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Review

Decoding the Cellular Green-to-Red Gold Transition: Harnessing AI-Integrated IoT-ML Framework for Boosting *Haematococcus pluvialis* Cultivation and Astaxanthin Bio-Production

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Abstract: *Haematococcus pluvialis* is a unicellular green microalga renowned as the natural source of astaxanthin with biotechnological applications. Its commercial value is closely linked to its distinctive two-stage life cycle. Under favourable conditions, *H. pluvialis* exists in a motile green vegetative stage, where cells actively grow and accumulate biomass. When exposed to environmental stressors such as high light intensity, nutrient limitation, salinity, or elevated temperature, the cells enter non-motile red cyst stage, characterised by the accumulation of astaxanthin as a protective response to oxidative stress. Despite the growing demand for microalgal products, cultivating *H. pluvialis* remains challenging due to the need for precise control of cultivation parameters, including biomass concentration, pH, light intensity and temperature. While the agriculture and aquaculture sectors have increasingly adopted Internet of Things (IoT) and artificial intelligence (AI) technologies, their application in microalgae farming is still at an early infant stage. This review provides an overview of *H. pluvialis* cultivation, emphasizing its two-stage growth strategy for astaxanthin production, and evaluates the potential of IoT-enabled smart farming systems. Monitoring this transformation is important for identifying the optimal harvesting stage to maximize astaxanthin production and biomass quality. The integration of sensors, automation, and machine learning (ML) for monitoring and predicting biomass growth is proposed to enhance efficiency and reduce reliance on manual analysis. However, challenges such as limited datasets and difficulties in accurate modelling persist, underscoring the need to further develop intelligent microalgae cultivation systems.

Keywords: smart farming; machine learning; Internet of Things; astaxanthin; *Haematococcus pluvialis*; artificial intelligence

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1. Introduction

Haematococcus pluvialis, a unicellular freshwater microalga, is widely regarded as one of the most promising natural sources of astaxanthin due to its superior antioxidant capacity and safety for human consumption in comparison with petrochemically synthesised astaxanthin [1]. Astaxanthin is a bright red secondary carotenoid belonging to the same family as lycopene, lutein, and β-carotene. Astaxanthin is biosynthesised by microalgae, plants, yeast, and certain bacteria. It is also commonly found in aquatic organisms, such as salmon, trout, red sea bream, shrimp, lobster, and fish eggs. Owing to its strong free-radical scavenging ability, astaxanthin has attracted significant interest across various industries, including food, feed, cosmetics, aquaculture, nutraceuticals, and pharmaceuticals [2]. Notably, its antioxidant activity is substantially higher than that of other antioxidants, being approximately 65 times more potent than vitamin C, 54 times stronger than β-carotene, 10 times stronger than carotenoids (β-carotene, canthaxanthin, zeaxanthin, and lutein), and up to 100 times more effective than α-tocopherol. Despite its advantages, approximately 95% of the astaxanthin currently available on the market is synthetically produced, while natural astaxanthin derived from *H. pluvialis* accounts for only 1% of the global production. Synthetic astaxanthin exhibits lower antioxidant activity and has limited approval for human consumption, as it is produced via chemical synthesis involving C10-dialdehyde and asta-C15-triarylphosphonium salt [3]. Consequently, the growing demand for natural astaxanthin, coupled with its higher market value, has driven increased interest in developing efficient, sustainable production strategies using *H. pluvialis*.

Several strategies have been developed for astaxanthin production from *H. pluvialis*, including photoautotrophic, heterotrophic, and mixotrophic cultivation in systems such as open raceway ponds and closed photobioreactors, operated in batch or fed-batch modes [3]. Recent advancements include two-stage mixotrophic cultivation systems and attached cultivation techniques that employ immobilised biofilm. Astaxanthin accumulation in *H. pluvialis* is closely associated with cellular carotenoid content and overall productivity, both of which are strongly influenced by cultivation conditions. Under optimal stress, carotenoid content in dried biomass can reach up to 5%, with astaxanthin comprising approximately 90% of total carotenoids. Astaxanthin biosynthesis is induced by environmental stressors, such as high light intensity, nutrient limitation, salinity, and temperature fluctuations, which are often applied in combination to enhance production. This response is linked to the disruption of cellular homeostasis and serves as a protective mechanism against oxidative stress and reduced photosynthetic efficiency [4]. The cultivation process typically involves two distinct stages: a green stage, where cells proliferate under favourable conditions to maximise biomass, followed by a red stage, in which stress conditions are applied to induce astaxanthin accumulation in non-motile cyst cells. Optimising the transition and balance between these two stages is critical for maximising astaxanthin yield.

The world is rapidly advancing towards the fourth industrial revolution, driven by advances in IoT and ML technologies. The integration of these tools into microalgae production systems presents a promising opportunity to overcome existing challenges by reducing operational costs and enabling real-time monitoring of growth and productivity. These technologies have shown increasing potential in detecting growth phases, predicting biomass productivity, and enabling real-time process control through data-driven decision-making. Studies focusing on *H. pluvialis* have explored image-based analysis, sensor integration, and predictive modelling to identify growth stages and monitor astaxanthin accumulation. However, despite these advancements, several challenges remain. Biological variability in microalgae cultures can introduce significant heterogeneity, affecting model robustness and generalisability. This challenge is further amplified in both monoculture and mixed-culture systems, where variations in cell morphology, species interactions, and pigment composition increase the complexity of accurately identifying growth stages and monitoring biomass dynamics. Addressing these limitations is essential to developing reliable, scalable AI-driven monitoring systems for microalgae cultivation.

This review provides a comprehensive overview of recent advances in astaxanthin production through the cultivation of *H. pluvialis*. The novelty of this review lies in integrating interdisciplinary approaches, bridging biological cultivation strategies with intelligent digital technologies. The significance of this review is to demonstrate how smart farming concepts can enhance productivity, efficiency, and scalability in microalgae systems. Furthermore, this review proposes an integrated IoT-ML framework for *H. pluvialis* cultivation, enabling automated monitoring, predictive analysis, and optimisation of key process parameters, thereby advancing the development of intelligent and sustainable microalgae production systems.

2. *Haematococcus pluvialis*

H. pluvialis is a well-known Chlorophyta microalgae capable of producing astaxanthin, a high-value carotenoid with various health properties, including antioxidant, anti-diabetic, anti-inflammatory, and eye-protective properties [5]. The accumulation of astaxanthin in *H. pluvialis* can reach approximately 3.8–5% of dry cell weight [6]. Astaxanthin was initially commercialised in the aquaculture sector as a feed additive to enhance the pigmentation of farmed salmonids, improving their characteristic orange-red colour [7]. Although astaxanthin can also be derived from other sources, such as yeast and marine by-products (e.g., shrimp and crayfish), *H. pluvialis* remains the most attractive source due to its superior antioxidant activity [8,9]. The commercial feasibility of large-scale cultivation of *H. pluvialis* has been demonstrated by companies such as Fuji Chemical Industry [8].

Despite these advances, several challenges persist, particularly high energy consumption and the complexity of cultivation strategies. The production process typically involves a two-stage cultivation system. The first stage, known as the green stage, focuses on biomass accumulation under favourable growth conditions, where cells are cultivated in nutrient-rich media to maximise growth rates. This stage is followed by the red stage, during which environmental stress is applied to induce astaxanthin accumulation. This stage is characterised by the transition of *H. pluvialis* cells from green to red under stress conditions. Exposure to environmental stressors such as high or low temperatures, nutrient deprivation, salinity, and intense light induces significant cellular and morphological changes, leading to the accumulation of astaxanthin [10]. During the green stage, the culture predominantly consists of macrozooids and palmella cells, whereas in the red stage, asexual aplanospore cells are formed and serve as the primary sites of astaxanthin accumulation. Initially, *H. pluvialis* appears as spherical, biflagellate green microalgal cells with a diameter of approximately 30 µm. As cultivation progresses to the intermediate stage, typically around two weeks after reaching maximum biomass, cells begin to undergo encystment, transitioning into greenish-orange forms. Under intense stress, flagella are lost, cell size increases, and thick-walled cysts form, accompanied by the accumulation of astaxanthin, leading to the characteristic red stage. This section reviews the fundamental mechanisms of astaxanthin production in *H. pluvialis* and recent technological developments, with a focus on cultivation strategies, bioprocess optimisation, and stress induction approaches to enhance astaxanthin yield.

2.1. Haematococcus pluvialis: Transition between Motile Green and Non-Motile Red Stages

At the initial stage, the cultivation of *H. pluvialis* (Figure 1) is comparable to that of most microalgal species, where growth occurs under favourable conditions to maximise biomass production. Depending on trophic conditions, *H. pluvialis* can adopt different metabolic modes. Under photoautotrophic cultivation, light serves as the primary energy source for photosynthesis, while CO_2, water, and essential nutrients (e.g., carbon, nitrogen, phosphorus, sulphur, potassium, and iron) support cell growth [11]. Following this growth phase, stress conditions are introduced to induce astaxanthin accumulation.

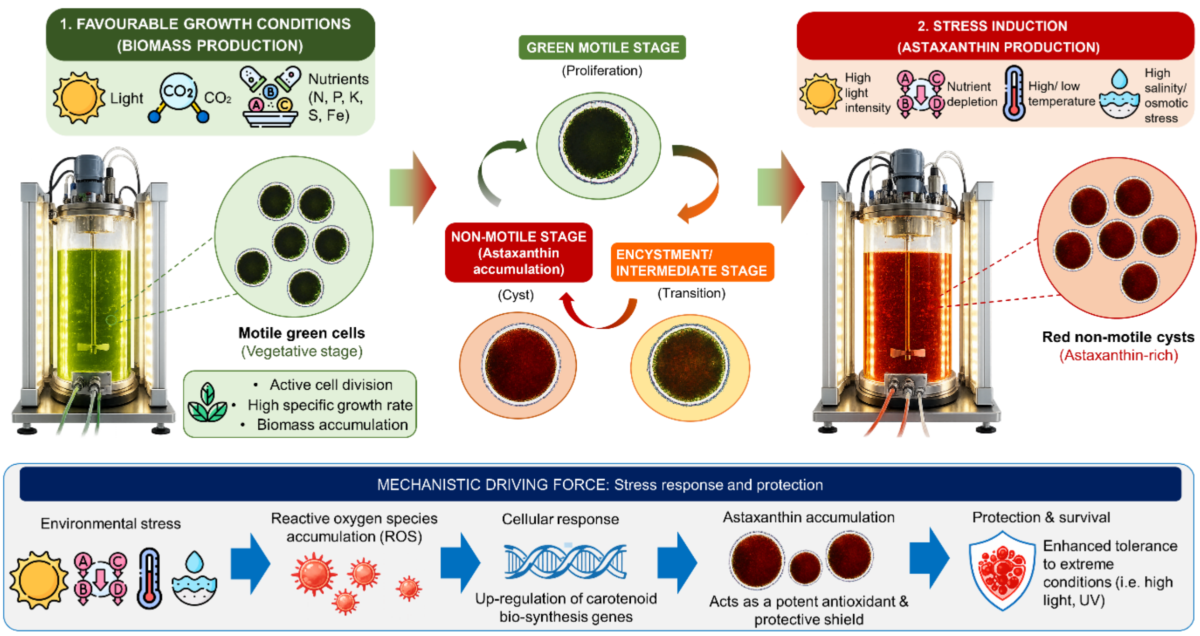

Figure 1. Schematic illustration of the two-stage cultivation strategy of *H. pluvialis* for biomass growth and astaxanthin production. Under favourable growth conditions, including sufficient light, CO_2, and nutrients, motile green vegetative cells proliferate rapidly during the green stage to maximise biomass accumulation. Upon exposure to environmental stressors such as high light intensity, nutrient deprivation, temperature stress, and salinity stress, the cells undergo encystment and transition into non-motile red cysts enriched with astaxanthin. The lower panel summarises the mechanistic pathway underlying stress-induced astaxanthin biosynthesis, in which reactive oxygen species (ROS) generated under stress trigger cellular defence responses and upregulate carotenoid biosynthesis, leading to astaxanthin accumulation that protects cells against oxidative and environmental damage.

Mixotrophic cultivation represents an alternative strategy, in which both organic and inorganic carbon sources are utilised. This approach enables continued growth after nutrient depletion while simultaneously promoting astaxanthin accumulation without the need for severe external stress application [12]. For instance, supplementation with acetate or acetic acid following nitrate depletion has been shown to enhance astaxanthin productivity by approximately 20% [13]. Nevertheless, despite its potential, mixotrophic cultivation remains less commercially developed than photoautotrophic systems due to challenges such as managing light stress and the risk of microbial contamination [12].

2.2. Impact of Cultivation Parameters on Astaxanthin Production

Astaxanthin production in *H. pluvialis* is generally achieved through the induction of environmental stress conditions that stimulate its accumulation. Key parameters, including salinity, nitrogen availability, light intensity, metal supplementation, and temperature, can be regulated to create a stress environment favourable for enhanced astaxanthin synthesis [14]. Table 1 summarises the key cultivation parameters that can be optimised to promote astaxanthin production, along with recent research trends associated with each factor.

Table 1. Overview of key parameters influencing astaxanthin production in *H. pluvialis*.

Parameter	Condition/Range	Effect on Astaxanthin Production	Reference
Chemical stress	Addition of salicylic acid (SA), 25–50 mg/mL	SA treatment upregulated astaxanthin biosynthesis genes. SA25 (25 mg/L) upregulated ipi-1, psy, pds, crtR-B, and lyc, while SA50 (50 mg/L) showed greater effects on ipi-1, ipi-2, pds, crtR-B, and lyc.	[15]
	Two-stage mixotrophic growth: BG-11 medium supplemented with nutrients, followed by second stage that induced astaxanthin production under varying light and metal-ion conditions.	Maximum biomass of 11.6 g/L. Green LED light (520 nm, 150 μmol photons m^{-2} s^{-1}) and sodium selenite (25 mg/L) produced the highest astaxanthin accumulation (1.85 mg/g). Copper (II) sulfate (5 μM) enhanced astaxanthin accumulation (1.7 mg/g).	[16]
	Arginine and calcium	Arginine supplementation increased astaxanthin and lipid accumulation by 25% and 24.5%, respectively, while co-supplementation with Ca^{2+} further enhanced productivity by 40.7% and 32.6%.	[17]
Salinity	NaCl gradient (5–12.5 g/L)	Moderate NaCl concentrations (5–10 g/L) enhanced astaxanthin content, with the highest level (7.51 mg/L) observed at 7.5 g/L. Excessive NaCl concentration negatively affected astaxanthin production.	[18]
Nitrogen sources	Carbon to nitrogen (C/N) ratio (fed-batch)	High C/N ratio enhanced acetyl-CoA production and carotenoid transport. Biomass reached 9.18 g/L with 100% immotile cyst cells and astaxanthin productivity of 15.45 mg/L/day.	[19]
	$NaNO_3$ concentration (0–0.13 g/L)	Nitrogen deficiency (0 g/L) enhanced astaxanthin accumulation, reaching a maximum of 5.60 mg/L. Astaxanthin accumulation accelerated after day 14 under nitrogen-limited conditions.	[18]
Illumination intensity	Monochromatic red light at 20 °C and red-blue mixed light at 30 °C	Red light favoured green cell growth, while mixed red-blue light enhanced astaxanthin production, reaching 25 mg/L/day.	[4]
	Photon flux density of 250 μmol_{hv} m^{-2} s^{-1} and mean rate of photon absorption of 7000 ± 100 μmol_{hv} kg_X^{-1} s^{-1}	Maximize astaxanthin productivity of 0.29 ± 0.05 g m^{-2} d^{-1}	[20]
Temperature	20–30 °C	Highest accumulation at 30 °C, reaching 50 mg/g with pigment productivity of 20–25 mg/L/day. Temperatures above 30 °C caused rapid cellular lysis and inhibited carotenogenesis.	[4]
Inoculum density and storage time	Different inoculum densities (2.5–10 g dry weight m^{-2}) and biomass storage durations at 4 °C (24–168 h)	Optimal biomass density (5–7.5 g m^{-2}), 24 h storage at 4 °C, and light intensity (400–600 μmol photons^{-2} s^{-1}) enhanced biomass (8.7 g m^{-2} d^{-1}) and astaxanthin productivity (170 m^{-2} d^{-1}), while longer storage time reduced performance.	[21]
Carbon dioxide	0–20% CO_2	Moderately high CO_2 (5%) enhanced growth and astaxanthin, whereas higher concentrations (10–20%) inhibit growth, photosynthesis, and carbon assimilation.	[22]
Extension of the green stage	Combined effects of four factors: temperature, sodium bicarbonate, nitrogen, and phosphorus.	Temperature was the most influential factor, with temperatures above 22 °C reducing growth. Optimal green-stage cultivation conditions (22 °C, 250 mg/L $NaHCO_3$, 150 mg/L nitrogen, and 40 mg/L phosphorus) produced 1.53 g/L biomass after 15 days. Inorganic carbon should be supplied carefully to maintain $NaHCO_3$ concentrations below 250 mg/L and avoid premature red-stage induction.	[23]

2.2.1. Salinity

Salinity is a key cultivation parameter influencing the growth and astaxanthin production of *H. pluvialis*. Elevated salinity levels will affect the oxygen-evolving complex (OEC) and the photosystem II (PSII) reaction centre, altering light absorption and photosynthetic efficiency [24]. While moderate salinity stress may enhance cellular responses and improve productivity, maintaining an optimal salinity range is essential to balance biomass growth and production efficiency [25]. Suboptimal salinity conditions can lead to osmotic and ionic imbalances, resulting in inhibited growth, reduced photosynthetic activity, and morphological changes in the cells. On the other hand, increasing salinity to the optimal range can promote lipid accumulation without significantly affecting fatty acid and protein composition. In addition, salinity regulation via salicylic acid addition has been shown to enhance astaxanthin production. Gao et al. (2012) demonstrated that supplementation with salicylic acid (25–50 mg/L) significantly influenced the transcription of key carotenogenic genes *(ipi-1, ipi-2, psy, pds, crtR-B, lyc, bkt,* and *crtO)* [15]. Notably, a concentration of 50 mg/L effectively upregulated these genes, thereby enhancing astaxanthin accumulation in *H. pluvialis*.

2.2.2. Nitrogen

Nitrogen is one of the main nutrients that supports microalgal growth; however, its limitation is widely used to induce astaxanthin accumulation in *H. pluvialis*. Nitrogen deprivation creates a stress condition that redirects cellular metabolism from biomass production towards carotenoid biosynthesis. Lu et al. (2019) demonstrated that maintaining a high carbon-to-nitrogen (C/N) ratio enhances acetate kinase activity, leading to a 5.76-fold increase during the early logarithmic phase. This promotes the accumulation of acetyl-CoA, which serves as a precursor for astaxanthin biosynthesis. Additionally, a high C/N ratio enhances carotenoid content within the cells, thereby increasing astaxanthin accumulation [19]. Nevertheless, complete nitrogen depletion may not be optimal for production. Imamoglu et al. (2009) reported that cultures grown in distilled water and nitrogen-free media accumulated comparable levels of astaxanthin (29.62 mg/g and 30.07 mg/g, respectively), indicating minimal difference in productivity [26]. These findings suggest that excessive nitrogen starvation may increase cell mortality, thereby limiting overall astaxanthin yield. Therefore, controlled nitrogen limitation, rather than complete depletion, is essential to balance cell viability and astaxanthin production.

2.2.3. Illumination Intensity

Microalgae are photosynthetic microorganisms that utilise light and carbon dioxide to generate energy. This process involves two main stages: the light-dependent reactions and the Calvin cycle. In the light-dependent stage, PSII and PSI absorb light energy, initiating electron transfer from PSII to PSI, which drives the reduction of NADPH and ferredoxin. These energy carriers are subsequently used in the Calvin cycle to fix CO_2 into organic compounds [27]. In *H. pluvialis*, exposure to high light intensity induces stress conditions that promote astaxanthin accumulation [28]. Light wavelength and intensity play a crucial role in regulating carotenogenic gene expression, thereby influencing astaxanthin biosynthesis.

Recent studies have demonstrated that both light wavelength and intensity significantly affect astaxanthin productivity [4]. Pereira and Otero (2020) reported that monochromatic red light at 20 °C supports optimal biomass productivity. A combination of 30 °C red and blue light maximises astaxanthin production, achieving rates of up to 25 $\mathrm{mg\,L^{-1}day^{-1}}$ [4]. Furthermore, the combined application of high light intensity and nitrogen starvation has been shown to produce the highest astaxanthin rate of 200 $\mathrm{\mu molm^{-2}s^{-1}}$ [29]. Overall, light-induced stress primarily enhances biomass and astaxanthin yield, whereas nutrient limitation increases the astaxanthin-to-biomass ratio, highlighting the importance of optimising both factors for efficient production.

2.2.4. Temperature

Temperature is a critical parameter influencing microalgal growth and astaxanthin production. The optimal temperature range for most microalgae is typically between 20 °C and 25 °C, within a broader range of 15 °C–30 °C, which supports photosynthesis and cell division [30]. Under suboptimal low-temperature conditions, increasing temperature enhances enzymatic activity in the Calvin cycle, thereby improving growth rates. Generally, for every 10 °C increase below the optimal range, the growth rate may increase by about 100%. However, temperatures exceeding the optimal range impose thermal stress, leading to enzyme denaturation, impaired photosynthesis, and reduced biomass productivity.

In *H. pluvialis*, similar trends are observed. The optimal growth temperature ranges from 17 °C to 23 °C [31], while elevated temperatures can act as stressors, inducing astaxanthin accumulation. Pereira and Otero (2020) reported that increasing the cultivation temperature from 20 °C to 30 °C increased pigment productivity by 100% [4], whereas further increases beyond 30 °C result in diminished productivity due to cellular lysis. Similarly, Giannelli et al. (2015) observed that raising the temperature from 20 °C to 30.5 °C reduces cell concentration by approximately 65%, indicating a trade-off between growth and stress-induced production [32]. On the other hand, astaxanthin production was measured under nitrogen starvation at both 20 °C and 27 °C. Under nitrogen starvation conditions, astaxanthin accumulation was higher at 27 °C than at 20 °C, indicating that moderately elevated temperatures from 27 °C to 30 °C are favourable during the red stage for enhanced astaxanthin production.

2.2.5. Inoculum Size

The interaction between inoculum density and light intensity plays a critical role in determining biomass yield in *H. pluvialis* cultivation. The effect of inoculum size depends strongly on the level of light exposure. At low inoculum densities, exposure to high light intensity can lead to photoinhibition or cell bleaching, adversely affecting growth. Pham et al. (2018) investigated the influence of inoculum size in 200 mL cultures with initial cell densities ranging from 0.5×10^4 to 4×10^4 cells mL^{-1} and reported a maximum cell density of approximately 2.87×10^5 cells [33]. However, excessive inoculum densities may reduce light penetration and limit photosynthetic efficiency. Similarly, Sarada et al. (2002) observed that cultures with higher initial inoculum densities (3×10^4 cells mL^{-1}) exhibited reduced maximum cell density and prolonged growth periods before reaching the stationary phase, ultimately decreasing overall productivity [34]. These findings highlight the importance of optimising inoculum size in relation to light intensity to achieve efficient biomass production.

2.2.6. Carbon Dioxide

CO_2 is a crucial factor influencing microalgal growth, serving as the primary carbon source for photosynthesis. In the two-stage cultivation of *H. pluvialis*, achieving high cell density and synchronisation during the vegetative (green) stage is essential for maximising productivity. Optimised CO_2 enrichment has been shown to enhance both biomass growth and astaxanthin accumulation. For instance, increasing the CO_2 concentration in the sparging gas mixture to approximately 5% promotes astaxanthin synthesis without negatively affecting photosynthetic performance. Recent studies have also reported the development of *H. pluvialis* mutants with improved tolerance to elevated CO_2 levels (6–15%), exhibiting enhanced growth and astaxanthin accumulation. These strains demonstrate adaptations, including low-pH resistance and upregulation of metabolic pathways involved in pyruvate metabolism, carotenoid biosynthesis, and fatty acid production. Additionally, the use of sequential photobioreactors enables direct utilisation of flue gas as a cost-effective CO_2 source, reducing reliance on inorganic carbon supplementation. However, excessive CO_2 concentrations (>10%) can impose stress on the photosynthetic system, leading to inhibition of growth, cellular damage, and reduced astaxanthin production [22]. Therefore, maintaining an optimal CO_2 level is essential to balance carbon availability with cellular tolerance, ensuring efficient biomass production and astaxanthin accumulation.

2.3. Application and Market of Astaxanthin

Currently, more than 95% of global astaxanthin production is derived from synthetic processes using petrochemical feedstocks, primarily because of lower production and operating costs compared with biological production from *H. pluvialis* [1]. However, concerns regarding safety and limited approval for direct human consumption have restricted the use of synthetic astaxanthin mainly to aquaculture, where it is used as a feed additive to enhance the pigmentation of salmonids and shrimp. The use of synthetic astaxanthin in animal feed has been approved by regulatory authorities, such as the Food and Drug Administration (FDA), with a limit of 10 mg/kg or less [35]. Furthermore, reliance on non-renewable resources raises concerns about environmental sustainability. As the health benefits of astaxanthin become increasingly recognised, demand across industries, including food, nutraceuticals, pharmaceuticals, cosmetics, and aquaculture, has grown dramatically [9]. In 2016, global demand was estimated at approximately 280 metric tons, with prices ranging from 2000 to 7500 USD/kg [1], depending on the production method and purity [36]. The market for natural astaxanthin, particularly for human consumption, has strong growth potential, driven by consumer preference for safe, natural bioactive compounds. Despite its higher market value, astaxanthin derived from *H. pluvialis* remains more costly to produce, with production costs reaching up to 2500 USD/kg compared to approximately 1000 USD/kg for synthetic alternatives [9]. To address these challenges, strategies such as biorefinery approaches have been proposed to improve economic feasibility by valorising by-products, specifically, polyhydroxybutyrate (PHB) and triglycerides, and enhancing resource efficiency [37]. Astaxanthin has diverse applications, ranging from aquaculture feed additives to high-value products in nutraceuticals, pharmaceuticals, and cosmetics. Commercial products developed by companies such as Fuji Chemical Industry, including AstaREAL and related formulations, highlight the growing interest in natural astaxanthin, marketed in forms such as oils, powders, and capsules for health supplements, functional food and performance enhancement [8].

3. Smart Farming: An Overview

The agricultural sector plays a crucial role in supporting food security, economic stability, and national sustainability. However, the industry faces ongoing challenges, including a declining workforce and limited participation from younger generations, which hinder its advancement and technological adoption [38]. These factors contribute to inefficiencies and slow progress in implementing sustainable and modern agricultural practices, particularly in developing regions [39]. In this context, the integration of advanced technologies offers potential to improve productivity, efficiency, and sustainability in specific agricultural applications. Smart farming, also known as smart agriculture, involves integrating IoT and AI technologies into agricultural systems to enable precise, automated monitoring and control of farming operations [40]. IoT enables connected devices, such as sensors and control systems, to collect and transmit data in real time, supporting continuous system monitoring. AI techniques can then be applied to analyse these datasets, identify patterns, and support decision-making. The combined use of IoT and AI enables data-driven approaches, such as sensor-based monitoring and automated control, that can support the optimisation of specific farming processes [41]. Such approaches are particularly relevant for microalgae systems, including *H. pluvialis*, which are highly sensitive to environmental conditions, as illustrated in Figure 2.

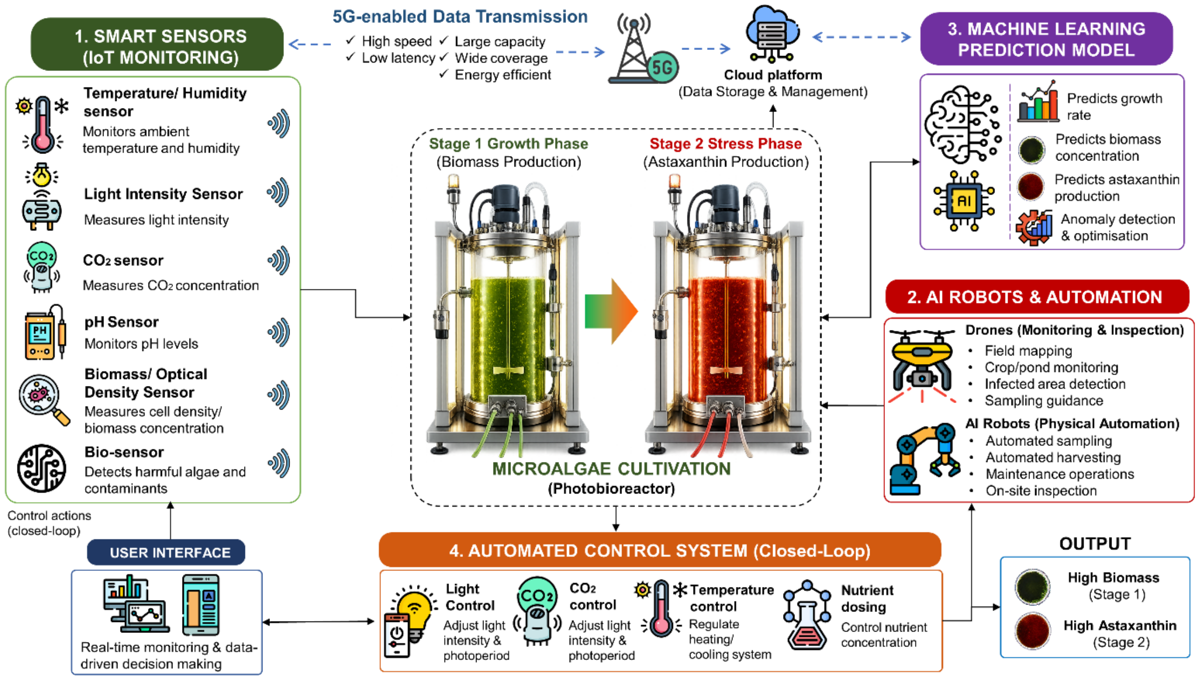

Figure 2. Conceptual framework of an IoT- and machine learning-integrated smart farming system for automated *H. pluvialis* cultivation and astaxanthin production. Smart sensors continuously monitor critical cultivation parameters, including temperature, humidity, light intensity, CO_2 concentration, pH, biomass density, and potential contaminants, and transmit real-time data via a 5G-enabled network to cloud-based storage and management systems. The cultivation process consists of a biomass production phase (green stage) followed by a stress-induced astaxanthin production phase (red stage). Machine learning models analyse the collected data to predict growth performance, biomass concentration, astaxanthin accumulation, and operational anomalies for process optimisation. AI-driven automation, including drones and robotic systems, supports monitoring, inspection, harvesting, and maintenance activities. The entire platform operates as a closed-loop control system that dynamically regulates environmental and nutritional conditions to maximise biomass productivity and astaxanthin yield.

3.1. Smart Sensors

Smart sensors are important components in smart farming system, enabling continuous monitoring of key operational conditions. Recent advancements in sensor technology allow the measurement of environmental variables, for example, temperature, humidity, CO_2, and illumination, which are relevant for *H. pluvialis* growth and astaxanthin production [42]. Furthermore, emerging biosensors offer enhanced capability for detecting harmful algal species, providing improved sensitivity compared to some conventional laboratory-based methods [43]. These sensors can be integrated into IoT networks to support real-time data collection and transmission to users and control systems. The collected data may then be analysed using AI-based approaches to support decision-making and adaptive control. Earlier implementations of such systems were constrained by communication limitations, including latency and bandwidth. However, advancements in wireless communication technologies, including the transition from 3G and 4G to 5G networks, have improved data transfer rates and reduced latency [44]. These developments can enhance the reliability and responsiveness of monitoring systems, supporting more timely adjustments in controlled environments such as photobioreactors and open pond systems used for *H. pluvialis* cultivation.

3.2. Artificial Intelligence (AI) Robots

Drones and robotic systems are being explored as components of IoT-enabled monitoring and automation frameworks, primarily for large-scale agricultural applications. Drones equipped with sensors and GPS technology can support environmental monitoring, data acquisition, and field mapping [45]. However, their direct application to microalgae cultivation, including *H. pluvialis*, remains limited. In microalgae-related studies, drones have mainly been used for environmental mapping and for identifying sampling locations in algal biodiversity research, for example, in the study of the biodiversity of Bulgarian Extremophilic algae [46]. These applications suggest potential for supporting large-scale outdoor systems, such as raceway ponds, but do not yet extend to routine cultivation control.

Similarly, AI-enabled robotic systems have been developed to automate selected tasks such as irrigation and pesticide application in agriculture [47]. While these systems demonstrate the feasibility of automation, their application to *H. pluvialis* cultivation is still at an early stage and may be limited to controlled or experimental settings. Emerging technologies, such as microalgae-based biohybrid microrobots, have shown capabilities in cellular-level imaging and analysis [48]. Although promising, these technologies remain largely experimental and have not yet been implemented in large-scale cultivation systems.

From an economic perspective, the benefits of automation depend on cost and scalability. Previous studies suggest that higher levels of automation do not necessarily translate to lower production costs, and that partial automation may provide more balanced outcomes [49]. While robotic systems can improve monitoring precision and process consistency, high implementation costs remain a key limitation [50]. Therefore, further research is needed to evaluate the feasibility and cost-effectiveness of integrating these technologies into *H. pluvialis* cultivation systems.

4. Machine Learning (ML)

ML has become a widely used approach for analysing complex datasets that are difficult to process using conventional methods. It has been applied in various data-intensive fields, including finance and pharmaceuticals, and is increasingly being explored in agricultural and biotechnological systems. The growing use of IoT technologies has generated large volumes of data, creating a need for advanced analytical tools to support data interpretation. ML techniques enable systems to learn from data and identify patterns, thereby assisting in data-driven decision-making.

A typical ML workflow involves training models on datasets with known variables to learn underlying relationships. These trained models can then be applied to new datasets for tasks such as prediction or classification. ML approaches are generally categorised into supervised and unsupervised learning, depending on the availability of labelled data [51]. Model selection is guided by dataset characteristics, and multiple models are often evaluated to identify the most suitable approach.

Model performance is typically assessed using a subset of data reserved for validation, with metrics such as accuracy used to evaluate predictive capability [52]. Based on these evaluations, models can be further optimised by adjusting parameters or selecting alternative algorithms. Once developed, ML models can be used to analyse new data and provide predictive insights that may support monitoring and optimisation processes in applications such as microalgae cultivation [53].

4.1. Learning Paradigms

The selection of ML techniques depends on the characteristics of the available data and the specific objectives of the application. In practice, multiple learning paradigms may be combined to improve performance. Techniques such as dimensionality reduction are often used alongside other methods to enhance efficiency and interpretability. This section provides an overview of key ML paradigms relevant to data-driven system analysis, including supervised, unsupervised, reinforcement, semi-supervised, and ensemble learning, as well as artificial neural networks (ANNs). Additional approaches such as instance-based learning, dimensionality reduction, and hybrid methods are also considered. Each paradigm offers distinct advantages and limitations depending on the data structure and application context. Table 2 summarises the main characteristics, general descriptions, and representative applications of these learning approaches.

Table 2. Description and representative use cases of different machine learning paradigms.

Learning Paradigms	Descriptions	Example Applications	References
Supervised Learning	• One of the most widely used ML approaches, where labelled data are used to train models for prediction. • The model learns relationships between input variables and known outputs, enabling it to predict outcomes for new data. It can be further divided into classification (discrete outputs) and regression (continuous outputs).	- Modelling electrodeionization (EDI) in wastewater treatment - Classification of microalgae species based on cell morphology - Prediction of astaxanthin accumulation using image-based colour intensity analysis	[54,55]
Unsupervised Learning	• A statistical-based approach to recognise unlabelled patterns. • It commonly involves clustering and grouping similar data points, enabling the model to classify new data based on similarity to existing clusters. • Typical tasks include clustering, dimensionality reduction, and anomaly detection.	- Classification of microalgal cells based on microscopy images	[56,57]
Reinforcement Learning	• Learn optimal actions for different situations to maximize cumulative rewards. • It operates as a closed-loop system, where actions influence future states. • Key components include a policy, reward signal, value function, and, optionally, an environment model.	- Optimisation of microalgae cultivation processes, such as adjusting light intensity, which was proven to increase production yield by 17%.	[58,59]
Semi-Supervised Learning	• A hybrid approach that combines supervised and unsupervised learning. • It leverages the limited availability of labelled data alongside larger unlabelled datasets to enhance learning accuracy and generalisation.	- Classification of microalgae based on microscopy images with improved accuracy (>90%) and enabling automated identification systems	[60–62]
Ensemble Learning	• An approach that combines multiple individual models to improve predictive performance. • The outputs from different predictions are aggregated, often using voting methods, to reduce bias and variance and enhance accuracy. • Classified into sequential approaches (e.g., AdaBoost) and parallel approaches (e.g., Random Forest).	- High-accuracy classification of microalgae cells based on microscopy images, achieving up to 97% prediction accuracy	[63,64]
Artificial Neural Network (ANN)	• A ML approach inspired by biological neural networks, consisting of interconnected layers, including input, hidden, and output layers • Each layer contains neurons that process information through weighted connections. • The model is trained by adjusting these weights to minimise error and improve prediction accuracy. • Advanced variants include deep neural networks (DNN), convolutional neural networks (CNN), and recurrent neural networks (RNN).	- Prediction of cell density of *H. pluvialis* based on environmental variables such as light intensity, illumination time, temperature, and pH, achieving up to 95% accuracy	[65,66]

4.2. ML-Integrated IoT Systems

The integration of ML with IoT technologies offers potential to enhance monitoring and control in microalgae production systems. IoT-enabled cultivation platforms facilitate continuous data collection from sensors that measure key parameters. In the context of microalgae cultivation, integrated IoT-ML systems can support decision-making through predictive tools such as Decision Support Systems (DSS). For example, Giannino et al. (2018) proposed a semi-automated framework using environmental data, such as temperature and airflow, to simulate microalgal growth and assist in adjusting operating conditions [67]. The combination of ML-based analysis with IoT-enabled monitoring can facilitate feedback-driven control strategies. Key parameters, such as cultivation time, temperature, and CO_2 supply, may be adjusted based on model outputs. While such approaches can enhance process monitoring and consistency, fully autonomous, real-time optimisation remains dependent on system design, data quality, and scalability considerations.

5. Integration of ML into Microalgae Cultivation

Microalgae cultivation has been attractive due to its diverse applications and environmental sustainability. Nevertheless, economic feasibility remains a major bottleneck for large-scale production, highlighting the need for continued research to improve and optimise current cultivation and harvesting technologies [68]. ML techniques offer opportunities to support data analysis and predictive modelling in microalgae systems. These approaches may reduce the time and complexity associated with conventional monitoring methods and enable estimation of growth behaviour and productivity. By applying predictive models, ML can assist in process optimisation and reduce reliance on manual analysis. Nevertheless, the effectiveness of such approaches depends on data availability, system variability, and model generalisability.

5.1. Prediction by Colour Intensity

Colour intensity is commonly used as an indicator of microalgal growth, as it correlates with biomass concentration [69]. At low biomass concentrations, cultures appear less opaque, allowing greater light transmission. As biomass increases, light absorption increases, resulting in darker cultures. This relationship can be quantified using absorbance measurements obtained from UV-Vis spectrophotometry. Recent studies have explored ML-based approaches for non-destructive estimation of microalgae growth using colour intensity. For instance, Salmi et al. (2022) developed a one-dimensional convolutional neural network (1D-CNN) model capable of simultaneously predicting microalgae species and biomass concentration [70]. The model utilised RGB images of samples, including both monoculture and mixed-species systems, combined with absorbance data as input features. The results demonstrated high classification accuracy, achieving approximately 95% accuracy for monocultures and 100% for mixed-species samples. Additionally, biomass prediction was achieved with mean errors of 22% and 24% for monoculture and mixed cultures, respectively. Although such approaches demonstrate potential, their applicability in large-scale *H. pluvialis* cultivation may depend on imaging conditions, environmental variability, and system calibration. Nevertheless, the use of image-based ML methods, including those based on accessible devices such as smartphone cameras, could support rapid, on-site monitoring and reduce reliance on laboratory analysis.

5.2. Biomass State Prediction

ML also shows potential for supporting downstream processing through predictive modelling of biomass characteristics. Various drying methods, including sun drying, spray drying, and freeze drying, present trade-offs between cost, efficiency, and product quality [71]. Economically viable methods, such as sun drying, often suffer from poor product consistency, whereas higher-quality techniques, including freeze and spray drying, remain costly for large-scale implementation. Additionally, variations in drying processes can affect cell morphology and structural integrity, further influencing downstream applications. Given that downstream processing can account for approximately 50–60% of total production costs in microalgae-based systems [72], integrating ML models offers a potential pathway to optimise operational decisions. To give an example, predictive models can be used to classify biomass states, estimate suitable drying strategies, and assess potential impacts on product quality. However, the application of ML to predict biomass viability and processing outcomes in microalgae systems remains limited. Recent studies have demonstrated the effectiveness of ML in predicting biomass production. Rodríguez-Rangel et al. (2022) compared multiple models, including ANN, CNN, LSTM, kNN, and Random Forest, and reported that a 1D-CNN model achieved the highest accuracy, with a mean squared error (MSE) of 0.0028 [73]. Similarly, Lopez-Exposito et al. (2019) showed that Random Forest Regression could accurately estimate floc size and geometry, improving harvesting efficiency and reducing production costs [74]. These findings suggest that ML approaches can support data-driven decision-making in microalgae processing, although further validation under practical cultivation conditions is required.

5.3. Prediction by Cell Count

Cell counting using microscopy and a haemocytometer remains a well-established method for assessing microalgal growth. Although more labour-intensive and time-consuming than optical methods such as colour intensity, it provides a direct and reliable measure of cell concentration, which is closely linked to biomass production. In contrast, optical approaches may be influenced by impurities or debris present in the culture, potentially reducing measurement accuracy [75]. Therefore, integrating ML with cell-counting approaches offers a promising pathway to accelerate analysis while retaining high accuracy.

While ML applications for microalgae cell counting remain limited, related approaches are well developed in biomedical imaging. As an example, CNN models have been successfully applied to classify different types of blood cells, such as red blood cells, white blood cells, and platelets, based on morphological features extracted from image datasets. One study achieved classification accuracies of 89.1%, 100%, and 96.61%, respectively, using a relatively small dataset of 500 images [76]. These results highlight the ability of CNNs to accurately distinguish cell types and segment individual cells in complex images, a critical step in automated cell counting. In addition, advanced image segmentation techniques have been developed to further automate counting processes. A hybrid framework combining a modified U-Net architecture with K-means clustering has been used to perform automated cell counting from stimulated Raman scattering (SRS) images of brain tumour samples, achieving a mean error of 12.08% [77]. These methods highlight the feasibility of combining deep learning and clustering techniques for cell detection and counting, providing a basis for adaptation to microalgae systems, including *H. pluvialis*.

In microalgae research, supervised and unsupervised ML approaches have been explored for cell counting [75,77]. Supervised models can be trained to perform multi-output predictions, such as estimating cell concentration while simultaneously classifying species. This may be particularly useful for analysing mixed cultures or monitoring dynamic changes in species composition. In contrast, unsupervised approaches are often used for image segmentation, such as identifying individual cells using clustering-based techniques. These methods can reduce the need for labelled data, thereby lowering the effort required for dataset preparation. Overall, ML-assisted cell counting is a promising approach to enhance the efficiency and scalability of microalgae growth monitoring. However, selecting appropriate methods involves trade-offs among accuracy, computational cost, and data requirements. Further work is needed to develop robust, scalable, and application-specific ML solutions for cell counting in *H. pluvialis* cultivation systems.

6. Integration of AI Technologies in *Haematococcus pluvialis* Cultivation and Astaxanthin Production

The integration of ML into astaxanthin production systems offers a powerful approach to enhance the efficiency and precision of *H. pluvialis* cultivation by analysing complex relationships between environmental conditions and biological responses (Figure 3). This enables accurate prediction of biomass growth, identification of the transition between green and red stages, and optimisation of stress conditions required for astaxanthin accumulation. In addition, advanced ML techniques, including CNNs, support non-invasive monitoring through image analysis, allowing real-time assessment of cell density, morphology, and pigment content. The integration of ML with automated control systems facilitates dynamic adjustment of cultivation parameters, creating a closed-loop system that continuously improves productivity and process stability. Consequently, ML-driven astaxanthin production systems contribute to more efficient resource utilisation, improved product consistency, and enhanced scalability of microalgae-based bioprocesses.

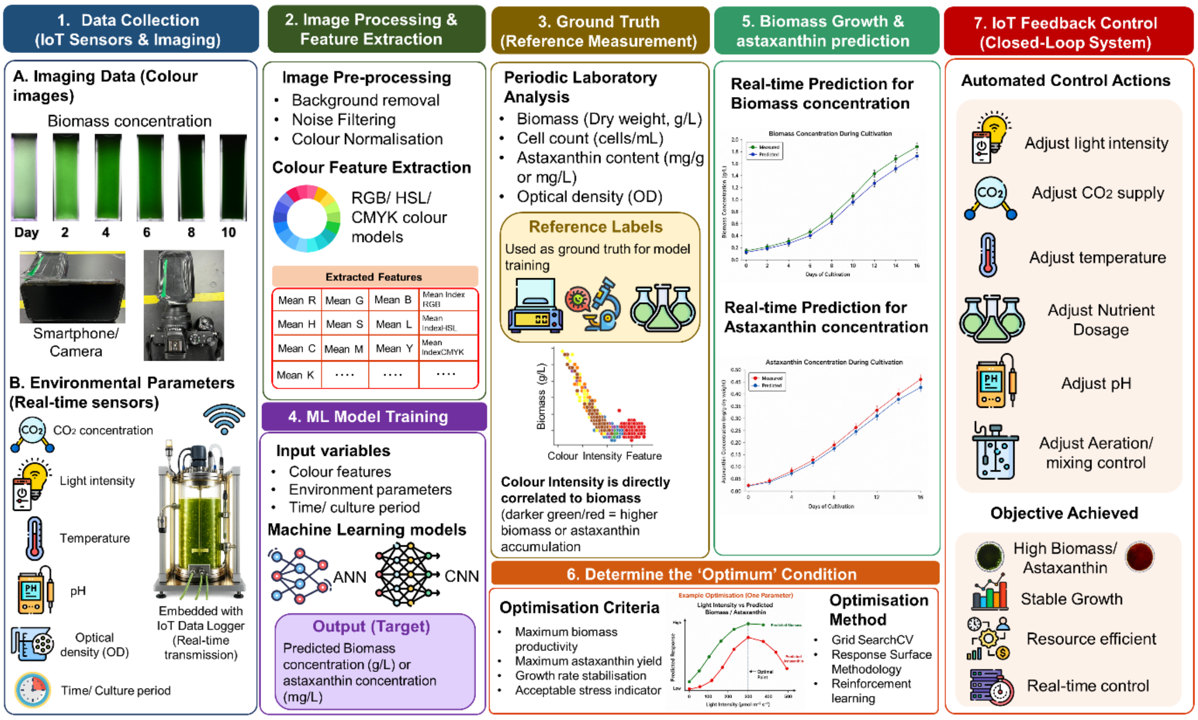

Figure 3. Workflow of an IoT- and machine learning-driven framework for biomass and astaxanthin prediction in *H. pluvialis* cultivation. The system begins with real-time data acquisition from imaging devices and embedded IoT sensors monitoring environmental and cultivation parameters, including CO_2 concentration, light intensity, temperature, pH, optical density, and cultivation time. Acquired images undergo preprocessing and colour feature extraction to obtain quantitative RGB/HSV/CMYK descriptors associated with biomass accumulation and astaxanthin production. Ground-truth laboratory measurements, including dry biomass weight, cell count, astaxanthin concentration, and optical density, are used to train machine learning models such as ANNs and CNNs. The trained models enable real-time prediction of biomass concentration and astaxanthin accumulation during cultivation. Prediction outputs are subsequently integrated into a closed-loop IoT feedback control system capable of dynamically adjusting cultivation parameters, including light intensity, CO_2 supply, temperature, nutrient dosage, pH, and aeration conditions, to achieve optimal biomass productivity, astaxanthin yield, and stable cultivation performance [78–80].

6.1. IoT and Smart Cultivation Systems

The integration of IoT technologies into *H. pluvialis* cultivation has enabled the development of smart and automated cultivation systems for improved process control and productivity. IoT-based systems use interconnected sensors, cameras, and automated control units to continuously monitor environmental parameters, such as temperature, light intensity, CO_2 concentration, and nutrient availability, in real time. These smart cultivation platforms enhance cultivation efficiency by enabling rapid data acquisition, remote monitoring, automated adjustments, and improved management of growth conditions, ultimately supporting stable biomass production and optimized astaxanthin accumulation (Figure 4).

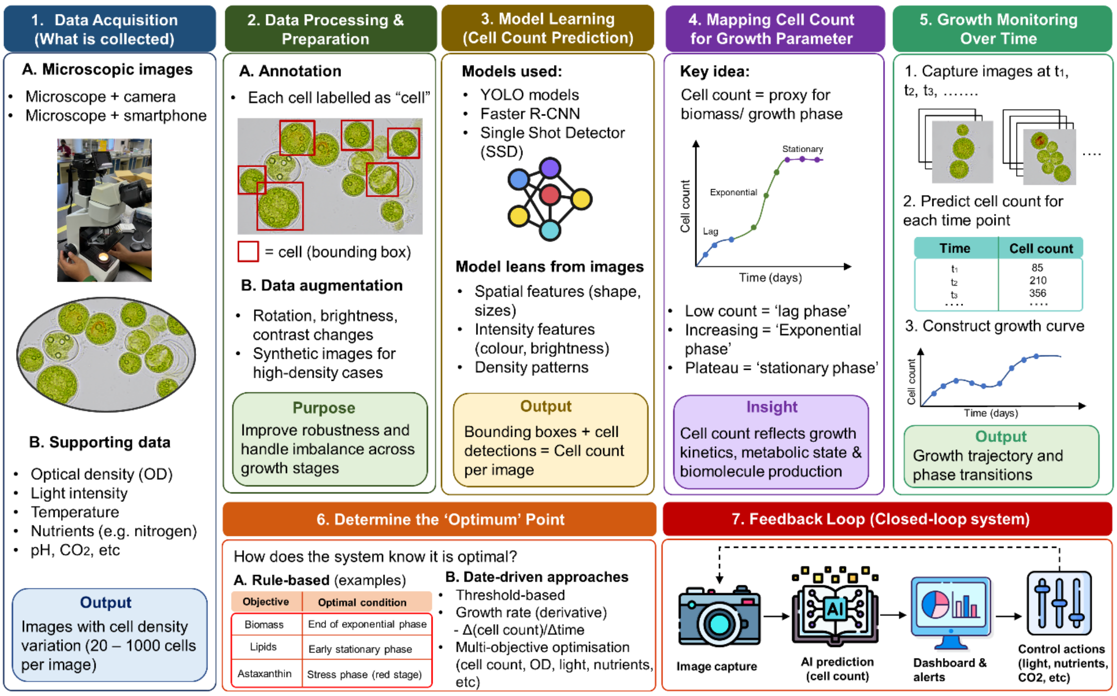

Figure 4. Workflow of a machine learning-based cell count prediction and growth monitoring framework for *H. pluvialis* cultivation. Microscopic images acquired using microscope-camera systems and supporting environmental data, including optical density, light intensity, temperature, nutrient concentration, pH, and CO_2 levels, are collected during cultivation. Acquired images are annotated and augmented before being used to train object detection models, including YOLO, Faster R-CNN, and Single Shot Detector (SSD), for automated cell detection and counting. The models learn spatial, intensity, and density-related image features to estimate cell abundance from cultivation images. Predicted cell counts are subsequently mapped to biological growth parameters, with cell density serving as a proxy for biomass accumulation and growth-phase transitions. Time-series predictions enable the construction of growth trajectories for monitoring lag, exponential, and stationary growth phases. Optimal cultivation conditions are determined using rule-based and data-driven optimisation approaches that account for biomass productivity, lipid accumulation, astaxanthin production, and growth stability. The framework is integrated into a closed-loop feedback system capable of automated monitoring, prediction, and dynamic adjustment of cultivation conditions to optimise microalgae productivity and astaxanthin yield [81,82].

Rudnicki et al. (2026) developed a 5 L automated stirred-tank photobioreactor (ST-PBR) for two-stage cultivation of *H. pluvialis*. Limitations of excessive shear stress and limited light penetration were addressed through phase-controlled illumination (60–300 μmol m^{-2} s^{-1}) and an integrated process control system with Keysight VEE Pro 9.33 software. The reactor design incorporated a dual marine impeller operating at a low tip speed (0.48 m/s) to maintain cell integrity, along with a precise gas-delivery strategy based on pH regulation. Performance evaluation demonstrated stable operation, achieving a biomass concentration of 1.315 g/L without sedimentation and an astaxanthin content of 4.12% dry weight, offering a scalable and cost-effective alternative to conventional photobioreactor designs for astaxanthin production [83].

To address the limitations of conventional analytical methods, Song et al. (2022) implemented a digital in-line holographic flow cytometry system based on a linear-array sensor to monitor *H. pluvialis* within a microfluidic platform. The system employed a two-dimensional hydrodynamic focusing chip and utilised a modified angular spectrum method to reconstruct holographic images, correcting distortions caused by differences between the flow velocity and the sensor acquisition rate. The imaging depth of focus was digitally enhanced to capture cells across the full depth of the microfluidic channel. This approach enables efficient and reliable analysis of microalgal growth characteristics under varying cultivation conditions, offering a cost-effective and scalable alternative to traditional measurement techniques [84]. Similarly, a predictive model was constructed to evaluate the growth dynamics of *H. pluvialis* during the cell proliferation phase. Key variables, including light intensity, pH, and cell radius, were selected as input parameters, with the microalgae's growth status as the output variable. Experimental data obtained from cultivation studies were analysed using a regression tree approach based on the Classification and Regression Tree (CART) algorithm. This model enables accurate prediction of algal growth behaviour by identifying relationships between environmental factors and cellular responses [85].

Sun et al. (2022) [86] explored ML-based image classification techniques for *H. pluvialis* cells. In this approach, features such as cell shape and pixel distribution were extracted from microscopic images, and classification models were trained to distinguish between different growth stages. Among the evaluated algorithms, including C4.5 decision trees, support vector machines (SVMs), and k-nearest neighbours (KNN), the decision tree model demonstrated superior performance, achieving an accuracy of approximately 97%, providing a practical solution for monitoring growth stages. Zhang et al. (2021) also established SVM-based classification model to distinguish the growth stages of *H. pluvialis*. A dataset of 150 samples was used for model training and evaluation, with separate training and test subsets. Different kernel functions were applied to assess their impact on classification performance, and model parameters were optimised using learning curve analysis. Model evaluation was conducted using confusion matrix metrics and receiver operating characteristic curve analysis. The results demonstrated that the SVM model effectively classified *H. pluvialis* cells into proliferation and induction stages, achieving an accuracy of 91.11% with a multi-directional kernel and sigmoid functions [87].

To identify the optimal time point for applying astaxanthin-inducing conditions and initiating the transition to the second production stage, Wei et al. (2017) designed an organic electrochemical transistor (OECT)-based sensing platform to monitor the cellular state of *H. pluvialis*. Variations in channel current, caused by the accumulation of *H. pluvialis* cells on a poly(3,4-ethylenedioxythiophene): polystyrene sulfonate (PEDOT: PSS) film, were used as indicators of growth progression. Distinct signal changes were observed following combined blue light irradiation and sodium bicarbonate treatment, with stabilisation occurring approximately 120 min after treatment, corresponding to the maturation of the senescent phase. This enabled precise identification of the optimal timing for astaxanthin induction, highlighting the potential of OECT-based biosensors as rapid and non-invasive tools for process control in microalgae cultivation [88]. Ma et al. (2026) also introduced a smart Optical Imaging-Assisted Acoustic Streaming Enrichment System (SOAES) to enhance the selective enrichment of *H. pluvialis* with high astaxanthin content. The system integrated a stress-inducing cultivation module with an intelligent perception–decision framework and acoustofluidic technology. A Morphology-Optics Enhanced YOLO (MOE-YOLO) algorithm was employed to analyse optical images and construct a dynamic map of microalgal growth states, capturing both morphological features and intracellular astaxanthin accumulation. This information was combined with a random forest regression model to optimise key operational parameters, including acoustic amplitude and flow rate, enabling precise, label-free enrichment under varying conditions. Experimental results demonstrated that, under blue light exposure and 20% seawater concentration, the system achieved an enrichment factor of 9.4, with astaxanthin content increasing by approximately 125% compared to initial levels [89]. Overall, smart cultivation systems offer significant potential to improve the efficiency, automation, and reliability of *H. pluvialis* cultivation while reducing manual intervention and operational complexity.

6.2. ML-Assisted Monitoring for Growth, Biomass, and Astaxanthin Prediction

ML technologies can be used for real-time monitoring and prediction of *H. pluvialis* growth, biomass accumulation, and astaxanthin production. By integrating image analysis, sensor data, spectroscopy, and cultivation parameters, ML models can identify growth phases, monitor the green-to-red transition, and estimate astaxanthin content with improved accuracy and efficiency. These approaches enable non-destructive and automated monitoring systems that support cultivation optimization, early stress detection, and determination of the optimal harvesting stage for enhanced astaxanthin productivity.

A study by Liyanaarachchi et al. (2026) demonstrated the use of an ANN to optimise astaxanthin production from *H. pluvialis* by accounting for multiple interacting cultivation factors. A central composite design (CCD) was used to structure the experiments, with light intensity and initial concentrations of nitrate, phosphate, and biomass selected as key variables. Data were collected over a 26-day cultivation period in 1 L photobioreactors, and the dataset was subsequently expanded to enhance model training. The model predicted a peak biomass concentration of 1.02 g/L at day 13 and an astaxanthin concentration of 28.76 mg/L (3.06%) at day 18 under nitrate-depleted conditions. Experimental validation confirmed strong agreement with the predicted outcomes, demonstrating the effectiveness of ANN-based optimisation for improving cultivation performance and supporting efficient, large-scale production [90]. Calderini et al. (2025) applied reflectance hyperspectral imaging (HSI), combined with a one-dimensional CNN (1D-CNN), to estimate astaxanthin content in *H. pluvialis* and high-performance liquid chromatography as a reference method. The model showed high predictive accuracy, with an average error of 5.9% under consistent cultivation conditions, although performance declined at very low astaxanthin levels (<0.6 μg/mg). Compared with conventional linear regression approaches based on limited wavelength data, the CNN model demonstrated superior performance. However, variations in cultivation conditions altered spectral profiles, thereby reducing model accuracy. This limitation was addressed through transfer learning, which partially retrained the model, improving adaptability and reducing prediction error to around 8.2% [91]. Besides, Lee et al., (2022) introduced an efficient screening method based on the negative phototaxis behaviour of *H. pluvialis* to identify mutants with enhanced astaxanthin production. A polydimethylsiloxane (PDMS)-based microfluidic platform, combined with controlled light exposure, was used to evaluate the phototactic responses of mutant strains. A photosynthesis-deficient mutant exhibited reduced sensitivity to blue light, confirming the relationship between photosynthetic efficiency and phototaxis. Using this principle, photosensitive mutants were selected from a large mutant library. The selected strain (M1) demonstrated improved performance, showing a 1.17-fold increase in growth rate and a 1.26-fold enhancement in astaxanthin accumulation compared to the wild type. These findings highlight the effectiveness of phototaxis-based screening as a rapid and reliable tool for identifying high-performing strains [92]. In short, ML-assisted monitoring systems provide a promising approach for the non-invasive assessment of astaxanthin accumulation to enhance astaxanthin productivity.

7. Future Prospects

The potential of ML in microalgae production is substantial, with advanced algorithms capable of addressing complex processes and enabling predictive optimisation when sufficient data are available. However, despite extensive research, the adoption of ML in routine industrial practice remains limited, not only in the microalgae sector but also in other data-rich fields such as healthcare [93]. This limitation is largely due to challenges in data availability, as well as the time, cost, and effort required for data collection, preprocessing, and model development, particularly for small and medium enterprises. Limited access to skilled and experienced personnel in artificial intelligence, data science, and system integration may also hinder the effective deployment and maintenance of these advanced systems. Implementing these sophisticated technologies requires expertise in data processing, model development, and system integration, which may not be readily available in all regions or industries. This skills gap can lead to challenges in system optimisation, reduced reliability, and increased dependency on external technical support. Consequently, insufficient technical capacity may slow down the adoption of intelligent microalgae cultivation systems and limit their scalability. Therefore, future efforts should also focus on capacity building, interdisciplinary collaboration, and training programs to support the adoption of AI technologies in the microalgae industry.

In the microalgae industry, species-specific variations in morphology, pigmentation, and growth characteristics further complicate model generalisation, requiring extensive data collection and model customisation. ML models developed for one species may not be directly transferable to others, necessitating extensive data acquisition and model retraining. Additionally, conventional data collection methods, such as microscopy-based cell counting, remain labour-intensive and often require laboratory-based analysis, limiting the efficiency gains offered by ML integration. Although emerging IoT-enabled imaging technologies offer potential solutions for real-time monitoring, these systems are not yet fully optimised or widely adapted for microalgae applications [48].

Economic considerations also play a critical role in adoption. The profit-to-investment (PIR) ratio represents an important consideration when transitioning from conventional monitoring approaches to IoT- and AI-driven automation systems. The economic feasibility of adopting higher levels of automation must be carefully evaluated to determine whether the associated benefits justify the required investment. Although previous studies suggest that increasing automation can reduce operational costs, these gains may reach a point of diminishing returns [50]. Furthermore, these findings are often based on general agricultural systems and may not directly reflect the specific economic dynamics of microalgae production. Therefore, more targeted studies are required to assess the cost-effectiveness of automation in microalgae cultivation and to guide informed decision-making for industrial adoption.

Nevertheless, ongoing technological advancements are expected to improve the accessibility and affordability of IoT and ML solutions. Future developments may include real-time monitoring systems that integrate image-based analysis of colour intensity with ML models to predict continuous growth. Furthermore, the establishment of large, shared datasets, particularly for microscopic images across different microalgae species, could enable the development of more generalised and transferable ML models. The development of shared datasets and generalised models, combined with improved sensor technologies, will enable scalable and efficient microalgae production systems. Such advancements would significantly reduce reliance on manual data collection and enhance scalability, paving the way for intelligent, efficient, and commercially viable microalgae cultivation systems.

8. Conclusions

The growing demand for natural astaxanthin has significantly increased the value of biologically derived products, driving extensive research into improving production processes. This review has examined the cultivation of astaxanthin from *H. pluvialis* and highlighted the potential of integrating IoT and ML technologies to enhance production efficiency. Current advancements in IoT-enabled monitoring and ML-based data analysis offer promising opportunities to optimise cultivation conditions, improve biomass productivity, and enable real-time decision-making. In parallel, the application of ML in microalgae systems is still in its early stages, with significant opportunities for developing robust predictive models for growth, productivity, and process optimisation. Despite their potential, the adoption of IoT and ML technologies in large-scale microalgae production is constrained by limited data availability, data quality issues, the need for specialised expertise, and implementation costs. Addressing these challenges will require continued research, improved data infrastructure, standardised datasets, and interdisciplinary collaboration. In summary, the integration of IoT and ML has the potential to transform microalgae cultivation into a more efficient, scalable, and sustainable process, supporting the growing global demand for natural astaxanthin.

Author Contributions

J.W.R.C.: Conceptualization, Data curation, Visualization, Writing—original draft, Funding acquisition, D.Y.Y.T.: Data curation, Investigation, Methodology, Funding acquisition, Writing—review and editing. S.-M.P. & T.S.S.: Writing—review and editing, Investigation, Supervision. P.L.S.: Project administration, Supervision. All authors have read and agreed to the published version of the manuscript.

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Conflicts of Interest

The authors declare no conflict of interest.

Use of AI and AI-Assisted Technologies

During the preparation of this work, the author(s) used ChatGPT (OpenAI) to assist with language editing and conceptual development of graphical illustrations. After using this tool/service, the author(s) reviewed and edited the content as needed and take(s) full responsibility for the content of the published article.

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