

Escherichia coli produttori di Verocototossina: Un microorganismo patogeno in continua evoluzione

Stefano Morabito

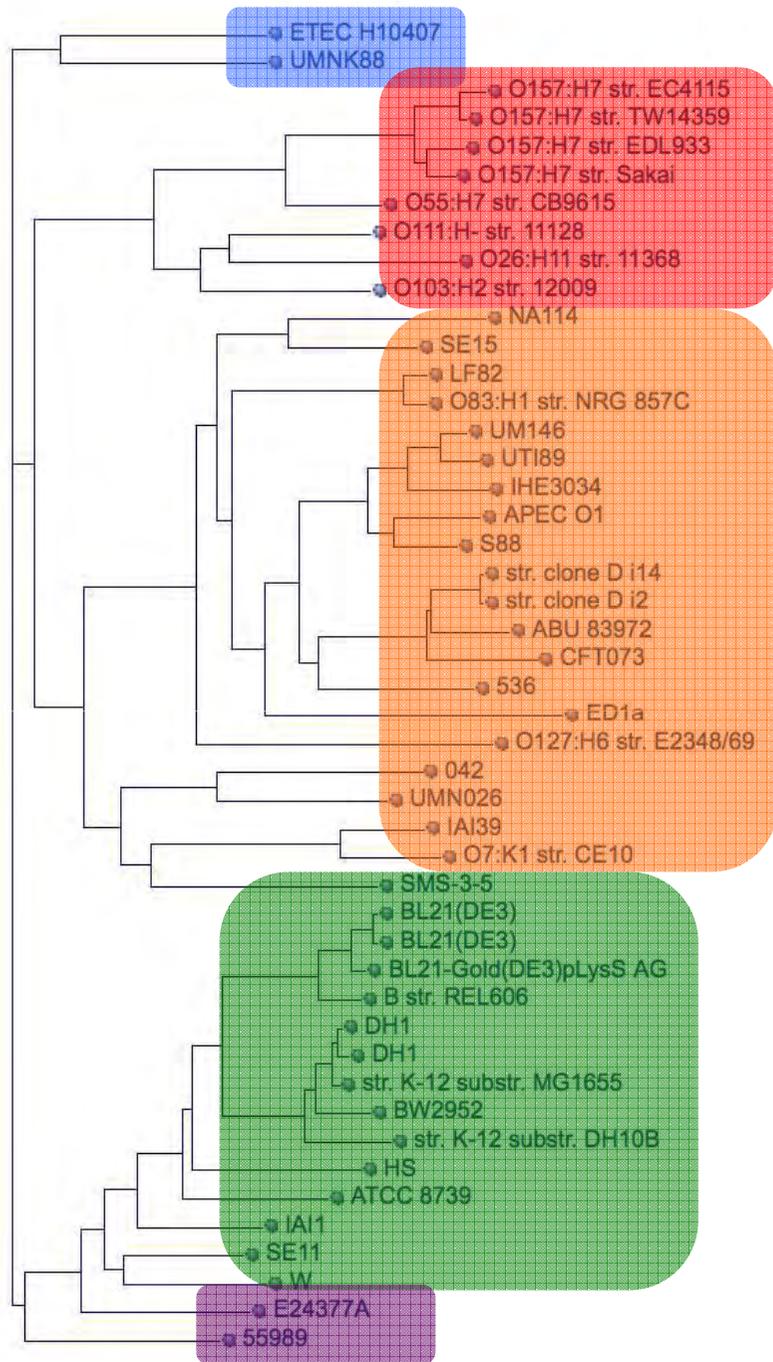
EU Reference Laboratory for E. coli

Dipartimento di Sanità Pubblica Veterinaria e Sicurezza Alimentare

Istituto Superiore di Sanità

Roma, Italia





ETEC

VTEC

EIEC
ExPEC

K/12
Commensali

EAEC

http://www.ncbi.nlm.nih.gov/sites/entrez?db=genome&cmd=Retrieve&dopt=Overview&list_uids=167



Verocytotoxin-producing *E. coli*

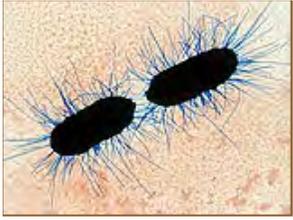
Tra i più temibili patogeni alimentari!



Manifestazione clinica: Molto grave

Dose infettante Molto bassa (10-100 CFU !)

Epidemie in comunità Di grandi dimensioni



VTEC: Manifestazione Clinica

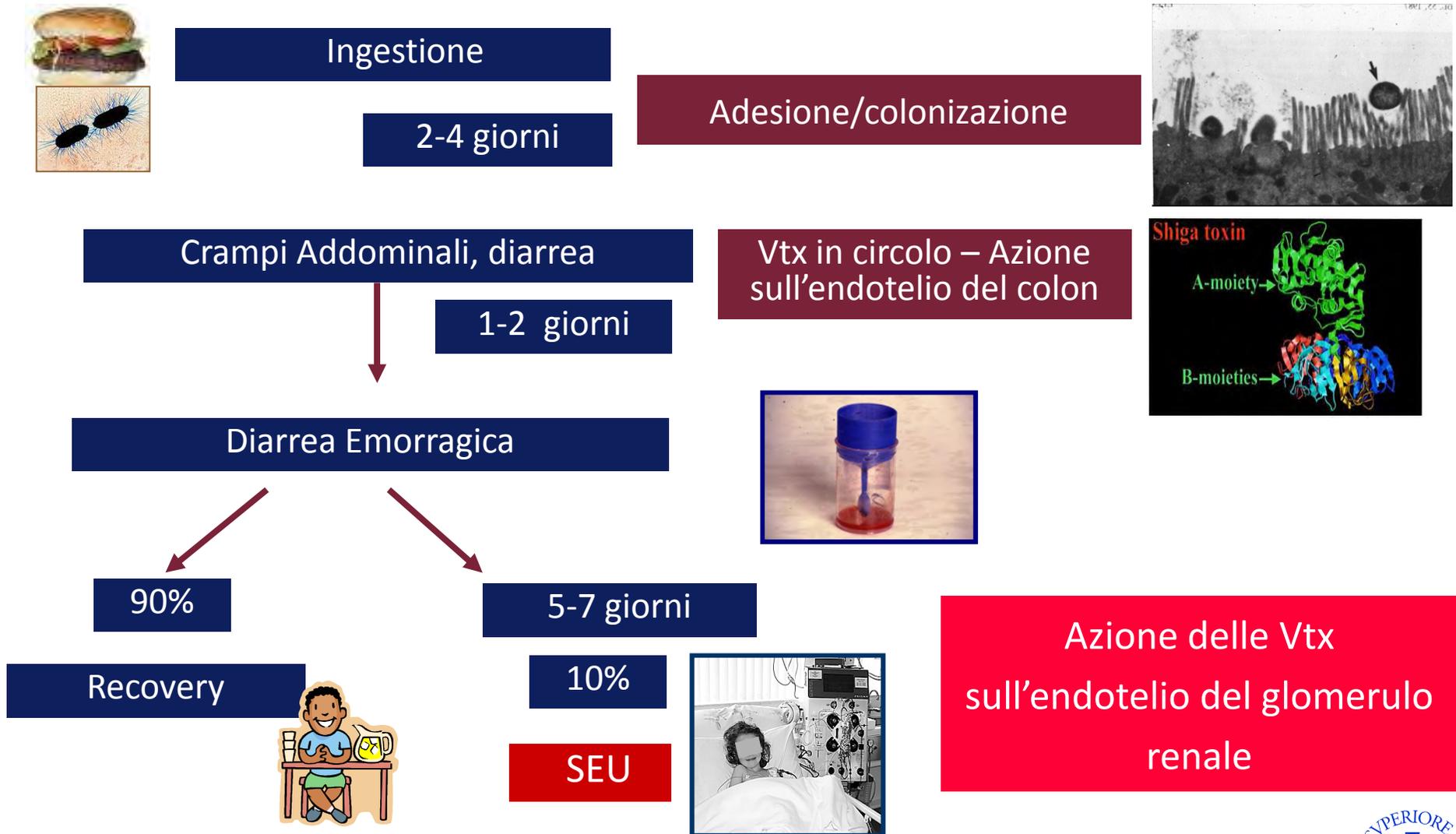
- **Intestinale**
 - Asintomatica
 - Diarrea Acquosa
 - Colite Emorragica

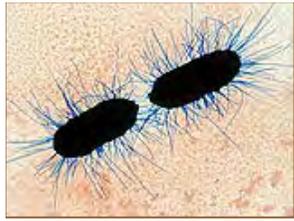


- **Sistemica**
 - Hemolytic Uremic Syndrome (HUS)
 - Interessamento Neurologico



Patogenesi delle infezioni da VTEC





Sindrome Emolitico Uremica (SEU)

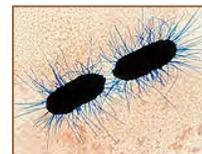
- Anemia Emolitica
- Trombocitopenia
- Insufficienza renale acuta



- **Principale causa di insufficienza renale nei bambini. Richiede dialisi**
- **Trattamento con antibiotici non è efficace ed è controindicato**

VTEC sono patogeni zoonotici

Alimenti di origine animale: Contaminazione Primaria



VTEC sono patogeni zoonotici

Contaminazione dell'ambiente



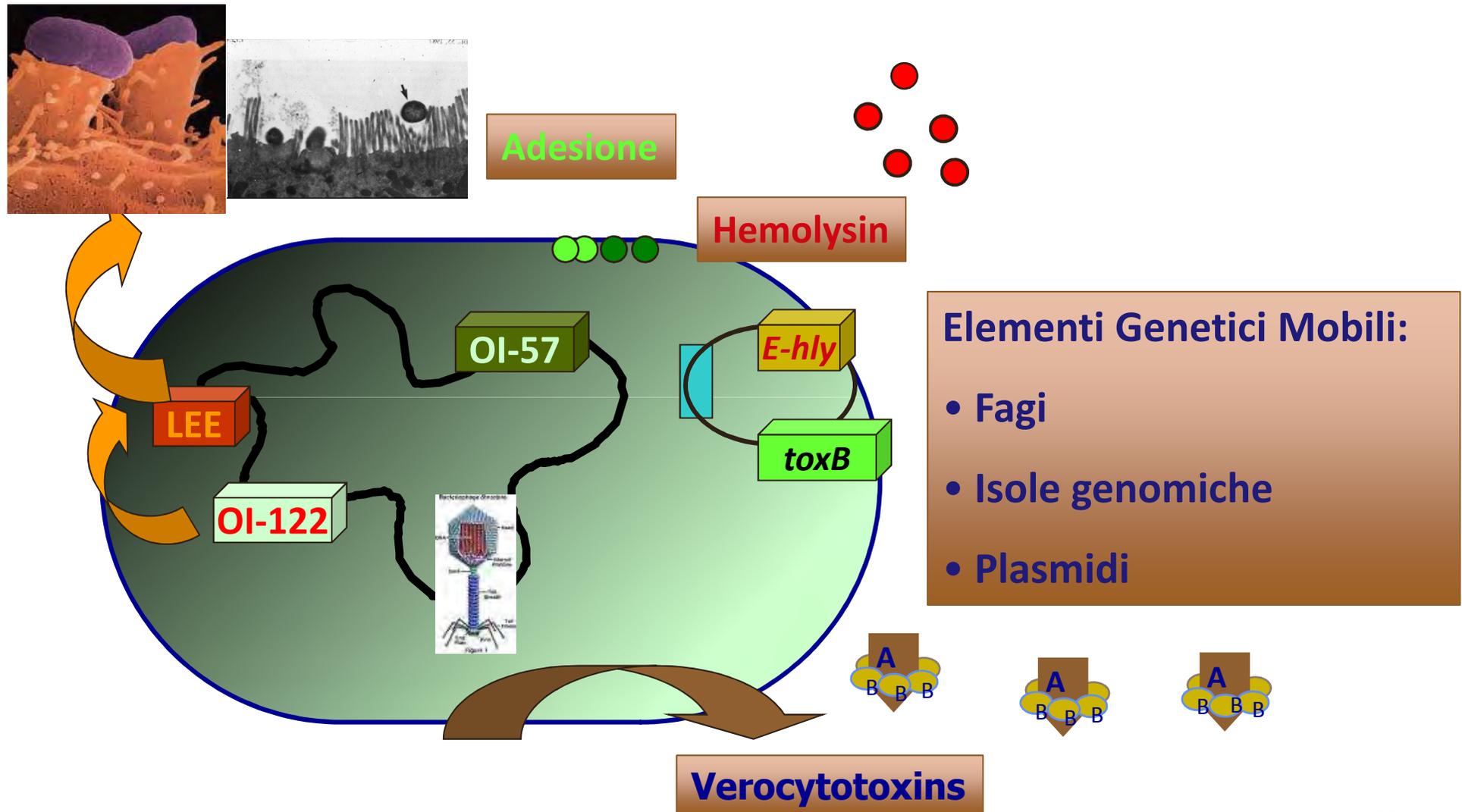
Suolo



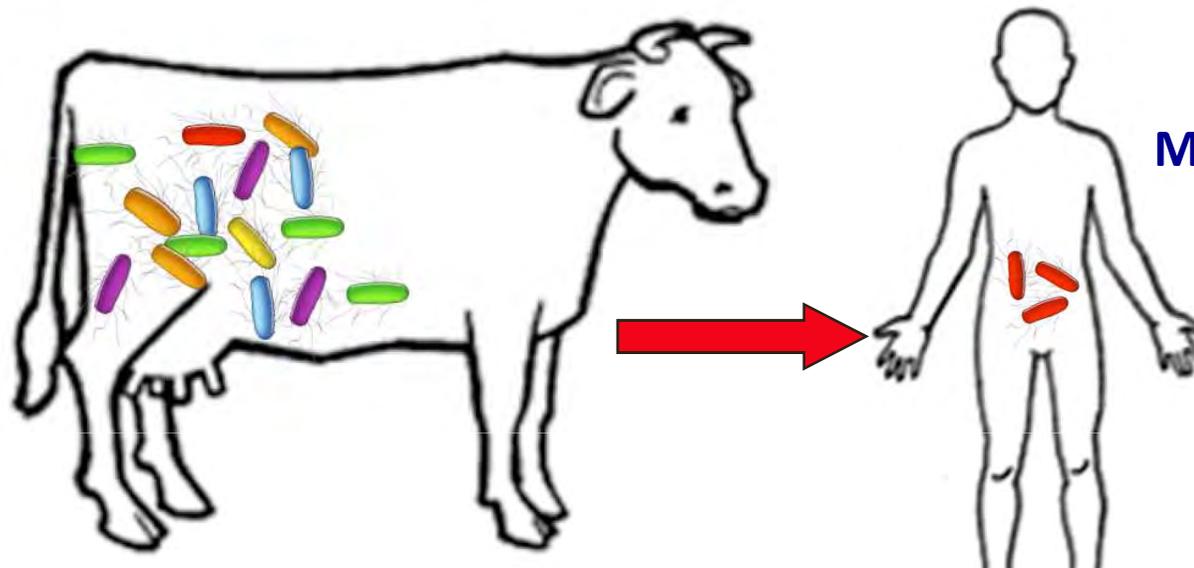
Acque di superficie



VTEC Complessità Genomica



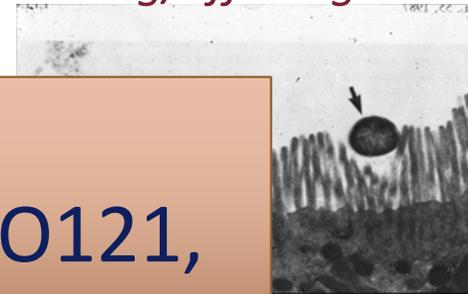
VTEC=patogeno?



VTEC

ASSOCIATI CON LE FORME
MORBOSE PIU' GRAVI (DE, SEU)

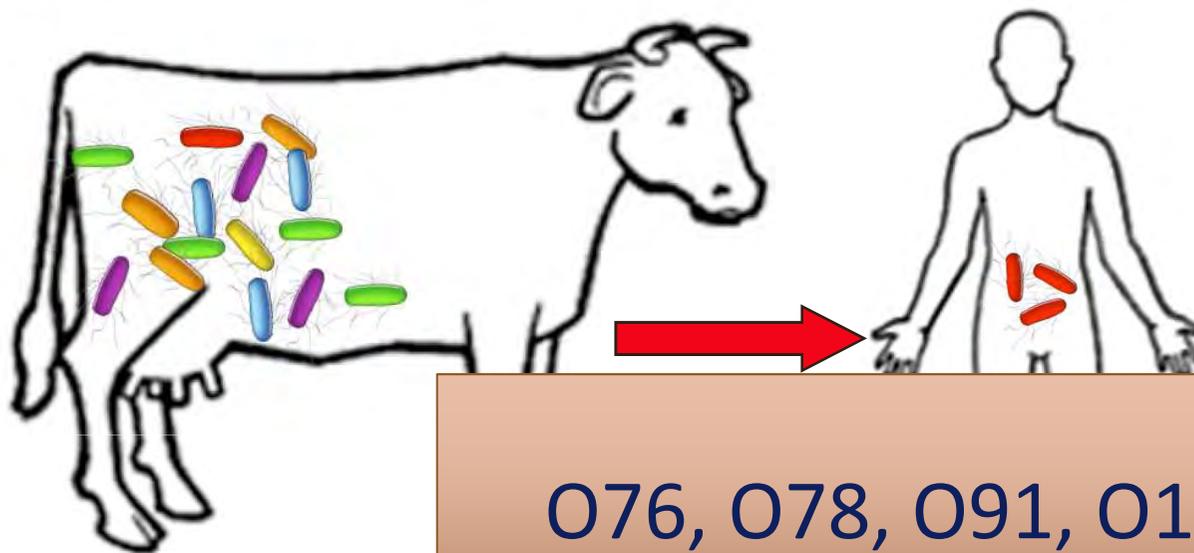
Fattori di Virulenza Addizionali
Attaching/Effacing adhesion



>100 sierotipi
Nel serbatoio
nell'ambiente

0157
026, 045, 0111, 0103, 0121,
0145

VTEC=patogeno?



**ASSOCIATI CON FORME
MORBOSE ANCHE GRAVI**

**O76, O78, O91, O113, O117,
O146, O174,**

Adizionali in
cus LEE
Subtilasi

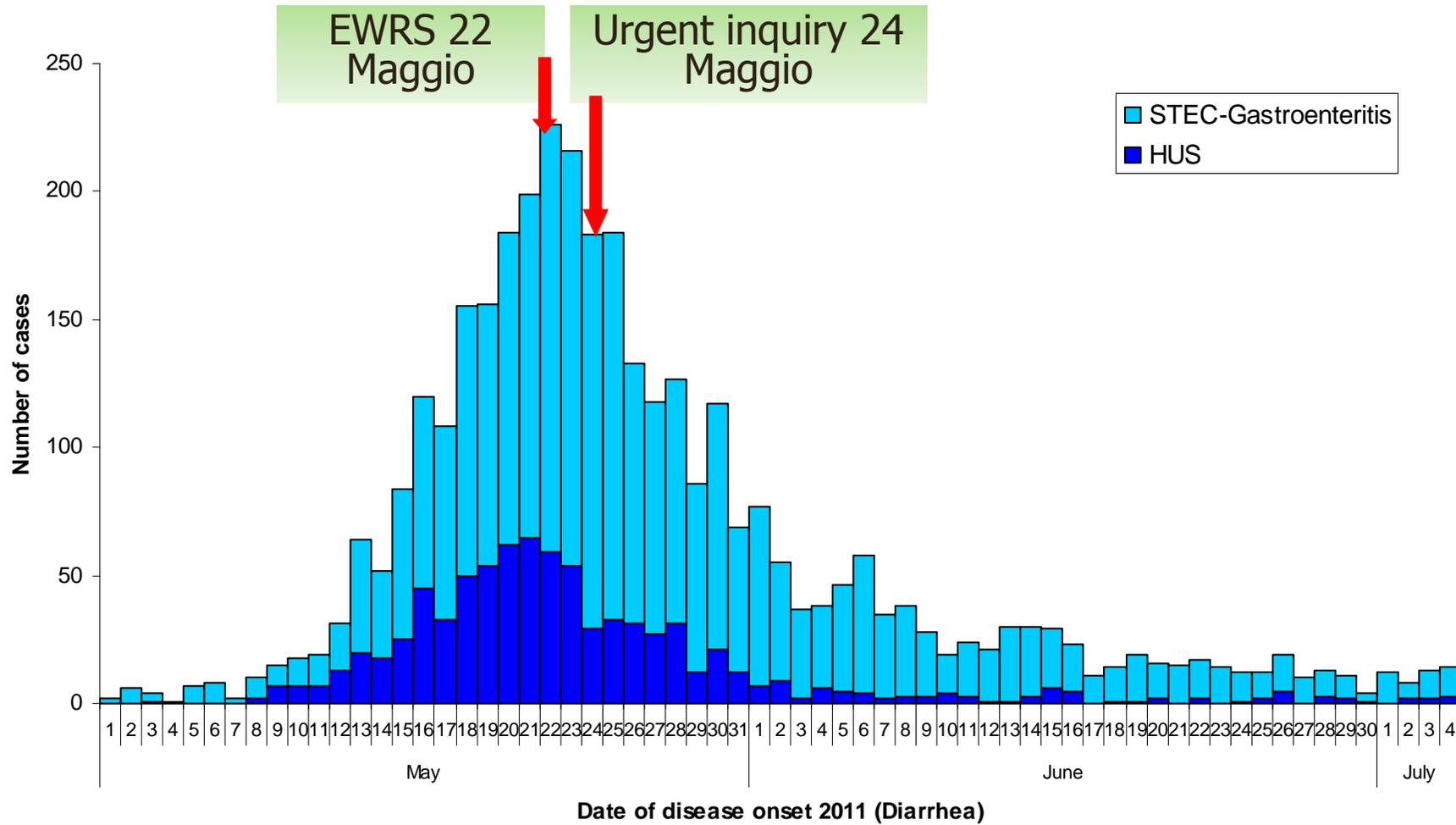


VTEC: Nomenclatura

- **VTEC:** *E. coli* che produce Vtx
- **STEC:** *E. coli* che produce Vtx
- **O157:** Sierograppo di *E. coli*. A volte produce Vtx
- **EHEC:** *E. coli* enteroemorraggici (LEE-positivi)
- **“Top five serogroups”:** Serogruppi VTEC associati a casi di SEU (O26, O111, O103, O145+O157)
- **HAS:** HUS-associated VTEC
- **Seropathotypes A-E:** gruppi di VTEC che causano malattia grave o leggera o presenti nel solo serbatoio animale.

VTEC O104:H4 Germania 2011

Source: Robert Koch Institute. Report: Final presentation and evaluation of epidemiological findings in the EHEC O104:H4 outbreak, Germany 2011. Berlin 2011.

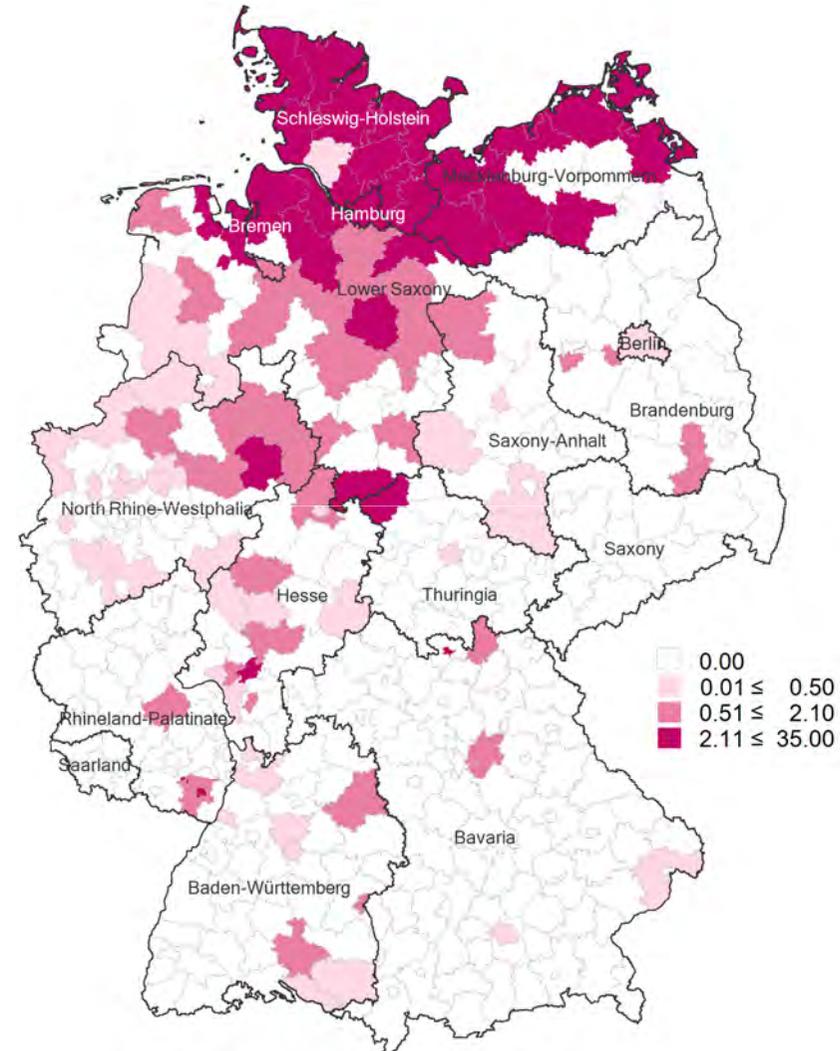


Incidenza di SEU durante l'epidemia

ROBERT KOCH INSTITUT



Casi/100,000



Da: Robert Koch Institute. Report: Final presentation and evaluation of epidemiological findings in the EHEC O104:H4 outbreak, Germany 2011. Berlin 2011.

VTEC O104:H4 Germania 2011

3,842 casi

2987
non-SEU

855
SEU

18 Decessi
(0.6%)

35 Decessi
(4.1%)

SEU

- 68% Donne

- **Mediana età 42 anni (0-91 anni)**

- Diarrea emorragica 79%

Emergenza di un nuovo tipo di VTEC

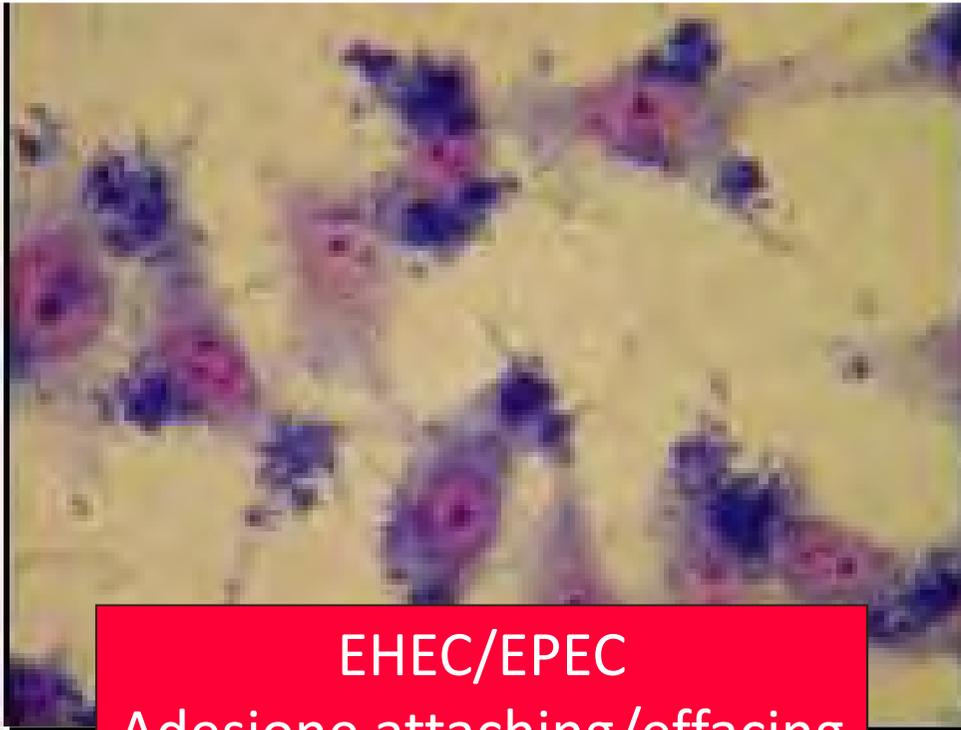
E-ALERT

Characteristics of the enteroaggregative Shiga toxin/ verotoxin-producing *Escherichia coli* O104:H4 strain causing the outbreak of haemolytic uraemic syndrome in Germany, May to June 2011

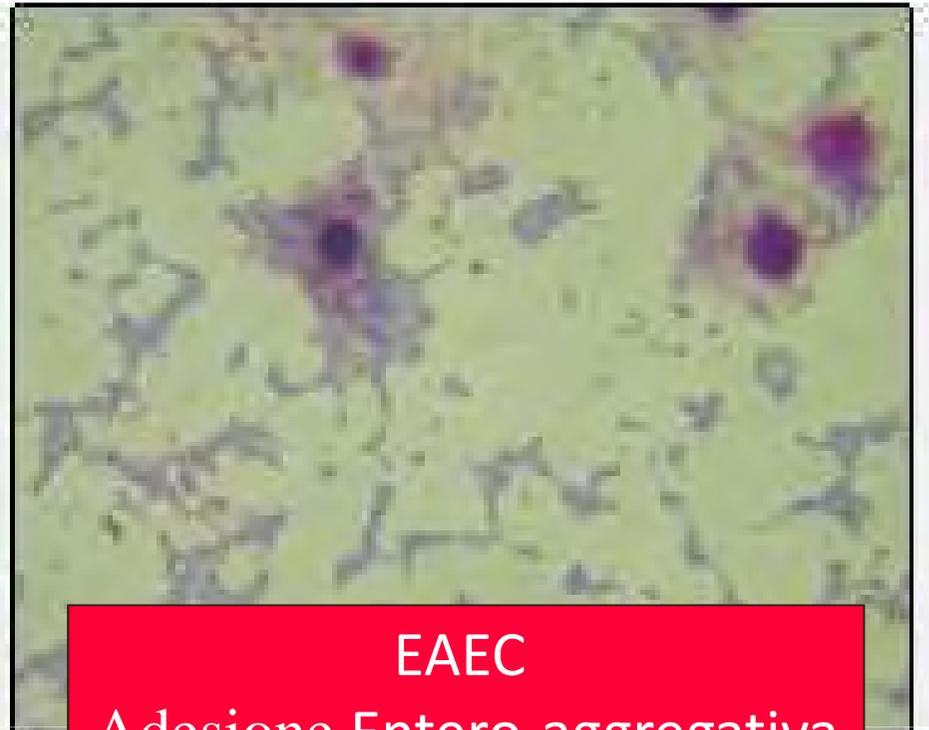
F Scheutz (fsc@ssi.dk)^{1,2}, E Møller Nielsen², J Frimodt-Møller^{1,3}, N Boisen^{1,2}, S Morabito⁴, R Tozzoli⁴, J P Nataro⁵, A Caprioli⁴

1. World Health Organization Collaborating Centre for Reference and Research on *Escherichia* and *Klebsiella*, Department of Microbiological Surveillance and Research, Copenhagen, Denmark
2. Food-borne pathogens and typing, Department of Microbiological Surveillance and Research, Statens Serum Institut, Copenhagen, Denmark
3. Department of Clinical Microbiology, Hillerød Sygehus, Hillerød, Denmark
4. European Union Reference Laboratory for *Escherichia coli*, Department of veterinary public health and food safety, Istituto Superiore di Sanità, Rome, Italy
5. University of Virginia School of Medicine, Charlottesville, United States





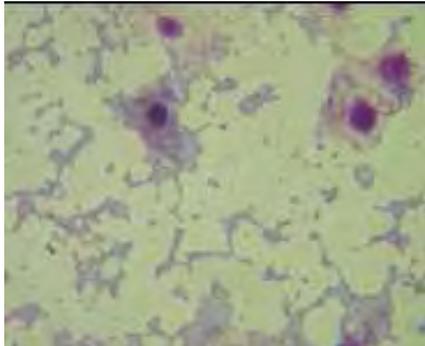
EHEC/EPEC
Adesione attaching/effacing



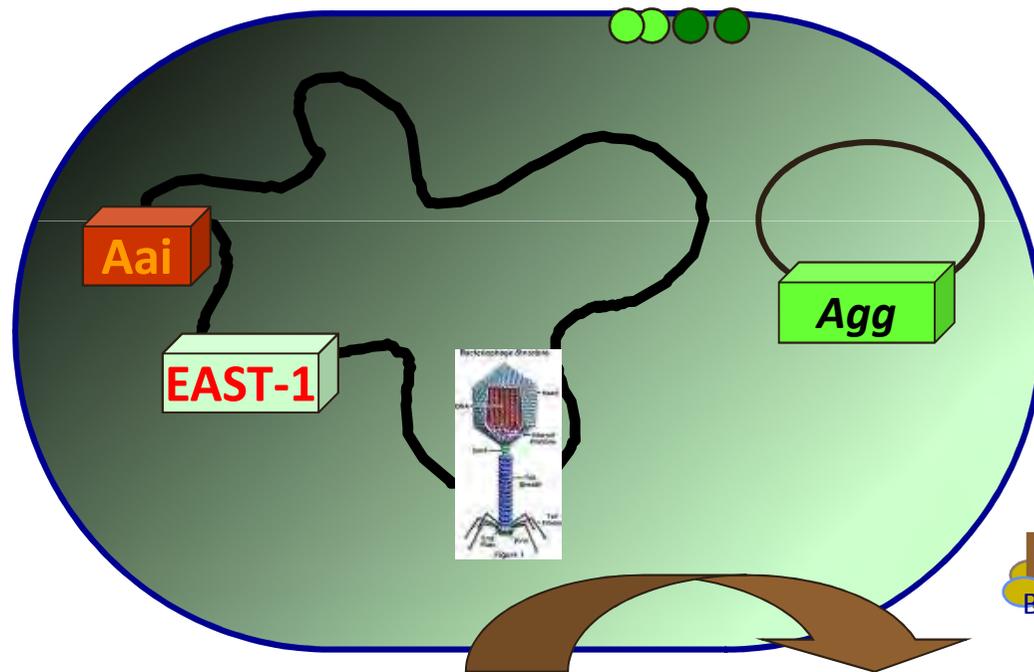
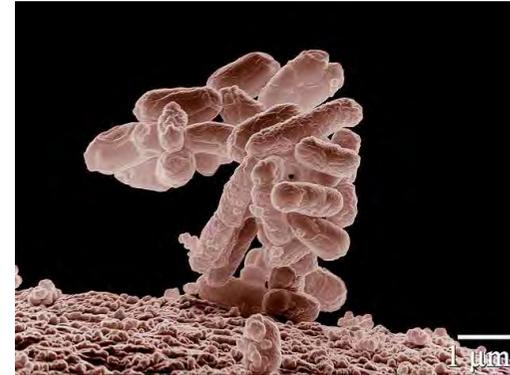
EAEC
Adesione Entero-aggregativa



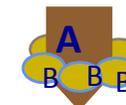
Vtx-producing EAEC



Adesione „Stacked brick“



E. coli con genoma EAEC ha acquisito un Fago-Stx



Shigatoxin-

Vtx-producing EAEC

JOURNAL OF CLINICAL MICROBIOLOGY, Mar. 1998, p. 840-842
 0095-1137/98/\$04.00+0
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Vol. 36, No. 3

Enteroaggregative, Shiga Toxin-Producing *Escherichia coli* O111:H2 Associated with an Outbreak of Hemolytic-Uremic Syndrome

STEFANO MORABITO,¹ HELGE KARCH,² PATRIZIA MARIANI-KURKDJIAN,³ HERBERT SCHMIDT,² FABIO MINELLI,¹ EDOUARD BINGEN,³ AND ALFREDO CAPRIOLI^{1*}

FEMS Microbiol Lett, 2000 Oct 1;191(1):7-10.

Inducible stx2 phages are lysogenized in the enteroaggregative and other phenotypic *Escherichia coli* O86:HNM isolated from patients.

Iyoda S¹, Tamura K, Itoh K, Izumiva H, Ueno N, Nagata K, Togo M, Terajima J, Watanabe H.

Emerg Infect Dis. 2011 Oct;17(10):1957-8. doi: 10.3201/eid1710.111072.

Similarity of Shiga toxin-producing *Escherichia coli* O104:H4 strains from Italy and Germany.

Scavia G, Morabito S, Tozzoli R, Michelacci V, Marziano ML, Minelli F, Ferreri C, Paolialonga F, Edefonti A, Caprioli A.

J Clin Microbiol. 2012 Dec;50(12):4116-9. doi: 10.1128/JCM.02047-12. Epub 2012 Oct 3.

Characterization of a verocytotoxin-producing enteroaggregative *Escherichia coli* serogroup O111:H21 strain associated with a household outbreak in Northern Ireland.

Dallman T, Smith GP, O'Brien B, Chattaway MA, Finlay D, Grant KA, Jenkins C.

Laboratory of Gastrointestinal Pathogens, Health Protection Agency, London, United Kingdom.

Questi VTEC sono in fase di stabilizzazione

EAEC O111:H10
8 HUS

1992
France

EAEC O86:NM
1 HUS

2000
Japan

EAEC O104:H4
1 HUS

2001
Germany

EAEC O104:H4
1 HUS

2009
Italy

EAEC O104:H4
1 HUS

2010
Finland

EAEC O104:H4
850 HUS

2011
Germany, France

EAEC O111:H21
1 HUS

2012
Northern Ireland

EAEC O127:H4
4 HUS

2013
Italy



EAEC sono patogeni a trasmissione interumana

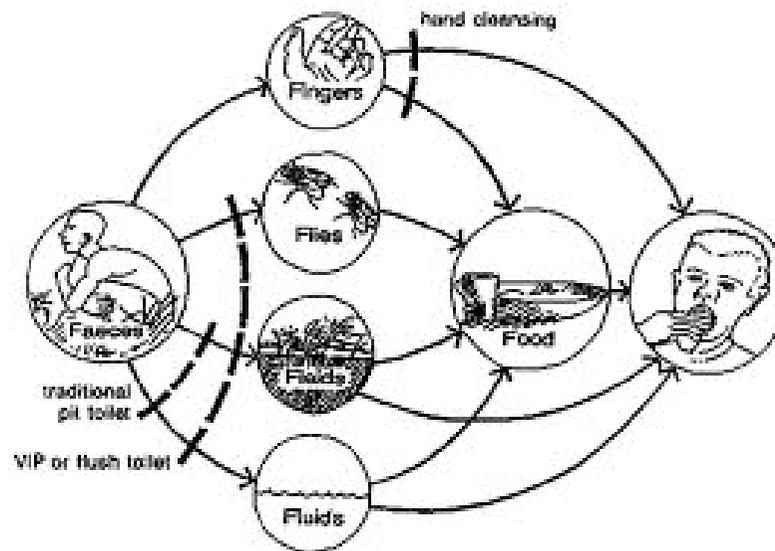
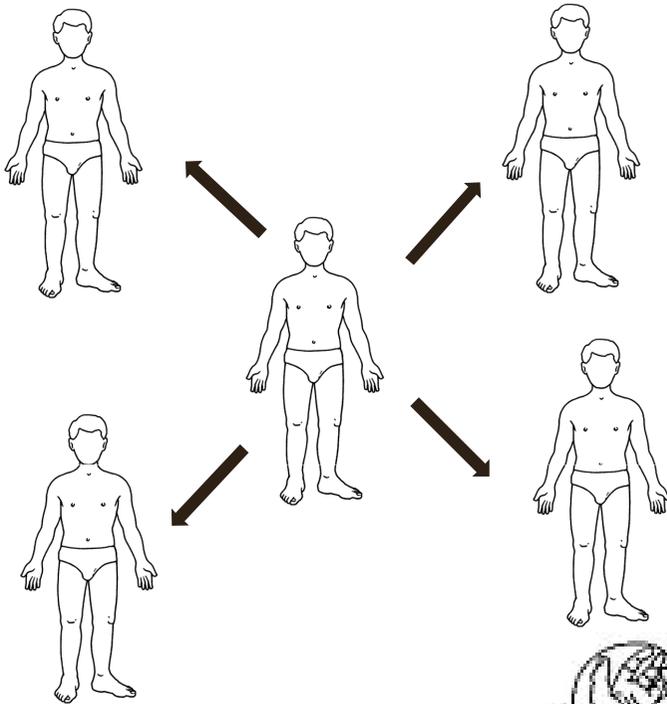






Fig. 1.1. A Monster Soup Commonly Called Thames Water, by William Heath (1795–1840), circa 1828. Philadelphia Museum of Art: Gift of Mrs. William Horstmann. Reproduced with permission.

Da: Nutrition and Health in Developing Countries. Richard D. Semba and martin W. Bloem Eds.
Humana Press. 2008





Taken from: [Hungry Planet: What the World Eats](#) by [Peter Menzel](#) and [Faith D'Aluisio](#) (Sep 1, 2007)

VTEC O104 entra in EU con semi di Fieno Greco



Epidemia in Germania

Assenza di conferma Microbiologica
ma forte evidenza epidemiologica

16 casi in una scuola in Francia



Tracing Back exercise
EFSA



Il mercato degli alimenti cambia

Trends of food Innovation In E

Trends

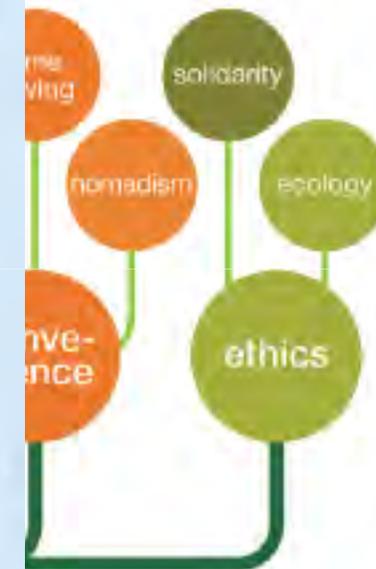
Axis

Consumer



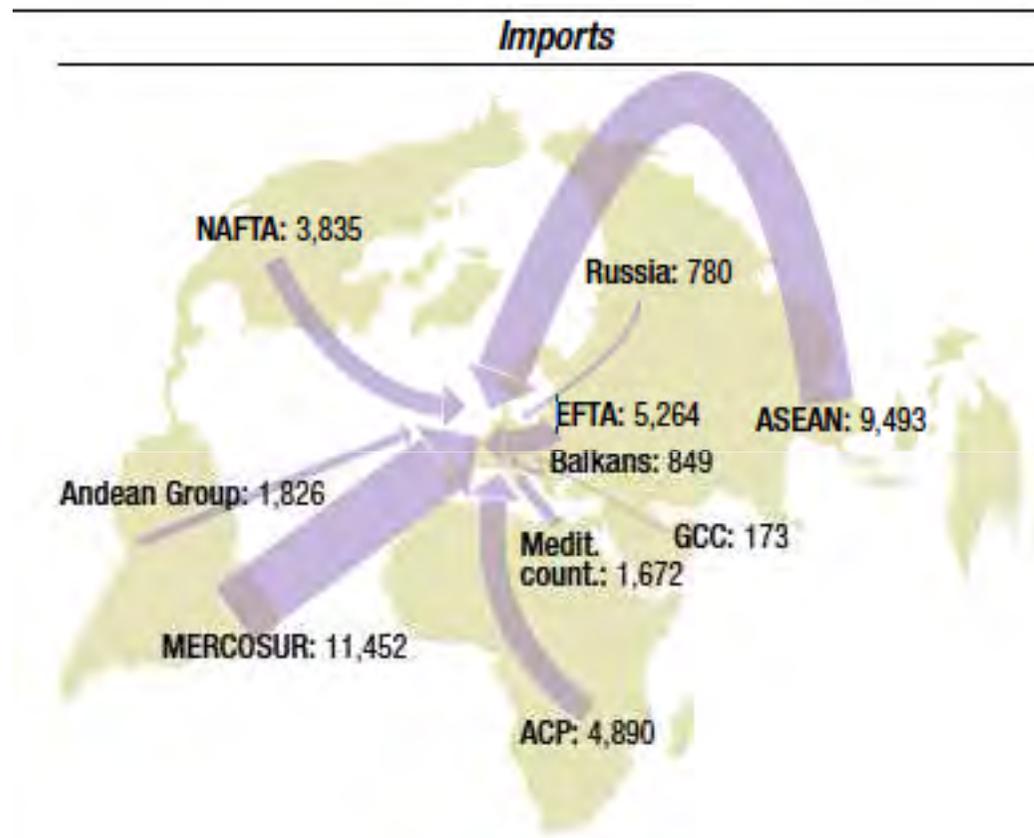
Fifty years ago, the average grocery store stocked about 200 items, of which 70 percent were grown, produced or processed within 100 miles of where they were eventually purchased.

Today, the average supermarket stocks close to 50,000 food items. By some estimates, the food Americans eat has traveled, on average, 2,000 miles before it is consumed. (Sources: Boston Globe and Mander)



Source: XTC World Innovation Panorama 2010; Copyright XTC 2010 (www.worldinnovation.com)

Importazioni di alimenti in EU (2009, Milioni di €)



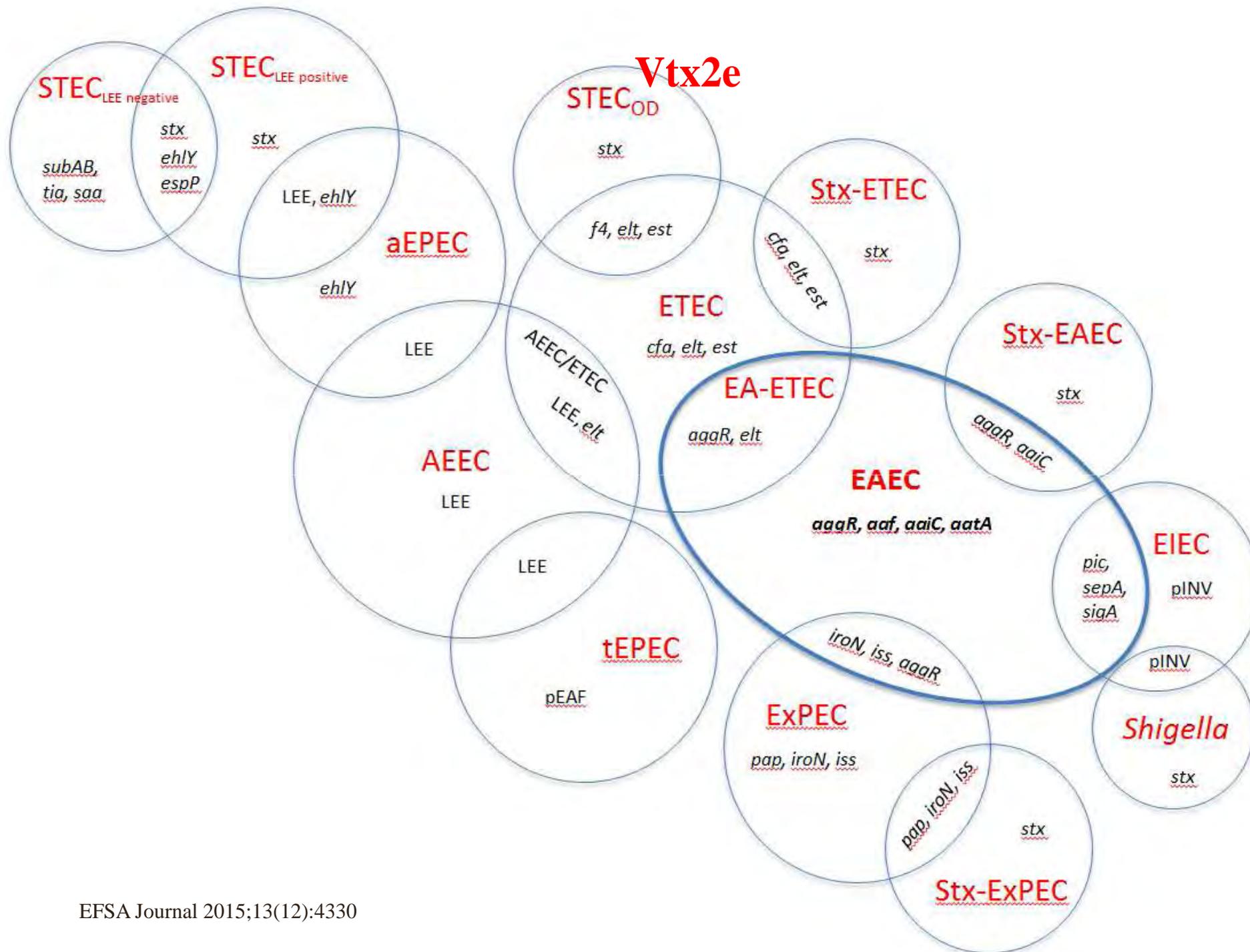
Source: Eurostat Comext

Patogeni emergenti possono essere introdotte in Europa con alimenti e materie prime provenienti dai paesi in via di sviluppo



Carte sur la géographie des grands flux migratoires.
 © Atlas des migrations, Le Monde-La Vie / CNHI / Sciences-Po





Il Piccione è un serbatoio naturale di VTEC

1998



Short Communications

Feral pigeons as a source of verocytotoxin-producing *Escherichia coli*

G. Dell'Omo, S. Morabito, R. Quondam, U. Agrimi,
F. Ciuchini, A. Macri, A. Caprioli

Veterinary Record (1998) **142**, 309-310

programme aimed at the control of pigeon populations, the presence of VTEC in faecal samples collected from pigeons living in the historical centre of Rome was examined.

Pigeons (*Columba livia*) were trapped between February 1 and March 30, 1997 in two different squares in Rome, the Piazza San Pietro and the Piazza Navona, which are situated about 2 km apart. One hundred and sixty birds were individually tagged and a faecal sample was collected from each animal soon after capture. Specimens were kept in a transport medium (Amies w/o charcoal; Difco Laboratories) until cultured in trypticase soy broth (Oxoid) at 37°C for 18 hours. The presence of VTEC was assessed by examining filter-sterilised supernatants from these enrichment cultures for the presence of verocytotoxins by the Vero cell cytotoxicity

2000

APPLIED AND ENVIRONMENTAL MICROBIOLOGY, Mar. 2000, p. 1205-1208
0099-2240/00/\$04.00+0

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Vol. 66, No. 3

A New Shiga Toxin 2 Variant (Stx2f) from *Escherichia coli* Isolated from Pigeons

HERBERT SCHMIDT,¹ JÜRGEN SCHEEF,¹ STEFANO MORABITO,² ALFREDO CAPRIOLI,²
LOTHAR H. WIELER,³ AND HELGE KARCH^{1*}

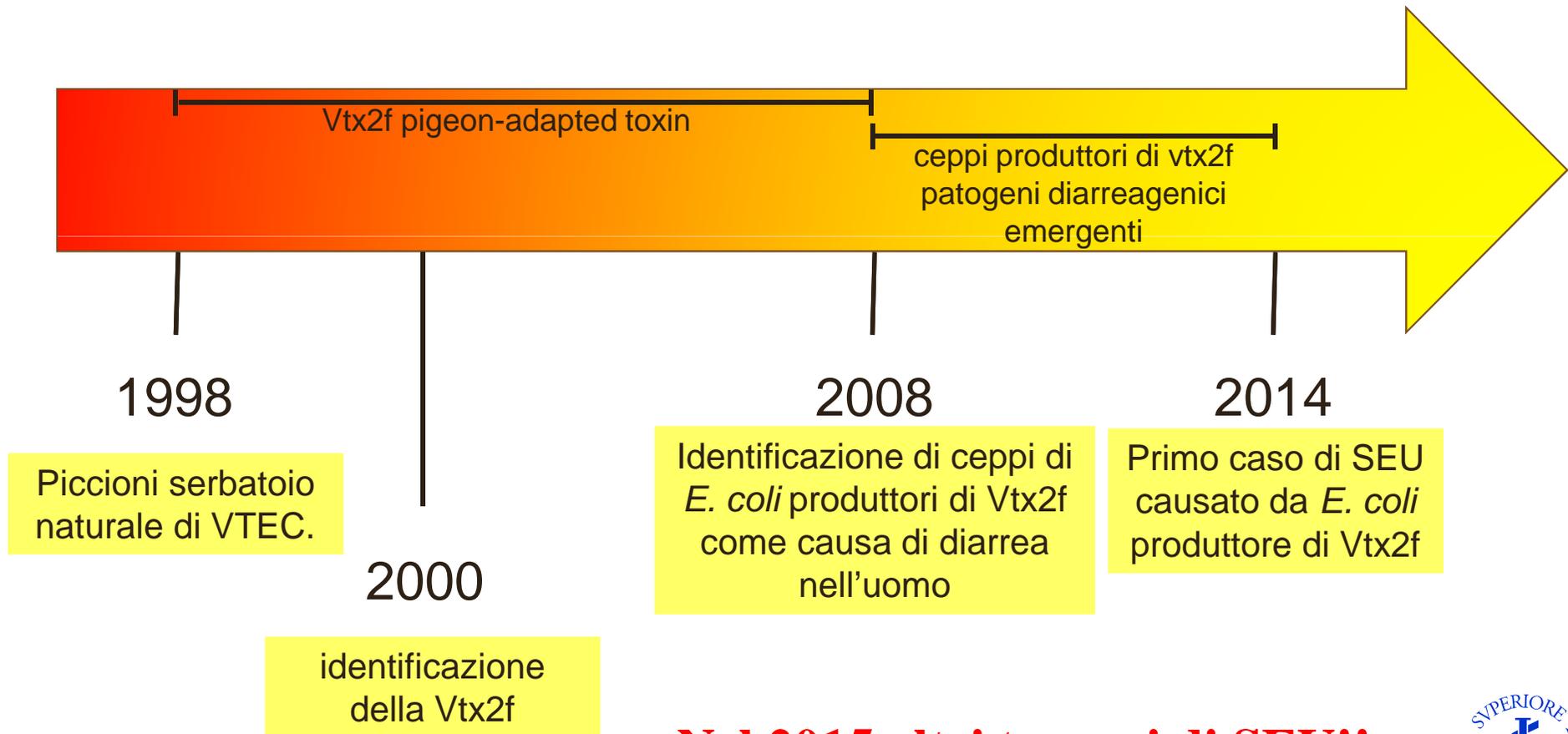
Institut für Hygiene und Mikrobiologie der Universität Würzburg, 97080 Würzburg,¹ and Institut für Mikrobiologie und Tierseuchen der Freien Universität Berlin, 10115 Berlin,² Germany, and Laboratorio di Medicina Veterinaria, Istituto Superiore di Sanità, Viale Regina Elena 299, 00161 Rome, Italy²

Received 14 July 1999/Accepted 10 December 1999



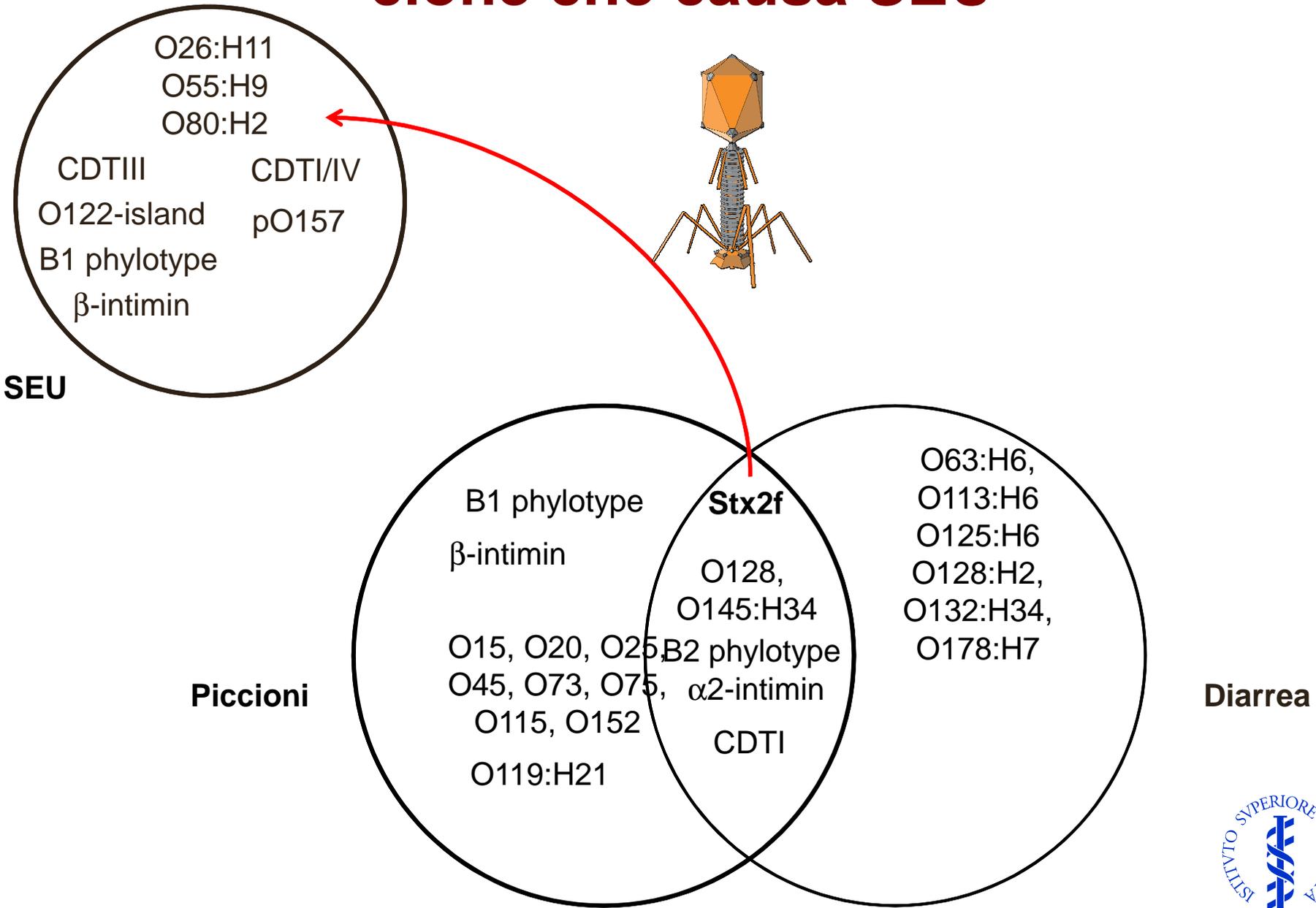
VTEC dei piccioni e malattia nell'uomo

Fino al 2004, Vtx2f-VTEC (LEE-positivi) erano considerati una variante "Host-adapted"



Nel 2015 altri tre casi di SEU!!

VTEC-Vtx2f: Ipotesi sull'emergenza di un clone che causa SEU



Enterobacter cloacae Producing a Shiga-Like Toxin II-Related Cytotoxin Associated with a Case of Hemolytic-Uremic Syndrome

ADRIENNE W. PATON AND JAMES C. PATON*

*Molecular Microbiology Unit, Women's and Children's Hospital,
North Adelaide, South Australia 5006, Australia*

Received 17 August 1995/Returned for modification 16 October 1995/Accepted 14 November 1995

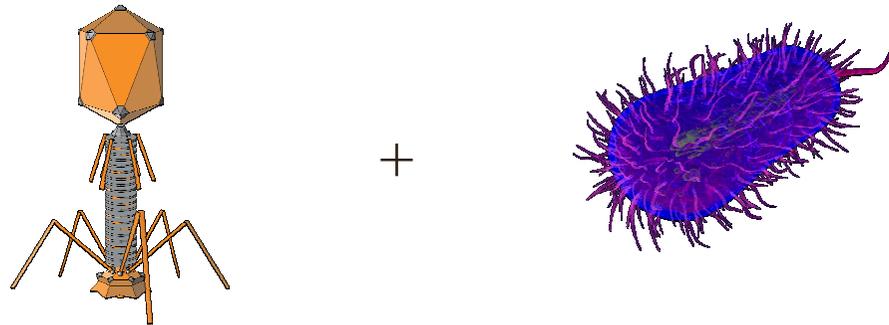
Two Shiga-like toxin-producing organisms were isolated from the feces of an infant with hemolytic-uremic syndrome by PCR followed by colony blot hybridization. One strain was identified as *Escherichia coli* OR:H9, while the other was identified as *Enterobacter cloacae*. Both isolates were highly cytotoxic for Vero cells, and Southern hybridization analysis of chromosomal DNA indicated that both contained a single *slt-II*-related gene and that these genes were located on similarly sized restriction fragments. Nucleotide sequence analysis indicated that the toxin encoded by the *E. cloacae* *slt-II*-related gene was very similar to Shiga-like toxin II variants from *E. coli*, differing from the most closely related toxin by 3 residues in the A subunit.

Altri reports: *C. freundii*, *E. albertii*



Un nuovo paradigma per la diagnostica clinica e l'epidemiologia delle infezioni

VTEC: sintesi di due microrganismi



I cloni VTEC stabilizzati che causano malattia nell'uomo sono diversi:

- Ceppi tipici che causano SEU (LEE positivi)
- Ceppi di *E. coli* Enteroaggregativi vtx2-produttori
- Ceppi LEE negativi produttori di SubAB
- Ceppi produttori di tossine "host-adapted" (Vtx2f)
- ETEC produttori di Vtx2d

...

Conclusioni

- ✓ I VTEC sono una classe ampia di ceppi di *E. coli* che producono Verocitotossina
 - ✓ Apparentemente non tutti i VTEC sono patogeni
 - ✓ Perchè causino la malattia nell'uomo devono avere un apparato per la colonizzazione del tratto intestinale
- ✓ I tipi di VTEC isolati da casi di malattia nell'uomo sono in continuo aumento
- ✓ Chiarire i meccanismi dell'emergenza delle nuove varianti aiuta a identificare i serbatoi animali e capire l'epidemiologia delle infezioni nell'uomo

Grazie per l'attenzione

