

# Genome-based phylogenetic analysis of O26 Verocytotoxin-producing *Escherichia coli*

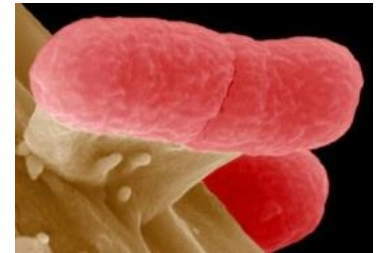
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[www.iss.it/vtec](http://www.iss.it/vtec)



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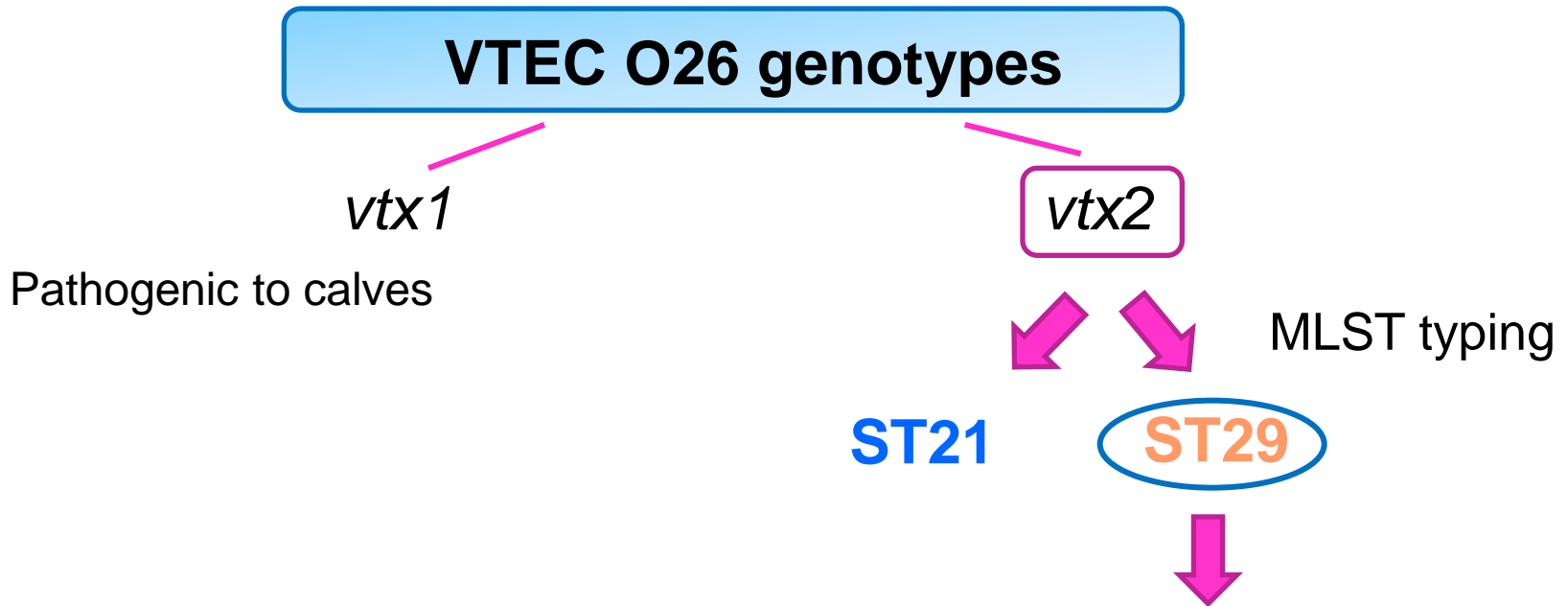
# Emergence of an O26 VT2 clone

Clinical Infectious Diseases 2013;56(10):1373–81

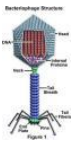
Enterohemorrhagic *Escherichia coli* O26:H11/H<sup>-</sup>:

A New Virulent Clone Emerges in Europe

Martina Bielaszewska,<sup>1,2</sup> Alexander Mellmann,<sup>1,2</sup> Stefan Bletz,<sup>1</sup> Wenlan Zhang,<sup>1</sup> Robin Köck,<sup>1</sup> Annelene Kossow,<sup>1</sup> Rita Prager,<sup>2</sup> Angelika Fruth,<sup>2</sup> Dorothea Orth-Höller,<sup>3</sup> Monika Marejková,<sup>4</sup> Stefano Morabito,<sup>5</sup> Alfredo Caprioli,<sup>5</sup> Denis Piérard,<sup>6</sup> Geraldine Smith,<sup>7</sup> Claire Jenkins,<sup>7</sup> Katarína Čurová,<sup>8</sup> and Helge Karch<sup>1</sup>



- Emerging since 1990s
- Survival after *vtx2*-encoding phage induction



# Clinical relevance of VTEC O26 in Italy



O26/tot	%tot	Year
9/42	21.4%	2011
17/43	39.5%	2012

20 O26 cases in **Apulia in August 2013**

12 O26 cases in the rest of Italy in 2013  
(7 in **August 2013**)

**Total 27  
O26 cases in Italy in  
August 2013**

## Epidemiological cluster



# Aims of the work

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- **Genome-based study on the evolution of VTEC O26**
- **Phylogenetic analysis of VTEC strains isolated in **Italy** in **2013****
- **Investigating the **ecology** of VTEC O26**

# Analysed strains

Human  
isolates

Tot 21

3 from USA (two from 1997, one from 2003)

16 Italy 2013

2 historical strains from Italy (1989 and 2010)



Strains from  
different  
sources

Tot 10

4 from cow feces (USA 2012)

1 from pig feces (USA 2012)

1 from unknown source (Japan 1989)

4 from food (Italy 2013)



# Methods

22 strains  
In house sequencing



9 strains  
GenBank retrieved data



Center for Genomic  
Epidemiology

Multi-Locus  
Sequence Typing  
(MLST)



Virulence genes

Single Nucleotidic  
Polymorphisms (SNPs)  
based phylogenetic tree

# Results



22 strains



MLST  
VirulenceFinder



Center for Genomic  
Epidemiology



9 strains

	ST21	ST29	<i>vtx1</i>	<i>vtx2</i>	<i>vtx-</i>
21 human isolates	15	6	5	15	1
5 isolates from animals	5	-	5	-	-
4 isolates from food	1	3	-	1	3
1 isolate of unknown origin	1	-	1	-	-

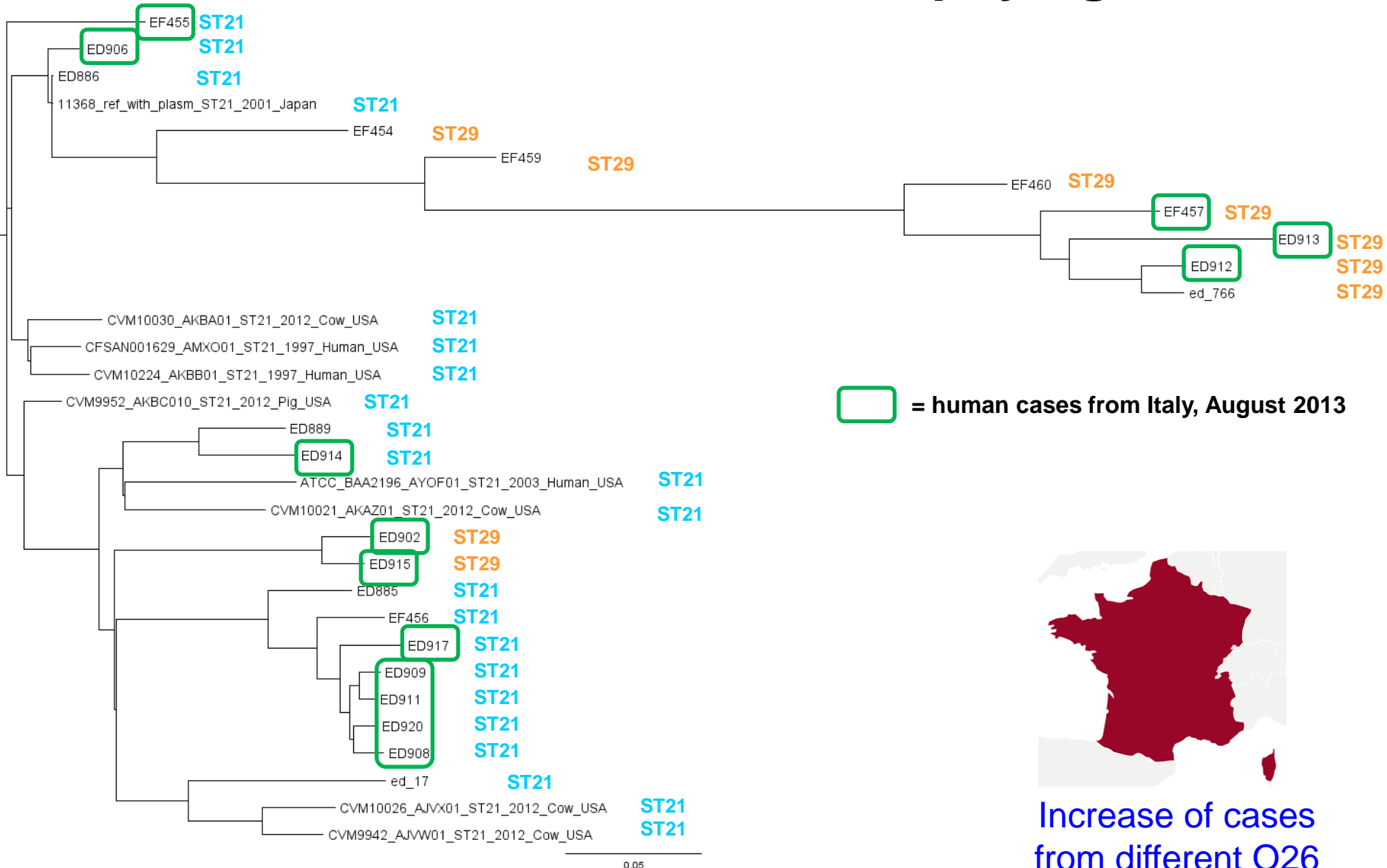
# Results

- No ST29 strains from animals
- All the *vtx1*-positive strains from human and animals belong to ST21. They cause diarrhea in calves and uncomplicated diarrhoea in humans.
- *vtx2*-positive strains are either ST21 or ST29

	ST21	ST29	<i>vtx1</i>	<i>vtx2</i>	<i>vtx-</i>
21 human isolates	15	6	5	15	1
5 isolates from animals	5	-	5	-	-
4 isolates from food	1	3	-	1	3
1 isolate of unknown origin	1	-	1	-	-
13 Human isolates from Italy August 2013	8	5	-	12	1



# SNPs based phylogenetic tree

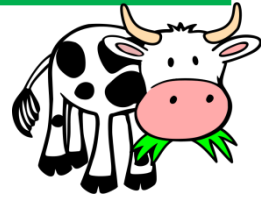


Increase of cases from different O26 VTEC isolates in France in 2013

# VTEC O26 ecology

O26 frequently isolated from ruminants

**RESERVOIR**



...but what about the ST?



Few studies investigating O26 isolated from animals and performing MLST

Zweifel C., Applied Environmental Microbiology 2013

Hauser E., Applied Environmental Microbiology 2013

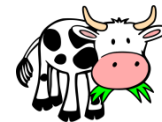
**ST21** Frequently isolated from ruminants

**ST29** Rarely isolated from animals

1 strain from cattle

1 strain from wild boar

**RESERVOIR**



# Source-sink evolution

ST29 O26 VTEC could have evolved according to a source-sink model

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## Source–sink dynamics of virulence evolution

*Evgeni V. Sokurenko, Richard Gomulkiewicz and Daniel E. Dykhuizen*

JULY 2006 | VOLUME 4 [www.nature.com/reviews/micro](http://www.nature.com/reviews/micro)

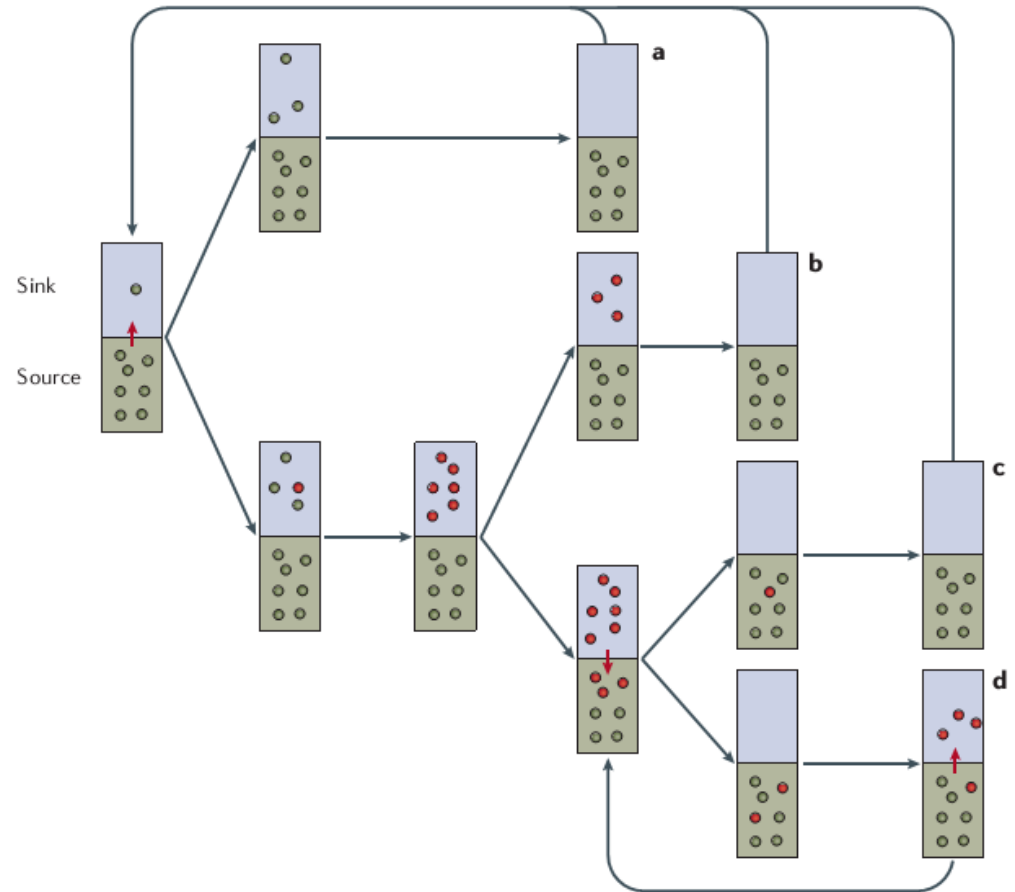
SINK



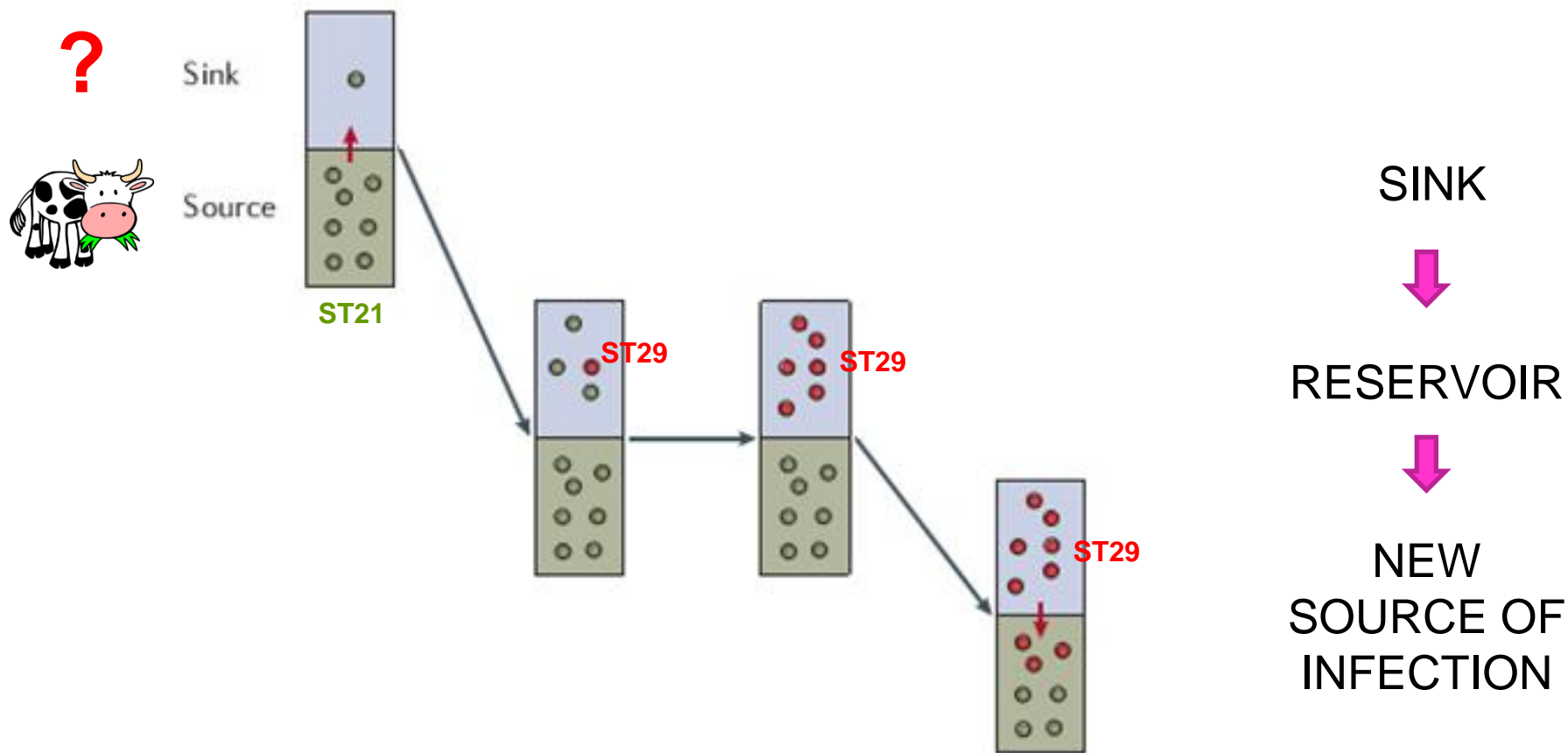
RESERVOIR



NEW SOURCE OF  
INFECTION



# Source-sink evolution in O26 VTEC



# Conclusions

## Source-sink hypothesis for ST29 O26 VTEC

SINK → RESERVOIR → NEW SOURCE

**Which is the new source???**

**Epidemiology** → Simultaneous cases, different isolates

- Foodborne zoonoses? Animal different from cattle? **No isolation of similar strains**
- Interhuman circulation? **Lack of familiar cases and correlated isolates**
- Environment



# Acknowledgements

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Claire Jenkins

Tim Dallman



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Antonio Parisi

Giovanna La Salandra

## Thank you for your attention!



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Laboratorio Europeo e Nazionale di Riferimento per *E. coli*

