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Optimizing the detection of Campylobacter spp. in Ukraine

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Introduction. Campylobacteriosis is the most frequent cause of acute intestinal infections in many countries In Ukraine, 501 confirmed cases of human campylobacteriosis were registered in 2019-2021. However, in one hospital in Dnipro alone, 235 cases were confirmed during the same period. This suggests that the real number of yearly campylobacteriosis cases in Ukraine is much higher. According to the Ukrainian agencies, no cases of animal or poultry campylobacteriosis have been reported in the last 10 years. This suggests incorrect diagnosis and weak surveillance. To improve surveillance of Campylobacter in animals in Ukraine, we optimized the isolation of Campylobacter according to the DSTU ISO 10272-1:2007 standard. The study aimed to compare the detection of Campylobacter in broiler chickens using 1) a direct inoculation method and 2) an enrichment method

Methods. DSTU ISO 10272-1:2007 was used for enrichment and detection of Campylobacter, and ISO 10272-1:2017(E) for direct culture and detection. Nutrient media and selective additives manufactured by HiMedia Laboratories (India) were used. A Thermo Scientific Heracell VIOS 160i CO2 incubator was used to create a microaerobic atmosphere. The samples were collected from blind appendages taken from 20 broiler chickens obtained from a slaughterhouse.

Results. We optimized 2 different approaches for Campylobacter isolation. For the enrichment protocol, samples were placed in Bolton's broth enrichment medium and supplemented with 5% lysed horse blood and antibiotic mixture (FD231). Antibiotics (FD042) were used to overcome contamination. Samples were cultivated in a CO2 incubator at 41.5°C for 44 hours. Samples were then plated on bloodless mCCD agar medium with cefoperazone and in parallel on Preston agar containing 5% lysed horse blood and a mixture of antibiotics (FD042). Characteristic colonies were noted. The morphology of Campylobacter was confirmed microscopically.
For the direct plating protocol, samples were plated directly on mCCD agar with cefoperazone and in parallel on Campylobacter Agar medium with 5% lysed horse blood with antibiotics (FD008), and incubated in a CO2 incubator for 44 hours at a temperature of 41.5°C. After cultivation on both media, samples showed characteristic growth. On mCCD agar, we identified round wet colonies of gray color with a metallic sheen. On Campylobacter agar, we identified wet flat homogeneous neoplasms with a tendency to spread.
Microscopic analysis and MALDI-TOF confirmed the identification of four isolates of Campylobacter jejuni and one isolate of Campylobacter coli.
The results show that direct plating is more advantageous as it is cheaper and faster.

Conclusions. We obtained 4 isolates of Campylobacter jejuni and 1 Campylobacter coli. We believe that in Ukraine: 1) the importance of Campylobacter underestimated; 2) clinical and diagnostic training of specialists is needed; 3) improved isolation methods are needed; 4) Campylobacter surveillance is needed; and 5) regulation is needed. We also recommend a direct seeding approach for culturing Ukraine.