Escherichia coli as a Multifaceted Pathogenic and Versatile Bacterium

Vânia Santos Braz†, Karine Melchior and Cristiano Gallina Moreira*†

Department of Biological Sciences, School of Pharmaceutical Sciences, São Paulo State University (UNESP), Araraquara, Brazil

Genetic plasticity promotes evolution and a vast diversity in Escherichia coli varying from avirulent to highly pathogenic strains, including the emergence of virulent hybrid microorganism. This ability also contributes to the emergence of antimicrobial resistance. These hybrid pathogenic E. coli (HyPEC) are emergent threats, such as O104:H4 from the European outbreak in 2011, aggregative adherent bacteria with the potent Shiga-toxin. Here, we briefly revisited the details of these E. coli classic and hybrid pathogens, the increase in antimicrobial resistance in the context of a genetically empowered multifaceted and versatile bug and the growing need to advance alternative therapies to fight these infections.

Keywords: treatment, genetic mobility, pathogenesis, Escherichia, multiresistant

INTRODUCTION

Escherichia coli (or E. coli) is a Gram-negative versatile bacterium, easily found and amenable to natural and random genetic alteration. There is a vast collection of sequenced E. coli genomes which exhibit different sizes and genomic diversity among commensal and pathogens, indicating a great assortment within the same bacterial species. They comprise of non-pathogenic bacteria that may act as commensals and belong to the normal intestinal microbiota of humans and many animals. There are also pathogenic variants, divided as diarrheagenic and extraintestinal pathogens, with different pathotypes and various natural hybrid strains (Tables 1 and 2). These variants can be facultative or obligate pathogens. The facultative bacteria are part of the intestinal tract and may act as opportunistic pathogens when outside of their natural habitat, causing various types of extraintestinal infections. On the other hand, intestinal obligate pathogenic variants cause infections in distinct conditions, from moderate diarrhea to more threatening cases, as lethal outcome (Kaper et al., 2004; Köhler and Dobrindt, 2011).

E. coli pan-genome studies indicate enormous capacity to evolve by gene acquisition and genetic modification. Besides, these genomes have a mosaic-like structure consisting of a core genome, encoding essential cellular functions, and an accessory genome with flexible strain-specific sequences. Thus, E. coli is a model well established for studying the interdependence of genome architecture and the lifestyle of bacteria (Touchon et al., 2009; Dobrindt et al., 2010).

Based on virulence factors in E. coli genomes and phenotypic traits, the human pathotypes of diarrheagenic E. coli (DEC) are differentiated from non-pathogenic E. coli and extraintestinal pathogenic E. coli (ExPEC). The ExPEC are classified as uropathogenic E. coli (UPEC), sepsis-causing E. coli (SEPEC) and neonatal meningitis-associated E. coli (NMEC) (Kaper et al., 2004). Recent pathogenomics and phenotypic classification have revisited the DEC group as nine distinct
| E. coli Pathotype (DEC and ExPEC) | Main virulence traits | Clinical manifestation | Antimicrobial resistance (AMR) commonly found | Mobile genetic resistance determinants | References |
|----------------------------------|-----------------------|-----------------------|-----------------------------------------------|---------------------------------------|------------|
| Shiga toxin-producing (STEC)     | Shiga-toxin           | Not associated with human diseases | Streptomycin, Ampicillin, Tetracyclines and sulfonamides | ND | Jerse et al., 1990; Kaper et al., 2004; Day et al., 2017; Knutton et al., 1989; Mellies et al., 1999; Kaper et al., 2004; Garmendia et al., 2005; Day et al., 2017 |
| Enterohemorrhagic (EHEC)         | EscF, EscC, EspA, EspB, EspD, Intimin, Tir, and Shiga-toxin | Bloody diarrhea and HUS | Streptomycin, Ampicillin, Tetracyclines and sulfonamides | Resistance plasmid-mediated (as pO157, pO111-CRL115, pO26-CRL125, pO145-13514) | Tobe et al., 1999; Trabulsi et al., 2002; Kaper et al., 2004; Ingle et al., 2018 |
| Enteropathogenic (EPEC)          | EscF, EscC, EspA, EspB, EspD, Intimin, Tir, EAF plasmid (tEPEC) and Bfp (tEPEC) | Watery diarrhea | Streptomycin, Ampicillin, Tetracyclines, Trimethoprim and Sulfamethoxazole | Resistance plasmid-mediated (as pEAF, MB80, pB171_90, pEO208) | Regu-Mangia et al., 2009; Aslani et al., 2011; Gomes et al., 2016; Pavlovskisa and Sobieszczanska, 2017; Chattaway et al., 2017 |
| Enteroaggregative (EAEC)         | pAA plasmid, aggregated fimbiae adhesion (AAF), AggR regulator and dispersin | Acute and chronic diarrhea | Ampicillin, Trimethoprim, Sulfamethoxazole, Naldixic acid, and ciprofloxacin | Resistance plasmid-mediated (as pAA), chromosomal gyrB and parC mutations | Kaper et al., 2004; Baylis et al., 2006; Gomes et al., 2016; Pavlovskisa and Sobieszczanska, 2017 |
| Enteroinvasive (IEEC)            | Plasmid pINV and invasins | Bacillary Dysentery | Carbapenem, fosfomycin-trimethanol, nitrofurantoin, chloramphenicol, β-lactams, naldixic acid, ampicillin and fluoroquinolones | Resistance plasmid-mediated, chromosomal gyrB and parC mutations | Kaper et al., 2004; Medina et al., 2015; Gomes et al., 2016; Pavlovskisa and Sobieszczanska, 2017 |
| Enterotoxigenic (ETEC)           | Thermstable (ST) and thermtolable (LT) enterotoxins | Watery diarrhea, known as traveler’s diarrhea | Ampicillin, sulfamethoxazole, tetracyclines and azithromycin | Resistance plasmid-mediated (distinct Inc type conjugative plasmids) | Kaper et al., 2004; Nash et al., 2010; Servin, 2014; Gomes et al., 2016 |
| Diffusely-adhering (DAEC)        | Afa/Dr adhesins       | Acute diarrhea to assymptomatic cases | Ampicillin, Trimethoprim, Sulfamethoxazole, Fosfomycin, piperacillin, tetracyclines, ciprofloxacin, co-trimoxazole, nitrofurantoin, oxacillin, bactericin, cloxacillin, chloramphenicol, and naldixic acid | Resistance plasmid-mediated, chromosomal gyrB and parC mutations | Kaper et al., 2004; Nash et al., 2010; Barrios-Villa et al., 2017 |
| Adherent-invasive (AIEC)         | type VI secretion system, type I pili, long polar fimbiae | Chronic gut inflammation and Crohn’s disease | Ampicillin and ciprofloxacin | Resistance plasmid-mediated, chromosomal gyrB and parC mutations | Kaper et al., 2004; Nash et al., 2010; Barrios-Villa et al., 2017 |
| Cell-detaching (CDEC)            | K-hemolysin, pyelonephritis-associated pili and cytotoxic necrotizing factor 1 (CNF1) | Diarrhea in infants, cell detaching, and linked to Crohn’s disease cases | Amoxicillin-clavulanic acid, ampicillin, mezlocillin, piperacillin, tetracycline, trimethoprim, trimethoprim-sulfamethoxazole, spectinomycin, streptomycin and sulfonamide | Resistance plasmid-mediated, integrons | Elliott et al., 1998; Fábrega et al., 2002; Okeke et al., 2002; Kaper et al., 2004; Rakotina et al., 2017 |
| Uropathogenic (UPEC)             | P fimbiae, certain other mannosate-resistant adhesins, and type 1 fimbiae, K capsule, Hemolysin, Aerobactin | Urinary and Bloodstream infections | Fluoroquinolone, aminoglycosides, trimethoprim-sulfamethoxazole and carbapenems | Resistance plasmid-mediated, transposons, integrons, chromosomal | Kaper et al., 2004; Mobley et al., 2009; Petty et al., 2014 |

(Continued)
pathotypes, proposed by their differential features and the essential virulence genes defining each subgroup, such as Shiga toxin-producing E. coli (STEC), enterohemorrhagic E. coli (EHEC), enteropathogenic E. coli (EPEC), enterotoxigenic E. coli (ETEC), enteroinvasive E. coli (EIEC), enteroaggregative E. coli (EAEC), diffusely-adhering E. coli (DAEC), adherent-invasive E. coli (AIEC), and cell-detaching E. coli (CDEC) (Kaper et al., 2004; Pawlowska and Sobieszczanska, 2017) (Table 1).

Herein, we briefly describe the diversity of these classic and novel emerging E. coli pathotypes and their genetic plasticity in a multifaceted organism. The mobile genetic elements are responsible for the appearance of novel hybrid strains with distinct assortment of virulence and antimicrobial resistance traits, bringing up the urgent need to reconsider the forms of treatment for these infections.

**TYPES OF E. COLI: MANY FLAVORS WITHIN A SINGLE BACTERIAL SPECIES**

E. coli is one of the most genetically versatile microorganisms and is able to colonize and persist in several niches, both in the environment or in hosts. Commensal E. coli strains colonize the gastrointestinal tract of humans a few hours after birth, resulting in a symbiotic relationship between the microbiota and its host (Ducarmon et al., 2019). However, the mechanisms by which E. coli ensures this efficient symbiosis is not well known. It could be related to its high ability to use nutrients in the colon (Fabich et al., 2008; Ducarmon et al., 2019). Several studies have shown that competition for nutrients between microbiota and pathogens limits the colonization of the pathogens, leading to fierce competition among these microorganisms (Lustri et al., 2017).

Occasionally, pathogenic E. coli cannot be distinguished from commensal E. coli, only based on specific virulence factors, as some previously described in ExPEC strains (Köhler and Dobrindt, 2011). However, this scenario is changing due to sophistication and availability of molecular typing methodologies. New computational approaches bring countless important information about host-pathogen relationships, reservoir, clinical diagnoses, and novel ExPEC transmission pathways (Johnson and Russo, 2018). Often, virulence genes are located in transmissible genetic elements such as genomic islands, bacteriophages, insertion sequences (ISs), integrons, plasmids, and transposons; hence, they can be easily exchanged among different bacteria (Hacker et al., 2003; Dobrindt et al., 2010). They also carry multiple antibiotic resistance genes that have been under strong selective pressure as consequence of the extensive use of antibiotics (Brzuskiewicz et al., 2009).

Common genetic changes in E. coli genomes ensure high diversity due to the gain and loss of genes through genetic modification events. There are many strains of ExPEC that normally colonize the gut asymptptomatically, as members of the intestinal microbiota. Nonetheless, only a subset of ExPEC as UPEC, SEPEC and NMEC are responsible for the vast majority of infections such as urinary tract infections, sepsis, and meningitis (Kaper et al., 2004). There is a great variety of virulence factors in ExPEC strains, such as adhesins (fimbrial and non-fimbrial), siderophores, toxins, invasins, the ability to survive in serum, among others. Moreover, many of these virulence factors may occur combined within the same strain and act synergistically. Despite extra factors, the septic strains always possess at least an adherence system, an iron uptake system and genes for serum survival (Biran and Ron, 2018; Johnson and Russo, 2018) (Table 1).

The genetic evolution in E. coli pathogenesis employs horizontal transfer mechanisms within same and across similar species. Therefore, the IS, transposons and integrons may facilitate novel rearrangements within the genome, such as duplication and suppression of genes and also capture of new genes. This genetic material transit can result in greater flexibility concerning various features, such as the transition of pathogenic bacteria between humans and animals, resistance to antimicrobials, appearance of emerging pathogens due to the gain of virulence genes, increased pathogenicity, among other features (Frost et al., 2005; Brigulla and Wackernagel, 2010; Dobrindt et al., 2010; Jackson et al., 2011; Sheppard et al., 2018). All these conditions may contribute to the virulence of these bacteria, like the bacteriophage importance in the pathogenesis. The horizontal transfer between different strains favors the emergence of new pathogenic strains with discrepancies in the bacteriophage repertoire affecting directly their virulence (Manning et al., 2008; Ogura et al., 2009; Dobrindt et al., 2010; Jackson et al., 2011).
TABLE 2 | Hybrid pathogenic (HyPEC) main features described.

| HyPEC     | Main features       | Hybrid virulence traits identified                                                                 | Clinical manifestation                                      | Antimicrobial resistance (AMR) described                      | References                                                                 |
|-----------|---------------------|-------------------------------------------------------------------------------------------------------|------------------------------------------------------------|----------------------------------------------------------------|--------------------------------------------------------------------------|
| O104:H4   | Hybrid EAEC with STEC | Aggregative typical fimbriae, Shiga toxin                                                           | Diarrhea, HUS                                              | Quinolones and β-lactams                                        | Bielaszewskia et al., 2011; Rasko et al., 2011; Muniesa et al., 2012; Navarro-Garcia, 2014; Ribeiro et al., 2019 |
| O157:H7   | Hybrid STEC with ExPEC | Intimin, Shiga toxin and pSS88-like plasmid                                                          | HUS, Bacteremia                                             | β-lactams                                                     | Peigne et al., 2009; Mariani-Kurkdjian et al., 2014                    |
| O2:H6     | Hybrid STEC with UPEC | α-hlyA, cnf1 and cib genes                                                                           | Diarrhea, Urinary tract infections, HUS                     | ND                                                           | Bielaszewskia et al., 2014                                              |
| ST131     | Hybrid UPEC with ExPEC | pAA plasmid                                                                                           | Urinary infections, Bloodstream infections and Diarrhea     | β-lactams                                                     | Boll et al., 2018                                                       |
| EPEC/ETEC | Hybrid EPEC and ETEC | Intimin, LEE island, ST and LT toxin                                                                  | Diarrhea, Mild fever and cough                             | β-lactams, SUT, and quinolones                                 | Dutta et al., 2015                                                      |
| STEC/ETEC | Hybrid STEC and ETEC | Intimin, Shiga toxin (Stx2) and LT toxin                                                              | Acute diarrhea and HUS                                     | ND                                                           | Lindstedt et al., 2018                                                   |
| O137:H6   | Hybrid EPEC and STEC | Intimin, BFP, Shiga toxin (Stx2) and AIDA-I autotransporter                                         | Diarrhea and HUS                                           | ND                                                           | Gioia-Di Chiacchio et al., 2018                                         |
| STEC/ETEC | Hybrid STEC and ETEC | Intimin, Shiga toxin, ST and LT toxins                                                                | Acute diarrhea and HUS                                     | ND                                                           | Nyholm et al., 2015                                                     |
| EPEC/ETEC | Hybrid EPEC and ETEC | Intimin, BFP, ST and LT toxins                                                                       | Diarrhea, Mild fever and cough                             | ND                                                           | Hazen et al., 2017                                                      |

HyPEC, Hybrid Pathogenic E. coli; HUS, Hemolytic Uremic Syndrome; ND, non-described; SUT, Trimethoprim-sulfamethoxazole.

The co-evolution of bacterial genomes with plasmids, besides potential genetic and phenotypic gain may impact cellular metabolism to ensure the maintenance and stability of the plasmid (Jackson et al., 2011). Many ExPEC virulence genes are encoded within plasmids, often belonging to the ColV family, which encodes colicin, serum survival factors and iron uptake systems (Biran and Ron, 2018). Similarly, intestinal pathogens carry a variety of types of plasmids, associated with virulence, majorly belonging to the incompatibility group IncF, which has transfer functions (Carattoli, 2009). There are virulence plasmids essential for some pathotypes of E. coli, such as pINV and pPAA, respectively, in EIEC and EAEC, according to each own group features (Kaper et al., 2004).

Although, all ExPEC and DEC pathotypes are not enough to fully classify all pathogenic E. coli strains, since these bacteria are so variable, allowing constant appearance of distinct hybrid-formed strains within this dynamic bacterial species. The carriage of virulence genes essential to the pathogenesis of each pathotype and the ability to adapt to different conditions allow the emergence of hybrid pathogenic E. coli (HyPEC).

GENETIC PLASTICITY AND EMERGENT E. COLI PATHOGEN: HYPEC

E. coli has an astonishing facility to amend very well, replicate and disseminate. These features allowed the advent of novel HyPEC. Acquired virulence genes and novel functions appear from mutation, recombination and other genetic changes. All these genetic differences have increased the occurrence of novel hybrid and antimicrobial resistance among DEC and ExPEC (Dobrindt et al., 2003; Bielaszewskia et al., 2007; Khan et al., 2018).

Recently, a HyPEC strain received widespread attention after an outbreak of foodborne bloody diarrhea and hemorrhagic uremic syndrome (HUS) in Germany. This outbreak of E. coli O104:H4 was associated with consumption of raw fenugreek sprouts, as a hybrid EAEC strain with STEC features, like Shiga toxin presence. This HyPEC was quickly sequenced and unraveled its intricate nature, but even with a quick response and identification it was not enough to avoid 3,842 hospitalizations with many fatalities in Europe and North Africa (Bielaszewska et al., 2011; Rasko et al., 2011). Emerging processes are responsible for the HyPEC occurrences. Herein, the combined enteroaggregative features in a rare serotype was responsible to high attachment to cells and a biofilm formation (Navarro-Garcia, 2014; Ribeiro et al., 2019). Moreover, this strain has gained stx2 gene lambdoid phage integrated in the genome, thus it may release the Shiga-toxin. These features have increased HUS occurrence during the outbreak on this HyPEC when compared to STEC (Muniesa et al., 2012).

Many distinct genetic hybrid examples are reported in E. coli, such as STEC/ExPEC O80:H2 serotype, which caused HUS and bacteremia due the presence of stx2 and eae genes from STECs and pSS88-like plasmid, described in meningitis, urosepsis and avian pathogenic strains of ExPEC (Peigne et al., 2009; Mariani-
Kurkdjian et al., 2014). The STEC/UPEC strain O2:H6 serotype, a STEC with virulence genes as α-hlyA, cnf1, and clb from UPEC that have ability to cause diarrhea and urinary tract infections (Bielaszewska et al., 2014). The EPEC/ETEC strain has acquired the LEE island and encodes the LT toxin (Dutta et al., 2015). The broadly reported multidrug resistant E. coli ST131 is example of highly virulent ExPEC associated with urinary and bloodstream infections. It has also acquired enteroaggregative diarrheagenic phenotype due to pAA plasmid presence (Boll et al., 2018). Many others HyPEC are described as case report, but not fully characterized. Here, we have briefly sampled some of the acquired genes by these strains, their direct impact in virulence and their hybrid nature (Table 2). Comparably to these HyPEC, the coined terms hybrid- and hetero-pathogenic E. coli have been recently described as new combination of virulence factors among classic E. coli groups. Together, they show differences between typical and atypical subgroups within the EAEC and EPEC pathotypes and hybrids, such as EPEC/STEC, ExPEC/EPEC and ExPEC/EAEC hybrids (Santos et al., 2020). Similar to our approach here, this study shows how this topic is critical in the field.

The high prevalence of classic pathogenic E. coli and appearance of HyPEC occur via similar genetic mechanisms, which also enable bacteria to resist the presence of distinct antimicrobials. Bacteria resistant to various classes of antibiotics are related to the complex combination of intrinsic and acquired resistance genes, which may act synergistically (Cag et al., 2016; Khan et al., 2018). Together that brings multiresistant bacteria, as an alarming factor reported worldwide in several bacterial species. WHO has prioritized studies on AMR bacteria, including Enterobacteriaceae, based on recent surveillance reports (WHO, 2018).

EMERGING HYBRIDS AND ALTERNATIVE THERAPIES

The complex combination of multidrug-resistant bacteria and emerging hybrid bacteria with intrinsic or acquired bacterial virulence factors disseminated by genetic mobility elements, the intense and inappropriate use of antibiotics have simultaneously favored the emergence of resistance to various antibiotics (Khan et al., 2018). That is a special challenge to these hybrid strains, since these HyPEC gathered virulence traits and acquired antibiotic resistance, together these points raise the importance to alternative treatments. These options are crucial to reduce the use of antibiotics and the consequent increase of antimicrobial resistance. Novel therapies are urgent to replace prophylactic and treatment with antibiotics by probiotics, prebiotics, enzymatic compounds, vaccines, monoclonal antibodies, phage therapy, antivirulence compounds, among other possibilities (Gadde et al., 2017).

Recently, different vaccine strategies have been used for pathogenic E. coli infection as an alternative to antibiotic therapy (Rojas-Lopez et al., 2018), including vaccines with attenuated toxins (McKenzie et al., 2007; Bitzan et al., 2009), attenuated bacterial cell (Calderon Toledo et al., 2011), individual components of virulence factors such as Shiga toxin (Liu et al., 2009), EspA or Intimin (Oliveira et al., 2012), small peptides (Zhang et al., 2011), DNA (García-Angulo et al., 2014) or polysaccharides (Ahmed et al., 2006; van den Dobbelsteen, 2016), as well detailed in the literature. Commercial vaccines have aimed the use to protect livestock, such as poultry, swine and bovine herds, against respectively to APEC, like Poulvac® E. coli, ETEC and EHEC infections (Sadeyesen et al., 2015; Nesta and Pizza, 2018). Vaccines with a modern approach and technology still are a promising strategy to protect against emergent HyPECs infections in humans and livestock.

Recent studies have revisited the phage therapy as a biological alternative, which employs strictly lytic phages uncappable of lysogenization (Carter et al., 2012). Studies have demonstrated ability of phages to decrease biofilm formation in UPEC (Chibeu et al., 2012), increased mice rate survival in E. coli-induced pneumonia (Dufour et al., 2015). Moreover, lytic bacteriophages were used to infect and kill bacteria harboring phage-dependent conjugative plasmid to avoid emergence of multiresistant bacteria (Ojala et al., 2013; Tagliaferri et al., 2019). The phages cocktail EcoShield™ is already commercialized (Intralytix) and it has been reported to significantly reduce the E. coli O157:H7 contamination on surfaces and food (Abuladze et al., 2008; Carter et al., 2012). Additionally, mutual use of phages with antibiotics have emerged, with SPR02 and DAF6 phages combined with enrofloxacin have shown promising data, rescuing chickens challenged with avian pathogenic E. coli infection (Tagliaferri et al., 2019).

The novel approach via antivirulence-directed compounds works disarming the pathogens' ability to cause disease by inhibiting their virulence factors, favoring the host's immune defenses during the bacterial clearance. These compounds do not induce bacterial resistance as antibiotics, because they disarm the pathogen, instead of directly targeting its growth. Therefore, as they are directed to specific factors for pathogenesis, they potentially reduce the selection of resistance and limit collateral damage to the microbiota. Some virulence inhibitors are effective against many pathogens, molecules such as LED209, HCl02A, HCl03A, Artemisinin, and Ethoxzolamide, by inhibit different two-component systems as QseBC in E. coli and other enteropathogens (Sperandio et al., 2003; Rasko et al., 2008; Yang et al., 2014; Xue et al., 2015; Kim et al., 2020), Bicycl 2-pyridones, Biaryl mannoside, Nitazoxanide and FN075, avoiding the initial bacterial adhesion; and compounds like Toxtazins A and B, Ebselen, 7086, 7812, 7832, BPT15, and BBH7, blocking toxins and secretion systems (Payne, 2008; Johnson and Abramovitch, 2017).

CONCLUSION

The forces that shape the evolution in E. coli comprise vast repertoire, affecting genetic flexibility and excessive permissiveness to acquire and donate DNA via horizontal gene
transfer. These features guarantee the spread of antibiotic resistance as well as virulence factors inherited among the various pathotypes of *E. coli*. The exact identification and assessment assist researchers to better understand this bacterium modification, diagnosis, public health and treatment. *E. coli* strains with multiple and distinct factors are probably very common but unreported, since these *E. coli* strains have developed many strategies to persist in different settings and successfully infect the host. These strategies result in an immense variety of microorganisms, ranging from avirulent to extremely virulent strains that can cause intestinal or extraintestinal diseases. *E. coli* strains have great potential for dissemination and capacity to pass along hereditary elements. Currently, these HyPEC strains are a very concerning threat that demands more studies and the development of novel treatment methods.

### REFERENCES

Abuladze, T., Li, M., Menetrez, M. Y., Dean, T., Senecal, A., and Sulakvelidze, A. (2008). Bacteriophages reduce experimental contamination of hard surfaces, tomato, spinach, broccoli, and ground beef by *Escherichia coli* O157:H7. *Appl. Environ. Microbiol.* 74, 6230–6238. doi: 10.1128/AEM.01465-08.21

Ahmed, A., Li, J., Shihba, Y., Robbins, J. B., and Sui, S. C. (2006). Safety and immunogenicity of *Escherichia coli* O157:O-specific polysaccharide conjugate vaccine in 2-5-year-old children. *J. Infect. Dis.* 193, 515–521. doi: 10.1086/499821

Aslani, M. M., Alikhani, M. Y., Zavari, A., Yousefizadeh, M., Bitzan, M., Poole, R., Mehran, M., Sicard, E., Brockus, C., Thuning-Roberson, C., Biran, D., and Ron, E. Z. (2018). Extraintestinal Pathogenic *Escherichia coli* ST131 O25:H4/H30-Rx virotypes. *Braz J. Infect. Dis.* 22 (7-8), 625–630. doi: 10.1016/j.bjid.2018.02.001

Carter, C. D., Parks, A., Abuladze, T., Li, M., Woolston, J., Magrone, J., et al. (2012). Bacteriophage cocktail significantly reduces *Escherichia coli* O157:H7 contamination of lettuce and beef, but does not protect against recontamination. *Bacteriophage* 2 (3), 178–185. doi: 10.4161/bact.22825

### AUTHOR CONTRIBUTIONS

VB: writing and organization. KM: writing and mentoring. All authors contributed to the article and approved the submitted version.

### FUNDING

Financially supported by FAPESP (grants 2014/06779-2, 2018/22412-2, 2018/22042-0, and 2019/03049-7), CNPq (307418/2017-0), and “Programa de Apoio ao Desenvolvimento Científico da Faculdade de Ciências Farmacêuticas da UNESP-PADIC). This study was financed in part by the Coordenação de Aperfeiçoamento de Pessoal de Nível Superior - Brasil (CAPES) - Finance Code 001.
Han, Z., Pinkner, J. S., Ford, B., Chorrell, E., Crowley, J. M., Cusumano, C. K., et al. (2012). Lead optimization studies on FimH antagonists: discovery of potent and orally bioavailable non-substituted biphenyl mannosides. J. Med. Chem. 55, 3945–3959. doi: 10.1021/jm300165m

Hazen, T. H., Michalski, J., Luo, Q., Shetty, A. C., Daugherty, S. C., Fleckenstein, J. M., et al. (2017). Comparative genomics and transcriptomics of Escherichia coli isolates carrying virulence factors of both enteropathogenic and enterotoxigenic E. coli. Sci. Rep. 7, 3513. doi: 10.1038/s41598-017-03489-z

Ingle, D. J., Levine, M. K., Klotz, K. L., Holt, K. E., and Robins-Browne, R. M. (2018). Dynamics of antimicrobial resistance in intestinal Escherichia coli from children in community settings in South Asia and sub-Saharan Africa. Nat. Microbiol. 3 (9), 1063–1073. doi: 10.1038/s41564-018-0217-4

Jackson, R. W., Vinatzer, B., Arnold, D. L., Dorus, S., and Murillo, J. (2011). The influence of the accessory genome on bacterial pathogen evolution. Mol. Genet. Genom. 1 (1), 55–65. doi: 10.1007/s00439-011-1643z

Jargas, C., Han, Z., Kalas, V., Klein, R., Pinkner, J. S., Ford, B., et al. (2016). Antivirulence Isouquinolone Mannosides: Optimization of the Biaryl Aglycone for FimH Lectin Binding Affinity and Efficacy in the Treatment of Chronic UTI. ChemMedChem 11 (4), 367–373. doi: 10.1002/cmdc.201600006

Jerse, A. E., Yu, J., Tall, B. D., and Kaper, J. B. (1990). A genetic locus of enteropathogenic Escherichia coli necessary for the production of attaching and effacing lesions on tissue culture cells. Proc. Natl. Acad. Sci. U.S.A. 87 (20), 7839–7843. doi: 10.1073/pnas.87.20.7839

Johnson, K. J., Bhat, S., and Abernethy, R. B. (2017). Small Molecules That Sabotage Bacterial Virulence. Trends Pharmacol. Sci. 38 (4), 339–362. doi: 10.1016/j.tips.2017.01.004

Johnson, J. R., and Russo, T. A. (2018). Molecular Epidemiology of Extrainestinal Pathogenic Escherichia coli. EcoSal Plus 8 (1), 4–22. doi: 10.1128/ecosalplus.ESP-0004-2017

Kaper, J. B., Nataro, J. P., and Mobley, H. L. (2004). Pathogenic Escherichia coli. Nat. Rev. Microbiol. 2 (12), 123–140. doi: 10.1038/nrmicro818

Khan, A., Miller, W. R., and Arias, C. A. (2018). Mechanisms of antimicrobial resistance among hospital-associated pathogens. Expert Rev. Anti. Infect. Ther. 16 (4), 269–287. doi: 10.1080/14787964.2018.1456919

Kim, C. S., Gatsios, A., Cuesta, S., Lam, Y. C., Wei, Z., Chen, H., et al. (2020). Characterization of Autoinducer-3 Structure and Biosynthesis in E. coli. ACS Cent. Sci. 6 (2), 197–206. doi: 10.1021/acscentsci.9b01076

Knutton, S., Baldwin, T., Williams, P. H., and McNeish, A. S. (1989). Actin accumulation at sites of bacterial adhesion to tissue culture cells: basis of a new diagnostic test for enteropathogenic and enterohemorrhagic Escherichia coli. Infect. Immun. 57 (4), 1290–1298. doi: 10.1128/IAI.57.4.1290-1298.1989

Köhler, C. D., and Dobrindt, U. (2011). What defines extraintestinal pathogenic Escherichia coli? Int. J. Med. Microbiol. 301 (8), 642–647. doi: 10.1016/j.ijmm.2011.09.006

Kostal, T. K., Valtonen, M. V., Parkkinen, J., Väisänen-Rhen, V., Finne, J., Orskov, F., et al. (1985). Serotypes, hemolysin production, and receptor recognition of Escherichia coli strains associated with neonatal sepsis and meningitis. Infect. Immun. 48 (2), 486–491. doi: 10.1128/IAI.48.2.486-491.1985

Lindstedt, B. A., Finton, M. D., Porcellato, D., and Brandal, L. T. (2018). High frequency of hybrid Escherichia coli strains with combined Intestinal Pathogenic Escherichia coli(IPEC) and Extrainestinal Pathogenic Escherichia coli (ExPEC) virulence factors isolated from human faecal samples. BMC Infect. Dis. 18, 544. doi: 10.1186/s12879-018-3489-2

Liu, J., Sun, Y., Feng, S., Zhu, L., Guo, X., and Qi, C. (2009). Towards an attenuated enterohemorrhagic Escherichia coli O157:H7 vaccine characterized by a deleted ler gene and containing apathogenic Shiga toxins. Vaccine 27, 5929–5935. doi: 10.1016/j.vaccine.2009.07.097

Logue, C. M., Doekott, C., Mangiamele, P., Wannemuehler, Y. M., Johnson, T. J., Tivendale, K. A., et al. (2012). Genotypic and phenotypic traits that distinguish neonatal meningitis-associated Escherichia coli from fecal E. coli isolates of healthy human hosts. Appl. Environ. Microbiol. 78 (16), 5824–5830. doi: 10.1128/AEM.07869-11

Lustri, B. C., Sperandio, V., and Moreira, C. C. (2017). bacterial chat: intestinal metabolites and signals in host-microbiota-pathogen interactions. Infect. Immun. 85 (12), e00476-17. doi: 10.1128/IAI.00476-17

Malby, R., Leatham-Jensen, M. P., Gibson, T., Cohen, P. S., and Conway, T. (2013). Nutritional basis for colonization resistance by human commensal
Escherichia coli strains HS and Nissle 1917 against E. coli O157:H7 in the mouse intestine. PloS One 11, e0155911. doi: 10.1371/journal.pone.0155911
Meling, S. D., McGowan, A. S., Sportman, A. C., Qi, W., Lach, D. W., Ouazette, L. M., et al. (2008). Variation in virulence among clades of Escherichia coli O157:H7 associated with disease outbreaks. Proc. Natl. Acad. Sci. U.S.A. 105 (12), 4868–4873. doi: 10.1073/pnas.0710834105
Mariani-Kurkdjian, P., Lemaitre, C., Bidet, P., Perez, D., Boggini, L., Kwon, T., et al. (2014). Haemolytic-uraemic syndrome with bacteremia caused by a new hybrid Escherichia coli pathotype. New Microbes New Infect. 2, 127–131. doi: 10.1016/j.nmni.2014.01.003
Medina, A. M., Rivera, F. P., Pons, M. J., Riveros, M., Gomes, C., Bernal, M., et al. (2015). Comparative analysis of antimicrobial resistance in enterotoxigenic Escherichia coli strains from two paediatric cohort studies in Lima, Peru. Trans. R. Soc Trop. Med. Hyg. 109 (6), 493–502. doi: 10.1093/trstmh/trv054
Mellies, J., Elliott, S. J., Sperandio, V., Veiga, P., Reeves, A. Z., Lavoie, S., et al. (1999). The Per reguol of enteropathogenic Escherichia coli: identification of a regulatory cascade and a novel transcriptional activator, the locus of enterocyte effacement (LEE)-encoded regulator (Ler). Mol. Microbiol. 33 (2), 296–306. doi: 10.1111/j.1365-2958.1999.01473.x
Mobley, H., Donnenberg, M., and Hagan, E. (2009). Pathogenic Enterobacteriaceae: the language of hormones. Int. J. Med. Microbiol. 295 (6–7), 455–462. doi: 10.1016/j.ijmm.2005.07.007
Muniesa, M., Hammerl, J. A., Hertwig, S., Appel, B., and Brüssow, H. (2012). Shiga toxin-producing Escherichia coli O104:H4: a new challenge for microbiology. Appl. Environ. Microbiol. 78 (12), 4065–4073. doi: 10.1128/AEM.00217-12
Nagarjuna, D., Mittal, G., Dhanda, R. S., Gaind, R., and Yadav, M. (2018). Alarming levels of antimicrobial resistance among sepsis patients admitted to ICU in a tertiary care hospital in India – a case control retrospective study. Antimicrob. Resist. Infect. Control 7, 150. doi: 10.1186/s13756-018-0444-8
Nash, J. H., Villegas, A., Kropinski, A. M., Aguilar-Velazuela, R., Konczy, P., Masccarenhas, M., et al. (2010). Genome sequence of adherent-invasive Escherichia coli and comparative genomic analysis with other E. coli pathotypes. BMC Genomics 11, 667. doi: 10.1186/1471-2164-11-667
Navarro-Alfonsin, S. C., Barnhart-Kaufman, A. L., Gómez-Plata, X., and Hori, S. (2014). Escherichia coli O104:H4 Pathogenesis: an EnterogeeogenicEcoli/Shiga Toxin-Producing E. coli Explosive Cocktail of HighVirulence. Microbiol. Spectr. 2 (6), 2–15. doi: 10.1128/microbiolspec.EHEC-0008-2013
Nesta, B., and Piazza, M. (2018). "Vaccines against Escherichia coli." In Escherichia coli, a Versatile Pathogen (Cham: Springer), 213–242. doi: 10.1007/82_2018_111
Nyholm, O., Halka-Laiti, J., Wiliund, G., Okeke, U., Paulin, L., Auvinen, P., et al. (2015). Comparative genomics and characterization of hybrid Shigatoxigenic and Enterotoxigenic Escherichia coli (STEC/ETEC) strains. PloS One 10, e0135936. doi: 10.1371/journal.pone.0135936
Ogura, Y., Oku, T., Iguchi, A., Toh, H., Asadulghani, M., Oshima, K., et al. (2009). Comparative genomics reveal the mechanism of the parallel evolution of O157 and non-O157 enterohemorrhagic Escherichia coli. Proc. Natl. Acad. Sci. U.S.A. 106 (42), 17939–17944. doi: 10.1073/pnas.0903585106
Ojala, V., Laitalaainen, J., and Jalasvuori, M. (2013). Fight evolution with evolution: plasmid-dependent phages with a wide host range prevent the spread of antibiotic resistance. Evol. Appl. 6, 925–932. doi: 10.1111/eva.12076
Okeke, I. N., Steinmückl, H., Kanack, K. J., Elliott, S. J., Sundström, L., Kaper, J. B., et al. (2002). Antibiotic-resistant cell-detaching Escherichia coli strains from Nigerian children. J. Clin. Microbiol. 40 (1), 301–305. doi: 10.1128/JCM.40.1.301-305.2002
Oliveira, A. F., Cardoso, S. A., Almeida, F. B., de Oliveira, L. L., Pitoondo-Silva, A., Soares, S. G., et al. (2012). Oral immunization with attenuated Salmonella
Tagliaferri, T. L., Mathias, J., and Hans-Peter, H. (2019). Fighting pathogenic bacteria on two fronts: phages and antibiotics as combined strategy. *Front. Cel. Infect. Microb.* 9, 22. doi: 10.3389/fcimb.2019.00022

Tobe, T., Hayashi, T., Han, C. G., Schoolnik, G. K., Ohitsu, E., and Sasakawa, C. (1999). Complete DNA sequence and structural analysis of the enteropathogenic *Escherichia coli* adherence factor plasmid. *Infect. Immun.* 67 (10), 5455–5462. doi: 10.1128/IAI.67.10.5455-5462.1999

Touchon, M., Hoede, C., Tenaillon, O., Barbe, V., Baeriswyl, S., Bidet, P., et al. (2009). Organised genome dynamics in the *Escherichia coli* species results in highly diverse adaptive paths. *PloS Genet.* 5 (1), e1000344. doi: 10.1371/journal.pgen.1000344

Trabulsi, L. R., Keller, R., and Tardelli Gomes, T. A. (2002). Typical and atypical enteropathogenic *Escherichia coli*. *Emerg. Infect. Dis.* 8 (5), 508–513. doi: 10.3201/eid0805.010385

van den Dobbelsteen, G., Fae, K. C., Serroyen, J., van den Nieuwenhof, I. M., Braun, M., Haeuptle, M. A., et al. (2016). Immunogenicity and safety of a tetravalent *E. coli* O-antigen bioconjugate vaccine in animal models. *Vaccine* 34, 4152e60. doi: 10.1016/j.vaccine.2016.06.067

World Health Organization. (2018). *Global antimicrobial resistance surveillance system (GLASS) report. Early implementanion 2016-2017*, ISBN: .

Xue, X. Y., Mao, X. G., Li, Z., Chen, Z., Zhou, Y., Hou, Z., et al. (2015). A potent and selective antimicrobial poly(amideamine) dendrimer conjugate with LED209 targeting QseC receptor to inhibit the virulence genes of gram-negative bacteria. *Nanomedicine* 11 (2), 329–339. doi: 10.1016/j.nano.2014.09.016

Yang, Q., Anh, N. D., Bossier, P., and Defoirdt, T. (2014). Norepinephrine and dopamine increase motility, biofilm formation, and virulence of *Vibrio harveyi*. *Front. Microbiol.* 5:584. doi: 10.3389/fmicb.2014.00584

Zhang, X. H., He, K. W., Zhang, S. X., Lu, W. C., Zhao, P. D., Luan, X. T., et al. (2011). Subcutaneous and intranasal immunization with Stx2B-Tir-Stx1B-Zot reduces colonization and shedding of *Escherichia coli* O157:H7 in mice. *Vaccine* 29, 3923–3929. doi: 10.1016/j.vaccine.2011.02.007

Zheng, B., Dong, H., Xu, H., Lv, J., Zhang, J., Jiang, X., et al. (2016). Coexistence of MCR-1 and NDM-1 in Clinical *Escherichia coli* Isolates. *Clin. Infect. Dis.* 63 (10), 1393–1395. doi: 10.1093/cid/ciw553

Zhong, L. L., Zhang, Y. F., Doi, Y., Huang, X., Zhang, X. F., Zeng, K. J., et al. (2017). Coproduction of MCR-1 and NDM-1 by Colistin-Resistant *Escherichia coli* Isolated from a Healthy Individual. *Antimicrob. Agents Chemother.* 61 (1), e01962–e01916. doi: 10.1128/AAC.01962-16

**Conflict of Interest:** The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Copyright © 2020 Braz, Melchior and Moreira. This is an open-access article distributed under the terms of the Creative Commons Attribution License (CC BY). The use, distribution or reproduction in other forums is permitted, provided the original author(s) and the copyright owner(s) are credited and that the original publication in this journal is cited, in accordance with accepted academic practice. No use, distribution or reproduction is permitted which does not comply with these terms.