

# Genome Sequence of *Ureaplasma diversum* Strain ATCC 49782

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**Here, we report the complete genome sequence of *Ureaplasma diversum* strain ATCC 49782. This species is of bovine origin, having an association with reproductive disorders in cattle, including placentitis, fetal alveolitis, abortion, and birth of weak calves. It has a small circular chromosome of 975,425 bp.**

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*Ureaplasma diversum* is a bovine-origin ureaplasma isolated in 1969. Initially, it was defined as a nonpathogenic species, but it recently has been shown to cause damage in bovine organs and cells (1–5). This ureaplasma has been associated with female infertility, placentitis, fetal alveolitis, and abortion or birth of weak calves (3–5). In bulls, it is an important pathogen of the genital tract and may cause low sperm motility, seminal vesiculitis, and epididymitis (1, 6). Nevertheless, the mechanisms by which this organism exerts its virulence and pathogenicity in cattle are mostly unknown (1, 7–9).

*U. diversum* ATCC 49782 was first cultured in 2 ml of *Ureaplasma* medium (UB) at 37°C, followed by propagation in 3,000 ml of the same broth. At the logarithmic growth phase (based on colorimetric changes), the culture was centrifuged at 20,600 × *g* for 30 min at 25°C. The DNA was extracted using a PureLink genomic DNA minikit (Life Technologies, Brazil). The whole genome was sequenced from a paired-end library using Illumina HiSeq 2000 (Illumina, Inc., San Diego, CA) at the Purdue University Genomics Core Facility. Average reads of about 100 bases were assembled using ABySS 1.2.7. After assembly, resulting in 4,552× genome coverage, eight remaining gaps were closed using conventional PCR, followed by Sanger sequencing in both directions. First-pass annotation was achieved using the National Center for Biotechnology Information (NCBI) Prokaryotic Genome Annotation Pipeline (PGAP).

The complete genome of *U. diversum* strain ATCC 49782 is composed of 975,425 bp in a single circular chromosome, with a G+C content of 28.1%. It appears to use the opal stop codon (UGA) for tryptophan. A total of 646 coding DNA sequences (CDSs), 6 rRNAs, and 22 tRNA genes were predicted and annotated. Three hundred seventy-four CDSs (57.9%) have putative functional identities; however, 272 CDSs (42.1%) are hypothetical proteins.

Comparing the data obtained for *U. diversum* ATCC 49782 with those for 14 ureaplasma serovars of human origin (10 serovars of *Ureaplasma urealyticum* and 4 serovars of *Ureaplasma parvum*) (10), a larger genome size was observed (*U. diversum*, 0.97

Mbp; *U. urealyticum*, 0.84 to 0.95 Mbp; *U. parvum*, 0.75 to 0.78 Mbp), with a slightly higher G+C content (*U. diversum*, 28.1%; *U. urealyticum*, 25 to 27%; *U. parvum*, 25%), number of CDSs (*U. diversum*, 646; *U. urealyticum*, average of 608; *U. parvum*, average of 608), and number of hypothetical CDSs (*U. diversum*, 237; *U. urealyticum*, average of 230; *U. parvum*, average of 201). These genetic differences may reflect for the host specificity of *U. diversum*. However, a detailed assessment of the genes needs to be performed.

The sequence described here represents a valuable new resource for the study of *U. diversum* on a genetic basis. Further, extensive analyses of these data may help us to better understand the host specificity of this mollicute and what defines its pathogenicity in bovines. Moreover, it will also aid the development of new strategies for the treatment, prevention, and control of ureaplasma infections.

**Nucleotide sequence accession number.** The genome sequence of *U. diversum* strain ATCC 49782 has been deposited in GenBank under the accession no. [CP009770](https://www.ncbi.nlm.nih.gov/nuccore/CP009770).

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## REFERENCES

- Marques LM, Ueno PM, Buzinhan M, Cortez BA, Neto RL, Yamaguti M, Oliveira RC, Guimarães AM, Monezi TA, Braga AC, Machado-Santelli GM, Timenetsky J. 2010. Invasion of *Ureaplasma diversum* in hep-2 cells. *BMC Microbiol* 10:83. <http://dx.doi.org/10.1186/1471-2180-10-83>.
- Miller RB, Ruhnke HL, Doig PA, Poitras BJ, Palmer NC. 1983. The effects of *Ureaplasma diversum* inoculated into the amniotic cavity in cows. *Theriogenology* 20:367–374. [http://dx.doi.org/10.1016/0093-691X\(83\)90071-7](http://dx.doi.org/10.1016/0093-691X(83)90071-7).

3. Buzinhani M, Yamaguti M, Oliveira RC, Cortez BA, Marques LM, Machado-Santelli GM, Assumpção ME, Timenetsky J. 2011. Invasion of *Ureaplasma diversum* in bovine spermatozooids. *BMC Res Notes* 4:455. <http://dx.doi.org/10.1186/1756-0500-4-455>.
4. Howard CJ, Gourlay RN, Brownlie J. 1973. The virulence of T-mycoplasmas, isolated from various animal species, assayed by intramammary inoculation in cattle. *J Hyg (Lond)* 71:163–170.
5. Kreplin CM, Ruhnke HL, Miller RB, Doig PA. 1987. The effect of intrauterine inoculation with *Ureaplasma diversum* on bovine fertility. *Can J Vet Res* 51:440–443.
6. Mulira GL, Saunders JR, Barth AD. 1992. Isolation of *Ureaplasma diversum* and mycoplasmas from genital tracts of beef and dairy cattle in Saskatchewan. *Can Vet J* 33:46–49.
7. Amorim A, Marques L, Santos AM, Martins H, Barbosa M, Rezende I, Andrade E, Campos G, Lobão T, Cortez B, Monezi T, Machado-Santelli G, Timenetsky J. 2014. Apoptosis in HEP-2 cells infected with *Ureaplasma diversum*. *Biol Res* 47:38. <http://dx.doi.org/10.1186/0717-6287-47-38>.
8. Buzinhani M, Buim MR, Yamaguti M, Oliveira RC, Mettifogo E, Timenetsky J. 2007. Genotyping of *Ureaplasma diversum* isolates using pulsed-field electrophoresis. *Vet J* 173:688–690. <http://dx.doi.org/10.1016/j.tvjl.2006.02.001>.
9. Kim JJ, Quinn PA, Fortier MA. 1994. *Ureaplasma diversum* infection *in vitro* alters prostaglandin E2 and prostaglandin F2a production by bovine endometrial cells without affecting cell viability. *Infect Immun* 62:1528–1533.
10. Paralanov V, Lu J, Duffy LB, Crabb DM, Shrivastava S, Methé BA, Inman J, Yooseph S, Xiao L, Cassell GH, Waites KB, Glass JI. 2012. Comparative genome analysis of 19 *Ureaplasma urealyticum* and *Ureaplasma parvum* strains. *BMC Microbiol* 12:88. <http://dx.doi.org/10.1186/1471-2180-12-88>.