3010BBS Molecular & Applied Microbiology (Assignment 1)

Background:

E. coli 16S rRNA sequence is 1548 nucleotide in length and is numbered from position 1 to 1548. In a cell, the 16S rRNA exists as secondary, tertiary and quaternary structures and is involved in protein synthesis. *E. coli* 16S rRNA secondary structure together with the numbering scheme is used as a template against which all other bacterial 16S rRNA sequences are compared and aligned.

You have been provided with secondary structures of the 16S rRNA of *E. coli*. You have to complete the following tasks for the assignment.

Task 1

The following are short sequences from the 16S rRNA molecules of five members of domain Bacteria.

(i) *Escherichia coli*	CUGCCAGUGAUAAACUGGAGGAA
(ii) *Thermus aquaticus*	CUGCCUGCGAAAGCAGGAGGAA
(iii) *Bacillus cereus*	CUGCCGGUGACAAACCGGAGGAA
(iv) *Campylobacter jejuni*	CUGCCUUCGUAAGGAGGAGGAA
(v) *Neisseria gonorrhoeae*	CUGCCGGUGACAAGCCGGAGGAA

(a) For *E. coli* the sequence begins at position 1161. Identify and write the position at which the sequence ends.

(b) You now have identified the sequence start and end positions which relates to the 16S rRNA secondary structure of *E. coli*. Using this information as a guide, draw the corresponding secondary structures for each of the remaining sequences (ii to v).

(c) Align the sequences shown in (a) by introducing gaps (-) using the secondary structures as a guideline. This type of alignment is known as "aligning sequences using secondary structure constraints"

(d) Align the sequences without the constraint, that is introduce gaps (-) randomly so that the nucleotides of all the sequence match.

(e) Can you se the difference between the alignment in (c) and (d).

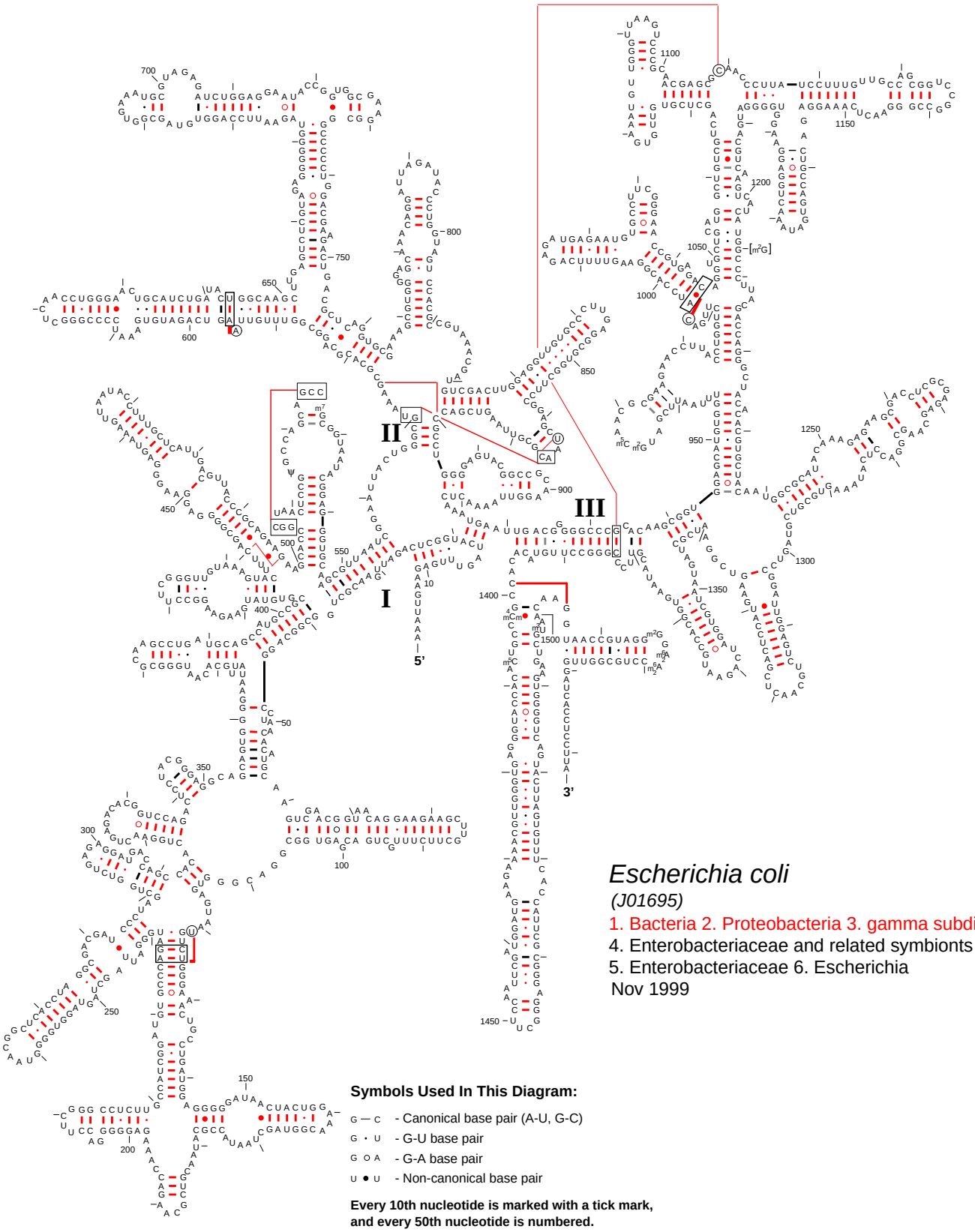
(f) The following sequence is from *Bacteroides fragilis* and is the same region as for the sequences shown in (a). Draw the secondary structure and modify your alignment to accommodate this new sequence.

Bacteroides fragilis CUGCCGUCGUAAGAUGUGAGGAA

Task 2

From the *E. coli* secondary structure, extract the sequence from positions 826 to 860. Extract the sequence for the same secondary structure region for the 5 other organisms and align all six sequences. See Fig

Secondary Structure: small subunit ribosomal RNA



Escherichia coli

(J01695)

1. Bacteria 2. Proteobacteria 3. gamma subdivision
4. Enterobacteriaceae and related symbionts
5. Enterobacteriaceae 6. Escherichia
Nov 1999

Symbols Used In This Diagram:

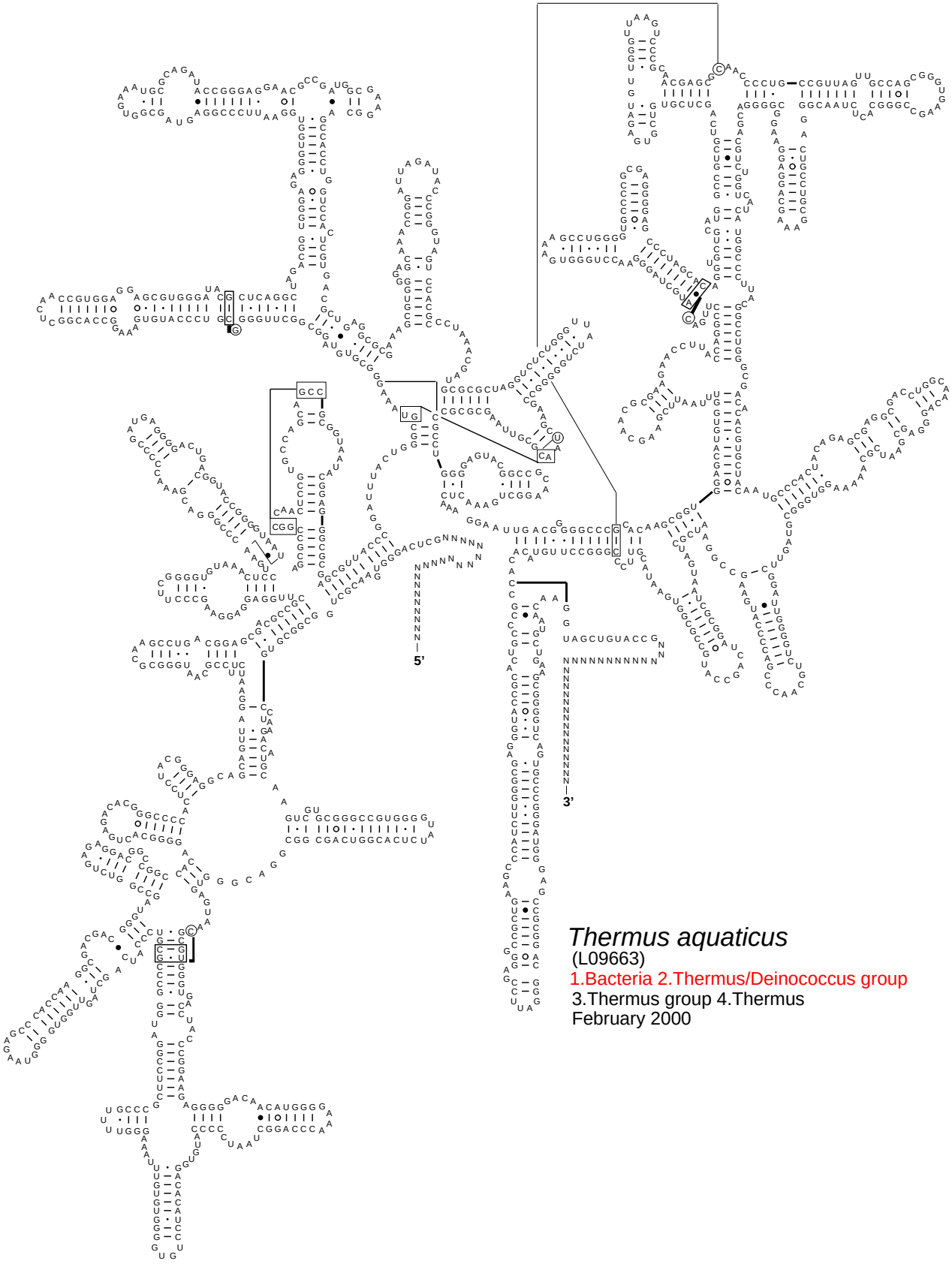
G — C - Canonical base pair (A-U, G-C)
G • U - G-U base pair
G o A - G-A base pair
U ● U - Non-canonical base pair

Every 10th nucleotide is marked with a tick mark, and every 50th nucleotide is numbered.

Tertiary interactions with strong comparative data are connected by solid lines.

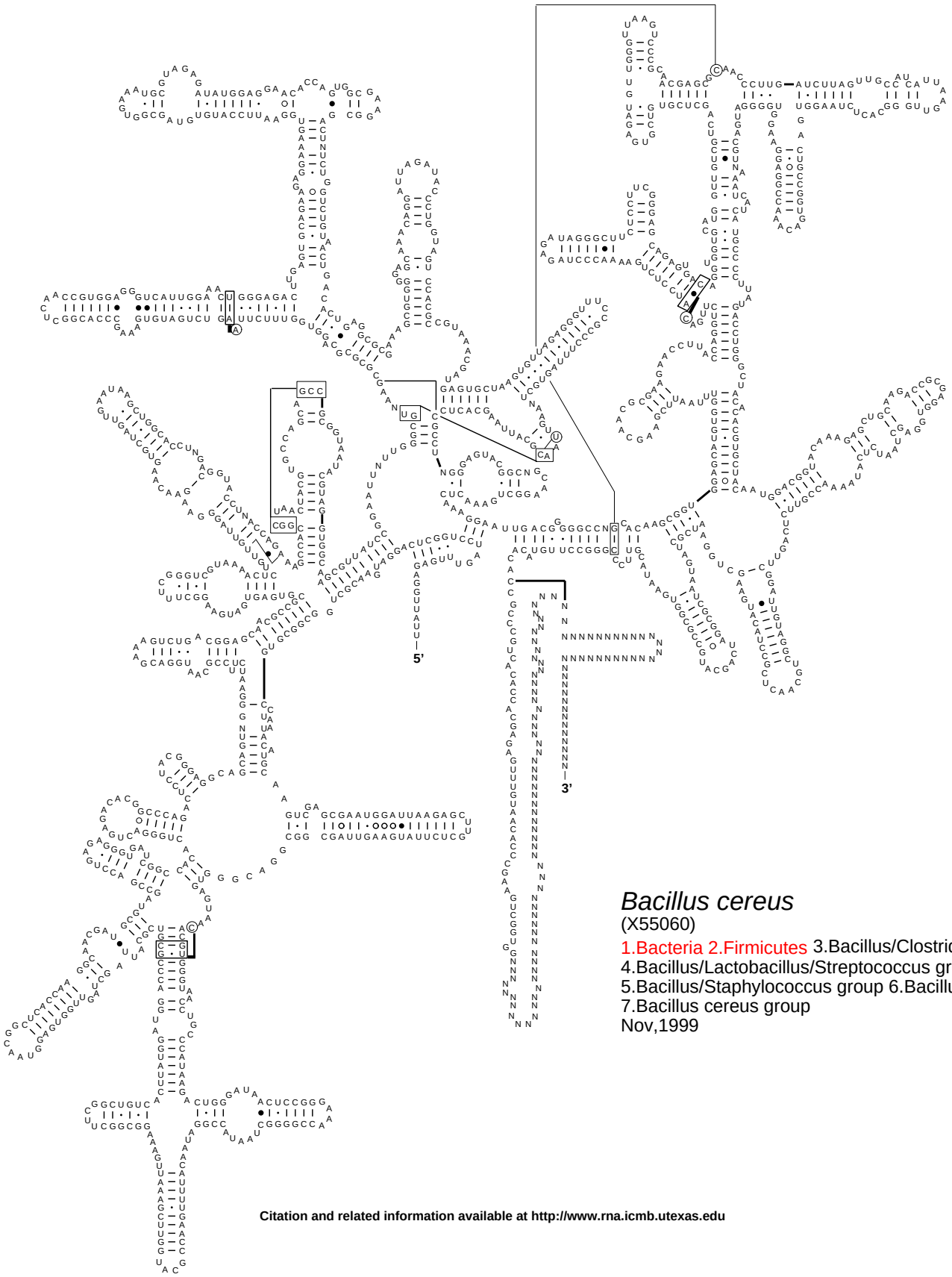
Citation and related information available at http://www.rna.icmb.utexas.edu

Secondary Structure: small subunit ribosomal RNA



Thermus aquaticus
(L09663)
1.Bacteria 2.Thermus/Deinococcus group
3.Thermus group 4.Thermus
February 2000

Secondary Structure: small subunit ribosomal RNA

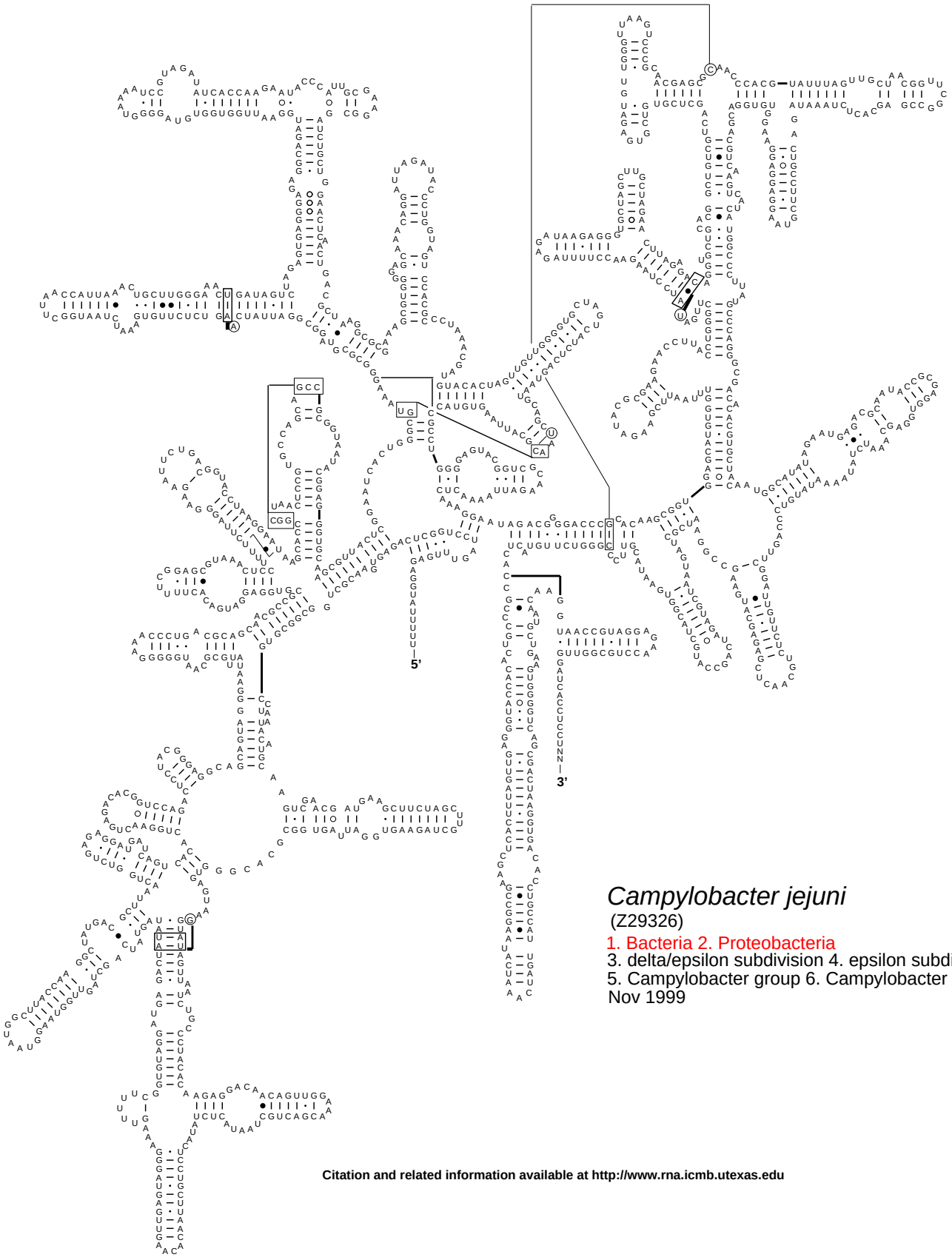


Bacillus cereus
(X55060)

1.Bacteria 2.Firmicutes 3.Bacillus/Clostridium group
4.Bacillus/Lactobacillus/Streptococcus group
5.Bacillus/Staphylococcus group 6.Bacillus
7.Bacillus cereus group
Nov,1999

Citation and related information available at http://www.rna.icmb.utexas.edu

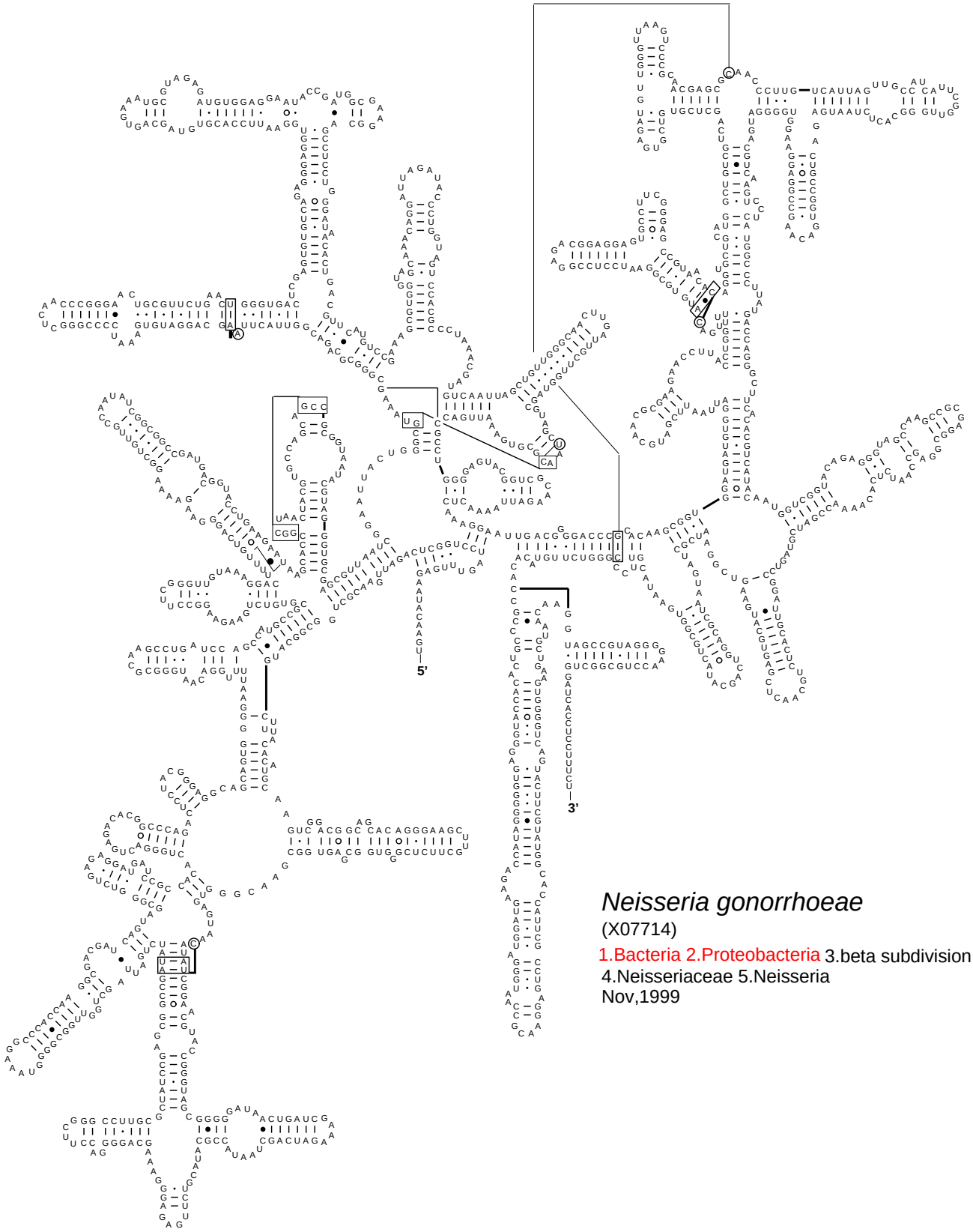
Secondary Structure: small subunit ribosomal RNA



Campylobacter jejuni
(Z29326)
1. Bacteria 2. Proteobacteria
3. delta/epsilon subdivision 4. epsilon subdivision
5. Campylobacter group 6. Campylobacter
Nov 1999

Citation and related information available at http://www.rna.icmb.utexas.edu

Secondary Structure: small subunit ribosomal RNA



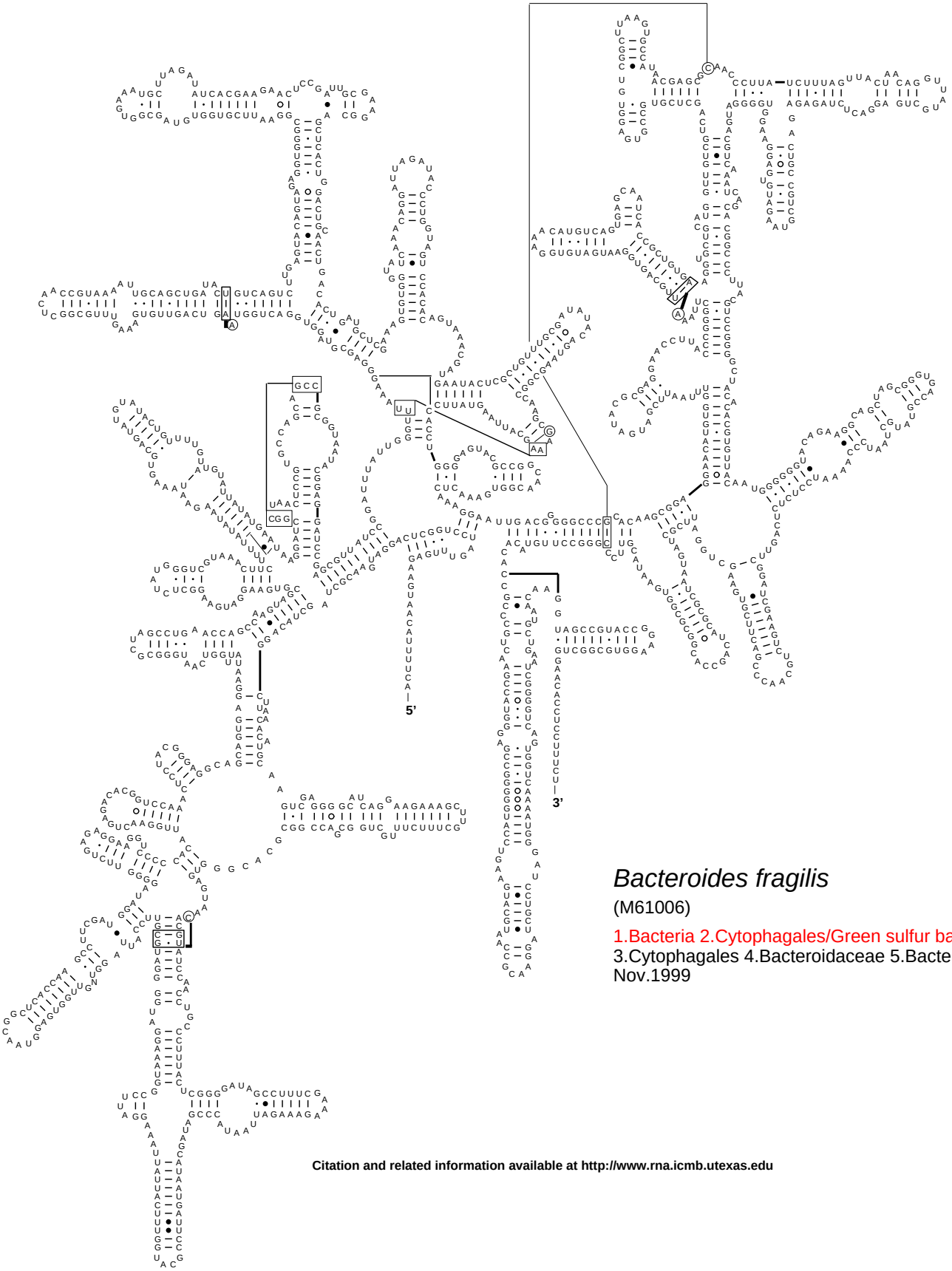
Neisseria gonorrhoeae

(X07714)

1.Bacteria 2.Proteobacteria 3.beta subdivision
4.Neisseriaceae 5.Neisseria
Nov,1999

Citation and related information available at http://www.rna.icmb.utexas.edu

Secondary Structure: small subunit ribosomal RNA



Bacteroides fragilis

(M61006)

1.Bacteria 2.Cytophagales/Green sulfur bacteria group
3.Cytophagales 4.Bacteroidaceae 5.Bacteroides
Nov.1999

Citation and related information available at http://www.rna.icmb.utexas.edu