



A scanning electron micrograph (SEM) showing several curved, hair-like Campylobacter bacteria against a dark background.

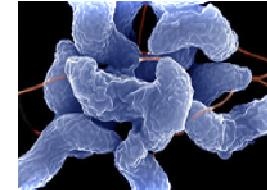
# La Diagnostica Rapida del **CAMPYLOBACTER**

3° Congresso NEWMICRO (Padenghe sul Garda 21 marzo 2013)

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# Campylobacteriosi



- **“Foodborne”: infezione di origine alimentare**
- **Prima causa di gastroenterite e diarrea acuta di origine batterica**
- **Incidenza elevata nella popolazione pediatrica**
- **Nei Paesi sviluppati Campylobacteriosi più frequente di Salmonellosi (casi sporadici)**
- **Bassa dose infettiva**
- **No trasmissione persona-persona**

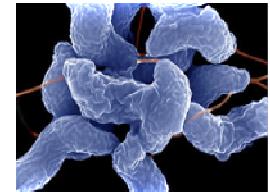
# Aspetti Clinici



- **Generalmente Autolimitante (2 settimane)**
- **Sintomi: gastroenterite acuta, diarrea, febbre, dolori addominali, crampi e talvolta sangue nelle feci**
- **Incidenza elevata nella popolazione pediatrica < 5 anni**
- **Diagnosi differenziale:**
  - **Altre infezioni/pseudoappendicite**
- **Complicazioni in pazienti**
  - **immunocompromessi – anziani – bambini piccoli**
- **Complicazioni post – infezione:**
  - **Sindrome di Guillain-Barré**
  - **Artrite reattiva**
  - **Appendicite/pancreatite**



# Le specie patogene

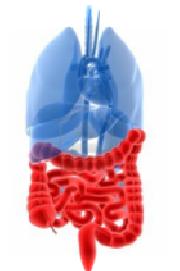


- ***Campylobacter jejuni* - 95-99%**
- ***Campylobacter coli* - 1-5%**

- ***Campylobacter lari***

- ***Campylobacter upsaliensis***

- ***Campylobacter fetus***



Gastro enteric infections

Extra-intestinal infections  
bacteremia

\* Tabella 11 da Not Ist Super Sanità 2011;24(1):3-10



**Tabella 11 - Isolamenti di ceppi di *Campylobacter* spp. da casi di infezione umana**

Specie	2007		2008		2009	
	n.	%	n.	%	n.	%
<i>Campylobacter jejuni</i>	445	65,6	458	77,2	389	73,2
<i>Campylobacter coli</i>	41	6,0	44	7,4	38	7,1
<i>Campylobacter lari</i>	6	0,9	1	0,2	6	1,2
<b>Totale speciati</b>	<b>492</b>	<b>72,6</b>	<b>503</b>	<b>84,8</b>	<b>433</b>	<b>81,5</b>
<i>Campylobacter</i> spp.	186	27,4	90	15,2	98	18,4
<b>Totale</b>	<b>678</b>	<b>100,0</b>	<b>593</b>	<b>100,0</b>	<b>531</b>	<b>100,0</b>

# Biology of *C. coli* - *C.jejuni*

- **Gram Negative**
- **Spiral or curved rods**
- **Motile**
- **Microaerophilic**
  - **5% oxygen with increased levels of carbon dioxide and nitrogen**
- **Thermophilic (42°C)**

Characteristic	Result
Growth at 25 °C	-
Growth at 35-37 °C	-
Growth at 42 °C	+
Nitrate reduction	+
Catalase test	+
Oxidase test	+
Growth on MacConkey agar	+
Motility (wet mount)	+
Glucose utilization	-
Hippurate hydrolysis	+
Resistance to naladixic acid	-
Resistance to cephalothin	+



- **Illness self-limited and usually resolves within 2 weeks.**
- **Symptoms: severe gastroenteritis, fever, abdominal pain and cramps. Sometimes bloody stool.**
- **Risks increased in people who have weak immune systems (cancer patients, people with AIDS, very young children and the elderly)**
- **In children the infection may be mistaken for appendicitis or a problem with the pancreas when abdominal pain is the most significant symptom**
- **Complications: bacteremia, relapsing enteritis, prolonged symptoms**
- **Post Infectious Complications:**
  - **Guillain-Barre' Syndrome: a rapidly progressive acute paralysis of peripheral nerves - *Incidence 1 per 1,000 to 3,000 infection***
  - **Reactive Arthritis: 2 to 7% of infections (most patients recover within 12 months)**



# Therapy Options

## REHYDRATATION



- **Campylobacter enteritis is a self-limiting disease**
- **Antimicrobial therapy: in severe cases only**
  - *often necessary for young children, pregnant women, or immunosuppressed patients because of the possibility of greater severity or duration of infection*

## ANTIBIOTICS OF CHOICE:

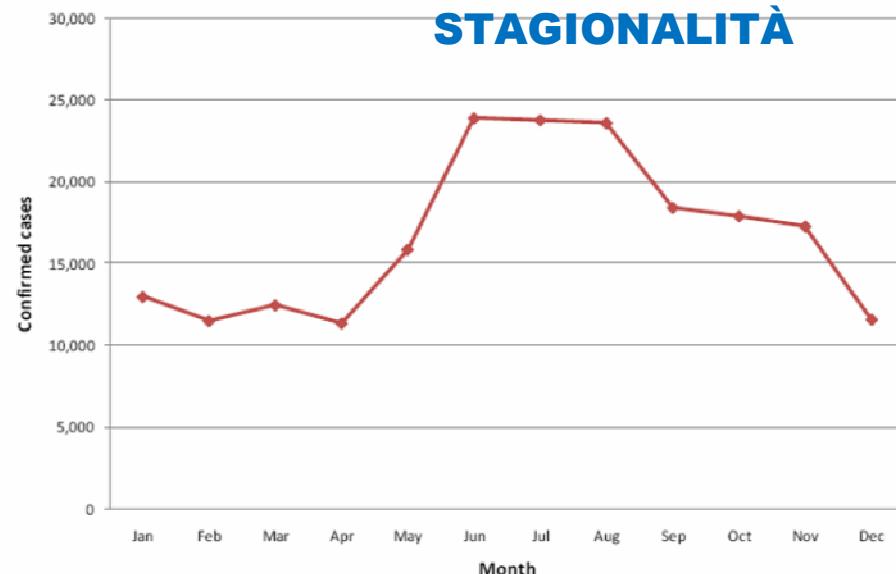
- **Macrolides such as Erythromycin or Azithromycin**
- **Fluorquinolones such as Ciprofloxacin may be used, but resistance rates are increasing**

# Reservoirs

Data	Total number of MSs reporting
Poultry meat	19
Pig meat	14
Bovine meat	11
Other types of meat	9
Milk and dairy products	10
Other food	10

[EFSA Journal 2012;10\(3\):2597](#)

Figure CA3. Number of reported confirmed campylobacteriosis cases in humans by month, TESSy data for reporting Member States, 2010



**Colonize the colon of  
farm animals and  
domestic pets**

# EPIDEMIOLOGIA del Campylobacter in Europa



EFSA JOURNAL

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The European Union Summary Report on Trends and Sources of Zoonoses, Zoonotic Agents and Food-borne Outbreaks in 2010

Figure CA1. | *Campylobacteriosis notification rates in humans in the EU (per 100,000 population), 2008*

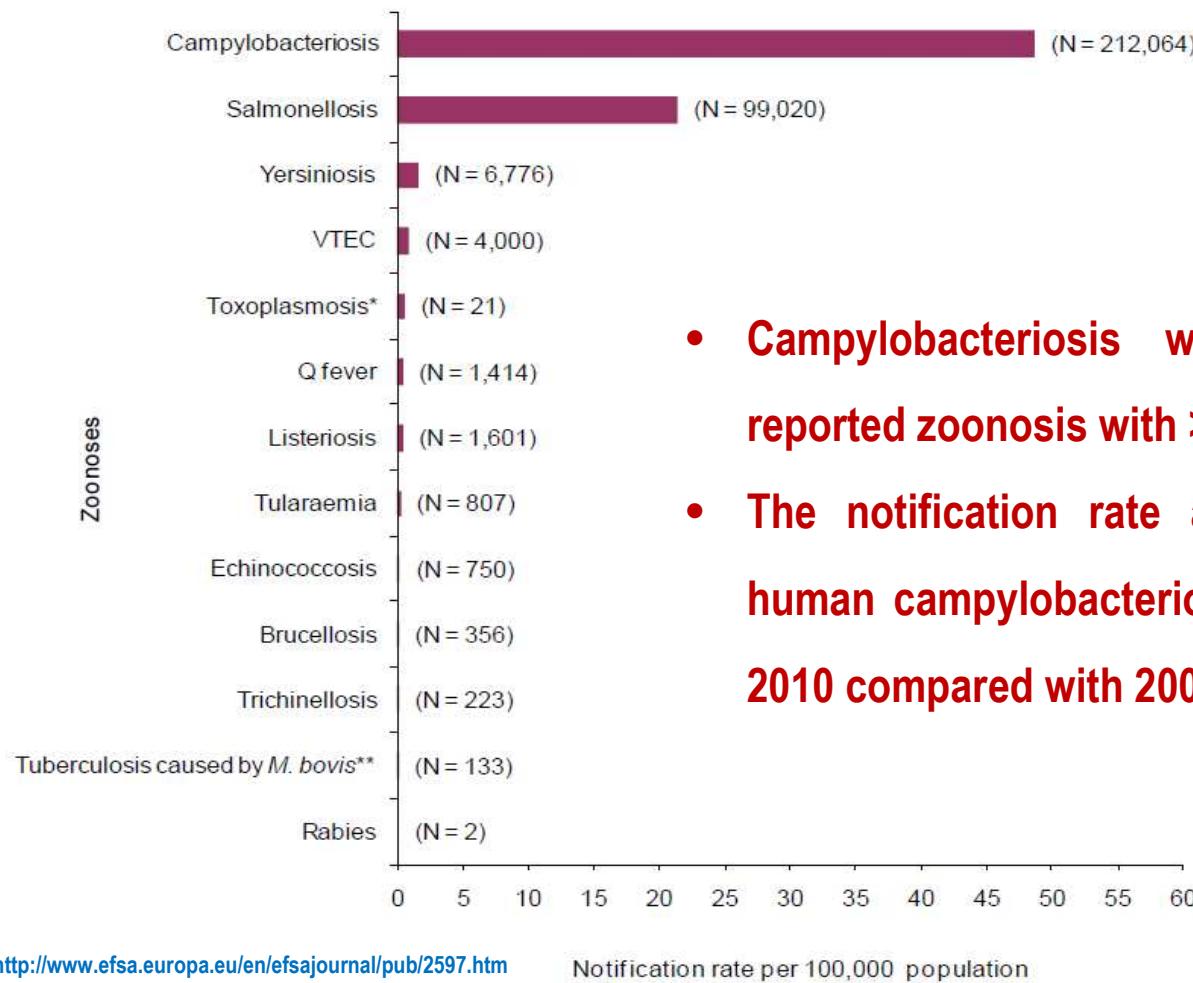


## The European Union Summary Report on Trends and Sources of Zoonoses, Zoonotic Agents and Food-borne Outbreaks in 2010<sup>1</sup>

The European Food Safety Authority and the European Centre for Disease Prevention and Control analysed the information on the occurrence of zoonoses and food-borne outbreaks in 2010 submitted by 27 European Union Member States.

### STATISTICHE CASI SPORADICI

Figure SU1. Reported notification rates of zoonoses in confirmed human cases in the EU, 2010



- Campylobacteriosis was the most commonly reported zoonosis with > 200'000 human cases
- The notification rate and confirmed number of human campylobacteriosis in the EU increased in 2010 compared with 2009 (+6%)

Table CA2. Reported campylobacteriosis cases in humans 2006-2010 and notification rates for 2010

EFSA Journal 2012;10(3):2597

Country	Report Type <sup>1</sup>	2010			2009	2008	2007	2006
		Cases	Confirmed cases	Confirmed cases/ 100,000				
Austria	C	4,405	4,405	52.60	1,516	4,280	5,822	5,020
Belgium	C	3,031	3,031	27.96	5,697	5,111	5,895	5,771
Bulgaria	A	6	6	0.08	26	19	38	75
Cyprus	C	55	55	6.85	37	23	17	2
Czech Republic	C	21,164	21,075	200.58	20,259	20,067	24,137	22,571
Denmark	C	4,037	4,037	72.94	3,353	3,470	3,868	3,239
Estonia	C	197	197	14.70	170	154	114	124
Finland	C	3,944	3,944	73.70	4,050	4,453	4,107	3,439
France	C	4,324	4,324	6.68	3,956	3,424	3,058	2,675
Germany	C	65,713	65,110	79.59	62,787	64,731	66,107	52,035
Greece	- <sup>4</sup>	-	-	-	-	-	-	-
Hungary	C	7,201	7,201	71.91	6,579	5,516	5,809	6,807
Ireland	C	1,662	1,660	37.15	1,810	1,752	1,885	1,812
Italy	C	457	457	0.76	531	265	676	801
Latvia	C	1	1	0.04	0	0	0	0
Lithuania	C	1,095	1,095	32.89	812	762	564	624
Luxembourg	C	600	600	119.51	523	439	345	285
Malta	C	204	204	49.40	132	77	91	54
Netherlands <sup>2</sup>	C	4,322	3,983	46.21	3,739	3,341	3,289	3,186
Poland	C	375	367	0.96	359	270	192	157
Portugal	- <sup>4</sup>	-	-	-	-	-	-	-
Romania	C	179	175	0.82	254	2	-	-
Slovakia	C	4,578	4,476	82.51	3,813	3,064	3,380	2,718
Slovenia	C	1,022	1,022	49.93	952	898	1,127	944
Spain <sup>3</sup>	C	6,340	6,340	55.14	5,106	5,160	5,331	5,889
Sweden	C	8,001	8,001	85.66	7,178	7,692	7,106	6,078
United Kingdom	C	70,298	70,298	113.37	65,043	55,609	57,849	52,134
<b>EU Total</b>		<b>213,211</b>	<b>212,064</b>	<b>48.56</b>	<b>198,682</b>	<b>190,579</b>	<b>200,807</b>	<b>176,440</b>
Iceland	C	55	55	17.32	74	98	93	117
Liechtenstein	-	-	-	-	-	2	0	10
Norway	C	2,682	2,682	55.21	2,848	2,875	2,836	2,588
Switzerland <sup>5</sup>	C	6,604	6,604	85.05	7,795	7,552	5,834	5,240

<http://www.efsa.europa.eu/en/efsjournal/pub/2597.htm>

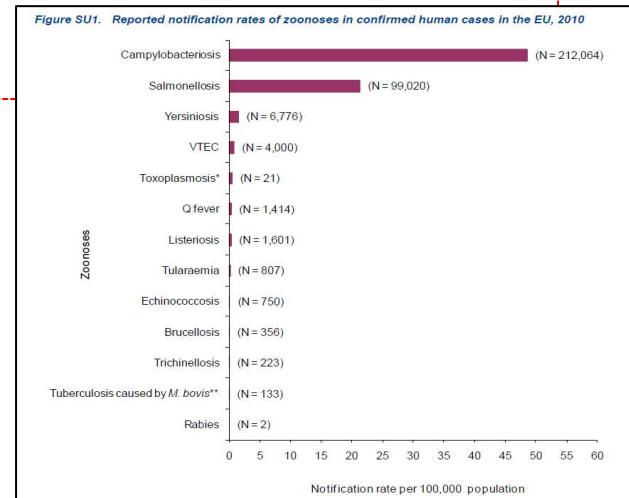


## DATI 3 ANNI\* (ENTERNET): 2007-2009

- Salmonella 11'714 casi
- Campy 1'802 casi

## PERCHE'?

- NO notifica obbligatoria per Campy in Italia
- Migliore sorveglianza/gestione derrate alimentari?
- Diagnostica di laboratorio?



\*Not Ist Super Sanità 2011;24(1):3-10

# CAMPY: THE LABORATORY DIAGNOSIS

- **Isolation from stool by Culture**
  - (filters/antibiotics)



- **Antigen Detection in stool**
  - Commercial EIAs



- **NAAT (commercial – homebrew)**



# DIAGNOSIS by CULTURE

## DRAWBACKS and LIMITS



- **Proper Sample collection and Transportation**

- 2-8°C in «modified Cary-Blair» within 24 h

organisms must be  
viable to grow

- **Culture on agar plate**

- Microaerophilic conditions: 5% O<sub>2</sub>, 10% CO<sub>2</sub>
  - Combined methods needed
    - A) Selective media (antibiotics) – 42°C after 24 h
    - B) Filter method at 37°C

more than one culture  
method needed

- **Biochemical Identification and typing:**

*at least*

- Gram stain
  - Oxidase test

LONG TAT

- Visible colonies at 40-48 hrs,  
• ≥ 72 hours to report negative

# Campylobacteriosis diagnosis: Culture drawbacks



- **Organisms must be viable to grow (collection - transportation to the lab must be strictly controlled)**
- **Must have optimal micro-aerobic conditions (equipment?)**
  - **37-42°C optimal temperature**
  - **5% O<sub>2</sub>, 10% CO<sub>2</sub>**
  - **Selective methods should be used (filtration/antibiotics)**
- **Biochemical confirmation of colonies is necessary**
- **Long TAT**
  - **Visible colonies at 40-72 hrs,**
  - **> 72 hours to report negative**

# Bessede et al, JCM 2011

## France Reference Center



CULTURE HAS A  
LOW SENSITIVITY!

JOURNAL OF CLINICAL MICROBIOLOGY, Mar. 2011, p. 941–944  
0095-1137/11/\$12.00 doi:10.1128/JCM.01489-10

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Vol. 49, No. 3

## New Methods for Detection of Campylobacters in Stool Samples in Comparison to Culture<sup>▽†</sup>

Emilie Bessède,<sup>1,2,3</sup> Adline Delcamp,<sup>2</sup> Elodie Sifré,<sup>2</sup> Alice Buissonnière,<sup>2</sup> and Francis Mégraud<sup>1,2,3\*</sup>

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Received 22 July 2010/Returned for modification 23 December 2010/Accepted 28 December 2010

*Campylobacter* species, especially *Campylobacter jejuni* and *Campylobacter coli*, are a major cause of human bacterial enteritis. Current detection in stools is done essentially by culture on selective and nonselective media with filtration. These methods were compared to 2 molecular biology methods, an in-house real-time PCR and a multiplex PCR named Seeplex Diarrhea ACE Detection, and 3 immunoenzymatic methods, Premier Campy, RidaScreen Campylobacter, and ImmunoCard Stat!Campy. Out of 242 stool specimens tested, 23 (9.5%) fulfilled the positivity criteria, i.e., they were positive by one or both culture methods or, in case of a negative culture, by a positive molecular method and a positive immunoenzymatic method. The striking feature of this study is the low sensitivity of culture, in the range of 60%, in contrast to immunoenzymatic and molecular tests.

# Bessede et al, JCM 2011

## France Reference Center

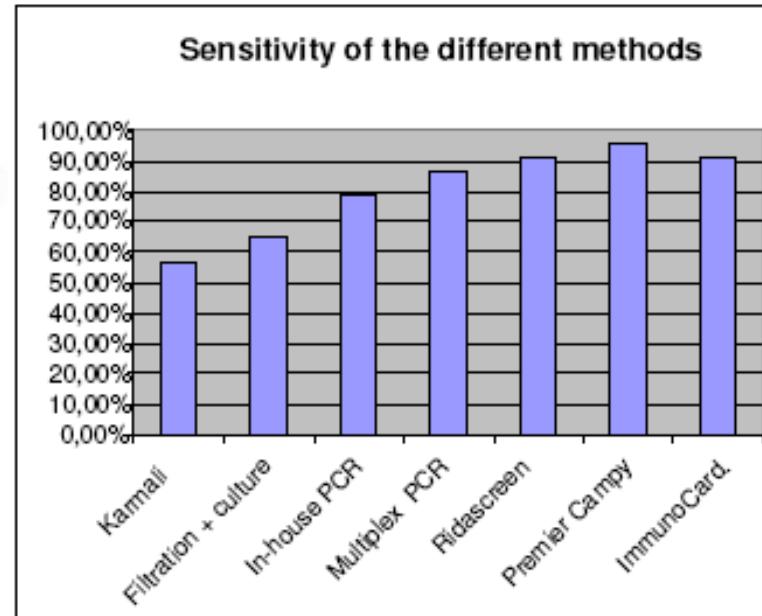
Figure 1. Sensitivity and positive predictive value of the different techniques used for *Campylobacter* detection.

## CONCLUSIONS

ImmunoCardSTAT!Campy<sup>®</sup> is its rapidity as, the results being obtained in less than 30 min, as well as its convenience in comparison to the ELISAs and PCRs.

In conclusion, this study highlights the limits of culture methods to detect campylobacters in stools. Currently the best accuracy is obtained by ELISAs followed by molecular methods.

However, the rapid detection by ImmunoCardSTAT!Campy<sup>®</sup> is attractive and further studies should be performed to specify the conditions of its use, and we may have to change the guidelines for *Campylobacter* detection in the future.





# L'esperienza nella Regione Piemonte

Sistema Sorveglianza Malattie da Alimenti (2001)

Sistema di Sorveglianza Laboratorio (2009)

AREA	CASI CONFERMATI in LAB ogni 100'000 abitanti (dati 2010) <sup>1,2</sup> :
Media EU	48
UK	113
Spagna	55
Italia	0.76
Piemonte <sup>2</sup>	10.3

<sup>1</sup> [www.efsa.europa.eu/en/efsajournal/pub/2597.htm](http://www.efsa.europa.eu/en/efsajournal/pub/2597.htm)

<sup>2</sup> Serra et al, Aspetti Clinici ed Epidemiologici dell'infezione da Campylobacter - 2012 - [www.regione.piemonte.it/sanita/cms/documentazione](http://www.regione.piemonte.it/sanita/cms/documentazione)

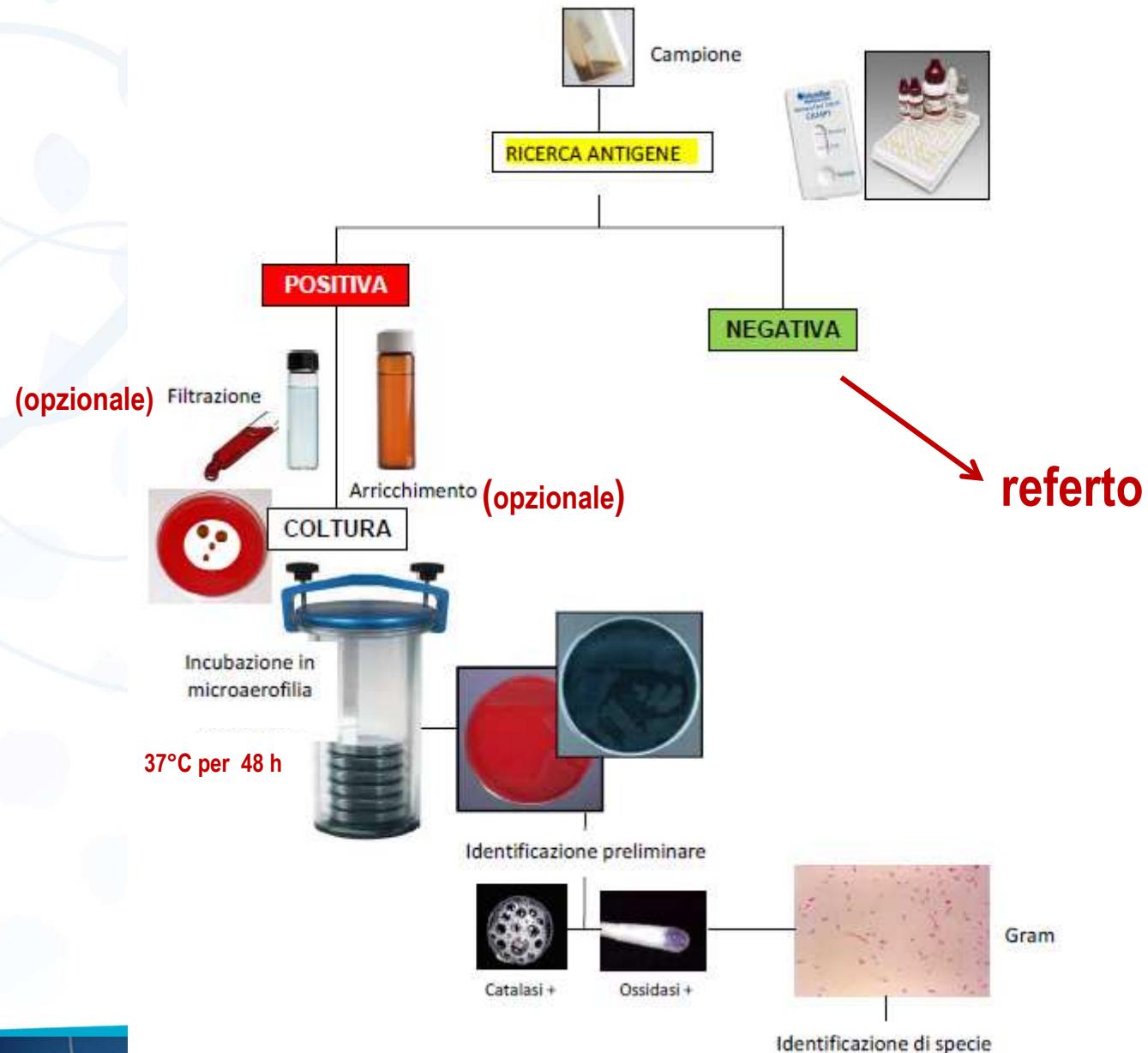
# Disposizioni per la sorveglianza Regione Piemonte – 2012\*

- Se possibile, fornire al laboratorio un campione prima di somministrare antibiotici.
- I campioni fecali devono essere inviati al laboratorio entro 2-4 ore dalla raccolta

## Diagnosi microbiologica: protocollo raccomandato

- Ricercare la presenza dell'antigene nel campione con metodo immunometrico CARD o ELISA
  - In caso di risultato negativo referire: “*Negativa la ricerca di Campylobacter spp.*”
  - In caso di risultato positivo emettere il referto preliminare “*Positiva la ricerca dell'antigene di Campylobacter: coltura in corso*” – Referto definitivo dopo 48 ore
  - Per campioni positivi agli antigeni e negativi alla coltura, verranno date informazioni supplementari dopo altre 48 ore di incubazione:  
*«Positivo alla ricerca di antigeni di Campylobacter, la coltura è tuttavia negativa. Possibile presenza nel campione di ceppi di Campylobacter non coltivabili a causa delle modalità di conservazione del campione non corrette»*

Fig. 1 - Algoritmo diagnostico (vedi protocollo diagnostico e relative note)



# Meridian Options:

## Premier®CAMPY



Catalog Number:	618096
Kit Size:	96 microwells
Specimen Type:	Unpreserved or Cary-Blair preserved stool
Sensitivity:	97.7%
Specificity:	95.3%
Time to Results (TAT):	2 hours
Shelf Life:	18 months – Storage 2-8°C



## ImmunoCard STAT!® CAMPY

Catalog Number:	751530
Kit Size:	30 tests
Specimen Type:	Unpreserved or Cary-Blair preserved stool
Sensitivity:	98.1 %
Specificity:	95.9 %
Time to Results (TAT):	20 min
Shelf Life:	18 months – Storage 2-8°C



# Para-Pak® C&S

Catalog Number:

900612

Assay Format:

Para-Pak® device - stool transport vial

Kit Size:

120 vials

Specimen Type:

Stool

Kit Storage Requirements:

Room Temp

Shelf Life:

18 months



- Sistema standardizzato per raccolta/trasporto/conservazione campioni fecali per ricerca di batteri patogeni enterici
- Variante del terreno Cary-Blair, ottimizzato per *Campylobacter*
- Con indicatore di Ph – fino a 96 ore
- Formulazione liquida – utilizzabile con piastratore automatico

# Grazie!

**per informazioni sui prodotti e bibliografia**

**visitate il nostro nuovo sito**

**www.meridianbioscience.eu**

**oppure:**

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