IDENTIFICATION OF *Escherichia coli* FROM CHICKEN SAMPLES USING CULTURE-BASED METHOD AND BIOINFORMATIVE TOOLS FOR *hlyF* GENE

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This Final Year Project Report entitled "**Identification of *Escherichia coli* from Chicken Samples Using Culture-Based Method and Bioinformative Tools for *hlyF* Gene"** was submitted by Puteri Imein Sufia binti Mohd Adly in partial fulfilment of the requirements for the Degree of Bachelor of Science (Hons.) Biology, in the Faculty of Applied Sciences, and was approved by

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ABSTRACT

IDENTIFICATION OF *Escherichia coli* USING CULTURE-BASED METHOD AND BIOINFORMATIVE TOOLS FOR *hlyF* GENE

Colibacillosis is a disease found in chickens that houses the zoonotic avian pathogenic *Escherichia coli* (APEC). Unhealthy chickens will exhibit signs such as no appetite, more tired and the anus being dark red with mostly white or yellow faeces. This is a serious issue that is arising in the poultry industry because it affects the well-beings of chicken consumers. This study aims to isolate APEC from unhealthy-living chicken samples using culture-based method, design specific primers of *hlyF* gene from genomic sequence of APEC and amplify the gene using *in-silico* PCR. Therefore, this study was involved sampling 30 unhealthy-living chickens via cloacal swabbing. Culture-based method was done to identify the presence of *E. coli* in the sample. The test includes culture of differential agar of MacConkey, malonate test to differentiate among Enterobacteriaceae strains and Gram staining for morphology identification. A total of 19 positive samples were obtained and further kept in 20% glycerol stock for upcoming use. For the molecular based detection, DNA extraction was performed on the 19 positive samples. The gene of interest, *hlyF*, was assessed using bioinformative tools such as BLASTn, FASTA, ClustalW and *in-silico* PCR. A set of primers were design according to Primer3 with a size of 228 bp and contained 40% and 45% of GC content for both forward and reverse primers respectively. *In-silico* PCR was used to test the capability of the primers to amplify specifically to the target gene of *hlyF*. In conclusion, this study managed to identify 19 positive samples infected by APEC and successfully designed primers for virulent gene *hlyF*. Completion of this study will benefit the poultry industry by minimizing the chances of APEC infections by producing a rapid detection in order to control infection, thus preventing economy loss and recession.

Keywords: *E. coli*, *hlyF* gene, APEC, chicken, poultry, zoonotic, primers, PCR, gel electrophoresis, bioinformative tools

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