CAMPYLOBACTER AND *ARCOBACTER* CONTAMINATION OF RAW MEAT AT RETAIL SALE IN ITALY

Marcella Pintori, Mariangela Frustoli, Paola Mutti, Stefania Quintavalla and Silvana Barbuti

Stazione Sperimentale per l'Industria delle Conserve Alimentari, V.le F. Tanara, 31/A, 43100 Parma, Italy

Key words:
Thermophilic *Campylobacter* and *Arcobacter* spp., pork and poultry meat, incidence.

Background
Campylobacter spp. are now recognized as a major cause of enteric infection in humans (campylobacteriosis). Numerous epidemiological reports link foods of animal origin as vehicles of this human disease. Raw or undercooked chickens recurrently have been implicated in outbreaks as have been unpasteurized milks. Other animal food products such as hamburger and pork have been implicated in epidemiological studies, suggesting the transmission of the organism from food to man. *Arcobacter* spp., specifically *A. butzleri*, have been associated with enteritis in humans and nonhuman primates. The isolation of *Arcobacter* spp. from meat, poultry and water has increased awareness of this organism as a potential food-safety concern. *Arcobacter* species are similar morphologically to *Campylobacter* species but differ particularly in being able to grow in air than microaerobically and at 15°C, which is lower than the temperatures used for the incubation of *Campylobacter* spp.

Objective
This study has been undertaken to provide information on the current status of the contamination of pathogenic bacteria such as thermophilic *Campylobacter* and *Arcobacter* spp. in food in Italy, and in particular to determine the incidence of these microrganisms in fresh poultry and pork meat samples destined for consumption.

Methods
Sampling:
104 samples of poultry meat and 76 samples of pork meat have been taken from commerce in various sale points in Parma, in the period of April-December 2000. The samples of approximately 200-400 g were prepackaged and represented various preparations and/or anatomical cuts.
The entire edible portion was taken and grounded under aseptic conditions, then 25 g were used for the determination of *Campylobacter* and 25 g for the determination of *Arcobacter*.

Isolation:
Campylobacter: 225 ml of Bolton broth (Oxoid CM983 with supplement SR183) was added to the 25 g sample in a sterile bottle. The bottles were placed for 4h at 37°C and then 18h at 42°C under microaerophilic conditions.
After incubation, surface plating was performed on m-CCDA agar (Oxoid) and Karmali agar (Oxoid). The two media were incubated at 37°C in microaerophilic atmosphere, by using Campy Gen (Oxoid) and were observed after 48h.
Arcobacter: 225 ml of *Arcobacter* broth (Oxoid) with C.A.T. supplement (Oxoid) were added to the 25 g sample in a sterile bottle. The bottles were placed for 24 h at 30°C under aerophilic condition. After incubation, surface plating was performed on Karmali agar (Oxoid), and CCDA agar with C.A.T supplement (Oxoid). The two media were incubated at 30°C in aerobic condition and growth was observed after 48h.
Identification:
Criteria for presumptive identification were based upon a translucent colonial appearance and microscopical examination for curved to spiral-shaped bacterial rods, showing typical darting corkscrew-like motility. For *Campylobacter* presumptive colonies the Dryspot *Campylobacter* Test (Oxoid) was also used. Up to 5 colonies were selected for confirmation. Colonies were streaked onto Columbia Blood Agar (bioMérieux, France) and incubated at 37°C for 24-48 h for *Campylobacter* spp. and at 30°C for 24-48 h for *Arcobacter* spp. Colonies on Columbia were confirmed by Gram-stain, catalase and oxidase tests and API CAMPY (bioMérieux).

Results and discussion
A total of 180 poultry and meat samples were examined, of which 20.5 % were positive for *Campylobacter* species and 13.9% were positive for *Arcobacter* species (Table 1). Isolation rates varied between sources, with poultry being the most frequently contaminated by *Campylobacter* (33.6 % of samples) compared to pork (2.6 %).

Sample type	N° samples analysed	N° of positive samples			
		Campylobacter		*Arcobacter*	
		Number	%	Number	%
Poultry	104	35	33.6	18	17.3
Pork	76	2	2.6	7	9.2
Total meat	**180**	**37**	**20.5**	**25**	**13.9**

Table 1. Incidence of *Campylobacter* spp. and *Arcobacter* spp. in food samples collected in the Parma area

Sample type	N° samples analysed	N° of positive samples			
		Campylobacter		*Arcobacter*	
		Number	%	Number	%
Shoulder	30	0	0	1	3.3
Minced	25	1	3.3	5	20
Meat and bone	10	0	0	0	0
Bacon	9	0	0	1	11.1
Pigskin	2	1	50	0	0
Total meat	**76**	**2**	**2.6**	**7**	**9.2**

Table 2. Incidence of *Campylobacter* and *Arcobacter* spp. in pork meat.

Sample type	N° samples analysed	N° of positive samples			
		Campylobacter		*Arcobacter*	
		Number	%	Number	%
Chicken	68	27	39.7	15	22.1
Turkey	24	3	12.5	1	11.1
Guinea-fowl	9	4	33.3	0	0
Duck	3	1	12.5	2	8.3
Total meat	**104**	**35**	**33.6**	**18**	**17.3**

Table 3. Incidence of *Campylobacter* and *Arcobacter* spp. in poultry meat

In pork, a higher frequency of contamination was found for *Arcobacter* than for thermophilic *Campylobacter*. In particular minced meat (ground and sausages) proved the highest percentage of isolation for *Arcobacter* (20 %) followed by bacon samples (11.1 %). The only two samples positive for *Campylobacter* were one shoulder and one pigskin (Table 2).
Chicken meat and guinea-fowl were particularly contaminated both by *Campylobacter* and *Arcobacter*, although the percentage regarding chicken was lower than that observed in other studies (Table 3).
The prevalence of individual *Campylobacter* species also varied according to meat type (Table 4).

Source	*Campylobacter* species	N° samples positive for each species
Poultry	*C. jejuni jejuni*	17
	C. coli	12
	C. fetus fetus	2
	C. hyontestinalis	1
Pork meat	*C. coli*	2

Table 4. Distribution of *Campylobacter* species between sample type

None sample was positive for more than one species. The most frequent isolation from chicken samples was *C. jejuni jejuni* (60 %) followed by *C. coli* (18.5 %), while all the *Campylobacter* isolated from guinea-fowl samples were *C. coli*. *C. fetus fetus* were isolated from the duck sample and from a single sample of chicken. Three strains were not identified by API CAMPY kit.
Only *C. coli* was isolated from pork samples.
As regard to *Arcobacter* spp., the identification system API CAMPY has allowed the identification of the single species *A. cryaerophilus*, which is the only profile included in the API index.

Conclusions

Our data confirm that chicken samples, and poultry in general, appear to be prominent reservoirs of *Campylobacter* and *Arcobacter* spp. Although thermophilic *Campylobacter* are microorganisms which grow under microaerophilic conditions, they succeed in surviving in retail packages, where the CO_2 concentration is relatively high.
Implementation of strict hygiene rules in slaughterhouses and in butcher shops or supermarkets, prevention of cross-contamination after processing are required to reduce the chance of foodborne poisoning by these significant pathogens.

References

- Atabay, H.I., Corry, J.E.L., On, S.L.W. Diversity and prevalence of *Arcobacter* spp. in broiler chickens. *J. Appl. Microbiol.*, *84*, 1007-1016 (1998).
- Gill, C.O., Harris, L.M. Hamburgers and broiler chickens as potential sources of human *Campylobacter* enteritis. *J. Food Protect.*, *47*, 96-99 (1984).
- Korsak, N., Daube, G., Ghafir,Y., Chahed, A., Jolly, S., Vindevogel, G. An efficient sampling technique used to detect four foodborne pathogens on pork and beef carcasses in nine Belgian abattoirs. *J. Food Protect.*, *61*, 535-541 (1998).
- Mafu, A. A., Higgins, R., Nadeau, M., Cousineau, G. The incidence of *Salmonella*, *Campylobacter*, and *Yersinia enterocolitica* in swine carcasses and the slaughterhouse environment. *J. Food Protect.*, *52*, 9, 642-645 (1989).
- Mulder, R.W.A.W. Impact of transport and related stesses on the incidence and extent of pathogens in pigmeat and poultry. *J. Food Nutrit. Press*, *15*, 239-246 (1995).
- Tokumaru, M., Konuma, H., Umesako, M., Konno, S., Shinagawa, K. Rates of detection of *Salmonella* and *Campylobacter* in meats in response to the sample size and the infection level of each species. *Int. J. Food Microbiol.*, *13*, 41-46 (1990)
- Zanetti, F., Varoli, O., Stampi, S., De Luca, G. Prevalence of thermophilic *Campylobacter* and *Arcobacter butzleri* in food of animal origin. *Int. J. Food Microbiol.*, *33*, 315-321 (1996).