



**FICHA TÉCNICA DE PROYECTOS**

<b>Nombre del proyecto:</b>	Estudio de Prevalencia y Resistencia a los antibacterianos de <i>Campylobacter</i> spp., <i>Escherichia coli</i> BLEE y <i>Salmonella</i> spp. en muestras gastrointestinales de pollos faenados en camales industriales de la Provincia de Pichincha-Ecuador.
<b>Investigador (es) principal (es)</b>	<b>Christian Vinueza</b>
<b>Investigador (es) asociado (s)</b>	<b>María Belén Cevallos, Marco Cisneros</b>
<b>Organismo Financiador</b>	<b>SENPLADES</b>
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<b>Fecha de Inicio:</b>	<b>01 de Julio de 2014</b>
<b>Fecha de Finalización:</b>	<b>06 de junio de 2017</b>
<b>Instituciones participantes:</b>	<ul style="list-style-type: none"><li>• <b>UNIETAR. Facultad de Medicina Veterinaria y Zootecnia. Universidad Central del Ecuador.</b></li></ul>
<b>Códigos:</b>	<b>DGI: 43</b> <b>Código Único del Proyecto (CUP): 9175.0000.0000.377771</b>
<b>Objetivos generales:</b>	Determinar la prevalencia y la resistencia a los antibacterianos de <i>Campylobacter</i> spp., <i>Escherichia coli</i> BLEE y <i>Salmonella</i> spp. en pollos faenados en camales industriales en la Provincia de Pichincha.
<b>Resumen Ejecutivo:</b>	<p><b>Salmonella</b></p> <p>Caeca content from 388 at random selected broiler batches were collected in 6 slaughterhouses during 1 year and analyzed by the ISO 6579/Amd1 protocol for the isolation for <i>Salmonella</i>. Isolates were serotyped and genotypic variation was acceded by pulsed field gel electrophoresis. MIC values for sulfamethoxazole, gentamicin, ciprofloxacin, ampicillin, cefotaxime, ceftazidime, tetracycline, streptomycin, trimethoprim, chloramphenicol, colistin, florfenicol, kanamycin and nalidixic acid were obtained. Presence of bla<sub>CTX-M</sub>, bla<sub>TEM</sub>, bla<sub>SHV</sub> and bla<sub>CMY</sub>; and <i>mcr-1</i> plasmid genes was investigated in resistant strains to cefotaxime and colistin respectively. Prevalence at batch level was 16.0%. The most common serotype was S. Infantis (83.9%) followed by S. Enteritidis (14.5%) and S. Corvallis (1.6%).</p> <p>The pulsed field gel electrophoresis analysis showed that S. Corvallis, S. Enteritidis and S. Infantis isolates belonged to 1, 2 and 12 genotypes respectively. S. Infantis isolates showed high resistance rates to 12 antibiotics ranging from 57.7%(kanamycin) up to 98.1% (nalidixic acid and sulfamethoxazole). All S. Enteritidis isolates showed resistance to colistin.</p> <p>High multiresistant patterns were found for all the serotypes. The bla<sub>CTX-M</sub> gene was present in 33 S. Infantis isolates while <i>mcr-1</i> was negative in 10 colistin resistant isolates.</p> <p><b>Escherichia coli</b></p>

	<p>Sampling was performed from June 2013 to July 2014 in 6 slaughterhouses that slaughter broilers from 115 farms totaling 384 flocks. Each sample of collected caeca was streaked onto TBX agar supplemented with cefotaxime (3 mg/l). In total, 176 isolates were analyzed for AR patterns by the disk diffusion method and for bla<sub>CTX-M</sub>, bla<sub>TEM</sub>, bla<sub>CMY</sub>, bla<sub>SHV</sub>, bla<sub>KPC</sub>, and mcr-1 by PCR and sequencing. ESBL and AmpC <i>E. coli</i> were found in 362 flocks (94.3%) from 112 farms (97.4%). We found that 98.3% of the cefotaxime-resistant isolates were multi-resistant to antibiotics. Low resistance was observed for ertapenem and nitrofurantoin. The most prevalent ESBL genes were the ones belonging to the bla<sub>CTX-M</sub> group (90.9%), specifically the bla<sub>CTX-M-65</sub>, bla<sub>CTX-M-55</sub> and bla<sub>CTX-M-3</sub> alleles. Most of the AmpC strains presented the bla<sub>CMY-2</sub> gene. Three isolates showed the mcr-1 gene.</p> <p><b><i>Campylobacter</i></b></p> <p>Caecal content from 379 randomly selected broiler batches originating from 115 farms were collected from 6 slaughterhouses located in the province of Pichincha during 1 year. Microbiological isolation was performed by direct plating on mCCDA agar. Identification of <i>Campylobacter</i> species was done by PCR. Minimum inhibitory concentration (MIC) values for gentamicin, ciprofloxacin, nalidixic acid, tetracycline, streptomycin, and erythromycin were obtained.</p> <p>Genetic variation was assessed by RFLP-<i>flaA</i> typing and Multilocus Sequence Typing (MLST) of selected isolates. Prevalence at batch level was 64.1%. Of the positive batches 68.7% were positive for <i>C. coli</i>, 18.9% for <i>C. jejuni</i>, and 12.4% for <i>C. coli</i> and <i>C. jejuni</i>. Resistance rates above 67% were shown for tetracycline, ciprofloxacin, and nalidixic acid. The resistance pattern tetracycline, ciprofloxacin, and nalidixic acid was the dominant one in both <i>Campylobacter</i> species. RFLP-<i>flaA</i> typing analysis showed that <i>C. coli</i> and <i>C. jejuni</i> strains belonged to 38 and 26 profiles respectively. On the other hand MLST typing revealed that <i>C. coli</i> except one strain belonged to CC-828, while <i>C. jejuni</i> except 2 strains belonged to 12 assigned clonal complexes (CCs). Furthermore 4 new sequence types (STs) for both species were described, whereby 2 new STs for <i>C. coli</i> were based on new allele sequences.</p>
<p><b>Productos:</b></p>	<p><b>Artículos científicos</b></p> <ul style="list-style-type: none"> <li>• Vinueza-Burgos C, Cevallos M, Ron-Garrido L, Bertrand S, De Zutter L (2016). <b>Prevalence and Diversity of <i>Salmonella</i> Serotypes in Ecuadorian Broilers at Slaughter Age.</b> PLoS ONE 11(7): e0159567. DOI:10.1371/journal.pone.0159567</li> <li>• Vinueza-Burgos C. <b><i>Salmonella</i> and <i>Campylobacter</i> in broilers at slaughter age: a possible source for carcasses contamination in Ecuador.</b> (2017) Ghent University Press. DOI: 10.13140/RG.2.2.20687.48803</li> <li>• Christian Vinueza-Burgos, Magali Wautier, Delphine Martiny, Marco Cisneros, Inge Van Damme, Lieven De Zutter; <b>Prevalence, antimicrobial resistance and genetic diversity of <i>Campylobacter coli</i> and <i>Campylobacter jejuni</i> in Ecuadorian broilers at slaughter age.</b> Poultry Science 2017 pew487. DOI: 10.3382/ps/pew487</li> <li>• Christian Vinueza-Burgos, Lieven De Zutter; <b>2P4 - Important foodborne bacteria and their antimicrobial resistances in Ecuadorian poultry.</b> Special Issue: Abstracts of the 10th European Congress on Tropical Medicine and International Health, 16-20 October 2017, Antwerp, Belgium October 2017 Volume 22, Issue Supplement S1, Page 144. DOI: 10.1111/tmi.12979</li> </ul>

- Vinueza-Burgos C, Cevallos M, Cineros M, Van-Damme M y De Zutter L (2018). **Quantification of the *Campylobacter* contamination on broiler carcasses during the slaughter of *Campylobacter* positive flocks in semi-industrialized slaughterhouses.** International Journal of Food Microbiology 269 (2018) 75-79. DOI:10.1016/j.ijfoodmicro.2018.01.021
- Vinueza-Burgos C, Ortega-Paredes D, Narváez C, De Zutter L, Zurita J (2019) **Characterization of cefotaxime resistant *Escherichia coli* isolated from broiler farms in Ecuador.** PLoS ONE 14(4): e0207567 DOI: 10.1371/journal.pone.0207567

#### Tesis

- Galárraga A. (2014). **Aislamiento y tipificación molecular de *Campylobacter jejuni* y *Campylobacter coli* en contenido cecal de pollos faenados en camales industriales en la provincia de Pichincha.**
- Poma V. (2014). **Aislamiento e identificación de *Campylobacter* spp. en el tracto gastrointestinal de pollos faenados en mataderos industriales de la provincia de Pichincha.**
- Valseca J. (2015). **Aislamiento e identificación de *Salmonella* spp. móviles en el contenido cecal de aves faenadas en plantas de procesamiento industrial ubicadas en la provincia de Pichincha.**
- Vásconez D. (2015). **Aislamiento e identificación de *Escherichia coli* BLEE en ciegos de pollos faenados en camales industriales en la provincia de Pichincha.**
- Larco N. (2015). **Aislamiento de cepas móviles e inmóviles de *Salmonella* spp. en contenido cecal de pollos faenados en camales industriales de la provincia de Pichincha.**
- Pérez E. (2015). **Cuantificación de *Escherichia coli* productor de  $\beta$ -lactamasas de espectro extendido (BLEE) en puntos críticos de control en camales industriales de la Provincia de Pichincha.**
- Falcón J. (2015). **Cuantificación de *Campylobacter* spp. en cinco puntos críticos de control de dos camales industriales de pollos de la provincia de Pichincha.**
- Medina J. (2015). **Cuantificación de *Campylobacter* spp. en un matadero semi-industrial de Aves y posterior identificación de especies (*C. jejuni* y *C. coli*) mediante técnicas de diagnóstico molecular.**
- Narváez C. (2015). **Identificación de genes de resistencia a antibióticos B-lactámicos en cepas BLEE y AmpC de *Escherichia coli* de origen Aviar.**
- De Janon D. (2016). **Determinación fenotípica de cepas de *Escherichia coli* resistente a betalactámicos, por la técnica de doble disco, en pollos faenados en seis camales industriales de la provincia de Pichincha.**
- Paguanquiza J. (2016). **Genotipificación del gen Flagelar A en *Campylobacter jejuni* provenientes de los procesos de faenamiento industrial de pollos broiler.**
- Chacha S. (2016). **Identificación de genes de resistencia en *Salmonella* a betalactámicos en pollos (Tesis de maestría).**
- Vinueza C. (2017). ***Salmonella* and *Campylobacter* in broilers at slaughter age: a possible source for carcasses contamination in Ecuador (Tesis de doctorado).**

#### Posters

- Vinueza-Burgos, C., Narváez-Manosalvas, C., Barba, P., Ortega-Paredes, D., Zurita J. (2017). **Intensive poultry production: an important hot-spot of multi-**

	<p><b>resistant <i>Escherichia coli</i> in Ecuador.</b> 27th European Congress of Clinical Microbiology and Infectious Diseases. Vienna, Austria.</p> <ul style="list-style-type: none"><li>• Vinueza Christian., Cevallos María., Cisneros Marco., De Zutter L. (2015). <b><i>Campylobacter</i> contamination of carcasses during the slaughter of broilers in Ecuador.</b> 18th International Workshop of <i>Campylobacter, Helicobacter</i> &amp; Related Organisms (CHRO). Rotorua, New Zealand.</li><li>• Vinueza Christian., Cevallos María., Cisneros Marco., De Zutter L. (2015). <b>Prevalence of <i>Campylobacter</i> and their antimicrobial resistance in broilers at slaughter in Ecuador.</b> 18th International Workshop of <i>Campylobacter, Helicobacter</i> &amp; Related Organisms (CHRO). Rotorua, New Zealand.</li><li>• Vinueza Christian., Cevallos María., Cisneros Marco., De Zutter L. (2015). <b>Prevalence of <i>Campylobacter</i> and <i>Salmonella</i> and their antimicrobial resistance in broilers at slaughter in Ecuador.</b> 20th Conference on Food Microbiology - Belgian Society for Food Microbiology 2015. Brussels, Belgium.</li></ul>
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