International Journal of Food Microbiology 166 (2013) 309–315

Contents lists available at ScienceDirect

International Journal of Food Microbiology

journal homepage: www.elsevier.com/locate/ijfoodmicro

An investigation of the immediate and storage effects of chemical treatments on *Campylobacter* and sensory characteristics of poultry meat

H. Meredith [a,b], D. Walsh [a], D.A. McDowell [b], D.J. Bolton [a,*]

[a] Teagasc Food Research Centre, Ashtown, Dublin, Ireland
[b] Food Microbiology Research Unit, School of Health and Life Sciences, University of Ulster, Jordanstown, Co. Antrim, Northern Ireland, UK

ARTICLE INFO

Article history:
Received 11 January 2013
Received in revised form 11 April 2013
Accepted 7 July 2013
Available online 12 July 2013

Keywords:
Chemical decontamination
Poultry
Campylobacter
Sensory analysis
'In-plant' studies

ABSTRACT

Campylobacteriosis is the most common foodborne bacterial infection in developed countries and many cases are associated with poultry. This study investigated the immediate and storage effect of dipping inoculated poultry skin samples in trisodium phosphate (TSP, 10 & 14%, w/v), lactic acid (LA, 1 & 5%, v/v), citric acid (CA, 1 & 5%, w/v), peroxyacids (POA, 100 & 200 ppm) and acidified sodium chlorite (ASC, 500 & 1200 ppm). Spray application was also tested using the higher concentrations in the laboratory. In a broiler processing plant the efficacy of using TSP (14%) and CA (5%) applied by immersion and spray was investigated using naturally contaminated carcasses and the effect of these treatments on the sensory attributes of a skin-on (drumstick) and skin-off (fillet) raw and cooked product was assessed using descriptive sensory analysis. In the laboratory, immersion in TSP (14%), LA (5%), CA (5%) and ASC (1200 ppm) significantly ($P < 0.05$) reduced the *Campylobacter* counts and a 2.5 to 3 $\log_{10}$ cfu/cm^2 reduction was observed within the shelf-life (3–5 days) of poultry meat. Spraying was ineffective even after storage. In the broiler processing plant, immersion in TSP (14%) or CA (5%) achieved *Campylobacter* reductions of 2.49 and 1.44 $\log_{10}$ cfu/cm^2, respectively. There were no significant differences between the treatments for any of the attributes measured in either raw or cooked drumsticks. The 'colour' of raw chicken fillets treated with both TSP (14%, w/v) and CA (5%, w/v) was significantly ($P \leq 0.05$) lighter than that of control samples. The 'intensity of chicken odour' and the perception of 'salt' in cooked chicken fillets treated with CA (5%, w/v) were also significantly ($P \leq 0.05$) higher than that of either control or TSP (14%, w/v) treated samples. It was concluded that TSP (14%) or CA (5%) could be applied to significantly reduce *Campylobacter* contamination of broilers without adversely affecting the sensory quality of the product.

1. Introduction

In Europe and elsewhere, more bacterial foodborne infections are caused by *Campylobacter* than any other bacterial agents (EFSA, 2011). Most infections with this organism are associated with the consumption of poultry meat and poultry meat products which have been contaminated during production or processing activities (Whyte et al., 2003). Many broiler flocks are infected with *Campylobacter* by the third or fourth week of rearing (Patriarchi et al., 2009). These bacteria colonise the ceca and are subsequently shed into the production environment at concentrations of up to 10^7 per gram of faeces, facilitating rapid spread throughout the flock.

The nature and practices involved in current chicken slaughter processes mean that it is not possible to prevent cross contamination of poultry carcasses with fecal matters from the feathers or alimentary tract of processed birds (Sampers et al., 2008). Cross contamination can also occur between processed flocks, with slaughter-line operations such as scalding, plucking and evisceration frequently transferring *Campylobacter* from contaminated birds to subsequently processed clean carcasses at rates in excess of 80% (EFSA, 2008).

While most cases of human campylobacteriosis manifest as mild and self-limiting gastroenteritis, a minority of individuals may suffer a range of more serious systemic effects including Guillain-Barré syndrome, a chronic and potentially fatal form of paralysis (EFSA, 2011). In addition to such individual impacts in terms of human suffering, campylobacteriosis is a major cost to the economies of developed countries in terms of lost working days and medical treatment. The development and application of effective means for reducing the incidence and counts of *Campylobacter* on poultry carcasses is both a key public health objective, and of considerable economic importance to the poultry industry. *Campylobacter* control at the farm stage is heavily reliant on stringent biosecurity and there is considerable evidence that adequate levels of biosecurity can be difficult to achieve and sustain (Newell et al., 2011).

Research in relation to the development of effective control initiatives has therefore moved along the poultry chain and into the slaughter house, at which stage a number of risk assessment studies have suggested that chemical treatments during slaughter and processing

* Corresponding author at: Teagasc Food Research Centre, Ashtown, Dublin 15, Ireland. Tel.: +353 1 805 9539.
E-mail address: declan.bolton@teagasc.ie (D.J. Bolton).

0168-1605/$ – see front matter © 2013 Elsevier B.V. All rights reserved.
http://dx.doi.org/10.1016/j.ijfoodmicro.2013.07.005

may be able to significantly reduce the risk posed to consumers by *Campylobacter* on chicken carcasses (Havelaar et al., 2005; Lindqvist and Lindblad, 2008; Rosenquist et al., 2003). As discussed by Loretz et al. (2010) these include decontamination technologies based on organic acids, chlorine and phosphates.

Despite the universality of the public health threat posed by *Campylobacter*, different jurisdictions have different views and legislation in relation to the application of chemical decontamination technologies. Chemical decontamination of poultry meat has been permitted within poultry processing in the USA, but not in the EU. The legal framework for the use of substances, other than potable water, for microbial decontamination of animal products in the European Union has been in place since 2004, however, no chemical decontamination products have yet been approved for the decontamination of poultry (Loretz et al., 2010). Approval has been inhibited by insufficient data in 4 key areas: [1] the potential beneficial effect of chemical decontamination during chilled storage; [2] the lack of studies where the treated poultry is rinsed (the EU will require the chemical be washed off shortly after application); [3] the dearth of studies using naturally contaminated product in commercial broiler processing plants and [4] the absence of data on the effect of chemical treatments on the sensory attributes of the product (EFSA, 2011). However, the continuing and increasing problems posed by *Campylobacter* within the EU (EFSA, 2012) and recent EFSA approvals for the use of lactic acid in beef (EFSA, 2006, 2011) and trisodium phosphate in poultry processing (EFSA 2005) have renewed interest in the validation and application of technologies for the chemical decontamination of poultry within the EU.

The objectives of this study was therefore to: [1] investigate the immediate and storage related effects of dipping and spraying (followed by rinsing in water) with trisodium phosphate (TSP), lactic acid (LA), citric acid (CA), peroxyacetic acid (POA) and acidified sodium chlorite (ASC) on *Campylobacter* numbers on inoculated poultry samples; [2] apply and investigate the most effective laboratory treatments in a commercial poultry processing plant; and [3] identify and investigate any sensory impacts of the most effective laboratory treatments on chicken meat products.

2. Materials and methods

2.1. Preparation of Campylobacter inoculum

Five poultry isolates (3 *Campylobacter jejuni* and 2 *Campylobacter coli*) from the Teagasc Food Research Centre (Ashtown) culture collection were used in this study. Cultures were produced by incubating a Protect© bead (Lab M, Lancashire, UK) of each isolate in 30 ml of Hunts broth (Nutrient broth (Oxoid, Basingstoke, UK) and yeast extract (Oxoid, Basingstoke, UK), 5% lysed horse blood and 0.4% *Campylobacter* growth supplement FBP (Hunt, 1992)) and incubating at 42 °C for 48 h in microaerobic (5% O_2, 10% CO_2 and 85% N_2) conditions using gas generating kits (Biomerieux, Marcy l'Etoile, France). Samples (1 ml) from each of the resultant suspensions were inoculated into 100 ml volumes of Hunts broth and incubated under microaerobic conditions at 42 °C for 24 h. Cells were recovered by centrifugation (10 min at 2655 g), washed three times with 9 ml maximum recovery diluent (MRD) (Oxoid Basingstoke, UK), resuspended in 50 ml MRD, combined and diluted to 500 ml in MRD to form an initial inoculum. This combined *Campylobacter* inoculum (approximately 7.5 $\log_{10}$ cfu/ml) was used immediately.

2.2. Chemical solution preparation

All chemical solutions were prepared in sterile distilled water (SDW) as follows; tri-sodium phosphate (TSP, VWR International) to 10 and 14% (w/v); lactic acid (LA, Sigma Aldrich) to 1 and 5% (v/v); citric acid (CA, Sigma Aldrich) to 1 and 5% (w/v); peroxyacids (POA, Ecolab, Bray, Ireland) to 100 and 200 ppm (v/v) and acidified sodium chlorite (ASC, Sigma Aldrich) to 500 and 1200 ppm. All dilutions were stored in 500 ml volumes at 20 °C and used within 24 h.

2.3. Sample inoculation

Skin samples (approximately 5 × 5 × 1 cm^2) were aseptically excising from the breast of full retail chickens previously tested to ensure they were *Campylobacter* negative. These were immediately immersed in the *Campylobacter* cocktail inoculum for 1 min and stored at room temperature for 30 min in a laminar flow cabinet to allow for bacterial attachment.

2.4. Chemical treatment — dipping

Inoculated chicken skin samples (n = 216) prepared as described above, were distributed in 12 sets (each containing 18 samples). One set of samples was left untreated (untreated control). Each of the other (11/12) sets of samples was treated (both sides) by immersion for 15 s in 500 ml volumes of 10% or 14% TSP; 1 or 5% LA; 1 or 5% CA; 100 or 200 ppm POA; 500 or 1200 ppm ASC or SDW. The chemical solution was not refreshed during the dipping process. Each set of samples was subsequently immersed in SDW for 15 s and allowed to drain (the SDW was changed after each treatment). Samples (3 from each control or treatment) were tested for surviving *Campylobacter* cells immediately and after storage aerobically in covered foil trays at 4 °C for 1, 3, 5, 10 and 15 days. All experiments were repeated on three separate occasions.

2.5. Chemical treatment — spraying

Chicken skin samples (n = 126), prepared and inoculated as described above, were distributed into seven sets (each containing 18 samples). One set of samples was left untreated. Each of the other (6/7) sets of samples was mist sprayed (both sides) with approximately 3 ml of solution using a spray bottle, on all surfaces, from a distance of 15 cm for a total of 15 s, with sterile solutions of 14% TSP, 5% LA, 5%, CA, 200 ppm POA, 1200 ppm ASC or SDW. Each set of samples was then immersed in SDW for 15 s and allowed to drain. Samples were tested for surviving *Campylobacter* cells immediately and after storage at 4 °C for 1, 3, 5, 10 and 15 days. All experiments were repeated on 3 separate occasions.

To enumerate *Campylobacter* cells, all skin samples were diluted with 90 ml MRD, pulsified (Pulsifier, Microgen Bioproducts) for 15 s, serially diluted in MRD and plated onto *Campylobacter* blood-free agar base (modified charcoal cefoperazone deoxycholate agar [mCCDA] Oxoid, Basingstoke, UK) and incubated at 42 °C for 48 h under microaerobic conditions.

2.6. 'In-plant' validation

This part of the study was carried out in a commercial broiler slaughter plant, processing approximately 11,700 birds per day. During production, 90 carcasses were removed from the processing line after evisceration. Each carcass was swabbed on the left hand side using a pre-moistened (MRD) sterile cellulose acetate sponge. The carcasses were then randomly allocated to five groups [1 to 5], each containing 18 carcass samples. Group 1 carcasses were treated with sterile distilled water applied as a spray and immediately immersed in a bucket of sterile distilled water at 20 °C for approximately 2 s. Three of the group 1 carcasses were immediately tested for *Campylobacter* by swabbing the right hand side and processing the samples as described below. The remaining 15 treated carcasses were stored at 4 °C and 3 carcasses were tested after each of 1, 3, 5, 10 and 15 days. Group 2 and 3 carcasses were treated with TSP (14%, w/v) applied as a dip and as a spray, respectively, dipped in sterile distilled water for 15 s and tested using the same schedule as group 1 birds. Group 4 and 5 carcasses were treated with CA (5%, v/v)