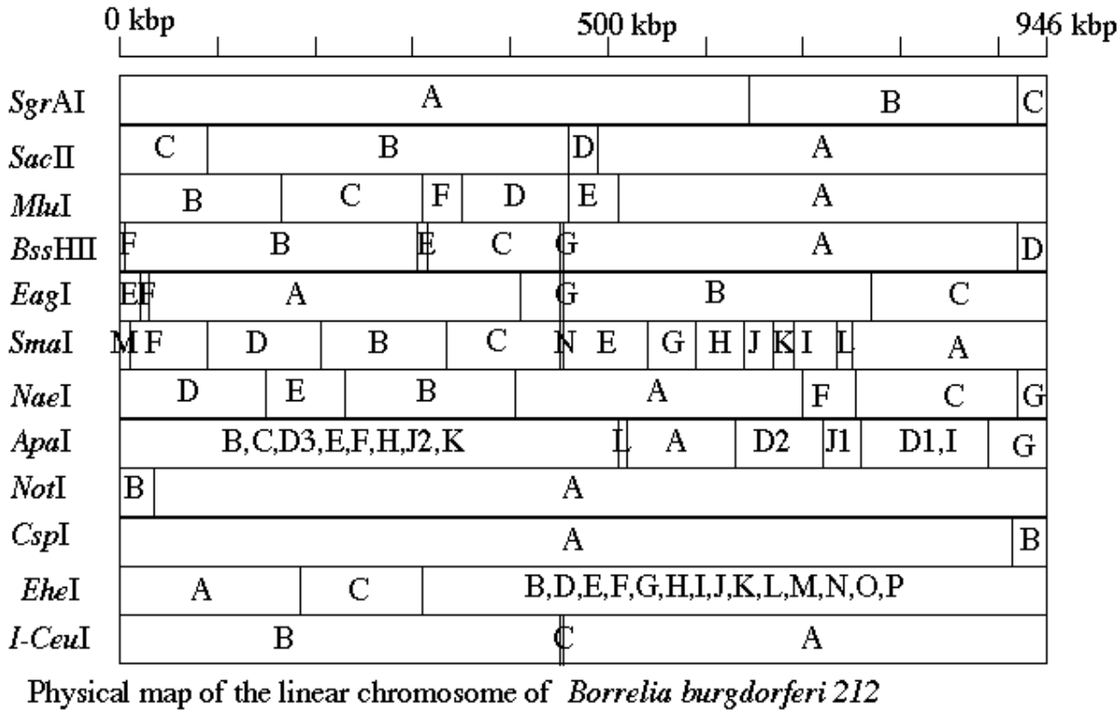


Fig. 4.

3. Genome features in *Borrelia burgdorferi*

Chromosome 910,725 bp (28.6% G+C)

Coding sequences (93%)

RNAs (0.7%)

Intergenic sequence (6.3%)

853 coding sequences
500 (59%) with identified database match
104 (12%) match hypothetical proteins
249 (29%) with no database match

Plasmids

cp9 9,386 bp (23.6% GC)
cp26 26,497 bp (26.3% GC)

lp17 16,828 bp (23.1% GC)
lp25 24,182 bp (23.3% GC)
lp28-1 26,926 bp (32.3% GC)
lp28-2 29,771 bp (31.5% GC)
lp28-3 28,605 bp (25.1% GC)
lp28-4 27,329 bp (24.4% GC)
lp36 36,834 bp (26.8% GC)
lp38 38,853 bp (26.1% GC)
lp54 53,590 bp (28.1% GC)
Coding sequences (71%)
Intergenic sequence (29%)
430 coding sequences
70 (16%) with identified database match
110 (26%) match hypothetical proteins
250 (58%) with no database match

Ribosomal RNA Chromosome coordinates

16S 444581–446118
23S 438590–441508
5S 438446–438557
23S 435334–438267
5S 435201–435312

Stable RNA

tmRNA 46973–47335
mpB 750816–751175

Transfer RNA

34 species (8 clusters,14 single genes)

*The telomeric sequences of the nine linear plasmids assembled as part of this study were not determined; estimation of the number of missing terminal nucleotides by restriction analysis suggests that less than 1,200 bp is missing in all cases. Comparisons with previously determined sequences of lp 16.9 and one terminus of lp28-1 indicate that 25, 60 and 1,200 bp are missing, respectively.

Chromosomally-encoded genes

- rRNA sequences
- fla sequences
- hbb sequences
- fesmid sequences

3.1 *Borrelia burgdorferi* rRNA sequences

Genbank mnemomic	Accession number	Description	Strain	Date of entry	Size
Gb_ba:Bbrnaopr	U03396	*alaT* (Ala-tRNA), *ileT* (Ile-tRNA), *rrs* (16S rRNA), *rrlA* & *rrlB* (23S rRNA) *rrfA* & *rrfB* (5S rRNA)	B31	10/93	11955bp
Gb_ba:Bbu44938	U44938	*rrs* (16S rRNA)	5MT	5/96	1,533bp
Gb_ba:Bor16rg	L39080	*rrs* (16S rRNA)	9MT	3/95	1,533bp
Gb_ba:Bbu44939	U44939	*rrs* (16S rRNA)	917Y	5/96	1,533bp
Gb_ba:Bb16s297	X85204	*rrs* (16S rRNA)	297	5/95	1,488bp
Gb_ba:Borrrd	L36160	*rrs* (16S rRNA)	934U	9/94	1,536bp
Gb_ba:Bor16rga	L39081	*rrs* (16S rRNA)	935T	3/95	1,542bp
Gb_ba:Borrrdq	M64309	*rrs* (16S rRNA)	1352	4/92	1,481bp
Gb_ba:Borrrd	M64310	*rrs* (16S rRNA)	20004	4/92	1,480bp
Gb_ba:Bb16srrna	X57404	*rrs* (16S rRNA)	B31	3/92	1,465bp
Gb_ba:Borssrna	M59293	*rrs* (16S rRNA)	B31	4/92	1,480bp
Gb_ba:Borrnaca	M89935	*rrs* (16S rRNA)	CA2-87	1/93	1,291bp
Gb_ba:Bb16sdk7	X85195	*rrs* (16S rRNA)	DK7	5/95	1,488bp
Gb_ba:Bb16sdk29	X85202	*rrs* (16S rRNA)	DK29	5/95	1,488bp
Gb_ba:Bb16sdunk	X85201	*rrs* (16S rRNA)	DUNKIRK	5/95	1,488bp
Gb_ba:Bbu28501	U28501	*rrs* (16S rRNA)	ESP-1	7/95	1,488bp
Gb_ba:Borrr16sa	M60967	*rrs* (16S rRNA)	G2	4/92	1,483bp
Gb_ba:Borrnail	M89936	*rrs* (16S rRNA)	Illinois 1	1/93	1,291bp
Gb_ba:Bb16skipp	X85196	*rrs* (16S rRNA)	KIPP	5/95	1,488bp
Gb_ba:Bb16slipz	X85203	*rrs* (16S rRNA)	LIPITZ	5/95	1,488bp
Gb_ba:Borrr16sc	M60969	*rrs* (16S rRNA)	Sh-2-82	4/92	1,476bp
Gb_ba:Borrnavs	M89938	*rrs* (16S rRNA)	VS219	1/93	1,350bp
Gb_ba:Borrgda	L40596	*rrs* (16S rRNA)		3/95	1,492bp

Genbank mnemomic	Accession number	Description	Strain	Date of entry	Size
Gb_ba:Borrg16s	M88329	*rrs* (16S rRNA)		11/93	1,537bp
Gb_ba:Bor23srrna	M93664	*rr1* (23S rRNA)	212	6/92	398bp
Gb_ba:Bb23s5s	X85745	*rrl rrs* (23S & 5S rRNA)	B31	7/95	2,093bp
Gb_ba:Borrg23s	M88330	*rr1* (23S rRNA)		1/93	2,926bp
Gb_ba:Bb523srr	X57791	*rrf* (5S) and *rrl* (23S rRNA genes).		6/93	616bp
Gb_ba:Borburssp	L30121	internal transcribed spacer	212	7/94	253bp
Gb_ba:Borburs2sp	L30127	internal transcribed spacer	B31	7/94	254bp
Gb_ba:Borburg7sp	L30123	internal transcribed spacer	CA2	7/94	255bp

Table 2.

3.2 *Borrelia burgdorferi* fla sequences

Genbank mnemomic	Accession number	Description	Strain	Date of entry	Size
Gb_ba:Borflass	L29234	*fla* (flagellin)	212	7/94	193bp
Gb_ba:Bbfaa	X16833	*fla* flagellum-associated 41kD antigen (flagellin)	B31	9/93	1,435bp
Gb_ba:Bbfla2	X15661	*fla* (flagellin)	B31	2/94	1,011bp
Gb_ba:Borflab31a	L29200	*fla* (flagellin)	B31	7/94	193bp
Gb_ba:Borflag	M34710	*fla* (flagellin)	B31	5/95	684bp
Gb_ba:Bbbop41	X69607	*fla* flagellum-associated 41kD antigen (flagellin)	BO	5/94	1,008bp
Gb_ba:Bbgehofla	X56334	*fla* (flagellin)	GeHo	4/93	1,426bp
Gb_ba:Bbfla	X15660	*fla* (flagellin)	GeHo	2/94	1,011bp

Genbank mnemomic	Accession number	Description	Strain	Date of entry	Size
Gb_ba:Bbflagen	X75200	*fla* (flagellin)	HB19	8/95	1,117bp
Gb_ba:Bbflagen	X75200	*fla* (flagellin)	HB19	12/93	1,117bp
Gb_ba:Bbhep41	X69609	*fla* flagellum-associated 41kD antigen (flagellin)	HE	5/94	1,011bp
Gb_ba:Bbkap41	X69611	*fla* flagellum-associated 41kD antigen (flagellin)	KA	5/94	1,008bp
Gb_ba:Bor2fla	L42881	*fla* (flagellin)	KL10	6/95	1,011bp
Gb_ba:Bor1fla	L42876	*fla* (flagellin)	NBS1ab	6/95	1,011bp
Gb_ba:Borflac	M67458	*fla* (flagellin)	PSto	5/92	226bp
Gb_ba:Bbtrop41	X69614	*fla* flagellum-associated 41kD antigen (flagellin)	TRO	5/94	1,011bp

Table 3.

3.3 *Borrelia burgdorferi hbb* sequences

Genbank mnemomic	Accession number	Description	Strain	Date of entry	Size
Gb_ba:Bbu48650	U48650	*hbb* (Histone like protein HBbu)	A44S	4/96	327bp
Gb_ba:Bbu48648	U48648	*hbb* (Histone like protein HBbu)	B31	4/96	327bp
Gb_ba:Bbu48652	U48652	*hbb* (Histone like protein HBbu)	IP1	4/96	327bp
Gb_ba:Bbu48653	U48653	*hbb* (Histone like protein HBbu)	IP2	4/96	327bp
Gb_ba:Bbu48648	U48648	*hbb* (Histone like protein HBbu)	B31	4/96	327bp
Gb_ba:Bbu48649	U48649	*hbb* (Histone like protein HBbu)	NY13-87	4/96	327bp
Gb_ba:Bbu48654	U48654	*hbb* (Histone like protein HBbu)	IP3	4/96	327bp
Gb_ba:Bbu35673	U35673	*hbb* (Histone like protein HBbu)	Sh-2-82	10/95	3,399bp

Table 4.

3.4 *Borrelia burgdorferi* fesmid sequences

Genbank mnemomic	Accession number	Description	Strain	Date of entry	Size
Gb_ba:Bbu43739	U43739	Genes noted below	B31	1/96	34,817bp

Table 5.

orf38 (open reading frame);
orf37 (open reading frame);
orf36 (open reading frame);
ylxH (putative ATP-binding protein);
flhF (flagella asociated putative GTP-binding protein protein);
flhA (flagellar protein required for flagellar formation);
flhB (flagellar protein required for flagellar formation);
fliR (flagellar protein required for flagellar formation);
fliQ (flagellar protein required for flagellar formation);
fliP (flagellar protein required for flagellar formation);
fliZ (flagellar protein required for flagellar formation);
fliN (flagellar switch protein);
fliM (flagellar switch protein);
orf25 (open reading frame);
motB (flagellar motor rotation protein B);
motA (flagellar motor rotation protein A);
flgE (flagellar hook protein);
ylxG (flagellar synthesis);
orf20 (open reading frame);
orf19 (open reading frame);
orf18 (open reading frame);
fliI (flagellar synthesis);
fliH (flagellar synthesis);
fliG (flagellar switch protein);
fliF (flagella basal-body M ring protein);
fliE (flagella basal-body protein);
flgC (flagella associated rod protein);
flgB (flagella associated rod protein);
hslU heat shock protein);
hslV (heat shock protein);
smg (?);
orf7 (open reading frame);
ftsZ (cell division protein);
ftsA (cell division protein);
divIB (cell division protein);
ftsW (cell division protein);
mraY (phosphotransferase);
murF (pentapeptide presynthetase)

3.5 *Borrelia burgdorferi* chromosomal sequences (Except rrn & fla genes)

Genbank mnemomic	Accession number	Description	Strain	Date of entry	Size
Gb_ba:Borp39ant	L24194	*bmpA bmpB* (immunodo-minant antigen P39 gene)	Sh-2-82	7/94	2,304bp
Gb_ba:Borbmpa	L35050	*bmpA bmpB* (membrane lipoproteins A & B	212	12/94	904bp
Gb_ba:Borbmpc	L34547	*bmpC* (membrane lipoprotein C)	297	11/94	1,293bp
Gb_ba:Bbu35450	L34547	*bmpD* (membrane lipoprotein D)	297	4/96	1,525bp
Gb_ba:Bbcheagen	X91907	*cheA1* (histidine kinase)	212	9/95	332bp
Gb_ba:Bbu28962	U28962	*cheA1* (histidine kinase)	CT-1	6/95	2,491bp
Gb_ba:Borchea	L39965	*cheA2* (histidine kinase)	B31	8/95	2,410bp
Gb_ba: Bbu34384	U34384	*cheW* (Positive regulator of CheA activity)	CT-1	9/95	660bp
Gb_ba:Bbu04527	U04527	*dnaA* (DNA replication initiatior), *dnaN* (DNA polymerase III beta subunit),*gyrB* (DNA gyrase B subunit), *rpmH* (ribosomal pro-tein L34) and *rnpA* (ribonuclease P protein component)	212	2/94	4943bp
Gb_ba:Borgrpepls	M96847	*dnaJ dnaK*and *grpE* (heat shock proteins)		2/93	3913bp
Gb_ba:Bordnaj	M97914	*dnaJ*(heat-shock protein)	CA12	12/92	1,094bp
Gb_ba:Borhsp70a	M97912	*dnaK* (70 kDa heat shock protein)	CA12	10/92	1,928bp
Gb_ba:S42385	S42385	*dnaK* (70 kDa heat shock protein)	CA12	10/92	1,911bp
Gb_ba:Bbhspro	X67646	*dnaK* (70 kDa heat-shock protein)	ZS7	8/92	2,116bp
Gb_ba:Bbu12870	U12870	*flgE* (flagellar hook polypeptide)	N40	4/95	1,552bp
Gb_ba:Bbu19712	U19712	*flgE* (flagellar hook polypeptide)	B31	1/95	571bp
Gb_ba:Borflge	L43849	*flgE* (flagellar hook polypeptide)	HB19	8/95	1499bp
Gb_ba:Borflif	L40501	*fliF* (Flagellar MS-ring protein)	212	2/96	1717bp
Gb_ba:BBU09711	U09711	*fliG* (Flagellar switch protein)	212	7/95	1035bp
Gb_ba:Borflih	L40502	*fliH* (export of flagellar proteins?)	212	1/96	921 bp
Gb_ba:Borflii	L43325	*fliI* (export of flagellar proteins?)	212	1/96	1311 bp

Genbank mnemomic	Accession number	Description	Strain	Date of entry	Size
Gb_ba:Bbftszg	Z12164	*ftsZ* (cell division protein)	212	5/94	261bp
Gb_ba:Bbu28760	U28760	*gapDH* (glyceraldehyde-3-phosphate dehydro-genase); *pgk* phosphogly-cerate kinase; *tpi* triose-phosphate isomerase	B31	6/95	798bp
Gb_ba:Bbgidag	Z12160	*gidA* (glucose inhibited division protein)	212	5/94	196bp
Gb_ba:Bbhsp60	X65139	*groEL*(common antigen)	ZS7	5/92	1,931bp
Gb_ba:Bbgyrag	Z12165	*gyrA* (DNA gyrase subunit A)	212	5/94	289bp
Gb_ba:Bbgyrbg	Z12166	*gyrB*(DNA gyrase subunit B)	212	5/94	253bp
Gb_ba:Borhtpg	L32145	*htpG* (C62.5 heat shock protein)	212	12/94	236bp
Gb_ba:Borlonaa	L77216	*lon* ATP-dependent protease	B31	4/96	2,946bp
Gb_ba:Bormetg	L32146	*metG* (methionyl tRNA synthetase)	212	12/94	346bp
Gb_ba:Borplsctop	L32861	*parE* (topoisomerase IV, B subunit), *plsC* (1-acyl-sn-glycerol-3-phosphate acetyltransferase)	212	5/94	677bp
Gb_ba:Bbysc1	X78708	*pep* APE1 (aminopepti-dase 1 homologue)	ZS7	4/94	1,776bp
Gb_ba:Borpgktpi	L32595	*pgk* (phosphoglycerate kinase), *tpi* (triose-phosphate isomerase)	212	5/94	370bp
Gb_ba:Borpthh	L32144	*pth* (peptidyl-tRNA hydrolase)	212	4/94	910bp
Gb_ba:Bbu23457	U23457	*recA* General recombi-nation & DNA repair	Sh-2-82	4/96	2,025bp
Gb_ba:Borrho	L07656	*rho* (Rho protein)	Sh-2-82	9/93	1,499bp
Gb_ba:Borrpob	L48488	*rpoB rpoC* (RNA polymerase, beta & beta prime subunits)	B31	11/95	3,682bp
Gb_ba:Borrhoa	L46347	*rho* (Rho protein)	212	8/95	571bp
Gb_ba:Bbu35673	U35673	*rpsT* (30S ribosomal protein S20)	Sh-2-82	10/95	3,399bp
Gb_ba:Bbrnasep	U17591	*rpoD* (primary sigma factor)	B31	12/94	4,165bp
Gb_ba:Bortufz	L23125	*tuf* (elongation factor EF-Tu)	B31	8/93	1,230bp

Table 6.

3.6 *Borrelia burgdorferi* chromosomal sequences (Antigens and proteins of unknown function)

Genbank mnemomic	Accession number	Description	Strain	Date of entry	Size
Gb_ba:Bororf	L32797	p21 (21 kDa protein)		5/95	1,152bp
Gb_ba:Borunk	L31615	p21A (21 kD protein)	297	8/94	700bp
Gb_ba:Bor22kdant	M90084	p22 (22 kD antigen)	B31	10/93	795bp
Gb_ba:Borp22x	L22530	p22X (22 kD outer surface lipoprotein)	N40	8/94	585bp
Gb_ba:Borp23a	L31616	p23 (23kD protein)	297	8/94	686bp
Gb_ba:Bbhypp	X63898	p38 (38 kD ATP-binding protein)	GeHo	2/92	1,435bp
Gb_ba:Bdna66kd	X87725	p66 (66 kD protein)	B31	6/95	2,180bp
Gb_ba:Borlyme	L32596	p66 (66 kD protein)	212	6/92	240bp
Gb_ba:Bbp831001	X81514	p93/p100 (93 kD protein)	297	7/95	287bp
Gb_ba:Bbbop93	X69601	p93 (93 kD protein)	BO	12/93	1,991bp
Gb_ba:Bbp97	X77749	p97 (97 kD protein)	GOE2	6/95	2,082bp
Gb_ba:Bbp831002	X81520	p93/p100 (93 kD protein)	pacificus	7/95	269bp
Gb_ba:Bbp83100	X81357	p83/p100 (100 kD protein)	PBre	4/96	287bp
Gb_ba:Bbp831003	X81528	p93/p100 (93 kD protein)	PKa2	7/95	287bp
Gb_ba:Bbp831004	X81531	p93/p100 (93 kD protein)	T255	7/95	287bp
Gb_ba:Bbtrop93	X69604	p93 (93 kD protein)	TRO	12/93	2,081bp
Gb_ba:Borsurant	L36037	surface antigen	Dk1	9/94	185bp
Gb_ba:Bbla7	X70826	LA7 (21 kD lipoprotein)	ZS7	11/93	821bp
Gb_ba:	X91965	*abp* (probable ATP binding protein)	212	9/95	285bp
Gb_ba:Boraaa	M60802	immunogen gene		12/92	2,258bp
Gb_ba:Bbu18292	U18292	"bbk2.10 gene"	297	7/95	1,799bp
Gb_ba:Bbu19105	U19105	"bbk2.10 gene"	N40	7/95	832bp
Gb_ba:Borlyme	L32596	PCR target	212	8/94	240bp

Genbank mnemomic	Accession number	Description	Strain	Date of entry	Size
Gb_ba:Borseqa	M58429	PCR target		3/91	379bp
Gb_ba:Borseqc	M58431	PCR target		3/91	1725bp
Gb_ba:Borseqd	M58432	PCR target		3/91	381bp
Gb_ba:Borseqe	M58433	PCR target		3/91	379bp
Gb_ba:Bbu35673	U35673	*orfH, orfR and hbbU* (putative proteins)	Sh-2-82	10/95	3,399bp

Table 7.

Plasmid-encoded genes

- ospA genes
- ospB genes
- ospC genes
- ospD genes

3.7 *Borrelia burgdorferi* Plasmid-encoded sequences (Except ospA, ospB, ospC & ospD)

Genbank mnemomic	Accession number	Description	Plasmid	Strain	Date of entry	Size
Gb_ba: Borgmpguaa	L25883	*guaA* (GMP synthetase)	26 kb cp	CA-11.2A	11/94	1,599bp
Gb_ba:Bbu13372	U13372	*guaB* (IMP dehydrogenase)	26 kb cp	CA-11.2A	11/94	1,212bp
Gb_ba:Borospea	L13924	*ospE* (outer surface protein E)	45kb lp	N40	3/94	644bp
Gb_ba:Borospfa	L13925	*ospF* (outer surface protein F)	45kb lp	N40	3/94	785bp
Gb_ba:Bbu19754	U19754	*ospF* (outer surface protein F)	45kb lp	297	7/95	690bp
Gb_ba:Bbospg	X82409	*ospG* & *bapA* (outer surface protein G & associated protein A)	48b lp	ZS7	11/95	1524bp
Gb_ba:Bbu22451	U22451	p12 (12kDa lipoprotein)	49kb lp	B31	3/95	285bp
Gb_ba: Borexpprtn	L16625	p20 (exported neuro-toxin-like protein)	9kb cp	B31	8/94	720bp
Gb_ba:S66708	S66708	PCR target sequence	30kb cp	B31	11/95	416bp

Genbank mnemomic	Accession number	Description	Plasmid	Strain	Date of entry	Size
Gb_ba:Bors1a	L34016	S1 antigen	49kb lp	N40	11/95	1,421bp
Gb_ba:Bors2a	L34016	S2 antigen	49kb lp	N40	11/95	837bp
Gb_ba: Bbptl4916	X53311	telomeres pTL16 and pTL49	16kb lp & 49kb lp		2/93	238bp
Gb_ba:Bbptr16	X53312	telomere pTR16	16 kb lp		2/93	191bp
Gb_ba:S65114	S65114	left terminal repeat, telomeric fragment	lp		7/92	38bp
Gb_ba:	X87127	repeated DNA element	30.5kb cp		4/96	5,500bp

Table 8.

3.8 *Borrelia burgdorferi* ospA sequences

Genbank mnemomic	Accession number	Description	Strain	Date of entry	Size
Gb_ba:Bb297ospa	X85442	*ospA* (outer surface protein A)	297	8/95	822bp
Gb_ba:Borospad	L23138	*ospA ospB* (outer surface proteins A & B)	19535NY2	8/94	1,653bp
Gb_ba:Borospah	L23141	*ospA ospB* (outer surface proteins A & B)	21343WI	8/94	1,653bp
Gb_ba:Borospac	L23137	*ospA ospB* (outer surface proteins A & B)	27985CT2	6/94	1,653bp
Gb_ba:Borospaf	L23140	*ospA ospB* (outer surface proteins A & B)	41552MA	8/94	1,653bp
Gb_ba:Borospae	L23139	*ospA ospB* (outer surface proteins A & B)	42373NY3	8/94	1,653bp
Gb_ba:Borospaa	L23136	*ospA ospB* (outer surface proteins A & B)	B19CT1	6/94	1,653bp
Gb_ba:Bbospab	X14407	*ospA ospB* (outer surface proteins A & B)	B31	9/94	1,915bp
Gb_ba:Boropsab	L19701	*ospA ospB* (outer surface proteins A & B)	B31	6/93	1,916b

Genbank mnemomic	Accession number	Description	Strain	Date of entry	Size
Gb_ba:Borospai	L23142	*ospA ospB* (outer surface proteins A & B)	CA3	8/94	1,653bp
Gb_ba:Borospaj	L23143	*ospA ospB* (outer surface proteins A & B)	CA7	8/94	1,653bp
Gb_ba:Borospak	L23144	*ospA ospB* (outer surface proteins A & B)	CA8	8/94	1,653bp
Gb_ba:Bbdk6ospa	X83622	*ospA* (outer surface protein A)	DK6	1/95	822bp
Gb_ba:Bbpospa	X63412	*ospA* (outer surface protein A)	DK29	1/94	825bp
Gb_ba:Bormajospr	L19702	*ospA* (outer surface protein A)	G2	6/93	2,123bp
Gb_ba:Bbaspa	X60300	*ospA* (outer surface protein A)	Goe2	12/92	1,361bp
Gb_ba:Borospaa	L23136	*ospA ospB* (outer surface proteins A & B)	HB19CT1	6/94	1,653bp
Gb_ba:Bbospa3	X65600	*ospA* (outer surface protein A)	HE	1/94	822bp
Gb_ba:Bbu33179	U33179	*ospA* (outer surface protein A)	HT29	9/95	270bp
Gb_ba:Bbopsaa	X70365	*ospA* (outer surface protein A)	IP3	5/94	822bp
Gb_ba:Bbka0spa	X69606	*ospA* (outer surface protein A)	KA	5/94	822bp
Gb_ba:Bbospcmul	X84779	*ospA* (outer surface protein A)	MUL	5/95	534bp
Gb_ba:Borfra	L38657	*ospA* (outer surface protein A)	N3	1/95	822bp
Gb_ba:Borospa	M57248	*ospA* (outer surface protein A)	N40	11/91	819bp
Gb_ba:Bbdnaospa	X85739	*ospA* (outer surface protein A)	PBre	9/95	822bp
Gb_ba:Bbpheiosp	X80251	*ospA* (outer surface protein A)	PHei	9/95	822bp
Gb_ba:Bbpkaospa	X80182	*ospA* (outer surface protein A)	PKa	9/95	822bp
Gb_ba:Bbpwud1	X80184	*ospA* (outer surface protein A)	PWud1	9/95	822bp
Gb_ba:Bbpwudi	X68540	*ospA* (outer surface protein A)	PWudI	3/93	333bp
Gb_ba:Bbpwudl6	X80185	*ospA* (outer surface protein A)	PWud1/6	9/95	822bp
Gb_ba:Bbpwudll	X80253	*ospA* (outer surface protein A)	PWud11	9/95	825bp

Genbank mnemomic	Accession number	Description	Strain	Date of entry	Size
Gb_ba:Bbpwudii	X68539	*ospA* (outer surface protein A)	PWudII	3/93	333bp
Gb_ba:Bor90ospa	L42873	*ospA* (outer surface protein A)	SIMON	6/95	582bp
Gb_ba:Bbt25ospa	X85443	*ospA* (outer surface protein A)	T255	9/95	822bp
Gb_ba:Borospaab	D29660	*ospA* (outer surface protein A)	tick isolate	4/95	911bp
Gb_ba:Bbospa1	X65598	*ospA* (outer surface protein A)	TRO	1/94	822bp
Gb_ba:Bbospa	X16467	*ospA* (outer surface protein A)	ZS7	9/93	942bp
Gb_ba:A22442	A22442	*ospA* (outer surface protein A)	ZS7	12/94	822bp
Gb_ba:Bbosproa	X66065	*ospA* (outer surface protein A)	ZQ1	7/93	825bp
Gb_ba:A24006	A24006	*ospA* (outer surface protein A)	ZQ1	2/95	825bp
Gb_ba:A04009	A04009	*ospA ospB* (outer surface proteins A & B)		4/93	1,915bp

Table 9.

3.9 *Borrelia burgdorferi* ospB sequences

Genbank mnemomic	Accession number	Description	Strain	Date of entry	Size
Gb_ba:Borospad	L23138	*ospA ospB* (outer surface proteins A & B)	19535NY2	8/94	1,653bp
Gb_ba:Borospah	L23141	*ospA ospB* (outer surface proteins A & B)	21343WI	8/94	1,653bp
Gb_ba:Borospac	L23137	*ospA ospB* (outer surface proteins A & B)	27985CT2	6/94	1,653bp
Gb_ba:Borospaf	L23140	*ospA ospB* (outer surface proteins A & B)	41552MA	8/94	1,653bp
Gb_ba:Borospae	L23139	*ospA ospB* (outer surface proteins A & B)	42373NY3	8/94	1,653bp
Gb_ba:Boropsab	L19701	*ospA ospB* (outer surface proteins A & B)	B31	6/93	1,916b
Gb_ba:Bbospab	X14407	*ospA ospB* (outer surface proteins A & B)	B31	9/94	1,915bp

Genbank mnemomic	Accession number	Description	Strain	Date of entry	Size
Gb_ba:Bbospbb31	X74808	*ospB* (outer surface protein B)	B31	7/94	934bp
Gb_ba:Bbospbev	X74810	*ospB* (outer surface protein B)	B31/EVB	7/94	934bp
Gb_ba:Bbospbbp	X74809	*ospB* (outer surface protein B)	BEP4	7/94	934bp
Gb_ba:Borospai	L23142	*ospA ospB* (outer surface proteins A & B)	CA3	8/94	1,653bp
Gb_ba:Borospaj	L23143	*ospA ospB* (outer surface proteins A & B)	CA7	8/94	1,653bp
Gb_ba:Borospak	L23144	*ospA ospB* (outer surface proteins A & B)	CA8	8/94	1,653bp
Gb_ba:Borospbvr	L31399	*ospB* (outer surface protein B)	HB19	3/95	891bp
Gb_ba:Borospaa	L23136	*ospA ospB* (outer surface proteins A & B)	HB19CT1	6/94	1,653bp
Gb_ba:A04009	A04009	*ospA ospB* (outer surface proteins A & B)		4/93	1,915bp

Table 10.

3.10 *Borrelia burgdorferi* ospC sequences

Genbank mnemomic	Accession number	Description	Strain	Date of entry	Size
Gb_ba:Bbospc272	X84785	*ospC* (outer surface protein C)	272	5/95	534bp
Gb_ba:Bbu08284	U08284	*ospC* (outer surface protein C)	297	9/94	579bp
Gb_ba:Bor26ospc	L42893	*ospC* (outer surface protein C)	297	6/95	576bp
Gb_ba:Bbu01892	U01892	*ospC* (outer surface protein C)	2591	1/94	824bp
Gb_ba:Bor32ospc	L42899	*ospC* (outer surface protein C)	21347	6/95	576bp
Gb_ba:Bor30ospc	L42897	*ospC* (outer surface protein C)	26815	6/95	579bp
Gb_ba:Bor29ospc	L42896	*ospC* (outer surface protein C)	27579	6/95	573bp
Gb_ba:Bor28ospc	L42895	*ospC* (outer surface protein C)	28354	6/95	579bp
Gb_ba:Bor27ospc	L42894	*ospC* (outer surface protein C)	28691	6/95	573bp

Genbank mnemomic	Accession number	Description	Strain	Date of entry	Size
Gb_ba:Bbb31ospc	X69596	*ospC* (outer surface protein C)	B31	5/93	633bp
Gb_ba:Bbu01894	U01894	*ospC* (outer surface protein C)	B31	1/94	980bp
Gb_ba:Borospca	D49497	*ospC* (outer surface protein C)	B31	5/95	633bp
Gb_ba:Bbospcbur	X84765	*ospC* (outer surface protein C)	BUR	5/95	534bp
Gb_ba:Borospc	L25413	*ospC* (outer surface protein C)	CA-11.2A	7/94	1,150bp
Gb_ba:Bbospce	X73626	*ospC* (outer surface protein C)	DK6	2/94	609bp
Gb_ba:Bbospcd	X73625	*ospC* (outer surface protein C)	DK7	2/94	618bp
Gb_ba:Bbospcc	X73624	*ospC* (outer surface protein C)	DK26	2/94	624bp
Gb_ba:Bbospcb	X73623	*ospC* (outer surface protein C)	DK27	2/94	624bp
Gb_ba:Bbospcduk	X84778	*ospC* (outer surface protein C)	DUNKIRK	5/95	528bp
Gb_ba:Bbu04281	U04281	*ospC* (outer surface protein C)	HB19	1/95	692bp
Gb_ba:Bor20ospc	L42887	*ospC* (outer surface protein C)	Ip2	6/95	576bp
Gb_ba:Bbospckip	X84782	*ospC* (outer surface protein C)	KIPP	5/95	534bp
Gb_ba:Bbu04240	U04240	*ospC* (outer surface protein C)	N40	8/94	689bp
Gb_ba:Bbdnaospc	X83555	*ospC* (outer surface protein C)	pacificus	6/95	630bp
Gb_ba:Bbospc1	X81522	*ospC* (outer surface protein C)	PBre	6/95	636bp
Gb_ba:Bbpkaospc	X69589	*ospC* (outer surface protein C)	PKa	2/94	633bp
Gb_ba:Bbt25ospc	X69592	*ospC* (outer surface protein C)	T25	2/94	636bp
Gb_ba:Bbospc2	X81524	*ospC* (outer surface protein C)	T255	5/95	633bp
Gb_ba:Bbospctxw	X84783	*ospC* (outer surface protein C)	TXGW	5/95	531bp
Gb_ba:Bbwudospc	X69590	*ospC* (outer surface protein C)	WudI	2/94	639bp
Gb_ba:Bor40ospc	L42868	*ospC* (outer surface protein C)	ZS7	6/95	579bp

Table 11.

3.11 *Borrelia burgdorferi* ospD sequences

Genbank mnemomic	Accession number	Description	Strain	Date of entry	Size
Gb_ba:Bbu05304	U05304	*ospD* (outer surface protein D)	3028	11/94	1,012bp
Gb_ba:Bbu05305	U05305	*ospD* (outer surface protein D)	27985	11/94	1,012bp
Gb_ba:Borospd	M97452	*ospD* (outer surface protein D)	B31	2/93	1,079bp
Gb_ba:Bbu05324	U05324	*ospD* (outer surface protein D)	CA12	11/94	991bp
Gb_ba:Borospdhb	L34055	*ospD* (outer surface protein D)	HB19	6/94	1,045bp
Gb_ba:Bbu05327	U05327	*ospD* (outer surface protein D)	lp7	11/94	1,064bp

Table 12.

3.12 *Borrelia burgdorferi* fusion sequences

Genbank mnemomic	Accession number	Description	Strain	Date of entry	Size
Gb_ba:A24010	A24010	*ospA* fusion	NS1	2/95	1,020bp
Gb_ba:A24012	A24012	*ospA* fusion	NS1	2/95	1,014bp
Gb_ba:A24014	A24014	*ospA* fusion	NS1	2/95	1,017bp
Gb_ba:A24016	A24016	*ospA* fusion	NS1	2/95	1,017bp
Gb_ba:Borbb1	L31427	*phoA* fusion	297	4/95	279bp
Gb_ba:Borbb10	L31421	*phoA* fusion	297	4/95	319bp
Gb_ba:Borbb11	L31424	*phoA* fusion	297	4/95	248bp
Gb_ba:Borbb13	L31422	*phoA* fusion	297	4/95	354bp
Gb_ba:Borbb14	L31423	*phoA* fusion	297	4/95	361bp
Gb_ba:Borbb16	L31425	*phoA* fusion	297	4/95	135bp
Gb_ba:Borbb17	L31426	*phoA* fusion	297	4/95	615bp
Gb_ba:Borbb4	L31417	*phoA* fusion	297	4/95	294bp
Gb_ba:Borbb4a	L31419	*phoA* fusion	297.	4/95	221bp
Gb_ba:Borbb5	L31418	*phoA* fusion	297	4/95	341bp
Gb_ba:Borbb9	L31420	*phoA* fusion	297	4/95	233bp

Table 13.

3.13 *Borrelia burgdorferi* promoter sequences

Genbank mnemomic	Accession number	Description	Strain	Date of entry	Size
Gb_ba:Borproma	M28680	promoter	B31	6/90	194bp
Gb_ba:Borpromb	M28681	promoter	B31	6/90	203bp
Gb_ba:Borpromc	M28682	promoter	B31	6/90	78bp

Table 14.

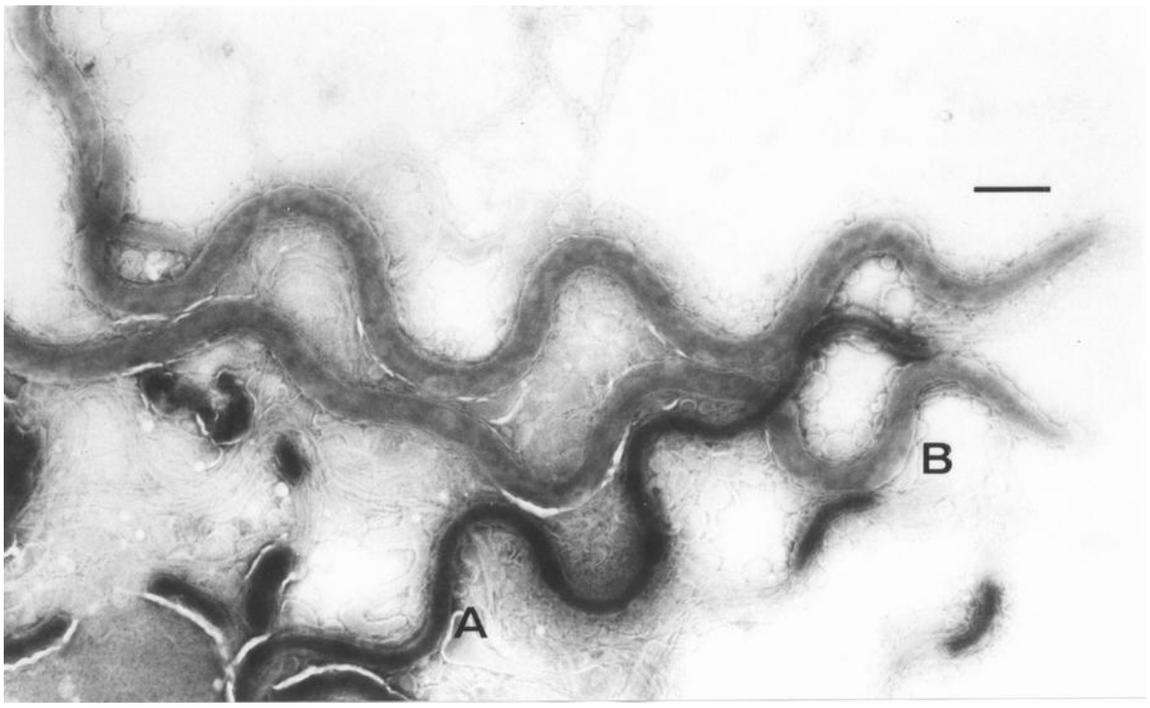


Fig. 5. Electron microscopy of unfixed, negative stained DK1 strain (skin isolate). This strain consist of two morphologically distinct borrelia A. small and B. larger *borrelia*. Bar 1 mm. Magnification 10,260 x.



Fig. 6. Electron microscopy of unfixed, negative stained DK1 strain (skin isolate).

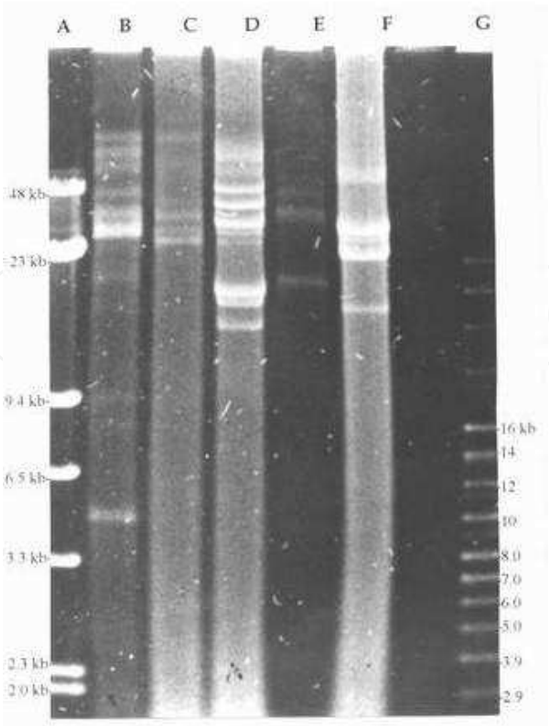


Fig. 7. Plasmids isolated from different strains of *Borrelia burgdorferi* : The Dk1 strain (B), Dk5 strain (C), DK6 strain (D), DK 2 strain (E), DK7 strain (F) and a super coiled circular molecular weight marker (G). Linear molecular markers (A) (HindIII fragments of Lambda DNA). Samples were separated in 0.3% gel at 14°C for 20 hr then stained with ethidium bromide.

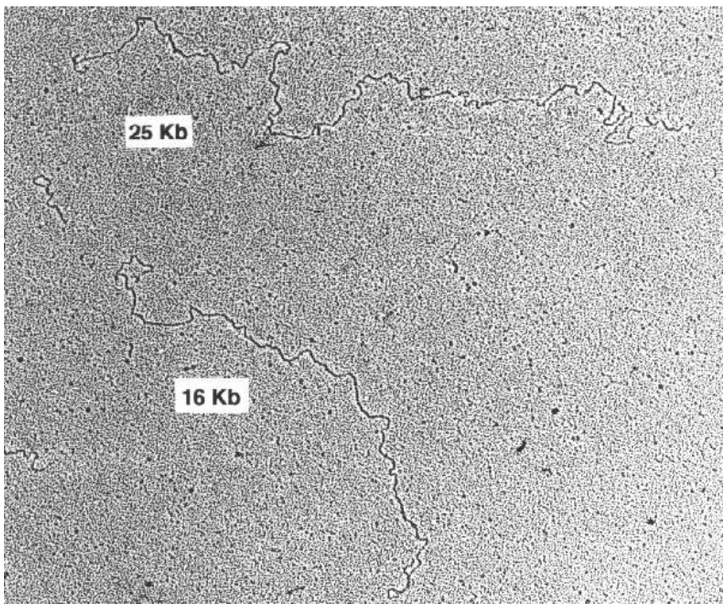


Fig. 8. Elrectron micrograph of 25 kb plasmid extracted from DK1 strain . One supercoiled plasmid. magnification 52000 x.

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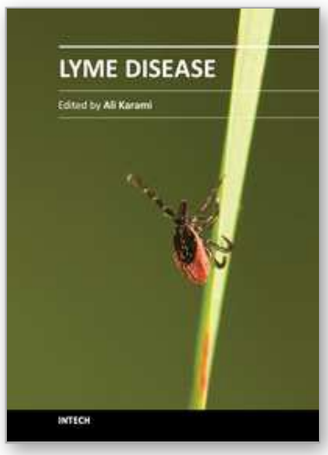
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Lyme disease, or Lyme borreliosis, is an emerging infectious disease caused by bacteria belonging to the genus borrelia. Borrelia burgdorferi, in the strict sense. This book deals mostly with the molecular biology of the Lyme disease agent orrelia burgdorferi. It has been written by experts in the relevant field and is tailored to the need of researchers, advanced students of biology, molecular biology, molecular genetics of microorganism. It will also be of use to infectious disease experts and people in other disciplines needing to know more about Lyme borreliosis. The book contains chapters on the molecular biology of the Lyme disease agent, zoonotic peculiarities of Bb, advancement in Bb antibody testing, the serology diagnostic schemes in Bb, discovering Lyme disease in ticks and dogs, adaptation to glucosamine starvation in Bb, and porins in the genus borrelia.

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