Extraintestinal pathogenic *Escherichia coli* (ExPEC) ST221 isolated in the microbiome of swine in South Africa

To the Editor: Extraintestinal pathogenic *Escherichia coli* (ExPEC) strains are versatile bacteria that have the ability to cause human extraintestinal infections such as urinary tract infections, neonatal meningitis, and sepsis worldwide.[1] They lead to significant medical and societal costs in both healthcare and community settings. ExPEC are known for their extensive intrinsic virulence potential based on the number and diversity of specific putative or proven virulence genes regardless of their isolation source. They usually inhabit the intestinal microbiome of humans and animals, from which they can subsequently emerge to cause extraintestinal infections. There is therefore a thin line between the non-pathogenic commensal *E. coli* and ExPEC strains, and it has been hypothesised that ExPEC virulence may be a by-product of the commensal lifestyle.[2] In addition, the emergence of the extended-spectrum beta-lactamase (ESBL) genes encoding for resistance to beta-lactam antibiotics has exacerbated their pathogenicity.

A total of 432 rectal and nasal samples from pigs collected from 5 abattoirs (3 in Cameroon and 2 in South Africa (SA)) were phenotypically screened and tested for antimicrobial susceptibility, with a selected subsample undergoing whole-genome sequencing (WGS) and bioinformatics analysis as previously described.[3] The Comprehensive Antibiotic Resistance Database (CARD), ResFinder, VirulenceFinder, VFanalyzer, MGEFinder and PlasmidFinder were used to identify resistance genes, virulence factors, mobile genetic elements and plasmids, respectively. The multilocus sequence type (MLST) was also determined from WGS data.

One of the ESBL-producing *E. coli* isolates, PN254E (GenBank accession number VKOR00000000), showed resistance to beta-lactam and non-beta-lactam antibiotics. This resistance phenotype was corroborated by the identification of the $bla_{\text{CTX-M-1}}$ and $bla_{\text{CTX-M-14}}$ genes through WGS, which also delineated a collection of resistance genes encoding target modification, antibiotic inactivation, antibiotic efflux pumps and regulators (Table 1). The PN254E isolate had a novel combination of known *E. coli*

Table 1. Demographic and genomic features of *Escherichia coli* PN254E

Isolate (ST)	Country (abattoir)	Sample type	P score	Antibiotic resistance genes	Virulence factors	Mobile genetic elements			
						Plasmids (pMLST)	Insertion sequence	Transposon	PAI
PN254E (221)	South Africa (SH005)	Nasal swab	0.924	*aph(3′)-IIa, strA, strB, aadA1, aadA2, aadA3, AmpC1, ampH, aph(3″)-Ib, aph(3″)-Ib, aadA5, aph(6)-Id, blaCTX-M-14, blaCTX-M-1, tet(A), cmlA1, fosA3, oqxA, oqxB, sul2, sul3, evgA, emrY, emrR, PmrF, mdtH, mdtG, baeS, acrA, msbA, AcrE, marA, acrF, qacH, KpnE, KpnF, PBP3 (D350N, S357N), gyrA (p.S83L), gyrA (p.D87N), mdf(A)*	*cfaA, cfaB, cfaD/cfaE, cgsD, cgsE, cgsF, cgsG, cgsA, cgsB, ecpA, ecpB, ecpC, ecpD, ecpE, elfA, elfC, elfD, elfG, eaeH, focC, focD, focG, focH, focI, hcpA, hcpB, hcpC, papA, papC, papD, papX, sfaC, sfaG, fimA, fimB, fimC, fimD, fimE, fimF, fimG, fimH, fimI, fimZ, htpB, flmH, nueA, plr/gapA, pilT, pilQ, pilR, pilS, pilW, agn43, ehaA, upaG/ehaG, vat, ibeB, ibeC, cheB, cheR, cheW, cheY, cheZ, motA, motB, iutA, iucA, iucB, iucC, iucD, chuA, sitC, sitD, iroC, iroD, iroE, iroN_fyuA, irp1, irp2, ybtA, ybtE, ybtP, ybtU, ybtX, fagC, ccmF, entB, entC, entE, entF, fepB, fepC, fepD, fepG, hemC, hemE, hemG, hemH, hemL, hemN, espL1, espL4, espR, espX1, espX4, espX5, ast, hlyA, cysC1, rmlB, wbjD/wecB, galF, wcaL, wzb, wzc, adeG, pgaC, msbA, lpxABCDK, kdtA, htrB, rfaDEF, wecA, galU, galE, mrsA/glmM, acpXL, rmlA*	IncI1(ST3),* IncI2, IncFIB(K89:A-:B1), IncFIC(FII), IncHI2, IncHI2A, ColpVC, ColRNAI, repA	IsVsa3,† ISEc9,‡ ISEc40,§ ISEc38,¶ IS629, ISEc13, IsKpn8, ISVsa5, IS421, IS3, ISEsa1, IS26, MITEEC1	ISSbo1, Cn_2198_ ISSbo1, Tn6082	*HPI*

ST = sequence type; P score = pathogenicity score; pMLST = plasmid multilocus sequence type; PAI = pathogenicity islands; *HPI* = highly pathogenic island.
Synteny of resistance and virulence genes and MGEs:
*IncI1 harboured the composite transposon Issbo1 and cn_2198_Issbo1.
†IsVsa3 encoded the resistance gene *sul2*.
‡ISEc9 encoded the resistance gene $bla_{\text{CTX-M-1}}$ and the virulence factor *cib*.
§ISEc40 encoded the virulence factor *focI*.
¶ISEc38 encoded the virulence factor *mchB*.

MLST alleles and was ascribed to the sequence type (ST) 221 and serotype O34:H9. It further harboured several virulence factors including the ferric aerobactin receptor (*iutA*), increased serum survival (*iss*), heat-resistant agglutinin (*hra*), temperature-sensitive haemagglutinin (*tsh*), P fimbrial adhesin (*papA*, *papC*, *papD*, *papX*), type I fimbriae (*fimA*, *fimB*, *fimD*, *fimE*, *fimF*, *fimG*, *fimI*), F1C fimbriae (*focC*, *focD*, *focG*, *focH*, *focI*), salmochelin siderophore (*iroN*), yersiniabactin siderophore (*fyuA*) and haemolysin *HlyA* that are commonly detected in several *E. coli* strains responsible for human extraintestinal infections. Numerous mobile genetic elements such as the IncFIB(AP001918 [F89:A-:B1]), IncI1 (ST3), and IncHI2 (unknown ST) plasmid incompatibility groups, the high-pathogenicity island (*HPI*), the transposon Tn6082 and an array of insertion sequences were also detected, as shown in Table 1.

To the best of our knowledge, this is the first report on the presence of an ExPEC ST221-fimH9 serotype O34:H9 harbouring the *HPI*, hypervirulent plasmid IncI1 ST3, and over 100 virulence factors isolated from the swine microbiome in SA, and indeed in Africa. Our report clearly shows that the gut microbiome of swine is also a reservoir of ESBL-producing ExPEC and a potential source of virulence and resistance genes that may be transferred to other bacteria prevailing in the microbiome. The combination of virulence and drug resistance in pathogenic bacteria highlights the worrisome situation of a likely dearth of therapeutic alternatives for some serious bacterial infections in the near future. This phenomenon, coupled with high prevalence of immunocompromised individuals in the sub-Saharan African region, calls for increased surveillance of the population structure of ExPEC in order to preserve the general public from highly virulent and resistant bacterial infections. Stringent efforts to ensure rational antibiotic use in agriculture are a further imperative to safeguard and preserve antibiotics for future generations.

Ethical approval. Ethical approval was obtained from the Biomedical Research Ethics Committee (ref. no. BE365/15) and the Animal Research Ethics Committee (ref. no. AREC/091/015D) of the University of KwaZulu-Natal, as well as from the National Ethics Committee for Research in Human Health of Cameroon (ref. no. 2016/01/684/CE/CNERSH/SP) prior to starting the study. Ministerial approvals were also obtained from the Cameroonian Ministry of Livestock, Fisheries and Animal Industries (ref. no. 061/L/MINEPIA/SG/DREPIA/CE) and Ministry of Scientific Research and Innovation (ref. no. 015/MINRESI/B00/C00/C10/C14). The study was further recorded by the Department of Agriculture, Forestry and Fisheries (ref. no. 12/11/1/5 (878).

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