



# Enteric bacteria as safety markers in Nile fishes in relation to public health

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## List of Abbreviations

<b>APC</b>	Aerobic Plate Count
<b>APHA</b>	American Public Health Association
<b>CDC</b>	<b>Center for Disease Control and Prevention</b>
<i>E.coli</i>	<i>Escherichia coli</i>
<b>EHEC</b>	Enterohemorrhagic <i>Escherichia coli</i>
<b>EIEC</b>	Enteroinvasive <i>Escherichia coli</i>
<b>EPEC</b>	Enteropathogenic <i>Escherichia coli</i>
<b>ETEC</b>	Enterotoxigenic <i>Escherichia coli</i>
<b>FAO</b>	Food and Agriculture Organization of the United Nations
<b>FDA</b>	Food and Drug Administration
<b>FSIS</b>	Food Safety and Inspection service
<b>HACCP</b>	Hazard Analysis Critical Control Point
<b>ICMSF</b>	International commission of Microbiological Specification for Foods
<b>ISO</b>	International Organization for Standardization
<b>NEA</b>	National Environment Agency
<i>S.aureus</i>	<i>Staphylococcus aureus</i>
<b>USFDA</b>	United StateDepartment of Health and Human Services Food and Drug Administration Center for Food Safety and Applied Nutrition
<b>WHO</b>	World Health Organization

## **7. SUMMARY**

Fish is the best animal protein due to its high nutritive value as well as its high quality. Also fish is rich in calcium, phosphorous and generous supply of B-complex vitamins as well as its lower content of cholesterol.

Fish flesh is generally sterile immediately after catching; however it may become contaminated with different microorganisms during subsequent handling as these microorganisms can penetrate from skin and the gut to the flesh. *Salmonella* act as a public health hazard if present, where it cause food poisoning. Also, *E. coli* may give rise to severe diarrhea in infants and young children as well as food poisoning and gastroenteritis among adult consumers.

This study was carried out for isolation and identification of *E.coli*, *Salmonella* and other *Enterobacteriaceae* from Fresh water fish.

A total of 150 random samples of Fresh water fish such as *Tilapia nilotica*, *Bagrus bayad* and *Clarias gariepinus* (50 of each) were collected from different Markets and farms in Kalyobia Governorate.

The samples were placed separately in clean sterile plastic bags and transferred in an insulated ice box to the laboratory under complete aseptic conditions without any delay. All collected samples were subjected to bacteriological examination.

The bacteriological examination of *Tilapia nilotica* revealed that, the mean value of APC, Enterobacteriaceae, Coliform and *Staphylococcus aureus* counts were  $8.32 \times 10^5 \pm 0.48 \times 10^5$ ,  $2.65 \times 10^3 \pm 0.16 \times 10^3$ ,  $1.81 \times 10^3 \pm 0.13 \times 10^3$  and  $1.24 \times 10^3 \pm 0.16 \times 10^3$  respectively. While the mean value of APC, Enterobacteriaceae, Coliform and



Staphylococcus aureus counts for Claries gariepinus were  $9.74 \times 10^5 \pm 0.42 \times 10^5$ ,  $3.25 \times 10^3 \pm 0.14 \times 10^3$ ,  $2.35 \times 10^3 \pm 0.15 \times 10^3$  and  $1.67 \times 10^3 \pm 0.17 \times 10^3$  respectively. Meanwhile for Bagrus bayad the mean value of APC, Enterobacteriaceae, Coliform and Staphylococcus aureus counts were  $6.54 \times 10^5 \pm 0.30 \times 10^5$ ,  $2.07 \times 10^3 \pm 0.13 \times 10^2$ ,  $1.74 \times 10^3 \pm 0.13 \times 10^2$  and  $0.71 \times 10^3 \pm 0.01 \times 10^3$  respectively.

Regarding to the bacterial species isolated, the results appeared that, 22 isolates of *E.coli* were isolated from examined Nile fish represented as 7(14%) from *Tilapia nilotica* with serotypes O27:H2 (8%), O63:H2 (2 %) & O158:H4 (4%) , 9(18%) from Claries gariepinus with serotypes O27:H2(8%) ,O63:H2(2%),O158:H4(6%)& O159:H7(2%) and 6(12%) from Bagrus bayad with serotypes O27:H2 (6%), O63:H2 (2 %) & O158:H4 (4%). Also, 23 isolates of coagulase positive *S.aureus* were isolated from examined Nile fish represented as 7 (14%) from *Tilapia nilotica* ; 9(18%) Claries gariepinus, and 7(14%) from Bagrus bayad samples. The results of *Y. enterocolitica* isolation revealed that, 7 isolates of *Y. enterocolitica* were isolated from examined Nile fish represented as 2(4%) *Tilapia nilotica* and 5(10%) from Claries gariepinus samples. Meanwhile, all examined samples of Bagrus bayad fish were free from *Y. enterocolitica*. Meanwhile, the present study failed to detect Salmonella serovars from examined *Tilapia nilotica* and Bagrus bayad samples but represented as 3(6%) from Claries gariepinus with serotypes Salmonella Essen, Salmonella Saint Paul & Salmonella Enteritidis. The most important *Enterobacteriaceae* isolated from fresh water fish are *Citrobacter diversus*, *Citrobacter freundii*, *Enterobacter aerogenes*, *Enterobacter agglomerans*, *Enterobacter cloacae*, *Klebsiella azanae*, *Klebsiella pneumoniae*, *Proteus mirabilis*, *proteus vulgaris*.

Also, these results cleared that, the unaccepted samples are +ve for the *E. coli* isolates as the agarose gel electrophoresis of multiple PCR gave +ve results of *stx1* (180 bp), *stx2* (255 bp) and *eaeA* (384 bp) genes for characterization of *Enteropathogenic E. coli*.

The incidence of virulence genes of Enteropathogenic *E. coli* isolated from the examined samples of fresh cleared that, the Shiga-toxin 1 gene (*STx1*), observed in the *E. coli* isolates of O158 : H4 and O63 : H2. While, the Shiga-toxin 2 gene (*STx2*), observed in the *E. coli* isolates of O27 : H2, O63 : H2 and O159 : H7 . While, the Intimin gene (*EaeA*), only in *E. coli* isolates of O159 : H7.

Agarose gel electrophoresis of multiplex PCR gave +ve results of *inv* (570 bp) and *ystA* (145 bp) genes for characterization of *Yersinia enterocolitica* , The results of PCR amplifications of different used genes of *Y. enterocolitica* strains cleared that, *inv* gene observed in 5 isolates. While, *ystA* gene observed in 7 isolates *Y. enterocolitica* strain . Finally, The public health significance of contaminants and the possible sources of contamination of Nile fish with these organisms as well as suggestive hygienic measures to improve the quality of such fish were discussed.