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Complete Genomic Sequence of an Avian Pathogenic *Escherichia coli* Strain of Serotype O7:HNT

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Avian pathogenic *Escherichia coli* (APEC) is associated with colibacillosis in poultry. Here, we present the first complete sequence of an APEC strain of the O7:HNT serotype and ST73 sequence type, isolated from a broiler with cellulitis. Complete genomes of APEC with distinct genetic backgrounds may be useful for comparative analysis.

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Escherichia coli is a versatile bacterium exhibiting a high degree of genomic plasticity (1). Most strains are harmless; however, a subset of *E. coli* cause intestinal (intestinal pathogenic *E. coli* [InPEC]) or extraintestinal (extraintestinal pathogenic *E. coli* [ExPEC]) diseases in humans and animals (2). In mammals, extraintestinal diseases caused by ExPEC include neonatal meningitis, urinary tract infections, septicemia, mastitis, and pyometra (3–6). ExPEC that produces disease in multiple avian species is known as avian pathogenic *E. coli* (APEC) (7). Collectively, these diseases are known as colibacillosis (7). Colibacillosis is responsible for remarkable losses in the poultry industry worldwide (7). Besides the detrimental impact of colibacillosis on poultry health and the economic viability of the poultry industry, there is evidence that ExPEC from animal origin might contaminate food and cause disease in human beings (8).

A number of APEC have been sequenced. Among them, there are strains belonging to the serogroups O1 (9), O2 (10), and O78 (11), and also nontypeable strains (12). Serogroup O7 strains are commonly detected in human ExPEC (13), where they are known to cause urinary tract infections (13), neonatal meningitis (3), and septicemia (14), and may also occur in APEC (15), where they have caused avian cellulitis in broilers (4) and colibacillosis in turkeys and chickens (16). Most O7 sequenced genomes represent human ExPEC strains, such as uropathogenic *E. coli* (UPEC) IAI39 (accession no. CU928164), UPEC UMN026 (17), and neonatal meningitis *E. coli* (NMEC) CE10 (18). To our knowledge, a complete genome with an APEC O7 strain has not been reported.

The plasmidless *E. coli* strain RS76 of the serotype O7:HNT and sequence type ST93 was isolated from the carcass of a slaughtered broiler diagnosed with avian cellulitis in Brazil (4). The genome was sequenced using 150-bp Illumina paired-end (15,697,322 reads) and 100-bp mate pair (24,920,310 reads) libraries. *De novo* assembly was performed with SPAdes 3.0 (19). The assembly presented an average coverage of 200×. The resulting contigs and scaffolds were ordered with progressiveMauve (20) using *E. coli* BL21(DE3) (accession no. AM946981) as a reference. Gaps were eliminated with PCR and subsequent Sanger sequencing. The APEC RS76 genome sequence was annotated by the NCBI Pro-

karyotic Genomes Automatic Annotation Pipeline (PGAAP). The consistency of the PGAAP annotation was verified against a previous Prokka 1.11 annotation (21).

The single chromosome presented two gaps with unknown length. The total length, excluding gaps, was 4,689,208 bp (50.7% GC content). This chromosome presented 4,407 coding sequences, 84 tRNA encoding genes, and 7 rRNA encoding operons.

There are two general issues regarding the impact of APEC that still need to be elucidated. The genetic determinants associated with APEC pathogenicity in avian species are not fully understood yet. Also, the role of APEC as a zoonotic agent is controversial. The sequencing of complete APEC genomes presenting distinct genetic backgrounds may be useful for future comparative genomic analysis addressing those issues.

Nucleotide sequence accession number. The complete sequence of strain RS76 was deposited in GenBank under the accession number CP013048.

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REFERENCES

1. Kaper JB, Nataro JP, Mobley HLT. 2004. Pathogenic *Escherichia coli*. *Nat Rev Microbiol* 2:123–140. <http://dx.doi.org/10.1038/nrmicro818>.

2. Moriel DG, Rosini R, Seib KL, Serino L, Pizza M, Rappuoli R. 2012. *Escherichia coli*: great diversity around a common core. *mBio* 3:e00118-12. <http://dx.doi.org/10.1128/mBio.00118-12>.
3. Ewers C, Li G, Wilking H, Kiebling S, Alt K, Antão E, Laturnus C, Diehl I, Glodde S, Homeier T, Böhnke U, Steinrück H, Philipp H-C, Wieler LH. 2007. Avian pathogenic, uropathogenic, and newborn meningitis-causing *Escherichia coli*: how closely related are they? *Int J Med Microbiol* 297:163–176. <http://dx.doi.org/10.1016/j.ijmm.2007.01.003>.
4. Maluta RP, Logue CM, Casas MRT, Meng T, Guastalli EAL, Rojas TCG, Montelli AC, Sadatsune T, de Carvalho Ramos M, Nolan LK, da Silveira WD. 2014. Overlapped sequence types (STs) and serogroups of avian pathogenic (APEC) and human extraintestinal pathogenic (ExPEC) *Escherichia coli* isolated in Brazil. *PLoS One* 9:e105016. <http://dx.doi.org/10.1371/journal.pone.0105016>.
5. Maluta RP, Borges CA, Beraldo LG, Cardozo MV, Voorwald FA, Santana AM, Rigobelo EC, Toniollo GH, Ávila FA. 2014. Frequencies of virulence genes and pulse field gel electrophoresis fingerprints in *Escherichia coli* isolates from canine pyometra. *Vet J* 202:393–395. <http://dx.doi.org/10.1016/j.tvjl.2014.08.016>.
6. Wenz JR, Barrington GM, Garry FB, Ellis RP, Magnuson RJ. 2006. *Escherichia coli* isolates' serotypes, genotypes, and virulence genes and clinical coliform mastitis severity. *J Dairy Sci* 89:3408–3412. [http://dx.doi.org/10.3168/jds.S0022-0302\(06\)72377-3](http://dx.doi.org/10.3168/jds.S0022-0302(06)72377-3).
7. Dziva F, Stevens MP. 2008. Colibacillosis in poultry: unravelling the molecular basis of virulence of avian pathogenic *Escherichia coli* in their natural hosts. *Avian Pathol* 37:355–366. <http://dx.doi.org/10.1080/03079450802216652>.
8. Bélanger L, Garenaux A, Harel J, Boulianne M, Nadeau E, Dozois CM. 2011. *Escherichia coli* from animal reservoirs as a potential source of human extraintestinal pathogenic *E. coli*. *FEMS Immunol Med Microbiol* 62:1–10. <http://dx.doi.org/10.1111/j.1574-695X.2011.00797.x>.
9. Johnson TJ, Kariyawasam S, Wannemuehler Y, Mangiamale P, Johnson SJ, Doetkott C, Skyberg JA, Lynne AM, Johnson JR, Nolan LK. 2007. The genome sequence of avian pathogenic *Escherichia coli* strain O1:K1:H7 shares strong similarities with human extraintestinal pathogenic *E. coli* genomes. *J Bacteriol* 189:3228–3236. <http://dx.doi.org/10.1128/JB.01726-06>.
10. Zhu Ghe X, Jiang J, Pan Z, Hu L, Wang S, Wang H, Leung FC, Dai J, Fan H. 2014. Comparative genomic analysis shows that avian pathogenic *Escherichia coli* isolate IMT5155 (O2:K1:H5; ST complex 95, ST140) shares close relationship with ST95 APEC O1:K1 and human ExPEC O18:K1 strains. *PLoS One* 9:e112048. <http://dx.doi.org/10.1371/journal.pone.0112048>.
11. Mangiamale P, Nicholson B, Wannemuehler Y, Seemann T, Logue CM, Li G, Tivendale KA, Nolan LK. 2013. Complete genome sequence of the avian pathogenic *Escherichia coli* strain APEC O78. *Genome Announc* 1(2):e0002613. <http://dx.doi.org/10.1128/genomeA.00026-13>.
12. Rojas TCG, Maluta RP, Parizzi LP, Koenigkan LV, Yang J, Yu J, Pereira GAG, Dias da Silveira W. 2013. Genome sequences of avian pathogenic *Escherichia coli* strains isolated from Brazilian commercial poultry. *Genome Announc* 1(2):e0011013. <http://dx.doi.org/10.1128/genomeA.00110-13>.
13. Smith JL, Fratacamico PM, Gunther NW. 2007. Extraintestinal pathogenic *Escherichia coli*. *Foodborne Pathog Dis* 4:134–163. <http://dx.doi.org/10.1089/fpd.2007.0087>.
14. Maluta RP, Logue CM, Tiba Casas MR, Meng T, Lopes Guastalli EA, Galvao Rojas TC, Montelli AC, Sadatsune T, de Carvalho Ramos M, Nolan LK, da Silveira WD. 2014. Overlapped sequence types (STs) and serogroups of avian pathogenic (APEC) and human extraintestinal pathogenic (ExPEC) *Escherichia coli* isolated in Brazil. *PLoS One* 9:e105016. <http://dx.doi.org/10.1371/journal.pone.0105016>.
15. Rodriguez-Siek KE, Giddings CW, Doetkott C, Johnson TJ, Fakhr MK, Nolan LK. 2005. Comparison of *Escherichia coli* isolates implicated in human urinary tract infection and avian colibacillosis. *Microbiology* 151:2097–2110. <http://dx.doi.org/10.1099/mic.0.27499-0>.
16. Pfaff-McDonough SJ, Horne SM, Giddings CW, Ebert JO, Doetkott C, Smith MH, Nolan LK. 2000. Complement resistance-related traits among *Escherichia coli* isolates from apparently healthy birds and birds with colibacillosis. *Avian Dis* 44:23–33. <http://dx.doi.org/10.2307/1592504>.
17. Touchon M, Hoede C, Tenaillon O, Barbe V, Baeriswyl S, Bidet P, Bingen E, Bonacorsi S, Bouchier C, Bouvet O, Calteau A, Chiapello H, Clermont O, Cruveiller S, Danchin A, Diard M, Dossat C, Karoui ME, Frapy E, Garry L, Ghigo JM, Gilles AM, Johnson J, Le Bouguéne C, Lescat M, Mangenot S, Martinez-Jéhanne V, Matic I, Nassif X, Oztas S, Petit MA, Pichon C, Rouy Z, Ruf CS, Schneider D, Tourret J, Vacherie B, Vallenet D, Médigue C, Rocha EP, Denamur E. 2009. Organised genome dynamics in the *Escherichia coli* species results in highly diverse adaptive paths. *PLoS Genet* 5:e1000344. <http://dx.doi.org/10.1371/journal.pgen.1000344>.
18. Lu S, Zhang X, Zhu Y, Kim KS, Yang J, Jin Q. 2011. Complete genome sequence of the neonatal-meningitis-associated *Escherichia coli* strain ce10. *J Bacteriol* 193:7005. <http://dx.doi.org/10.1128/JB.06284-11>.
19. Bankevich A, Nurk S, Antipov D, Gurevich AA, Dvorkin M, Kulikov AS, Lesin VM, Nikolenko SI, Pham S, Prjibelski AD, Pyshkin AV, Sirotkin AV, Vyahhi N, Tesler G, Alekseyev MA, Pevzner PA. 2012. SPAdes: a new genome assembly algorithm and its applications to single-cell sequencing. *J Comput Biol* 19:455–477. <http://dx.doi.org/10.1089/cmb.2012.0021>.
20. Darling AE, Mau B, Perna NT. 2010. progressiveMauve: multiple genome alignment with gene gain, loss and rearrangement. *PLoS One* 5:e11147. <http://dx.doi.org/10.1371/journal.pone.0011147>.
21. Seemann T. 2014. Prokka: rapid prokaryotic genome annotation. *Bioinformatics* 30:2068–2069. <http://dx.doi.org/10.1093/bioinformatics/btu153>.