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Assessment of Scientific Payload Carrying Spirulina Onboard Blue Origin's New Shepard Vehicle

By Pedro J. Llanos, Morgan Shilling, Kristina Andrijauskaite, Kody Kidder & Vijay V. Duraisamy

Abstract- The research team at ERAU and UTHSCSA analyzed the effects of suborbital flight stressors and various light conditions (red, white, no light) on the *Arthrospira platensis* (Spirulina), onboard Blue Origin's New Shepard vehicle. Commercially available cyanobacterium species were cultivated and closely monitored in mother colonies several months before the suborbital flight mission. The aim of this study was to estimate the biomass production and growth as a potential dietary alternative for prospective human spaceflight's life support system. Spirulina samples were flown in a NanoLab with adjacent avionics supporting the light conditions and sensors to monitor the temperature, relative humidity, and accelerations. The various flight parameters measured in the NanoLab were validated with the flight data gathered by Nanoracks, the flight integrator. Thus, we also assessed the effect of microgravity and different light conditions on the gene expression. Our data indicates that the Spirulina samples onboard the rocket had significant ($p < 0.01-0.0001$) downregulation of majority of the gene expression.

Keywords: blue origin's new shepard, microalgae, spirulina, suborbital flight, microgravity, life support system, gene expression, food source.

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Assessment of Scientific Payload Carrying Spirulina Onboard Blue Origin's New Shepard Vehicle

Pedro J. Llanos ^α, Morgan Shilling ^α, Kristina Andrijauskaite ^ρ, Kody Kidder ^ω & Vijay V. Duraisamy[‡]

Abstract—The research team at ERAU and UTHSCSA analyzed the effects of suborbital flight stressors and various light conditions (red, white, no light) on the *Arthrospira platensis* (Spirulina), onboard Blue Origin's New Shepard vehicle. Commercially available cyanobacterium species were cultivated and closely monitored in mother colonies several months before the suborbital flight mission. The aim of this study was to estimate the biomass production and growth as a potential dietary alternative for prospective human spaceflight's life support system. Spirulina samples were flown in a NanoLab with adjacent avionics supporting the light conditions and sensors to monitor the temperature, relative humidity, and accelerations. The various flight parameters measured in the NanoLab were validated with the flight data gathered by Nanoracks, the flight integrator. Thus, we also assessed the effect of microgravity and different light conditions on the gene expression. Our data indicates that the Spirulina samples onboard the rocket had significant ($p < 0.01-0.0001$) downregulation of majority of the gene expression. However, when comparing different light effects, the red light had the most significant effect on most genes, especially for genes involved in magnesium (mgtE) and nitrate-nitrite (nrtP) transport. Finally, we used laser-scanning confocal microscopy to provide high-resolution imaging visualizations of the Spirulina under different conditions (ground, flight, and light conditions). Results indicate that flight samples exposed to red light had the most profound effect on gene expression and showed an enhanced behavior suggesting that photosynthetic organisms are influenced by light energy. Our findings indicate that Spirulina may be able to survive and grow under various light conditions and at lower temperatures than optimally cultivated conditions for several weeks.

Keywords: blue origin's new shepard, microalgae, spirulina, suborbital flight, microgravity, life support system, gene expression, food source.

I. INTRODUCTION

Future human spaceflight exploration to expand and sustain human presence in low-Earth orbit (LEO) and eventually to the Moon requires hybrid systems that can support human health, life support and habitation systems, such as food preparation, processing, and preservation in various environments. These are several research factors that space agencies (NASA and ESA) are further exploring to better understand key technologies that can support and

enhance human health performance and environmental control life support systems (Fahrion J, Mastroleo F, Dussap C-G & Leys, 2021). Plant growth has no major obstacles in space if its environment has proper ventilation, lighting, and temperature and humidity control. To promote a sustainable human presence in space, large scale food production is required (most food supplied to astronauts is ferried with space vehicles to the International Space Station) but large-scale test for food production in reduced gravity is still a paradigm since special electrical equipment sets a limit for the large-scale plant production (Poulet, Fontaine & Dussap, 2016)

Spirulina (*Arthrospira sp.*) is a blue-green microalgae dated back to 3.5 billion years and is the most well-known of algae among other types, such as the spirulina maxima and fusiformis (Moraes, Arruda, Maresca, Antune & Moraes, 2012). Spirulina is a filamentous cyanobacteria (micro-algae) characterized by cylindrical, multicellular trichomes in an open left-handed helix. Spirulina platensis has been used as human food in Africa, Asia, and South America, whereas Spirulina maxima is confined to Central America. Spirulina platensis flourishes around 25°C-26°C with high alkaline pH (9.5-11).

Spirulina platensis is considered a potent nutrient-dense whole food with about 65%-71% protein content, rich in vitamins A in the form of beta carotene, B complex, and highly rich in minerals, such as potassium, calcium, zinc, magnesium, manganese, selenium, iron, and phosphorus (Mukhopadhyay, 2015). It is low in fat, but does contain vital essential fatty acids, including very high amounts of Gamma Linolenic Acid (GLA). GLA is a hormone precursor and is found conducive to healthy heart functioning and circulation. It also has anti-inflammatory properties, which are beneficial for skin and hair. It has been hailed as the "food of the future", besides being considered as an ideal food for astronauts by NASA (Poulet, 2016). Spirulina was consumed by the ancient Aztecs but became popular again when NASA proposed that it could be grown in space for use by astronauts.

With our present study, we intend to provide further insight of the environment in which our experiment was exposed to including preflight, launch site and post-flight operations after our experiment was

Author: e-mail: llanosp@erau.edu

successfully flown in the suborbital flight aboard Blue Origin's New Shepard vehicle launched on May 2, 2019, from West Texas Launch Site. This study also provides laboratory findings about the cultivation and production of biomass of the *Spirulina platensis*, ribonucleic acid (RNA) extraction and confocal microscopy – tools that can help maturing prospective experiments and leveraging flight operation efforts. Finally, in this study, we share our insights on gene expression analyses obtained from flown spirulina samples which were compared to the ground controls.

Previous work on the cultivation of *Spirulina platensis* up to 25 days conducted by Kumar, Kulshreshtha, and Singh (2011) has shown a wide range of temperature tolerance (20.0°C - 40°C) and various light intensity (lux) ranges (500 lux – 3,500 lux with dry weight of about 0.5 g/L and 0.6 g/L for 1,000 lux and 1,500 lux, respectively at the end of the 25th day. The ideal temperature for *Spirulina platensis* is around 35°C. Any deviation from this value will affect its productivity and at 17°C, *Spirulina* can enter a dormant state where the biological processes stop but the viability continues (Keller et al., 2021).

Research by Delrue et al. (2017) conducted *Spirulina platensis* cultivation for 28 days (dry weight 0.7-1.1 g/L) and 40 days (dry weight 2-2.1 g/L). Research work by Yim et al. (2016) showed spirulina platensis cultivation times of 10 days under various light sources and light intensities (200 lux – 2,000 lux) with overall dry weight up from about 0.01 g/L to near 1.2 g/L and most cases around 0.4 g/L. Other studies by Kumari, Pathak and Guria (2015) showed the spirulina growth under various light emitting diodes intensities with yields between 0.8 g/L to 1.2 g/L after 18 days cultivation. More recent work by Prates, Radmann, Duarte, Greque de Moraes and Costa (2018) revealed *Spirulina platensis* biomass production yields about 0.8 - 1.8 g/L for various LEDs for 10 days cultivation period. Research by Moraes et al. (2012) cultivated *Spirulina platensis* for 23 days under controlled temperature (30°C). Research work by Wang, Fu, and Liu (2007) provided *Spirulina platensis* dry weights of up to 0.4 g/L for 5 days cultivation period using several light intensities (I) and a mathematical growth model which can be used for our system.

$$\mu_{red} = 0.02 \frac{g}{day} + \frac{0.44 \frac{g}{day} \cdot \left| \left(I - 391.3 \frac{\mu mol}{s \cdot m^2} \right) \right|}{461.2 \frac{\mu mol}{s \cdot m^2} + \left| \left(I - 391.3 \frac{\mu mol}{s \cdot m^2} \right) \right|}$$

$$\mu_{white} = 0.02 \frac{g}{day} + \frac{0.32 \frac{g}{day} \cdot \left| \left(I - 398.4 \frac{\mu mol}{s \cdot m^2} \right) \right|}{773.5 \frac{\mu mol}{s \cdot m^2} + \left| \left(I - 398.4 \frac{\mu mol}{s \cdot m^2} \right) \right|}$$

Other research by Pandey, Tiwari, Singh, and Tiwari (2011) showed *Spirulina maxima* incubated at 30 ± 2°C.

II. DESIGN, METHODOLOGY AND APPROACH

a) *Spirulina* Feasibility Studies

Spirulina Alga-Gro seawater medium was obtained from Carolina Biological Supply Company. To make 1 L of Alga-Gro, we added 20 mL of concentrate to 980 mL of pasteurized seawater from Daytona Beach Shores. The Nitrogen, Phosphorus and Potassium (NPK) rating of the Alga-Gro is 10 ppm phosphates and 72 ppm nitrates (no potassium concentration is given). The optimum growth for this spirulina is 22°C.

The algae were cultured in media prepared from "pasteurized" seawater heated to about 70°C for 45 minutes, then left undisturbed at ambient temperature for about 1 day before using with the algae concentrate. Two identical 2L tanks (40mL of concentrate and 1960 mL of pasteurized sea water in each tank) were used in the cultivation process (see Figure 1).

Before flight, cultivation (Figure 1) of the *Spirulina platensis* was conducted for about 27 days

with a photoperiod of 12 hours light/dark provided by white fluorescent lamps at a light intensity of 2,500 lux and temperature of 24.0 ± 2.0°C. After 27 days, small samples of spirulina were taken and placed on 1.5 ml Eppendorf conical tubes and studied for feasibility before flight.



Figure 1: Spirulina growth chamber preflight feasibility studies. Day 1. Day 7. Day 10. Day 17. Day 19. Day 21. Day 27. Spirulina samples in tubes at initial state, after 18 hours (showing discoloration) and after 38 hours.

Two main colonies (Figure 1) for about 27 days were used to assess the preliminary survivability tests on the algae before flight. Both tanks have been cultured and were periodically replenished by sea water with added nutrients to allow the algae to grow and flourish.

We hypothesized the algae to survive for at least several days without showing any signs of deterioration. However, most samples showed signs of damage within the first day of the test or after 24 hours. The cause of this deterioration was present since the ratio of algae to water in the test tube was too high (0.5 g of algae and 1.0 ml of water), and thus the oxygen present within the water was used up rapidly, resulting in the early onset of algae damage and loss. The

survivability test was repeated with less algae present in each test tube to gain more accurate results.

Some observations were extracted after the survivability test. Within 24 hours, most of the samples were already showing signs of damage. The expected damage could be observed in the algae slowly turning color from a healthy deep green to a light green and finally to a sickly brown (as seen in some samples after 38 hours in Figure 1). This change in color is to be expected in the algae that is suffering from a lack of light. However, we observed the algae turned from dark green to blue, and the watercolor changed from clear to a cloudy blue.

The blue discoloration is not from the algae, but from cyanobacteria present within the water with the spirulina algae. We know that cyanobacteria thrive within low oxygen environments and when in a low oxygen environment, the cyanobacteria multiply, turning the water within the test tube blue. This is not an ideal situation as cyanobacteria produce toxins which are harmful to algae and other forms of life, including humans. This blue discoloration is a sign that damage is being done to the algae and that consumption of the algae affected by cyanobacteria may be harmful to humans. Cyanobacteria toxins can cause skin irritation, gastrointestinal distress, allergic reactions, and life-threatening liver damage in humans. The emergence of a blue discoloration in the water of a sample will immediately count a sample as dead due to the sample no longer being fit for human consumption.

To counteract this discoloration, a lower ratio of algae to medium was necessary. After lowering the ratio of algae to water, some samples did not show signs of cyanobacteria until 65 hours within the survivability test.

This survivability test unknowingly showed us the results of a lack of oxygen and cyanobacteria toxins

upon spirulina. However, this was not what the survivability test was intended to study. The results of this survivability test ask for another survivability test to evaluate the effects of a lack of light as opposed to a lack of oxygen. The main change in this survivability test was to have reduced mass of algae present within the tubes. To maximize the science outcome, the team decided to have only 0.1 grams of wet mass spirulina to be present in the next survivability test so that the ratio would be 0.1g of algae to 1.0 ml of water. Ideally this change would show that spirulina could survive up to five days with no light while avoiding the dangers of cyanobacteria.

b) *Payload Integration at Payload Processing Facility*

The team integrated the payload in the Payload Processing Facility (PPF) at the West Texas Launch Site (Figure 2a, Figure 2b). Spirulina samples were kept under the sample thermal conditions prior to flight (Figure 2c, Figure 2d) for about 12 hours. Both flight and control NanoLabs are shown in Figure 2e and Figure 2f, respectively.



a.



b.



c.



d.

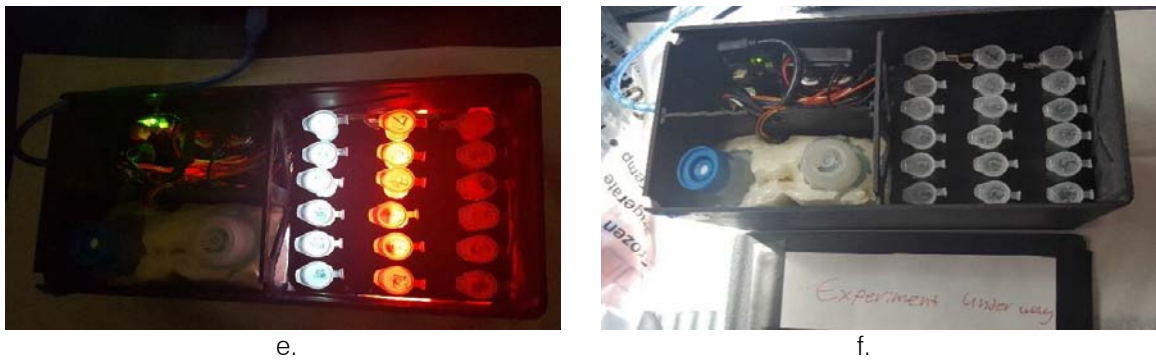


Figure 2: Preparation of samples at the PPF. a. Integration of Payload at Payload Processing Facility (PPF). b. Samples kept in the electric heater at PPF. c. Placing tubes with Spirulina in the NanoLab. d. Two sets of 18 tubes with Spirulina or flight and control. e. NanoLab housing the Spirulina flight samples. f. NanoLab housing the Spirulina control samples.

c) *Spirulina* Postflight Analysis

After the suborbital flight, some samples were transported to the University of Texas Health Science Center at San Antonio (UTHSCSA) facilities for further processing for RNA analysis. The rest of the samples were transported (the same day of the launch) back to ERAU. Figure 3 shows the growth evolution of a flight sample cultivated for 20 days.

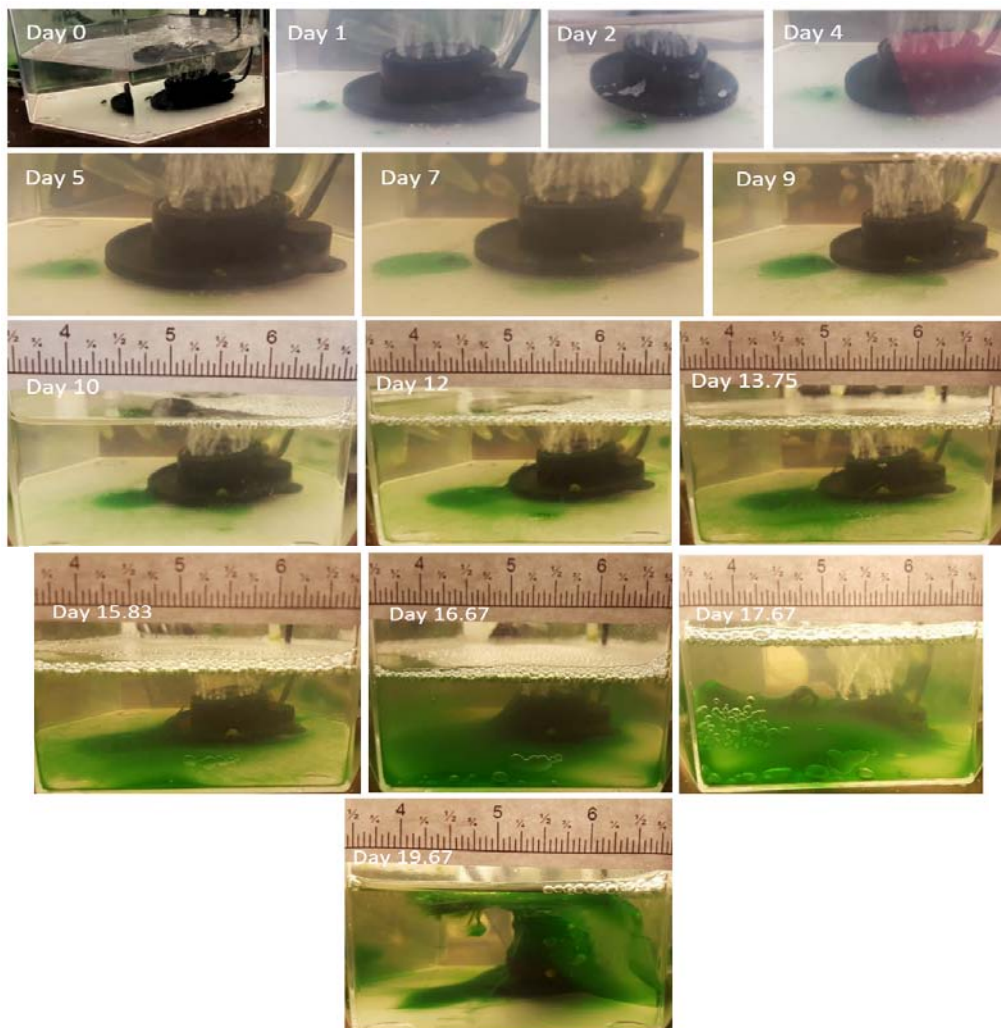


Figure 3: Spirulina growth chamber postflight study showing a flight sample that was cultivated for 20 days.

III. RNA EXTRACTION, CDNA SYNTHESIS AND QRT-PCR

RNAs from spirulina were isolated using Qiagen RNA mini kit (Bio-Rad) according to the manufacturer's standard protocol. RNA concentrations were measured using Nanovue Plus spectrophotometer (GE Healthcare Life Sciences, Pittsburgh, PA). cDNAs were synthesized using iScript™ cDNA Synthesis Kit (Bio-Rad) with 0.1 µg total RNA template loaded for each reaction in all samples. qRT-PCRs were done with SsoAdvanced™ universal SYBR® green supermix (Bio-Rad) following the manufacturer's recommended protocol in a CFX Connect™ real-time PCR detection system (Bio-Rad).

cDNA templates for each sample and reaction were set at 20 µl. Spirulina primers were designed using the Primer 3 Plus Software and listed in Table 1. qRT-PCRs were done with the following parameters: initial denaturation (95°C, 2 min); 40 cycles of denaturation (95°C, 15 s), annealing (55°C, 30 s), and plate reading; melt curve analysis from 65°C-95°C at 0.5°C increment for 5 sec. qRT-PCR data were analyzed using the comparative ($2^{-\Delta\Delta Ct}$) method and values were normalized to housekeeping gene 16S expression. All samples were run at least in duplicate with at least two independent experiments.

Table 1: Gene Specification from Uniport/Org.

<i>Gene Name</i>	<i>Forward Primer (5'->3')</i>	<i>Reverse Primer (5'->3')</i>	<i>Relevance (molecular function; biological process)</i>
<i>CipB</i>	ACGCTGTTAGACAACGCTGA	TACCTCCGAGTCCGCTCACT	ATP binding; protein refolding, response to heat
<i>RcbX</i>	TGCTAAGGACACGACCAAGG	GTTGCGTCATTCGTTCCAGG	Protein folding chaperone; carbon fixation, photosynthesis,
<i>MgtE</i>	ACAGGCGGACGGAGATAACT	ACATCAACGGCGGTGGTAAT	Putative ion magnesium transmembrane transporter; metal ion binding
<i>MpsA</i>	GATCCGACCTAGCCACTTCC	CCAATACGATCGCCTACAGC	ATP binding, metal ion binding, water dikinase activity; gluconeogenesis, Phosphoenolpyruvate synthase
<i>NrtP</i>	AGCCACAGCTATTCAAGACG	CAACCACCATTAACCTCGACC	Nitrate transmembrane transporter, nitrite transmembrane transporter; nitrate assimilation, nitrate import, nitrite transport.
<i>GroES</i>	TTGTCGGTGTGGTGAACAA	GACGGGCGATCGTAAATCCT	ATP binding; protein folding
<i>DnaK</i>	GGGGAACGAGCCATGGTTAA	GTTCTTCAGTCGCTGACCCA	ATP binding, unfolded protein binding; protein folding
<i>Tsf</i>	GCTTTGTTGCCTATCCCT	CACCACGCAGACACTCATA	Translation elongation factor activity; protein biosynthesis
<i>GroEL</i>	TAGCCTTTGACGAAGAATC	ACCATCTCCAGCCACATC	ATP binding; protein refolding
<i>GroEL 1</i>	ATAAGGCGACAGCTTTCTTGTA G	ATTGATTGCTACTTGCCCACTC TT	ATP binding, unfolded protein binding; protein refolding
<i>HspA</i>	GGCAATCATTAGTTTAAATTGGGA A	AGCCAGATCGACCTTAACCAC	Small heat shock protein molecular chaperone; stress response
<i>NrtD</i>	CTGTAACCAACTCCACCTCC	CGTAGTATTCTGGCTGTCC	ATPase-coupled transmembrane transporter activity, ATP

			binding, Nitrate ABC transporter ATPase subunits C and D
<i>FoIE</i>	TTCTGACCAACGGCTACAGT	GAAGACACCAATCATGGCAC	Putative GTP cyclohydrolase I binding, GTP cyclohydrolase I activity, zinc ion binding; once-carbon metabolic process, tetrahydrofolate biosynthetic process
<i>16S</i>	TAGCGGTGAAATGCGTAGAT	AAGTGCCCAACTGAATGATG	Gene housekeeping

IV. CONFOCAL MICROSCOPY

To better visualize the structure of flown and ground control spirulina samples, we placed them in petri dishes and exposed them to confocal microscopy using Olympus FV3000 Confocal Laser Scanning Microscope. We imaged spirulina samples at different excitation wavelengths ranging from 488 (green/Fluo-3), 561 (red/tdTomato), and 650 (blue/FuraRed) using 2X and 10X resolution.

V. RESULTS

a) Suborbital Flight Characteristics

The suborbital flight characteristics are displayed in Figure 4. The maximum altitude during the suborbital flight was 105.68 km. Figure 4a shows the acceleration map generated with the data from our NanoLab (maximum acceleration is 2.16 g) and Figure 4b displays the acceleration map obtained with the data from a NanoRacks sensor (maximum acceleration sensed is 7.06 g during parachute deployment and 4.30 g during reentry). Although our sensor maximum acceleration seems lower than the value for one of the NanoRacks sensors, it is consistent with other NanoRacks sensors measurements (Figure 4c, 4d). This lower measured acceleration may have been caused by a power interruption which affected the data collection. Figure 4c and Figure 4d are the acceleration profiles in function of time for both the NanoLab sensor and one of the NanoRacks sensors, both showing the difference of phase events during the suborbital flight. Figure 4e depicts the descent, parachute deployment and landing phases of the flight, and Figure 4f depicts a zoom section of the parachute phase. Previous research (Llanos et al., 2019) showed the acceleration profile for another suborbital experiment which was used to leverage the efforts of this research.

Figure 4g and Figure 4h display the acceleration profiles for other two NanoRacks sensors with maximum accelerations of 2.13 g and 2.68 g, respectively. Another two NanoRacks sensors measured maximum accelerations of 2.48g and 2.49g. Note that

the acceleration measured by our NanoLab sensor falls in between the acceleration measured by all four NanoRacks sensors. As we can observe, different sensors provided slightly different acceleration profiles since the sensors were placed in various locations within the Nano Feather Frame (NFF) where all the NanoLabs are integrated.

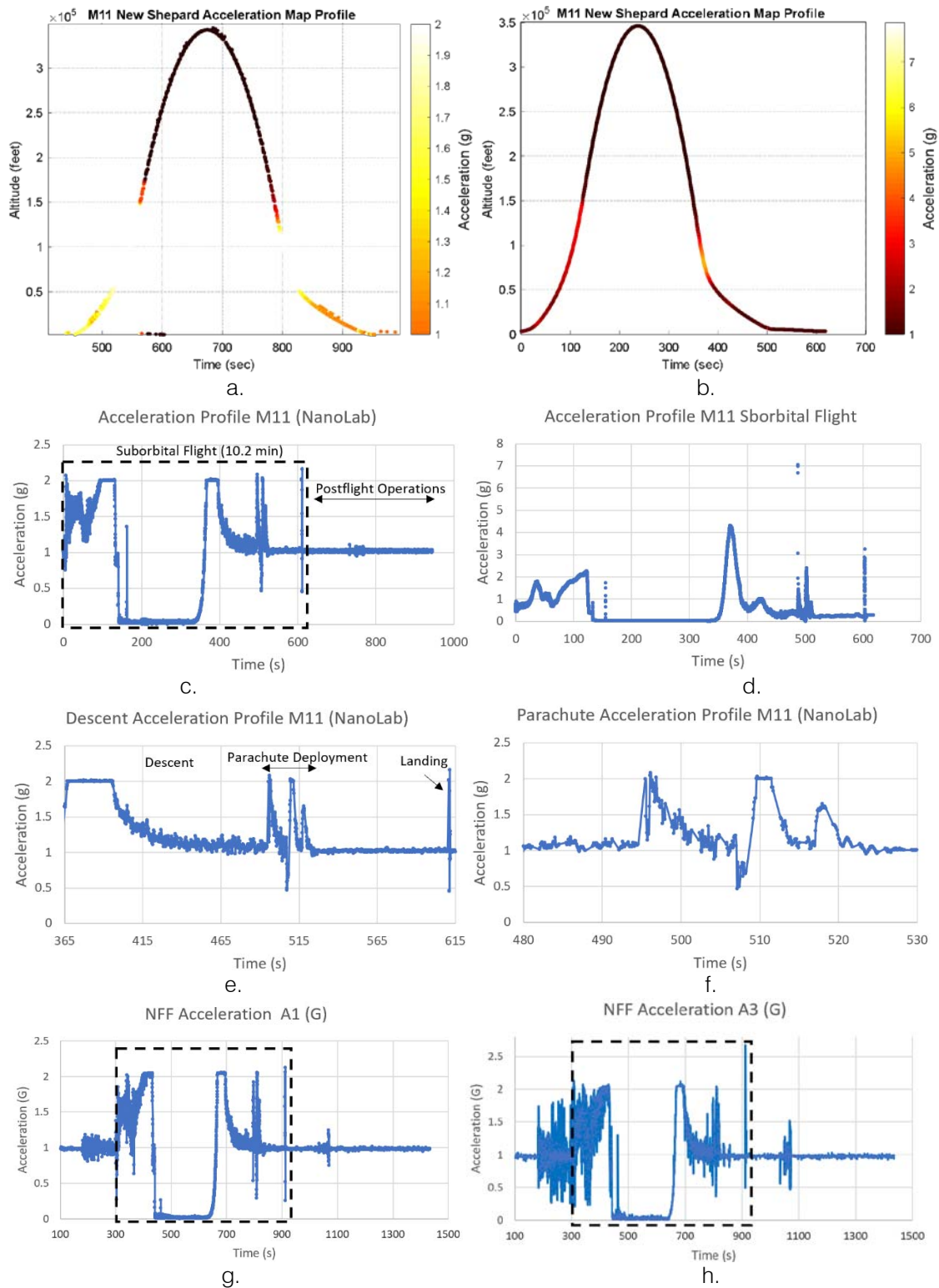


Figure 4: Acceleration profiles during various mission phases. a. Acceleration map profile measured with NanoLab sensor. b. Acceleration map profile measured with NanoRacks sensor. c. Acceleration measured by NanoLab during preflight, suborbital flight and postflight mission phases. d. Acceleration during descent. e. Acceleration during parachute deployment. f. Acceleration during landing. g. Acceleration measured inside the Nano Feather Frame by NanoRacks. h. Acceleration measured by another sensor inside the Nano Feather Frame by NanoRacks.

Our Nanolab sensor measured a microgravity between $10^{-4}g$ and $10^{-1}g$. It was measured that 3.6% of time of about 3.4 minutes with microgravity quality of the time the NanoLab measured between microgravity

levels between $10^{-4}g$ and $10^{-2}g$, 16.5% of the time between $10^{-2}g$ and $2 \cdot 10^{-2}g$, 44.5% of the time between $2 \cdot 10^{-2}g$ and $3 \cdot 10^{-2}g$, 16.8% of the time between $3 \cdot 10^{-2}g$ and $4 \cdot 10^{-2}g$, 12.5% of the time between $4 \cdot 10^{-2}g$ and $5 \cdot 10^{-2}g$ and 6.1% of the time between $5 \cdot 10^{-2}g$ and $10^{-1}g$. The NanoRacks sensor measured microgravity time of about 3.5 minutes with about 4.1% of time between $10^{-4}g$ and $10^{-2}g$, 79.7% of the time between $10^{-2}g$ and $2 \cdot 10^{-2}g$, 12.6% of the time between $2 \cdot 10^{-2}g$ and $3 \cdot 10^{-2}g$, 2.0% of the time between $3 \cdot 10^{-2}g$ and $4 \cdot 10^{-2}g$, and 0.3% of the time between $4 \cdot 10^{-2}g$ and $5 \cdot 10^{-2}g$, and 1.3% of the time between $5 \cdot 10^{-2}g$ and $10^{-1}g$.

b) *Spirulina Mass Growth Evolution*

Using equations for the growth model by Wang, Fu, and Liu (2007), we present an approximate estimated growth model nine hours before launch and until payload recovery a few hours after landing. The average light intensity for both red and white LEDs were 1100 lux and 875 lux, respectively, approximately the same as for the light intensities measured during ground control (Figure 5a).

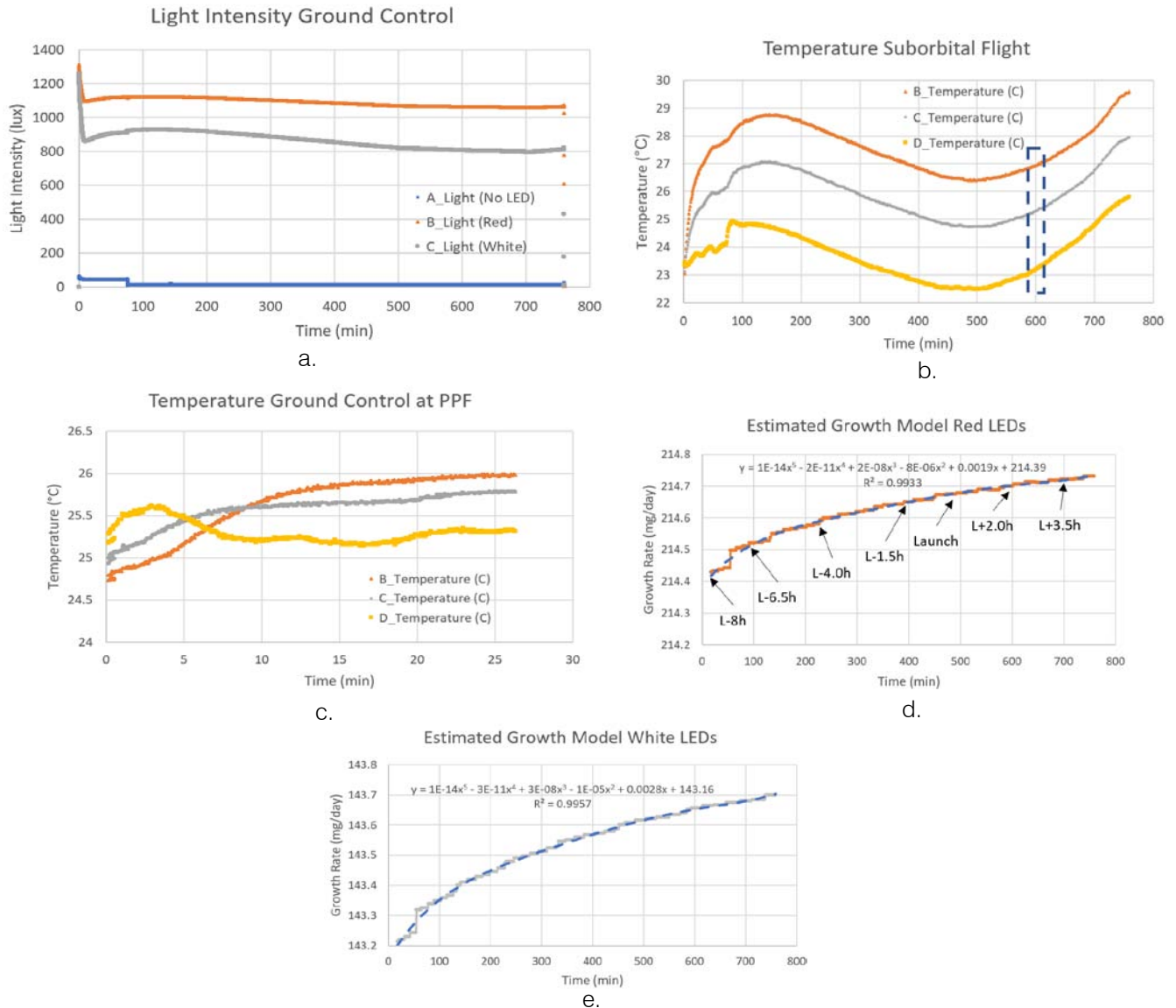


Figure 5: a. Light intensity (a) and temperature variations for both NanoLabs. b. Temperature profile during suborbital flight including preflight and postflight operations. c. Temperature profile test at PPF. d. Growth rate for red LEDs. e. Growth rate for white LEDs.

Figure 5b shows the temperature profile for the flight samples during preflight operations, flight (see blue dashed box) and post flight operations. Note that the different temperature profiles correspond to the temperature measured in each compartment for

different light conditions (B_Temperature for white LED, C_Temperature for red LED, D_Temperature for no LED light). During this period, the growth rate of the algae for the red LEDs ranged from about 214.4 mg/day to about 214.7 mg/day with about 0.3 mg in about 12 hours (see

Figure 5d), and for the white LEDs, the samples growth was between 143.2 mg/day and 143.7 mg/day with about 0.5 mg in about 12 hours (see Figure 5e). Similarly, the algae exposed to the white LEDs grew about 0.5 mg in 12 hours. The algae kept in the NanoLab at the PPF was exposed to the thermal profile (see Figure 5c).

From the samples (control and flight) brought back to ERAU we took nine control samples and nine flight samples that were kept in individual 50 ml tubes

with 5ml sea water and about 0.15 g of algae. All these samples were kept for seven weeks to analyze their growth evolution. Samples were replenished with media and proper nutrients from their respective mother colony. Three samples were taken for each light intensity condition (C1, C2, C3: no light), (C7, C8, C9: red) and (C13, C14, C15: white). Similarly, for flight samples we had (F4, F5, F6: no light), (F10, F11, F12: red) and (F16, F17, F18: white).

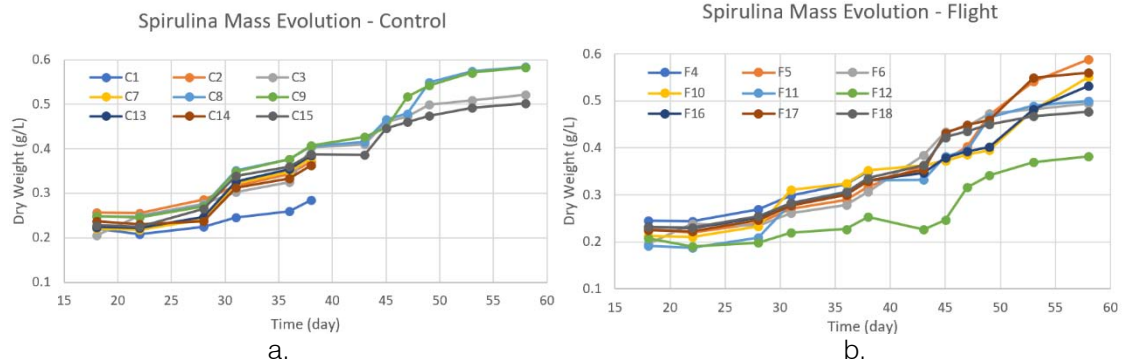


Figure 6: Time behavior of spirulina growth post flight for both control and flight samples under the same lab conditions. a. control samples. b. flight samples.

The estimated spirulina dry weight for all the samples (control and flight) are provided in Figure 6. Under the conditions examined, the ones that yielded more biomass production across all control samples were 0.584 g/L and 0.583 g/L corresponding to the controls C8 and C9, respectively, both under red light. The next conditions which yielded more biomass production were the control samples C3 (under no light) and C15 (under white light) with 0.521 g/L and 0.502 g/L, respectively. Summing all the biomass production for all control samples, we obtained an average of 1.546 g/L (red light), 1.250 g/L (white light), and 1.176 g/L (no light) during the seven weeks span. The rest of the control samples generated dry weights of about 0.28 g/L to 0.38 g/L. As for the flight samples, they all survived yielding similar dry weights at the end of the seven weeks. The total dry weight for samples under red light, white light, and no light were 1.432 g/L, 1.570 g/L and 1.432 g/L, respectively. The total dry weight from for the tank with control samples and flight samples were 3.971 g/L and 4.435 g/L, respectively.

An important observation from Figure 6 is that the growth pattern slowed down at around 20-25 days after the start of the growth (17 days after suborbital flight) showing a dip of lowest growth that lasted for a few days. This low dip in growth mass has previously been seen with the dry weight or biomass concentration found by previous research groups (Delrue et al., 2017 and Kumari et al., 2015), yet these groups do not show further insight into the growth of the spirulina after 20 or 27 days.

Phosphate is the major nutrient for the growth of microalgae to produce nucleic acids and phospholipids (Kumari et al., 2015), and the uptake of this micronutrient is greater when exposed to red LED light than to white LED light even using several mediums for about 20 days.

c) *Spirulina Ph, Ammonia (NH₃/NH₄⁺), Nitrate (NO₂⁻), Nitrite (NO₃⁻) Assessment*

We next assessed the pH, ammonia (NH₃/NH₄⁺), nitrate (NO₂⁻), nitrite (NO₃⁻) in flown spirulina samples as well as in ground controls (Table 2).

Our results show that the highest pH of 8.6 was observed in control samples exposed to red light, whereas the pH for flown samples under the red light was 7.8. In general, most control samples show pH values between acceptable and ideal values while the flight samples show pH values between acceptable to low values. This behavior was expected since control samples remained at almost constant temperatures of 25-26 °C (Figure 5c) inside the PPF, and they were not exposed to any vibrations or high-g accelerations during the suborbital flight like the flight samples. The flight samples were exposed to more temperature fluctuations as seen in Figure 5b from the time of delivery of the payload to the NanoRacks team to integration of the payload inside the NFF inside the New Shepard capsule. Most control samples showed signs of some level of stress for ammonia and ammonium while all the flight samples showed higher levels of stress. In terms of nitrite contents, both control and flight samples did not

show any stress levels, and as for the nitrate contents, most control and flight samples showed ideal to good conditions.

Table 2: Spirulina Ph, Ammonia (NH₃/NH₄⁺), Nitrate (NO₂⁻), Nitrite (NO₃⁻) Test.

Sample	pH	NH ₃ /NH ₄ ⁺ (ammonia/ammonium)	NO ₂ ⁻ (Nitrite)	NO ₃ ⁻ (Nitrate)
Control Samples				
C1 (no light)	7.8 (acceptable)	3.0 (danger-)	0	40 (ok-)
C2	7.4 (low)	1.0 (stress)	0	20 (ok+)
C3	7.8 (acceptable)	2.0 (stress+)	0	20 (ok+)
C7 (red light)	7.8 (acceptable)	1.0 (stress)	0	0 (ideal)
C8	8.4 (ideal)	1.0 (stress)	0	0 (ideal)
C9	8.6 (ideal)	0 (ideal)	0.5 (caution)	20 (ok+)
C13 (white light)	7.8 (acceptable)	0.5 (ideal)	0	20 (ok+)
C14	7.4 (low)	6.0 (danger+)	0	0 (ideal)
C15	7.8 (acceptable)	3.0 (danger-)	0	0 (ideal)
Flight Samples				
F4 (no light)	7.8 (acceptable)	3.0 (danger-)	0	20 (ok+)
F5	7.4 (low)	3.0 (danger-)	0	20 (ok+)
F6	7.4 (low)	3.0 (danger-)	0	0 (ideal)
F10 (red light)	7.0 (too low)	3.0 (danger-)	0	0 (ideal)
F11	7.8 (acceptable)	3.0 (danger-)	0	20 (ok+)
F12	7.4 (low)	3.0 (danger-)	0	20 (ok+)
F16 (white light)	7.8 (acceptable)	3.0 (danger-)	0	20 (ok+)
F17	7.4 (low)	6.0 (danger+)	0	0 (ideal)
F18	7.6 (acceptable-)	3.0 (danger-)	0	20 (ok+)

d) Gene Expression Analysis

In our study, we selected genes that have been previously studied by Panyakampol et al. (2015). This selection was generated based on the thermal effects on some of these genes regarding the cellular processes, inorganic ion transporters and carbon and nitrogen metabolism mechanisms.

Our data indicate that spirulina samples exposed to suborbital flight had downregulated expression of all genes as compared to ground controls regardless of the light exposure type (Figure 7a, b, c).

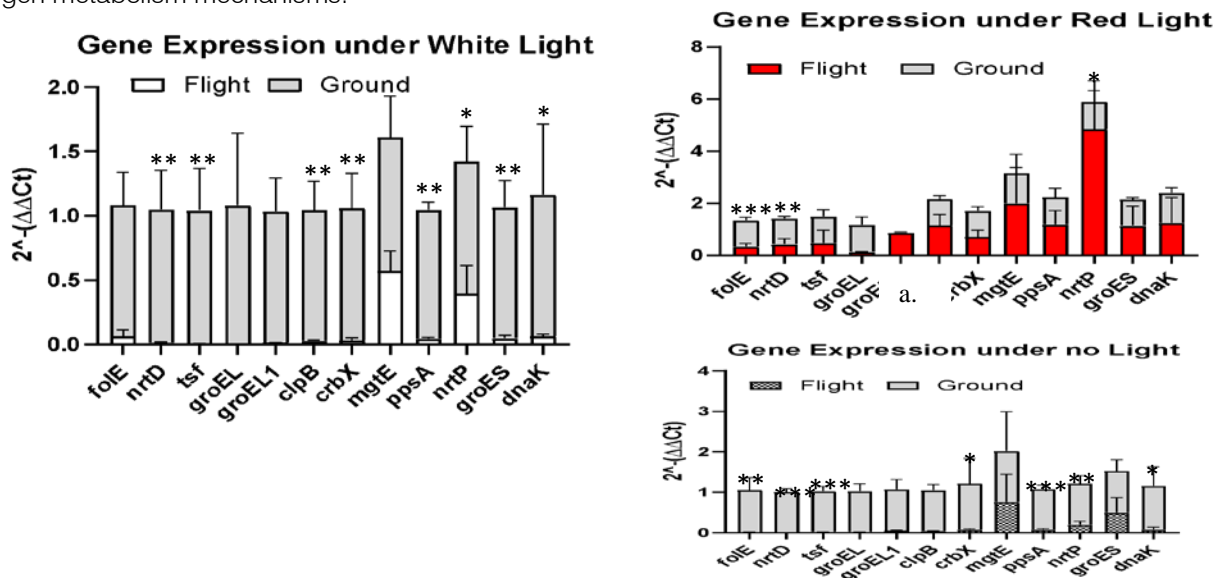


Figure 7: Gene change expression (2⁻(ΔΔCt)) among different flight and ground conditions (a, b, c). Values indicate mean ± SD. * p ≤ 0.05; ** p ≤ 0.01; *** p ≤ 0.0001.

However, certain genes were elevated more than others. We saw that exposure to light led to the highest expression of *mgtE*, *nrtP*, and *dnaK* genes across all conditions. The most profound effect of white light had on the *ppsA* gene ($p < 0.001$) (Figure 7), while exposure to red and no light conditions elevated the

expression of *folE* (Figure 7b) and *nrtD*, *tsf*, *ppsA* genes (Figure 7c). In addition to comparing flight and ground control samples, we also sought to investigate which light condition had the most profound effect on flown spirulina samples (Figure 8).

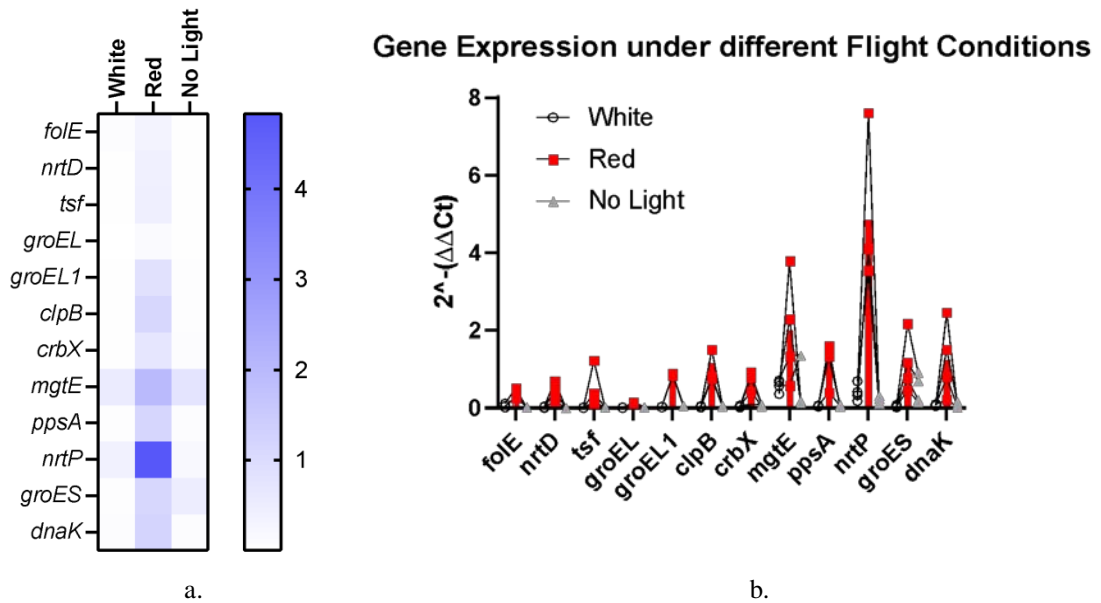
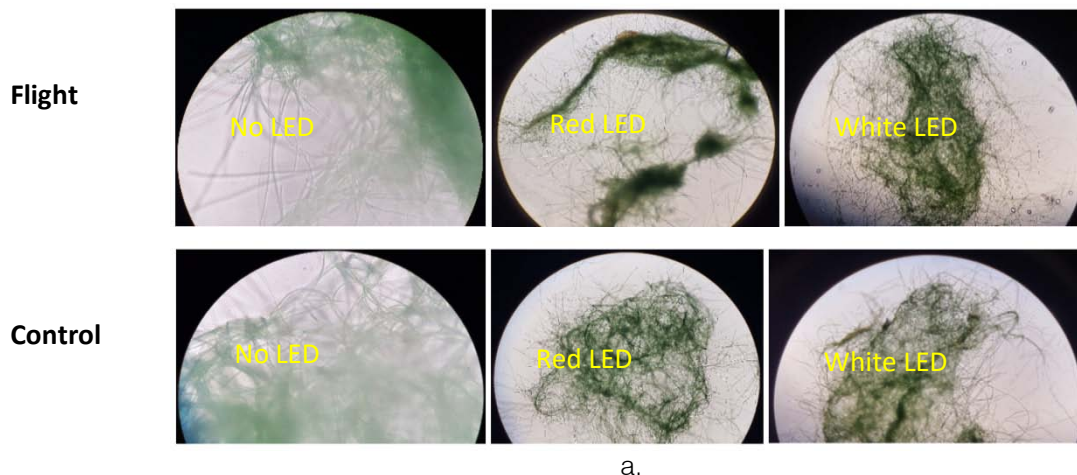


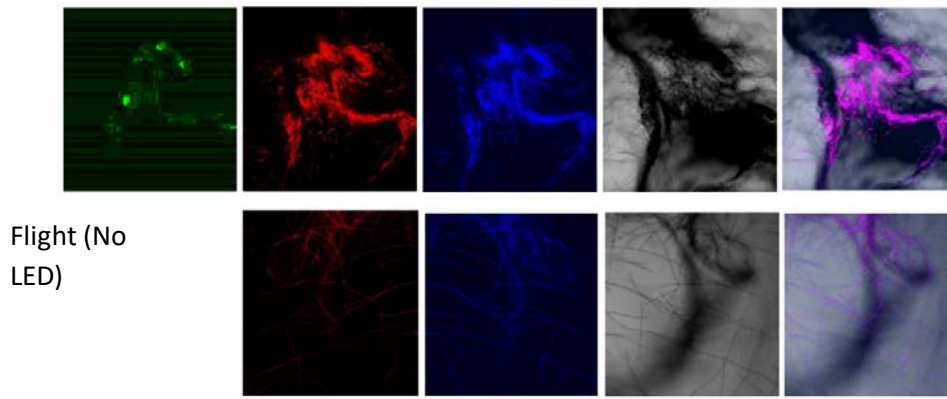
Figure 8: Heat map of genes altered by different light conditions in flown spirulina samples (a). Gene change expression ($2^{-\Delta\Delta Ct}$) among different flight conditions. Values indicate mean \pm SD (b).

Based on the heat map generated from the quantitative RT-PCR analysis (Figure 8a), red light led to the highest expression of all genes, with the highest expression of the *nrtP* and *mgtE* genes. White light led to the highest expression of the *mgtE* and *nrtP* genes as well, while no light condition elevated *mgtE* and *groES* genes. Similar patterns were observed in the bar graph where each dot represents an individual experiment (Figure 8b). This graph shows individual samples distribution across different conditions and indicates reasonable variability among different conditions.

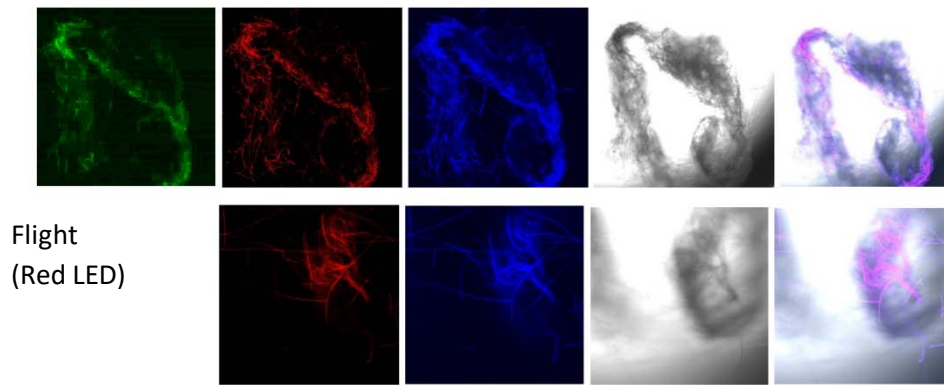
VI. CONFOCAL MICROSCOPY

We next assessed the structure of ground and flown spirulina samples using inverted and confocal microscopy (Figure 9b-Figure 9f) where the top row represents a 1X resolution while the bottom row represents a 2X resolution or 10X resolution for Figure 9g. Figure 9a depicts the pictures of the selected samples under the Motic AEZI Inverted Phase Microscope.

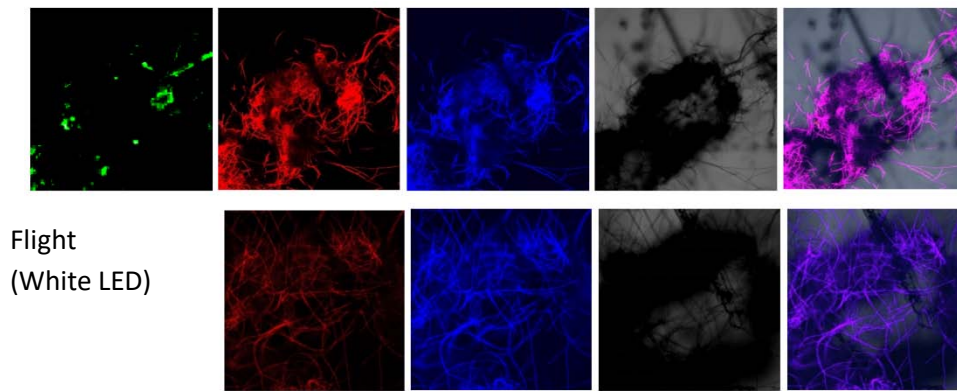




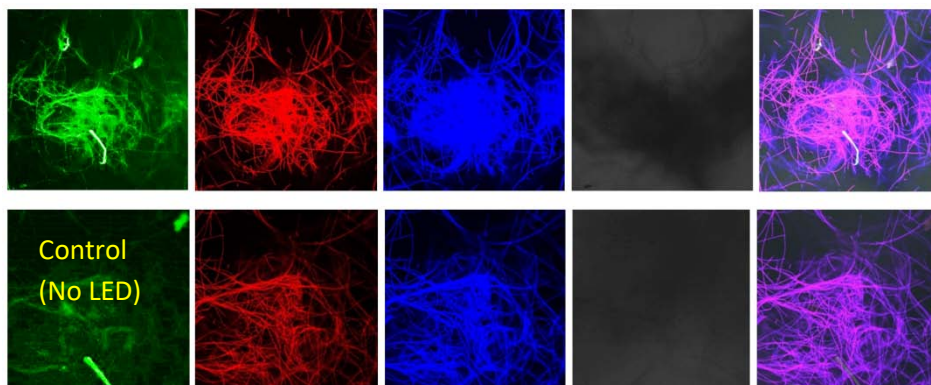
b.



c.



d.



e.



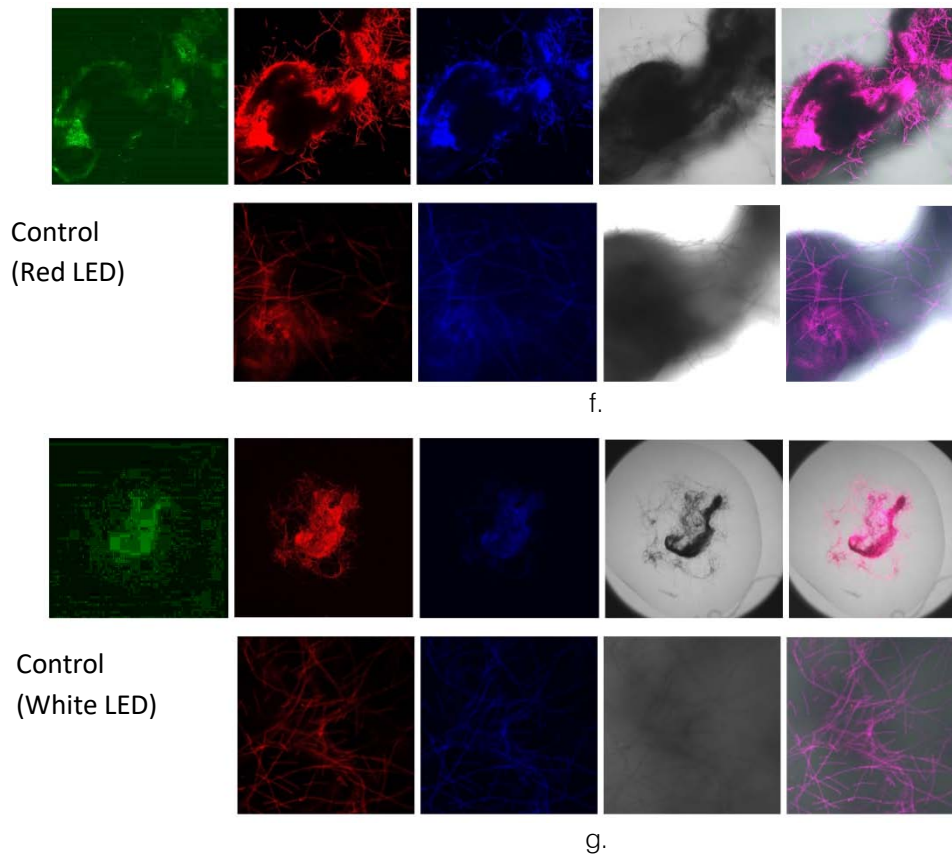


Figure 9: Confocal microscopy for control and flight samples. a. Flight samples (top) and control samples (bottom) as seen under the microscope. b. Flight-6 (no LED). b. Flight-10 (red LED). c. Flight-18 (White LED).d. Control-2 (No LED). e. Control-8 (red LED). f. Control-15 (white LED).

Based on images taken under inverted microscope (Figure 9a), control samples had a more defined structure when compared to flown samples. In flown samples (Figure 9b, c, d), no light and white light led to the highest laser intensity and better-defined structures when compared to the red light. For control samples (Figure 9e, f, g), no light led to a more defined spirulina structure when compared with samples exposed to white and red light.

VII. CONCLUSIONS

Given the significant interest in future human space flights, finding an alternative food source is an important subject of current and future space research. In this study, we sought to determine whether exposing spirulina (*Arthrospira platensis*) to a different light condition during a suborbital flight could influence the expression of genes involved in many important cellular processes, such as ATP regulation, photosynthesis, nitrate transport, etc.

Spirulina flight samples yielded more mass than the control samples as an overall. In particular, the flight samples exposed to red light yielded about 7% less mass than the controlled samples exposed to the same light conditions at the PPF; but both the flight samples

exposed to white light and no light yielded about 25% and 21% more mass than their controlled samples at the PPF. The low biomass for red LED light may be associated with the uptake rate of micronutrients, including phosphorus, which takes place at a higher rate for red light than for white light.

Our results suggest that red light led to the most profound weight mass reduction in flown samples when compared to other light conditions. Thus, it downregulated the expression of many genes suggesting that exposure to red light might not be beneficial.

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Declaration of Conflicting Interests

The authors declared no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

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Artificial Vision Prototype for People and Vehicle Characteristics Recognition

By Fanny Suárez Mosquera, Cristian Dangelis Ballén Martínez
& Gerardo Alberto Castang Montiel

Universidad Distrital Francisco José de Caldas

Abstract- In a world increasingly connected with technology and with the growing need for security in all aspects of life, and the need of security in the transport of child in educational establishments is a great challenge. In other areas, the need of security in areas such as hospitals, parking lots, securities transporters, airports, etc. This solution aims to recognize patterns (vehicle plates) and characteristics (facial patterns); one of the main premises of the prototype was the use of free software to create a successful, low cost and easy-to-use solution.

Keywords: *telematicprototype, patterns recognition, arduino, artificial vision, free software.*

GJRE-B Classification: *DDC Code: 387.7 LCC Code: HE9765*



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Artificial Vision Prototype for People and Vehicle Characteristics Recognition

Fanny Suárez Mosquera ^α, Cristian Dangelis Ballén Martínez ^σ & Gerardo Alberto Castang Montiel ^ρ

Abstract- In a world increasingly connected with technology and with the growing need for security in all aspects of life, and the need of security in the transport of child in educational establishments is a great challenge. In other areas, the need of security in areas such as hospitals, parking lots, securities transporters, airports, etc. This solution aims to recognize patterns (vehicle plates) and characteristics (facial patterns); one of the main premises of the prototype was the use of free software to create a successful, low cost and easy-to-use solution.

Keywords: *telematic prototype, patterns recognition, arduino, artificial vision, free software.*

I. INTRODUCTION

The prototype was create to provide an accurate and easy solution in security. With this prototype, different schools can know about different characteristics of drivers and vehicles that provide school route service, providing a validation tool that can be integrate with the mobility registration system. A recognition of facial patterns and features can be made all of this supported by *free software recognition libraries*, through 128-point feature vectors which makes face representation for comparison. Allowing it to be easy to use and a low cost solution it will be apply in a number of scenarios making it scalable. Enhancing the centralization of data by using a distributed database generating availability and a higher security level.

II. ANALYSIS STAGE

An analysis of the chosen hardware and software architecture is carry out, and the scope and limitations of this are define for subsequent implementation. The main objective is design an artificial vision prototype for facial recognition of drivers and school route plates for entry and/or exit from the institution.

III. DESIGN STAGE

The recognition of the involved agents with the system is developed and the interaction between these with the prototype. Besides relating the model of

hardware and software of the system, it is divide in the Software component and Hardware component.

IV. HARDWARE MODEL

This item is scalable and configurable since inside this prototype is necessary to build a hardware component, which would allow the driver to move into the institution when start the validation circuit of the prototype (IP camera and the rest of the services of facial recognition and registration). This component consist of a circuit with an Arduino card given the capacity of interaction that it can have with the system. It could be easily adapted to systems of sensors, automatic doors, registers and other access devices.

V. SOFTWARE MODEL

The software make the *interaction* of the system with the final user. The system has components of graphic interface where the administration and the configuration modules will be visualize besides seeing the alerts with each one of the different entries that occur. The development with *free and open source technologies* such as *Python, Java, JSF, Java programming languages* were use with free distribution tools for its development such as *NetBeans, notepad ++, VisualStudioCode, Mysql database engine and Maria DB.*

VI. DATABASE MODEL

The database modeling is an important point in the software development since it is from this design that *the system's planning and interaction* begins in addition to managing the distributed database to maintain the reliability and availability standards required for proper operation. The MARIA DB database engine was use with a GPL license in which the configuration is carry out in order to establish the distributed database.

VII. SYSYEM AGENTS

User interactions were define for this system and the specifications may change according to the environment to be use with the Hardware and Software solution.

Author ^{α σ ρ}: *Universidad Distrital Francisco José de Caldas, Facultad Tecnológica, Bogotá D.C. Colombia.*

e-mails: *fsuarezm@correo.udistrital.edu.co,*

cdballenm@correo.udistrital.edu.co, gacastangm@udistrital.edu.co

Table 1: Definition of System Agents.

Agent	Description
Admin	Responsible for the administration of alerts, personnel and vehicles authorized to enter to the institution.
Watchman	Agent who receives notifications of unauthorized entry into the institution, person responsible for granting or refusing entry into and/or leaving the institution
Driver	Agent that interacts with the system through the Arduino board.

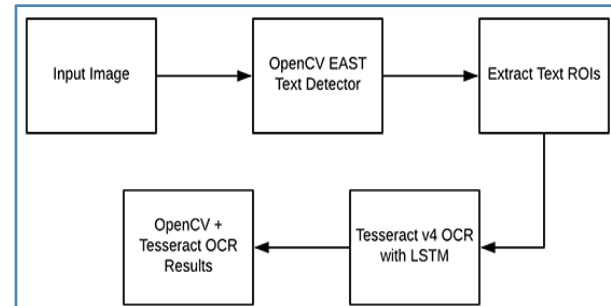
Source: authors

VIII. ARCHITECTURE

It is indispensable to think about a good architecture because it defines a path to begin to build the system. The way in which the components will interact with other systems or libraries and thus achieve the main objective to have a functional prototype ready for implementation. The used libraries and protocols are OpenALPR, OpenCV and RSTP. *OpenALPR* is an open source library used in automatic license plate pattern recognition written in C++ with links in C#, Java, Node.js, Go and Python. The library analyzes images and video sequences to identify license plate patterns. *OpenCV* is a free machine vision library originally developed by Intel. It contains more than 500 functions that cover a wide range of areas in the vision process such as facial recognition, camera calibration, stereo vision and robotic vision. *RSTP* (Real Time Streaming Protocol) is a non-connection-oriented protocol. The server maintains a session associated with an identifier in most cases RTSP uses TCP for player control data and UDP for audio and video data.

IX. LICENSE PLATE RECOGNITION WITH OPENALPR

We use OpenALPR library, which is an open source library that helps the automatic recognition of license plates. It operates for the video stream as follows: The image flow will be constantly extract from the IP camera in MJPEG format through RTSP on a video format, and OpenALPR will start the validation. The agent will start the process automatically and the library processes the flow as fast as possible while searching for plate images. When the plates are detect, the information will be write to a local beanstalk queue as JSON data. The library save the image in a configurable location as a jpeg image and run a separate process that empty the beanstalk queue upload the data to the server via HTTP. This library works with OpenCV for character recognition.

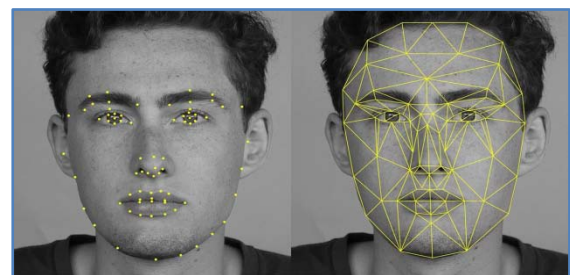


source: <https://www.pyimagesearch.com/2018/09/17/>

Figure 1: Pipeline for Opencv

X. FACE RECOGNITION

This library built in python from the *Dlib toolkit* made in C++ contains deep learning algorithms, which allows to model abstractions providing accurate results reaching good accuracy model. There are different methods for face comparison such as the waterfall, which analyzes hundreds of small patterns and characteristics that must match, similar to detecting a fingerprint but this method is not suitable due to its impossibility of detecting profiled faces or low occlusion. Others based on deep metric learning rely on 128-point characteristic vectors making a representation of the face are used in this prototype. The algorithms can be accurate with the right distance threshold between points.

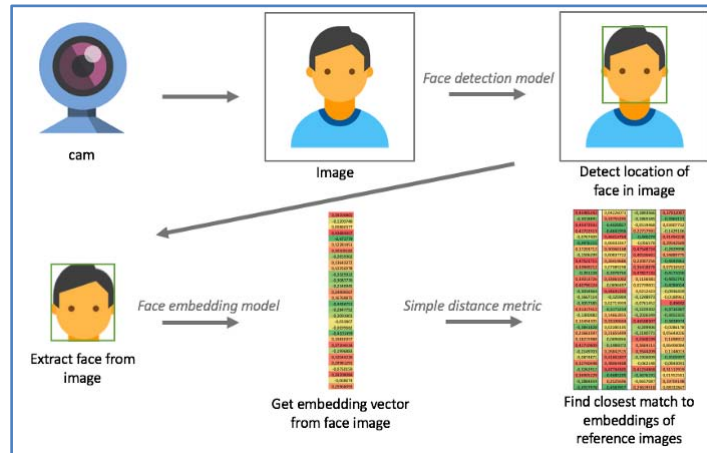


Source: <https://encryptedtbn0.gstatic.com/images?q=tbn:ANd9GcS0M>

Figure 2: Dotted vector

The following steps identify the one face: The face detection model identifies the face's location inside the image. The embedding fed model is used to obtain a

vector of facial characteristics of size 128 points. It compares this vector with those of its "friends" and finds the most similar.

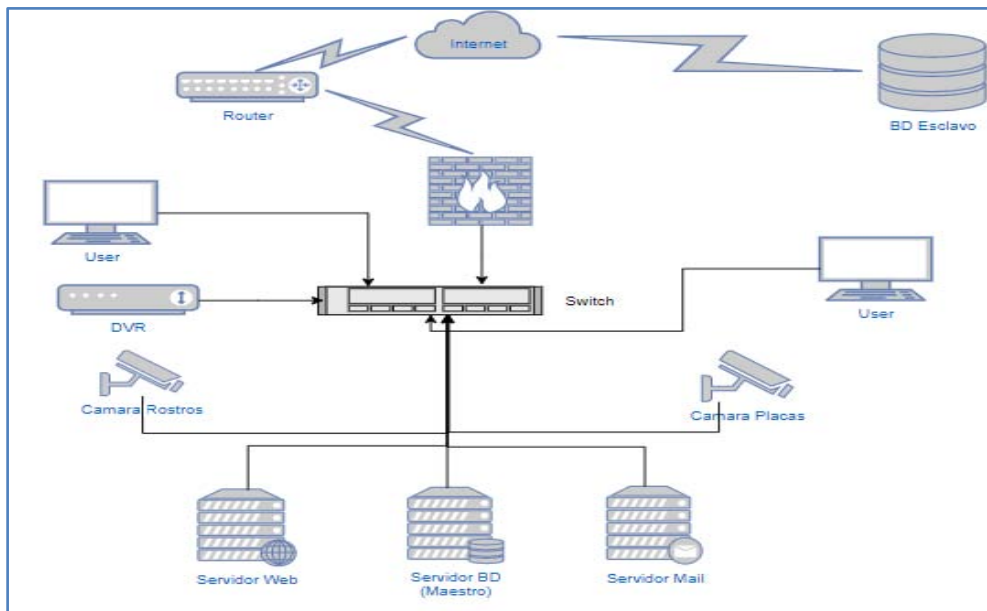


Source: https://miro.medium.com/max/700/1*R-ObQiGjDK4Njd5tSQEz5g.png

Figure 3: Recognition cycle

XI. IMPLEMENTATION STAGE

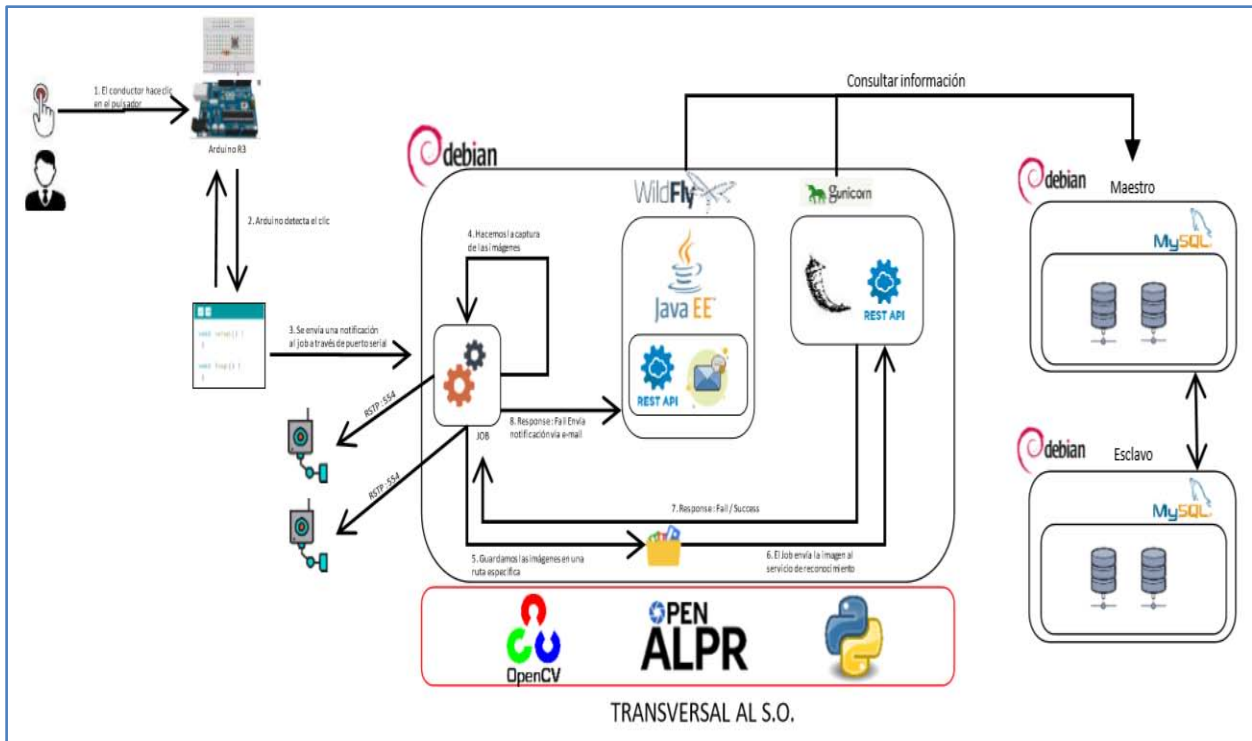
The infrastructure defined for the system operation as shown in the below figure. Figure 4. Implementation of the prototype



Source: Authors

The prototype has components of hardware y software that interoperate in this form: *Arduino Wiring*: This code is stored in Arduino's microcontroller to start the request and begin the whole process. It can be replaced by the access component that the application requires. *Python Code*: It is the backend of the application where the processing of the images and comparison of these are located. *Java Code*: It is the frontend of the system since it provides a friendly interface to the Administrator and Watchman actors that

make up the system. To have a better view of the system operation the following diagram has been validate.



Source: Authors

Figure 5: Prototype Overview

The diagram above describes the technologies used and the flow of information. An Arduino card and a push button is use to start the flow, which could be replaced by an access bar or a movement sensor. The Arduino board algorithm processes the message sent through the serial port and sends an alert to the JOB (Automatic Task). It takes a picture of the face and the license plate of the vehicle and stores it in a folder. The JOB (Scheduled Task) sends the 64-based images to the recognition service for analysis. The service generates a response indicating whether the recognition was successful or not to the JOB. If everything was successful the corresponding access is given, otherwise it sends an email to a previously configured account indicating that there was an unauthorized access.

XII. HARDWARE AND SOFTWARE INTEGRATION

We make the two components interact with each other and give functionality to the final product and the following elements were define for its operation: The Hardware model, Job and Web Services and Web Application (JEE).

XIII. TEST STAGE

The accuracy of a project is define by the test cycle aim to identify achievements and limitations obtained in the development of the prototype. For this is need a vehicle and a person who will carry out the entry simulation to the institution. When analyzing the image the results are:

```
plate0: 10 results
- UGU286 confidence: 94.1289
- UGU286 confidence: 85.1463
- U6U286 confidence: 84.3539
- UGUZ86 confidence: 83.4433
- UG0286 confidence: 81.8547
- UGD286 confidence: 81.659
- 0GU286 confidence: 81.6427
- DGU286 confidence: 81.6177
- 0GU286 confidence: 81.5763
- UG0286 confidence: 81.4162
```

Source: Authors

Figure 6: Vehicle Plate Identification

A list of ten possible plates with percentage of accuracy are see. We can obtain from 10 to 50 results. The percentage indicates which characters have more similarity to the image of the vehicle plate so getting the right plate. Regarding facial recognition the first thing to have when starting are images with people faces that are intend to be identify and getting two profiles of the person and a front photo in order to ensure greater accuracy in recognition. With this, the algorithm is able to recognize whether the face to be analyze is the desired person. Some guidelines that must be take into account for proper operation such as the light distance, camera resolution and the number of faces that can appear in the image. The algorithm makes the comparison between previously saved images and the take image for the identification. The validation algorithm returns a vector with the analysis response. If true value is return, means that it recognized a match with one of the base images.


```

import face_recognition

picture_of_me = face_recognition.load_image_file("me.jpg")
my_face_encoding = face_recognition.face_encodings(picture_of_me)[0]

unknown_picture = face_recognition.load_image_file("unknown.jpg")
unknown_face_encoding = face_recognition.face_encodings(unknown_picture)[0]

results = face_recognition.compare_faces([my_face_encoding], unknown_face_encoding)

if results[0] == True:
    print("It's a picture of me!")
else:
    print("It's not a picture of me!")

```

Source: <https://github.com/openalpr/openalpr>

Figure 7: Face Recognition Algorithm

XIV. RESULTS STAGE

The prototype's confidence levels may change with conditions of distance, comparative and luminosity. Recognition levels are high when the prototype has the right photo profiles for optimal comparison. In the plate case found that the prototype could be highly adaptive. The OCR not only recognizes the model of the plates that are hand as standard but also recognizes those parameterized in the system.

XV. CURRENT WORKS

To have a more complete overview of the potential of the Open ALPR tool, which uses OpenCv, it is research and is evident that these libraries have a wide use from implementation of face recognition in major airports, and too be a solution for optimal fruit sorting with high quality standards.

XVI. FUTURE WORKS

This application is highly scalable and modular to be use in other works where it can be integrate in a simple way. The version implements pattern recognition and we can will detect plates from various countries, color, vehicle brand and model; and the library for face recognition can be integrate easily.

XVII. CONCLUSIONS

The face recognition effectiveness ranges between 96% and 99% depending on the image type to be analyzed having a higher effectiveness if it has the three angles of the face which are the 128 point feature vectors for OpenCV works and which it makes the comparison.

The Optical Character Recognition (OCR) component has an effectiveness of approximately 94% that depends on factors such as brightness, image capture angle, camera resolution and that the plate does not have any kind of obstruction because there may be a poor character reading.

These two great components integrated to the software component allow a powerful tool with a high rate of effectiveness where it can be evidenced that with the available tools in its free versions can provide very useful solutions in all common life areas.

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Brazil Motorcycles Categories and Hybrid Electric Technology Comparison with Powertrain Sizing

By Marcelo Fernandes De Almeida

Rio de Janeiro State University

Abstract- The world is changing and the vehicle technology is changing together to adapt to new customers behaviors, one new customer behavior is to use the electrical powertrain to traction the vehicles and decrease the transport pollution. The electrical powertrain could be detached in two groups: battery electrical vehicles (BEV), its use only an electrical engine on the vehicle, and hybrid electrical vehicle (HEV), its use the electrical engine and the internal combustion engine (ICE) together. Both are very widespread among the cars, but they do not have the same attention for the motorcycles. The BEV technology is under progress for motorcycle, while HEV has a modestly progress among the motorcycles and this study focus on this powertrain. Using the Brazil federation informs and crossing with the electrical powertrain categories definitions, this study define which motorcycle categories is adequate to use the HEV on Brazil and the powertrain specifications of these motorcycle categories.

Keywords: battery electrical vehicle (BEV), hybrid electrical vehicle (HEV), internal combustion engine (ICE), motorcycle.

GJRE-B Classification: DDC Code: 629.47 LCC Code: TL795.5



BRAZIL MOTORCYCLES CATEGORIES AND HYBRID ELECTRICAL TECHNOLOGY COMPARISON WITH POWERTRAIN SIZING

Strictly as per the compliance and regulations of:



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Brazil Motorcycles Categories and Hybrid Electric Technology Comparison with Powertrain Sizing

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Keywords: battery electrical vehicle (BEV), hybrid electrical vehicle (HEV), internal combustion engine (ICE), motorcycle.

I. INTRODUCTION

a) Motivation

New times are coming and its means we need adapt to this new times. Thinking in a new world and in the future, the mobility is changing to adapt to new mindset and be healthier.

One of the contributor to pollution is the ICE (internal combustion engine), present in each city in the world, the ICE operation spread a lot of particles in the air and increase the pollution on the cities. According IEA report 2017, "transport sector alone contributes to 24% of CO2 emissions in 2015".

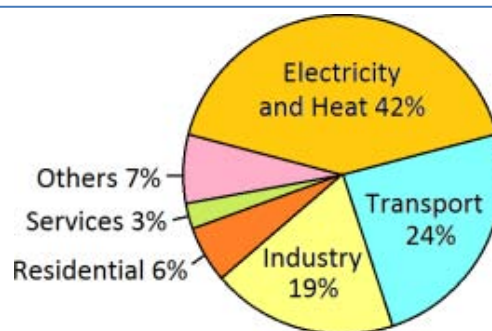


Figure 1: World CO2 Emissions from Fuel Combustion by Sector (IEA Report 2017)

The internal combustion engine was a huge step to society development and their evolution, but to achieve new targets with new mindset, the society are looking to be healthy. To achieve this new healthy target, we are walking to new technologies to decrease the pollution emissions from the transports, as the BEV (Battery Electric vehicles) and the HEV (hybrid electric vehicles).

i. BEV Definition

According Vidyanandan (2018), "Battery electric vehicles are propelled by electric motors by using energy stored on board in batteries". Therefore, BEV vehicles does not have a presence of the ICE to help the propulsion system. Basic, the BEV has the Battery, Engine and the Transmission

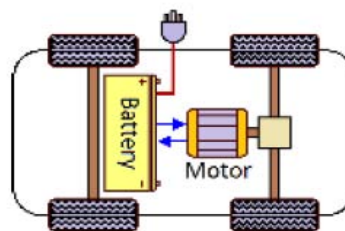


Figure 2: Basic BEV System (Vidyanandan, 2018)

ii. HEV Definition

HEV (Hybrid vehicles) have the internal combustion engine with the electric engine and, according Vidyanandan (2018), "Hybrid electric vehicles have the benefits of both ICE vehicles and electric vehicles, and overcome their individual disadvantages".

Author: Graduated in a Mechanical production engineering from the Rio de Janeiro State University (UERJ, 2014), Master business administration in project management from Getulio Vargas Foundation (FGV, 2017). Actually a R&D Project engineer in a multinational automotive company from France/Pays-Bas working in worldwide projects. e-mail: marc_falm@yahoo.com.br

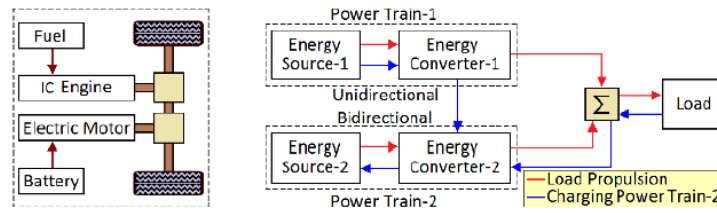


Figure 3: Arrangement of a HEV with Power Flow Paths(Vidyanandan, 2018)

For cars, the BEV or HEV technology are very solid and society has many examples used by the world-renowned brands as:

BEV: BMW i3 (BEV), Nissan Leaf, Chevrolet Bolt, Audi e-Tron and Renault Zoe;

HEV: Toyota Prius, Audi A7 Sportback, Ford Fusion Hybrid, BMW i3 (HEV), Volvo XC60 and many others.

However, the motorcycles category does not have the same scenario for both. The motorcycle world-renowned brands do not have the same presence of BEV or HEV and between both technologies have a difference, the companies have more BEV in comparison than HEV as.

BEV: Voltz EV01, Aima Tiger X6, MUUV Custom S, Magias Italiane Maranello and Energie Mobi Super Soco TC;

HEV: Honda PCX.

In addition, these BEV motorcycles have a low autonomy, being common to have between 60 and 80km.

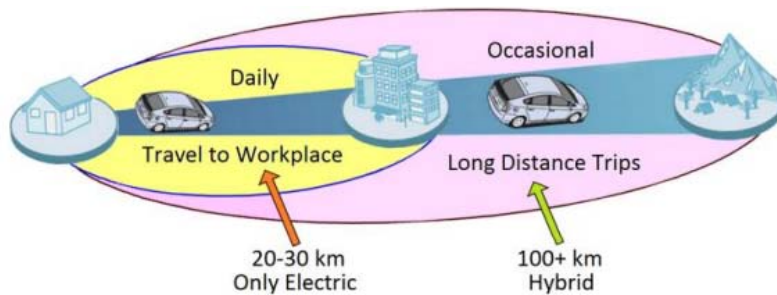


Figure 4: Travel Range of a Typical PHEV in Different Modes (Vidyanandan, 2018)

b) *Main Goals*

Knowing about the motorcycle technology opportunity for electric vehicle with large area to explore, this present article have as main goals:

- Identify the Brazilian motorcycle customer behavior;
- Identify the best match motorcycle for HEV;
- Define the specifications to sizing the hybrid engine.

II. COMPARISON BETWEEN BEV X HEV X ICE

According Vidyanandan (2018), the main difference between BEV and the HEV vehicle is the autonomy. BEV is better when the customer need to drive low distances and do not use the vehicle to travel, otherwise is better use the HEV for long distances.



Mass comparison between ICE x HEV x BEV

Table 1: Mass comparison ICE x BEV x HEV(Sources: Fiat Italy website; Volkswagen Portugal website; Peugeot UK website; Jac China website; Toyota USA website; Mitsubishi North America website; Nissan Japan website; Car and Drive website)

Vehicle	ICE	HEV	BEV	Difference BEV - ICE	Difference HEV - ICE	Difference BEV - HEV
Fiat 500	Rockstar ICE 930 kg	500 Hybrid HEV 980 kg	500e BEV 1351 kg	+421 kg	+50 kg	+371 kg
Volkswagen Golf	Golf 115CV 200Nm ICE 1240 kg		E-Golf 136CV BEV 1615 kg	+375 kg		
Volkswagen up	up 65CV 91Nm ICE 991 kg		e-up 83CV BEV 1235 kg	+244 kg		
Peugeot 208	1220cm ³ turbo ICE 1158 kg		136HP 260Nm BEV 1455 kg	+297 kg		
Jac S2 (IEV7S)	112HP 146Nm ICE 1110 kg		270Nm BEV 1495 kg	+385 kg		
Toyota Camry	2.5L 203HP 203HP ICE 1470 kg	2.5L 208HP 149Nm HEV 1610 kg			+140 kg	
Toyota Avalon	205HP 163lbf ICE 1620 kg	215HP 149lbf HEV 1640 kg			+20 kg	
Mitsubishi Outlander	166HP 220Nm ICE 1510 kg	80kW per engine 195Nm HEV 1915 kg			+405 kg	
Nissan Note	142Nm ICE 1090 kg	e-Power 254Nm HEV 1230 kg			+140 kg	
BMW i3		BMW i3 181HP 199Nm HEV 1500 kg	181HP 199Nm BEV 1379 kg			-121 kg

Using the last information, we can define the table below:

Table 2: Categories Main Points

Powertrain	Main points
ICE	Highest in CO2 emission in comparison than HEV / BEV High autonomy Lighter vehicle in comparison than HEV / BEV Quickly fuel (Easy for travels)
HEV	Low CO2 emission



BEV

- High autonomy
- Little heaviest
- Quickly fuel (Easy for travels)
- No CO2 emission due to engine
- Low autonomy
- Heaviest than ICE and HEV
- Lengthy fuel

III. BRAZILIAN MOTORCYCLE CATEGORIES

According Izo (2019), Brazil has the bellow main motorcycle categories:

Scooter: Scooter has 50cm³ to 150cm³ and aimed at younger customers. Normally, the gearbox is automatic; you have a good driveability inside the cities and have a pocket to keep small things. Scooters does not have the same comfort & safety than the biggest motorcycles and the pilot drive in the sitting position.



Figure 5: Scooter: Yamaha Nmax 160 (Izo, 2019)

Cub: Looks like scooters, but the pilot have a foot pegs to put your feet. The fuel economy is this motorcycle spotlight.



Figure 6: Cub: Honda Biz 125 2018 (Izo, 2019)

Sport: Sport motorcycles were created for strong accelerations. However, this motorcycle do not have a comfortable seat due to design made to optimize the aerodynamic (pilot must to put his chest close the tank to increase the aerodynamic). The suspension is very rigid and the seat usually is uncomfortable. This motorcycle can achieve easily 1200cm³.



Figure 7: Sport: Honda Cbr 1000rr Fireblade (Izo, 2019)

Naked: The customers usually drive in the cities or highways. Naked motorcycles have few fairing, only the necessary. This motorcycle has a large range of sizes (200cm³ - 1000cm³).



Figure 8: Naked: YAMAHA MT-07 ABS 2019 (Izo, 2019)

Custom: Made for roadways. Design for pilot comfort with low seat, long suspensions and high handlebar. Normally have a range of sizes (800cm³ - 1000cm³).



Figure 9: Custo: Harley-Davidson Sportster 883 2018 (Izo, 2019)

Trail: Tall motorcycle with tall seat, suspension and handlebar. Trail motorcycle has a good driveability for city and travels. Normally the customer drive in different roads (dust, asphalt and others). Displacement average close the 1000cm³.



Figure 10: Trail: Triumph Tiger 800 Xrx (Izo, 2019)

Below the table with the motorcycle categories description:

Table 3: Categories Main Points

Category	Main points
Scooter	City usage
	Young customers
	Low displacement (50cm ³ - 150cm ³)
	Economic
	Uncomfortable (Drive in sitting position)
Cub	City usage
	Young customers
	Low displacement (100cm ³ - 125cm ³)
	Economic
	Average comfortable (Drive with foot pegs)
Sport	Sport usage
	“Sport” customers
	High displacement (as 1200cm ³)
	High consumption
	Uncomfortable
Naked	City / highway usage
	Large range of customers
	Large range of displacement (200cm ³ - 1000cm ³)





	Large range of consumption
	Average comfort
Custom	Highway / Travel usage
	Traveling Customers
	High displacement (until 1800cm ³)
	Average to high consumption
	Very comfortable
Trail	City / Travel / Trail usage
	Daily usage with some travels
	High displacement (average of 1000cm ³)
	Average to high consumption
	Comfortable

IV. COMPARISON BETWEEN MOTORCYCLE AND POWERTRAINS

Below the table crossing the information from motorcycles categories and the engine types:

Table 4: Comparison Between Categories and Engines

	ICEV	HEV	BEV
	Highest in CO2 emission in comparison than HEV / BEV	Low CO2 emission	No CO2 emission due to engine
	High autonomy	High autonomy	Low autonomy
	Lighter vehicle in comparison than HEV / BEV	Little heaviest	Heaviest than ICE and HEV
	Quickly fuel (Easy for travels)	Quickly fuel (Easy for travels)	Lengthy fuel
Scooter			
City usage			
Young customers			
Low displacement (50cm ³ - 150cm ³)	3	2	1
Economic			
Uncomfortable (Drive in sitting position)			
Cub			
City usage			
Young customers			
Low displacement (100cm ³ - 125cm ³)	3	2	1
Economic			
Average comfortable (Drive with foot pegs)			
Sport			
Sport usage	1	3	2
"Sport" customers			

High displacement (as 1200cm ³) High consumption Uncomfortable			
Naked City / highway usage Large range of customers Large range of displacement (200cm ³ - 1000cm ³) Large range of consumption Average comfort	1	2	3
Custom Highway / Travel usage Traveling Customers High displacement (until 1800cm ³) Average to high consumption Very comfortable	2	1	3
Trail and Big Trail City / Travel / Trail usage Daily usage with some travels High displacement (average of 1000cm ³) Average to high consumption Comfortable	2	1	3

According to the table, Custom and Trail are the most compatible motorcycle categories with HEV technology because they are used for long travels, need a quick fuel, high autonomy and use to decrease pollution.

V. SPECIFICATION FOR CUSTOM AND TRAIL

Fenabrave is a national automotive federation from Brazil and the best in class motorcycle for each category could be identified through the Fenabrave informs. Below the Fenabrave informs ranking table from December 2019 for Trail motorcycles and Customs:



Figure 11: Ranking Fenabrave December 2019 (Fenabrave, 2020)

Based on the sales ranking from Fenabrave, the best sales motorcycle specification will be used to define the motorcycle specification target.

Table 5: Specification table (Sources: Triumph Brazil website; BMW Brazil website; Suzuki Brazil website; Harley Davidson Brazil website; Kawazaki Brazil website; Royal Enfield Brazil website)

Motorcycle	Torque	Power	Energy	Mass
Triumph / Tiger800	79 Nm (8,0 kgf.m) @ 7,850 rpm	95 CV @ 9,250 rpm	70 kW @ 9,250 rpm	199 kg
BMW / F850 GS	88 Nm (9,0 kgf.m) @ 6,250rpm	80 CV @ 6,250 rpm	58 kW @ 6,250 rpm	229 kg
BMW / R1250GS	143 Nm (14,6 kgf.m) @ 6,250 rpm	136 CV @ 7,750 rpm	100 kW @ 7,750 rpm	249 kg
BMW / R1200	125 Nm (12,7 kgf.m) @ 6,500 rpm	92 CV @ 7,750 rpm	92 kW @ 7,750 rpm	232 kg
Triumph / Tiger 1200	122 Nm (12,4 kgf.m) @ 7,600rpm	141 CV @ 9,350 rpm	104 kW @ 9,350 rpm	242 kg
Suzuki / Vstrom650	62 Nm (6,32 kgf.m) @ 6,500RPM	71 CV @ 8,800 rpm	52 kW @ 8,800 rpm	199 kg
H.Davison / FL FB	145 Nm (14,8 kgf.m) @ 3,000 rpm	71 CV @ 4,560 rpm	52 kW @ 4,560 rpm	304 kg
Kawazaki / Vulcan S	63 Nm (6,4 kgf.m) @ 6,600 rpm	61 CV @ 7,500 rpm	45 kW @ 7,500 rpm	228 kg
H.Davison / XL 883	68 Nm (6,9 kgf.m) @ 4,750 rpm	52 CV @ 5,750 rpm	38 kW @ 5,750 rpm	247 kg
H.Davison / XL 1200	96 Nm (9,8 kgf.m) @ 3,500 rpm	66 CV @ 6,000 rpm	49 kW @ 6,000 rpm	248 kg
Royal enfield / Classic	52 Nm (5,3 kgf.m) @ 5,250 rpm	47 CV @ 7,250 rpm	35 kW @ 7,250 rpm	202 kg

Follow the train of thought, below the specification comparison and the analysis to define the targets for Custom and Trail motorcycle categories.

Table 6: Motorcycle Categories Analysis

Motorcycle	Peso	Torque	Torque / kg	Torque/kg variation (Unid / Cat average)
Triumph / Tiger800	199 kg	79 Nm	0,40 Nm/kg	-12%
BMW / F850 GS	229 kg	88 Nm	0,38 Nm/kg	-15%
BMW / R1250GS	249 kg	143 Nm	0,57 Nm/kg	27%
BMW / R1200	232 kg	125 Nm	0,54 Nm/kg	19%
Triumph / Tiger 1200	242 kg	122 Nm	0,50 Nm/kg	12%
Suzuki / Vstrom650	199 kg	62 Nm	0,31 Nm/kg	-31%
H.Davison / FL FB	304 kg	145 Nm	0,48 Nm/kg	43%
Kawazaki / Vulcan S	228 kg	63 Nm	0,28 Nm/kg	-17%
H.Davison / XL 883	247 kg	68 Nm	0,28 Nm/kg	-18%
H.Davison / XL 1200	248 kg	96 Nm	0,39 Nm/kg	16%
Royal enfield / Classic	202 kg	52 Nm	0,26 Nm/kg	-23%
Average MaxTrail	225 kg	103 Nm	0,45 Nm/kg	
Min MaxTrail	199 kg	62 Nm	0,31 Nm/kg	
Max MaxTrail	249 kg	143 Nm	0,57 Nm/kg	
Average Custom	246 kg	85 Nm	0,33 Nm/kg	
Min Custom	202 kg	52 Nm	0,26 Nm/kg	
Max Custom	304 kg	145 Nm	0,48 Nm/kg	
Geral Average	234 kg	95 Nm	0,40 Nm/kg	

According the Table 6. Motorcycle categories analysis, the Trail specifications are:

Weight: Average of 225kg, range between 199Kg and 249Kg;

Torque: Average of 103Nm, range between 62Nm and 143Nm;

Correlation between torque and weight: Average of 0,45Nm/Kg, range between 0,31Nm/kg and 0,57Nm/Kg. And the Custom specification are:

Weight: Average of 246kg, range between 202Kg and 304Kg;

Torque: Average of 85Nm, range between 52Nm and 145Nm;

Correlation between torque and weight: Average of 0,33Nm/Kg, range between 0,26Nm/kg and 0,48Nm/Kg.

VI. CONCLUSION

According to these work data, the motorcycle categories with best match to hybrid electric vehicle (HEV) technology are Custom and Trail motorcycle categories due the necessities to do travels and, consequently, need more autonomy and a quickly fuel.

Need to consider some points to design the HEV powertrain for motorcycle. Following these work analysis:

- Correlation between torque and weight is important because it demonstrates how much torque the motorcycle needs to meet the customer's behavior;
- Weight demonstrate the range of mass the motorcycle could be to meet the customer's behavior.

For example, the trail motorcycles customer drives in different roads and some roads, as dirt or bumpy roads, the customer need a lighter and taller motorcycle with high torque (as 0,45Nm/Kg), in comparison the custom motorcycle customer basically use on asphalt and needs a heaviest and lower motorcycle with a reasonable torque (0,33Nm/Kg). Therefore, the trail motorcycle must be lighter than custom motorcycle and, normally, the trail motorcycle has more torque than custom motorcycle.

In addition, considering the analysis, the good motorcycle target to apply HEV technology is:

- Trail motorcycles: Triumph/Tiger800 is the motorcycle close the average with 0,40Nm/kg against the average of 0,45Nm/kg and this is the best-selling motorcycle for Trail category;
- Custom motorcycles: H.Davison/XL883 is the motorcycle close the average with 0,28Nm/kg against the average of 0,33Nm/kg and H.Davison is the brand best-selling motorcycles for Custom categories;

Abbreviations:

ICE – Internal combustion engine

HEV – Hybrid electrical vehicle

BEV – Battery electrical vehicle

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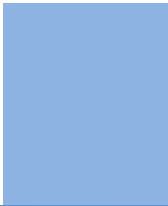
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The abstract is the foundation of the research paper. It should be clear and concise and must contain the objective of the paper and inferences drawn. It is advised to not include big mathematical equations or complicated jargon.

Many researchers searching for information online will use search engines such as Google, Yahoo or others. By optimizing your paper for search engines, you will amplify the chance of someone finding it. In turn, this will make it more likely to be viewed and cited in further works. Global Journals has compiled these guidelines to facilitate you to maximize the web-friendliness of the most public part of your paper.

Keywords

A major lynchpin of research work for the writing of research papers is the keyword search, which one will employ to find both library and internet resources. Up to eleven keywords or very brief phrases have to be given to help data retrieval, mining, and indexing.

One must be persistent and creative in using keywords. An effective keyword search requires a strategy: planning of a list of possible keywords and phrases to try.

Choice of the main keywords is the first tool of writing a research paper. Research paper writing is an art. Keyword search should be as strategic as possible.

One should start brainstorming lists of potential keywords before even beginning searching. Think about the most important concepts related to research work. Ask, "What words would a source have to include to be truly valuable in a research paper?" Then consider synonyms for the important words.

It may take the discovery of only one important paper to steer in the right keyword direction because, in most databases, the keywords under which a research paper is abstracted are listed with the paper.

Numerical Methods

Numerical methods used should be transparent and, where appropriate, supported by references.

Abbreviations

Authors must list all the abbreviations used in the paper at the end of the paper or in a separate table before using them.

Formulas and equations

Authors are advised to submit any mathematical equation using either MathJax, KaTeX, or LaTeX, or in a very high-quality image.

Tables, Figures, and Figure Legends

Tables: Tables should be cautiously designed, uncrowned, and include only essential data. Each must have an Arabic number, e.g., Table 4, a self-explanatory caption, and be on a separate sheet. Authors must submit tables in an editable format and not as images. References to these tables (if any) must be mentioned accurately.



Figures

Figures are supposed to be submitted as separate files. Always include a citation in the text for each figure using Arabic numbers, e.g., Fig. 4. Artwork must be submitted online in vector electronic form or by emailing it.

PREPARATION OF ELETRONIC FIGURES FOR PUBLICATION

Although low-quality images are sufficient for review purposes, print publication requires high-quality images to prevent the final product being blurred or fuzzy. Submit (possibly by e-mail) EPS (line art) or TIFF (halftone/ photographs) files only. MS PowerPoint and Word Graphics are unsuitable for printed pictures. Avoid using pixel-oriented software. Scans (TIFF only) should have a resolution of at least 350 dpi (halftone) or 700 to 1100 dpi (line drawings). Please give the data for figures in black and white or submit a Color Work Agreement form. EPS files must be saved with fonts embedded (and with a TIFF preview, if possible).

For scanned images, the scanning resolution at final image size ought to be as follows to ensure good reproduction: line art: >650 dpi; halftones (including gel photographs): >350 dpi; figures containing both halftone and line images: >650 dpi.

Color charges: Authors are advised to pay the full cost for the reproduction of their color artwork. Hence, please note that if there is color artwork in your manuscript when it is accepted for publication, we would require you to complete and return a Color Work Agreement form before your paper can be published. Also, you can email your editor to remove the color fee after acceptance of the paper.

TIPS FOR WRITING A GOOD QUALITY ENGINEERING RESEARCH PAPER

Techniques for writing a good quality engineering research paper:

1. Choosing the topic: In most cases, the topic is selected by the interests of the author, but it can also be suggested by the guides. You can have several topics, and then judge which you are most comfortable with. This may be done by asking several questions of yourself, like "Will I be able to carry out a search in this area? Will I find all necessary resources to accomplish the search? Will I be able to find all information in this field area?" If the answer to this type of question is "yes," then you ought to choose that topic. In most cases, you may have to conduct surveys and visit several places. Also, you might have to do a lot of work to find all the rises and falls of the various data on that subject. Sometimes, detailed information plays a vital role, instead of short information. Evaluators are human: The first thing to remember is that evaluators are also human beings. They are not only meant for rejecting a paper. They are here to evaluate your paper. So present your best aspect.

2. Think like evaluators: If you are in confusion or getting demotivated because your paper may not be accepted by the evaluators, then think, and try to evaluate your paper like an evaluator. Try to understand what an evaluator wants in your research paper, and you will automatically have your answer. Make blueprints of paper: The outline is the plan or framework that will help you to arrange your thoughts. It will make your paper logical. But remember that all points of your outline must be related to the topic you have chosen.

3. Ask your guides: If you are having any difficulty with your research, then do not hesitate to share your difficulty with your guide (if you have one). They will surely help you out and resolve your doubts. If you can't clarify what exactly you require for your work, then ask your supervisor to help you with an alternative. He or she might also provide you with a list of essential readings.

4. Use of computer is recommended: As you are doing research in the field of research engineering then this point is quite obvious. Use right software: Always use good quality software packages. If you are not capable of judging good software, then you can lose the quality of your paper unknowingly. There are various programs available to help you which you can get through the internet.

5. Use the internet for help: An excellent start for your paper is using Google. It is a wondrous search engine, where you can have your doubts resolved. You may also read some answers for the frequent question of how to write your research paper or find a model research paper. You can download books from the internet. If you have all the required books, place importance on reading, selecting, and analyzing the specified information. Then sketch out your research paper. Use big pictures: You may use encyclopedias like Wikipedia to get pictures with the best resolution. At Global Journals, you should strictly follow [here](#).



6. Bookmarks are useful: When you read any book or magazine, you generally use bookmarks, right? It is a good habit which helps to not lose your continuity. You should always use bookmarks while searching on the internet also, which will make your search easier.

7. Revise what you wrote: When you write anything, always read it, summarize it, and then finalize it.

8. Make every effort: Make every effort to mention what you are going to write in your paper. That means always have a good start. Try to mention everything in the introduction—what is the need for a particular research paper. Polish your work with good writing skills and always give an evaluator what he wants. Make backups: When you are going to do any important thing like making a research paper, you should always have backup copies of it either on your computer or on paper. This protects you from losing any portion of your important data.

9. Produce good diagrams of your own: Always try to include good charts or diagrams in your paper to improve quality. Using several unnecessary diagrams will degrade the quality of your paper by creating a hodgepodge. So always try to include diagrams which were made by you to improve the readability of your paper. Use of direct quotes: When you do research relevant to literature, history, or current affairs, then use of quotes becomes essential, but if the study is relevant to science, use of quotes is not preferable.

10. Use proper verb tense: Use proper verb tenses in your paper. Use past tense to present those events that have happened. Use present tense to indicate events that are going on. Use future tense to indicate events that will happen in the future. Use of wrong tenses will confuse the evaluator. Avoid sentences that are incomplete.

11. Pick a good study spot: Always try to pick a spot for your research which is quiet. Not every spot is good for studying.

12. Know what you know: Always try to know what you know by making objectives, otherwise you will be confused and unable to achieve your target.

13. Use good grammar: Always use good grammar and words that will have a positive impact on the evaluator; use of good vocabulary does not mean using tough words which the evaluator has to find in a dictionary. Do not fragment sentences. Eliminate one-word sentences. Do not ever use a big word when a smaller one would suffice.

Verbs have to be in agreement with their subjects. In a research paper, do not start sentences with conjunctions or finish them with prepositions. When writing formally, it is advisable to never split an infinitive because someone will (wrongly) complain. Avoid clichés like a disease. Always shun irritating alliteration. Use language which is simple and straightforward. Put together a neat summary.

14. Arrangement of information: Each section of the main body should start with an opening sentence, and there should be a changeover at the end of the section. Give only valid and powerful arguments for your topic. You may also maintain your arguments with records.

15. Never start at the last minute: Always allow enough time for research work. Leaving everything to the last minute will degrade your paper and spoil your work.

16. Multitasking in research is not good: Doing several things at the same time is a bad habit in the case of research activity. Research is an area where everything has a particular time slot. Divide your research work into parts, and do a particular part in a particular time slot.

17. Never copy others' work: Never copy others' work and give it your name because if the evaluator has seen it anywhere, you will be in trouble. Take proper rest and food: No matter how many hours you spend on your research activity, if you are not taking care of your health, then all your efforts will have been in vain. For quality research, take proper rest and food.

18. Go to seminars: Attend seminars if the topic is relevant to your research area. Utilize all your resources.

19. Refresh your mind after intervals: Try to give your mind a rest by listening to soft music or sleeping in intervals. This will also improve your memory. Acquire colleagues: Always try to acquire colleagues. No matter how sharp you are, if you acquire colleagues, they can give you ideas which will be helpful to your research.

20. Think technically: Always think technically. If anything happens, search for its reasons, benefits, and demerits. Think and then print: When you go to print your paper, check that tables are not split, headings are not detached from their descriptions, and page sequence is maintained.



21. Adding unnecessary information: Do not add unnecessary information like "I have used MS Excel to draw graphs." Irrelevant and inappropriate material is superfluous. Foreign terminology and phrases are not apropos. One should never take a broad view. Analogy is like feathers on a snake. Use words properly, regardless of how others use them. Remove quotations. Puns are for kids, not grunt readers. Never oversimplify: When adding material to your research paper, never go for oversimplification; this will definitely irritate the evaluator. Be specific. Never use rhythmic redundancies. Contractions shouldn't be used in a research paper. Comparisons are as terrible as clichés. Give up ampersands, abbreviations, and so on. Remove commas that are not necessary. Parenthetical words should be between brackets or commas. Understatement is always the best way to put forward earth-shaking thoughts. Give a detailed literary review.

22. Report concluded results: Use concluded results. From raw data, filter the results, and then conclude your studies based on measurements and observations taken. An appropriate number of decimal places should be used. Parenthetical remarks are prohibited here. Proofread carefully at the final stage. At the end, give an outline to your arguments. Spot perspectives of further study of the subject. Justify your conclusion at the bottom sufficiently, which will probably include examples.

23. Upon conclusion: Once you have concluded your research, the next most important step is to present your findings. Presentation is extremely important as it is the definite medium through which your research is going to be in print for the rest of the crowd. Care should be taken to categorize your thoughts well and present them in a logical and neat manner. A good quality research paper format is essential because it serves to highlight your research paper and bring to light all necessary aspects of your research.

INFORMAL GUIDELINES OF RESEARCH PAPER WRITING

Key points to remember:

- Submit all work in its final form.
- Write your paper in the form which is presented in the guidelines using the template.
- Please note the criteria peer reviewers will use for grading the final paper.

Final points:

One purpose of organizing a research paper is to let people interpret your efforts selectively. The journal requires the following sections, submitted in the order listed, with each section starting on a new page:

The introduction: This will be compiled from reference matter and reflect the design processes or outline of basis that directed you to make a study. As you carry out the process of study, the method and process section will be constructed like that. The results segment will show related statistics in nearly sequential order and direct reviewers to similar intellectual paths throughout the data that you gathered to carry out your study.

The discussion section:

This will provide understanding of the data and projections as to the implications of the results. The use of good quality references throughout the paper will give the effort trustworthiness by representing an alertness to prior workings.

Writing a research paper is not an easy job, no matter how trouble-free the actual research or concept. Practice, excellent preparation, and controlled record-keeping are the only means to make straightforward progression.

General style:

Specific editorial column necessities for compliance of a manuscript will always take over from directions in these general guidelines.

To make a paper clear: Adhere to recommended page limits.

Mistakes to avoid:

- Insertion of a title at the foot of a page with subsequent text on the next page.
- Separating a table, chart, or figure—confine each to a single page.
- Submitting a manuscript with pages out of sequence.
- In every section of your document, use standard writing style, including articles ("a" and "the").
- Keep paying attention to the topic of the paper.



- Use paragraphs to split each significant point (excluding the abstract).
- Align the primary line of each section.
- Present your points in sound order.
- Use present tense to report well-accepted matters.
- Use past tense to describe specific results.
- Do not use familiar wording; don't address the reviewer directly. Don't use slang or superlatives.
- Avoid use of extra pictures—include only those figures essential to presenting results.

Title page:

Choose a revealing title. It should be short and include the name(s) and address(es) of all authors. It should not have acronyms or abbreviations or exceed two printed lines.

Abstract: This summary should be two hundred words or less. It should clearly and briefly explain the key findings reported in the manuscript and must have precise statistics. It should not have acronyms or abbreviations. It should be logical in itself. Do not cite references at this point.

An abstract is a brief, distinct paragraph summary of finished work or work in development. In a minute or less, a reviewer can be taught the foundation behind the study, common approaches to the problem, relevant results, and significant conclusions or new questions.

Write your summary when your paper is completed because how can you write the summary of anything which is not yet written? Wealth of terminology is very essential in abstract. Use comprehensive sentences, and do not sacrifice readability for brevity; you can maintain it succinctly by phrasing sentences so that they provide more than a lone rationale. The author can at this moment go straight to shortening the outcome. Sum up the study with the subsequent elements in any summary. Try to limit the initial two items to no more than one line each.

Reason for writing the article—theory, overall issue, purpose.

- Fundamental goal.
- To-the-point depiction of the research.
- Consequences, including definite statistics—if the consequences are quantitative in nature, account for this; results of any numerical analysis should be reported. Significant conclusions or questions that emerge from the research.

Approach:

- Single section and succinct.
- An outline of the job done is always written in past tense.
- Concentrate on shortening results—limit background information to a verdict or two.
- Exact spelling, clarity of sentences and phrases, and appropriate reporting of quantities (proper units, important statistics) are just as significant in an abstract as they are anywhere else.

Introduction:

The introduction should "introduce" the manuscript. The reviewer should be presented with sufficient background information to be capable of comprehending and calculating the purpose of your study without having to refer to other works. The basis for the study should be offered. Give the most important references, but avoid making a comprehensive appraisal of the topic. Describe the problem visibly. If the problem is not acknowledged in a logical, reasonable way, the reviewer will give no attention to your results. Speak in common terms about techniques used to explain the problem, if needed, but do not present any particulars about the protocols here.

The following approach can create a valuable beginning:

- Explain the value (significance) of the study.
- Defend the model—why did you employ this particular system or method? What is its compensation? Remark upon its appropriateness from an abstract point of view as well as pointing out sensible reasons for using it.
- Present a justification. State your particular theory(-ies) or aim(s), and describe the logic that led you to choose them.
- Briefly explain the study's tentative purpose and how it meets the declared objectives.



Approach:

Use past tense except for when referring to recognized facts. After all, the manuscript will be submitted after the entire job is done. Sort out your thoughts; manufacture one key point for every section. If you make the four points listed above, you will need at least four paragraphs. Present surrounding information only when it is necessary to support a situation. The reviewer does not desire to read everything you know about a topic. Shape the theory specifically—do not take a broad view.

As always, give awareness to spelling, simplicity, and correctness of sentences and phrases.

Procedures (methods and materials):

This part is supposed to be the easiest to carve if you have good skills. A soundly written procedures segment allows a capable scientist to replicate your results. Present precise information about your supplies. The suppliers and clarity of reagents can be helpful bits of information. Present methods in sequential order, but linked methodologies can be grouped as a segment. Be concise when relating the protocols. Attempt to give the least amount of information that would permit another capable scientist to replicate your outcome, but be cautious that vital information is integrated. The use of subheadings is suggested and ought to be synchronized with the results section.

When a technique is used that has been well-described in another section, mention the specific item describing the way, but draw the basic principle while stating the situation. The purpose is to show all particular resources and broad procedures so that another person may use some or all of the methods in one more study or referee the scientific value of your work. It is not to be a step-by-step report of the whole thing you did, nor is a methods section a set of orders.

Materials:

Materials may be reported in part of a section or else they may be recognized along with your measures.

Methods:

- Report the method and not the particulars of each process that engaged the same methodology.
- Describe the method entirely.
- To be succinct, present methods under headings dedicated to specific dealings or groups of measures.
- Simplify—detail how procedures were completed, not how they were performed on a particular day.
- If well-known procedures were used, account for the procedure by name, possibly with a reference, and that's all.

Approach:

It is embarrassing to use vigorous voice when documenting methods without using first person, which would focus the reviewer's interest on the researcher rather than the job. As a result, when writing up the methods, most authors use third person passive voice.

Use standard style in this and every other part of the paper—avoid familiar lists, and use full sentences.

What to keep away from:

- Resources and methods are not a set of information.
- Skip all descriptive information and surroundings—save it for the argument.
- Leave out information that is immaterial to a third party.

Results:

The principle of a results segment is to present and demonstrate your conclusion. Create this part as entirely objective details of the outcome, and save all understanding for the discussion.

The page length of this segment is set by the sum and types of data to be reported. Use statistics and tables, if suitable, to present consequences most efficiently.

You must clearly differentiate material which would usually be incorporated in a study editorial from any unprocessed data or additional appendix matter that would not be available. In fact, such matters should not be submitted at all except if requested by the instructor.



Content:

- Sum up your conclusions in text and demonstrate them, if suitable, with figures and tables.
- In the manuscript, explain each of your consequences, and point the reader to remarks that are most appropriate.
- Present a background, such as by describing the question that was addressed by creation of an exacting study.
- Explain results of control experiments and give remarks that are not accessible in a prescribed figure or table, if appropriate.
- Examine your data, then prepare the analyzed (transformed) data in the form of a figure (graph), table, or manuscript.

What to stay away from:

- Do not discuss or infer your outcome, report surrounding information, or try to explain anything.
- Do not include raw data or intermediate calculations in a research manuscript.
- Do not present similar data more than once.
- A manuscript should complement any figures or tables, not duplicate information.
- Never confuse figures with tables—there is a difference.

Approach:

As always, use past tense when you submit your results, and put the whole thing in a reasonable order.

Put figures and tables, appropriately numbered, in order at the end of the report.

If you desire, you may place your figures and tables properly within the text of your results section.

Figures and tables:

If you put figures and tables at the end of some details, make certain that they are visibly distinguished from any attached appendix materials, such as raw facts. Whatever the position, each table must be titled, numbered one after the other, and include a heading. All figures and tables must be divided from the text.

Discussion:

The discussion is expected to be the trickiest segment to write. A lot of papers submitted to the journal are discarded based on problems with the discussion. There is no rule for how long an argument should be.

Position your understanding of the outcome visibly to lead the reviewer through your conclusions, and then finish the paper with a summing up of the implications of the study. The purpose here is to offer an understanding of your results and support all of your conclusions, using facts from your research and generally accepted information, if suitable. The implication of results should be fully described.

Infer your data in the conversation in suitable depth. This means that when you clarify an observable fact, you must explain mechanisms that may account for the observation. If your results vary from your prospect, make clear why that may have happened. If your results agree, then explain the theory that the proof supported. It is never suitable to just state that the data approved the prospect, and let it drop at that. Make a decision as to whether each premise is supported or discarded or if you cannot make a conclusion with assurance. Do not just dismiss a study or part of a study as "uncertain."

Research papers are not acknowledged if the work is imperfect. Draw what conclusions you can based upon the results that you have, and take care of the study as a finished work.

- You may propose future guidelines, such as how an experiment might be personalized to accomplish a new idea.
- Give details of all of your remarks as much as possible, focusing on mechanisms.
- Make a decision as to whether the tentative design sufficiently addressed the theory and whether or not it was correctly restricted. Try to present substitute explanations if they are sensible alternatives.
- One piece of research will not counter an overall question, so maintain the large picture in mind. Where do you go next? The best studies unlock new avenues of study. What questions remain?
- Recommendations for detailed papers will offer supplementary suggestions.



Approach:

When you refer to information, differentiate data generated by your own studies from other available information. Present work done by specific persons (including you) in past tense.

Describe generally acknowledged facts and main beliefs in present tense.

THE ADMINISTRATION RULES

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CRITERION FOR GRADING A RESEARCH PAPER (COMPILATION)
BY GLOBAL JOURNALS

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Topics	Grades		
	A-B	C-D	E-F
<i>Abstract</i>	Clear and concise with appropriate content, Correct format. 200 words or below	Unclear summary and no specific data, Incorrect form Above 200 words	No specific data with ambiguous information Above 250 words
<i>Introduction</i>	Containing all background details with clear goal and appropriate details, flow specification, no grammar and spelling mistake, well organized sentence and paragraph, reference cited	Unclear and confusing data, appropriate format, grammar and spelling errors with unorganized matter	Out of place depth and content, hazy format
<i>Methods and Procedures</i>	Clear and to the point with well arranged paragraph, precision and accuracy of facts and figures, well organized subheads	Difficult to comprehend with embarrassed text, too much explanation but completed	Incorrect and unorganized structure with hazy meaning
<i>Result</i>	Well organized, Clear and specific, Correct units with precision, correct data, well structuring of paragraph, no grammar and spelling mistake	Complete and embarrassed text, difficult to comprehend	Irregular format with wrong facts and figures
<i>Discussion</i>	Well organized, meaningful specification, sound conclusion, logical and concise explanation, highly structured paragraph reference cited	Wordy, unclear conclusion, spurious	Conclusion is not cited, unorganized, difficult to comprehend
<i>References</i>	Complete and correct format, well organized	Beside the point, Incomplete	Wrong format and structuring



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