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ASSESSING THE ECOLOGICAL AND MICROBIAL IMPACTS OF SHIP BALLAST WATER DISCHARGE IN GLOBAL SHIPPING

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ABSTRACT

The global shipping industry, responsible for 90% of world trade, plays a pivotal role in facilitating economic growth. However, its impact on marine environments, notably through pollution from ballast water discharges, poses environmental challenges. The discharge of ballast water emerges as a significant threat, introducing non-native species and pathogens into new ecosystems, leading to ecological and economic consequences. The study aims to understand the impact of these invasive species on native ecosystems, biodiversity, ecosystem function, and potential economic implications. Ten vessels from diverse regions were sampled, revealing zero total coliform counts and *Escheria coli* presence, indicating relatively clean ballast water. Microbiological analysis highlighted variations in heterotrophic counts across samples with zero total coliform counts and *Escheria coli*. The study identified the presence of pathogenic bacteria, including Bacillus sp., *Vibro cholera*, and Staphylococcus aureus. The study highlights the potential for the spread of invasive species through ballast water discharge, necessitating strategies to mitigate negative impacts.

Keywords: Invasive species, Ecological risk, Human health risk, Maritime transport

INTRODUCTION

The role of the shipping industry in facilitating global trade is indisputable. As per data provided by the International Maritime Organization (IMO), maritime transportation accounts for 90% of world trade, transporting over 10 billion tons of containers, solid, and liquid bulk cargo annually (IMO, 2017; Walker, 2016). These goods range from essential items such as food, beverages, and clothing to larger items like home appliances and electronics. The efficient and reliable transportation provided by the shipping industry is crucial to global trade, which in turn, spurs economic growth and development. However, maritime commerce has played a role in the emergence of pollution in the marine environment, with approximately 20% of global emissions are attributed to the ten major Asian ports, including Shanghai, Shenzhen, Hong Kong in China, and Busan in South Korea (Wan et al. 2016).

The discharge of ballast water from ships specifically poses a significant threat to the marine environment (Walker et al. 2019). Ballast water is used by ships to maintain stability and balance while at sea, but when released in a new location, it can introduce non-native species and pathogens into the ecosystem, leading to ecological and economic consequences (Iswantoro et al. 2023). For example is the introduction of the zebra mussel through ballast water discharge in the Great Lakes (Roberts, 1990). The zebra mussel, originally from the Caspian Sea, was inadvertently transported to the Great Lakes through ballast water discharge in the 1980s. This invasive species quickly spread throughout the region, outcompeting native species and causing significant ecological damage. The zebra mussels attached themselves to various surfaces, including water intake pipes, boat hulls, and native mussel shells, leading to clogged pipes, reduced water flow, and disrupted ecosystems.

Approximately 12 billion metric tons of ballast water transport an estimated 15,000 species each year (Wright, 2021). This immense volume highlights the potential for the introduction and spread of invasive species through ballast water discharge. Invasive species, which are transferred outside of their natural habitats and transported to new areas where they do not typically appear, can become established and threaten the original ecosystem and its species (Morissette et al. 2023; O'Hara et al. 2023). The introduction of invasive species into marine ecosystems through activities such as discharging ballast water is a global concern (Bailey, 2015). These invasive species can disrupt ecosystem functionality and put marine biodiversity at risk. The impacts of invasive species on native populations and food webs can be significant, leading to changes in species biomass and composition. The untreated ballast water can lead to the spread of invasive species, which can have negative effects on the marine ecosystem, biodiversity, fisheries, aquaculture, and public health (Ivče et al. 2022; Walker et al. 2019). Therefore, it is crucial to identify and quantify the proliferation of invasive species that are released into new environments when ballast water is discharged, in order to develop strategies to mitigate the negative impacts of ballast water invasions. The study aims to understand the impact of these invasive species on native ecosystems, including changes in biodiversity, ecosystem function, and potential economic implications.

MATERIALS AND METHODS

Study Area

Ten (10) different vessels labelled Sample A to J were selected for this study (Figure 1). Sample A- Lome, Sample B- Atlantic, Sample C- Philippine, Sample D- Switzerland, Sample E- Senegal, Sample F- Indian, Sample G- Cameroon, Sample H- Lagos anchorage, Sample I- Escravos River Nigeria, Sample J- South Africa

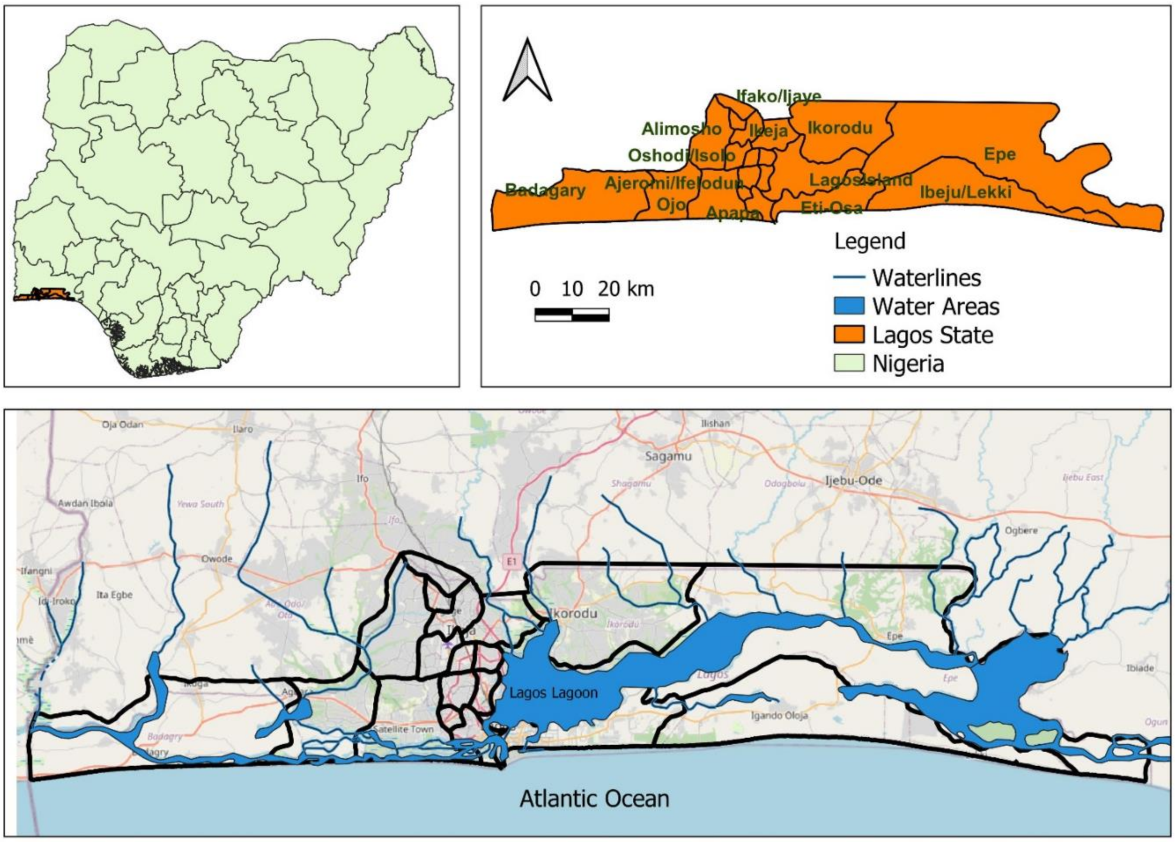

Figure 1: Map showing the countries from which ballast water samples were collected

Sample collection

Ten (10) samples of ship ballast water were collected from 12th -22nd December, 2023. from10 different vessels through an open manhole from a single tank per vessel. The ten water samples from conveyor vessels were collected in a 2 litres sterilized polythene bottles each and transported in ice blocks to the Department of Microbiology laboratory, University of Ibadan, Ibadan Oyo state for microbiological analysis.

Determination of Microbiological analysis of Ballast water samples

The study involved conducting a microbiological analysis on samples of ship ballast water to investigate the presence of marine species in the maritime environment. The analysis utilized the standard microbiological plate method to determine various parameters, including total heterotrophic counts (THC), total coliform count, total Staphylococcal count (TSC), halophilic bacterial count (HBC), *Escherichia coli*, and Vibrio count. Additionally, the most probable number (MPN) technique was employed for assessing total and faecal coliform counts in the water samples.

Determination of Total Coliform Count and Fecal Coliform Count

The examination of bacteriological features in water samples involved the application of the Most Probable Number (Multiple Tube Fermentation Method) for the enumeration of both total coliform and faecal coliform counts. Lauryl Tryptose Broth (LTB) and fermentation tubes (Durham tube) were utilized in this process (Bukar et al. 2015). The water samples to be tested underwent a serial dilution and were then incubated at 35°C for a duration of 20-24 hours to determine the total coliform count. Tubes with positive results were subsequently transferred to Brilliant Green Lactose Bile Broth and incubated for 48 hours at 35°C. Following a 24-hour incubation period, tubes exhibiting gas production were moved to the confirmation experiment, and the quantification of coliforms was determined using established statistical tables (APHA, 1998).

Heterotrophic Plate Count

A series of dilutions for water samples was created using sterile distilled water, employing the 10th dilution. Aseptically, 1 ml of the water sample was transferred to the center of a sterile petri dish containing the appropriate medium. The water sample was evenly spread using a sterilized glass rod, and the medium was then incubated at 37˚C for 24 hours, following the methodology outlined by APHA (1998) and used by Bukar et al. (2015).

RESULTS AND DISCUSSION

The microbiological analysis of ship ballast water samples collected from various vessels belonging to different countries indicated that all ten samples analysed exhibited zero total coliform counts (TCC) and no presence of *Escheria coli* (*E. coli*) bacteria. The heterotrophic counts (THC) varied from 0.3 to 23.7 (103 cfu/mL), with Sample J showing the highest heterotrophic counts and Sample F exhibiting the lowest count. None of the samples, including A, C, E, F, I, and J, showed any Total Staphylococcus count (TSC). However, Samples A and D displayed the lowest TSC value of 1.2 cfu/ml, followed by sample H with a value of 3.5cfu/ml, and sample G showed the highest TSC count of 4.9 cfu/ml (Table 1).

The halophilic bacterial count revealed the lowest count in sample D with a value of 1.6cfu/ml and the highest count in Sample B with a value of 19.4cfu/ml. Samples E and F displayed the lowest Vibrio count of 0.1cfu/ml, followed by

Sample D with a value of 0.3cfu/ml, and the highest Vibrio count was found in Sample A with a value of 2.2 cfu/ml. However, no Vibrio count was detected in Samples B, C, G, H, I, and J. The presence of pathogenic bacteria in the samples suggests that appropriate measures should be taken to prevent the spread of potential diseases.

The result of the ship ballast water samples collected from different vessels of different countries showed that the samples contaminated with pathogenic bacteria which include micro flora in sample A- is *Bacillus* sp., *Vibro cholerae, Staphylococcus aureus;* Sample B- *Bacillus* sp., *S. aureus;* Sample C- *Bacillus* sp. ; Sample D- *Bacillus* sp., *V. cholerae, S. aureus;* Sample E- *Bacillus* sp., *V. cholera;* Sample F- *Bacillus* sp., *V. cholera;* Sample G- *S. aureus*, *Bacillus* sp.; Sample H- *S. aureus, Bacillus* sp.; Sample I- *Bacillus* sp; Sample J- *Bacillus* sp respectively (Table 1). There was no faecal coliform detected in all the ten ship ballast water samples collected from different vessel and total coliform recorded ≤ 2 (MPN/100mL)in all samples (Table 2).

Table 1: Bacterial counts on different culture media (x 10^3cfu/mL)

Sample code	**Sample Location**	**Nature of sample**	**THC**	**TSC**	**HBC**	***E. coli***	**TCC**	**Vibrio count**	**Major microflora obtained**
Sample A	Lome	Water	8.1	1.2	12.8	NG	NG	2.2	*Bacillus* sp., *Vibro cholerae, Staphylococcus aureus*
Sample B	Atlantic	Water	12.3	4.6	19.4	NG	NG	NG	*Bacillus* sp., *S. aureus*
Sample C	Philippine	Water	NG	NG	14.8	NG	NG	NG	*Bacillus* sp.
Sample D	Switzerland	Water	1.6	1.2	1.6	NG	NG	0.3	*Bacillus* sp., *V. cholerae, S. aureus*
Sample E	Senegal	Water	0.9	NG	3.8	NG	NG	0.1	*Bacillus* sp., *V. cholerae*
Sample F	Indian	Water	0.3	NG	1.8	NG	NG	0.1	*Bacillus* sp., *V. cholerae*
Sample G	Cameroon	Water	15.6	4.9	10.2	NG	NG	NG	*Staphylococcus aureus, Bacillus* sp.
Sample H	Lagos anchorage	Water	17.9	3.5	9.5	NG	NG	NG	*Staphylococcus aureus, Bacillus* sp.
Sample I	Nigeria Escravos River	Water	24.2	NG	8.9	NG	NG	NG	*Bacillus* sp.
Sample J	South Africa	Water	23.7	NG	9.2	NG	NG	NG	*Bacillus* sp.

KEY: NG: No growth was observed after the incubation period., THC- Total heterotrophic counts; TSC- total Staphylococcal count; HBC- Halophilic bacterial count; TCC- Total coliform count;

Table 2: Total and Faecal Coliform count of the water samples

Sample code	**Sample Location**	**Nature of sample**	**Total Coliform (MPN/100mL)**	**Faecal Coliform**
A	Lome	Water	≤2	ND
B	Atlantic	Water	≤2	ND
C	Philipine	Water	≤2	ND
D	Switzerland	Water	≤2	ND
E	Senegal	Water	≤2	ND
F	Indian	Water	≤2	ND
G	Cameroon	Water	≤2	ND
H	Lagos anchorage	Water	≤2	ND
I	Nigeria Escravos River	Water	≤2	ND
J	South Africa	Water	≤2	ND

KEY: ND: Not detected.

Faecal coliforms, including E. coli, are significant indicators of faecal contamination and are commonly used to assess the bacteriological quality of water (Lu et al. 2021). The absence of total coliform counts and *E. coli* in all ten samples aligns with the understanding that these bacteria are indicative of faecal contamination. This is a positive result, as the absence of such indicators suggests a lower risk of waterborne diseases and contamination in the marine environment. The presence of E. coli and other indicator organisms indicates the potential for the transfer of aquatic organisms and pathogens through ballast water exchange (Akyala et al. 2014). The recorded low levels of total coliform further emphasize the relatively clean nature of the analysed ballast water.

The variation in heterotrophic counts (THC) across samples is consistent with previous studies that highlight the dynamic nature of heterotrophic bacteria in marine environments (Çiftçi Türetken and Altuğ, 2016). Sample J exhibiting the highest THC and Sample F with the lowest count indicate differences in microbial abundance, possibly influenced by factors such as vessel origin, water source, and environmental conditions (Hassard et al. 2017). The absence of Total Staphylococcus count (TSC) in most samples, except for varying counts in Samples A, D, G, and H, suggests that Staphylococcus may not be prevalent in ballast water or might be present in low concentrations. Halophilic bacterial counts reveal a wide range of variability, with Sample B exhibiting the highest count and Sample D the lowest. This variability in halophilic bacterial counts may be attributed to the different salinity levels in the ballast water samples. It is possible that Sample B came from a region with higher salinity, which favours the growth of halophilic bacteria, whereas Sample D may have originated from a location with lower salinity.

The transport of invasive species through ship ballast water may have severe consequences on ecological, economic, and public health systems (Iswantoro et al. 2023). The primary ecological impacts related to invasive species include predation, parasitism, competition, the introduction of novel pathogens, genetic changes, habitat alterations, species shifts, and loss of biodiversity (Havel et al. 2015; Siddiqui et al. 2021). The global movement of ballast water serves as a means of dispersing long-term human pathogens, thereby contributing to the proliferation of waterborne diseases affecting both humans and other organisms (Sayinli et al. 2022). Various countries around the world, such as Africa, the United States, Australia, and certain European nations, grapple with the issue of exotic species invasions, which can lead to alterations in ecosystem function, nutrient cycles, and reductions in water quality (Christopher and Richard, 2002; Tamelander et al. 2010; Kholdebarin et al. 2020). Some invasive species transported by ballast water also pose a threat to public health by increasing the risk of pathogens and parasitism.

For instance, Martins et al. (1991; 1993) reported the detection of Vibrio cholera in Latin America, resulting in over 1.2 million cases of cholera and 12,000 fatalities. The study collected samples of ballast water from ships and identified the presence of Vibrio cholera, *Bacillus* sp., and *Staphylococcus aureus*, which poses a significant threat to public health. According to Battle et al. (2009), viruses such as *Vibrio cholera* and *Giardia duodenalis* can cause public health issues, with Vibrio cholera being transmitted through contaminated water and seafood consumption.

Different areas with varying numbers of invasive species are at high risk, as per published studies (Díaz-de-León and Díaz-Mondragón, 2013; Early et al. 2016; McGeoch et al. 2016). Nonetheless, Díaz-de-León and Díaz-Mondragón (2013) found different invasive species in ship ballast water, including Zebra mussel, Chinese mitten crab, European green crab, Round goby, Comb jelly, North Pacific sea star, and Red mysid shrimp, which were not present in the current study's collected ballast water samples.

The results of this study concur with those of Takahashi et al. (2008), which investigated the presence of *V. cholera*, *Staphylococcus aureus*, and fecal coliforms in nine Brazilian ports. The discrepancy in findings suggests that the composition of invasive species and microbial contaminants in ship ballast water may vary across different regions and time periods. Therefore, further research is necessary to elucidate the factors that influence the presence and distribution of these organisms and to develop effective strategies for prevention and control.

CONCLUSION

The Critical role of the shipping industry in global trade cannot be overstated, contributing significantly to economic growth and development. However, this study highlights the potential environmental risks associated with the discharge of ballast water, a crucial aspect of maritime commerce. The microbiological analysis of ballast water samples from ten different vessels revealed a diverse microbial landscape. Notably, the absence of total coliform counts and Escherichia coli in all samples aligns with the aim of mitigating waterborne diseases and contamination.

The study highlights a critical concern due to the presence of pathogenic bacteria, including *Bacillus* sp., *Vibrio cholera*, and *Staphylococcus aureus* in the ballast water samples. This poses a significant threat to marine ecosystems, public health, and economic activities. Invasive species, once introduced, can disrupt ecosystem functionality, alter biodiversity, and have economic implications. The study emphasizes the global concern surrounding the impact of invasive species on marine ecosystems, fisheries, aquaculture, and public health. The results suggest a need for robust strategies to identify, quantify, and mitigate the proliferation of invasive species through ballast water discharge.

It is imperative to continue research efforts to understand the factors influencing the presence and distribution of invasive species and pathogens in ballast water. Developing effective prevention and control strategies is crucial to protect marine ecosystems, biodiversity, and human health.

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