RESEARCH PAPER

Relationship between the Survival of White Spot Syndrome Virus (WSSV) and the Physico-Chemical Properties of Soil at Different Depths in Brackish water Ecosystems

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ABSTRACT

White Spot Syndrome Virus (WSSV) was first reported as an epidemic in penaeid shrimp farms in China in 1993 and soon spread to other countries in Asia and eventually worldwide. The survival and infectivity of shrimp populations are influenced by both environmental and biological conditions. Understanding the physico-chemical properties of pond sediment and water, as well as the interactions among these components and the resident organisms, is crucial for effective shrimp pond management and cultivation. In this study, soil characteristics such as texture, moisture, pH, organic carbon content, and electrical conductivity were analyzed in three different shrimp farms (Farms 1, 2, and 3). Moisture levels were highest in Farm 1 throughout the sampling period, while Farms 2 and 3 showed moderate levels. Electrical conductivity remained within acceptable limits in all farms, although Farm 3 exhibited relatively higher values across all soil layers. Organic carbon content in the pond soils was low, with all samples measuring less than 0.5%, and soil pH fell within the recommended range for shrimp farming. PCR analysis confirmed the persistence of WSSV in pond soils for up to 33 days. Soil samples from all three farms were positive for WSSV from day 1 to day 21 in all layers, but the virus was undetectable on days 27 and 33. The viral load declined from an average of 654,300 g^{-1} on day 1 to 517 g^{-1} by day 21, with no significant counts detected afterward. Overall, the measured soil parameters indicate that the pond conditions are suitable for shrimp culture, suggesting the potential for high production yields.

Key words: Soil, Brackish water, WSSV, Viability, Different depth, Physico chemical properties

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Introductions

The aquaculture sector is one of the fastest-growing segments within the food industry. In India, shrimp production has shown a consistent upward trend from 1990 to 2003 (Gnana Selvam et al., 2012). Aquaculture plays a critical role in providing a reliable supply of aquatic species for human consumption, as wild fisheries cannot meet the increasing demand from a growing population. Consequently, aquaculture not only fulfills human needs but also reduces pressure on wild stocks, allowing them to remain a valuable resource.

The sustainability of shrimp aquaculture is increasingly threatened due to ecological and pathological challenges in most shrimp-producing countries. Shrimp production is frequently impacted by environmental degradation as well as infectious and non-infectious diseases (Bachere, 2000). Among infectious diseases, bacterial and viral infections—whether single or multiple pathogens—are responsible for the majority of production losses. White Spot Syndrome Virus (WSSV) first emerged as an epidemic in penaeid shrimp farms in China in 1993 and rapidly spread across Asia and then globally (Chou et al., 1995).

WSSV is a highly virulent, large, enveloped, double-stranded DNA (dsDNA) virus that replicates rapidly in shrimp. The International Committee on Taxonomy of Viruses (ICTV) classifies it under the genus Whispovirus within the family Nimaviridae

(Fauquet et al., 2005). Since its emergence in 1992, WSSV has caused estimated global economic losses exceeding US$6 billion in shrimp aquaculture (Lightner et al., 2012). Reports indicate widespread prevalence of WSSV in both wild and cultured shrimp populations. For example, wild Penaeus monodon in Chennai, Tamil Nadu showed a prevalence of 56.2%, whereas Digha, West Bengal and Visakhapatnam, Andhra Pradesh recorded 10.9% and 0.6%, respectively (Dutta et al., 2013). In contrast, studies along the Pacific coast of Mexico reported WSSV prevalence of about 3.6% in wild invertebrates such as crabs and various shrimp species, as well as in vertebrates near shrimp farms (Macias-Rodriguez et al., 2014).

In culture ponds, shrimp are vulnerable to disease, which often occurs when the balance between shrimp and pathogens is disrupted, typically due to environmental stress. Horowitz and Horowitz (2003) recommended enhancing environmental and biological conditions to strengthen disease resistance in infected populations. Suggested measures include: (a) physical improvements, such as better aeration, temperature control, optimized feeding, sludge and organic matter removal, and wastewater treatment; (b) chemical measures, including management of pH and salinity, reduction of ammonia and nitrite, and selective use of antibiotics; and (c) biological strategies, mainly employing probiotics to establish beneficial microbial communities.

Studies on viruses in soil have largely focused on their fate, movement, and detection, particularly for pathogenic viruses introduced externally. Factors affecting viral adsorption include soil solution properties such as ionic strength, pH, and dissolved organic matter; virus characteristics like isoelectric point and hydrophobicity; and soil features including water content, clay and organic matter content, and the presence of organic coatings. These findings have been critical in understanding virus-soil interactions and improving detection methods.

Metagenomic studies have revealed that marine sediments contain the most diverse viral assemblages known. While viruses clearly influence bacterial mortality in soils, the broader ecological roles of viruses in terrestrial soils remain poorly understood. Compared to marine systems, the ecological significance of viruses in soils is still largely unknown. Detailed knowledge of sediment and water physico-chemical properties, their interactions, and ecological relationships among organisms is essential for assessing pond suitability and managing shrimp aquaculture effectively (Pankaj Kumar et al., 2012).

In this study, WSSV abundance in soil was assessed using PCR-based detection. Viral abundance was analyzed in relation to soil samples to determine correlations with abiotic and biotic soil factors. Virus survival in soil is influenced by multiple factors, including virus type, physical state, suspended particles, organic matter, salts, pH, antiviral chemicals, and relative humidity, moisture, and water activity.

Materials and Methods

Soil Sampling

A total of 10 kg of soil was collected from Farm 1 (Ponds A, B, and C) located at Sadras, Kalpakkam, South Chennai, Tamil Nadu; Farm 2 (Ponds A, B, and C) located at Marakkanam, north of Pondicherry, Tamil Nadu; and Farm 3 (Ponds A, B, and C) located at Gangapattinam, Nellore, Andhra Pradesh. The collected soils were pooled, thoroughly mixed, and sun-dried for four days.

The dried soil was then mixed with three liters of sterile water. Subsequently, 15 mL of WSSV-positive extract was diluted in 500 mL of TNE buffer. The diluted WSSV mixture was thoroughly mixed with the soils collected from Farms 1, 2, and 3. The mixed soil was transferred into 20 L capacity buckets (23 cm diameter and 37 cm height) containing 21 cm depth of soil. The buckets were labeled as Farm 1, Farm 2, and Farm 3, and were kept under sunlight for drying. This procedure was carried out in triplicate for soils from Farm 1, Farm 2, and Farm 3.

Soil Analysis

Soil cores were collected using a hand-operated core sampler with a diameter of 5 cm. Core samples were taken from the buckets using the sampler. The collected soil cores were divided into three portions: the first 7 cm was designated as the upper layer, the second 7 cm as the middle layer, and the last 7 cm as the bottom layer. This procedure was carried out on seven different sampling days: day 1, 3, 9, 15, 21, 27, and 33. The core segments were transported to the laboratory for further analysis.

Soil Physico-Chemical Properties and WSSV Viability Experiments

Experiments on soil physico-chemical properties and WSSV counts in soil were conducted using soil samples collected from three sets of buckets (Farm 1, Farm 2, and Farm 3). Soil samples collected on days 1, 3, 9, 15, 21, 27, and 33 from the upper, middle, and bottom layers were analyzed for moisture, texture, pH, electrical conductivity, and organic carbon content using standard methods (Jackson, 1973) at the soil testing laboratory of the Central Institute of Brackishwater Aquaculture (CIBA, ICAR), Chennai.

The viability of WSSV in the upper, middle, and bottom soil layers on days 1, 3, 9, 15, 21, 27, and 33 was determined. DNA from soil samples was extracted following the method described by Satheesh Kumar et al. (2013), and the presence of

WSSV was confirmed using the PCR protocol recommended by OIE (2012). The WSSV DNA copy number was estimated using a WSSV detection and quantitative real-time PCR kit (LabIndia Life Sciences, Gurgaon, India) according to the manufacturer's instructions, using an Applied Biosystems StepOne™ Real-Time PCR system (California, USA).

The WSSV DNA copy number in the original stock was estimated to be 12,070,299 copies μL^{-1}, which was diluted 60,000-fold in TNE buffer to obtain a working suspension containing 186 WSSV copies μL^{-1}, as determined by quantitative real-time PCR.

Results

Soil Physico-Chemical Properties

Soil texture, moisture, pH, organic carbon, and electrical conductivity were estimated for Farms 1, 2, and 3. The parameters recorded in the ponds were found to be suitable for shrimp culture. The details of the soil physico-chemical parameters are presented in the following table, and comparisons are illustrated in the graph.

Soil Texture

The soil texture of Farm 1 was classified as sandy clay loam, Farm 2 also exhibited sandy clay loam soil, while Farm 3 showed sandy soil texture. These classifications were determined based on the relative proportions of sand, silt, and clay obtained through soil texture analysis (Table 1)

Farm-1 Sandy clay loam			Farm-2 Sandy clay loam			Farm-3 Sandy loam		
Sand (%)	Silt (%)	Clay(%)	Sand(%)	Silt(%)	Clay(%)	Sand (%)	Silt(%)	Clay(%)
87.6	0.1	32.3	72.5	7.28	20.23	74.53	7.63	17.85

Table 1: Soil texture determination

Days	Pond A(%)			Pond B(%)			Pond C(%)		
	Upper	Middle	Bottom	Upper	Middle	Bottom	Upper	Middle	Bottom
1	25	25	25	22.5	22.5	22.5	20	20	20
3	20.5	22.2	24.57	20.5	21.5	22	17.5	18.5	19
9	17.5	20	22.5	17.5	19	20	15	16.5	17
15	5	5.5	6.5	2.5	3	3.5	1	1	1.5
21	0.5	0.5	1.5	0	0.5	1	0	0	0
27	0	0	0.5	0	0	0	0	0	0
33	0	0	0	0	0	0	0	0	0

Table 2: Soil Moisture analysis

Soil Moisture

Soil moisture content refers to the quantity of water present in the soil. At low relative humidity, moisture mainly consists of adsorbed water, whereas at higher relative humidity, liquid water becomes increasingly important depending on pore size. The effect of moisture on the survival of viruses in the environment varies with virus type. Viruses with higher lipid content tend to be more persistent at lower relative humidity, while viruses with little or no lipid content are generally more stable at higher relative humidity. In the present study, the moisture content in Farm 1 was 25% in the upper and middle layers on the first day of sampling and decreased to 0.5% on the 21st day. No moisture (0%) was recorded from the 27th to the 33rd day. In the bottom layer of Farm 1, the moisture content was 25% on the first day and decreased to 0.5% on the 27th day. In Farm 2, the upper layer showed 22.5% moisture on the first day, which decreased to 2.5% on the 15th day. The middle layer recorded 22.5% on the first day and decreased to 0.5% on the 21st day. In the bottom layer, the moisture content was 22.5% on the first day and decreased to 1% on the 21st day. In Farm 3, the upper layer showed 20% moisture on the first day and decreased to 1.5% on the 15th day. Similarly, the middle layer recorded 20% on the first day and decreased to 1.5% on the 15th day. In the bottom layer, the moisture content was 20% on the first day and decreased to 0.5% on the 21st day of the experiment (Table 2)

The present study indicated that the moisture content was highest in Farm 1 throughout the sampling period, while intermediate levels were observed in Farms 2 and 3. Enveloped viruses such as WSSV tend to survive better under higher moisture conditions than under intermediate moisture levels.

Days	Farm-1			Farm-2			Farm-3		
	Upper	Middle	Bottom	Upper	Middle	Bottom	Upper	Middle	Bottom
1	2.1	2.09	2.46	3.07	3.32	3.38	12.68	10.89	11.95
3	2.75	2.4	2.31	3.09	3.07	3.82	12.64	10.92	11.33
9	3.91	1.86	2.1	3.21	3.38	3.58	13.49	11.68	11.25

15	3.9	2.75	2.12	3.94	3.44	3.65	12.31	12.2	10.89
21	3.92	2.78	2.17	3.91	3.41	3.61	12.65	12.89	11.04
27	3.86	2.69	2.4	3.78	3.53	3.74	12.75	12.26	10.97
33	3.94	2.87	2.43	4.02	3.49	3.96	12.96	12.28	11.06

Table 3: Soil Electrical conductivity analysis

Electrical Conductivity

Electrical conductivity (EC) is a measure of how well a solution conducts electricity and is closely related to the salt content present in the medium. It can also be used to provide a rough estimate of the total dissolved solids (TDS) in water. Changes in EC are associated with the release or depletion of soluble ions in soil–water systems. Therefore, EC may play an indirect role in shrimp pond productivity (Mandal and Chattopadhyay, 1990). In the present study, the EC values in Farm 1 ranged from 2.1 to 3.9 in the upper layer, 2.09 to 2.87 in the middle layer, and 2.43 to 2.6 in the bottom layer. In Farm 2, the EC values ranged from 3.07 to 4.02 in the upper layer, 3.32 to 3.49 in the middle layer, and 3.38 to 3.96 in the bottom layer. In Farm 3, the EC values ranged from 12.68 to 12.96 in the upper layer, 10.89 to 12.28 in the middle layer, and 11.06 to 11.95 in the bottom layer. The present study indicates that the electrical conductivity values of all farms were within acceptable limits for shrimp culture. However, Farm 3 exhibited comparatively higher EC values, although they remained within the desirable range across all soil layers.

Days	Farm-1			Farm-2			Farm-3		
	Upper	Middle	Bottom	Upper	Middle	Bottom	Upper	Middle	Bottom
1	0.22	0.28	0.30	0.17	0.18	0.21	0.13	0.13	0.12
3	0.31	0.30	0.28	0.15	0.17	0.19	0.11	0.13	0.14
9	0.30	0.29	0.26	0.11	0.15	0.15	0.10	0.11	0.15
15	0.32	0.30	0.25	0.15	0.13	0.17	0.11	0.10	0.13
21	0.30	0.31	0.26	0.15	0.13	0.17	0.13	0.12	0.16
27	0.32	0.29	0.25	0.16	0.15	0.17	0.12	0.13	0.15
33	0.29	0.30	0.27	0.17	0.15	0.19	0.12	0.12	0.17

Table 4: Soil organic carbon contents

Organic Carbon

Organic carbon content is an important factor in determining the fertility status of soil. In the present investigation, the organic carbon content in Farm 1 ranged between 0.22–0.32%**,** 0.28–0.31%**,** and 0.25–0.30% in the upper, middle, and bottom soil layers, respectively. In Farm 2**,** the organic carbon content ranged between 0.15–0.17%**,** 0.13–0.18%**,** and **0**.15–0.21% in the upper, middle, and bottom layers, respectively. In Farm 3**,** the organic carbon content ranged between **0.10–0.13%** in all three soil layers.Mantoura reported that brackishwater pond soils are generally poor in organic carbon. Similarly, the present study also indicated that all pond soil samples were poor in organic carbon content, as the values recorded were less than **0**.5% (Table 4)

No of days of sampling and Soil pH

Days	Farm-1			Farm-2			Farm-3		
	Upper	Middle	Bottom	Upper	Middle	Bottom	Upper	Middle	Bottom
1	8.15	7.9	7.38	8.51	7.98	7.99	8.05	7.65	7.28
3	7.28	7.59	7.32	7.82	7.9	7.82	7.63	7.76	7.72
9	7.13	7.57	7.24	7.44	7.5	7.49	7.36	7.38	7.33
15	7.42	7.58	7.59	7.48	7.55	7.56	7.34	7.36	7.39
21	7.54	7.76	7.74	7.46	7.76	7.74	7.38	7.76	7.67
27	7.36	7.64	7.86	7.68	7.71	7.82	7.39	7.63	7.42
33	7.81	7.56	7.98	7.55	7.84	7.58	7.41	7.35	7.86

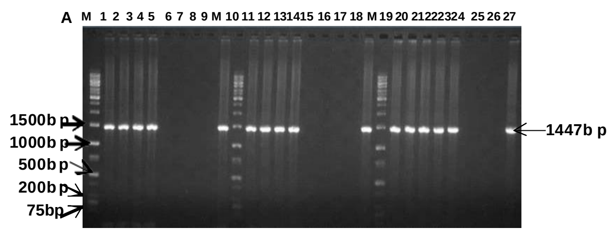
Table 5: No of days of sampling and Soil pH value

Soil pH

The normal pH range required for brackishwater shrimp culture is 6.5–7.5. In Farm 1, the maximum soil pH recorded was 8.15 in the upper layer and 7.9 in both the middle and bottom layers. The minimum pH recorded was 7.13 in the upper layer, 7.56 in the middle layer, and 7.24 in the bottom layer. In Farm 2, the maximum pH recorded was 8.51 in the upper layer and 7.9 in both the middle and bottom layers. The minimum pH recorded was 7.44 in the upper layer, 7.50 in the middle layer, and 7.49 in the bottom layer. In Farm 3, the maximum pH recorded was 8.05 in the upper layer, 7.76 in the middle layer, and 7.86 in the bottom layer. The minimum pH recorded was 7.34 in the upper layer, 7.35 in the middle layer, and 7.33 in the bottom layer (Table 5). The present study indicates that the soil pH values of all ponds were within the desirable limits for shrimp culture. Zafar et al. (2015) reported that soil quality parameters such as pH, organic carbon, and total nitrogen showed no significant differences between farms. Similarly, Abu Hena et al. (2004) reported that the dry soil pH did not show significant differences among the ponds studied throughout the culture period.

WSSV viability in shrimp pond soil

WSSV viability was evident in shrimp pond soil for up to 33 days, as revealed by PCR. Soil samples from Farms 1, 2, and 3 tested positive by first-step PCR from day 1 to day 21 in all soil layers, whereas samples were negative on days 27 and 33 (Fig 1 to 3) (Table 6). The WSSV count in the sediment decreased from an average of 654,300 g^{-1} on day 1 to 517 g^{-1} on day 21. No significant WSSV counts were detected after day 27 (Table 7).



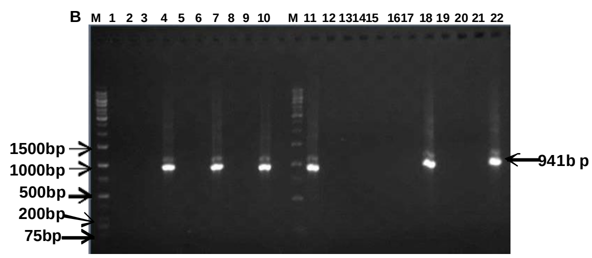


Figure. 1 A and B. WSSV presence in different depth of sandy clay loam (farm-1) soil (A: First step; B: Nested step). 1A: Lane M: 1kb marker, Lane 1to7:1st dayto33 day upper layer; Lane 10to16:1st day to 33rd day middle layer; Lane 19 to 25: 1st day to 33rd day bottom layer sample; Lane 8, 17 and 26: Negative control; Lane 9, 18 and 27: WSSV positive control (1447bp). **1B**. Lane M: 1kb marker; Lane1to10 farm-1, Lane1, 2, 3:27, 27 and 33 day upper layer; Lane 4 ,5 and 6:21, 27 and 33 day middle layer; Lane 7 and 8:27 and 33 day bottom layer. Lane11, 12, 13 and 14: Farm 2-15, 21, 27 and 33rd day upper layer; Lane15, 16 and 17: Farm-2-21, 27 and 33 day middle layer; Lane18, 19 and 20: Farm-2 -21, 27 and 33 day bottom layer. Lane9 and 21: Negative control; Lane10 and 22 WSSV positive control (941bp).

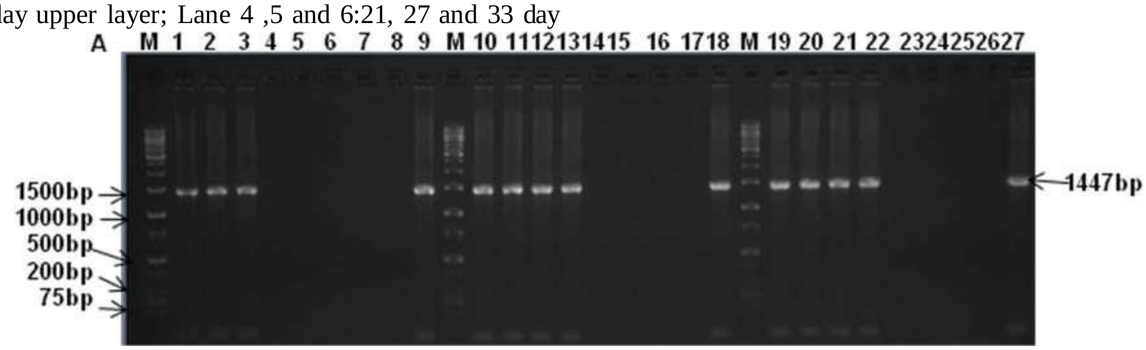
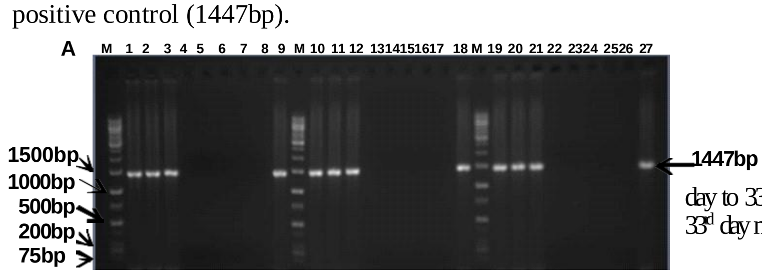
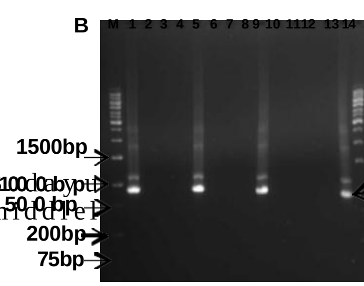


Figure. 2. WSSV presence in different depth of sandy clay loam (farm-2) soil (First step)
Lane M: 1kb marker; Lane 1to7: 1st day to 33 day upper layer; Lane 10 to16:1st day to 33rd day middle layer; Lane 19 to 25: 1st day to 33rd day bottom layer sample; Lane 8, 17 and 26: Negative control; Lane 9, 18 and 27: WSSV positive control (1447bp).





day to 33 ... 6:1st day to 33rd day

Fig. 3 A and B: WSSV presence in different depth of sandy loam soil (Farm-3) (A-First step and B-Nested step) 3 A. Lane M: 1kb marker; Lane 1to7:1st day bottom layer sample; Lane 8, 17 and 26: Negative control; Lane 9, 18 and 27: WSSV positive control (1447bp). 3 B**:** LaneM:1kb Marker; Lane1, 2, 3 and 4:15, 21, 27 and 33 day upper layer; Lane 5, 6, 7 and 8:15, 21 , 27 and 33 day middle layer; Lane9,10,11 and 12: 15, 21, 27 and 33 day bottom layer; Lane 13:Negative control; Lane14: WSSV positive control (941bp).

Days	WSSV presence in farm-1 soil			WSSV presence in farm-2 soil			WSSV presence in farm-3 soil		
	Upper	Middle	Bottom	Upper	Middle	Bottom	Upper	Middle	Bottom
1	+	+	+	+	+	+	+	+	+
3	+	+	+	+	+	+	+	+	+
9	+	+	+	+	+	+	+	+	+
15	+	+	+	+*	+	+	+*	+*	+*
21	ND	+*	+	ND	ND	+*	ND	ND	ND
27	ND	ND	+*	ND	ND	ND	ND	ND	ND
33	ND	ND	ND	ND	ND	ND	ND	ND	ND

Table 6: **Days of sampling and WSV presence in soil by PCR (ND-Not detected; +-positive)**

Days	WSSV count in sediment by real-time PCR(g^{-1}) (Pond A)			WSSV count in sediment by real-time PCR(g^{-1}) (Pond B)			WSSV count in sediment by real-time PCR(g^{-1}) (Pond C)		
	Upper	Middle	Bottom	Upper	Middle	Bottom	Upper	Middle	Bottom
1	654300	654300	654300	654300	654300	654300	654300	654300	654300
3	401050	432115	632115	232115	332115	432115	132115	332115	332175
9	115075	267124	367124	80124	80124	94320	2035	50124	50124
15	876	2012	2012	168	837	1250	35	175	250
21	57	150	517	ND	15	87	ND	ND	ND
27	ND	ND	23	ND	ND	ND	ND	ND	ND
33	ND	ND	ND	ND	ND	ND	ND	ND	ND

Table 7: Days of sampling and WSSV load in Soil by real-time PCR (ND- Not detected)

Discussion

The culture of brackishwater shrimp has increased substantially due to its desirable taste and strong demand in both domestic and international markets. Although shrimp are bottom-dwelling organisms, the depth and volume of water in ponds act as a buffer, mitigating the effects of weather fluctuations on their environment (Gunalan et al., 2010). Abiotic factors such as temperature, salinity, dissolved oxygen, and pH play a critical role in shrimp health and susceptibility to disease, and suboptimal pond conditions often exacerbate WSSV outbreaks in intensive farming systems (Turner et al., 2020). Shrimp, as invertebrates, lack an adaptive immune system like that of vertebrates, making them more vulnerable to environmental stress and infectious agents. Disease outbreaks are frequently linked to degraded environmental conditions that impair innate immune responses (De Souza Valente et al., 2020; Antony et al., 2021). Recent studies have also demonstrated that WSSV infection can disrupt the gut microbiota of shrimp, lowering microbial diversity and potentially reducing disease resistance, which highlights the complex interactions among the environment, host health, and pathogen virulence (Valente et al., 2020). In this study, high soil moisture and slightly lower pH may have contributed to extended WSSV persistence in pond sediments. Viral survival in subsurface environments is strongly influenced by soil moisture, with higher moisture favoring longer survival, while desiccation tends to inactivate viral particles (Gerba, 2005). Soil texture and particle size also affect water retention and adsorption, which can protect viruses from rapid degradation. Our findings showed that WSSV viability was higher in Pond A (sandy clay loam) and Pond B (sandy loam) compared to Pond C (sandy soil), with the highest viral persistence observed in the bottom sediment layers, likely due to better moisture retention and adsorption in finer-textured soils (Sobsey et al., 1986).

Sediments at the pond bottom play a key role in aquaculture pond ecology, as they significantly influence water quality. Variations in electrical conductivity and organic content affect virus survival: higher dissolved salts and organic matter can bind viral particles and reduce their viability, while lower levels may allow viruses to persist longer by reducing inactivation processes (Gerba, 2005; De Gryse et al., 2020). Organic matter also contributes to virus stability, as viral particles associated with organic matrices are better protected from environmental stressors (Sobsey et al., 1986). Although WSSV can persist in pond sediments for long periods, the actual infectivity depends on sediment properties and environmental conditions. WSSV DNA can remain detectable for over 30 days after harvest and may induce chronic infections in shrimp following exposure (De Gryse et al., 2020). In this study, sediments with high initial viral loads (654,300 copies g^{-1}) retained viable virus up to 21 days (517 g^{-1}) despite sun-drying, confirming the durability of viral particles in sediments. Biological vectors also play a role in WSSV persistence. For example, filter-feeding bivalves can accumulate WSSV particles from water, acting as temporary reservoirs that may increase transmission risks to shrimp under high viral concentrations (Min et al., 2024; Akshaya et al., 2021). This emphasizes that both biological interactions and sediment properties contribute to the persistence and spread of WSSV in aquaculture systems. Multiple environmental and biological factors, including proteolytic microorganisms, soil pH, temperature, and salinity, influence viral persistence in soils and sediments (Gerba, 2005). Higher temperatures, for instance, have been shown to reduce viral replication and disease severity in shrimp, suggesting that environmental manipulation may help control viral outbreaks (Elbahnaswy et al., 2025; NP 2020).

In the present study, differences in soil properties among farms showed that Farm 1 had significantly higher ($p \leq 0.05$) soil moisture, organic carbon, and WSSV load compared to other farms. Farm 3 displayed significantly higher electrical conductivity ($p \leq 0.05$), whereas soil pH and organic carbon were relatively consistent across farms. Comparisons among soil layers revealed that moisture and WSSV load were significantly higher in the bottom layer, while EC, pH, and organic carbon remained stable. Over the sampling period, moisture and viral load decreased significantly ($p \leq 0.05$) from day 1 to day 7, whereas other soil parameters showed no significant changes (Fig.4 to Fig 9).

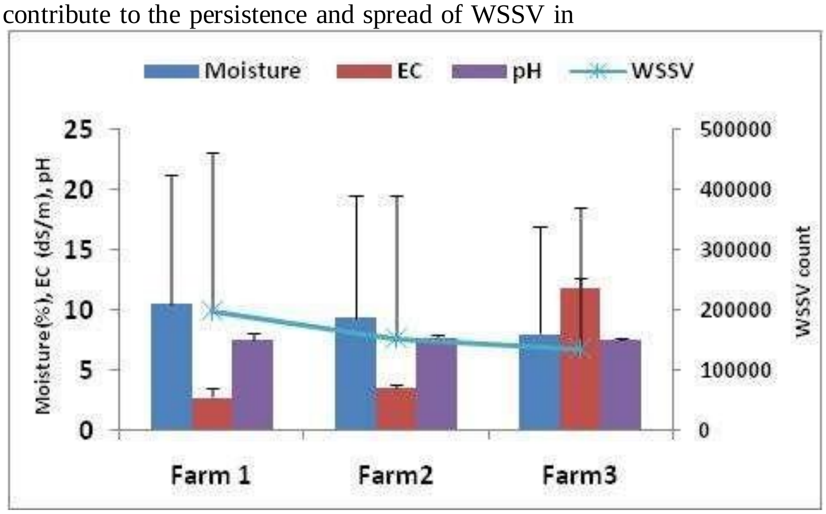


Figure 4. Correlation coefficient (r) between WSSV load and soil Moisture, EC and pH farm wise

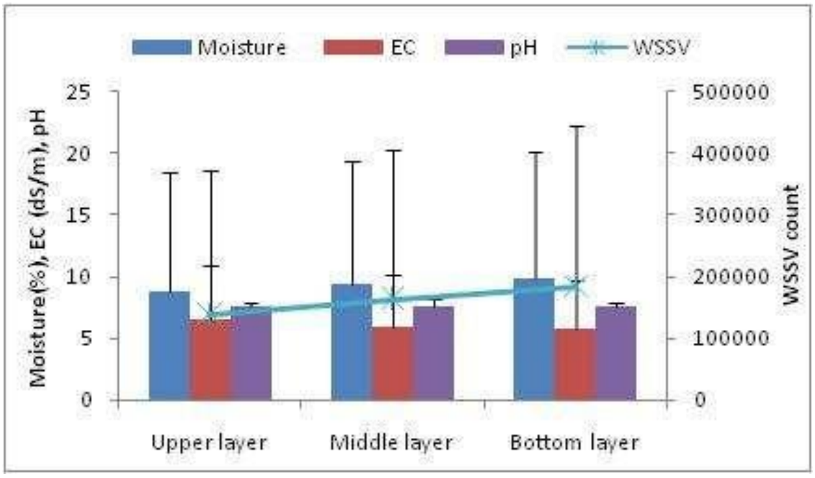


Figure 5. Correlation coefficient (r) between WSSV load and soil Moisture, EC and pH on the soil layer wise

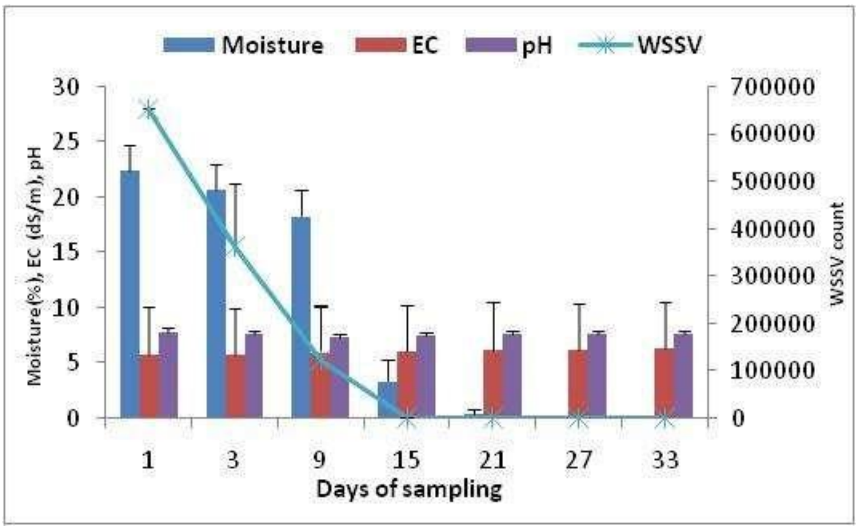


Figure 6. Correlation coefficient (r) between WSSV load and soil Moisture, EC and PH on days of soil sampling

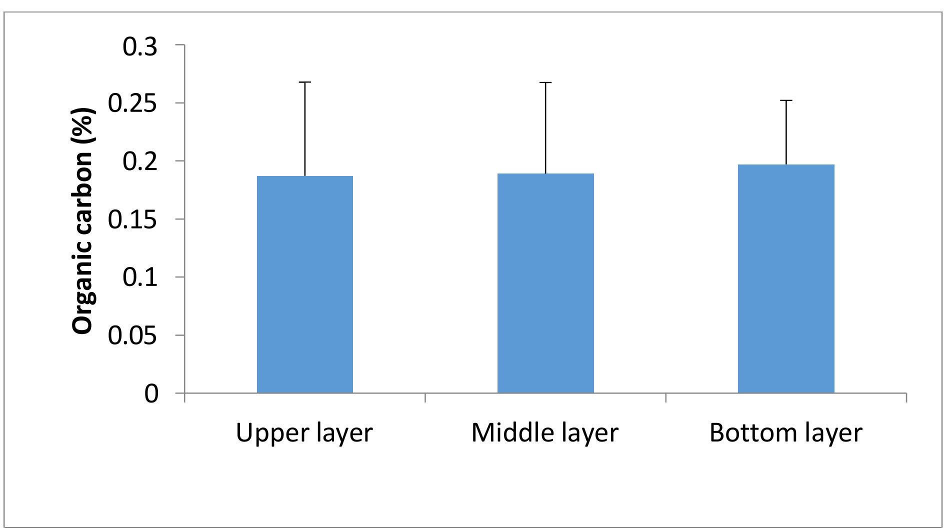


Figure 7. Correlation coefficient (r) between WSSV load and soil organic carbon soil, layer wise

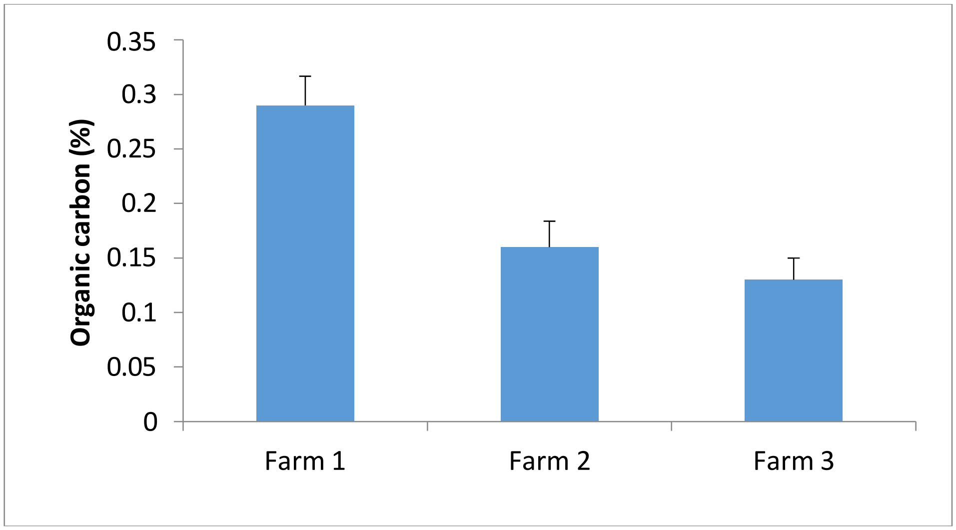


Figure 8. Correlation coefficient (r) between WSSV load and soil organic carbon, farm wise

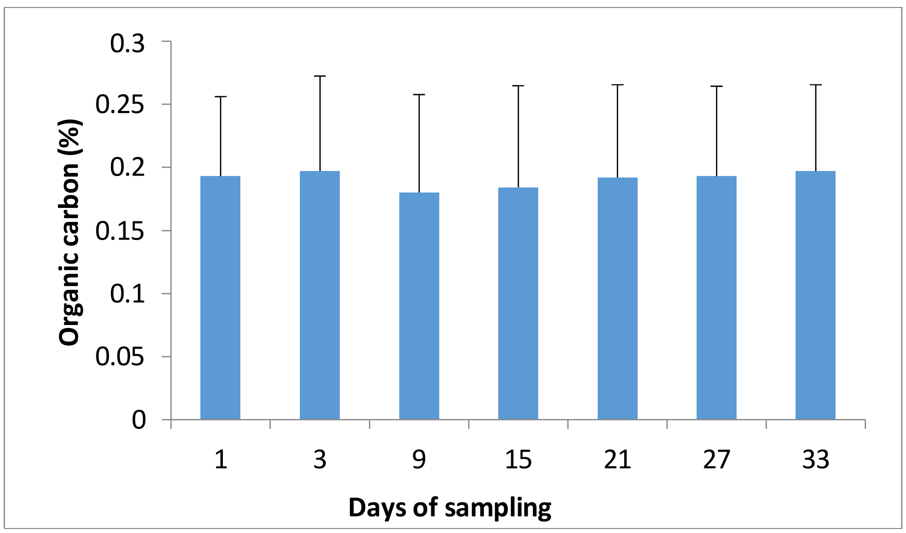


Fig 9. Correlation coefficient (r) between WSSV load and soil organic carbon on the days of sampling

Conclusion

This study demonstrates that White Spot Syndrome Virus (WSSV) can remain viable in shrimp pond sediments for prolonged periods, with the greatest persistence observed in the bottom sediment layers. Soil characteristics, particularly moisture content and texture, were found to strongly influence viral survival. Finer-textured soils, such as sandy clay loam and sandy loam, combined with higher moisture levels, supported longer virus persistence, whereas coarser sandy soils and lower moisture reduced WSSV viability. Organic carbon and electrical conductivity also affected virus survival, with moderate organic matter and lower EC promoting persistence, while higher levels tended to reduce it. Over time, WSSV load and soil moisture decreased significantly within the first week, whereas pH, EC, and organic carbon remained largely unchanged.

These results underscore the complex interactions between physical, chemical, and biological factors that regulate WSSV persistence in pond ecosystems. Effective pond management, including monitoring water quality, managing sediment composition, and maintaining balanced organic matter, can help limit virus survival and reduce the risk of disease outbreaks. Regular sediment management, such as partial removal, treatment of bottom layers, and careful drying practices, can further decrease viral persistence. The study also highlights the importance of farm-specific assessments, as variations in soil properties between ponds can influence viral load and survival. Overall, integrating sediment and water quality management with early disease monitoring is critical to safeguarding shrimp health, improving production efficiency, and minimizing the economic losses caused by WSSV in brackishwater aquaculture.

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Conflict of Interest

Authors declare that no conflict of interest

Funding Declaration

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