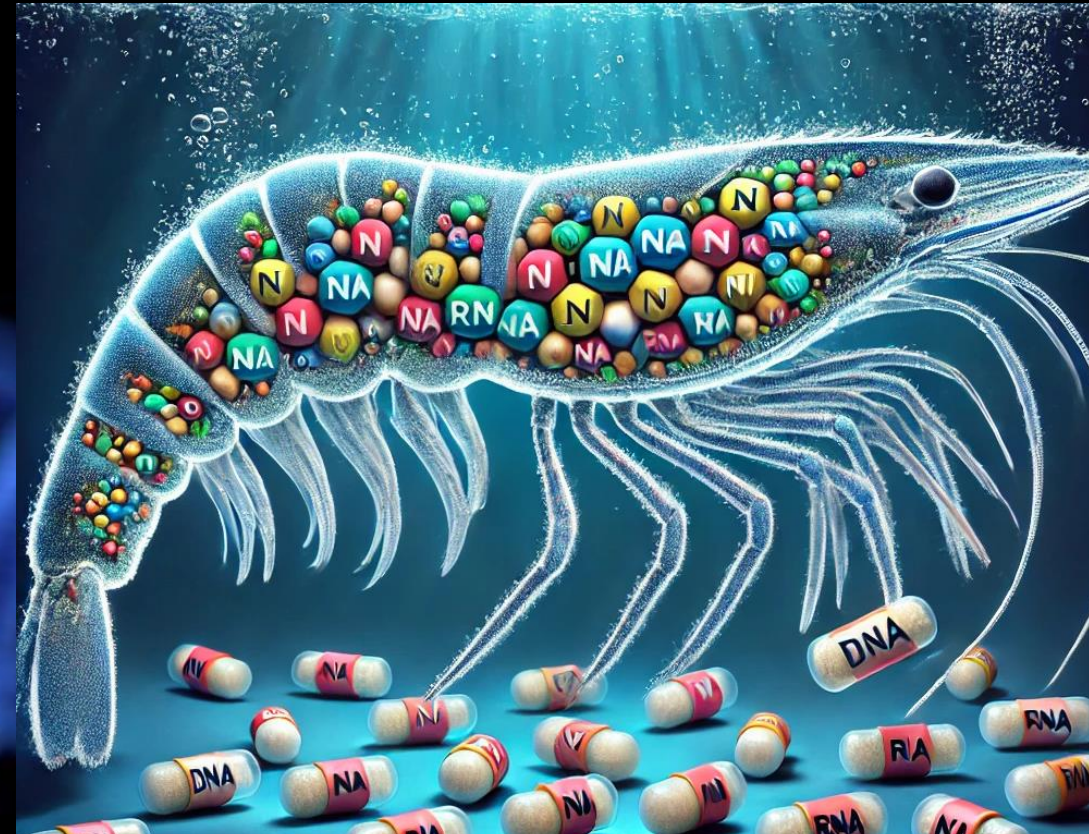
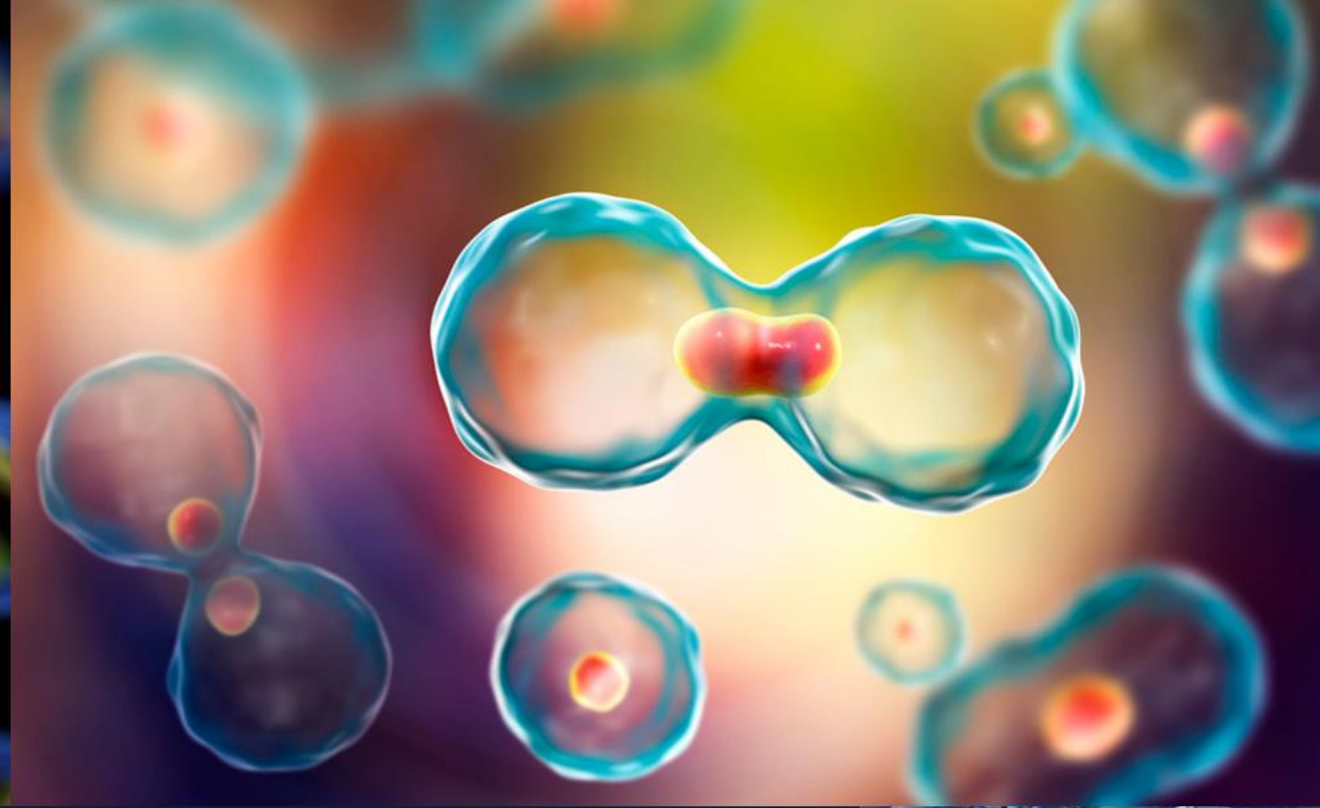


SUSTAINABLE FEEDS for Sustainable Aqua Farming

Marcelo Borba





**Each New Cell Requires 1 Billion
Nucleotides to Duplicate**



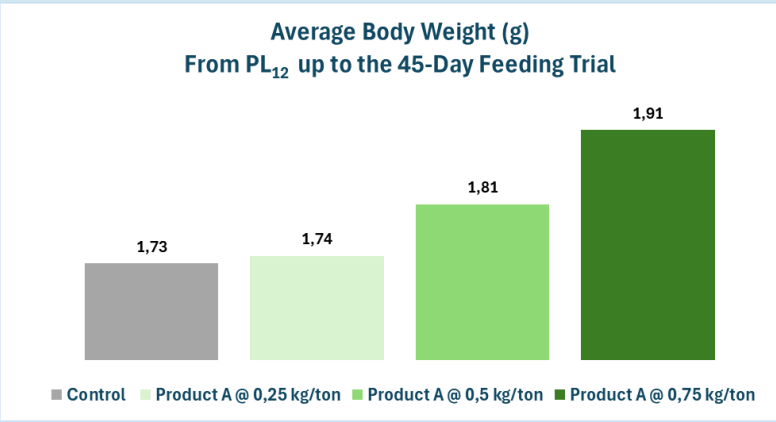
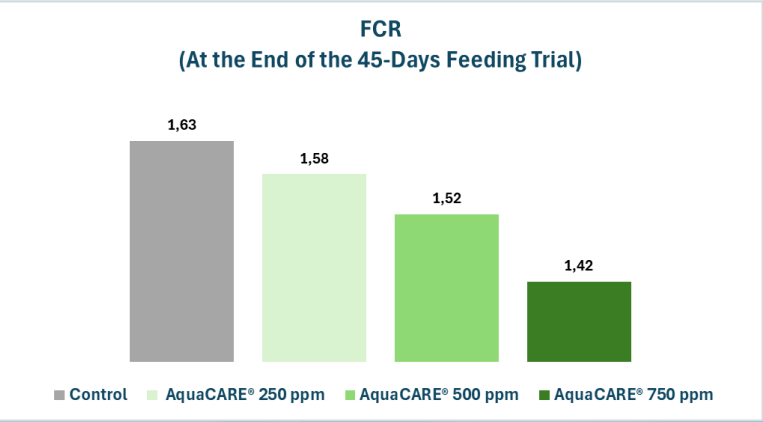
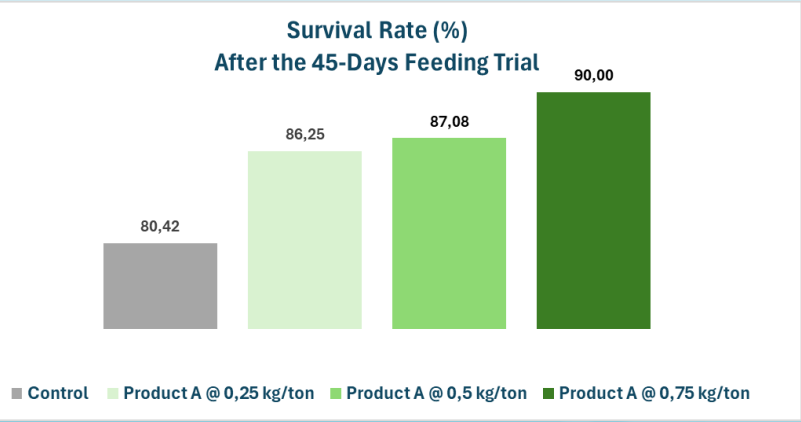
Effects of dietary yeast-derived nucleotide and RNA on growth performance, survival, immune responses, and resistance to *Vibrio parahaemolyticus* EMS/AHPND infection in Pacific white shrimp (*Litopenaeus vannamei*)


Scientific Trial 01: Kasetsart University, Thailand

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Zootechnical KPIs (PL₁₂ up to Young Juveniles) Before Bacterial Challenge






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Effects of dietary yeast-derived nucleotide and RNA on growth performance, survival, immune responses, and resistance to *Vibrio parahaemolyticus* infection in Pacific white shrimp (*Litopenaeus vannamei*)

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Nucleotide
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ABSTRACT

Nucleotides (NT) and RNA from yeast extracts are gaining interest as high-value feed additives. The present study intended to evaluate the influence of yeast-derived NT and RNA on the growth performance, survival, immune responses, and resistance to *Vibrio parahaemolyticus* infection in Pacific white shrimp. In Experiment 1, postlarvae were distributed into 7 groups, corresponding to 7 experimental diets: control, NT 0.25, NT 0.50, NT 0.75, RNA 0.25, RNA 0.50, and RNA 0.75 g/kg feed. They were fed the experimental diets for 45 days. Then, their body weights, survival rates, immune parameters, and *Vibrio* spp. counts in the hepatopancreas and intestines were determined. In Experiment 2, the shrimp from Experiment 1 were challenged by immersion with *V. parahaemolyticus* at 10⁶ CFU/mL. Each group was fed the same diet for another 10 days to assess the disease resistance performance. The results revealed that the shrimp body weights of all groups were similar, suggesting that neither NT nor RNA exerts the growth-promoting effect. However, the average survival rates of the NT and RNA groups were in the range of 80–93 %, significantly higher than that of the control (83 %). These increased survivals were in line with the reduction in the hepatopancreatic and intestinal *Vibrio* spp. counts and the elevated immune parameters in the NT and RNA-fed shrimp. At day 10 after the bacterial challenge, the highest survival rates were observed in the RNA 0.50 and 0.75 g/kg feed groups (91 % and 92 %, respectively), followed by the RNA 0.25 (79 %) and NT 0.75 g/kg feed (68 %), and significantly higher than the positive control (24 %). In short, both yeast-derived NT and RNA, especially the RNA at the dose of 0.50–0.75 g/kg feed groups, showed promising health benefits effects in the Pacific white shrimp, notably the improved immune function and disease resistance.

1. Introduction

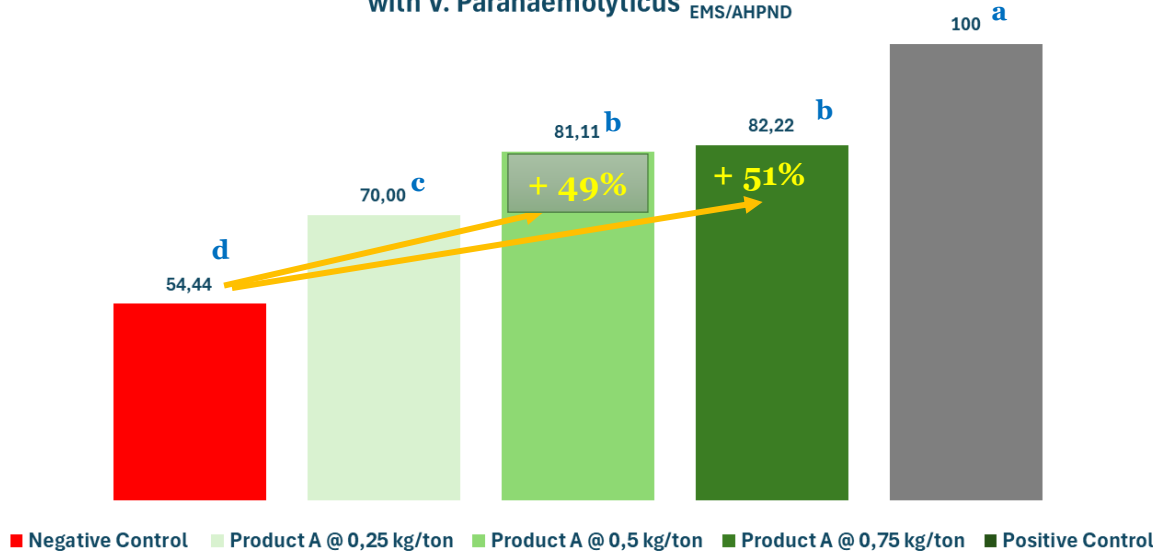
Nucleotides (NT) are basic units of nucleic acids (RNA and DNA) and play a key role in several biochemical processes. NT consists of nitrogenous bases (i.e., purines and pyrimidines), pentose sugar (i.e., ribose and deoxyribose), and phosphate groups. However, NT are traditionally not considered to be an essential nutrient because they can be produced endogenously under normal conditions via a salvage pathway in which the NT is synthesized from the nitrogenous bases and nucleosides formed during RNA and DNA degradation and *de novo* synthesis from amino acids and other molecules (Carver and Walker, 1995; Grønhaug and Westvold, 2001; Li and Gurtin, 2006). However, the production of NT may be insufficient to fulfill its demand in certain conditions such as infection, stress, or during rapid growth (Hess and Greenberg, 2012; Hossain et al., 2020). Thus, supplementation of exogenous NT can potentially be useful in these circumstances. The beneficial effects of dietary NT on human health regarding gastrointestinal growth and development, hepatic function, and immune system are documented (Carver and Walker, 1995; Hess and Greenberg, 2012). In addition, the advantages of using dietary NT in fish and shrimp

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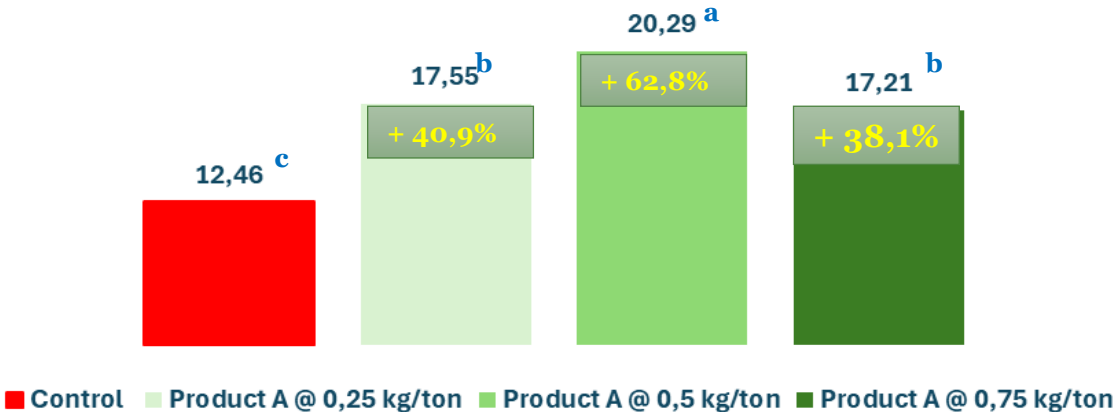
[https://doi.org/10.1016/j.aqurep.2022.101352](#)
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2222-5334/© 2022 The Authors. Published by Elsevier B.V. This is an open access article under the CC BY-NC-ND license (<http://creativecommons.org/licenses/by-nc-nd/4.0/>).

Zootechnical KPI After Bacterial Challenge

Survival Rate After the Bacteria Challenge
with *V. Parahaemolyticus* EMS/AHPND



Weight Gain (%)
After the *Vibrio parahaemolyticus* EMS/AHPND Challenge



Overall Trials' Stats

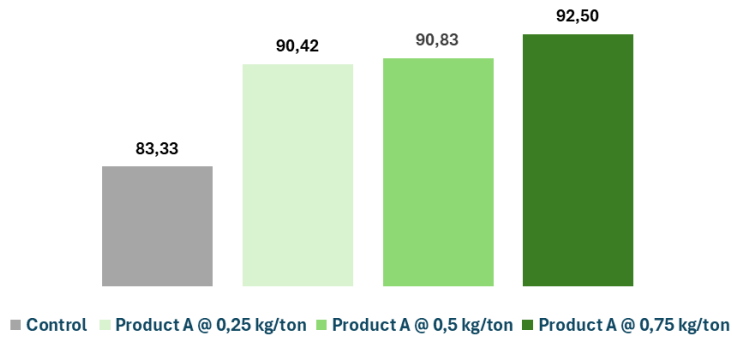
| Key Parameters | | Numerical and Statistical Differences Across Treatments | | | | | |
|----------------------|--|---|------------------------|-----------------------|------------------------|--|--|
| | | Control | AquaCARE (0,25 kg/ton) | AquaCARE (0,5 kg/ton) | AquaCARE (0,75 kg/ton) | | |
| Immunological | Total Hemocyte Count - THC (10 ⁶ cell/mL) | 1,95 ^c | 3,23 ^b | 3,94 ^a | 4,09 ^a | | |
| | Phagocytic Activity (%) | 51,67 ^c | 61,00 ^b | 64,67 ^a | 65,67 ^a | | |
| Gut Health | Total Vibrio spp. count Hepatopancreas (10 ³ CFU / g) | 9,60 ^c | 6,77 ^b | 3,67 ^a | 4,10 ^a | | |
| | Vibrio spp. count Intestines (10 ² CFU / g) | 10,27 ^c | 3,53 ^b | 1,47 ^a | 1,70 ^a | | |
| Anti-Oxidant Enzymes | Superoxide Dismutase - SOD (% inhibition) | 33,33 ^c | 40,28 ^b | 42,71 ^a | 44,79 ^a | | |
| | Phenoloxidase - PO (units/min/mg protein) | 220,32 ^c | 243,41 ^b | 255,36 ^a | 256,06 ^a | | |
| Zootechnical | Weight Gain (%) after Bacterial Challenge | 12,26 ^c | 17,55 ^b | 20,29 ^a | 17,21 ^b | | |
| | Vibrio parahaemolyticus (AHPND) | | | | | | |
| | Survival after Bacterial Challenge Vibrio parahaemolyticus (AHPND) | 54,44 ^c | 70,00 ^b | 81,11 ^a | 82,22 ^a | | |

Scientific Trial 02: Kasetsart University, Thailand (2025):

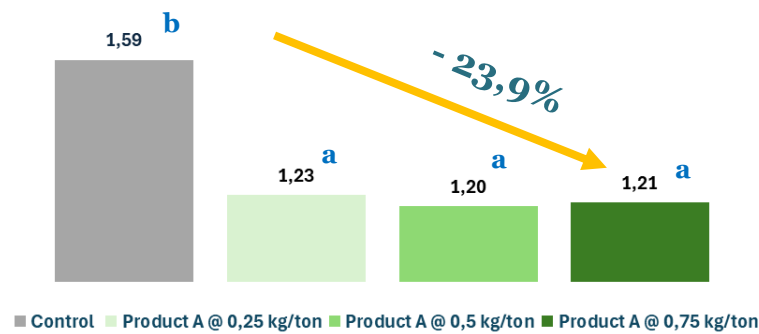
Investigating the Impact of Yeast-Derived Nucleic Acids and Nucleotides

Supplementation on Growth Performance, Condition of Hepatopancreas, Immunity, the Total Number of *Vibrio* spp. in the Gut, Stress Test with Low Oxygen, and Challenged Test with *Vibrio parahaemolyticus* AHPND in Pacific White Shrimp (*Litopenaeus vannamei*)

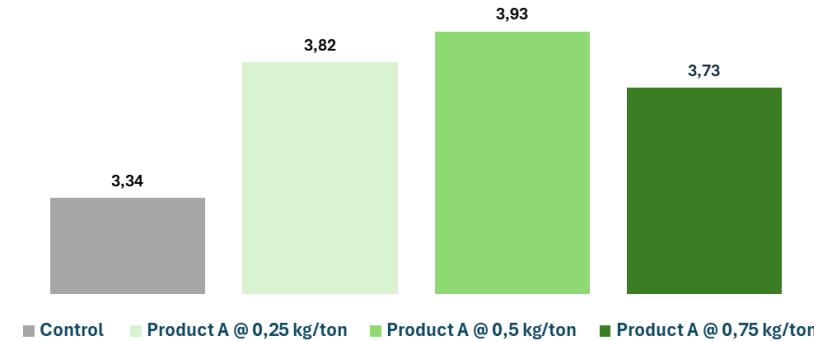
Survival Rate (%)
After the 45-Days Feeding Trial



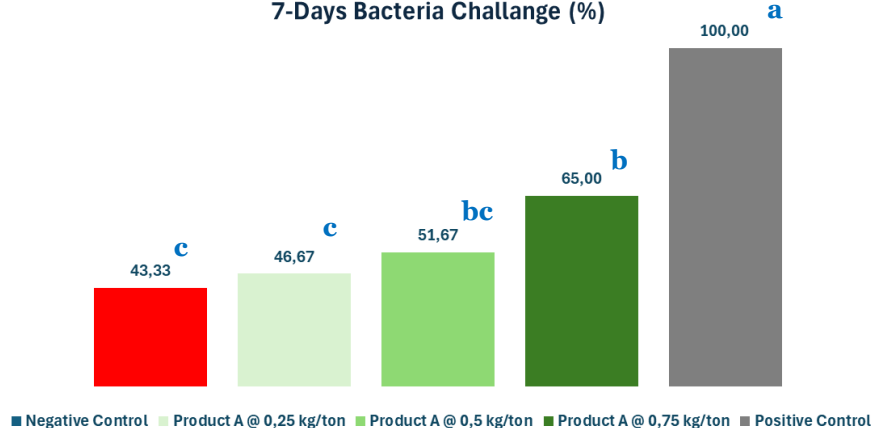
FCR
(At the End of the 45-Days Feeding Trial)



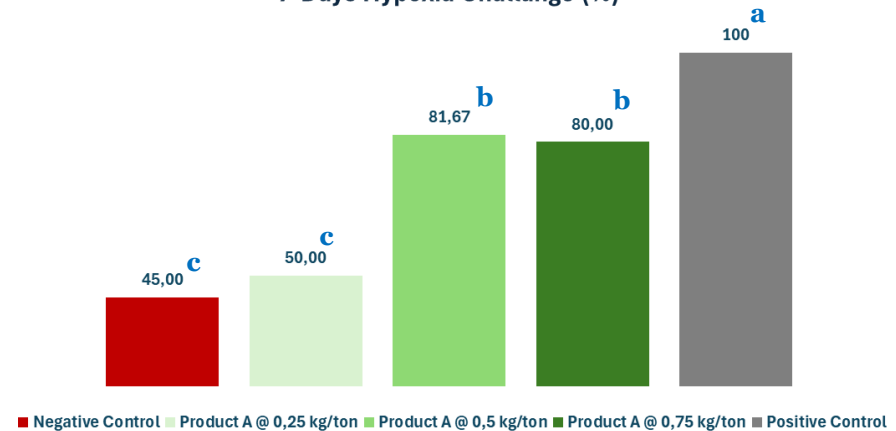
Shrimp Average Final Body Weight (g/shrimp)
(By the End of the 45 Days Feeding Trial)



Average Survival Rate After
7-Days Bacteria Challenge (%)



Average Survival Rate After
7-Days Hypoxia Challenge (%)



Zootechnical Performance of *Penaeus vannamei* Post-Larvae (PL₁₀ – PL₁₆) during the Acclimation Phase to Oligohaline Water in Intensive Nurseries under Different Application Doses of Nucleotides and Nucleic Acids (RNA).

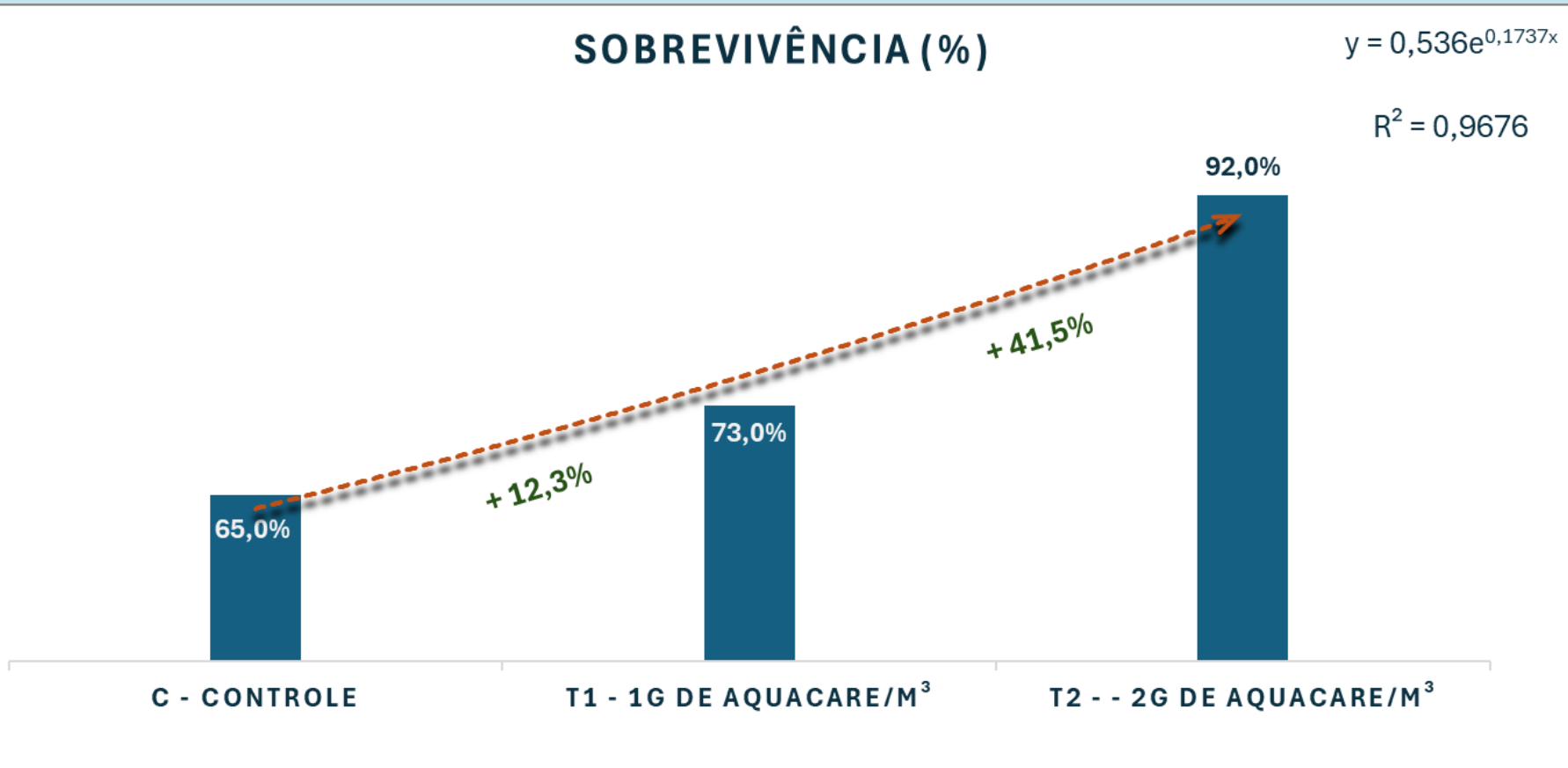


Survival Rate of *Penaeus vannamei* Post-Larvae _(10 a 16) of *durig the aclimattion Phase*

Water Salinity upon Arrival: 13 ppt

Farms' Underground Water Source: 0,6 ppt

Acclimatation Time in the Nursery Tanks: 6 Days



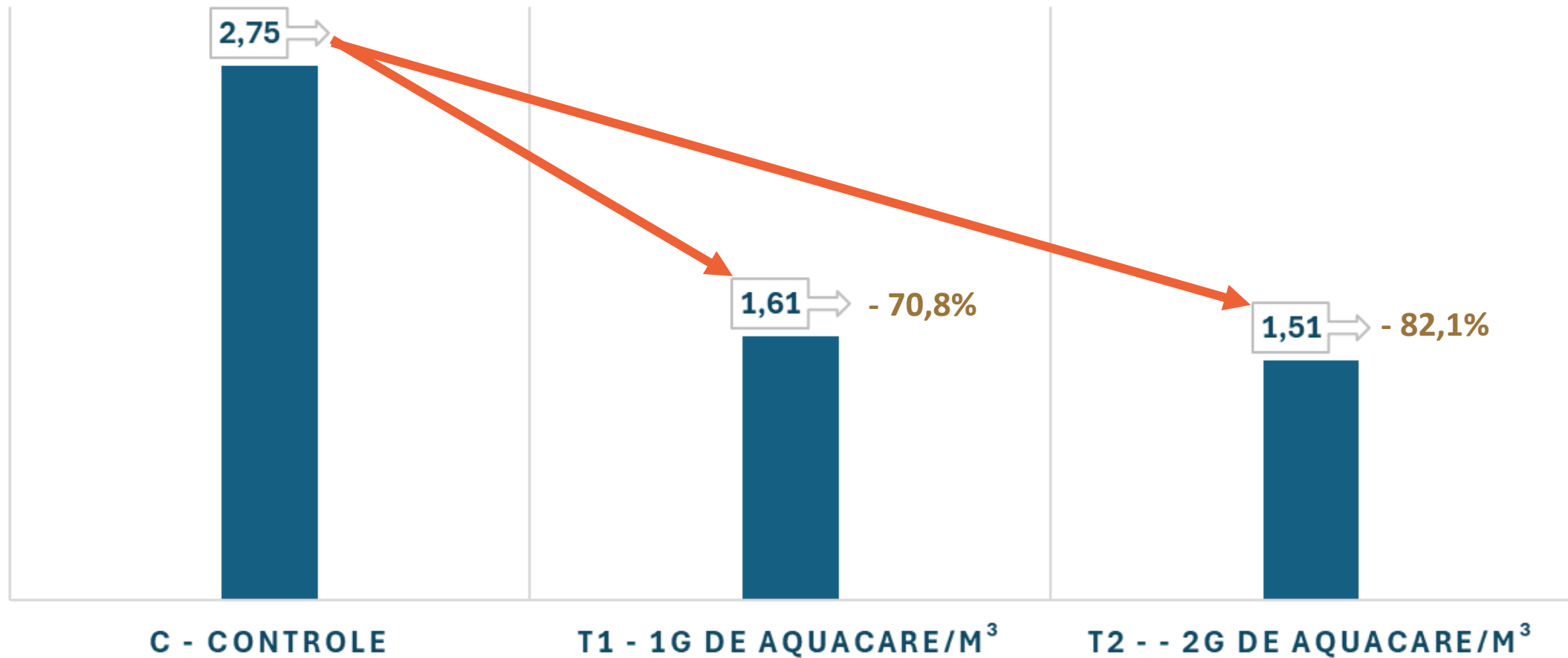
Tanks of 60 m³ Treatments

C - Control

T1 - Product A @ 1 g/m³/day

T2 - Product A @ 2 g/m³/day

FCR



Final Biomass After the Acclimation Phase

