

# Genomic Characteristics of Emerging Intraerythrocytic *Anaplasma capra* and High Prevalence in Goats, China

## Appendix

**Appendix Table 1.** Nucleotide sequence of primers used in the study

| Target       | Primer name | Primer sequence (5'-3')     | Annealing temperature (°C) | Amplicon size (bp) | References |
|--------------|-------------|-----------------------------|----------------------------|--------------------|------------|
| <i>rrs</i>   | Eh-out1     | TTGAGAGTTTGATCCTGGCTCAGAACG | 50                         | 1500               | (1)        |
|              | 3-17U       | WAAGGWGGTAATCCAGC           |                            |                    |            |
| <i>gltA</i>  | Outer-f     | GCGATTTTAGAGTGYGGAGATTG     | 55                         | 1076               | (2)        |
|              | Outer-r     | TACAATACCGGAGTAAAAGTCAA     | 51                         | 792                | (2)        |
|              | Inner-f     | GGTTCMTGTCYACTGCTGCGTG      |                            |                    |            |
| <i>groEL</i> | Inner-r     | TTGGATCGTARTTCTTGAGACC      | 56                         | 1264               | (2)        |
|              | Forward     | GCGAGGCGTTAGACAAGTCCATT     |                            |                    |            |
| <i>msp4</i>  | Reverse     | TCCAGAGATGCAAGCGTGTATAG     | 55                         | 799                | (2)        |
|              | Forward     | CAGTCTGCGCCTGCTCCCTAC       |                            |                    |            |
| <i>msp2</i>  | Reverse     | AGGAATCTTGCTCCAAGGTTA       | 51                         | 1139               | (2)        |
|              | Forward     | GCGTGTGATGGCTCTGGT          |                            |                    |            |
| <i>gltA*</i> | Reverse     | ACCAGTATCCTTATTTTTACC       | 60                         | 200                | This study |
|              | Forward     | CGAATCTATTTGCCTGCTT         |                            |                    |            |
|              | Reverse     | ATCGTAATTCTTGTAGACCTT       |                            |                    |            |

\*This pair of primers is used for the quantitative PCR.

## References

1. Wen B, Jian R, Zhang Y, Chen R. Simultaneous detection of *Anaplasma marginale* and a new *Ehrlichia* species closely related to *Ehrlichia chaffeensis* by sequence analyses of 16S ribosomal DNA in *Boophilus microplus* ticks from Tibet. J Clin Microbiol. 2002;40:3286–90. [PubMed](https://doi.org/10.1128/JCM.40.9.3286-3290.2002)  
<https://doi.org/10.1128/JCM.40.9.3286-3290.2002>
2. Li H, Zheng YC, Ma L, Jia N, Jiang BG, Jiang RR, et al. Human infection with a novel tick-borne *Anaplasma* species in China: a surveillance study. Lancet Infect Dis. 2015;15:663–70. [PubMed](https://doi.org/10.1016/S1473-3099(15)70051-4)  
[https://doi.org/10.1016/S1473-3099\(15\)70051-4](https://doi.org/10.1016/S1473-3099(15)70051-4)

**Appendix Table 2.** Probe sequences of fluorescence in situ hybridization (FISH)

| Probe sequence (5' to 3') | Probe sequence name |
|---------------------------|---------------------|
| TTCTGAGCCAGGATCAAAC       | AC16S-1             |
| TCGACTTGCATGTGTTAAGC      | AC16S-2             |
| AGCAAGCTACAGATTTGGTC      | AC16S-3             |
| CGTCTGCCACTAACCAAATC      | AC16S-4             |
| AGATTCCTATGCATTAACA       | AC16S-5             |
| TGGCTATCCCATACTACTAG      | AC16S-6             |
| GGATTATACGGTATTACCCA      | AC16S-7             |
| ATAGCGATAAAATCTTTCCCC     | AC16S-8             |
| CCAAGTAGCTAATCCGACAT      | AC16S-9             |
| ACAGATCACTGCCTTGGTAG      | AC16S-10            |
| TGATCATCCTCTCAGACCAG      | AC16S-11            |
| CATTGTCCAATATTTCCCAC      | AC16S-12            |
| CATAGCTGGATCAGGCTTGC      | AC16S-13            |
| TTTTACAACCCTAAGGCCTT      | AC16S-14            |
| TCATTATCTTCCCTACTGAA      | AC16S-15            |
| GGGACTTCTTCTGTAGGTAC      | AC16S-16            |
| CGCCCAATAATTCGAACAA       | AC16S-17            |
| TAACTTACCAAACCGCCTA       | AC16S-18            |
| GTTAAGCCCTGGTATTTTAC      | AC16S-19            |
| TGCAGTATTAAGAGCAGCCC      | AC16S-20            |
| TATCCTCTTCCGGACTCTAG      | AC16S-21            |
| ATTTACCTCTACACTAGGA       | AC16S-22            |
| GTGTTCTCCTAATATCTAC       | AC16S-23            |
| CAGGGTATCTAATCTGTTT       | AC16S-24            |
| TCAGCACTCATCGTTTACAG      | AC16S-25            |
| CAACACAGAGGCAAAAGCCC      | AC16S-26            |
| CGGAGTGCTTAACGCGTTAG      | AC16S-27            |
| CCTTTGAGTTTTAGTCTTGC      | AC16S-28            |
| CGAATTAACCAATGCTCC        | AC16S-29            |
| TGGTAAGGTTTTTCGCGTTG      | AC16S-30            |
| ATCTAACCTCCATGTCAAGA      | AC16S-31            |
| AACTGCGCCTTTCTGTAAAG      | AC16S-32            |
| ACGAGCTGACGACAGCCATG      | AC16S-33            |
| ACTTAACCCAACATCTCACG      | AC16S-34            |
| ATGAGGGTTACGCTCGTTGC      | AC16S-35            |
| CATTACCCGCTGGTAACTAA      | AC16S-36            |
| CACCGGCAGTTTCCCTTAAAG     | AC16S-37            |
| CGTGCTGACTTGACATCATC      | AC16S-38            |
| CATTGTAGCACGTGTGTAGC      | AC16S-39            |
| CGACGTTGCAACCTATTGTG      | AC16S-40            |
| CTTTTACGGATTAGCTCAGC      | AC16S-41            |
| CTCGAGTTGCAGAGGACAAT      | AC16S-42            |
| TCCACGATTACTAGCGATTTC     | AC16S-43            |
| CGAGAACGTATTCACCGTGG      | AC16S-44            |
| TGACGGGCAGTGTGTACAAG      | AC16S-45            |
| TTTGAGTTAAGCCAATTCCC      | AC16S-46            |
| CACCGACCCAACCTTAAATG      | AC16S-47            |
| TACAGCTACCTTGTTACGAC      | AC16S-48            |

**Appendix Table 3.** Genomic characteristics of *Anaplasma capra* strains BIME1 and BIME2 compared with that of representative *Anaplasma* species strains\*

| Characteristic   | Strain (GenBank accession no.)                  |   |   |   |  |   |  |
|------------------|---|---|---|---|--|---|--|
|                  | <i>A. capra</i> str. BIME1<br>(GCA_025628785.1) | <i>A. capra</i> str. BIME2<br>(GCA_025628805.1) | <i>A. ovis</i> str. Haibei<br>(NZ_CP015994.1) | <i>A. centrale</i> str. Israel<br>(NC_013532.1) | <i>A. marginale</i> str.<br>Florida<br>(NC_012026.1) | <i>A. platys</i> str. S3<br>(NZ_CP046391.1) | <i>A. phagocytophilum</i><br>str. JM (NC_021880) |
| Size (bp)        | 1,066,874                                       | 1,059,758                                       | 1,214,674                                     | 1,206,806                                       | 1,202,435  | 1,196,811                                   | 1,481,598  |
| GC rate (%)      | 48.32   | 48.32   | 48.9  | 50.0  | 49.8   | 45.5  | 41.6   |
| Gene counts (n)  | 929   | 932   | 1021  | 993   | 992  | 940   | 1155   |
| CDS counts (n)   | 862   | 863   | 945   | 922   | 913  | 882   | 997  |
| Pseudogenes (n)  | 27  | 29  | 32  | 27  | 35   | 17  | 114  |
| rRNAs (n)        | 3   | 3   | 3   | 3   | 3  | 3   | 3  |
| tRNAs (n)        | 37  | 37  | 37  | 37  | 37   | 34  | 37   |
| Completeness (%) | 99.79   | 99.36   | NA  | NA  | NA   | NA  | NA   |

\*bp, base pair; CDS, coding sequence; rRNA, ribosomal ribonucleic acid; tRNA, transfer ribonucleic acid; NA, not applicable.

**Appendix Table 4.** The estimated values of average nucleotide identity (ANI) and DNA-DNA hybridization (DDH) between *Anaplasma capra* and the other *Anaplasma* species

| Species                   | ANI                          |                              | DDH                        |                            |
|---------------------------|------------------------------|------------------------------|----------------------------|----------------------------|
|                           | <i>A. capra</i> str. BIME1   | <i>A. capra</i> str. BIME2   | <i>A. capra</i> str. BIME1 | <i>A. capra</i> str. BIME2 |
| <i>A. ovis</i>            | 78.0783<br>(GCA_002849345.1) | 78.0878<br>(GCA_002214625.1) | 17.4<br>(GCA_002849345.1)  | 17.5<br>(GCA_002849345.1)  |
| <i>A. marginale</i>       | 78.2897<br>(GCA_008801305.1) | 77.9471<br>(GCA_000020305.1) | 17.9<br>(GCA_000172475.1)  | 17.9<br>(GCA_000172475.1)  |
| <i>A. centrale</i>        | 77.9688<br>(GCA_000024505.1) | 77.8613<br>(GCA_000024505.1) | 17.4<br>(GCA_000024505.1)  | 17.4<br>(GCA_000024505.1)  |
| <i>A. phagocytophilum</i> | (-)                          | (-)                          | 13.0<br>(GCA_023476575.1)  | 13.0<br>(GCA_023278635.1)  |
| <i>A. platys</i>          | (-)                          | (-)                          | 13.1<br>(GCA_012790675.1)  | 13.1<br>(GCA_012790675.1)  |

**Appendix Table 5.** Virulence genes in *Anaplasma capra* str. BIME1 and BIME2

| Gene          | Description                                    | GenBank accession number   |  |
|---------------|--|--|--|
|               |  | <i>A. capra</i> str. BIME1   | <i>A. capra</i> str. BIME2                                   |
| <i>virB2</i>  | type IV secretion system protein VirB2 family  | MCU7611221.1<br>MCU7611222.1<br>MCU7611775.1<br>MCU7611780.1<br>MCU7611781.1<br>MCU7611782.1 | MCU7612774.1<br>MCU7612775.1<br>MCU7612776.1                 |
| <i>virB3</i>  | type IV secretion system protein VirB3         | MCU7611541.1   | MCU7612020.1   |
| <i>virB4</i>  | type IV secretion system protein VirB4 family  | MCU7611542.1<br>MCU7611779.1   | MCU7612019.1<br>MCU7612773.1                                 |
| <i>virB6</i>  | type IV secretion system protein VirB6 family  | MCU7611543.1<br>MCU7611544.1<br>MCU7611545.1<br>MCU7611546.1                                 | MCU7612018.1<br>MCU7612017.1<br>MCU7612016.1<br>MCU7612015.1 |
| <i>virB7</i>  | type IV secretion system protein VirB7         | MCU7611364.1   | MCU7612438.1   |
| <i>virB8</i>  | type IV secretion system protein VirB8 family  | MCU7611203.1<br>MCU7611581.1   | MCU7612293.1<br>MCU7611980.1                                 |
| <i>virB9</i>  | type IV secretion system protein VirB9 family  | MCU7611202.1<br>MCU7611762.1   | MCU7612294.1<br>MCU7612488.1                                 |
| <i>virB10</i> | type IV secretion system protein VirB10        | MCU7611201.1   | MCU7612295.1   |
| <i>virB11</i> | type IV secretion system ATPase VirB11         | MCU7611200.1   | MCU7612296.1   |
| <i>virD4</i>  | type IV secretion system component VirD4       | MCU7611199.1   | MCU7612297.1   |
| <i>Ats-1</i>  | <i>Anaplasma</i> T4SS translocated substrate-1 | MCU7611426.1   | MCU7612135.1   |
| <i>ompA</i>   | outer membrane protein OmpA                    | MCU7611514.1   | MCU7612047.1   |
| <i>Asp14</i>  | 14-kDa <i>Anaplasma</i> surface protein Asp14  | MCU7611843.1   | MCU7612563.1   |

**Appendix Table 6.** Genes predicted to be unique in *Anaplasma capra* str. BIME1 and BIME2

| Gene          | Protein  |                                | Function                              |
|---------------|--|--------------------------------|---------------------------------------|
| <i>menA</i>   | 1,4-dihydroxy-2-naphthoate polyprenyltransferase | Metabolic processing           | Menaquinone (vitamin K2) biosynthesis |
| unknown       | Glycosyltransferase 2 family protein             |                                | Glycan metabolism                     |
| <i>MKK9</i>   | Mitogen-activated protein kinase kinase 9        |                                | Ethylene and camalexin biosynthesis   |
| <i>MqnX</i>   | Aminodeoxyfutosine deaminase                     |                                | Menaquinone Biosynthesis              |
| <i>CPS1</i>   | Peregrinol diphosphate synthase                  |                                | Metabolism                            |
| <i>atuF</i>   | Geranyl-CoA carboxylase $\alpha$ subunit         |                                | Geraniol degradation                  |
| <i>Zbtb46</i> | Zinc finger and BTB domain-containing protein 46 | Genetic information processing | Transcription factor                  |
| <i>UFL1</i>   | E3 UFM1-protein ligase 1                         |                                | Cellular regulation                   |
| <i>Hmbox1</i> | Homeobox-containing protein 1                    |                                | Transcription factor                  |
| <i>RSF1</i>   | Remodeling and spacing factor 1                  |                                | DNA repair                            |
| <i>Ara54</i>  | E3 ubiquitin-protein ligase                      |                                | Cellular regulation                   |

| Gene          | Protein   | Function   |
|---------------|---|--|
| <i>MACC1</i>  | Metastasis-associated in colon cancer protein 1 | Signaling and cellular processing<br>Signaling regulator |
| <i>dia</i>    | Diaphanous protein                              | Cytokinesis  |
| <i>desK</i>   | Membrane-associated kinase DesK                 | Membrane-associated kinase                               |
| <i>hbhA</i>   | Heparin binding hemagglutinin                   | Virulence factor   |
| <i>Mrgprg</i> | Mas-related G protein-coupled receptor member G | G protein-coupled receptor                               |
| unknown       | Membrane protein                                | Protein with domain of unknown function                  |
| unknown*      | unclassified protein                            | Function unknown   |

\*Including 37 unclassified genes.

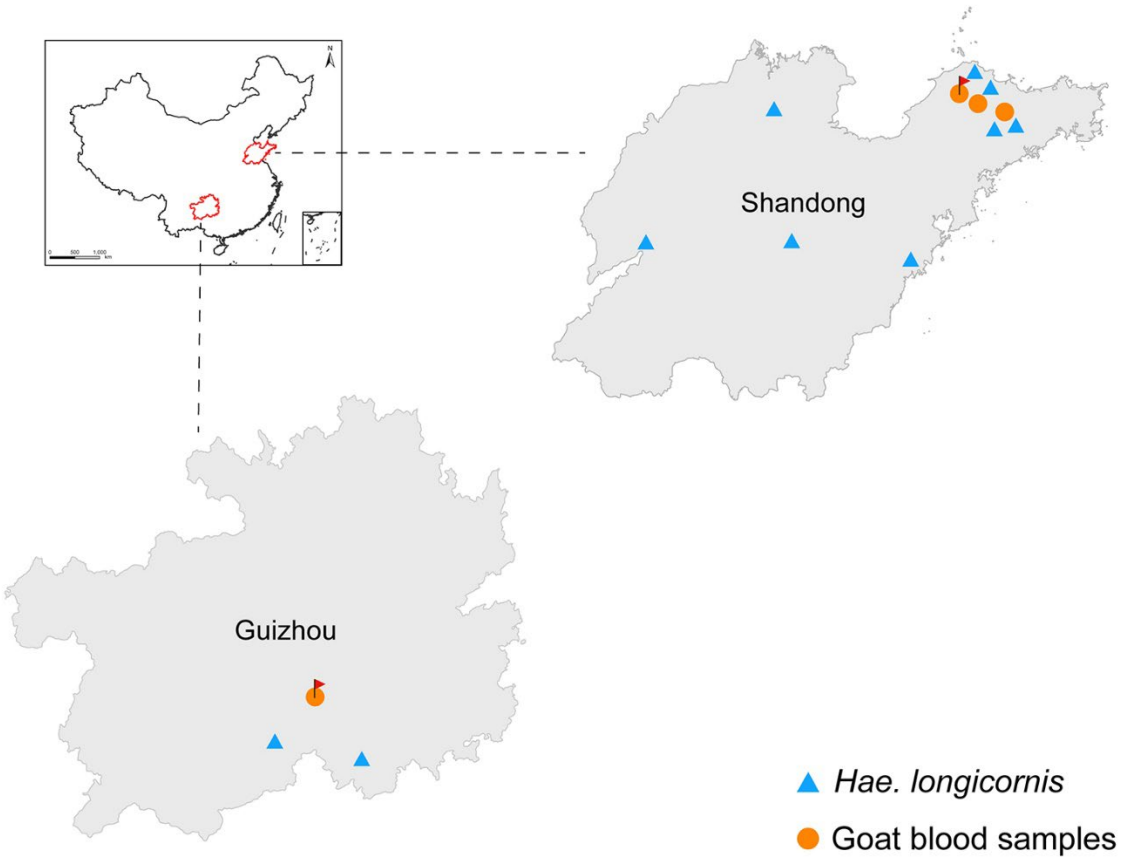
**Appendix Table 7.** Functional Clusters of Orthologous Groups of protein-coding genes from the representative *Anaplasma* species strains

| Category | Functional category  | <i>A. capra</i> | <i>A. capra</i> | <i>A. ovis</i> | <i>A. centrale</i> | <i>A. marginale</i> | <i>A. platys</i> | <i>A. phagocytophilum</i> |
|----------|--|-----------------|-----------------|----------------|--------------------|---------------------|------------------|---------------------------|
|          |  | str. BIME1      | str. BIME2      | str. Haibei    | str. Israel        | str. Florida        | str. S3          | str. JM                   |
| A        | RNA processing and modification                              | 1               | 1               | 1              | 1                  | 1                   | 1                | 1                         |
| B        | Chromatin structure and dynamics                             | 0               | 0               | 0              | 0                  | 0                   | 0                | 0                         |
| C        | Energy production and conversion                             | 67              | 67              | 74             | 70                 | 68                  | 71               | 74                        |
| D        | Cell cycle control, cell division, chromosome partitioning   | 16              | 15              | 18             | 17                 | 18                  | 11               | 14                        |
| E        | Amino acid transport and metabolism                          | 31              | 31              | 35             | 34                 | 35                  | 30               | 25                        |
| F        | Nucleotide transport and metabolism                          | 54              | 54              | 55             | 54                 | 54                  | 52               | 56                        |
| G        | Carbohydrate transport and metabolism                        | 21              | 20              | 26             | 26                 | 25                  | 27               | 22                        |
| H        | Coenzyme transport and metabolism                            | 65              | 65              | 67             | 64                 | 66                  | 58               | 68                        |
| I        | Lipid transport and metabolism                               | 28              | 28              | 30             | 29                 | 28                  | 29               | 29                        |
| J        | Translation, ribosomal structure and biogenesis              | 126             | 127             | 131            | 130                | 130                 | 128              | 137                       |
| K        | Transcription  | 18              | 17              | 21             | 20                 | 21                  | 19               | 21                        |
| L        | Replication, recombination and repair                        | 52              | 53              | 53             | 54                 | 52                  | 51               | 57                        |
| M        | Cell wall/membrane/envelope biogenesis                       | 48              | 47              | 62             | 58                 | 60                  | 35               | 62                        |
| N        | Cell motility  | 2               | 2               | 2              | 2                  | 2                   | 2                | 2                         |
| O        | Posttranslational modification, protein turnover, chaperones | 44              | 44              | 43             | 43                 | 43                  | 43               | 45                        |

| Category | Functional category   | <i>A. capra</i><br>str. BIME1 | <i>A. capra</i><br>str. BIME2 | <i>A. ovis</i><br>str. Haibei | <i>A. centrale</i><br>str. Israel | <i>A. marginale</i><br>str. Florida | <i>A. platys</i><br>str. S3 | <i>A. phagocytophilum</i><br>str. JM |
|----------|---|-------------------------------|-------------------------------|-------------------------------|-----------------------------------|-------------------------------------|-----------------------------|--------------------------------------|
|          |   | Number of genes               |                               |                               |                                   |                                     |                             |                                      |
| P        | Inorganic ion transport and metabolism                        | 31                            | 31                            | 36                            | 35                                | 34                                  | 35                          | 34                                   |
| Q        | Secondary metabolites biosynthesis, transport and catabolism  | 12                            | 12                            | 12                            | 12                                | 12                                  | 11                          | 12                                   |
| R        | General function prediction only                              | 0                             | 0                             | 0                             | 0                                 | 0                                   | 0                           | 0                                    |
| S        | Function unknown  | 90                            | 91                            | 89                            | 91                                | 92                                  | 83                          | 88                                   |
| T        | Signal transduction mechanisms                                | 9                             | 9                             | 10                            | 10                                | 10                                  | 10                          | 9                                    |
| U        | Intracellular trafficking, secretion, and vesicular transport | 38                            | 38                            | 38                            | 38                                | 38                                  | 40                          | 40                                   |
| V        | Defense mechanisms  | 4                             | 4                             | 4                             | 4                                 | 4                                   | 4                           | 4                                    |
| W        | Extracellular structures                                      | 0                             | 0                             | 0                             | 0                                 | 0                                   | 0                           | 0                                    |
| X        | Mobilome: prophages, transposons                              | 0                             | 0                             | 0                             | 0                                 | 0                                   | 0                           | 0                                    |
| Y        | Nuclear structure   | 0                             | 0                             | 0                             | 0                                 | 0                                   | 0                           | 0                                    |
| Z        | Cytoskeleton  | 0                             | 0                             | 0                             | 0                                 | 0                                   | 0                           | 0                                    |

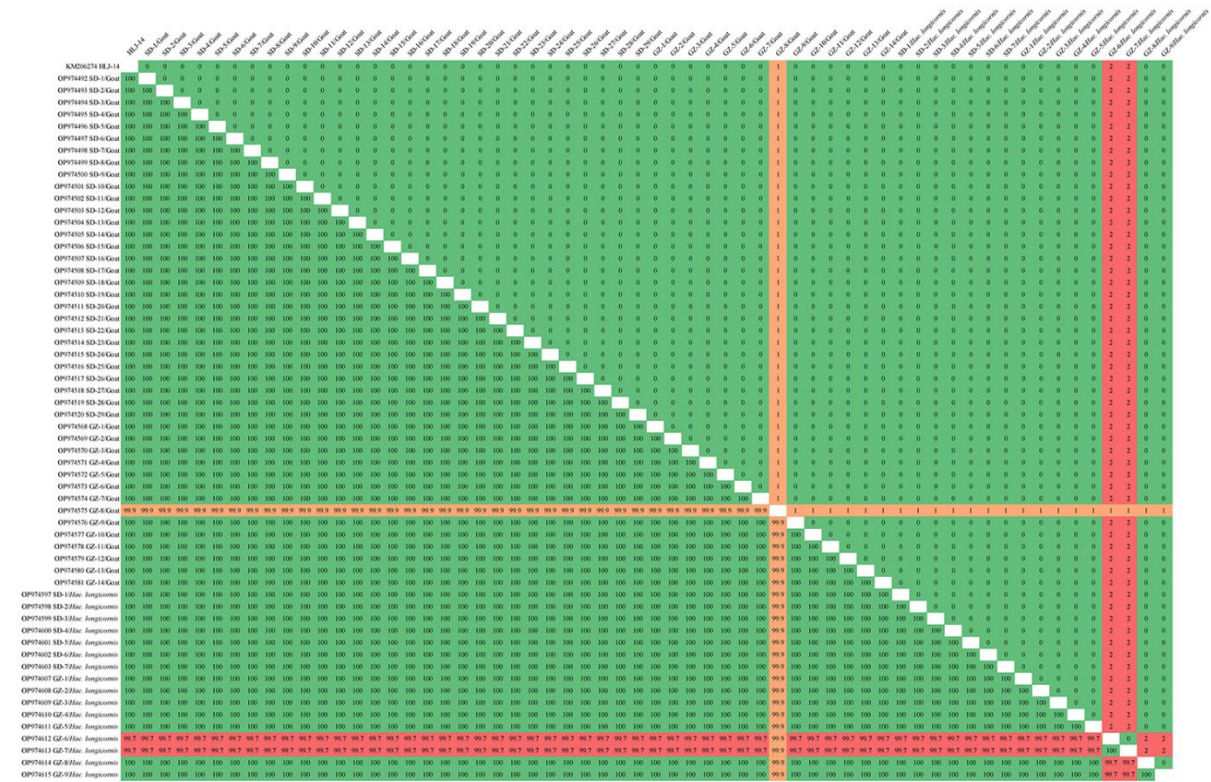
**Appendix Table 8.** The *Anaplasma capra*-positive numbers of goats and *Hae. Longicornis* in this study by PCR toward different gene loci

| Gene loci  | Goats from Shandong | Goats from Guizhou | <i>Hae. longicornis</i> from Shandong | <i>Hae. longicornis</i> from Guizhou |
|--|---------------------|--------------------|---------------------------------------|--------------------------------------|
| No. of tested  | 54                  | 18                 | 144                                   | 57                                   |
| 16S rRNA, <i>gltA</i> , <i>groEL</i> , <i>msp4</i> (+) | 14                  | 1                  | 0                                     | 0                                    |
| 16S rRNA, <i>gltA</i> , <i>msp4</i> (+)                | 4                   | 4                  | 0                                     | 3                                    |
| 16S rRNA, <i>gltA</i> , <i>groEL</i> (+)               | 1                   | 0                  | 0                                     | 0                                    |
| <i>gltA</i> , <i>groEL</i> , <i>msp4</i> (+)           | 7                   | 2                  | 0                                     | 0                                    |
| 16S rRNA, <i>gltA</i> (+)                              | 1                   | 0                  | 0                                     | 0                                    |
| <i>gltA</i> , <i>msp4</i> (+)                          | 0                   | 5                  | 3                                     | 2                                    |
| <i>gltA</i> (+)  | 2                   | 2                  | 4                                     | 4                                    |



**Appendix Figure 1.** The sampling sites where ticks and blood samples were collected in this study.

Different color and marks represent the types of samples collected in different areas. The flags indicate the locations, where the goat blood samples were collected for next-generation sequencing of *Anaplasma capra* genomes.



**Appendix Figure 2.** The comparison between each *gftA* gene sequences of *Anaplasma capra* this study and sequence from human. The upper right part represents the number of bases that differ from each sequence and the lower left part represents the nucleotide identity (%) between each sequence from others.

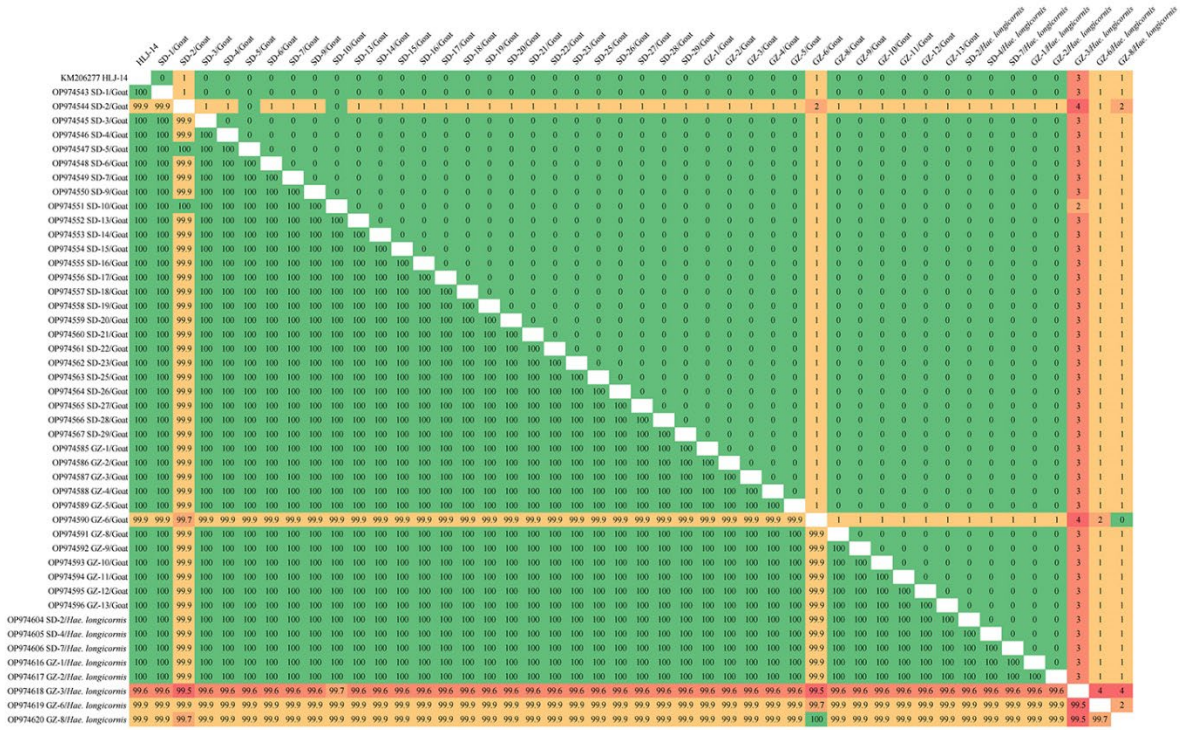


|                     | HLJ-14 | SD-1/Goat | SD-2/Goat | SD-3/Goat | SD-4/Goat | SD-5/Goat | SD-6/Goat | SD-7/Goat | SD-9/Goat | SD-12/Goat | SD-13/Goat | SD-14/Goat | SD-15/Goat | SD-16/Goat | SD-17/Goat | SD-18/Goat | SD-20/Goat | SD-21/Goat | SD-22/Goat | SD-23/Goat | SD-25/Goat | SD-26/Goat | SD-27/Goat | SD-29/Goat | GZ-3/Goat | GZ-5/Goat | GZ-11/Goat |
|---------------------|--------|-----------|-----------|-----------|-----------|-----------|-----------|-----------|-----------|------------|------------|------------|------------|------------|------------|------------|------------|------------|------------|------------|------------|------------|------------|------------|-----------|-----------|------------|
| KM206275 HLJ-14     | 0      | 0         | 0         | 0         | 0         | 0         | 0         | 0         | 0         | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 2         | 0         | 0          |
| OP974521 SD-1/Goat  | 100    | 0         | 0         | 0         | 0         | 5         | 0         | 0         | 0         | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 2         | 0         | 0          |
| OP974522 SD-3/Goat  | 100    | 100       | 0         | 0         | 0         | 5         | 0         | 0         | 0         | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 2         | 0         | 0          |
| OP974523 SD-4/Goat  | 100    | 100       | 100       | 0         | 0         | 5         | 0         | 0         | 0         | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 2         | 0         | 0          |
| OP974524 SD-5/Goat  | 100    | 100       | 100       | 100       | 0         | 5         | 0         | 0         | 0         | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 2         | 0         | 0          |
| OP974525 SD-6/Goat  | 100    | 99.6      | 99.6      | 99.6      | 99.6      | 5         | 5         | 5         | 5         | 5          | 5          | 5          | 5          | 5          | 5          | 5          | 5          | 5          | 5          | 5          | 5          | 5          | 5          | 7          | 5         | 5         |            |
| OP974526 SD-7/Goat  | 100    | 100       | 100       | 100       | 100       | 99.6      | 0         | 0         | 0         | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 2         | 0         | 0          |
| OP974527 SD-9/Goat  | 100    | 100       | 100       | 100       | 100       | 99.6      | 100       | 0         | 0         | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 2         | 0         | 0          |
| OP974528 SD-12/Goat | 100    | 100       | 100       | 100       | 100       | 99.6      | 100       | 100       | 0         | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 2         | 0         | 0          |
| OP974529 SD-13/Goat | 100    | 100       | 100       | 100       | 100       | 99.6      | 100       | 100       | 100       | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 2         | 0         | 0          |
| OP974530 SD-14/Goat | 100    | 100       | 100       | 100       | 100       | 99.6      | 100       | 100       | 100       | 100        | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 2         | 0         | 0          |
| OP974531 SD-15/Goat | 100    | 100       | 100       | 100       | 100       | 99.6      | 100       | 100       | 100       | 100        | 100        | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 2         | 0         | 0          |
| OP974532 SD-16/Goat | 100    | 100       | 100       | 100       | 100       | 99.6      | 100       | 100       | 100       | 100        | 100        | 100        | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 2         | 0         | 0          |
| OP974533 SD-17/Goat | 100    | 100       | 100       | 100       | 100       | 99.6      | 100       | 100       | 100       | 100        | 100        | 100        | 100        | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 2         | 0         | 0          |
| OP974534 SD-18/Goat | 100    | 100       | 100       | 100       | 100       | 99.6      | 100       | 100       | 100       | 100        | 100        | 100        | 100        | 100        | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 2         | 0         | 0          |
| OP974535 SD-20/Goat | 100    | 100       | 100       | 100       | 100       | 99.6      | 100       | 100       | 100       | 100        | 100        | 100        | 100        | 100        | 100        | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 2         | 0         | 0          |
| OP974536 SD-21/Goat | 100    | 100       | 100       | 100       | 100       | 99.6      | 100       | 100       | 100       | 100        | 100        | 100        | 100        | 100        | 100        | 100        | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 2         | 0         | 0          |
| OP974537 SD-22/Goat | 100    | 100       | 100       | 100       | 100       | 99.6      | 100       | 100       | 100       | 100        | 100        | 100        | 100        | 100        | 100        | 100        | 100        | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 2         | 0         | 0          |
| OP974538 SD-23/Goat | 100    | 100       | 100       | 100       | 100       | 99.6      | 100       | 100       | 100       | 100        | 100        | 100        | 100        | 100        | 100        | 100        | 100        | 100        | 0          | 0          | 0          | 0          | 0          | 0          | 2         | 0         | 0          |
| OP974539 SD-25/Goat | 100    | 100       | 100       | 100       | 100       | 99.6      | 100       | 100       | 100       | 100        | 100        | 100        | 100        | 100        | 100        | 100        | 100        | 100        | 100        | 100        | 0          | 0          | 0          | 0          | 2         | 0         | 0          |
| OP974540 SD-26/Goat | 100    | 100       | 100       | 100       | 100       | 99.6      | 100       | 100       | 100       | 100        | 100        | 100        | 100        | 100        | 100        | 100        | 100        | 100        | 100        | 100        | 100        | 0          | 0          | 0          | 2         | 0         | 0          |
| OP974541 SD-27/Goat | 100    | 100       | 100       | 100       | 100       | 99.6      | 100       | 100       | 100       | 100        | 100        | 100        | 100        | 100        | 100        | 100        | 100        | 100        | 100        | 100        | 100        | 100        | 0          | 0          | 2         | 0         | 0          |
| OP974542 SD-29/Goat | 100    | 100       | 100       | 100       | 100       | 99.6      | 100       | 100       | 100       | 100        | 100        | 100        | 100        | 100        | 100        | 100        | 100        | 100        | 100        | 100        | 100        | 100        | 100        | 0          | 2         | 0         | 0          |
| OP974582 GZ-3/Goat  | 99.8   | 99.8      | 99.8      | 99.8      | 99.8      | 99.4      | 99.8      | 99.8      | 99.8      | 99.8       | 99.8       | 99.8       | 99.8       | 99.8       | 99.8       | 99.8       | 99.8       | 99.8       | 99.8       | 99.8       | 99.8       | 99.8       | 99.8       | 99.8       | 2         | 2         | 0          |
| OP974583 GZ-5/Goat  | 100    | 100       | 100       | 100       | 100       | 99.6      | 100       | 100       | 100       | 100        | 100        | 100        | 100        | 100        | 100        | 100        | 100        | 100        | 100        | 100        | 100        | 100        | 100        | 100        | 99.8      | 0         | 0          |
| OP974584 GZ-11/Goat | 100    | 100       | 100       | 100       | 100       | 99.6      | 100       | 100       | 100       | 100        | 100        | 100        | 100        | 100        | 100        | 100        | 100        | 100        | 100        | 100        | 100        | 100        | 100        | 100        | 99.8      | 100       | 0          |

**Appendix Figure 3.** The comparison between each *groEL* gene sequences of *Anaplasma capra* this study and sequence from human. The upper right part represents the number of bases that differ from each sequence and the lower left part represents the nucleotide identity (%) between each sequence from others.

|                                | HLJ-14 | SD-1/Goat | SD-2/Goat | SD-3/Goat | SD-A/Goat | SD-6/Goat | SD-10/Goat | SD-11/Goat | SD-12/Goat | SD-13/Goat | SD-14/Goat | SD-15/Goat | SD-16/Goat | SD-18/Goat | SD-19/Goat | SD-20/Goat | SD-21/Goat | SD-23/Goat | SD-27/Goat | SD-28/Goat | SD-29/Goat | GZ-1/Goat | GZ-8/Goat | GZ-10/Goat | GZ-11/Goat | GZ-13/Goat | GZ-1/Hae. longicornis | GZ-2/Hae. longicornis | GZ-6/Hae. longicornis |
|--------------------------------|--------|-----------|-----------|-----------|-----------|-----------|------------|------------|------------|------------|------------|------------|------------|------------|------------|------------|------------|------------|------------|------------|------------|-----------|-----------|------------|------------|------------|-----------------------|-----------------------|-----------------------|
| KM206273 HLJ-14                | 7      | 0         | 10        | 11        | 6         | 0         | 0          | 0          | 0          | 0          | 1          | 0          | 0          | 1          | 0          | 0          | 0          | 0          | 6          | 0          | 8          | 1         | 7         | 0          | 0          | 0          | 0                     | 7                     |                       |
| OQ148549 SD-1/Goat             | 99.5   | 7         | 1         | 1         | 1         | 7         | 6          | 7          | 7          | 7          | 7          | 7          | 7          | 7          | 7          | 7          | 7          | 7          | 2          | 7          | 8          | 1         | 6         | 6          | 6          | 7          | 7                     | 2                     |                       |
| OQ148550 SD-2/Goat             | 100    | 99.5      | 10        | 11        | 6         | 0         | 0          | 0          | 0          | 0          | 1          | 0          | 0          | 1          | 0          | 0          | 0          | 0          | 6          | 0          | 8          | 1         | 7         | 0          | 0          | 0          | 0                     | 7                     |                       |
| OQ148551 SD-3/Goat             | 99.3   | 99.9      | 99.3      | 9         | 4         | 10        | 10         | 10         | 10         | 10         | 9          | 10         | 10         | 11         | 10         | 10         | 10         | 10         | 4          | 10         | 8          | 1         | 7         | 10         | 10         | 10         | 10                    | 0                     |                       |
| OQ148552 SD-4/Goat             | 99.2   | 99.9      | 99.2      | 99.4      | 5         | 11        | 11         | 11         | 11         | 11         | 12         | 11         | 11         | 12         | 11         | 11         | 11         | 11         | 7          | 11         | 8          | 1         | 12        | 11         | 11         | 11         | 11                    | 1                     |                       |
| OQ148553 SD-6/Goat             | 99.6   | 99.9      | 99.6      | 99.7      | 99.7      | 6         | 6          | 6          | 6          | 6          | 7          | 6          | 6          | 7          | 6          | 6          | 6          | 6          | 2          | 6          | 8          | 1         | 7         | 6          | 6          | 6          | 6                     | 1                     |                       |
| OQ148554 SD-10/Goat            | 100    | 99.5      | 100       | 99.3      | 99.2      | 99.6      | 0          | 0          | 0          | 0          | 1          | 0          | 0          | 1          | 0          | 0          | 0          | 0          | 6          | 0          | 8          | 1         | 7         | 0          | 0          | 0          | 0                     | 7                     |                       |
| OQ148555 SD-11/Goat            | 100    | 99.6      | 100       | 99.3      | 99.2      | 99.6      | 100        | 0          | 0          | 0          | 1          | 0          | 0          | 1          | 0          | 0          | 0          | 0          | 6          | 0          | 8          | 1         | 7         | 0          | 0          | 0          | 0                     | 7                     |                       |
| OQ148556 SD-12/Goat            | 100    | 99.5      | 100       | 99.3      | 99.2      | 99.6      | 100        | 100        | 0          | 0          | 1          | 0          | 0          | 0          | 0          | 0          | 0          | 0          | 6          | 0          | 8          | 1         | 7         | 0          | 0          | 0          | 0                     | 7                     |                       |
| OQ148557 SD-13/Goat            | 100    | 99.5      | 100       | 99.3      | 99.2      | 99.6      | 100        | 100        | 100        | 0          | 1          | 0          | 0          | 1          | 0          | 0          | 0          | 0          | 6          | 0          | 8          | 1         | 7         | 0          | 0          | 0          | 0                     | 7                     |                       |
| OQ148558 SD-15/Goat            | 100    | 99.5      | 100       | 99.3      | 99.2      | 99.6      | 100        | 100        | 100        | 100        | 1          | 0          | 0          | 1          | 0          | 0          | 0          | 0          | 6          | 0          | 8          | 1         | 7         | 0          | 0          | 0          | 0                     | 7                     |                       |
| OQ148559 SD-16/Goat            | 99.9   | 99.5      | 99.9      | 99.4      | 99.2      | 99.5      | 99.9       | 99.9       | 99.9       | 99.9       | 1          | 1          | 2          | 1          | 1          | 1          | 1          | 5          | 1          | 8          | 1          | 6         | 1         | 1          | 1          | 1          | 1                     | 7                     |                       |
| OQ148560 SD-18/Goat            | 100    | 99.5      | 100       | 99.3      | 99.2      | 99.6      | 100        | 100        | 100        | 100        | 99.9       | 0          | 1          | 0          | 0          | 0          | 0          | 0          | 6          | 0          | 8          | 1         | 7         | 0          | 0          | 0          | 0                     | 7                     |                       |
| OQ148561 SD-19/Goat            | 100    | 99.5      | 100       | 99.3      | 99.2      | 99.6      | 100        | 100        | 100        | 100        | 99.9       | 100        | 0          | 0          | 0          | 0          | 0          | 0          | 6          | 0          | 8          | 1         | 7         | 0          | 0          | 0          | 0                     | 7                     |                       |
| OQ148562 SD-20/Goat            | 99.9   | 99.5      | 99.9      | 99.2      | 99.2      | 99.5      | 99.9       | 99.9       | 100        | 99.9       | 99.9       | 99.9       | 99.9       | 100        | 0          | 1          | 0          | 1          | 6          | 1          | 8          | 1         | 7         | 0          | 1          | 1          | 1                     | 7                     |                       |
| OQ148563 SD-21/Goat            | 100    | 99.5      | 100       | 99.3      | 99.2      | 99.6      | 100        | 100        | 100        | 100        | 99.9       | 100        | 100        | 100        | 0          | 0          | 0          | 0          | 6          | 0          | 8          | 1         | 7         | 0          | 0          | 0          | 0                     | 7                     |                       |
| OQ148564 SD-22/Goat            | 100    | 99.5      | 100       | 99.3      | 99.2      | 99.6      | 100        | 100        | 100        | 100        | 99.9       | 100        | 100        | 99.9       | 100        | 100        | 100        | 100        | 6          | 0          | 8          | 1         | 7         | 0          | 0          | 0          | 0                     | 7                     |                       |
| OQ148565 SD-23/Goat            | 100    | 99.5      | 100       | 99.3      | 99.2      | 99.6      | 100        | 100        | 100        | 100        | 99.9       | 100        | 100        | 100        | 100        | 100        | 100        | 100        | 6          | 0          | 8          | 1         | 7         | 0          | 0          | 0          | 0                     | 7                     |                       |
| OQ148566 SD-27/Goat            | 100    | 99.5      | 100       | 99.3      | 99.2      | 99.6      | 100        | 100        | 100        | 100        | 99.9       | 100        | 100        | 99.9       | 100        | 100        | 100        | 100        | 6          | 0          | 8          | 1         | 7         | 0          | 0          | 0          | 0                     | 7                     |                       |
| OQ148567 SD-28/Goat            | 99.6   | 99.9      | 99.6      | 99.7      | 99.5      | 99.9      | 99.6       | 99.6       | 99.6       | 99.6       | 99.6       | 99.6       | 99.6       | 99.6       | 99.6       | 99.6       | 99.6       | 99.6       | 6          | 8          | 1          | 6         | 6         | 6          | 6          | 6          | 2                     |                       |                       |
| OQ148568 SD-29/Goat            | 100    | 99.5      | 100       | 99.3      | 99.2      | 99.6      | 100        | 100        | 100        | 100        | 99.9       | 100        | 100        | 99.9       | 100        | 100        | 100        | 100        | 99.6       | 8          | 1          | 7         | 0         | 0          | 0          | 0          | 7                     |                       |                       |
| OQ148572 GZ-1/Goat             | 99.4   | 99.4      | 99.4      | 99.4      | 99.4      | 99.4      | 99.4       | 99.4       | 99.4       | 99.4       | 99.4       | 99.4       | 99.4       | 99.4       | 99.4       | 99.4       | 99.4       | 99.4       | 99.4       | 99.4       | 99.4       | 99.4      | 99.4      | 99.4       | 99.4       | 99.4       | 99.4                  | 99.4                  | 99.4                  |
| OQ148573 GZ-8/Goat             | 99.9   | 99.9      | 99.9      | 99.9      | 99.9      | 99.9      | 99.9       | 99.9       | 99.9       | 99.9       | 99.9       | 99.9       | 99.9       | 99.9       | 99.9       | 99.9       | 99.9       | 99.9       | 99.9       | 99.9       | 99.9       | 99.9      | 99.9      | 99.9       | 99.9       | 99.9       | 99.9                  | 99.9                  | 99.9                  |
| OQ148574 GZ-10/Goat            | 99.5   | 99.5      | 99.5      | 99.5      | 99.5      | 99.5      | 99.5       | 99.5       | 99.5       | 99.5       | 99.6       | 99.5       | 99.5       | 99.5       | 99.5       | 99.5       | 99.5       | 99.5       | 99.6       | 99.5       | 99.4       | 99.5      | 7         | 7          | 7          | 7          | 6                     |                       |                       |
| OQ148575 GZ-11/Goat            | 100    | 99.6      | 100       | 99.3      | 99.2      | 99.6      | 100        | 100        | 100        | 100        | 99.9       | 100        | 100        | 100        | 100        | 100        | 100        | 100        | 99.6       | 100        | 99.4       | 99.9      | 99.5      | 0          | 0          | 0          | 7                     |                       |                       |
| OQ148576 GZ-13/Goat            | 100    | 99.6      | 100       | 99.3      | 99.2      | 99.6      | 100        | 100        | 100        | 100        | 99.9       | 100        | 100        | 99.9       | 100        | 100        | 100        | 100        | 99.6       | 100        | 99.4       | 99.9      | 99.5      | 100        | 0          | 0          | 7                     |                       |                       |
| OQ148569 GZ-1/Hae. longicornis | 100    | 99.5      | 100       | 99.3      | 99.2      | 99.6      | 100        | 100        | 100        | 100        | 99.9       | 100        | 100        | 99.9       | 100        | 100        | 100        | 100        | 99.6       | 100        | 99.4       | 99.9      | 99.5      | 100        | 100        | 0          | 7                     |                       |                       |
| OQ148570 GZ-2/Hae. longicornis | 100    | 99.5      | 100       | 99.3      | 99.2      | 99.6      | 100        | 100        | 100        | 100        | 99.9       | 100        | 100        | 99.9       | 100        | 100        | 100        | 100        | 99.6       | 100        | 99.4       | 99.9      | 99.5      | 100        | 100        | 100        | 0                     | 7                     |                       |
| OQ148571 GZ-6/Hae. longicornis | 99.5   | 99.9      | 99.5      | 100       | 99.9      | 99.9      | 99.5       | 99.5       | 99.5       | 99.5       | 99.5       | 99.5       | 99.5       | 99.5       | 99.5       | 99.5       | 99.5       | 99.5       | 99.9       | 99.5       | 99.4       | 99.9      | 99.5      | 99.5       | 99.5       | 99.5       | 99.5                  |                       |                       |

**Appendix Figure 4.** The comparison between each 16S rRNA gene sequences of *Anaplasma capra* this study and sequence from human. The upper right part represents the number of bases that differ from each sequence and the lower left part represents the nucleotide identity (%) between each sequence from others.



**Appendix Figure 5.** The comparison between each *msp4* gene sequences of *Anaplasma capra* this study and sequence from human. The upper right part represents the number of bases that differ from each sequence and the lower left part represents the nucleotide identity (%) between each sequence from others.