IMPACT OF COVID-19 ON THE MICROBIAL LOAD OF MUD CRAB, *SCYLLA SERRATA*: A STUDY FROM SOUTH 24 PARGANAS DISTRICT OF WEST BENGAL, INDIA

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Abstract

This study was conducted to evaluate the microbial load in the flesh and gills of the mud crab (*Scylla serrata*) collected from the Nagendra Bazar area in Diamond Harbour in South 24 Parganas district of West Bengal, India. The period of collection coincided with the Pre-COVID-19 phase (June 2018 and June 2019) and the COVID lockdown phase (June 2020 and June 2021). It is observed that the total coliform loads in both muscle and gills are more in the pre-COVID phase compared to the COVID lockdown phase. ANOVA analysis also confirmed significant variations between years and body parts (muscle and gills) for both Total coliform (TC) and Fecal Coliform (FC) (p < 0.01). The present study is of considerable importance as crabs are consumed by local people as well as exported in several foreign countries like China, Singapore, Japan, and the U.S.A.

Keywords: Mud crab (*Scylla Serrata*), microbial load, Total Coliform (TC), Fecal Coliform (FC)

I. Introduction

Mud crabs, *Scylla Serrata* (Portunidae) are large portunids with significant commercial value owing to their edibility (Fig. 1).
It is an edible, commercially important, and valuable arthropod species that are abundant in the brackish water ecosystem of deltaic Sundarbans (I). Since the early 1980s, mud crabs have emerged as an important food in India because of their high commercial values and since then the export of mud crabs to other countries has received great momentum. The species occurs abundantly in the intertidal mudflats of mangrove ecosystem and can easily be grown in less saline water and hence culture water has considerable influence on the microbial flora of the mud crabs. Mud crab farming is carried out in ponds connected with the brackish water of the estuaries in Indian Sundarbans. Recently mud crabs have emerged as a potential export commodity and their farming is increasing rapidly in the maritime states of Odisha and West Bengal because of high demand in the international markets. Crab farming is, therefore, an emerging aquaculture sector with tremendous economic potential.

In this context, it is important to maintain the quality of crab not only for local consumption but also from the export point of view. The industrial waste and other discharges from domestic units in the water bodies like estuaries boost up the growth, nourishment, and sustenance of many microorganisms. Certain crab farms are in the periphery of the sewage canals, which are highly susceptible to the problem of fecal contamination, in many developing countries where the discharge of sewage is not always properly controlled.

The shapes of the Coliform bacteria are tubular and they are Gram-negative in nature. Coliform groups are commonly found in the fecal matter of warm-blooded animals. They are also widely distributed in the aquatic and terrestrial environment. While coliform bacteria themselves do not normally cause any serious illness of high degree like lethality, but their presence in drinking water or food items indicates that disease-causing organisms (pathogens) of fecal origin could be present in the system. The entire coliform population is referred to as total coliform out of which a subset

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represents the fecal coliforms. The presence of total coliform corresponds to environmental contamination whereas fecal coliform corresponds to fecal contamination. Despite the increased production and the demand of mud crabs for their value for export, little attention has been given to the microbiological quality of crab.

On this background the present study was carried out with two main objectives namely:

a. Monitor the Total Coliform (TC) and Fecal Coliform (FC) in both muscle and gills as an assurance of its quality.

b. Observe the impact of the COVID-19 lockdown phase on the microbial load of mud crabs, as during this phase estuarine water was relatively upgraded due to complete closure of anthropogenic activities, industrial discharge, and release of the market wastes.

The estuarine environment has a great influence on the microbial load of mud crabs as the water required for crab farming and aquaculture is sourced from the adjacent estuaries (II), (III), (IV), (V). Mud crabs acquire a large number of microbes in their body parts from water, sediments, and food. The increased demand for mud crab production and their significant contribution of the species in the export basket of the country have imparted the present research a special status.

II. Materials and Methods

Sampling of the Crab

Healthy crabs were collected from the Nagendra Bazar area in Diamond Harbor in South 24 Parganas district of West Bengal (Fig. 2) and were scrubbed, rinsed with distilled water (several times) and the meat and gills were aseptically extracted using a sterile knife as described in APHA (VI). Further microbiological analysis was carried out in the laboratory with the flesh and gill samples.

Fig 2. Map showing the collection site (Nagendra Bazar) at Diamond Harbour

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Determination of total coliform, fecal coliform from crab tissue:

Assessment of total coliform and fecal coliform was carried out using LTB (Lauryl Tryptose Broth) as per the standard MPN (Most Probable Number) procedure (VI). Briefly, 10 ml of $10^{-3}$ dilution was added separately in a test tube containing 10 ml volume of single strength LTB broth. In the next step, incubation was carried out at $35\pm 0.5^\circ C$ for 24 hrs with the total sets, and the growth and gas production were carefully observed. Based on our observation MPN was enumerated and expressed through the standard unit of presumptive coliform MPN/100gm. Then the positive culture was inoculated into Brilliant Green Lactose Bile Broth and the tubes were incubated at $35\pm0.5^\circ C$ for 24 hrs and examined for the presence of growth with gas production. The MPN of total coliform (TC) was calculated and the results were expressed as confirmed coliform MPN/100gm. To enumerate the fecal coliform (FC) inocula from the 24 hrs positive presumptive tubes were aseptically transferred to tubes of EC medium. These tubes were incubated at $44\pm0.5^\circ C$ for 24 hrs and examined for the presence of growth with gas production. Results were expressed as fecal coliform MPN/100gm.

III. Results and Discussions

It is observed that mud crabs were collected from Nagendra Bazar in the South 24 Parganas district of West Bengal, India during the pre-COVID phase (June 2018 and June 2019) and the COVID lockdown phase (June 2020 and June 2021) were significantly different in terms of microbial load. However, in both the phase total coliform and the fecal coliform loads were relatively high in the gills compared to the muscles. Gills, being the respiratory and filtering organs of aquatic crustaceans are continuously exposed to the ambient environment, which may be the reason for the presence of high microbial loads in the organ.

The high microbial load during the pre-COVID phase in both muscle (7800 MPN/100gm in June 2018 and 8400 MPN/100gm in June 2019) and gills (10500 MPN/100gm in June 2018 and 13300 MPN/100gm in June 2019) compared to COVID lockdown phase (3700MPN/100gm in June 2020 and 4600 MPN/100gm in June 2021 in muscle and 5900 MPN/100gm in June 2020 and 7100 MPN/100gm in June 2021 in gills) may be due to closure of several anthropogenic activities during COVID lockdown phase (Fig. 3 and 4).

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Fig 3. Total coliform load (MPN/100gm) in the muscle and gills of mud crab (*Scylla serrata*) collected from Diamond Harbour area along the Hooghly estuary.

Fig 4. Fecal coliform load (MPN/100gm) in the muscle and gills of mud crab (*Scylla serrata*) collected from the Diamond Harbour area along the Hooghly estuary.

In context to the present locality, these anthropogenic activities include the release of the wastes from the market places, fish landing activities, plying of fishing and passenger vessels, operation of recreational and tourism units, etc. The complete closure of these activities was in accordance with the COVID-19 lockdown rules to contain the spreading of the disease, which might be the reason for the low microbial load in the crab tissue during this phase that is primarily sourced from the ambient aquatic phase.

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ANOVA results carried out considering the data sets reveal significant variations (p < 0.05) between years and body parts that strongly recommend the role of the COVID-19 lockdown phase in upgrading the tissue quality of aquatic organisms in terms of microbial load (Table 1 and 2).

Table 1: ANOVA results based on the TC data of Scylla serrata

<table>
<thead>
<tr>
<th>Source of Variation</th>
<th>SS</th>
<th>df</th>
<th>MS</th>
<th>Fcalculated</th>
<th>P-value</th>
<th>Fcrit</th>
</tr>
</thead>
<tbody>
<tr>
<td>Between Years</td>
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<td>1590125</td>
<td>20.8883</td>
<td>0.01635</td>
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</tr>
<tr>
<td>Between Body parts</td>
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<td>1891125</td>
<td>24.8424</td>
<td>0.01553</td>
<td>10.128</td>
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<tr>
<td>Error</td>
<td>2283750</td>
<td>3</td>
<td>761250</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Total</td>
<td>68898750</td>
<td>7</td>
<td></td>
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<td></td>
</tr>
</tbody>
</table>

Table 2: ANOVA results based on the FC data of Scylla serrata

<table>
<thead>
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<th>Source of Variation</th>
<th>SS</th>
<th>df</th>
<th>MS</th>
<th>Fcalculated</th>
<th>P-value</th>
<th>Fcrit</th>
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<tbody>
<tr>
<td>Between Years</td>
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<td>7506146</td>
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<td>9.276628</td>
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<tr>
<td>Between Body parts</td>
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<td>8302813</td>
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<tr>
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<td>Total</td>
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</tbody>
</table>

IV. Conclusion

COVID-19 is a deadly contagious disease caused by a virus that has the potential of changing its genetic configuration. All the nations of the world have enacted travel bans and put major cities and towns under strict lockdown to slow down the spread of coronavirus. This has resulted in the complete closures of several anthropogenic activities due to which the environmental quality has been upgraded. The aquatic environment is no exception to this and the organisms thriving in this aquatic phase also showed better health in relatively pollution-free water. Mud crabs collected from Nagendra Bazar in Diamond Harbour exhibited significantly low microbial load in the muscle and gills during the COVID-19 lockdown phase (June 2020 and 2021) compared to the pre-COVID phase (June 2018 and 2019) as revealed from ANOVA and thus lead us to conclude the production and yield of a better quality of edible crab for consumption during the lockdown period.

Conflicts of Interest:

The authors declare that they have no conflicts of interest regarding the paper.

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References


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