

Analysis of two cross-contamination cases of *Campylobacter jejuni* foodborne disease in fragile subjects in the territory of a Local Health Authority in Tuscany, Italy

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Abstract

Campylobacteriosis is the most reported foodborne disease in the European Union, with more than 100,000 confirmed cases annually. Human infection can be caused by a low infectious dose, and in fragile populations, the food disease can manifest itself in acute and severe forms. This study aims to analyze two cases of

campylobacteriosis in fragile people caused by *Campylobacter jejuni* in 2023 in Tuscany and the actions of the Local Health Competent Authority. From the results of the related investigations, it was possible to attribute both cases of foodborne diseases to unsafe food management during preparation/administration. Given the peculiar characteristics of the etiological agent, it is necessary to focus the attention of the population, especially those who deal with fragile subjects, on the good hygiene practices to be followed both at home and in collective catering.

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Key words: campylobacteriosis, *Campylobacter jejuni*, fragile people, official control, cross-contamination.

Contributions: all the authors made a substantial intellectual contribution, read and approved the final version of the manuscript, and agreed to be accountable for all aspects of the work.

Conflict of interest: the authors declare that they have no competing interests, and all authors confirm accuracy.

Ethics approval and consent to participate: not applicable.

Patient consent for publication: not applicable.

Funding: none.

Availability of data and materials: data available from the corresponding author upon request.

Received: 6 November 2023.
Accepted: 21 December 2023.
Early access: 21 February 2024.

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Licensee PAGEPress, Italy
Italian Journal of Food Safety 2024; 13:12053
doi:10.4081/ijfs.2024.12053

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Introduction

Campylobacteriosis is an infectious disease caused by gram-negative, spiral-shaped, S-shaped, or curved, flagellated, non-spore-forming, microaerophilic bacteria (Epps *et al.*, 2013). The disease is mostly linked to the species *Campylobacter jejuni* and *Campylobacter coli*. Worldwide, 4% of infections are caused by *C. coli* and the rest by *C. jejuni* (Costa and Iraola, 2019).

The infection can be transmitted directly or indirectly through food, animals, and humans. Since 2005, campylobacteriosis has been the most common gastrointestinal disease in the European Union, with 128,000 cases reported in 2021 (European Food Safety Authority and European Center for Disease Prevention and Control, 2022). Economic repercussions, in terms of health care costs and loss of productivity, are estimated at around 2.4 billion euros a year (European Food Safety Authority-Panel on Biological Hazards, 2011).

The high diffusion of campylobacteriosis may be attributable to *Campylobacter* spread in the environment and animals, mainly poultry, its adaptation to survival outside the host, and its low infectious dose (500-800 CFU) (Janssen *et al.*, 2008; Bronowski *et al.*, 2014; Teunis *et al.*, 2018).

Although *Campylobacter* is unable to grow in food, undercooking of raw foods and/or cross-contamination from raw food harboring even low microbial counts to ready-to-eat food is a major risk factor for human infection (Hansson *et al.*, 2018).

Campylobacteriosis is mainly characterized by non-specific gastroenteric symptoms, sometimes accompanied by fever, headache, vomiting, and muscle pain (Chlebicz and Śliżewska, 2018). However, more serious evolution, often chronic, can include reactive arthritis, hepatitis, pancreatitis, septicemia, and the neurological disorder known as Guillain-Barré syndrome (Finsterer *et al.*, 2022). As far as the fragile population (young, old, pregnant, and immunocompromised) is concerned, the disease can manifest itself in severe acute forms (Igwaran and Okoh, 2019). The incubation lasts 2-5 days, and the duration of symptoms varies from

3 to 7 days (Centers for Disease Control and Prevention, 2019).

Cases and outbreaks are reported every year in the European territory. In the European Union, outbreaks are mostly attributable to the consumption of contaminated water, raw dairy products, and mainly chicken meat (Heuvelink *et al.*, 2009; Hyllestad *et al.*, 2020; Wensley *et al.*, 2020), often through cross-contamination that may occur from the contact between ready-to-eat foods and raw chicken meat (European Food Safety Authority and European Center for Disease Prevention and Control, 2022).

Campylobacteriosis is also a big concern in the catering industry, representing a potential source of infection due to the large number of meals served and their specific manufacturing requirements (Osimani and Clementi, 2016). Hospital and school catering services deserve a special mention because of the high number of served meals and capillary distribution. Furthermore, the vulnerability of consumers receiving these food services and the consequences of any adverse effects related to different hazards are of the utmost importance and need to be thoroughly considered (Lupattelli *et al.*, 2022).

This study aims to analyze two cases of campylobacteriosis in vulnerable subjects that occurred between February and March 2023 in the Tuscan territory and the consequent actions of the Local Health Authority (LHA).

Methods

Regional setting and case definition

Tuscany, one of Italy's 20 regions, has a population of 3.6 million people (Italian National Institute of Statistics, 2023), 1 million of whom live in the metropolitan area of the city of Florence. Tuscany is divided into ten provinces and counts three LHA: North West, Center, and South East Tuscany.

The three LHA are the territorial branches of the Regional Health Service and ensure the uniformity of care in the different areas of the region. Organized into district units, they manage and plan the activities defined in the uniform and essential levels of care, including highly integrated social-health services and official control activities delegated to local authorities. The two cases occurred within the Center LHA (C-LHA) territory, and C-LHA assumed responsibility for managing these cases of campylobacteriosis and coordinating subsequent control activities.

In Italy, campylobacteriosis is subject to mandatory notification, and according to the routine procedure, suspected cases must be reported to the LHA within 48 hours of the diagnosis (Italian Ministry of Health, 1990). Personnel of the C-LHA performed an epidemiological investigation of the two suspected cases, including a laboratory investigation, contact tracing, and inspections.

Description of the cases

Case 1 (February 2023)

The first reported case concerned a 3-month-old newborn girl, born in Sweden in October 2022 from parents of Romanian nationality and fed exclusively with stage 1 ultra-high temperature-treated infant milk produced by an officially recognized European company. The child had been brought from Sweden to Romania from December 22nd until January 23rd, and from there she had arrived in Italy. During the journey, the child continued to be fed with the same formula.

The newborn girl made access to the emergency department of

a pediatric ward of the local hospital, where the evacuation of diarrheal stools persisted for approximately 3 days. Starting on February 2nd, mucus and blood traces appeared in the stools, accompanied by a fever spike (approximately 38.5°C).

The newborn girl was initially discharged on February 3rd, afebrile, after a stay in the short intensive observation unit. Antigen tests for Rotavirus and Adenovirus in feces resulted in negative results; a complete abdominal ultrasound yielded normal results, and the gastroenterological assessment suggested an infectious origin. Consequently, a stool culture was performed. Therefore, clinical monitoring at home was advised, along with the continuation of feeding with a stage 1 infant formula. A follow-up was scheduled for February 17th. However, on February 4th, there was a new admission to the pediatric ward of the local hospital due to the persistence of semi-formed stool evacuations (approximately 7-8/day) with traces of mucus and bright red blood, along with malnourishment. Due to the persistence of symptoms, the newborn was subsequently admitted to the medical pediatrics department. The stool culture taken on February 2nd tested positive for *C. jejuni*. On February 7th, campylobacteriosis notification and reporting were carried out to the C-LHA, and an interview and questionnaire regarding cases of gastroenteritis transmitted through food agents were conducted with the parents of the newborn. Following the clinical examination and in agreement with the infectious diseases specialist, antimicrobial therapy was initiated using azithromycin. The newborn was discharged in a safe condition on February 8th, with some scheduled follow-up visits.

Case 2 (March 2023)

The second case of campylobacteriosis was reported in March 2023. The case concerned an immunocompromised individual undergoing chemotherapy treatment who was hospitalized in the hematology department of the local hospital. The man was admitted to the hospital on March 1st, and the onset of symptoms (3-4 episodes of diarrhea/day) started on March 12th. The enteric symptomatology (persistent diarrhea) occurred during the hospitalization period. *C. jejuni* was isolated on March 20th via stool culture derived from a patient's sample obtained within the hospital ward. At the time of the investigation, the patient still exhibited mild symptoms. Throughout the hospital stay, the patient reported having exclusively consumed meals provided by the hospital facility and specifically intended for immunocompromised patients. However, it was not possible to sample the meals eaten. Consumed products included bottled mineral water, veal, turkey, parmesan cheese, peas, broccoli, peeled oranges, and orange juices.

Discussion

Actions taken by the Local Health Authority

The cases described cannot be defined as an outbreak; instead, they are considered sporadic cases. This is because a foodborne outbreak is defined as an incidence, observed under specific circumstances, involving two or more human cases of the same disease or infection. It also encompasses situations where the observed number of cases exceeds the expected number and where the cases are linked to or probably linked to the same food source (European Commission, 2003). The actions taken by the LHA were therefore distinct and specific for each individual case.

In Case 1, upon receiving notification of a campylobacteriosis case from the hospital, the LHA contacted the milk-producing

company *via* email on February 10th, informing it about the planned sampling and analysis for *Campylobacter* research. The email included specific details about the product batch and the product label. The company was also notified that the analysis would be conducted at the Official Laboratory of the Experimental Zooprophyllactic Institute of Lazio and Tuscany. It was emphasized that the analysis could not be repeated due to product inconsistency. A milk sample that the child had consumed was collected by LHA operators. The sampling report confirmed the delivery of an intact 200-mL brick of milk without any visible signs of tampering, stored at room temperature. The proof of purchase, indicating that the milk was bought in Sweden between December 15th and 17th, 2022, was also provided to the LHA. The sample was analyzed using reverse transcription polymerase chain reaction (method AOAC 031209:2021) and gave a negative result (absence in 25 mL). Based on the negative result, the newborn's campylobacteriosis was attributed to cross-contamination that occurred during milk preparation and other food manipulation. In this case, it could have been useful to provide education to the parents on the correct use of teats/bottles and their segregation from ordinary food.

In Case 2, the LHA decided to conduct an official control through an audit of the kitchen of the hospital canteen, which had as its object the management of special diets. The audit was conducted by the manager of the C-LHA's audit program with the director of the area to which the hospital belonged. The audit technique was chosen over an inspection because the LHA aimed to thoroughly investigate the entire management system of special diets. Additionally, since the hospital canteen was under contract with a service company, the presence of the company's quality assurance manager, along with hospital management, was deemed essential. Consequently, providing advanced notice of the official control, although brief, was necessary. For these reasons, the inspection technique was not considered sufficiently effective. The audit revealed some critical issues, primarily concerning the use of the same space for the preparation of all types of special diets. As a result, the audit team concluded that there was a need to differentiate between the various types of special diets and designate specific areas for the preparation of diets with distinct hygienic requirements (*e.g.*, allergies, intolerances, celiac disease, low microbial load diets). Additionally, the audit team determined that it was necessary to provide specific training to the food operators, focusing on good hygiene practices (GHP) and good manufacturing practices (GMP) to be followed during the preparation and administration of special diets. Then, the LHA agreed on timing with the company for resolving the identified non-conformities and checking compliance with the prescriptions. Also, in this case, it is important to note that the campylobacteriosis was likely to be connected to cross-contamination that occurred during the food preparation/administration. It is imperative for catering services to ensure the strict application of GHP and GMP principles, along with an appropriate hazard analysis tailored to the specific context, which should take into consideration the risk of cross-contamination (Lupattelli *et al.*, 2022).

Conclusions

The analysis of two cases of campylobacteriosis in different vulnerable individuals due to cross-contamination has highlighted the necessity for a collaborative effort among the stakeholders involved in the prevention of this foodborne disease. The adoption of proper control measures by food handlers remains essential,

especially when food is intended for potentially vulnerable individuals, as is the case in collective catering, daily meal services, or special dietary facilities. In these scenarios, it becomes evident that training must incorporate a strong emphasis on raising awareness and cultivating a mature food safety culture among the personnel (Food and Drug Administration, 2020). Furthermore, initiatives to support these establishments, such as guidelines for managing collective catering, could encompass hygienic measures and examples to prevent cross-contamination during meal production. Simultaneously, it is desirable that the competent authorities at various levels, including ministries, regional bodies, and local districts, engage in public awareness campaigns aimed at promoting proper food handling and safety practices, with a particular focus on vulnerable populations. Targeted initiatives, such as those designed for new parents, can offer practical guidance and support, empowering them to protect their families and individuals with fragile health from campylobacteriosis. Similar initiatives have been implemented in other EU countries, as exemplified by the "Don't Wash Raw Chicken" campaign (British Food Standard Agency, 2014). Lastly, vigilant surveillance and rigorous official controls on companies engaged in food production and food service/catering industries remain crucial, as does the prompt activation of notifications to LHA and dialogue among stakeholders for the investigation and proper management of cases and outbreaks.

In conclusion, the cases analyzed have emphasized the importance of a dual-pronged approach that combines public awareness campaigns and regulatory oversight to prevent campylobacteriosis, often linked to inadequate manipulation of raw or undercooked food by food handlers. This approach, which encompasses education and enforcement, is pivotal in safeguarding the health of vulnerable populations and the wider community.

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