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Assessment of risk management strategies among poultry farmers in Anambra State, Nigeria

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Abstract

The greatest threat to profitability and sustainability of the poultry enterprise is risk. Identification and assessment of risk mitigation level is important for policy formulation. Therefore, this study assessed risk management strategies among poultry farmers in Anambra State, Nigeria. A two-stage sampling technique that purposively selected six Local Government Areas: Ogbaru, Ihiala, Awka South, Anambra West, Anaocha and Idemili North based on the State Agricultural Development Program information on poultry farmers' concentration. Twenty poultry farmers were then selected randomly from each LGA. A structured questionnaire was used to elicit information from farmers. Data collected were socio-economic characteristics, risks encountered, and the management strategies used. Data were analyzed using descriptive (mean, standard-deviation and bar-chart) and inferential statistics, Multinomial logit model at 0.05 level. The categories of risk identified with the level of exposure were production/technological and health risks (100%), market/price risk (81.2%), institutional risk (55.2%), financial/credit risk (94.2%), environmental risk (67.2%) and human and personnel risk (59.1%). The categorization of farmer by their risk management strategies showed: biosecurity strategies (4.32±0.22), production strategies (4.22±0.43), mitigation/risk reduction strategies (3.30±0.68), other risks coping strategies (2.42±0.89), transfer measures/institutional strategies (1.05±0.30), marketing strategies (3.98±0.71), and financial strategies (2.4±0.87). Capital invested ($p<0.001$), cooperative society ($p<0.001$), disease outbreaks ($p<0.001$), livestock insurance ($p<0.001$), and government policy($p<0.001$), were the major determinants of the risk management level. The study recommended adequate funding, insurance policy and training to minimize risk.

Keywords: Poultry risk, Management strategies, Assessment, Multinomial logistic regression

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1. Introduction

Eliminating hunger by 2030 in the continent of Africa seems to be a mirage. Malnutrition has risen from 17.6 percent in 2014 to 19.1 percent in 2019 (FAO, 2019) and is still increasing. Challenges of food insecurity and hunger in Nigeria have remained a critical subject for consideration by stakeholders (Ejikeme et al., 2017; Osabohien et al., 2020). It is well-documented that Nigeria has a gap in its protein consumption (Nigerian Protein Deficiency Report, 2020 as cited in Daily Trust, 2021; Komolafe et al., 2023). By the targeted year 2030 for the elimination of hunger, this problem is bound to escalate if adequate measures to deal with the situation are not put in place.

Poultry production which is eyed to be the way out of the problem is cumbered with numerous risks and uncertainty such as theft, flood, fire outbreaks, and other unpredictable and unplanned events (Eleri et al., 2012). The contribution of poultry to animal proteins consumed globally was about 30% (AGRA, 2014). Globally poultry population is about

16.2 billion, 71.6% of which were in developing nations, producing 67,718,544 tons of meat and 57,861,747 tons of eggs (Gueye, 1998; Komolafe et al., 2023). The Nigerian poultry firms contribute to the economy by providing employment, income and quality protein thereby, reduces food security and alleviating poverty (Nasiru et al., 2012). Poultry contribution to Nigeria's agricultural GDP is above 25% (Oduntan, 2016) as sited in National Daily Newspaper, 2016. Poultry products (meat and egg) offer considerable potential for bridging the nutritional gap due to the adaptability capacity of high-yielding exotic birds to Nigerian weather and the production technology is simple with high returns on investment (Yusuf et al., 2016). The contributions of poultry to Nigeria's GDP could have been greater than what it is at present provided the risks associated with the sector could be reduced to the barest minimum.

The poultry industry in Nigeria has suffered losses in the past which has affected poultry farmers as well as consumers (Ogoke, 2009). Little improvement was recorded recently in the sector, but despite this improvement, local production only meets 30 percent of the demand for poultry products. A high percentage of Nigerian poultry farmers are less equipped to mitigate risks associated with production and income and this could lead to the eventual collapse of the industry if collaborative efforts are not made by the government and stakeholders to salvage the situation (Adepoju et al., 2013). Inadequate and appropriate measures in this regard will lead to a reduction in poultry production and protein intake of people, malnutrition, ill health and lower productivity and output, and poor welfare of the farmers (Bamiro et al., 2009; Obike et al., 2017). The risks involved in the poultry business need to be addressed to be able to manage and increase the productivity of the sector to meet the demand of the consumers. It is also important to assess the risks and uncertainty surrounding one of the most important livestock sectors (poultry) with the aim of understanding and finding better ways of improving the enterprise. Some farmers do not have the understanding of risks and uncertainties as well as risk management skills or approach required to manage problems and reduce the consequences of risk and uncertainties.

The concept of risks and uncertainties are related consequently, they are used interchangeably, since there is no risk without some level of uncertainty and vice-visa. Risk entails uncertainty in the occurrence of possible damage, loss or injury, that agricultural investment will yield the expected outcome or not, while uncertainty is that there is the availability of more than one possible outcome to a course of action and the form of each possible outcome is unknown (Aimin, 2010). The danger of risk and uncertainties can result in impressive loss of money, psychological displacement, and complete business failure; thus risk management becomes imperative (Adeyemo & Onikoyi, 2012). Producers' attitude toward risk is jamming in input allocation decisions, and hence in output supply. The degree of uncertainty, the consequences of the various possible outcomes and the personality of the individuals determine the behaviour of investors under the circumstance and the choice of strategies to be adopted to minimize the effects of risks. Most farmers are risk averse, this implies that a farmer will withdraw from a production plan with the highest expected profit if it is associated with a wide range of alternative profit outcomes; he may instead opt for a lower expected profit with a narrower range of profit outcomes (Ullah et al., 2015). Risk-averse decision rules differ from risk neutrality due to the existence of marginal risk premium, which is the wedge between input cost and expected marginal product at the optimum level of input use (Ramaswami, 1993; Russo et al., 2022).

Risk-averse farmers are the most cautious risk-takers, but they do take some risks. They lose because they miss economic opportunities for profit. Risk-neutral farmers understand they must take some chances to get ahead but recognize that there are degrees of risk in every situation. Before making a decision or taking action they gather information and analyze the odds. They try to be realistic and recognize the risks and try to reduce risks to acceptable levels. Risk lovers are individuals who enjoy risks as challenging and exciting and look for the chance to take risks. Many farmers may be in this category concerning their marketing plans. As long as financial survival is not at stake, they may enjoy the adventure of playing the market. Many speculators are in this category. Some close their eyes to risk, ignore facts, and go ahead and commonly fail because they refuse to take precautions. The sources and types of risk and uncertainty in poultry are numerous and diverse, the industry is bedeviled with multiple constraints such as outbreaks of diseases, continuous increase in the cost of inputs, inadequate market, continuous rise in the price of feed, substandard day-old chicks, weather fluctuation, inadequate veterinary services, transportation problems among others (Obike et al., 2017). Livestock farming risks are classified into: Production risk (drought, heavy rainfall and diseases and pests); Marketing risk (supply/cost of inputs, demand for a product/price and cost of production); Financial risk (loan and its cost); Institutional risk (change in policy at the local, national and international levels) and Personal/human risk (accidents, illness, civil unrest and death) (Kahan, 2008). In poultry farming, farmers express their risk mitigation in diverse ways, some of which are forward pricing, production practices, insurance, holding liquid reserves, diversification, and liability management (Adnan et al., 2020). Generally, these ways of risk mitigation are at a cost too high for most poultry farmers that are mostly smallholders with little or no opportunity for diversification and insurance.

Risk management in poultry farming is expedient, failure of which the farmer bears the brunt of low income, market instability and food insecurity (Lamine, 2018). Risk management strategies in livestock can be categorized majorly into three: Preventive strategies (reducing the probability of an adverse event); Mitigation strategies (reducing the potential impact of an adverse event); and Coping strategies (relieving the impact of the risky event after occurrence). Risk managing decisions begin with identifying. The greatest risks for farmers were understanding the potential impacts and likelihood of desirable outcomes and identifying the possible steps to lessen the impacts and avert failure (Howell & Hazard, 2012). The identification of the sources of risk is key to choosing an appropriate management strategy even at the planning stage of an investment to minimize loss, improve profitability and ensure the sustainability of the business. In developing countries, farmers lack access to adequate risk management policies such as futures contracts or guarantee funds, agricultural insurance and government assistance. Consequently, farmers rely on traditional coping risk mitigation strategies, unfortunately, most of these traditional techniques are inefficient. Adequate risk management in poultry farming will keep debt low, therefore lowering the cost of production and affording farmers good liquidity status (Asogwa et al., 2014). Since risk and uncertainties influence investment decisions, it is, therefore, imperative to assess the risks and uncertainty surrounding poultry enterprise which is one of the most important sectors of livestock with the vision of finding ways to improve the enterprise which this study addressed.

Much work has been done on the analysis of risk in agriculture and poultry production. Some of these works were the work of Vihi et al. (2018) on the analysis of farm risk and coping strategies; Aminu et al. (2019) on-farm risks and management strategies; Akinbile et al. (2013) on risk management strategies utilized by poultry farmers; Adeyonu et al. (2021) on risk perceptions of poultry farmers and the management strategies; Ebong and Awatt (2023) on analysis of risk management in poultry production enterprises and Obike et al. (2017) on risk management and output determinants among poultry farmers. Of all these studies none made mention of the determinants of the level of risk strategies employed by farmers which is the focus of this study.

The objectives of this study to assess the risks management strategies employed by poultry farmers were therefore: (*i*) to identify the risks and uncertainties encountered by the poultry farmers, (*ii*) to identify the risk management strategies employed by the poultry farmers, and (*iii*) to estimate the determinants of the level of risk management strategies.

2. Material and Method

The population of the study was poultry farmers in Anambra state. This study adopted a multistage sampling technique. Purposively selected six Local Government Areas (LGAs): Ogbaru, Ihiala, Awka South, Anambra West, Anaocha and Idemili North based on the State Agricultural Development Program (ADP) list of poultry farmers showing a high concentration of the farmers in the areas. In the second stage, twenty poultry farmers were selected randomly from each LGA; five in each community as shown in Table 1. A structured questionnaire was then used to elicit the required information from a total of one hundred and twenty poultry farmers taking 5 respondents randomly from four communities/towns in each GA. Data collected were socio-economic characteristics, risks encountered, and the risk management strategies employed by the farmers.

Table 1. Sampling technique and sample size

State	L.G.A.	Commun	Towns/Villages		No analyzed
Anambra	Ogbaru	Atani	5		
		Mputu	5		
		Odekpe	5		
		Umuodu	5	Total=20	20
	Ihiala	Ihala	5		
		Mbosi	5		
		Azia	5		
		Iseke	5	Total=20	20
	Awka South	Ifite-awka	5		
		Isiagu	5		
		Nibo	5		
		Nise	5	Total=20	20
	Anambra West	Odekpe	5		
		Oroma-etiti	5		
		Ukalla	5		
		Owelle	5	Total=20	20
	Anaocha	Agulu	5		
		Adazi-enu	5		
		Neni	5		
		Obeledu	5	Total=20	20
	Idemili North	Abatete	5		
		Umuoji	5		
		Nkpor	5		
		Ogidi	5	Total=20	20
				Total	120

2.1. Data analysis

Model specification

Risk management strategies level index:

The risk management strategies level index for poultry farmers was constructed to classify the farmers into different classes of the severity of management strategies levels. This was based on the number of identified management strategies variables that a poultry farmer is exposed to, based on their responses on the questionnaire yes=1 and no=0. The maximum number of actionable steps allowed to state in the questionnaire was thirty-four. A farmer who did not take any action get a score of zero, if 1 action was taken the farmer get a score of 1, and 2 actions get a score of 2 up to score of 34. The composite score was then used to classify farmers to different risk severity levels: high, intermediate and low; classification is done based on: High level: mean plus standard deviation and above, Intermediate level: between upper and lower categories, Low level: between mean minus standard deviation point to zero.

The result was used as the dependent variable (Y_i) for the multinomial logistic regression model.

Ordered probit regression:

Ordered probit regression is a statistical modelling technique. It is used in place of ordinary least squares regression, which assumes a continuous dependent variable, when the dependent variable is ordinal (ordered categories with unequal distances between them (McKelvey & Zavoina, 1975). Ordered probit regression model the probability that an observation is within a specific category. In ordered probit regression it is assumed that there is an underlying continuous latent variable, which determines the observed ordinal outcome. The latent variable can be expressed as: $Y^* = X'\beta + \varepsilon$ where X represents a vector of independent variables and β is a vector of coefficients to be estimated, and ε is a normally distributed error term with mean zero and constant variance (Greene, 2000). The observed ordinal categories Y are determined by threshold values such that:

Y=0 if $Y^* \leq 0$, $\quad Y$=1 if $0 < Y^* \leq \mu_1$, $\quad Y$=2 if $\mu_1 < Y^* \leq \mu_2$

where $\leq \mu_1$ and $\leq \mu_2$ are the cut points (threshold variables in the probit model).

The model is estimated using maximum likelihood estimation which finds parameter values that maximize the probability of observing the given data. The coefficients in an ordered probit model do not directly represent marginal effects but instead indicate the direction and relative strength of the relationship between each independent variable and the latent outcome variable (Greene, 2000). To interpret results meaningfully, researchers often compute marginal effects, which provide the change in the probability of each outcome category for a unit change in an independent variable (Long & Freese, 2014).

The ordered probit regression model;

$Y^* = X'\beta_i + \varepsilon$

Where; Y: the response categories for the level of adaptation or mitigation strategies, β_i: parameters to be estimated, X_i: vectors of socioeconomics, ε: a normally distributed error term with mean of zero and constant variance.

The independent variables were itemized below:
X_1: Age (years), X_2: Sex (*1* if male and *0* if female), X_3: Education (years), X_4: Capital invested, X_5: Years of experience (years), X_6: Flock size (*1* if size is big; > 5000 birds, *0* if size is small; < 5000 birds), X_7: Distance of farm to residence (km), X_8: Access to agricultural extension (*1* if farmer has access and *0* if otherwise), X_9: Membership of cooperative society (*1* member, *0* otherwise), X_{10}: Management system (battery cage system =*1*, deep litter system =*0*), X_{11}: Diseases prevention &treatment, X_{12}: Frequency disease outbreak (X_{12}=*1* if outbreak is frequent and 0 if otherwise), X_{13}: Involvement in livestock insurance (*1* if yes and *0* if otherwise), X_{14}: Credit Constraint (*1* if yes and *0* if otherwise), X_{15}: Fluctuating in prices of products (*1* Fluctuating and *0* if otherwise), X_{16}: Government policy (*1* if favorable and *0* if not).

3. Results and Discussion

From Figure 1 below as classified by the authors seven major risks were identified among the poultry farmers included in the study. These risk categories were production/technological risk, market/price risk, institutional risk, financial/credit risk, environmental risk, human and personnel risk and health risk. Production/technological and health risks (100%) were experienced by all the farmers included in the study. Market/price risk was experienced by 81.2% of the farmers, while institutional risk was experienced by only 55.2%, financial/credit risk (94.2%), environmental risk (67.2%) and human and personnel risk (59.1%). This corroborates the findings of Obike et al. (2017) and Olarinde et al. (2010). The result implies that the majority of farmers were exposed to various categories of risk which makes it very important to analyze and come up with policies that will help farmers reduce exposure to risk.

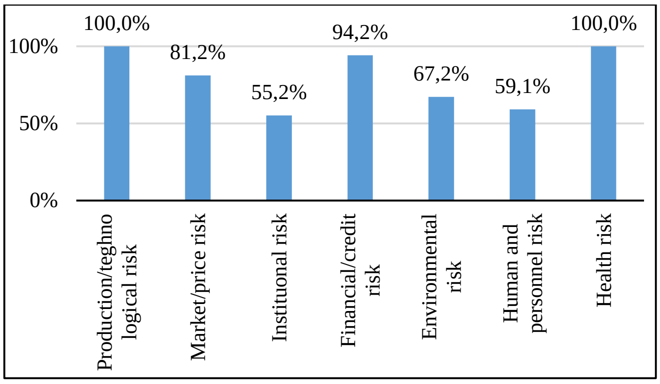

Figure 1. Poultry farmers' distribution by main sources of risks (*Source: Authors' computation from field survey data, 2023*)

Following Akinola (2014), Olarinde et al. (2010) and Obike et al. (2017), the categories of risk identified were decomposition into various components and the mean rating of the components was done in Table 2. The Health Risk was decomposition into eight categories with high cost of medication and vaccines (4.62±0.33) ranking first followed by frequent cases of a disease outbreak (4.39±0.56), then medication and vaccine failure (3.54±0.23). others in this group that were considered severe and of great importance, because they meet the cut-off point of 3, were inadequate veterinary services (3.37±0.22), inadequate space for poultry business expansion (3.34±0.52) and droppings

Table 2. Decomposition of the categories of risk identified

SN	Identified risk	Mean ($\bar{X}$)	Std. Dev.
1. Health Risk	High cost of medication and vaccines	4.62	0.33
	Frequent cases of disease outbreak	4.39	0.56
	Medication and vaccines failure	3.54	0.23
	Inadequate veterinary services	3.37	0.22
	Inadequate space for poultry business expansion	3.34	0.52
	Droppings accumulation	3.26	0.55
	Overcrowding/ Stampeding in poultry	2.48	0.62
	Cannibalism in poultry	2.28	0.32
2. Human and Personnel Risk	Inadequate improved poultry information on management technic	2.64	0.62
	Poor personal management	2.57	0.64
	Ill-health of farmer/worker	1.25	0.22
3. Environmental Risk	Weather variability	3.61	0.34
	Snake attack	3.23	0.42
	Shortage of water	2.94	0.35
	Natural disaster	1.44	0.66
4. Financial/Credit Risk	Credit constraint	4.56	0.48
	High interest rate	4.46	0.62
5. Production/Technological Risk	Inadequate technical know-how	4.12	0.54
	High labour cost	4.10	0.53
	Low output	3.23	0.22
	Poor quality day old chicks	3.01	0.22
	Poor quality feed	2.93	0.75
	Theft and burglary	2.09	0.80
	High mortality rate	2.67	0.67
6. Market/Price Risk	Fragility of poultry products (eggs)	4.37	0.74
	Lack of storage facilities	4.03	0.72
	Unstable price of products in the market	3.99	0.67
	Transportation problems	3.85	0.33
	Rise in cost of inputs	3.64	0.57
	Fluctuation in prices of output	3.32	0.23
	Fluctuations in price of exotic day old chicks	2.99	0.52
	Erratic demand for poultry products	2.77	0.46
	Accident during egg transportation	2.67	0.32
7. Institutional Risk	Low institutional support from government	3.39	0.70
	Erratic power supply	3.92	0.44
	Unfavourable Government policy	2.68	0.62
	Insecurity	2.67	0.64

Source: Authors' computation from field survey data, 2023

accumulation (3.26±0.55). The least in this group is cannibalism in poultry (2.28±0.32). This showed that there are eight major variables identified as health risks and six of them required serious attention. Human and Personnel Risk was decomposed into the second category. The components were just 3 inadequate improved poultry information on management technic (2.64±0.62), poor personal management (2.57±0.64) and ill-health of farmer/worker (1.25±0.22). These results corroborate the findings of Effiong et al. (2014) and Ebong and Awatt (2023). The third category was Environmental Risk with the following components: Weather variability (3.61±0.34), snake attack (3.23±0.42), shortage of water (2.94±0.35) and natural disaster (2.44±0.66). The next category was Financial/ Credit Risk of which credit constraint (4.56±0.48) and high-interest rate (4.46±0.62) were the components. Production/Technological Risk has five components but only four meet the cut-off point of 3. These components were inadequate technical know-how (4.12±0.54), high labour cost (4.10±0.53) output (3.23±0.22), poor quality day-old chicks (3.01±0.22), poor quality feed (2.93± 0.75), theft and burglary (2.09± 0.80) and high mortality rate (2.67±0.67). Market/Price Risk was decomposed into nine components, but only seven were severe. These were fragility of poultry products (eggs) (4.37±0.74), lack of storage facilities (4.03±0.72), unstable price of products in the market (3.99±0.67), transportation problems (3.85±0.33), rise in cost of inputs (3.64±0.57), fluctuation in prices of output (3.32±0.23) and fluctuations in price of exotic day-old chicks (2.99±0.52). Institutional Risks were decomposed into four, but only two were server: Low institutional support from the government (3.39±0.70) and erratic power supply (3.92±0.44).

Table 3 categorized the farmers on the basis of their risk behaviour and it was discovered that 65.83% of the farmers averse risk, 22.50% were risk neutral and only 11.67% were risk takers. This corroborates the findings of Ebong and Awatt (2023).

Table 3. Risk attitude of poultry farmers

Risk	Frequency	Percentage (%)
Risk averse	79	65.83
Risk neutrality	27	22.50
Risk takers/preference	14	11.67
Total	120	100

Source: Authors' computation from field survey data, 2023

Following Olarinde et al. (2010) and Obike et al. (2017) the farmers were categorized by their adoption of risk management strategies as shown in Table 4. The categorization with mean and standard deviation as follows: biosecurity strategies (4.32±0.22), production strategies (4.22±0.43), mitigation/ risk reduction strategies (3.30±0.68), other risks coping strategies (2.42±0.89), transfer measures/institutional strategies (1.05±0.30) marketing strategies (3.98±0.71), and financial strategies (2.4±0.87). Biosecurity strategies have the highest mean follow by production and then marketing strategies, followed by mitigation/risk reduction strategies. All the strategies had a mean value of ≥3 that is the cut-off point except financial strategies, transfer measures/institutional strategies and other risks coping strategies.

In Table 5, the categories of risk management strategies adopted were decomposed as follows. For the Biosecurity strategies category, we have adequate and timely vaccination (94.17%), adequate ventilation of poultry buildings (95.83%), quarantine of sick birds (95.00%), controlled visitors access to poultry buildings (93.33%), proper disinfection pens/cages (85.00%), prevention of rodents/pests (73.33%), adequate fencing and netting (68.33%) and foot dips with effective disinfectant (63.33%). For Production strategies the following were identified: avoid overcrowding (95.83%), separation of birds by species (95.83%), aeparation of birds by age (94.17%), use of wood shaven (82.50%), disease tolerant breed (74.17%), and adequate feed /nutrition in feed (72.50%). For the Mitigation/risk reduction strategies category, source day-old chicks from certified/authorized sellers (96.67%), administration of safe water always (92.50%), proper collection of eggs (73.33%), enterprise diversification (55.83%), feed from certified millers (53.33%), feed producing by self (46.67%), buying input in bulk/advance (43.33%), ensuring constant lightening/power supply (37.50%), adequate/proper record keeping (25.00%). The Coping strategies category has off-farm income (38.33%), adequate storage of input/products (21.67%), training/workshops attendance (20.63%) and cash liquidity for emergencies (17. 50%). Transfer measures/Institutional strategy has only insurance policies (6.67%). Marketing strategies have securing the market in advance (72.50%) and credit sales (69.17%). Financial strategies have loaned from friends and relatives (82.50%), loan from cooperatives (51.67%), advance from buyers (36.67%) and loan from bank/financial institutions (6.67%).

Table 6 presented the result of the ordered probit model that revealed the determinants of the level of risk management strategies. The dependent variable was derived using composite score to order the level of risk management strategies of farmers into low, medium and high as explained in the methodology. Sixteen variables were included in the model, but only five were significant at 1% and 5% The Wald chi2(16) = 74.4, Log likelihood = -194.26222, Prob > chi2 = 0.0000 which revealed a model that is statistically significant at 1%.

The determinants of the level of risk management strategies were explained based on initial categorization:

Biosecurity strategies: Frequency disease outbreak has marginal effect t (0.65) is significant ($p<0.001$). This implies that the likelihood of the level of risk management increase by 65%. This is in line with Robertson (2020) work with high frequencies of disease outbreak will a farmer to be proactive in adopting risk management strategies that aid the likelihood of disease outbreak reduction.

Production strategies: Years of experience (0.26), flock size (0.017) and management system (0.48) both were significant ($p<0.01$). This implies that the likelihood of the level of risk management increase by 26% 1.7% and 0.48 respectively with 10% increase in these variables. Years of formal education marginal effect ($p<0.001$). This implies that with a 5% increase in years of formal education, the likelihood of improving the risk management strategies would increase by 38%.

Mitigation/risk reduction strategies: Farm distance to residence (0.27) was significant ($p<0.01$). This implies that with a 10% increase in years of formal education, the likelihood of improving the risk management strategies would increase by 38%.

Table 4. Categorization of risk management strategies adopted by poultry farmers

Categorization of risk management strategies adopted by poultry farmers	Mean ($\bar{X}$)	Std. Dev.
1. Biosecurity strategies	4.32	0.22
2. Production strategies	4.22	0.43
3. Mitigation/risk reduction strategies	3.30	0.68
4. Other risks coping strategies	2.42	0.89
5. Transfer measures/ Institutional strategies	1.05	0.30
6. Marketing strategies	3.98	0.71
7. Financial strategies	2.43	0.87

Source: Authors' computation from field survey data, 2023

Table 5. Decomposition of categories of risk management strategies adopted by poultry farmers

Strategies employed		Frequency	Percentage (%)
1. Biosecurity strategies	Adequate and timely vaccination	113	94.17
	Adequate ventilation of poultry buildings	115	95.83
	Quarantine of sick birds	114	95.00
	Controlled visitors access to poultry buildings	112	93.33
	Proper disinfection pens/cages	102	85.00
	Prevention of rodents/pests	88	73.33
	Adequate fencing and netting	82	68.33
	Foot dips with effective disinfectant	76	63.33
2. Production strategies	Avoid overcrowding	115	95.83
	Separation of birds by species	115	95.83
	Separation of birds by age	113	94.17
	Use of wood shaven	99	82.50
	Disease tolerant breed	89	74.17
	Adequate feed/nutrition in feed	87	72.50
3. Mitigation/risk reduction strategies	Source DOC from certified/ authorized sellers	116	96.67
	Administration of safe water always	111	92.50
	Proper collection of eggs	88	73.33
	Enterprise diversification	67	55.83
	Feed from certified millers	64	53.33
	Feed producing by self	56	46.67
	Buying input in bulk/advance	52	43.33
	Ensuring constant lightening/power supply	45	37.50
	Adequate/proper record keeping	30	25.00
4. Coping strategies	Off farm income	46	38.33
	Adequate storage of input/products	26	21.67
	Training/workshops attendance	25	20.63
	Cash liquidity for emergency	21	17.50
5. Transfer measures/Institutional strategies	Insurance polices	8	6.67
6. Marketing strategies	Securing market in advance	87	72.50
	Credit sales	83	69.17
7. Financial strategies	Loan from friends and relative	99	82.50
	Loan from cooperative	62	51.67
	Advance from buyers	44	36.67
	Loan from bank/financial institutions	8	6.67

Source: Authors' computation from field survey data, 2023

Table 6. Determinants of the level of risk management strategies

Variables	coefficient	p>|t|	coefficient (Marginal effect)	p>|t| (Marginal effect)
1. Age	0.03	0.16	0.004*	0.06
2. Sex	0.16	0.40	-0.22	0.59
3. Education	0.43	0.007	0.38**	0.005
4. Capital invested	0.44	0.000	0.34***	0.000
5. Years of experience	0.26	0.05	0.336*	0.062
6. Flock size	0.017	0.08	0.005*	0.08
7. Farm distance to residence	0.27	0.089	0.33*	0.069
8. Access to extension	0.56*	0.06	0.461*	0.05
9. Membership of cooperative	0.36	0.000	0.25***	0.000
10. Management system	0.477	0.091	0.477*	0.09
11. Diseases prevention &treatment	0.77	0.07	0.67*	0.09
12. Frequency disease outbreak	0.72	0.000	0.65***	0.000
13. Livestock insurance	0.054	0.002	0.044**	0.002
14. Credit constraint	0.022	0.04	0.006*	0.04
15. Output prices fluctuation	-0.59	0.905	-0.57	0.905
16. Government policy	-0.621**	0.006	-0.52**	0.005

Wald chi2(16) = 74.49   Log likelihood = -194.26222   Prob > chi2 = 0.0000   Obs=120

/cut1	-0.63	0.85	-1.99	0.52
/cut2	-0.45	0.64	-1.63	0.78

Source: Authors' computation from field survey data, 2023

Transfer measures/Institutional strategies: The coefficient of membership of cooperative is positive, and the marginal effect (0,25) is significant (p<0.001). This implies that been a member of the cooperative would increase the likelihood of risk management by 25%. This aligns with the work of Sugiyanto and Anggi (2018). This happened because farmers that belong to cooperative can access loans and advise for better risk management. The livestock insurance marginal effect (0.044) is significant (p<0.001). This implies that the likelihood of the level of risk management to increase by 4.4% for farmers that were on agricultural insurance policy. This occurs because they will be guided to put the right measure in place and avoid moral hazard claim by the insurance company. Government policy marginal effect (0.52) was negative and significant (p<0.001). This implies that the level of risk management has the likelihood of decreasing by 52% if government policies were favourable to poultry farmers.

Financial strategies: The model showed that capital invested marginal effect is significant at (p<0.001). This implies that the likelihood of the level of risk management to increase by 44% for every one percent increase in capital invested. This corroborates the work of Harvey, Liu, & Zhu et al, (2015) This may be so since most of the farmers that operate large stock had access to credit which they must repay and would not be willing to fail.

Marketing strategies: Output prices fluctuation (-0.59) was negative and not significant.

4. Conclusion

The study provided empirical evidence of the assessment of risk management strategies level determinants. Based on the result of the Multinomial logit model, it was evident capital invested, membership in cooperative society, frequency of disease outbreaks and ınvolvement in livestock insurance, credit constraint, flock size, educational and government policy were the major determinants of the level of farmers' risk management strategies

Capital invested, flock size and membership in cooperative society through which farmers can access credit were positively determinants of risk mitigation level, therefore, credit should be made available by stakeholders through cooperative societies so as to reduce farmers level of credit Constraint.

Education, frequency of disease outbreaks and involvement in livestock insurance were also positive determinants of risk mitigation level, therefore, farmers should be encouraged to be involved in livestock insurance and be trained on the best way to prevent diseases by extension workers.

Unfavourable government policies should be annulled and favourable ones should be developed and implemented.

Conflict of interest

The authors declare no conflict of interest.

Ethical Approval

This arcticle does not require ethics committee approval.

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Identification of *Vibrio parahaemolyticus* found on plastics via matrix assisted laser desorption/ionization time of flight mass spectrometry

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Abstract

Plastic pollution has become a pervasive environmental threat in aquatic ecosystems worldwide, leading to the formation of microplastics that act as substrates for microbial colonization and potential pathogen transmission. This study investigated plastic-associated bacterial communities, with a focus on *Vibrio parahaemolyticus*, along the Western Mediterranean coast of Türkiye, particularly in the Manavgat river and adjacent coastal waters (0-5 m depth). The aim of this study was to characterize plastic-associated bacterial communities, particularly Vibrio parahaemolyticus, in plastic and water samples collected from the Manavgat River and adjacent Western Mediterranean coastal waters using FTIR, PCR, and MALDI-TOF MS. Plastic and water samples were analyzed using a combination of Fourier Transform Infrared Spectroscopy (FTIR), Polymerase Chain Reaction (PCR) and Matrix-Assisted Laser Desorption/Ionization Time of Flight Mass Spectrometry (MALDI-TOF MS). FTIR results identified polyethylene (PE), polypropylene (PP), polystyrene (PS), polyethylene terephthalate (PET) and high-density polyethylene (HDPE) as the dominant polymers. PCR amplification targeting the *GyrB* gene confirmed the presence of *V. parahaemolyticus* in biofilm-forming bacterial isolates from plastics and seawater. MALDI-TOF MS analyses further supported these findings, yielding genus-level identification scores (1.7-1.9) consistent with established classification thresholds. The results indicate that plastics serve as persistent reservoirs and transport vectors for potentially pathogenic bacteria, facilitating their survival and dissemination in aquatic habitats. This study underscores the significance of the plastisphere as a microbial niche and highlights the public health risks associated with plastic-associated biofilms. Further metagenomic and functional analyses are recommended to elucidate gene exchange dynamics and pathogenic potential within these biofilm communities.

Keywords: Plastic pollution, *Vibrio parahaemolyticus*, Plastisphere, MALDI-TOF MS, PCR, Mediterranean coast, Biofilm

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1. Introduction

Plastic pollution has emerged as a significant environmental concern to aquatic ecosystems globally (Barnes et al., 2009). Plastics accumulate in rivers, coastal zones and marine habitats due to their durability, low cost, and widespread commercial use (Morritt et al., 2014). Plastics undergo physical, chemical and biological degradation and eventually creating microplastics (<5 mm) (Thompson et al., 2004). After decades of persistence, these particles are now found in sediments, water bodies, biota and even air compartments (O'Brien et al., 2023). Between 1.15 and 2.41 million tons of plastic are thought to enter the ocean annually via rivers (Lebreton et al., 2017). Research indicates that approximately 900 species have been affected by marine litter with plastic constituting 92% of these encounters (Gall & Thompson, 2016). The International union for conservation of nature (IUCN) red list of threatened species including 17% of the species impacted by plastic (Gall & Thompson, 2016). A study by Kühn and Van Franeker (2020) identified 914 species of marine megafauna (including 226 seabird species, 86 marine mammal species, all sea turtle species and 430 species of fish) that are affected by entanglement and/or ingestion. Because microplastics can change water quality, enter food webs, and interact with a variety of chemical and biological pollutants, their ubiquity and endurance generate increasing worries about their effects on the environment and human health (Winiarska et al., 2024).

In addition to their chemical impacts, microplastics serve as carriers of a wide variety of microorganisms, including opportunistic and clinically significant diseases (Zettler et al., 2013). Their hydrophobic surfaces easily absorb contaminants and organic compounds, forming ideal microhabitats that improve microbe adhesion and survival during long distance transport (Bowley et al., 2021). Microplastics can help bacteria like *Vibrio*, *Pseudomonas*, and *Aeromonas* spp. spread throughout riverine and coastal habitats, according to studies, which raises worries about the introduction of dangerous germs into new biological niches (Zettler et al., 2013; Zhang et al., 2020). In areas where anthropogenic pollution and extensive aquaculture activities coexist, microplastics may contribute to the spread of waterborne diseases in addition to posing an ecological danger, according to this vector like activity (Bowley et al., 2021; Odioko & Becer, 2025).

Plastics are rapidly colonized by diverse microbial communities that create resilient biofilms on their surfaces after entering aquatic systems; this phenomenon is known as the "plastisphere" (Amaral-Zettler et al., 2020). These biofilms can create hotspots for gene exchange and microbial survival by enriching pathogenic bacteria and antibiotic-resistant strains at levels higher than those found in the water surrounding those (Amaral-Zettler et al., 2020). Because biofilms offer structural defense, bacteria become more resistant to environmental stress, disinfectants and antibiotics, which may improve the survival of pathogenic species like *Vibrio alginolyticus* and *V. parahaemolyticus* (Kirstein et al., 2016; Oberbeckmann et al., 2018). Because plastics, bacteria and antibiotic residues may coexist and interact in coastal locations, plastic-associated biofilms are a rising health risk for both ecosystems and human populations (Kirstein et al., 2016; Amaral-Zettler et al., 2020).

Studies have shown that *Vibrio* spp. species, isolated from various aquatic organisms and known to cause vibriosis disease, can be accurately identified using MALDI-TOF MS (Yavuzcan et al., 2022; Çağatay, 2024; Gökdağ & Çağatay, 2024). Dieckmann et al. (2010) compared the MALDI-TOF method and reported that a total of 83 *Vibrio* strains were identified. Erler et al. (2015) identified *Vibrio* species in approximately 100 environmental samples by comparing MALDI-TOF MS and some genes.

This study aims to isolate and identify plastic-colonizing bacteria from Türkiye's Western Mediterranean coast, specifically from the Manavgat River and coastal water at depths of 0-5 m, using genomic PCR proteomics-based matrix assisted laser desorption/ionization time of flight mass spectrometry (MALDI-TOF MS) techniques together.

2. Material and Method

2.1. Plastic sampling

16 samples were collected in April 2023 at the Manavgat River, discharging into the Western Mediterranean Coast of Türkiye, as well as in adjacent marine areas (0-5 m depth) (Table 1, Figure 1). 14 plastic particles were collected, while water sampling consisted of one seawater sample and two river water samples. Water samples and plastic fragments found along river and coastal zone were collected and placed separately into sterile falcon tubes and 250 ml bottles. During transport to the laboratory, insulated containers with cold chains were used to preserve sample integrity. Upon arrival, water samples were filtered through Whatman filters (Whatman, UK) with a pore size of 0.45 µm and stored at -20°C until further analysis. All collected samples were inoculated into tryptic soy broth (TSB) (Condalab, Spain) and alkaline peptone water (Condalab, Spain) within 24 hours of arrival.

Table 1. Coordinates of the sampling stations

Stations	Lang	Long
MD1	36,805	31,340
MN2	36,747	31,472
MN3	36,744	31,478

2.2. Bacterial growth

Solid and liquid media were used for bacterial isolation from reference bacteria (*V. parahaemolyticus* ATCC 18802), water and plastic samples. To promote the selective growth of *Vibrio* spp., commonly used as selective agars such as chromogenic agar (VCA) (Condalab, Spain) and Thiosulfate-citrate-bile salts-sucrose agar (TCBS) (Condalab, Spain) were applied in parallel with TSA, TSB and nutrient agar (Difco™, France) for the same samples (Di Pinto et al., 2011; Kirstein et al., 2016). Samples were incubated at 25-30°C for 24-48 hours.

2.3. Polymer analyses of plastics with fourier transform infrared spectroscopy (FTIR)

Biofilm structures were examined on eleven different types of plastic samples collected from a total of three stations located at Manavgat River and the coastal area (0-5 m depths). Polymer analyses were carried out using a spectrum 400 fourier transform infrared spectroscopy (FTIR) and FT-NIR spectrometer (Perkin Elmer, USA).

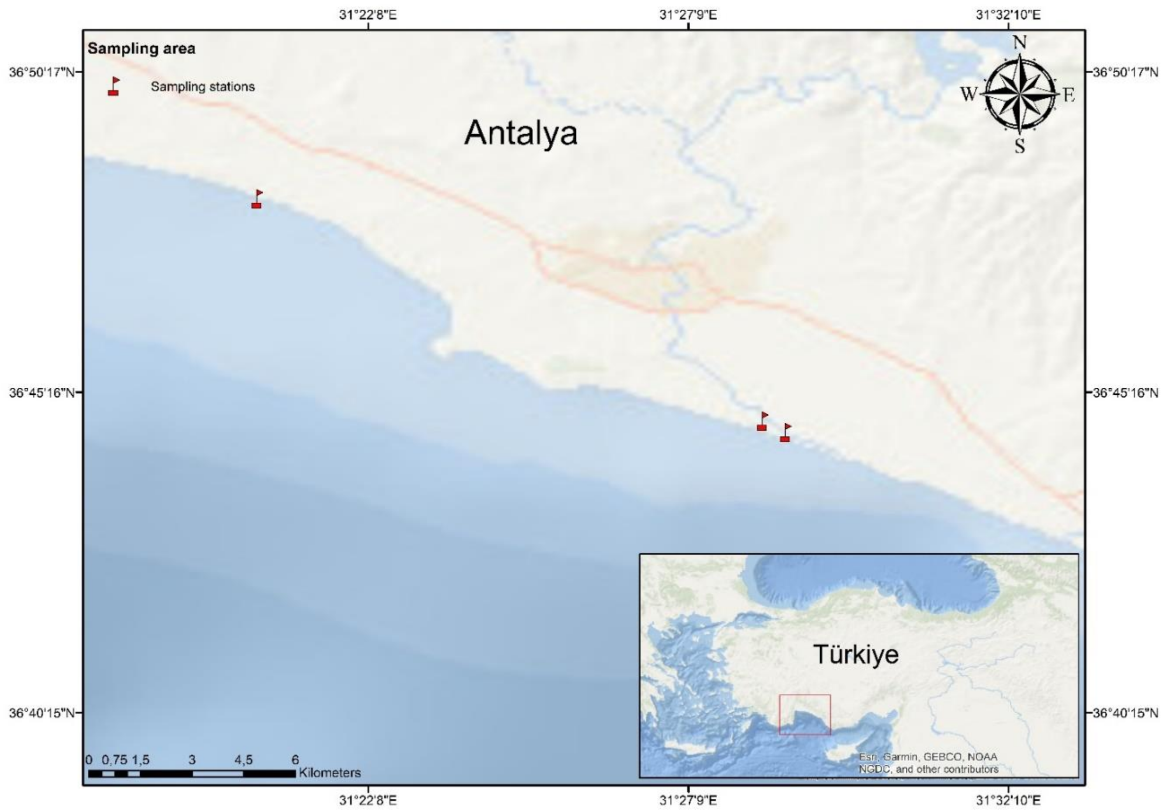


Figure 1. Sampling area

2.4. Molecular detection of *Vibrio* spp. on plastics

DNA isolation method was developed by modifying the protocols described by Comey et al. (1994) and Butler (2011). 1.5 mL of bacterial culture grown in medium for 24-48 hours was transferred into sterile tubes using sterile micropipettes and centrifuged at 5000 rpm for 10 minutes for genomic DNA extraction. The process was repeated until a sufficient pellet was obtained. 500 µL of SET buffer (75 mM NaCl, 25 mM EDTA, and 20 mM Tris-HCl, pH 7.5) (Sigma-Aldrich, Germany) was added to the pellet after removing the supernatant and vortexed until fully resuspended. The tubes were incubated at 95°C for 10 minutes and then allowed to cool to room temperature.

To denature proteins, 50 µL of SDS (sodium dodecyl sulfate) (Sigma-Aldrich, Germany) and 50 µL of Proteinase K (1 mg/mL) (Sigma-Aldrich, Germany) were added. Samples were incubated at 55°C for 2 hours to ensure complete protein degradation. Following incubation, 500 µL of phenol/chloroform/isoamyl alcohol (24:25:1) (Sigma-Aldrich, Germany) was added and the tubes were gently inverted for 5 minutes at room temperature. Samples were centrifuged at 10,000 rpm for 5 minutes, and the upper aqueous phase was transferred to a new sterile tube. An equal volume of chloroform/isoamyl alcohol (24:1) was added, mixed by gentle inversion for 5 minutes, and centrifuged again at 10,000 rpm for 5 minutes. The resulting upper phase was transferred to a new tube, and 1 mL of cold 99% ethanol was added to precipitate the DNA. After centrifugation at 10,000 rpm for 5 minutes, the supernatant was carefully removed. The pellet was washed with 1 mL of 70% ethanol by gentle inversion, followed by centrifugation at 10,000 rpm for 5 minutes. The supernatant was discarded, and the tubes were air-dried at 37°C under frequent monitoring until completely dry. The dried DNA pellets were dissolved in 50-100 µL of 1×TE buffer (10 mM Tris, 1 mM EDTA) depending on pellet size. The purified DNA samples were stored at -20°C for subsequent analyses.

2.5. PCR analyses

Vibrio parahaemolyticus specific primer targeting the *GyrB* gene (DNA Gyrase subunit B) was used to detect bacteria. PCR analysis was conducted by modifying the methods described by Venkateswaran et al. (1998). The primers, targeted gene regions, PCR thermal cycling conditions and references are presented in Table 2.

The PCR reaction mixture consisted of 10 µL PCR buffer (×10), 10 µL $MgCl_2$ (100 mM), 1 µL dNTP mix (10 mM each), 3.1 µL of each forward and reverse primer (50 µM), and 1 µL Taq DNA polymerase (5 U/µL) (Qiagen,USA), adjusted to a final volume of 25 µL with deionized water (Pascual et al., 2010). Amplified PCR products were verified by electrophoresis on 1.5% agarose (Sigma-Aldrich, Germany) gels containing 2 µL ethidium bromide (15 mg/mL) (Merk, Germany). To confirm the accuracy of the amplified gene regions, one randomly selected amplicon from each target gene was submitted for sequencing through a commercial sequencing service.

Table 2. Primers and PCR conditions

Primer name	Primer sequences (5'-3')	Amplicon size (bp)	PCR Conditions	Reference
GyrB	Forward -CGG CGT GGG TGT TTC GGT AGT Reverse-TCC GCT TCG CGC TCA TCA ATA	285	94ºC 1min 94ºC 1min 58ºC 1:30 sec X30 72ºC 2:30 sec 72ºC 7 min	Venkatesnran et al. (1998)

2.6. MALDI-TOF MS analyses

The formic acid extraction method was applied for bacterial identification using the MALDI-TOF MS system (Bruker Microflex) (Wu et al., 2020). Prior to analysis, bacterial colonies exhibiting distinct morphological characteristics were individually cultured in liquid media to obtain pure isolates. These isolates were sub-cultured as single colonies on TSA, VCA, TCBS, and nutrient agar plates for further bacterial growth. The direct transfer method was employed for MALDI-TOF MS analysis.

MALDI-TOF MS (Bruker Microflex, USA) analyses were performed within 12 hours of bacterial cultivation. For sample preparation, a single bacterial colony from each plate was picked using a sterile toothpick and gently smeared as a thin layer onto the metal 96-well target plate specific to the instrument. Subsequently, 1 µL of formic acid solution was applied to each spot and allowed to dry at room temperature. Within one hour after drying, 1 µL of matrix solution was added to each spot. Once dried at room temperature, the prepared target plate was loaded into the instrument for analysis.

3. Results and Discussion

3.1. Morphologic identification

Isolates were taken from biofilm from plastic surface were inoculated on VCA, TCBS, TSA, TSB and nutrient agar at 25-30°C for 24 hours. Colonies on VCA and TCBS plates were generally green, orange, circular with diameters between 0.2 and 0.4 mm (Figure 2).

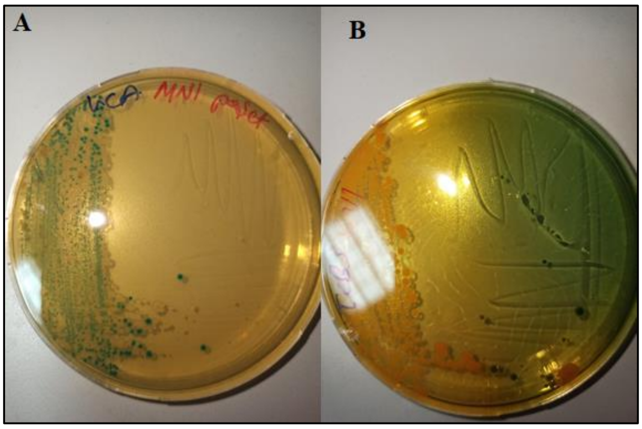


Figure 2. Colony morphology of *Vibrio parahaemolyticus* isolated from (A)Vibrio Chromogenic Agar (VCA) and (B) Thiosulfate-Citrate-Bile Salts-Sucrose Agar (TCBS)

3.2. Identification of Vibrio parahaemolyticus isolates on biofilm-form with PCR

To detect *Vibrio* spp. in the plastisphere (biofilm), species-specific PCR was performed using *GyrB*-Forward and *GyrB*-Reverse specific primers. As a result of this study, *V. parahaemolyticus* species were detected in different samples. The PCR amplicon bands obtained for *V. parahaemolyticus* were visualized on agarose gels. The resulting amplicon sizes were 285 bp for *V. parahaemolyticus*, as shown in Figure 3. For *V. parahaemolyticus*, the *GyrB* primers were used in studies by Vongxay et al. (2006) and Venkateswaran et al. (1998). The PCR bands are presented in Figure 3 and demonstrate consistency between the results of this study and the previous literature.

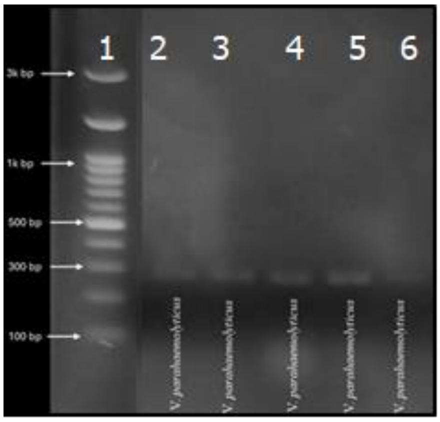


Figure 3. Molecular marker (1), *GyrB* gene amplicons from biofilm of plastics (2-6)

3.3. FTIR analyses of plastic samples

The photographs of plastics (Figure 4) and the FTIR based polymer analysis graphs are shown (Figure 5). The polymer readings were measured in the range of 650-4000 cm^{-1} and the spectra of the identified polymers are presented in the Figure 5.

FTIR analyses revealed the presence of polyethylene (PE), polyethylene terephthalate (PET), polypropylene (PP) (isotactic), polystyrene (PS), propylene-acrylic acid copolymer, and high-density polyethylene (HDPE) polymers. According to Plastics Europe (2023), polyethylene (PE), polypropylene (PP), polyvinyl chloride (PVC), polystyrene (PS), polyethylene terephthalate (PET), polyurethane (PUR), thermoplastics and thermoset plastics together accounted for 90% of the global plastic production of 400.3 million tons in 2022. Munari et al.

(2016) reported that plastics along the Adriatic Sea coasts mainly consisted of polyethylene (HD-PE and LD-PE), polypropylene (PP), and polyamide (PA). In Lebanon, polypropylene (PP), polyethylene (PE), polystyrene (PS), polyamide (PA), polyethylene terephthalate (PET), and polyurethane (PUR) were the most frequently detected polymers in seawater, sediment, and fish digestive tract samples (Kazour et al., 2019). Constant et al. (2017) found PE, PS, and PP as the most common polymers in coastal Spain, while Baini et al. (2018) identified PE, PP, PS, ethylene-vinyl acetate (EVA), and styrene-butadiene (SBR) as dominant polymers. Studies from the Turkish Mediterranean coasts have also reported PS, PE, and PP copolymers as dominant in seawater samples (Güven et al., 2017). *V. parahaemolyticus* was detected in four PET plastic fragments and in one seawater sample. However, because all positive plastics belonged to the same polymer category (PET) and the overall sample size was limited, no meaningful comparison could be made regarding polymer-specific detection frequency or potential patterns in MALDI-TOF scores or PCR positivity across different plastic types. The small number of samples represents a methodological limitation, and therefore polymer-dependent inferences should be interpreted with caution

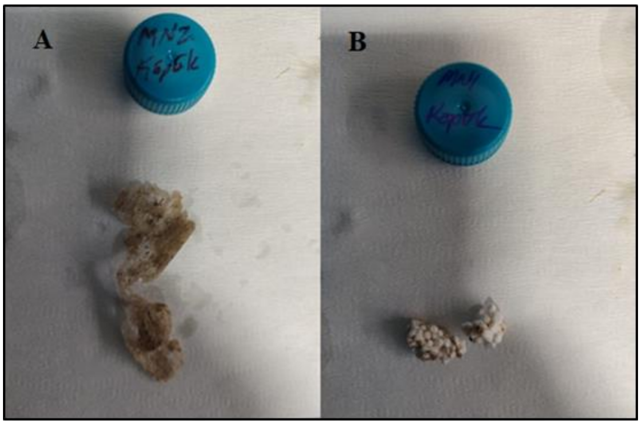
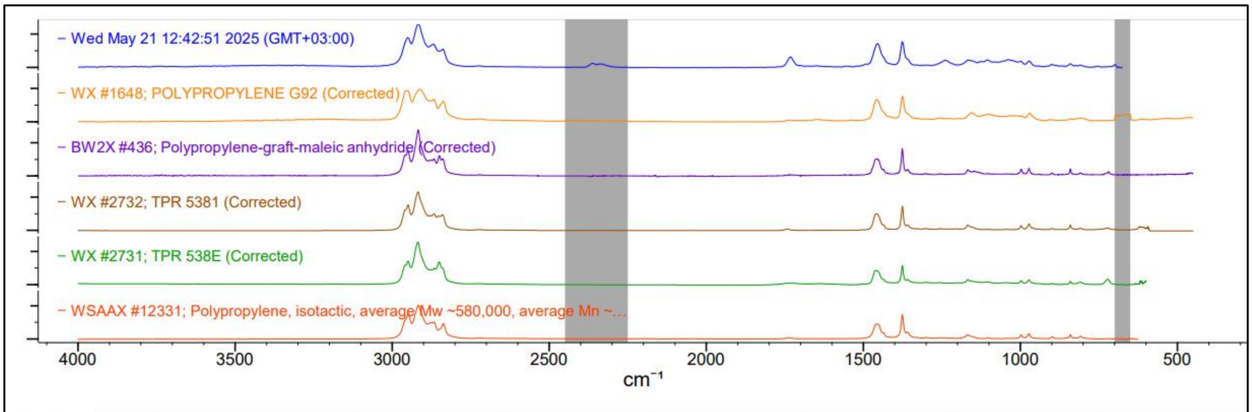


Figure 4. The photographs of plastics



HQI	ID	Name	hemical Descriptio	Classification	Sample Description	Solubility	ctraBase Compoun	Use
86.84	1648	POLYPROPYLENE G92	POLYPROPYLEN E				Acivbqy4oLI	
86.63	436	Polypropylene-graft-maleic anhydride			pellets; x=98, y=2		D09RrvA5XAA	
86.16	2732	TPR 5381	THERMOPLASTI C ELASTOMER	SYNTHETIC POLYISOPRENE S AND NATURAL			IKVxJjwOAvs	

Figure 5. Sample of FTIR analyses result

The high abundance of PE and PP polymers in these studies is linked to their low specific gravity (0.89-0.95 g/cm^3 and 0.85-0.92 g/cm^3, respectively), which allows them to float and accumulate along coastal areas due to water currents (Andrady, 2011; Jayasiri et al., 2013; Enders et al., 2015; Zhao et al., 2015; Munari et al., 2016). PE polymers, with an annual production of about 80 million tons, are primarily used in packaging, plastic bags, films, and bottle manufacturing (Thompson et al., 2004). PP polymers, with an annual production of around 50 million tons, are widely used in packaging and in manufacturing reusable containers, stationery, and textiles (Thompson et al., 2004; Jayasiri et al., 2013; Zhao et al., 2015; Munari et al., 2016). Single use PE and PP based items such as plastic bags, ropes, and fishing lines are commonly detected along both Mediterranean and global coastlines (Eriksen et al., 2014; Suaria et al., 2016).

3.4. Identification of Vibrio parahaemolyticus isolates on biofilm form with using MALDI-TOF MS

A bacterial colony of *Vibrio* spp. for identification with using MALDI-TOF MS were placed on a plate and exposed to laser shots. To facilitate efficient absorption of laser energy by the bacterial colony and proton transfer to the molecules, a matrix was added, enabling molecular ionization. Following ionization, gas phase ions were released as free molecules. The signal intensity peaks of the ionized molecules or component proteins were carried to the detector and compared with the reference database for species-level microbial identification. The bacteria identified using the MALDI-TOF MS method and their score values are listed in Table 3.

Table 3. MALDI-TOF scores of *Vibrio parahaemolyticus*

Sample	Species Identification	Score value
Polyethylene terephthalate (PET)	*V. parahaemolyticus*	1.741
Sea water	*V. parahaemolyticus*	1.783
Polyethylene terephthalate (PET)	*V. parahaemolyticus*	1.737
Polyethylene terephthalate (PET)	*V. parahaemolyticus*	1.727
Polyethylene terephthalate (PET)	*V. parahaemolyticus*	1.985

The identification of *Vibrio* spp. is often time-consuming due to their diverse phenotypic traits and long culture durations, which make biochemical identification difficult (Wu et al., 2020). The MALDI-TOF MS method allows rapid and accurate species-level identification using minimal sample volumes (De Bruyne et al., 2011; Malainine et al., 2013; Afanasev et al., 2014; Kirstein et al., 2016; Kapetanović et al., 2023; Bielen et al., 2024; Gökdağ & Çağatay, 2024). It also enables large-scale analysis of isolates from clinical and environmental samples (Dieckmann et al., 2010). This study did not evaluate virulence factors such as the tdh and trh genes or antibiotic resistance profiles; therefore, the virulence potential of the detected *V. parahaemolyticus* isolates remains unknown. Accordingly, statements regarding public health implications should be interpreted cautiously, as the presence of Vibrio on plastics and in seawater does not indicate clinical relevance. Future studies should include virulence-gene screening to better assess pathogenic potential.

Malainine et al. (2013) analyzed seawater, sediment, and mussel samples from Khnifiss Lagoon (Morocco) and identified *V. parahaemolyticus* using MALDI-TOF MS, highlighting its reliability. Afanasev et al. (2014) emphasized the device's high efficiency, ease of use, and low operational cost. Bielen et al. (2024) compared MALDI-TOF MS with 16S rDNA sequencing in mussel isolates and concluded that an expanded database would make it the fastest and most effective tool for culturable bacterial identification. Kirstein et al. (2016) were the first to identify biofilm-forming *Vibrio* spp. (*V. parahaemolyticus*, *V. vulnificus*, and *V. cholerae*) on plastics using MALDI-TOF MS. Kapetanović et al. (2023) also confirmed its high specificity and sensitivity for *Vibrio* detection in plastics, seawater, and sediment samples.

MALDI Biotyper RTC (Bruker®) software assigns score values between 0.00-3.00 (Jansson et al., 2020; Çağatay, 2024). According to the traffic-light classification: green (2.0-3.00) indicates reliable species-level identification, yellow (1.7-1.99) reliable genus-level identification, and red (<1.70) low-confidence or unidentified results. Species (*V. alginolyticus* and *V. parahaemolyticus*) had score values ≥1.7, consistent with previous literature findings, as shown in Table 3.

4. Conclusion

Plastics, as pervasive hydrophobic and hydrophilic pollutants, accumulate in both terrestrial and aquatic environments due to improper disposal and degradation. Their fragmentation into microplastics (<5 mm) enables ingestion by marine organisms, leading to mortality and potential biomagnification through the food chain, posing health risks to humans and other species. Given the continuous rise in global plastic production and consumption, plastic-associated pollution and its ecological impacts are expected to intensify.

This study demonstrated that plastics serve as substrates for microbial colonization of *V. parahaemolyticus*. These biofilms not only promote microbial survival and interaction but also host pathogenic taxa such as Vibrio spp., potentially facilitating pathogen persistence and gene transfer. These findings highlight the need for region-specific monitoring programs and evidence-based policies aimed at reducing plastic inputs and assessing pathogen-bearing biofilms on aquatic plastics. Further research should investigate interactions among resistance-related genes and the impact of single or multiple copies of resistance genes on antibiotic resistance in *V. parahaemolyticus*. Understanding the mechanisms governing biofilm development on microplastics, along with their roles in pollutant adsorption, antibiotic resistance, and bioremediation, requires further investigation, including whole-genome analyses of associated microbial communities.

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Conflict of interest

The authors declare no conflict of interest.

Ethical Approval

This arcticle does not require ethics committee approval.

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Effect of silicon and application of potassium on *Puccinellia distans* grown under saline-alkali conditions

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Abstract

Salinity and alkaline conditions are among the most important abiotic stress factors that limit plant growth and development, especially in arid and semi-arid areas. Indeed, approximately 30,000 hectares of plain land located in the Dumlu region of Erzurum cannot be adequately utilized due to high groundwater levels, salinity, and alkalinity. Reclaiming this type of soil is quite costly, and one of the prominent alternatives is to identify plant species and varieties that are tolerant to high soil salinity and ensure their cultivation in such areas. For this purpose, in our study, along with the halophyte plant *Puccinellia distans*, silicon dioxide (SiO_2) and potassium nitrate (KNO_3) fertilizers were used. The research was conducted in greenhouses at Atatürk University Faculty of Agriculture, using soil samples appropriately taken from four different sections of the affected region with varying salt content, according to a completely randomized experimental design. For this purpose, a total of 144 pots (4×1×4×3×3=144) were used across 4 different locations, with 1 halophyte plant *Puccinellia distans*, 4 different silicon (SiO_2) doses (0, 1, 2, and 4 mM), 3 different potassium nitrate (KNO_3) doses (0, 20, and 40 mM) with 3 replications. Plant height, number of main stems, number of leaves, fresh forage yield and dry matter yield, crude protein content and yield, and ADF and NDF contents were examined in the plants obtained at the end of the trial. As a result, a decrease in all yield and yield components was observed in plants grown under saline-alkali conditions compared to plants grown in normal soil. However, in saline-alkaline conditions, the application of silicon and potassium generally reduced the negative effects of salinity stress on the plant. The highest dry matter yield, crude protein content, and yield value were obtained from the application of 40 mM KNO_3 fertilizer without the addition of silicon. Based on these results, we can say that the plant Puccinella distans is promising for the evaluation of saline-alkali areas with similar conditions, and that potassium fertilization in particular is effective in increasing its yield.

Keywords: *Puccinellia distans*, Salinity, Halophytes, Silicon, Alkali

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1. Introduction

Salinity and alkalinity are among the most important abiotic stress factors that limit plant growth and development, especially in arid and semi-arid areas (Cui et al., 2021). Saline-alkali soils have poor physicochemical conditions, which damage the chemical, physical, and microbiological properties of the soil, thereby limiting crop growth (Zhao et al., 2020; Hao et al., 2021). Indeed, approximately 20% of the world's irrigated land is affected by salinity and alkalinity, making this a significant problem for global agriculture and food security (Baloch et al., 2025). Therefore, reclaiming problematic lands like saline-sodic soils and bringing them back into agricultural production is of great importance for agricultural sustainability. Among the reclamation methods for this type of soil are physical, chemical, and biological methods such as sand mixing and mulching, the use of gypsum and humic acid, phytoremediation, and microbial reclamation, as well as some modern technologies like genetic (Wang et al., 2023; Zhu et al., 2024) and electrokinetic reclamation (Reuss, 1809; Özentürk, 2022) . However, the most economical of these alternatives is phytoremediation, which involves identifying and cultivating plant species and varieties, often halophytes, that are tolerant to high soil salinity in such areas (Amombo et al., 2022).

Actually, saline-alkali soil conditions restrict the growth of many cultivated plants and allow only a small fraction of the herbaceous species in the vegetation to grow. Similarly, in a study conducted to determine the forage plant species that could grow in the research area, it was observed that grasses were more resistant, while none of the examined plants (alfalfa, birdsfoot trefoil, white clover, tall fescue, meadow fescue, perennial ryegrass, tall oatgrass, and smooth brome) showed promise in the land soils (Tan et al., 2002). However, it has been reported that halophytic plants can be used in such marginal areas for the remediation of saline soils, and can also serve as an alternative animal feed-protein source (Li et al., 2019; Çınar et al., 2021; Behera & Ramachandran, 2021). *Puccinellia distans* is known as a salt-tolerant halophyte plant that can grow naturally in saline areas (Soleimannejad et al., 2019). Studies have shown that the plant is used in Iran for the reclamation of saline lands (Bandani & Abdolzadeh, 2007). Additionally, it has been reported to increase the productivity of saline lands and can be used as a forage crop in animal nutrition (Alshammary et al., 2004; Rao et al., 2017). In addition, recent studies have concluded that the combined use of biological and chemical methods in the reclamation of saline-alkali soils could be a more effective alternative (Cuevas et al., 2019).

It is known that the first application of phytoremediation for the reclamation of saline-alkali soils, using gypsum with green manure plants, was carried out by Kelley (1937). On the other hand, it is known that silicon (Si) and potassium (K) increase the tolerance of plants under abiotic stress, especially salinity, and improve growth (Abbas et al., 2015; Ahanger & Agarwall, 2017; Servet & Eşitken, 2018). It has been found that silicon increases plant salt tolerance by controlling the transport and accumulation of Na^+ and K^+, which are the main salt tolerance mechanisms in plants under salinity stress (Zhu & Gong, 2014; Kınay & Erdem, 2022). In addition, potassium increases the tolerance of plants under salt stress due to its specific functions in important metabolic events such as plant water consumption, protein synthesis, energy metabolism, and photosynthesis (Li et al., 2017; Ahanger & Agarwall, 2017). For this purpose, it was aimed to determine both the adaptation of the halophyte *Puccinellia distans* and the effect of the applications on yield and yield components in the plant, along with SiO_2 and KNO_3 fertilizers in the research area.

2. Material and Method

The research was conducted in the Dumlu (Erzurum) region in 2009-2010, using soil samples taken from four different locations with varying salt content, which were exposed to the degradation process and had agricultural potential. The samples were collected from the greenhouses of Atatürk University Faculty of Agriculture. Soil samples taken from a depth of 0-30 cm in the test field were air-dried, sieved thru a 4 mm sieve, and then placed in 15 cm diameter plastic pots with 1000 g of soil per pot. In the trial greenhouse conditions, according to the factorial design, the Completely Randomized Block design was used with 4 different locations (1. Location: N 40°01'13.4", E 041°20'54.1"; 2. Location: N 39°58'07.2", E 041°17'57.3", 3.location: N 39°59'07.6"; E 041°17'56.7", 4.location: N 39°58'49.9", E 041°21'24.8"); 1 plant (*Puccinellia distans*), 4 different silicon doses (0, 1, 2, and 4 mM), 3 different KNO_3 doses (0, 20, and 40 mM) were carried out in a total of 144 pots (4×1×4×3×3=144) with 3 replications over a period of two years. However, since plant emergence occurred in only two soil groups (1 and 2), the experiment was conducted with these two soil type, and these two soil type were also evaluated in the statistical analysis.

The analyzes of the soils were conducted in the laboratories of the Soil Science Department in Faculty of Agriculture, according to Kacar (1994). Some properties of the problematic and soil materials used in the experiment are shown in Table 1. Analysis of soil samples revealed that soil sample 1 is normal, samples 2 and 3 are saline-alkaline, and sample 4 is alkaline soil (Table 1). It is known that the soils of the Dumlu region generally have a "saline-alkali" soil characteristic, making them heavier in texture compared to normal soil, with very high electrical conductivity and rich in lime, Na, Ca and Mg.

Based on initial soil analyzes, ammonium sulfate (20.5% N) and triple superphosphate (48% P_2O_5) fertilizers were applied as a standard basal fertilizer at a rate of 10 kg N/da and 5 kg P_2O_5/da for *Puccinellia distans* to ensure optimal plant development. SiO_2 (0, 1, 2, and 4 mM) and KNO_3 (0, 20, and 40 mM) applications were applied to pots along with seed sowing. Following fertilizer application, 40 seeds

Table 1. Ec, Ph, CEC values of soil samples

Soil type	pH	Ec (dS/m)	Ca (me $100g^{-1}$)	K (me $100g^{-1}$)	Mg (me $100g^{-1}$)	Na (me $100g^{-1}$)
1	7.91	1000	1.76	0.25	1.17	14.50
2	8.94	8060	1.45	0.49	0.65	22.14
3	8.79	8527	1.69	0.70	0.78	23.36
4	8.86	4480	0.47	1.00	0.52	22.95

of *Puccinella distans* were sown per pot at a rate of 1 kg per decare. The soil samples with plant emergence (soil samples 1 and 2) were thinned after germination, leaving 20 plants in each pot.

During the plants' growth period, their water needs were met with normal irrigation water, and the soil moisture level was attempted to be maintained at 50% of field capacity. The temperature and humidity values of the greenhouse conditions throughout the day were measured using a data logger (Testo 175-H2 V01.10), and approximate irrigation intervals were determined by weight calculation. The number of main stems, plant height, number of leaves, fresh forage yield and dry matter yield, and crude protein, ADF, and NDF analyzes were performed on the plants harvested from the pots where each treatment was applied. The data obtained from the research results were subjected to variance analysis using the JMP 5.1 statistical software package, and the significant means were compared according to the LSD test.

3. Results and Discussion

3.1. Plant height

According to the research results, the effects of the applied treatments were found to be significant on the soil types used and between years (p<0.01). In addition, the soil type × application interaction (ST×A) was significant (Table 2 and 3). In the second year, a decrease in plant height was observed compared to the first year. A 35% reduction was observed in saline-alkali soil compared to normal soil (Table 2). Among the applications, the highest value (17.50 cm) was obtained from the S_1K_2 application compared to the control group (14.33 cm) (Table 2). Aridity and high salt accumulation have reduced plant height. Studies have shown that salt stress reduces growth parameters such as root and shoot length, plant height, leaf area, and biomass accumulation (Parihar et al., 2015; Ahmed et al., 2022; Balasubramaniam et al., 2023). In addition, it is known that the combined application of silicon and potassium enhances plant growth by reducing Na accumulation and ESP (exchangeable sodium percentage), and increasing the K/Na ratio, especially under stress conditions (Zargar et al., 2019; Ghazi et al., 2021).

3.2. Number of main branch

The number of main branch in *Puccinella distans* was found to be 2.61 in normal soil and 2.32 in plants grown in saline-alkali soil (Table 2). This difference was determined to be statistically insignificant (p>0.05). This situation may be due to the osmotic stress caused by salt stress leading to a decrease in plant growth rate (Ouertani et al., 2021; Kınay & Erdem 2022). The ST×A interaction was statistically significant (p<0.05) with the applied treatments (p<0.01). Among the applications, the highest main branch number was found in the S_2K_1 and S_3K_1 applications, with a value of 3.25. Similarly, in a study conducted, it was found that silicon and potassium applications improved growth parameters such as the number of branch, number of seeds, seed yield, and plant height (Çakır, 2020).

Table 2. Plant height (cm), mean branch number, leaf number, ADF (%), NDF (%), crude protein ratio (%), crude protein yield (g), fresh forage yield (g/pot), and dry matter yield (g/pot) of *Puccinella distans* grown under different soil and different doses of silicon and potassium applications

Treatments		Plant height (cm)	Main branch number	Leaf number	ADF (%)	NDF (%)	Crude protein ratio (%)	Crude protein yield (g)	Fresh forage yield (g/pot)	Dry matter yield (g/pot)
Year	2009	14.56^{a}	2.58	5.79	32.84^{a}	44.96	11.34^{b}	19.15	5.80	1.69
	2010	14.10^{b}	2.35	5.61	32.49^{b}	44.14	11.82^{a}	20.64	6.04	1.75
	Mean	14.33	2.47	5.70	32.67	44.55	11.58	19.90	5.92	1.72
Soil Type	1	17.40^{a}	2.61	5.53	32.66	44.95	10.87^{b}	20.36	6.17^{a}	1.87^{a}
	2	11.21^{b}	2.32	5.88	32.66	44.14	12.28^{a}	19.44	5.67^{b}	1.56^{b}
	Mean	14.31	2.47	5.70	32.66	44.55	11.56	19.90	5.92	1.72
Application	S_0K_0	14.33^{cd}	2.08^{df}	5.08^{cd}	32.06^{bd}	46.16^{a}	12.13^{ab}	17.07^{df}	5.35^{cd}	1.41^{d}
	S_0K_1	16.17^{ab}	1.50^{f}	3.50^{e}	31.88^{d}	43.64^{cd}	11.74^{b}	20.15^{bd}	6.21^{ac}	1.75^{ac}
	S_0K_2	16.83^{ab}	2.00^{df}	4.75^{de}	32.94^{a}	44.86^{ac}	12.96^{a}	25.75^{a}	7.15^{a}	2.03^{a}
	S_1K_0	13.92^{de}	1.92^{ef}	4.58^{de}	33.06^{a}	45.72^{ab}	12.11^{ab}	17.89^{df}	5.13^{d}	1.48^{cd}
	S_1K_1	16.00^{b}	2.17^{de}	5.25^{cd}	31.95^{cd}	44.99^{ac}	12.35^{ab}	19.54^{ce}	5.76^{bd}	1.60^{bd}
	S_1K_2	17.50^{a}	2.42^{ce}	5.83^{bd}	33.15^{a}	44.76^{bc}	12.45^{ab}	25.03^{a}	7.17^{a}	2.02^{a}
	S_2K_0	13.00^{df}	2.33^{de}	6.33^{bc}	32.74^{ab}	44.92^{ac}	12.36^{ab}	22.06^{ac}	4.83^{d}	1.81^{ab}
	S_2K_1	13.58d^{e}	3.25^{a}	7.83^{a}	32.18^{bd}	44.06^{c}	10.31^{c}	18.43^{ce}	5.75^{bd}	1.76^{ac}
	S_2K_2	12.50^{ef}	3.00^{ac}	5.33^{cd}	33.15^{a}	42.54^{d}	10.19^{c}	16.37^{ef}	6.51^{ab}	1.61^{bd}
	S_3K_0	10.50^{g}	2.58^{bd}	6.08^{bc}	33.12^{a}	44.43^{bc}	10.00^{c}	14.38^{f}	5.28^{cd}	1.46^{d}
	S_3K_1	15.50^{bc}	3.25^{a}	7.08^{ab}	32.66^{ac}	44.62^{bc}	11.78^{b}	23.51^{ab}	6.35^{ac}	2.00^{a}
	S_3K_2	11.83^{fg}	3.08^{ab}	6.75^{ab}	33.14^{a}	43.84^{cd}	10.54^{c}	18.58^{ce}	5.89^{bd}	1.76^{ac}
	Mean	14.31	2.47	5.70	32.67	44.55	11.58	19.90	5.95	1.72

Table 3. Analysis of variance (ANOVA) for plant height, number of main branches, leaf number, ADF (%), NDF (%), crude protein content (%), crude protein yield (g), fresh forage yield (g/pot) and dry matter yield (g/pot) of *Puccinella distans* as affected by year (Y), soil type (ST) and application (A) of silicon and potassium

Sources of variation	DF	Plant height (cm)	Main branch number	Leaf number	ADF (%)	NDF (%)	Crude protein ratio (%)	Crude protein yield (g)	Fresh forage yield (g/pot)	Dry matter yield (g/pot)
Years (Y)	1	**	ns	ns	**	ns	*	ns	ns	ns
Soil type (ST)	1	**	ns	ns	ns	ns	**	ns	*	**
Application (A)	11	**	**	**	**	**	**	**	**	**
Y×ST	11	ns	ns	ns	ns	ns	ns	ns	ns	ns
Y×A	11	ns	ns	ns	ns	ns	ns	ns	ns	ns
ST×A	11	**	*	**	**	**	**	**	**	**
Y×ST×A	11	ns	ns	ns	ns	ns	ns	ns	ns	ns

**p<0.05; **p<0.01; ns: not significant.*

3.3. Number of leaf

The applications on the number of leaf and the ST×A interaction were significant (p<0.01) (Table 2 and Table 3). Among the applications made, the S2K1 application resulted in the highest leaf count value, increasing by 54% compared to the control (7.83) (Table 2). The osmotic effect of salt stress indicates itself in plants thru reduced growth rates, changes in leaf color, alterations in the root/shoot ratio, and changes in the speed of plant maturation, while the ionic effect of salt stress is primarily seen thru damage to meristematic tissues or leaves (Ouertani et al., 2021). The application of silicon and potassium to plants has been adopted as an effective method because it alleviates the negative effects of stress and increases stress tolerance (Zhu & Gong, 2014; Walsh & Walsh, 2020). Similar to our study, Maghsoudi et al. (2019), Yıldırım and Güneş (2021), and Walsh and Walsh (2020) reported that the applied silicon and potassium improved the adverse conditions caused by stress in the plant's growth parameters.

3.4. Fresh forage yield

Soil type was found to be 5% significant for fresh forage yield, while the interaction between applications and ST×A was determined to be significant (p<0.01) (Table 2 and 3). The fresh forage yield, determined as 6.17 g/pot in normal soil, decreased to 5.67 g/pot in saline-alkali soil. Over the years, fresh forage yield, which was 5.80 g/pot in the first year, was determined to be 6.04 in the second year, but this increase was found to be statistically insignificant. Among the applications performed, an increase was generally determined compared to the control, while the S_1K_2 application provided the highest increase in dry matter yield (7.17 g/pot) (Table 2). Kınay and Erdem (2022) observed decreases in the fresh forage yield and dry matter yield under increasing salt applications. However, similar to our study, it was determined that these decreases were reduced with silicon applications. This situation may have resulted from the application of silicon, which increased the plant's salt tolerance by reducing Na concentrations and increasing K concentrations. Indeed, silicon (Si) is known to promote plant growth and improve plant biomass and productivity under various stress conditions (Deshmukh et al., 2017). In addition to this, potassium plays an important role in maintaining the water content of plant tissues, as well as in the basic physiological functions of plants such as protein synthesis and the production of sugars and starches (Pandey & Mahiwal, 2020).

3.5. Dry matter yield

According to the research results, the effects of soil type, applied practices, and the interaction of ST×A on the dry matter yield of *Puccinellia distans* were found to be statistically significant (p<0.01) (Table 2 and 3). The dry matter yield increased depending on the applications and the fertilizer doses applied. The highest dry matter yield was obtained from the application of 40 mM potassium without silicon (S_0K_2, 2.03 g/pot) (Table 2). Growth reduction is a common symptom of any plant under high salinity stress due to disrupted ionic balance (Kundu et al., 2018). In the study conducted, it was also determined that salinity stress negatively affects plant growth and development (El Sabagh et al., 2021).

Potassium is a macronutrient essential for physiological processes in plants, such as enzyme activation, regulation of osmotic pressure, and stomatal movement (Wang et al., 2013).

It is also known that increasing potassium and calcium concentrations in plants under salt stress can improve the plant's osmoregulatory adaptation to salt stress, thereby mitigating the harmful effects of salinity on growth and yield (Nelson, 1978). Ju et al. (2021) and Hafeez et al. (2024), similar to our study, stated that potassium application has positive effects on salinity and mitigates the negative effects of salinity on wheat seedlings.

3.6. Crude protein ratio

The effect of soil type, applications, and the ST×A interaction on crude protein content was determined to be statistically significant (p<0.01) (Table 2 and 3). Additionally, the difference between the years has been significant (p<0.05). The highest crude protein ratio, at 12.96%, was obtained from the 40 mM potassium application (S_0K_2) without silicon. The crude protein ratio, which was 11.34% in the first year, increased to 11.82% in the second year (Table 2). The crude protein ratio was found to be 10.87% in normal soil and 12.28% in saline soil. Generally, it has been reported that salinity reduces the crude protein (CP) ratio in forage crops however, the study determined that salinity did not affect the protein content in canola (Francois, 1994).

Additionally, another study found that potassium silicate application increased carbohydrate and protein ratio in plants (Hellal et al., 2020). This situation could be a result of potassium silicate's role in increasing the transport of nutrients essential for protein and carbohydrate biosynthesis in plants, which is facilitated by its enhancement of the photosynthetic process.

3.7. Crude protein yield

According to the research results, the crude protein yield was found to be statistically significant for the applied treatments and the ST×A interaction (p<0.01) (Table 2 and 3). The crude protein yield in the second year increased compared to the first year and was determined to be statistically insignificant. The crude protein yield in saline-alkali soil, which was 19.44 g, was 20.36 g in normal soil. Because the high salt concentration reaches a threshold that inhibits nutrient uptake and plant growth (Wang et al., 2023; Abebe & Tu, 2024). The highest crude protein yield of 25.75 g was obtained from the S_0K_2 application (Table 2). Indeed, in their study, Walsh and Walsh (2020) reported that potassium can contribute to protein synthesis by increasing the nitrogen content in wheat.

3.8. ADF ratio

The change in the ADF ratio was found to be significant across years and applications (p<0.01). Additionally, the ST×A interaction was found to be statistically significant. The ADF ratio decreased in the second year compared to the first year (Table 2 and 3). The S_0K_1 (31.88%) and S_1K_1 (31.95%) applications caused a decrease in ADF ratio compared to the control (Table 2). Salinity and alkalinity stress can cause changes in the crude protein and fiber content of forage crops, leading to reduced nutrient uptake and impaired metabolic processes, which can decrease their nutritional value. The applications performed have improved the digestibility of the plant by reducing the ADF ratio. The results obtained approached the desired ADF ratio for forage crops (31%) (Attia-Ismail, 2015) and varied between 31 and 33 %.

3.9. NDF ratio

The effect of applications on the NDF ratio was found to be significant (p<0.01). Additionally, the ST×A interaction was statistically significant (Table 2 and 3). All the applications performed reduced the NDF ratio compared to the control (46.16%). The applications were evaluated positively as reducing the NDF ratio in the rumen leads to better digestibility and energy efficiency. Indeed, in forage crops, it is desirable for the NDF content to be below the upper limit of 40% in terms of quality (Attia-Ismail, 2015).

4. Conclusion

In this study, the effects of different doses of silicon and potassium fertilizers on the yield and yield components of the halophyte plant *Puccinellia distans*, adapted to regional conditions, were investigated in terms of bringing saline-alkali areas into production. According to our results, growth parameters such as plant height, leaf number, fresh forage yield and dry matter yield showed a decrease in saline-alkali soil, while silicon and potassium applications significantly reduced these negative effects. Especially the S_1K_2 and S_0K_2 applications provided the highest values in terms of growth and yield. The crude protein content and yield were also positively affected by the applications, with the highest value determined in the silicon-free application at a potassium dose of 40 mM (S_0K_2). When evaluated in terms of feed quality, the ADF and NDF ratios were reduced thru the applications, which was positively evaluated in terms of the plant's digestibility. In conclusion, silicon and potassium applications have been effective in improving the growth, yield, and quality parameters of *Puccinella distans*, especially under saline-alkali soil conditions. These findings make significant contributions to the sustainability of agricultural production in these areas.

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Conflict of interest

The authors declare no conflict of interest.

Ethical Approval

This arcticle does not require ethics committee approval.

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Species composition of seaweed biofoulers in fish cage aquaculture in Bongao Channel, Tawi-Tawi, Philippines

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Abstract

The excessive growth of seaweeds (macroalgal biofouling) on submerged cage structures is a severe operational constraint for fish cage aquaculture in tropical environments. This study investigated the species composition of macroalgal biofouling growing on the submerged structures of the fish cage aquaculture in Bongao Channel, Tawi-Tawi, Philippines, via a qualitative survey (e.g., snorkeling around the cage systems). The primary objective was to provide a taxonomic inventory of the colonizing seaweed species to understand the biological makeup of the fouling community. Through morphological examination of the collected samples, a total of 26 species belonging to 16 distinct genera were identified, representing the three major algal divisions: Chlorophyta (green algae) exhibited the highest generic richness, including *Boergesenia*, *Caulerpa*, *Chaetomorpha*, *Cladophora*, *Halimeda*, *Boodlea*, and *Ulva*. Rhodophyta (red algae) were represented by *Acanthophora*, *Actinotrichia*, *Chondrophycus*, *Yonagunia*, *Gracilaria*, *Hypnea*, and *Endosiphonia*, while Ochrophyta (brown algae) included *Padina* and *Sargassum*. The presence of various taxa, particularly filamentous *Chaetomorpha* and stoloniferous *Caulerpa*, identifies these groups as the key structural components of the biofouling community on the cage nets. Additionally, the inventory recorded the occurrence of economically valuable red algae (e.g., *Gracilaria* and *Hypnea*) and ecologically significant brown algae (e.g., *Sargassum*) within the assemblage. These findings establish a critical taxonomic baseline, providing the essential species-level data required to develop targeted mitigation strategies and sustainable management practices for the floating cage aquaculture in Tawi-Tawi.

Keywords: Aquaculture, Biofouling, Macroalgae, Seaweeds

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1. Introduction

Aquaculture is a critical pillar of global food security, evolving through intensive production systems to meet the rising demand for aquatic products (Bostock et al., 2010; Verdegem et al., 2023; Tahiluddin et al., 2025). Among these systems, cage culture remains a cornerstone of Asian aquaculture, originating from traditional practices in the Mekong Basin and evolving into a sophisticated global industry (De Silva & Phillips, 2007). Floating net cages are particularly advantageous in tropical regions because they utilize natural water currents for oxygenation and waste removal, theoretically eliminating the need for active water-quality management (Nagler et al., 2003; Alcantara & Noro, 2006).

However, the continuous immersion of aquaculture infrastructure in nutrient-rich tropical waters facilitates the rapid colonization of submerged surfaces by a wide array of organisms. This phenomenon, known as biofouling, represents one of the most significant operational constraints in the aquaculture industry (Bannister et al., 2019). Macroalgal biofouling, specifically, creates a physical barrier on nets, ropes, and frames. The accumulation of these seaweeds restricts water exchange, increases hydrodynamic drag on the structures, and may lead to localized depletions of dissolved oxygen (Madin et al., 2010; Fitridge et al., 2012; Mascorda Cabre et al., 2021). Furthermore, heavy fouling can decrease the effective volume of the cage by causing net deformation, thereby increasing stocking density and stressing the cultured fish (Fitridge et al., 2012).

Tawi-Tawi, located at the heart of the Coral Triangle in the southern Philippines, is a region of immense marine biodiversity (Muallil et al., 2020). The Bongao Channel serves as a vital waterway in this province, supporting a high richness of wild fish, corals, and macroalgae. While the cage culture industry in Tawi-Tawi has expanded gradually over the last two decades (Imlani et al., 2022), it faces persistent challenges from local biofouling communities. At the Mindanao State University–Tawi-Tawi College of Technology and Oceanography (MSU-TCTO) experimental floating cages, seaweed colonization is a visible and constant factor affecting cage maintenance.

Despite the clear operational impacts of biofouling, there is a notable lack of site-specific data regarding the taxonomic makeup of these communities in Tawi-Tawi. Understanding the species composition is a prerequisite for any mitigation strategy, as different seaweed taxa (e.g., filamentous vs. leathery algae) have varying rates of growth and impacts on water flow. Currently, baseline information on the macroalgal biofoulers of the Bongao Channel remains fragmented.

The present study aimed to address this knowledge gap by documenting the species composition and generic richness of macroalgal biofoulers on the MSU-TCTO floating fish cages. By establishing this taxonomic baseline, this research provides the essential biological data needed to develop sustainable management practices and effective fouling mitigation strategies for the burgeoning aquaculture industry in the region.

2. Material and Method

2.1. Study area

The study was conducted at the experimental floating fish cage facility of MSU-TCTO, situated within the Bongao Channel, Tawi-Tawi, southern Philippines (Figure 1). This area is characterized by high marine biodiversity and constant water exchange, making it a representative site for tropical small-scale cage aquaculture.

2.2. Sample collection

A qualitative survey of attached seaweeds was conducted via snorkeling to document the species composition on various submerged substrates, including nets, mooring ropes, and floating HDPE drums.

This study was designed as a preliminary taxonomic baseline assessment to provide a high-resolution snapshot of the fouling community during September, e.g., the wet season. While macroalgal communities exhibit seasonal shifts, this single-event sampling focused on achieving a comprehensive inventory of the taxa present during peak fouling periods to establish a primary reference for the region. Collected specimens were placed in labeled polyethylene bags with ambient seawater and transported immediately to the Seaweed Post-Harvest Laboratory, College of Fisheries, for processing. To maintain the study's focus on taxonomic richness, environmental parameters were not recorded, as the primary objective was the establishment of a species-level checklist.

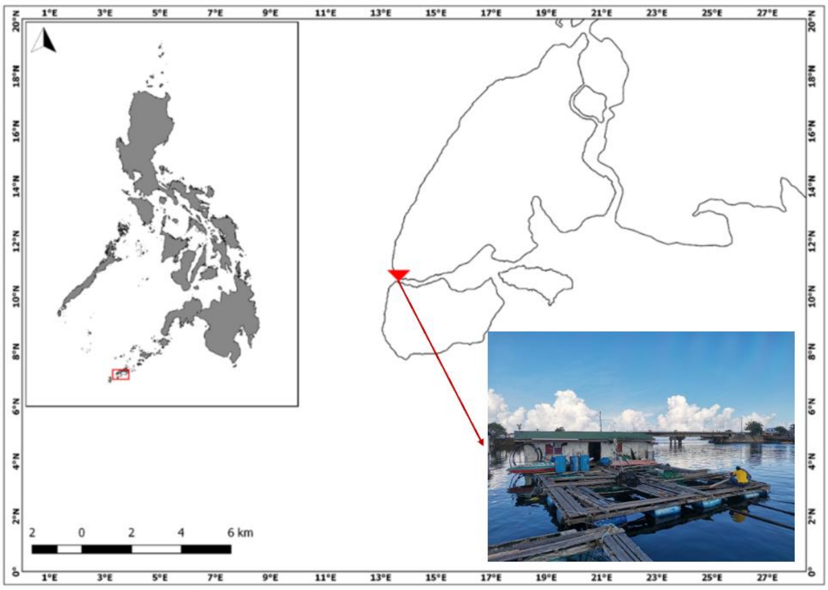

Figure 1. Map of Bongao, Tawi-Tawi, Philippines showing the MSU-TCTO floating fish cage

2.3. Laboratory procedures and species identification

Sorting and cleaning

Upon arrival at the laboratory, the macroalgal samples were allowed to acclimate in seawater for 30 minutes to maintain tissue integrity. The specimens were then meticulously cleaned of debris, sediment, and mobile epifauna using filtered seawater, followed by a brief rinse in distilled water to remove external salts.

The samples were processed into herbarium vouchers following standard phycological techniques (Tsuda & Abbott, 1985). Specimens were air-dried at room temperature (25-27 °C) for approximately one hour before being arranged on acid-free paper. The specimens were then pressed using a traditional wooden plant press, with blotting paper and corrugated cardboard changed every 24 hours to prevent fungal growth and ensure rapid desiccation. This process was maintained for three to five days until the specimens were fully dehydrated.

Taxonomic identification

The identification of the macroalgal biofoulers was based on a detailed examination of morphological and vegetative characters. Diagnostic features such as branching patterns, thallus structure, and specialized attachment organs (holdfasts) were analyzed. Taxonomic nomenclature and classification were verified using the following authoritative references: AlgaeBase (Guiry & Guiry, 2025) for current valid names and authorities, and Field Guide and Atlas of the Seaweed Resources of the Philippines (Trono, 1997) for regional morphological variations.

3. Results and Discussion

A total of 26 macroalgal species belonging to 16 genera were identified from the MSU-TCTO floating fish cage facility (Table 1). Chlorophyta (green algae) exhibited the highest taxonomic richness with 14 species (54% of the total), followed by Rhodophyta (red algae) with 8 species (31%), and Ochrophyta (brown algae) with 4 species (15%).

The Chlorophyta group was represented by seven genera: *Boergesenia*, *Caulerpa*, *Chaetomorpha*, *Cladophora*, *Halimeda*, *Boodlea*, and *Ulva*. As illustrated in Figure 2, these taxa ranged from stoloniferous forms to filamentous mats. Notably, *Caulerpa* and the filamentous *Chaetomorpha* were the most prominent genera observed colonizing the ropes and nets of the aquaculture structure.

The Ochrophyta were represented by two genera, *Padina* and *Sargassum* (Figure 3). These brown algae are characterized by their larger, more complex thalli compared to the green algae found on the site. Finally, the Rhodophyta displayed a diverse generic composition with seven identified genera: *Acanthophora*, *Actinotrichia*, *Chondophycus*, *Yonagunia*, *Gracilaria*, *Hypnea*, and *Endosiphonia* (Figure 4).

The prevalence of Chlorophyta in the Bongao Channel suggests a highly productive environment characterized by shallow depths and high light penetration. Genera such as *Caulerpa*, *Ulva*, and *Halimeda* are ubiquitous in tropical reef-associated ecosystems in the Philippines (Trono, 1997), where stable substrates—in this case, aquaculture infrastructure—allow for rapid colonization (Mineur et al., 2012). The presence of a taxonomically rich assemblage, including climax-stage genera like *Sargassum*, indicates relatively stable environmental conditions and minimal anthropogenic disturbance within the channel (Littler & Littler, 1984; Steneck & Dethier, 1994; Schaffelke et al., 2007).

While biofouling communities can act as natural filtration systems, their proliferation on aquaculture systems poses significant technical risks. As noted by Fitridge et al. (2012), the primary concerns involve the restriction of water exchange and the potential for cage deformation under increased hydrodynamic loads.

Table 1. Taxonomic checklist of seaweed biofoulers in the MSU-TCTO floating fish cage

No.	Phylum	Genus	Species
1	Chlorophyta	*Boergesenia*	*Boergesenia forbesii*
2		*Caulerpa*	*Caulerpa brachypus*
3			*Caulerpa nummularia*
4			*Caulerpa oligophylla*
5			*Caulerpa racemosa*
6		*Chaetomorpha*	*Chaetomorpha linum*
7			*Chaetomorpha crassa*
8		*Cladophora*	*Cladophora* sp.
9		*Halimeda*	*Halimeda cuneata*
10			*Halimeda discoidea*
11			*Halimeda copiosa*
12		*Boodlea*	*Boodlea composita*
13		*Ulva*	*Ulva reticulata*
14			*Ulva lactuca*
15	Ochrophyta	*Padina*	*Padina gymnospora*
16		*Sargassum*	*Sargassum aquifolium*
17			*Sargassum oligocystum*
18			*Sargassum polycystum*
19	Rhodophyta	*Acanthophora*	*Acanthophora spicifera*
20		*Actinotrichia*	*Actinotrichia fragilis*
21		*Chondrophycus*	*Chondrophycus* sp.
22		*Yonagunia*	*Yonagunia millardii*
23		*Gracilaria*	*Gracilaria salicornia*
24			*Gracilaria corticata*
25		*Hypnea*	*Hypnea musciformis*
26		*Endosiphonia*	*Endosiphonia* sp.

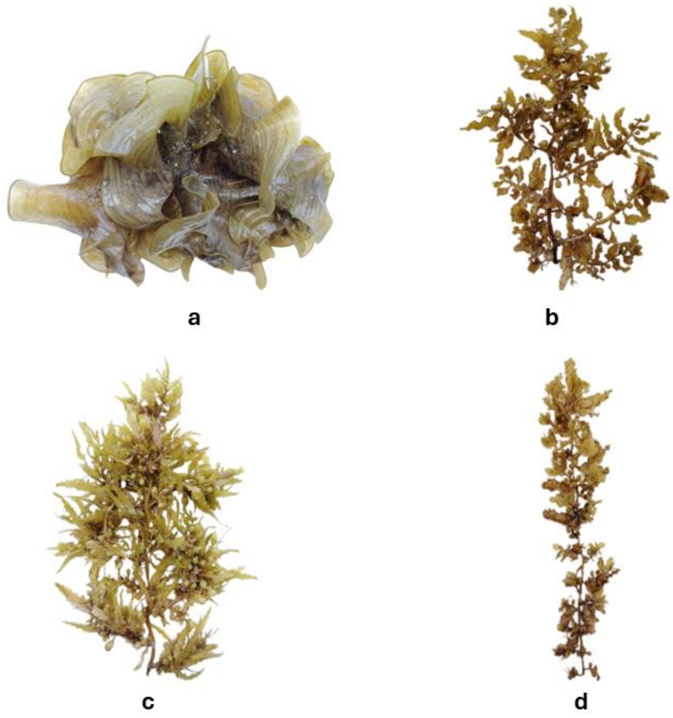

Figure 2. Ochrophyta, the brown seaweeds. a) *Padina gymnospora*, b) *Sargassum aquifolium*, c) *Sargassum oligocystum*, and d) *Sargassum polycystum*

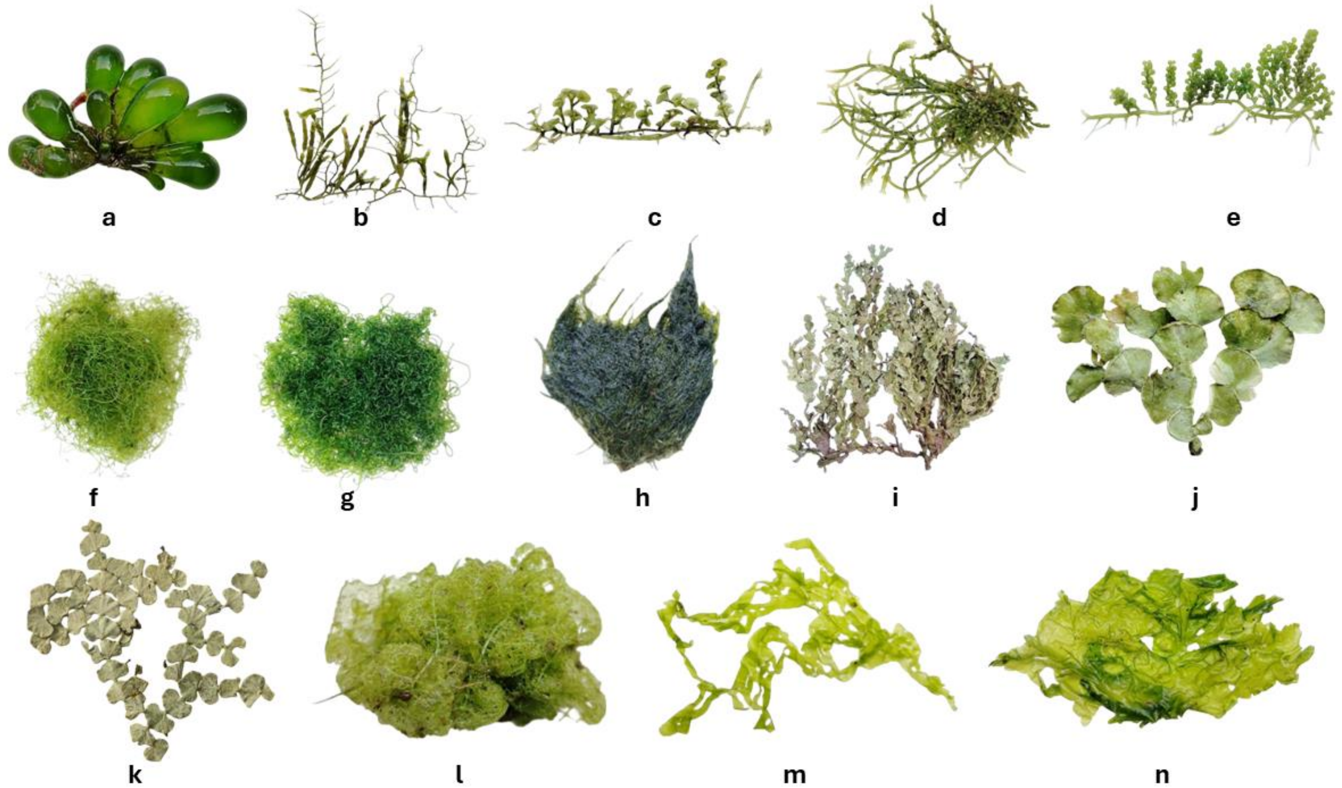

Figure 3. Chlorophyta, the green seaweeds. a) *Boergesenia forbesii*, b) *Caulerpa brachypus*, c) *Caulerpa nummularia*, d) *Caulerpa oligophylla*, e) *Caulerpa racemosa*, f) *Chaetomorpha linum*, g) *Chaetomorpha crassa*, h) *Cladophora* sp., i) *Halimeda cuneata*, j) *Halimeda discoidea*, k) *Halimeda copiosa*, l) *Boodlea composita*, m) *Ulva reticulata*, and n) *Ulva lactuca*

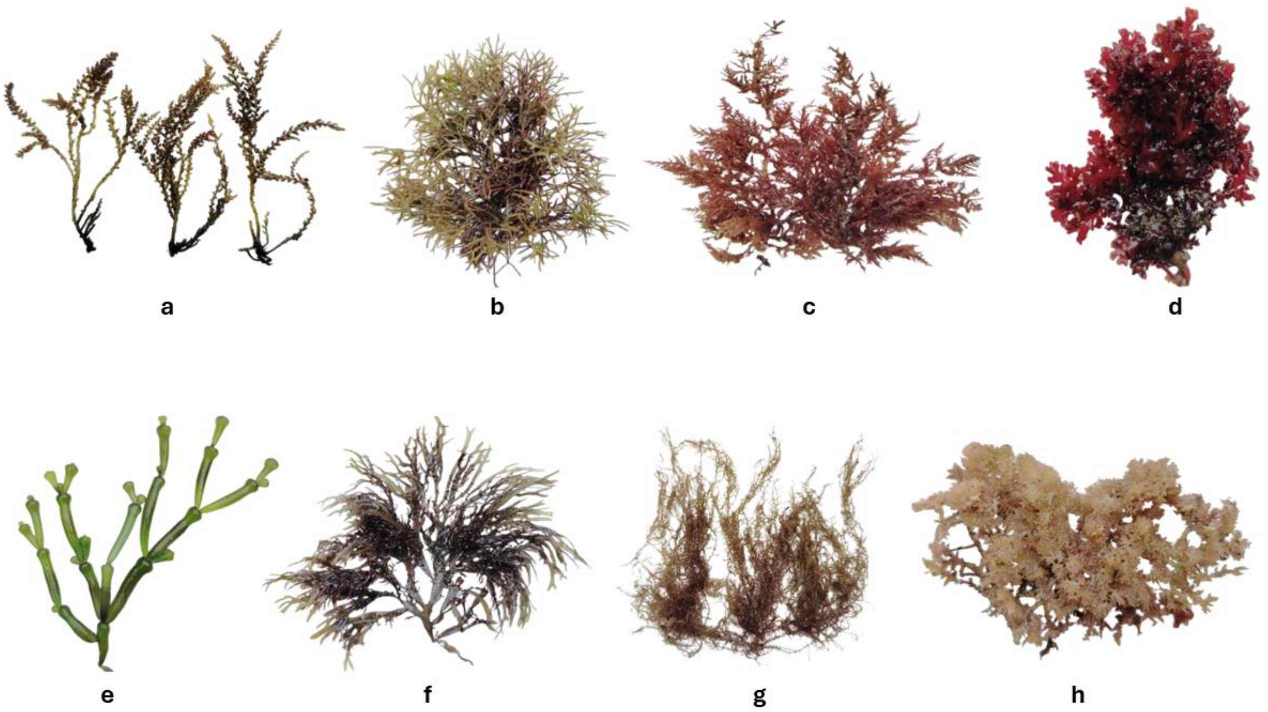

Figure 4. Rhodophyta, the red seaweeds. a) *Acanthophora spicifera*, b) *Actinotrichia fragilis*, c) *Chondrophycus* sp., d) *Yonagunia millardii*, e) *Gracilaria salicornia*, f) *Gracilaria corticata*, g) *Hypnea musciformis*, and h) *Endosiphonia* sp.

Observations from cage personnel at the MSU-TCTO facility confirm that while seaweed attachment does not appear to directly inhibit fish growth, it significantly reduces the effective culture volume. This "occupational fouling" can lead to localized reductions in water flow, hindering the removal of metabolic wastes and potentially facilitating the transmission of pathogens by providing a biological substrate for viral or bacterial hosts (Fitridge et al., 2012).

Conversely, the fouling community provides several ecosystem services that enhance the sustainability of the floating cage system. The identified *Sargassum* species are ecologically vital, acting as complex three-dimensional habitats and nursery grounds for juvenile fish and invertebrates (Casazza & Ross, 2008). These brown algae concentrate prey resources and offer protection from predators, effectively turning the aquaculture site into a localized biodiversity hotspot.

Furthermore, the documentation of Rhodophyta genera such as *Gracilaria*, *Hypnea*, and *Acanthophora* underscores the economic potential of the Bongao Channel. These taxa are high-value sources of agar and carrageenan, polysaccharides with increasing demand in the global food and industrial sectors (McHugh, 2003; Bixler & Porse, 2011; Jayakody et al., 2022). The natural abundance of these red algae suggests that the biofouling present on aquaculture structures could be viewed not just as a waste product, but as a potential secondary resource for sustainable seaweed farming ventures in the region.

4. Conclusion

This study provides the first comprehensive taxonomic checklist of seaweed biofoulers in the MSU-TCTO floating fish cage facility in Bongao Channel, Tawi-Tawi, Philippines. A total of 26 species were identified, with Chlorophyta being the most taxonomically diverse group. The results confirm that the fouling community is composed of a mix of opportunistic green algae, ecologically significant brown algae, and economically valuable red algae. As a baseline inventory, these findings provide the necessary biological data for local aquaculture managers to design targeted mitigation strategies, such as mesh size adjustments or scheduled mechanical cleaning, to maintain optimal cage performance in the Bongao Channel.

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Conflict of interest

The authors declare no conflict of interest.

Ethical Approval

This arcticle does not require ethics committee approval.

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Parenthetical citation:

(Sakici, 2020), (Unal & Sakici, 2020), (Bayram et al., 2020), (Sakici, 2020; Bayram et al., 2021)

References

The references should be written in APA Style. https://apastyle.apa.org/

Journal article:

Sakici, O. E., Saglam, F., & Seki, M. (2018). Single- and Double-entry Stem Volume Equations for Crimean Pine Stands in Kastamonu Regional Directorate of Forestry. *Turkish Journal of Forestry*, *19*(1), 20-29. https://doi.org/10.18182/tjf.394876

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Husch, B., Beers, T. W., & Kershaw, J. A. (2003). *Forest mensuration* (4th ed.). Wiley.

Chapter in an edited book:

Köhl, M. (2004). Forest inventory and monitoring. In J. Burley, J. Evans, & J. A. Youngquist (Eds.), *Encyclopedia of Forest Science* (pp. 403-409). Elsevier.

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Sakıcı, O. E., & Gulsunar, M. (2012). *Diameter distributions of Bornmullerian fir in mixed stands* [Oral presentation]. 14th International Fir Symposium, Kastamonu, Turkey.

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Saglam, F. (2016). *Constructing of above-ground biomass tables and developing of compatible biomass-volume equations for black pine (Pinus nigra J.F. Arnold) stands in Taskopru Forest Enterprise* [Master's thesis, Kastamonu University].

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