The Present & the Past: A Review of Strains Accessioned into the National Collection of Type Cultures

Jake David Turnbull, Sunita Gurung, Julie E. Russell, Sarah Alexander

The National Collection of Type Cultures, Public Health England, Colindale, London, UK

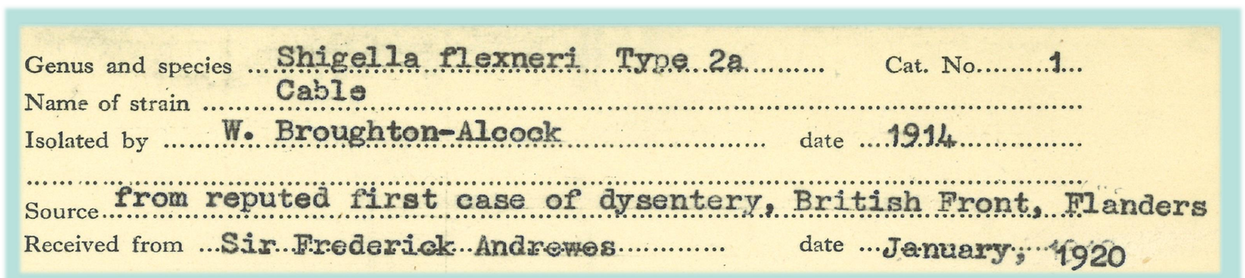
Public Health England

Protecting and improving the nation's health

INTRODUCTION

The UK's National Collection of Type Cultures (NCTC) is one of the world's longest established collections of bacterial strains, currently in its 100th year of operation. The collection houses considerable taxonomic and biological diversity, holding approximately 6000 bacterial strains from more than 800 species of bacteria of medical and veterinary importance.

New strains are added to the collection every year to ensure that it remains relevant to the biomedical diagnostic and research community that it serves. Here we review their value to the scientific community, both in their own right and in the context of bacterial strains already held by the NCTC.



Genus and species ...Shigella flexneri Type 2a..... Cat. No.....1...
Name of strain .....Cable.....
Isolated by .....W. Broughton-Alcock..... date ...1914.....
.....
Source...from reputed first case of dysentery, British Front, Flanders
Received from ...Sir Frederick Andrewes..... date ...January, 1920

Figure 1. An accession record for NCTC 1; the first strain deposited into the collection

METHODS

The two main ways of adding to the collection are from microbiologists offering to donate strains of scientific interest, or the collection's curators requesting strains; for example strains cited in scientific publications. All strains and associated metadata submitted to the collection are reviewed to ensure they meet the NCTC remit. It can take up to six months for a newly deposited isolate to become a fully accessioned NCTC strain.

The depositor sends a sample of the bacteria to NCTC, in duplicate, via an appropriate courier

A NCTC number is assigned. NCTC preserves the strain by freeze drying, and carries out internal quality control checks ensuring the purity, viability, identity and select traits of the strain

An ampoule of the freeze dried strain is sent to the depositor for additional checks that the strain remains unchanged by the preservation process

Following a successful depositors check, the strain is added to the NCTC online catalogue and the depositor is informed it has been released for distribution to the scientific community

Some strains (such as novel species) are not made available until the relevant peer-reviewed manuscripts are published.

RESULTS

OVERVIEW

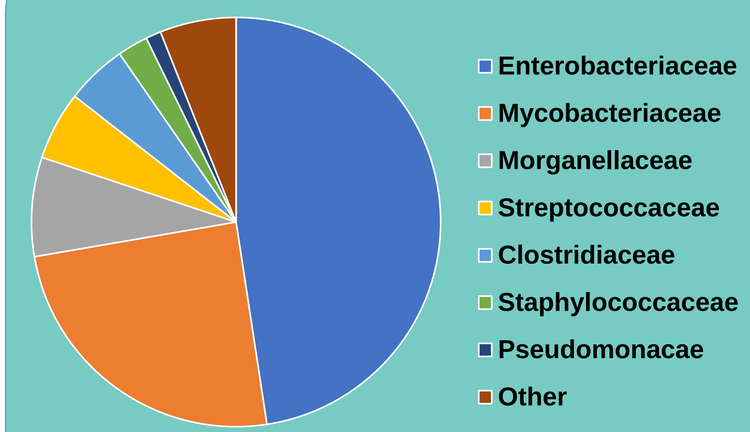


Fig. 2. The proportion of strains made available from the NCTC in 2018, by taxonomic family

166 bacterial strains were made available from the National Collection of Type Cultures in 2018.

The Murray Collection of pre-antibiotic era (1917-1954) Enterobacteriaceae

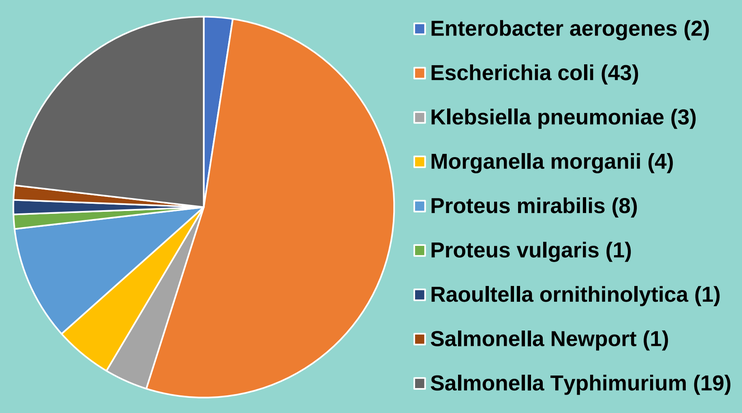


Fig. 3. The 82 strains from the Murray Collection were made available from the NCTC in 2018, by species

The 43 *Escherichia coli* strains were screened via whole genome sequencing for the virulence genes *eae, aggR*, *ipaH* and *stx.* All 43 strains are negative for all genes, with the exception of NCTC 13967 and NCTC 13973, which are positive for *ipaH* (associated with invasive infection)

Preterm infant-associated *Clostridium sp.*

NCTC / Strain No.	Organism	ENA WGS Accession
14034 / LH009	*Clostridium tertium*	GCA_900217175.1
14035 / LH025	*Clostridium paraputrificum*	GCA_900217185.1
14036 / LH052	*Clostridium cadaveris*	GCA_900217165.1
14037 / LH058	*Clostridium paraputrificum*	GCA_900217195.1
14038 / LH141	*Clostridium paraputrificum*	GCA_900217205.1

Table 1. 5 strains deposited by R. Kiu and L. Hall, following a wider study examining the microbiota of infants[1].

Recently described species & type strains

4 recently described species were made available in 2018

NCTC 13658 *Porphyromonas loveana* **Bird et al. 2016[2]**
Obligately anaerobic, pigmented, Gram negative species from a marsupial

NCTC 13847 *Treponema ruminis* **Newbrook et al. 2017[3]**
Spirochaete from rumen contents of slaughtered bull

NCTC 13950 *Staphylococcus cornubiensis* **Murray et al. 2018[4]**
A *Staphylococcus intermedius* group organism, isolated in pure culture from a human skin infection

NCTC 14063 *Staphylococcus caeli* **MacFadyen et al. 2018[5]**
A strain from industrial rabbit holding, with novel mecC gene allele mecC3

Six type strains not previously included in the NCTC were made available in 2018:

NCTC 13760 *Streptococcus infantarius;* **NCTC 13772** *Carnobacterium divergens;* **NCTC 13807** *Streptococcus oligofermentans;* **NCTC 13936** *Listeria borriae;* **NCTC 14061** *Macrococcus caseolyticus;* **NCTC 14210** *Burkholderia thailandensis*

NCTC 14052: An Emergent Pathogen

A strain of *Klebsiella pneumoniae* described by MMC Lam et al. 2018[6] as having a genome representative of an emerging hyper-virulent pathotype was submitted to NCTC, is now available as **NCTC 14052**

- The strain belongs to the CG23-I sub-lineage predominantly found in liver abscess infection
- Carries the HvKp virulence plasmid, intact copies of all virulence genes and no atypical accessory genes.

Contemporary antimicrobial resistant (AMR) strains

NCTC No.	Organism	Determinant / Phenotype
13713	*P. aeruginosa*	Upregulated intrinsic AmpC activity
13780	*E. faecalis*	vanA type glycopeptide resistance, from a contemporary hospital adapted lineage.
13952	*Salmonella* Typhimurium	mcr-1 mediated colistin resistance
13953	*Salmonella* Senftenberg	Carries a blaNDM-1 positive plasmid
13954	*Salmonella* Typhimurium	Carries a blaOXA-48a-like positive plasmid
14055	*Enterobacter asburiae*[7]	Novel blaFRI-2 non-metallo-carbapenemase gene
14056	*Pseudomonas guariconensis*	blaDIM metallo-carbapenemase gene
14089	*C. freundii*	blaGIM metallo-carbapenemase gene
14143	*S. pneumoniae*	Resistant to clindamycin, erthromycin, tetracycline, intermediate resistance to ampicillin, cefotaxime, penicillin
14208	*N. gonorrhoeae*	High-level azithromycin and high-level ceftriaxone resistance

Table 2. 10 strains with clinically relevant AMR mechanisms or determinants were made available from the NCTC in 2018. These strains were all deposited by PHE Bacteriology Reference Department, with the exception of NCTC 13780 which was deposited by K. E. Raven (University of Cambridge, UK)

Streptococcus agalactiae

NCTC No	Antigen	ENA WGS Accession
14091	III	ERS2510481
14092	Ib	ERS2510482
14093	II	ERS2510483
14094	Ia	ERS2510484
14095	V	ERS2510485

Table 3. Whole genome sequenced *Streptococcus agalactiae*, for use in opsonophagocytic killing assays and vaccine development. Deposited by A. Gorringe (PHE Pathogen Immunology Group, UK)

DISCUSSION

- For a bacterial species to be formally described, the type strain must be deposited into two recognized culture collections in two different countries. As demonstrated by the 10 type strains made available in 2018, NCTC continues to fulfil its role in the description of prokaryotic species, particularly those of clinical and veterinary interest.
- Progress continues to be made in making the historic (1917-1954) Murray collection of *Enterobacteriaceae* available for research
- **NCTC 14052** belongs to an emergent pathotype and has been made available with the aim of improving understanding the virulence factors that lead to specific HvKp pathologies and how well this lineage retains AMR determinants.
- Strains such as **NCTC 14034-14038** are linked to neonatal pathologies. Access to these strains and whole genome sequence data will help elucidate the pathogenicity of these species in neonates
- **NCTC 14055**, **NCTC 14056**, **NCTC 14089,** all produce either novel or less common types of carbapenemase and their addition to the collection provides clinical microbiologists adequate controls for their detection. **NCTC 14055**, **NCTC 14056**, **NCTC 14089**, supplement the other more widely disseminated types of carbapenemase producing strains already well represented in the NCTC.
- **NCTC 14208** has been included in the WHO panel of *Neisseria gonorrhoeae* strains for interlaboratory antimicrobial susceptibility testing quality assurance as **WHO Q,** complementing **NCTC 13477-84** (WHO F, G, K, L, M, N, O, P) and **NCTC 13817-22** (WHO U, V, W, X, Y, Z). It is the first globally reported strain of N. gonorrhoeae with both high-level azithromycin and ceftriaxone resistance.
- **NCTC 14091-14095** develop the NCTC's ability to support public health improvements via vaccine development

ACKNOWLEDGEMENTS

The authors would like to thank all NCTC staff members past and present for their work in curating the collection, and the depositors of the strains made available in 2018: A. K. Murray, Culture Collection University of Gothenburg, Dr. G. Paterson, Dr. Lindsay Hall , J. Slager, K. E. Raven, K. Newbrook, P. S. Bird, the units of the PHE Bacterial Reference Department, Prof. G. Yunn-Hwen, Prof. J. Vazquez-Boland, R. G. E. Murray, R. Kui, The late Prof. Naomi Datta, The Collection de L'Institut Pasteur, The Deutsche Sammlung von Mikroorganismen und Zellkulturnen.

CONCLUSIONS

For almost 100 years, present and past curators have ensured that the NCTC remains scientifically valuable. The number of type strains available for use as taxonomic reference, including those of novel species, has increased, as has the number and diversity of pathogens which range from strains of great clinical significance to more obscure or emerging pathogens.

If you are interested in depositing bacterial strains into the National Collection of Type Cultures, please contact

NCTCOperations@PHE.gov.uk

REFERENCES

1. Kiu R. et al. Preterm Infant-Associated Clostridium tertium, Clostridium cadaveris, and Clostridium paraputrificum Strains: Genomic and Evolutionary Insights. Genome Biol Evol. 2017 Oct 1;9(10):2707-2714.
2. Bird, P. S., et al. 2016. Porphyromonas loveana sp. nov., isolated from the oral cavity of Australian marsupials. IJSEM, 66, 3771-3778.
3. Newbrook, K., et al. 2017. Trepnonema ruminis sp. nov., a spirochaete isolated from the bovine rumen. IJSEM, 67, 1349-1354.
4. Murray, A. K, et al. 2018. *Staphylococcus cornubiensis* sp. nov., a member of the *Staphylococcus intermedius* Group (SIG). IJSEM, 68, 3404-3408.
5. MacFadyen A.C., et al. 2018. *Staphylococcus caeli* sp. nov., isolated from air sampling in an industrial rabbit holding. IJSEM, 69, 82-86.
6. Lam, M.M.C., Wyres, K. L., et al. Population genomics of hypervirulent Klebsiella pneumoniae clonal-group 23 reveals early emergence and rapid global dissemination. Nat Commun. 2018; 9: 2703.
7. Meunier D, et al. FRI-2 carbapenemase-producing Enterobacter cloacae complex in the UK. J Antimicrob Chemother. 2017 Sep 1;72(9):2478-2482.