

Assessing the Influence of Climate Change and Its Relationship on Mass Culture of Live Feed (Microalgae) in Outdoor System Conditions

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ABSTRACT

Climate change poses significant challenges to the cultivation of live feeds, crucial for the aquaculture industry. This study aims to explore the impact of climate change on the growth performance of microalgae used as live feed for marine aquatic animals, focusing on key challenges and potential solutions. Data from cultivating three microalgae species in a mass culture system under outdoor conditions in the Klongwan Fisheries Research Station (KFRS), Prachuap Khiri Khan province, Thailand, from January 2022 to December 2023, along with Geographic Information System (GIS) data on air temperature changes in the province during the same period, were analyzed. The assessment revealed that temperature fluctuation (TF) directly affected the specific growth rate (μ) of each live feed. Higher TF leading to a decreased in μ for two cultured live feeds, that is *Chlorella* and *Isochrysis*, with a negative temperature coefficient (Q10) of 0.04 and 0.00, respectively, for every 0.5°C increase in TF ($r^2 = 0.94$ and 0.38, respectively) from 31°C. In contrast, *Chaetoceros* showed a positive correlation between TF and μ , with a Q10 of 0.07, for every 0.5°C increase in TF ($r^2 = 0.97$) from 31°C. To mitigate these challenges, strategies such as using temperature-control measures (e.g., shading or insulation) and selecting temperature-tolerant strains of live feed can enhance the resilience of outdoor systems to temperature change, thereby maintaining productivity in fluctuating temperature conditions.

Keyword: Climate change/ Live feed culture/ Temperature fluctuations

1. INTRODUCTION

Climate change is a global phenomenon that poses significant challenges to various sectors, including agriculture and aquaculture [1]. In the aquaculture industry, the cultivation of live feeds, such as microalgae, is crucial for supporting the growth and development of marine aquatic animals. Microalgae were rich in essential nutrients and serve as an important source of food for many aquaculture species, including larval stages of fish and shrimp [2, 3].

The mass cultivation of microalgae in outdoor systems is preferred for its cost-effectiveness, scalability, and ability to produce large biomass. However, success in these systems heavily relies on environmental conditions such as temperature, light, and nutrients [4]. Climate change, with rising temperatures and more frequent extreme weather, poses significant challenges, particularly in outdoor settings where conditions are less controllable. Higher temperatures may boost growth rates but also increase vulnerability to thermal stress and shifts in species composition. Unpredictable weather can further destabilize optimal growth conditions [1, 5].

Research indicates that climate change affects microalgae productivity, composition, and diversity, but most studies are lab-based, with little data on large-scale outdoor systems. Understanding how rapidly changing climates, especially in regions like Southeast Asia, impact these systems is crucial for developing strategies to sustain and improve microalgae cultivation under changing conditions.

Thailand, with its robust aquaculture industry, presents an ideal setting to study the impacts of climate change on outdoor microalgae cultivation. The country has witnessed notable climatic changes in recent years, including rising temperatures and increasingly erratic weather patterns, making it a valuable case study for this research. Understanding these dynamics in relation to microalgae cultivation could offer important insights for other regions facing similar challenges. This study aims to evaluate the impact of climate change on the mass culture of live feed microalgae under outdoor conditions in Thailand. By assessing the growth performance of three microalgae species (*Chlorella* spp., *Isochrysis galbana*, and *Chaetoceros calcitrans*) and correlating this with Geographic Information System (GIS)

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data on temperature fluctuations, the research seeks to identify key challenges and potential solutions for maintaining productivity amidst variable temperatures. The outcomes of this study are expected to inform the development of strategies to strengthen the resilience of microalgae cultivation systems against climate change. These strategies will be essential for promoting sustainable aquaculture practices and supporting the broader goal of food security in regions vulnerable to environmental shifts.

2. METHODOLOGY

2.1 Study site and data source

The study was conducted at the Klongwan Fisheries Research Station (KFRS) in Prachuap Khiri Khan Province, Thailand. Prachuap Khiri Khan Province is located in the upper southern region of Thailand, covers an area of 6,367.62 square kilometers. It is positioned between latitude 10.9° - 12.6° N and longitudes 99.2° - 100.0° E. Three species of microalgae used as live feeds, including *Chlorella*, *Isochrysis* and *Chaetoceros* were cultivated in a mass culture system under outdoor conditions in KFRS area, from 2022 to 2023 (Figure 1).



Figure 1. A map showing the location of the live feed cultivation site (11°45'19"N, 99°47'35"E) at the Klongwan Fisheries Research Station (KFRS), located in Prachuap Khiri Khan province, Thailand, and the cultivation tanks for mass culturing of live feeds under outdoor conditions.

The original inoculum samples of the three species of marine microalgae (the diatom strains: *Chaetoceros calcitrans*; the green microalgal strains: *Chlorella* spp.; and the flagellate strains: *Isochrysis galbana*) were obtained from the Prachuap Khiri Khan Coastal Fisheries Research and Development Center, Department of Fisheries, Thailand.

Microalgal cultures were raised in 250 mL Erlenmeyer flasks containing sterilized sea water adjusted to a salinity of 28 ppt and enriched with liquid medium, Conway medium [6], with silicate added only for the diatoms and inoculated at 10% (v/v). The temperature-controlled room was maintained at 25±1°C under a 12 h light-to-12 h darkness photoperiod (12L:12D) using cool-white fluorescent lamps at a light intensity of about 1,000 Lux. These microalgae were cultured in the Erlenmeyer flasks until the cell density reached approximately 10⁶ cells/mL. Each culture was finally scaled up into 1 L glass bottles and then used for mass culture [7].

For microalgal cultivation in mass culture system, the information utilized in this assessment will be derived from KFRS's outdoor mass cultivation of each microalgae type, spanning from January 2022 to December 2023. The cultivation process began with the inoculation of microalgae obtained from laboratory cultivation into a 200 L culture tank, with an inoculum to culture water ratio of 1:25 and a water salinity of 26±1 ppt. Nutrients were added based on the specific requirements of each microalgae type [6]. The microalgae were initially cultured in a 200 L tank for approximately 3 days before being transferred to a 2,000 L fiber tank (Figure 1), where they were cultivated for an extended period under an average water salinity of 26±1 ppt. The cultivation in this tank continues for around 5-7 days, or

until the microalgae reach the end of their life cycle (death phase), repeated this cultivation method for each microalgae type on a monthly basis.

During the cultivation, algal cell samples were collected daily for estimation of cell density (in cells/mL). Cells were fixed with 5% formalin and then counted using a hemacytometer under a compound microscope at 40X magnification [7]. Subsequently, the data was averaged and the monthly growth was summarized for further analysis and synthesis.

2.2 Data collection

The specific growth rate (μ) of each live feed was measured every month of the mass cultivation under outdoor condition, with three replicate per month during of the culture period in January 2022 to December 2023, and the temperature fluctuation (TF) was recorded from Geographic Information System (GIS) data of ©WeatherSpark.com on air temperature changes in Prachuap Khiri Khan province during the same period and analyzed. In addition, the consistent rate of change in growth with temperature or temperature coefficient (Q10) of each live feed was analyzed. The μ and Q10 were calculated using equations 1 and 2, respectively:

$$\mu = \ln N_2 - \ln N_1 / t_2 - t_1 \quad (1)$$

$$Q_{10} = (\mu_2/\mu_1)^{10/T_2-T_1} \quad (2)$$

Where; μ = the specific growth rate (day^{-1}), N_1 = the cell count at time t_1 (cells/mL), N_2 = the cell count at time t_2 (cells/mL), t_1 = the first sampling time (day), t_2 = the second sampling time (day), Q_{10} = the temperature coefficient, μ_1 = the specific growth rate at temperature T_1 (day^{-1}), μ_2 = the specific growth rate at temperature T_2 (day^{-1}), T_1 = lowest value of the highest temperature in the year ($^{\circ}\text{C}$), T_2 = maximum temperature of the year ($^{\circ}\text{C}$), and \ln indicates the Napierian logarithm.

2.3 Data analysis

The collected data were consolidated and tallied in a spreadsheet and consequently analyzed using a statistical software package. The simple mean and percentage were the main descriptive statistics used to estimate the qualitative data. In addition, the relationship between TF and μ of each species of microalgae were examined using a simple linear correlation analysis and Pearson's correlation coefficient. A probability plot was used to test for normality before performing ANOVA. All analyses were performed using the IBM SPSS Statistics for Windows software (version 21.0; IBM Corp., Armonk, NY, USA).

3. RESULTS AND DISCUSSION

Figure 2 illustrates the geographic information depicting changes in air temperature in Prachuap Khiri Khan province from January 2022 to December 2023. The synthesis of this data reveals that the average lowest temperature of the highest temperature (l -T) and maximum temperature (m -T) in 2022 were 31.0°C and 36.0°C , respectively, and in 2023, were 31.0°C and 39.0°C , respectively. The total average l -T and m -T for the two years was 31.1°C and 37.0°C , respectively.

From average TF of the year, the assessment revealed that TF directly affected the μ of each live feed. For *Chlorella* and *Isochrysis*, higher TF led to a decrease in μ , with a negative Q10 of 0.04 and 0.00, respectively, for every 0.5°C increase in TF ($r^2 = 0.94$ and 0.38 , respectively) from 31°C . In contrast, *Chaetoceros* showed a positive correlation between TF and μ , with a Q10 of 0.07, for every 0.5°C increase in TF ($r^2 = 0.97$) from 31°C . An r-square (r^2) value closer to 1 indicates a stronger tendency of the growth rate (μ) to be influenced by temperature fluctuation (TF). For example, in the study, *Chlorella* and *Chaetoceros* showed r^2 values of 0.94 and 0.97, respectively, which represent strong correlations. In contrast, *Isochrysis* had an r^2 of 0.38, indicating a weaker correlation. Therefore, the tendency is best represented when r^2 is close to 1 (Figures 3 and 4).

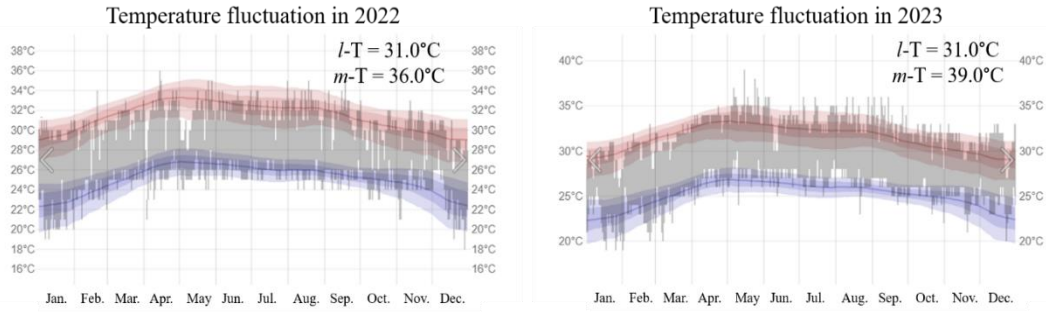


Figure 2. Annual temperature in Prachuap Khiri Khan province, Thailand (in 2022-2023); the graph illustrates the daily range of reported temperatures (gray bars), alongside the 24 h high (red bars) and low (blue bars) temperatures overlaid on the average daily high temperature values (faded red line) and daily average high and low temperature values (faded blue line), and showed average lowest temperature of the highest temperature ($l-T$) and maximum temperature ($m-T$) of the year (Data source: Adapted from geodatabase of ©WeatherSpark.com).

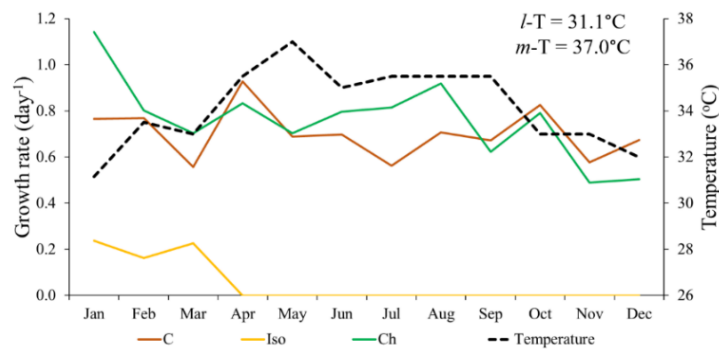


Figure 3. Average maximum temperatures fluctuation (TF) and specific growth rate (μ) of cultured three live feeds (C = *Chaetoceros calcitrans*, Iso = *Isochrysis galbana* and Ch = *Chlorella* spp.) in a mass culture system under outdoor conditions; $l-T$ is lowest temperature of the highest temperature and $m-T$ is maximum temperature of the year.

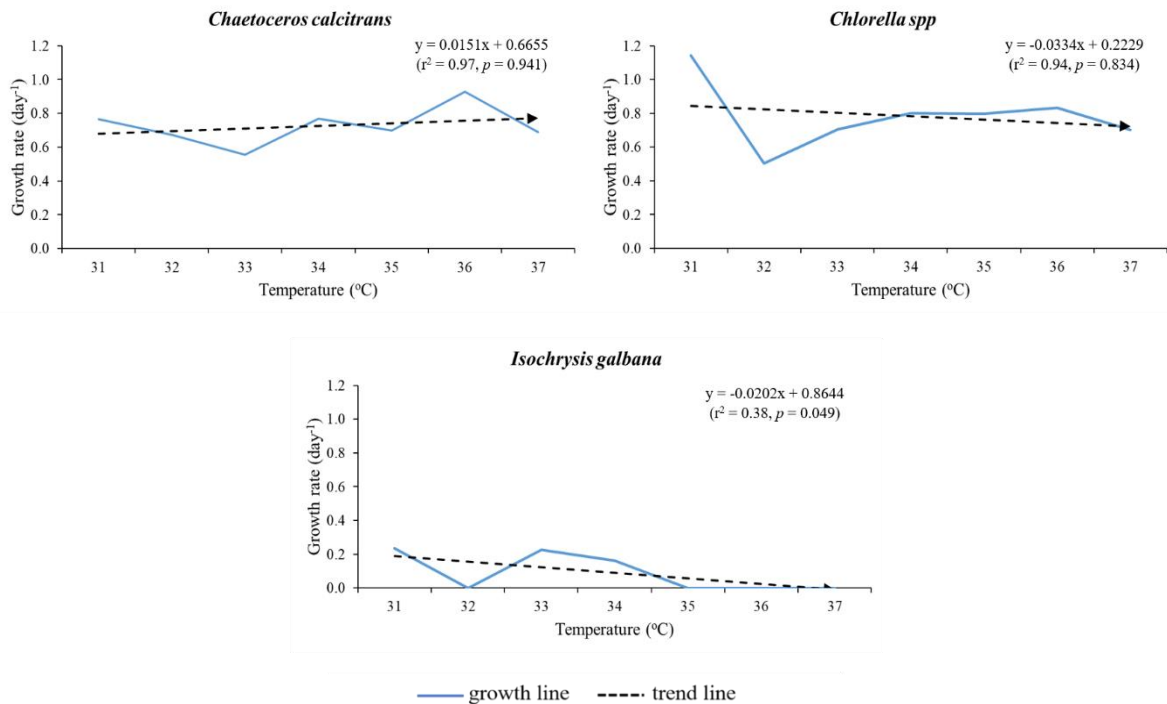


Figure 4. Correlation between maximum temperatures fluctuation (TF) and specific growth rate (μ) of each live feeds cultured in a mass culture system under outdoor conditions.

In current study indicates that higher TF leads to decreased growth rates of *Chlorella* and *Isochrysis* used as live feed. This finding is consistent with previous studies that had shown the negative effects of temperature fluctuations on the growth and productivity of some microalgae, for example, *Chlorella vulgaris* [5, 8]. In contrast, *Chaetoceros* exhibited a positive correlation between TF and μ , suggesting that these species may be more resilient to temperature fluctuations. The ability of these species to thrive in fluctuating temperature conditions is an important factor to consider when selecting microalgae species for aquaculture systems exposed to variable environmental conditions. To mitigate the challenges posed by climate change, aquaculture practitioners can employ several strategies. Temperature-control measures, such as shading or insulation, can help stabilize water temperatures in outdoor culture systems, reducing the impact of temperature fluctuations on microalgae growth [9-11]. Selecting temperature-tolerant strains of microalgae, can also enhance the resilience of culture systems to temperature changes [12, 13], such as from this study, *Chaetoceros calcitrans* had the resilience of mass culture system under outdoor conditions to temperature changes than *Chlorella* spp. and *Isochrysis galbana*.

Additionally, the use of GIS data to analyze air temperature changes in the cultivation area provides valuable insights into the local climate dynamics and its effects on microalgae cultivation. By incorporating GIS data into culture management practices, practitioners can make informed decisions about site selection and microalgae species selection, ensuring the sustainability of live feeds cultivation operation in the face of climate change. Previous studies had demonstrated the extensive application of GIS in aquaculture, highlighting their ability to enhance efficiency in farming and production [14]. GIS can also play a crucial role in identifying strategies to mitigate or resolve issues stemming from climate change, which were anticipated to impact aquatic animals significantly [15].

Overall, this study demonstrates the importance of understanding the impact of climate change on microalgae cultivation and developing strategies to mitigate its negative effects. By implementing temperature-control measures and selecting temperature-tolerant strains of microalgae, aquaculture practitioners can enhance the resilience of their operations to climate change, ensuring a stable supply of live feed for marine aquatic animals.

4. CONCLUSIONS

This study emphasizes the significant impact of climate change, especially temperature fluctuations, on the mass cultivation of live feed microalgae in outdoor systems. The inverse relationship between temperature fluctuations and the specific growth rate of *Chlorella* spp. and *Isochrysis galbana* highlights the susceptibility of these species to temperature changes. Conversely, *Chaetoceros calcitrans* demonstrated a positive relationship, suggesting their potential resilience to temperature fluctuations. Understanding and addressing the effects of climate change on live feed cultivation are vital for aquaculture practitioners, as this knowledge can greatly contribute to the aquaculture industry's sustainability in the face of a climate change.

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