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Research article

Combo chloro-photosynthetic device and applications for greenhouse gas reduction campaign and smart agriculture

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ABSTRACT

The increasing carbon dioxide (CO_2) levels in the air pose a direct threat to all living organisms and the environment. Leveraging the ability of plants to absorb CO_2 is one of the most effective methods for countering these rising CO_2 levels. The present study aimes to develop a combo photosynthetic and chlorophyll-a sensor based on Non-Dispersive Infrared (NDIR) spectroscopy and an optical method. This sensor enables simultaneous, intensive measurement of net photosynthesis and chlorophyll-a content and yields accurate information. Comparative analysis of the efficacy of the sensors to that of a commercial instrument demonstrated that the measurement values obtained from the developed photosynthetic and chlorophyll-a sensors were not significantly different from those acquired with the commercial instrument (portable photosynthesis system LI-6400) and chlorophyll metre (SPAD-502), with a 95 % confidence level. Furthermore, the developed photosynthetic sensor could be used as a new correlation unit for chlorophyll-a content and net photosynthesis. Therefore, the sensor can be used to propose effective plantation processes to reduce atmospheric CO_2 levels and in smart farming systems to control the quality of yields.

1. Introduction

Climate change has emerged as a major global threat, with forests playing a major role in regulating and alleviating its impacts by lowering the levels of atmospheric CO_2 [1–4]. Forests, the world's largest carbon sinks, aid in mitigating climate change via carbon sequestration. Hence, to implement effective policy measures and management planning, the assessment of carbon stocks in forests is pivotal [5]. Approximately 30 % of the Earth's surface area is covered by forests, harbouring 19 % of the global biomass and carbon pool [6,7]. Forests play a significant role in the global carbon balance by acting as both carbon stocks and sources, thus influencing atmospheric CO_2 levels. Forests store carbon in trees and soil through photosynthesis; therefore, CO_2 stocks in tree biomass resulting from photosynthesis play a major role in effectively sequestering atmospheric CO_2 levels, a key factor in climate change. Carbon stocks

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in trees and soils can be categorised into five carbon pools: aboveground biomass, belowground biomass, deadwood, litter, and soil organic matter [8]. However, the potential carbon stock in natural forests may vary depending on various factors, including the type of forest, plant composition, density, climate, and other environmental factors. Rich natural forests typically feature dense populations of large trees and, hence, high levels of biomass and carbon stocks. Furthermore, carbon stocks in tree biomass in forestry are mainly dependent on the biomass type, which varies depending on the plant species, age, growth stage, environment, and the silvicultural practices adopted. Generally, different plantation operations result in varying tree densities within an area. Consequently, forests, which consist primarily of trees of the same age, exhibit a higher tree density and, as a result, accumulate more biomass and carbon [9].

The net photosynthetic value of each plant species varies depending on internal, external, and environmental factors. Internal factors, including plant species and chlorophyll content, directly influence the net photosynthesis value. Plants can be categorised into three groups based on carbon fixation: C3, C4, and crassulacean acid metabolism (CAM). C3, C4, and CAM plants utilise the Calvin cycle to produce sugar from CO_2. Each pathway used to fix CO_2 has benefits and limitations, allowing plants to adapt and survive in different habitats [10]. For example, the C4 and CAM plants can survive effectively in hot and dry regions, whereas C3 plants thrive best in cool climates. In hot and dry environments, C4 plants exhibit higher rates of CO_2 assimilation than those of C3 plants. Moreover, they perform optimally under high light intensities [11], owing to their increased energy requirements. Therefore, being productive in places with high temperatures and a low water supply is ideal for crop plants.

Another internal factor that directly influences plants is chlorophyll, a vital internal factor that affects plants and their ability to capture, transfer, and convert light energy. Chlorophyll is essential for plant growth and development [12,13]. Therefore, to enhance photosynthetic activity, it is crucial to maintain a high concentration of chlorophyll in leaves [14,15]. However, the ability of plants to absorb light for food production depends on their total photosynthetic pigment content [16]. In addition, chlorophyll can influence plant physiology, and the chlorophyll content of a leaf can indicate the physiological state, stress, and nutritional status of the plant [17–20]. Notably, a decrease in the chlorophyll content of leaves leads to a reduction in the primary production and photosynthetic activities of plants [21], indicating that the chlorophyll content in leaves plays a vital role in plant productivity [22].

External factors, including light, carbon dioxide, temperature, relative humidity, and airflow [23], can directly influence the rate of photosynthesis as they affect the rates of chemical reactions in the photosynthetic pathway [24]. Therefore, environmental factors can significantly impact plant growth and development; alterations in these factors may result in severe damage or even the death of the plant, ultimately affecting the quality of the produce. For example, when the temperature in the environment exceeds 35 °C, various stages of plant development, such as seed germination, seedling growth, vegetative growth, flowering, and seed maturity, may be negatively impacted [25,26]. Consequently, plantation planning aimed at reducing CO_2 levels in the atmosphere relies heavily on highly accurate tools capable of measuring the rate of photosynthesis, which fluctuates according to the amount of CO_2 absorbed by each plant for photosynthesis.

In addition to CO_2, which is vital for photosynthesis, chlorophyll is an essential component. Chlorophyll, a green pigment present in plants, algae, and cyanobacteria, performs an essential function during the initial stage of photosynthetic energy transduction by absorbing sunlight. Chlorophyll is involved in harvesting light and converting it into an electrochemical potential across the photosynthetic membrane. This results in reactions that convert atmospheric CO_2 to carbohydrates, thereby preserving the energy of short-lived photons into longer-lasting products. Initially, chlorophyll molecules are energised, creating excited states that last for several nanoseconds (10^{-9} s). Subsequently, they contribute to the membrane potential, stabilising for several seconds. High-energy products, such as adenosine triphosphate (ATP) and reduced nicotinamide adenine dinucleotide phosphate (NADPH), which have lifetimes of several minutes or even hours, are then generated. Ultimately, this process results in the production of storage substances such as starch and sugar. However, various photosynthetic organisms may exhibit different photopigment compositions and photosynthetic mechanisms, owing to their adaptations to their specific physical environments [27]. The chlorophyll content (Chl) present in leaves serves as a sensitive indicator of the external environmental stress [28], photosynthetic capacity [29,30],vigour, and metabolic activity [31] of the plant. Overall, both CO_2 and chlorophyll content have major impacts on plant photosynthesis. In addition, chlorophyll content directly affects photosynthesis and reflects the growth status of crops and their exchange of materials and energy with the surrounding exterior environment [32–34]. Therefore, limited light availability significantly affects chlorophyll pigment content and photosynthesis [35].

In a previous study, an alternative photosynthetic sensor with comparable performance to commercial tools without any significant difference [36], was successfully developed, enabling the simultaneous measurement of the correlation between net photosynthesis and chlorophyll content. This simultaneous measurement can reduce measurement errors and provide more precise readings. This developed sensor can also offer a new unit in real-time and record additional parameters such as light intensity, temperature, and relative humidity. The combo chloro-photosynthetic sensor serves as a valuable tool for the in-depth study of plants, as they can rapidly and accurately measure parameters related to plant growth. Furthermore, the aim of developing this combo chloro-photosynthetic sensor is to establish a cost-effective and user-friendly alternative device, rendering it more accessible to interested parties, including educational institutions, government agencies, private organisations, and farmers. In addition, this sensor can facilitate the proposal of a plantation process tailored to a given environment and climate, as the same plant species may undergo different rates of photosynthesis when in different environments and climates. Notably, a sensor system can regulate the functioning of smart farming systems. The primary objective of smart farming technologies is to improve productivity by minimising costs and waste generation, reducing the impact on crops and livestock, and improving overall quality [37]. Smart farming operations use a variety of sensors to gather data, allowing for a more comprehensive understanding of the operational environment, which encompasses the dynamic conditions of crops, soil, weather, other environmental factors, and the operation itself. This increase in data can lead to enhanced accuracy and accelerated decision-making processes [38–40]. Therefore, planting in an optimal environment and climate for the plant species can enhance yield quality and contribute to the effective reduction of atmospheric CO_2.

The present study extends upon basic research on a portable photosynthetic sensor [36], a tool used to measure net photosynthesis. While net photosynthesis alone is sufficient to indicate plant growth rate, it may not capture all aspects of growth in a statistically significant manner. Therefore, incorporating chlorophyll content as an additional parameter enhances the accuracy of predicting plant growth correlated with photosynthesis. This study assessed the efficiency of our developed device, coupled with a photosynthetic and chlorophyll-a sensor, compared to that of another commercial tool. Consequently, the simultaneous of measurement net photosynthesis and chlorophyll content simultaneously enabled the calculation of their correlation as a new unit.

2. Materials and methods

2.1. Photosynthetic sensor

The photosynthetic sensor [36] was developed and compared to the commercial device.

Generally, plant growth or changes depend on both internal and external factors. Internal factors include characteristics of the leaves such as their construction, surface area, stomata, weight-to-area ratio, Nitrogen, and chlorophyll levels. External factors, or environmental factors, that affect plant growth include light, temperature, water, humidity, and nutrition [41–43]. During the design stage, sensors were carefully selected and installed to measure external or environmental factors. These sensors detect CO_2 concentration, light intensity, temperature, and relative humidity, which directly influence the photosynthesis process. The data from these sensors were used to calculate the net photosynthesis in the general unit, which can be compared and utilised in other research endeavours.

2.2. Systematic design of the chlorophyll sensor

The combo chloro-photosynthesis device was developed by integrating a chlorophyll-a sensor to measure and predict plant growth more accurately. The rationale behind developing a chlorophyll-a sensor is the higher concentration of chlorophyll-a than that of chlorophyll-b in plants. In general, for every gram of leaves, there are approximately 1.21 mg of chlorophyll-a and 0.43 mg of chlorophyll-b [44]. Accordingly, chlorophyll-a constitutes approximately three-quarters of the pigment content in plants [45], playing a crucial role in converting solar energy into chemical energy. The workings and components of the commercial tool explained the theory behind the design as follows. The absorption spectrum of chlorophyll is distinctive, as it absorbs photosynthetically active radiation (PAR) during photosynthesis. However, chlorophyll molecules absorb blue and red wavelengths more effectively than green molecules. As a result, green light is reflected from or transmitted through the leaves, making them appear green. This is depicted by a peak near 550 nm in the leaf transmittance (or reflectance) spectrum. In addition, chlorophyll is less effective at absorbing wavelengths

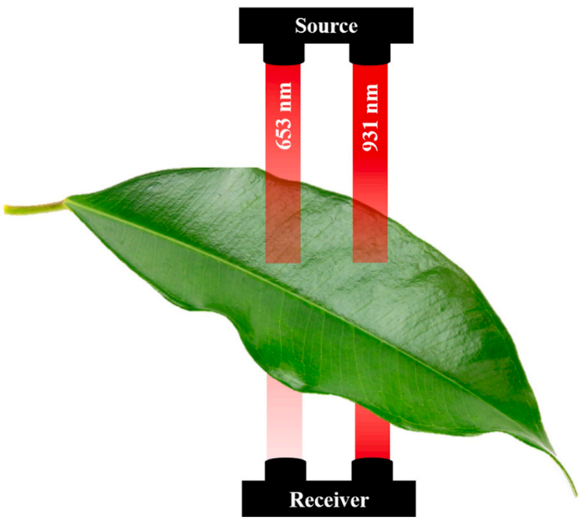

Fig. 1. The light source and receiver of the chlorophyll sensor.

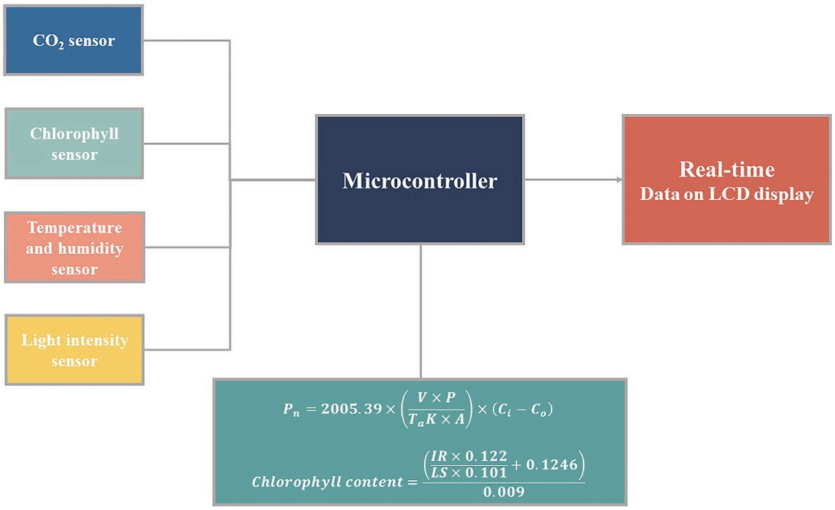

Fig. 2. Components of the advanced photosynthetic sensor.

above 700 nm. Hence, as the wavelength approaches 700 nm, the graph experiences a rapid change from low to high transmittance, as observed in the leaf transmittance spectrum. This sudden change in a leaf transmittance spectrum is called the 'red edge', where wavelengths above approximately 700 mm—or 'near-infrared (NIR)—do not get absorbed by the leaves for photosynthesis [46]. The commercial tool incorporates two light-emitting diodes (LEDs), one centred at 653 nm and the other at 931 nm, as the light source. Once light passes through the leaf sample, the paired detectors measure the light intensity to examine the transmittance of the leaf.

The relative chlorophyll content was determined by examining the light transmittance ratio at 931 nm to that at 653 nm, as 653 nm is within the PAR range and can be absorbed by chlorophyll for photosynthesis. However, 931 nm is an NIR wavelength, indicating that chlorophyll cannot absorb it. Therefore, this reading serves as a reference for investigating the mechanical differences between the leaves. 'Chlorophyll Content Index' (CCI) refers to the ratio of light transmittance between two wavelengths 931 nm and 653 nm (Fig. 1) [47]. The wavelength of infrared radiation can measure leaf thickness, while red light, or laser, measures leaf 'greenness'.

Additionally, during the design process of the prototype combo chlorophotosynthesis device, general units of measurement were used. This ensured that the tool could measure and display standardised values consistently, facilitating comparisons with other research findings in the study.

2.3. Chlorophyll content calculation

The ratio of leaf light transmittance between wavelengths 653 nm and 931 nm, as used in the equation below [46], can be used to calculate the chlorophyll content:

$$Chlorophyll\ content = \frac{\left(\frac{IR \times 0.122}{LS \times 0.101} + 0.1246\right)}{0.009} \quad (1)$$

where IR and LS are transmissions at 653 and 931 nm, respectively.

2.4. Developing the combo chloro-photosynthetic sensor

Fig. 2 shows the configuration of the combo chloro-photosynthetic sensor. A microcontroller was connected to each sensor to detect the chlorophyll content, residual CO_2 concentration, and ambient parameters. To detect the CO_2 concentration, an air pump and flow metre transport the residual CO_2 from the leaf chamber through a tube to the carbon dioxide sensor at a flow rate ranging from 0.3 to 1.0 L/min [48]. The chlorophyll sensor required two light sources with different wavelengths: infrared light and a laser with 653 and 931 nm wavelengths to measure chlorophyll content. It is also necessary to use a receiver as the light source to measure light transmission through the medium. The light source and receiver were placed opposite each other, as shown in Fig. 1. The net photosynthetic and chlorophyll contents were compiled using ambient CO_2 and the ratio of leaf light transmittance data under the operating command of the microcontroller.

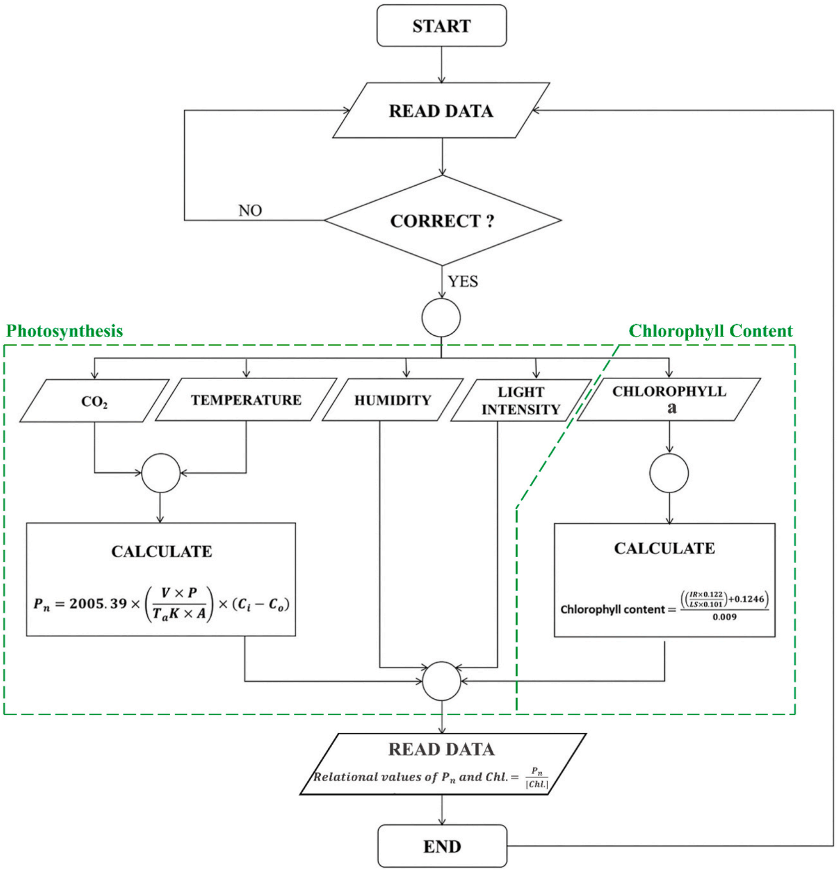

Fig. 3. The advanced photosynthetic sensor process flowchart.

Fig. 4. Advanced photosynthetic sensor.

2.5. Operation of the combo chloro-photosynthetic sensor

Fig. 3 illustrates the mechanism of the combo chloro-photosynthetic sensor. Each sensor began taking measurements at the beginning of the operation. During the data acquisition stage, once the smart sensor node received a measurement command, measurements of the five parameters – CO_2 concentration, temperature, relative humidity, light intensity, and chlorophyll content – were also collected from the sensors. Subsequently, two response variables were utilised to determine the net photosynthesis of the plants. Data from the receiver were then used to calculate the chlorophyll content using a microcontroller that could adjust and compute the data. Finally, these parameters were printed out in real-time, including net photosynthesis and chlorophyll content. This process was

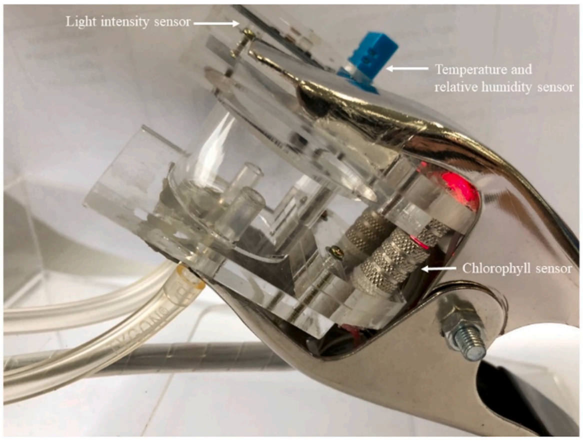

Fig. 5. Leaf chamber and other sensors.

repeated every 2 s.

2.6. Function of the combo chloro-photosynthetic sensor

The prototype of the combo chloro-photosynthetic sensor, shown in Fig. 4, was operated by attaching the leaf chamber to a leaf. An air pump was then utilised to circulate CO_2 from the chamber to the CO_2 sensor for measurement. Simultaneously, other sensors, namely the temperature sensor, relative humidity sensor, light intensity sensor, and chlorophyll sensor, were utilised to gather data on the environmental factors and the chlorophyll content. The CO_2 concentration and temperature results were then used to calculate net photosynthesis in $\mu molCO_2/m^2/s$ [23,36]. In addition, data from the light source and receiver were calculated to determine the chlorophyll content using Equation (1). Once completed, the overall results were displayed in real-time on the LCD panel.

2.7. Evaluation of the combo chloro-photosynthetic sensor

Given the high precision and accuracy of the photosynthetic sensor [36] only a chlorophyll sensor was tested and compared with a commercial tool (Minolta SPAD-502, Konica Minolta). The chlorophyll metre measured the transfer of light at red and infrared wavelengths of 650 and 940 nm, respectively, in plant leaves. This non-destructive evaluation produces a numerical output indicating the level of chlorophyll concentration and leaf greenness [15]. In addition, the chlorophyll content values obtained from the chlorophyll sensor and commercial tool on the same plants with leaves of different shades were stored and analysed to evaluate their efficiency and accuracy.

2.8. Statistical analysis

The chlorophyll content values obtained from both the developed chlorophyll sensor and the commercial tool, which collected data from identical leaf samples, were statistically analysed to evaluate the efficiency and accuracy of the chlorophyll sensor compared to the commercial tool. This analysis was performed using SPSS software (SPSS In., Chicago, IL, USA). An independent-sample *t*-test was conducted to compare the average chlorophyll content obtained from the chlorophyll sensor and those from the commercial tool. The significance was set at 0.05.

3. Results

3.1. Overview and functionality of the combo chloro-photosynthetic sensor

The combo chloro-photosynthetic sensor consisted of a CO_2 sensor, temperature sensor, relative humidity sensor, light intensity sensor, and an LED light source for the chlorophyll sensor. These primary sensors take measurements, calculate and process the values through the microcontroller, and display the measurements in real-time on an LCD.

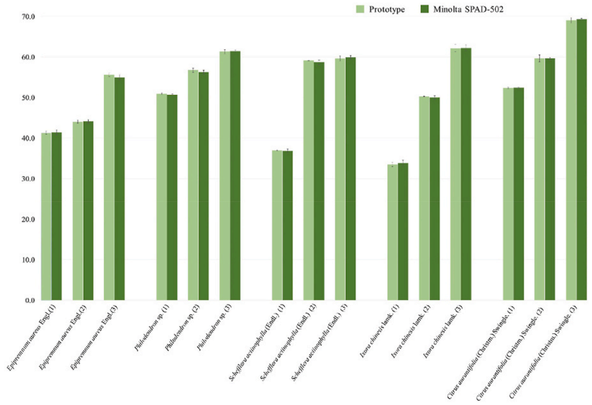

Fig. 6. The mean chlorophyll contents obtained from the advanced photosynthetic sensor (prototype) in comparison to the commercial tool (Minolta SPAD-502) in different plant samples.

The combo chloro-photosynthetic sensor was operated by attaching the leaf chamber to the plant leaf to measure its CO_2 concentration. The air pump within the sensor draws CO_2 from the leaf chamber through the CO_2 sensor to measure the leaf CO_2 concentration. At the same time, other sensors, which consist of the temperature, relative humidity, light intensity, and chlorophyll sensors, were also installed near the leaf chamber (Fig. 5) for measurements. These values were then calculated and displayed on an LCD screen. These measurements consisted of net photosynthesis (in $\mu molCO_2/m^2/s$), chlorophyll content (in SPAD), temperature (in °C), relative humidity (%RH), and light intensity (in LUX). In addition, the relationship between net photosynthesis and chlorophyll content was also shown in $mmolCO_2/molChl.s$.

3.2. Performance of the chlorophyll sensor

The mean chlorophyll contents of several plant species and various leaf shades were measured using the prototype and the commercial tool Minolta SPAD-502 to test the performance. The results are summarised in Fig. 6 and Table 1. The chlorophyll content of *Epipremnum aureus* Engl., *Philodendron* sp., *Schefflera actinophylla* (Endl.) Harms, *Ixora chinesis* Lamk., *Citrus aurantifolia* (Christm.) Swingles with three different leaf shades, as measured by the prototype, differed from the commercial tool by averages of 0.48 %, 0.51 %, 0.23 %, 0.32 %, and 0.61 %. This result indicates that the readings obtained from the prototype differed from those of the commercial tool by less than 1 %. Furthermore, the statistical analysis, as shown in Table 1, revealed significance levels greater than 0.05. Therefore, the mean chlorophyll content obtained from the prototype was not significantly different from that obtained using the commercial tool.

Table 1 presents the data and statistics obtained from the chlorophyll sensor and a commercial tool (Minolta SPAD-502; Konica Minolta) using an independent-sample *t*-test. This study focused on the chlorophyll content of three leaves under different shades from each plant species. The independent-sample *t*-test revealed a P-value of 0.82, 0.82, 0.07, 0.05, 0.10, 0.63, 0.30, 0.08, 0.36, 0.31, 0.23, 0.91, 0.57, 0.99 and 0.31 for the three leaves with different shades from each plant species. Given that all P-values exceeded 0.05 (95 % confidence level), the effectiveness and accuracy of the chlorophyll sensor developed in this study were not significantly different compared to those of the commercial tool.

Fig. 7 shows that the chlorophyll content obtained from the sensor did not differ from that obtained from the commercial tool. Similarly, the relationships between chlorophyll concentration (g/m^2) and SPAD values from the Minolta SPAD-502 were curvilinear for all three species. The slope of the relationship between the chlorophyll concentration and SPAD increased with increasing SPAD, and the data fit well with exponential functions with the two parameters [49].

These data confirm a direct correlation between both CCI and SPAD and chlorophyll content. By developing a regression model that correlates foliar chlorophyll content with SPAD chlorophyll content [50], the relationship was established between chlorophyll content obtained using a conventional method (mg/cm^2), and a chlorophyll metre (SPAD CCI). The graph reveals a significant correlation ($r^2 = 0.9029$) between the chlorophyll content obtained from the conventional method and that measured using the chlorophyll metre, indicating that SPAD-502 is an effective tool for non-destructively estimating total foliar chlorophyll content. Moreover, this tool can be used on plants of different ages, growing conditions, and genotypes [50,51].

Table 1
The chlorophyll contents and other statistical data obtained from the chlorophyll sensor and the commercial tool (Minolta SPAD-502, Konica Minolta).

Plants		Commercial	Chlorophyll Sensor	Significant (P-value)
		SPAD unit (±SD)		
Epipremnum aureus Engl. (1)		41.38	41.45 (±0.59)	0.82
Epipremnum aureus Engl. (2)		44.12	44.06 (±0.36)	0.82
Epipremnum aureus Engl. (3)		55.02	55.64 (±0.41)	0.07
Philodendron sp. (1)		50.72	50.95 (±0.15)	0.05
Philodendron sp. (2)		56.27	56.77 (±0.88)	0.10
Philodendron sp. (3)		61.45	61.34 (±0.80)	0.63
Schefflera actinophylla (Endl.) (1)		36.82	37.03 (±0.08)	0.30
Schefflera actinophylla (Endl.) (2)		58.72	59.19 (±0.10)	0.08
Schefflera actinophylla (Endl.) (3)		59.93	59.63 (±0.60)	0.36
Ixora chinesis Lamk. (1)		33.88	33.51 (±0.49)	0.31
Ixora chinesis Lamk. (2)		50.02	50.30 (±0.08)	0.23
Ixora chinesis Lamk. (3)		62.23	62.18 (±0.91)	0.91

(*continued on next page*)

Table 1 (*continued*)

Plants		Commercial	Chlorophyll Sensor	Significant (P-value)
		SPAD unit (±SD)		
Citrus aurantifolia (Christm.) Swingle. (1)		52.45	52.40 (±0.15)	0.57
Citrus aurantifolia (Christm.) Swingle. (2)		59.67	59.67 (±0.85)	0.99
Citrus aurantifolia (Christm.) Swingle. (3)		69.38	69.07 (±0.66)	0.31

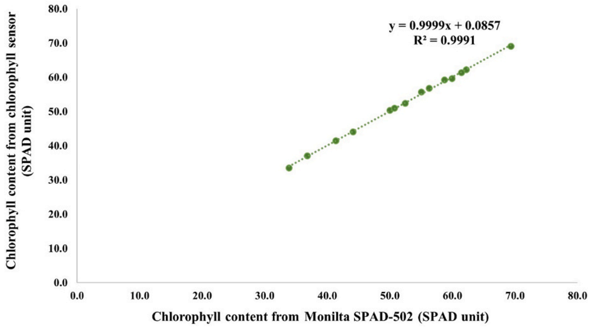

Fig. 7. Correlation between chlorophyll sensor and commercial tool.

Table 2
Converting the correlation between net photosynthesis and chlorophyll content to a new unit.

Plants	Photosynthesis (μmolCO_2/ m^2/s)	Chlorophyll Content (SPAD unit)	Chlorophyll Content (μmolChl./m^2)	Pn/Chl. (mmolCO_2/ molChl.s)
Tradescantia spathacea Swartz.	4.83	39.90	264.32	18.27
Citrus aurantifolia (Christm.) Swingle.	9.99	45.60	325.60	30.68
Aglaonema modestum Schott.	9.67	45.70	326.71	29.60
Excoecaria cochinchinensis Lour.	4.84	57.60	466.32	10.38
Dracaena loureirin Gagnep.	6.91	58.05	471.90	14.64
Bougainvillea spp.	15.00	58.20	473.77	31.66

3.3. *Correlation of net photosynthesis and chlorophyll content as a new unit of measurement*

In this study, a correlation was observed between net photosynthesis and chlorophyll content, which is expected as plants naturally maximise their ability to absorb light for photosynthesis by adapting their chlorophyll content as a survival mechanism. This adaptation positively impacts growth and long-term survival [52]. Conversely, as the chlorophyll content decreases, the photosynthetic rate of the plants also decreases. Decreased chlorophyll content limits the ability of leaf tissues to absorb light for photosynthesis [53]. Additionally, stress-induced damage to chloroplasts results in redox imbalance and decreased photosynthetic efficiency, which may eventually result in cell death [54].

In the present study, calculations of the relational values between net photosynthesis and chlorophyll content, through the simultaneous measurement and conversion of data using Equation (2), enabled more precise predictions regarding plant growth. The results of this process are summarised in Table 2. The relational values of net photosynthesis and chlorophyll content were calculated as follows:

$$Relational\ values\ of\ P_n\ and\ Chl. = \frac{P_n}{[Chl.]} \tag{2}$$

where P_n is the net photosynthesis in $\mu molCO_2/m^2/s$ and [Chl.] is chlorophyll content $\mu molChl./m^2$. This equation can convert chlorophyll content (SPAD unit) to chlorophyll content ($\mu molChl./m^2$) as follows:

$$Total\ Chl. = 0.055(SPAD^2) + 6.05(SPAD) - 64.64 \tag{3}$$

where total chlorophyll is total chlorophyll in $\mu molChl./m^2$ and SPAD is the chlorophyll content in the SPAD unit [55].

Table 2 presents the net photosynthesis and chlorophyll contents measured from six plant species: *Tradescantia spathacea* Swartz., *Citrus aurantifolia* (Christm.) Swingle., *Aglaonema modestum* Schott., *Excoecaria cochinchinensis* Lour., *Dracaena loureiri* Gagnep., and the *Bougainvillea* spp. The net photosynthetic values of the plants were 4.83, 9.99, 9.67, 4.84, 6.91, and 15.00 $\mu mol/m^2/s$, and their chlorophyll contents (SPAD) were 39.90, 45.60, 45.70, 57.60, 58.06, and 58.20 respectively. The chlorophyll contents obtained from the chlorophyll sensor were then mathematically converted to $\mu mol/m^2$ to enable the calculation of the correlation between the two variables. A linear equation of the relationship between the leaf chlorophyll content ($\mu mol/m^2$) and the SPAD reading of orange trees grown using a hydroponic system was established, enabling the conversion of chlorophyll content from SPAD to $\mu mol/m^2$ possible [54] using Equation (3). The chlorophyll contents (in $\mu mol/m^2$) of the six plant species were 264.32, 325.60, 326.71, 466.32, 471.90, and 473.77 $\mu mol/m^2$, respectively. Mathematical Equation (2) was then used to determine the correlation between net photosynthesis and chlorophyll content. The correlations between the two variables of the six plant species were 18.27, 30.68, 29.60, 10.38, 14.64, and 31.66 $mmolCO_2/molChl.s$, respectively. The correlations are presented in $mmolCO_2/molChl.s$, a new unit of measurement that allows for accurate prediction of plant activity (besides net photosynthesis and chlorophyll content).

Therefore, as shown in Table 2, calculating and converting the net photosynthesis and chlorophyll content into the unit $mmolCO_2/molChl.s$ can be beneficial during the plant selection process to identify plant species with high CO_2 absorption potential. Moreover, the results indicated a direct variation in the photosynthetic rate and a correlation between net photosynthesis and chlorophyll content. As shown in Table 2, although the plants had similar net photosynthetic values, their chlorophyll content differed depending on their species, age, and leaf position for light absorption. For example, the net photosynthetic value of *Tradescantia spathacea* Swartz., and *Excoecaria cochinchinensis* Lour. were 4.83 and 4.84, whereas their chlorophyll contents were 39.90 and 57.60, respectively. The correlation between the net photosynthesis and chlorophyll content, displayed in $mmolCO_2/molChl.s$, were 18.27 and 10.38, respectively. These values differed owing to the differences in chlorophyll content. Therefore, *Excoecaria cochinchinensis* Lour. had high chlorophyll content owing to its long leaf age, which is consistent with a previous study [56].

In addition, the findings of the present revealed that the dynamic values of the leaf greenness index measured using the SPAD chlorophyll reading, the leaf nitrogen concentration, and the absorption range of chlorophyll correlated with leaf development. This finding suggests a relationship between leaf age and location and their greenness and nitrogen content. Therefore, the correlation between the net photosynthesis and chlorophyll content in *Tradescantia spathacea* Swartz., and *Excoecaria cochinchinensis* Lour. indicated that *Tradescantia spathacea* Swartz. exhibits higher growth and CO_2 absorption rates than *Excoecaria cochinchinensis* Lour.

The data presented in Table 2, collected during the plant selection process, indicated *Aglaonema modestum* Schott., and *Citrus aurantifolia* (Christm.) Swingle. as the optimal species for mitigating CO_2 levels in the atmosphere, as they have a relatively low chlorophyll content compared to their high net photosynthesis values. Consequently, they can photosynthesise and absorb CO_2 in places with low light availability, such as in dense forests with obstructed sunlight, act as ground cover, or grow in areas with low light.

3.4. Economic analysis

During the development of the combo chloro-photosynthetic sensors, it was found that the developed invention, which is equivalent to the commercial tool, costs only 0.5 % of the commercial tool's price (Table 3). Although the invention's functionality may differ from that of the commercial device in certain aspects, it is not notably different from its ability to measure and display net photosynthesis and chlorophyll content. Using this invention as an alternative to a commercial tool will help reduce the land costs of imported products. In addition, it will positively and indirectly impact the economy if productively used in various fields. Owing to its affordability compared to commercial tools, the invention will become a more accessible alternative for students, agricultural scientists, and researchers. For instance, the combo chloro-photosynthetic sensor can measure the photosynthesis rate, facilitating strategic planning of the plantation process to efficiently reduce greenhouse gas emissions. Users can also utilise the invention in a smart farming system to ensure high-quality yields that meet customer needs and thus generate long-term, sustainable income.

Table 3
The percentage of the price of the advanced photosynthetic sensor compared to a commercial instrument.

Commercial instrument		Prototype (USD)	The percentage of the price of the prototype exposed to commercial instrument
Model	(USD)		
Portable photosynthesis system LI-6400	55,000+	280	0.51 %
Minolta SPAD-502 (Chlorophyll meter)	2,500+	15	0.60 %

4. Discussion and recommendations

4.1. Accuracy of the combo chloro-photosynthetic sensor

Table 1 presents the standard deviations (SD), which represent the accuracy of repeated tests and indicated deviations in measurements, of the comparison of the chlorophyll content values obtained from the developed tool and the commercial tool. Typically, smaller SD values indicate higher precision and accuracy of the data. The measurement results shown in Table 1 have SD values in the range of ±0.08–0.91. Notably, throughout this study, repeated measurements were conducted to test and compare the tools using identical samples, thus ensuring accurate data. However, during these repeated measurements, the tools must be reset to zero by removing them from their initial position each time. The tools are then repositioned to repeat the measurements. During this process, discrepancies in the data may occur, as the resulting position may vary from the original one. As the probes used for measurement are small, approximately 0.5–1 cm, achieving the same measurement location is challenging, leading to different measured values for each repeated measurement.

Leaves are an essential part of plants that are sensitive to changes in environmental factors. Therefore, the function of leaves varies depending on environmental conditions [57,58]. Adjustments in leaf area, thickness, and chlorophyll content enable plant leaves to adapt to different light intensities and qualities. For instance, under low-light conditions, the leaf area expands, facilitating better light capture and utilisation by the plant, thus promoting growth [59–61]. Furthermore, under drought conditions, plant leaves become smaller, thicker, and more densely packed. This adaptation reduces the transpiration rate, minimises water loss, and enhances the ability of the plant to withstand drought stress [62,63]. Therefore, plant leaves can adapt to environmental conditions for survival. Consequently, the colour or appearance of the leaves varies due to adaptations to different environmental conditions. This variation could contribute to fluctuations in chlorophyll content measurements, depending on the position of the leaf being measured.

4.2. Factors affecting leaf chlorophyll content

Plant development is influenced by a combination of internal and external factors. External factors, such as light, temperature, humidity, and abiotic stress, significantly impact the expression of both nuclear and chloroplast genes. Specifically, light is crucial in controlling various aspects of plant development, such as chloroplast formation and chlorophyll metabolism [64–67]. Abiotic stress factors, such as drought, extreme temperatures, and strong light, can alter gene expression within the chlorophyll biosynthesis pathway, resulting in decreased enzyme activity. This reduction in enzyme activity can hinder chlorophyll biosynthesis, thereby impacting chlorophyll content [67]. Furthermore, adequately watered plants exhibit higher chlorophyll-a content across all genotypes, whereas those subjected to water scarcity exhibit a lower content [68].

In addition, the chlorophyll content of each plant differs depending on the leaf age. The estimation of chlorophyll levels in aromatic coconut leaves revealed that leaves of different ages contain different chlorophyll contents [56]; young leaves have the lowest chlorophyll content, followed by mature leaves, with older leaves having the highest chlorophyll content. Moreover, in other plant species, such as Tulsi and soybean leaves, chlorophyll levels tend to increase from the youngest leaves to those deemed 'photosynthetically mature'. Subsequently, after reaching peak chlorophyll concentration, the levels gradually decrease [69,70]. In the present study, different leaf positions affected chlorophyll content. Specifically, the leaf apex exhibited the lowest chlorophyll content, followed by the leaf apex, and the leaf base exhibited the highest chlorophyll content.

4.3. Relationship between net photosynthesis and chlorophyll content in relation to CO_2 absorption and emission

The photosynthetic capacity of a plant is typically affected by both internal and external factors. Externally, factors such as solar radiation, atmospheric CO_2 levels, temperature, wind speed, vapour pressure, and the availability of water and nutrients play crucial roles in influencing photosynthesis. For example, the photosynthetic process tends to slow down with reduced sunlight intensity, lower temperatures, and limited water and nutrients. In addition, internal factors encompass leaf characteristics, such as their construction, surface area, stomata, weight-to-area ratio, N, and chlorophyll levels [71], all of which can influence the photosynthetic capacity.

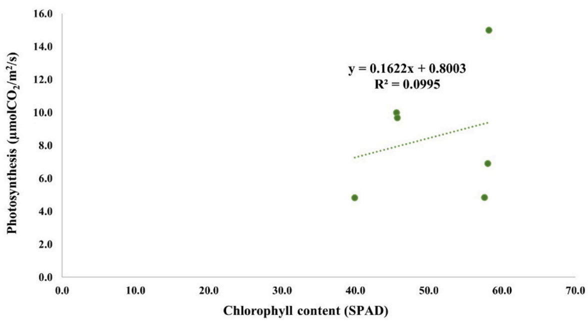

Fig. 8. Correlation between net photosynthesis and chlorophyll content.

Consequently, there is a direct correlation between the chlorophyll content in a plant and its photosynthetic efficiency (Fig. 8). Higher chlorophyll content enhances plant photosynthesis, resulting in improved crop yield [72–75]. According to the relationship between net photosynthesis and chlorophyll content (Equation (2)), if the net photosynthesis is high and the chlorophyll content is low, the plant accumulates more carbon in its biomass due to a lower respiration rate, resulting in less carbon being released from respiration. Furthermore, the amount of chlorophyll in chloroplasts is related to mitochondrial function, as plants utilise light energy to photosynthesise and convert atmospheric CO_2 into carbohydrates. These carbohydrates are subsequently broken down through respiration, which occurs in the cytosol and mitochondria, to produce the energy and carbon intermediates necessary for biosynthesis. Thus, for optimal carbon fixation and growth in plants, strict coordination between the two energy-transforming organelles is required [76], as respiration utilises substrates provided by photosynthesis, while cellular photosynthesis depends on respiration to create compounds such as ATP. Typically, a plant with a low chlorophyll content may exhibit a low number of mitochondria present [77]. In addition, a previous study using quantitative hybridisation analysis indicated generally higher mitochondrial DNA levels in white sectors than in green sectors. However, despite these higher mitochondrial DNA levels, dark respiration rates are generally lower in white sectors than those observed in green sectors [78]. This, in turn, can cause a lower respiration rate, resulting in plants using less accumulated CO_2 than plants with high chlorophyll content and mitochondria. Therefore, these plant species can serve as suitable sources of CO_2 (carbon sinks), particularly for reducing atmospheric CO_2 levels.

Using this correlation, the proposed new unit obtained from the invention presented in this study provides comprehensive information related to the carbon stock rate of the plant. This information can be used for carbon reduction planning.

4.4. Limitations in the reproducibility of the tool

As the combo chloro-photosynthetic sensor integrates the previously developed net photosynthesis sensor [36] with the chlorophyll sensor, it enables in-depth plant measurements and simultaneously displays results across all parameters.

Notably, in designing the chlorophyll sensor, careful selection of the appropriate light source was crucial. Furthermore, meticulous consideration was given to the design and positioning of the light source and receiver for accurate and precise measurements. The coding part consisted of two main sections: command code to initiate sensor operation and code for calculations and displaying measurement results on a display screen. This included parameters such as light intensity, temperature, relative humidity, net photosynthesis, chlorophyll content, and the correlation between net photosynthesis and chlorophyll content. These measurement results could be used to enhance the accuracy of predictions regarding plant growth.

5. Conclusions

The combo chloro-photosynthetic sensor, equipped with CO_2, temperature and humidity, light intensity, and chlorophyll sensors, offers high accuracy in measuring parameters and enables real-time data presentation. In comparative tests against a certain commercial instrument, the readings from each sensor were found to be statistically similar, with a 95 % confidence level. Similarly, there was no significant variance observed between the measurements of the invented tool and those from other commercial tools, also at a 95 % confidence level. Furthermore, the introduction of a new unit concerning the relationship between net photosynthesis and chlorophyll content provides an efficient tool for predicting plant growth. Notably, the combo chloro-photosynthetic sensor is a cost-effective, accessible, and convenient tool for studying plants and can be utilised in various fields. For example, it can be used to propose an ideal plantation process tailored to a specific area or plant species to maximise CO_2 absorption and reduce CO_2 emissions during plant respiration. In addition, government agencies and the general public can access the data acquired from such inventions for natural resource management. Involving the public in managing resources with the government and granting them access will foster a sense of ownership, leading to increased awareness and participation in resource restoration and conservation, thereby prolonging resource longevity. Moreover, the combo chloro-photosynthetic sensor can aid in smart farming systems by utilising sensors to accurately measure and control variables within an enclosed greenhouse. This smart system differs from the control system used in general greenhouses, as the invention allows the user to control the plant's photosynthetic rate, directly affecting their growth. They can also be used to control plant tissue cultures or establish optimal conditions for the growth of aquatic plants, including algae. This approach preserves the quality of the product and reduces external risks, promotes environmentally friendly production practices, contributes to food security, and positively impacts the country's economy.

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Data availability statement

The data presented in this study are available upon request to the corresponding author.

CRediT authorship contribution statement

Natsuda Khampa: Writing – original draft, Methodology, Formal analysis, Data curation. **Suwanna Kitpati Boontanon:**

Supervision. **Sayam Aroonsrimorakot:** Supervision. **Narin Boontanon:** Writing – review & editing, Supervision, Conceptualization.

Declaration of competing interest

The authors declare the following financial interests/personal relationships which may be considered as potential competing interests:Dr.Narin Boontanon reports financial support was provided by National Research Council of Thailand. Dr.Narin Boontanon reports a relationship with National Research Council of Thailand that includes: funding grants. If there are other authors, they declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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