Conservation and Aquaculture Research and Development Project for *Glossogobius giuris* (Biyang puti) in Naujan Lake (Goby Project)

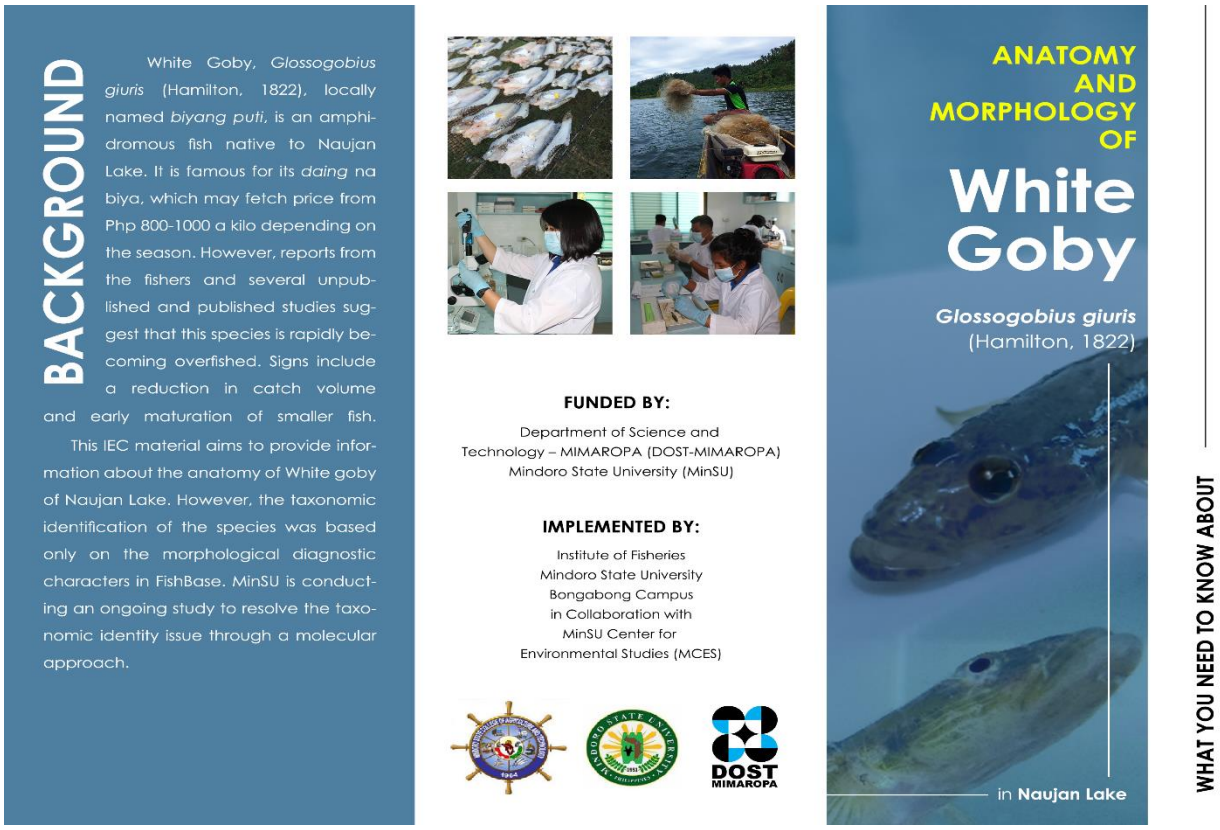
Description:

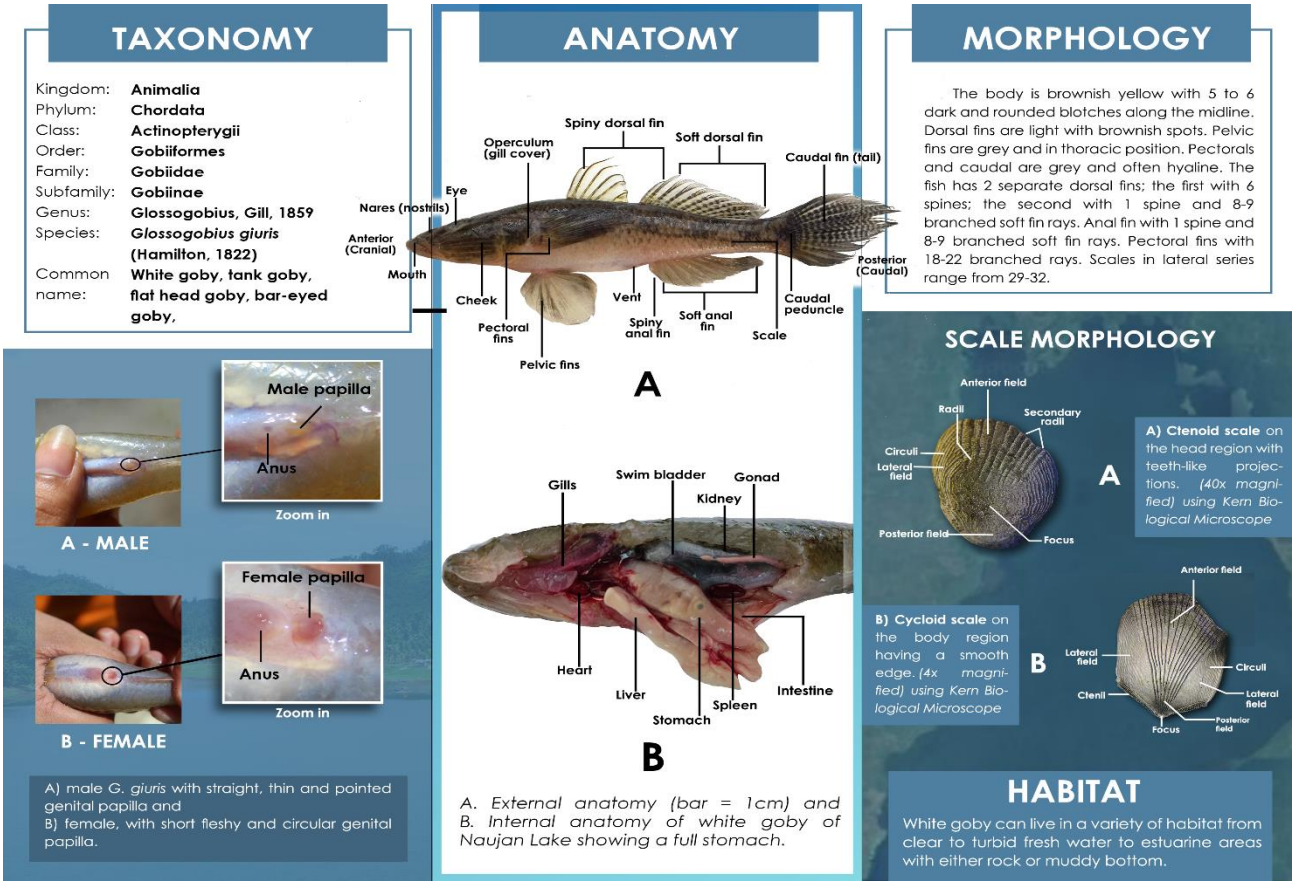
The Institute of Fisheries offers a Bachelor of Science in Fisheries, preparing graduates to enhance and increase aquatic wildlife populations while addressing community and industry needs. The program enhances student practical skills and industry readiness through hands-on training in food processing techniques like smoking, bottling, canning, and baking. It fosters community engagement and learning by conducting at least four community training sessions per semester. Additionally, the program aims to develop the food processing center into a valuable income-generating facility for the university by producing and selling processed food products, supporting both academic and economic objectives to ensure sustainability and growth within the institution.

Reproductive Biology of *G. aureus*

This study aims to address and identify the reproductive biology of the Naujan White Goby for aquaculture potential in inland fisheries. This also provides knowledge on the community on the cycle of the species on the lake.

Instructional Materials produced:





Information Dissemination:

Gut Content Analysis of White Goby

Gonadosomatic Index and Fecundity Analysis of White Goby

S

Fieldwork at Naujan Lake for White Goby

Publication Evidence:

Gmail — Elsie Guibone <gebrielle10@gmail.com>

short comm_submission
2 messages

katherine sanchez-escalona <nyctarinia@gmail.com> Tue, Dec 13, 2022 at 8:06 PM
To: TPJF NFRDI <journal.nfrdi@gmail.com>
Cc: Elsie Guibone <gebrielle10@gmail.com>, Abelle Balagtas <jaeniushp99@gmail.com>

Manuscript Cover Letter

13 December 2022

Mudjekeewis D. Santos, Ph.D.
Editor-in-Chief
The Philippine Journal of Fisheries

Dear Dr. Santos,

We are pleased to submit our manuscript entitled ***Lernaea sp.* (Copepoda, Lernaeidae) infestation in wild-caught Naujan White Goby, *Glossogobius sp.* broodstock from Naujan Lake** for consideration by The Philippines Journal of Fisheries (TPJF) as a short communication.

We confirm that this manuscript is our original work and has not been previously published or submitted for publication elsewhere. All authors listed on the title page have participated in the research and preparation of the manuscript. Specifically, the following is our author statement.

Abelle F. Balagtas: Conceptualization, Methodology, Writing, Data curation
Elsie Y. Guibone: Conceptualization, Methodology, Writing, Data Curation
Katherine P. Sanchez-Escalona: Writing-Reviewing and Editing, Supervision

The researchers followed all institutional and national guidelines for the care and use of laboratory animals.

To the best of our knowledge, no conflict of interest exists.

Please find below the list of suggested reviewers who have not read our manuscript prior to submission to TPJF:

1. Dr. Casiano H. Choresca Jr.
DA-National Fisheries Research and Development Institute
cchorescajr@gmail.com

2. Dr. Erlinda C. Lacierda
University of the Philippines Visayas (retired Professor)
eclacierda@yahoo.com

Exploration of the aquaculture potentials of the Naujan White Goby Glossogobius sp.

Communication letter

Mindoro State University
Victoria, Oriental Mindoro 5205, Philippines

E-mail: [illegible]
Website: www.[illegible].edu.ph
Phone: +63 [illegible]

21 January 2022

DR. REYNALDO V. EBORA
Executive Director
PCAARRD

Sir:

Greetings!

It is with great honor and pride to submit to you the first Annual Progress Report of the Goby Project reflecting the achievements of the Project for 2021.

Thank you for working with us in conservation.

Respectfully yours,

Sdg

DR. LEVY B. ARAGO, JR.
University President

Mindoro State University
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MELGAR G. FA[illegible]
DATE: _____

Sdg

14 February 2023

DR. REYNALDO V. EBORA
Executive Director
PCAARRD

DOST Form 7

EXECUTIVE SUMMARY FOR THE ANNUAL PROGRESS REPORT

(Attach DOST Forms 6, 8, 9, 11, 12)

(1) Program Title:
Project Title: **Conservation and Aquaculture Research and Development Project for Glossogobius giuris (Biyang Puti) in Naujan Lake (Goby Project)**
Project Leader/Gender: **Katherine P. Sanchez-Escalona/F**
Agency: **Mindoro State University**
Address/Telephone/Fax/Email: **Brgy. Alcate, Victoria, Oriental Mindoro/09166094464/nyctarinia@gmail.com**

(2) Cooperating Agency/ies

(3) Site/s of Implementation (Barangay / Municipality / District / Province / Region / Country)
Base Station: Mindoro State University Victoria Campus
Site/s of Implementation: Brgy. Balatasan, Bulalacao
Brgy. Maasin, Bulalacao
Brgy. Manaul, Mansalay

(4) Project Duration (number of months)	(5) Project Start Date	(6) Project End Date
24 months	**January 2021**	**December 2022**

(7) **Major Accomplishments**
a. Drafted 5 MOA for the projects: Provincial Government of Oriental Mindoro, Municipality of Bulalacao, tripartite agreement with Municipality of Mansalay and BFARMC of Manaul, BFARMC of Brgy. Balatasan, and BFARMC of Brgy. Maasin
b. Conducted project inception meetings and consultations with the beneficiary barangays
c. Conducted social profiling for all the beneficiary communities that was used as a basis in drafting the training modules
d. Conducted entrepreneurship training in Brgy. Manaul, Mansalay, and Brgys. Maasin and Balatasan in Bulalacao

A. Actual accomplishment of the project (vis-a-vis the objectives)

OBJECTIVES	ACCOMPLISHMENTS		
	TARGET	ACTUAL	PERCENTAGE (%)
To establish an experimental seaweed farm plot using viable laboratory-reared cultivars from PSU MSL culture facilities, 1,000m2	Established 1000 m2 experimental seaweed farm	All supplies for the construction had been purchased One of the three beneficiary communities has already started work i.e. net sewing, bamboo felling, collection of empty bottles as floaters	60%
To construct one (1) units of floating seaweed dryers	Constructed 1 unit floating seaweed dryer	All supplies had been purchased	50%
To select good quality seedstocks	Selected good quality seedstock	Locally available seaweed seedstock had been sourced out for planting	20%

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DOST Form 7

EXECUTIVE SUMMARY FOR THE ANNUAL PROGRESS REPORT

(Attach DOST Forms 6, 8, 9, 11, 13)

(1) Program Title:
Project Title: Conservation and Aquaculture Research and Development Project for *Glossogobius giuris* (Biyang Puti) in Naujan Lake (Goby Project)
Project Leader/Gender: Katherine P. Sanchez-Escalona/F
Agency: Mindoro State University
Address/Telephone/Fax/Email: Brgy. Alcate, Victoria, Oriental Mindoro/09166094464/nyctarinia@gmail.com

(2) Cooperating Agency/ies

(3) Site/s of Implementation (Barangay / Municipality / District / Province / Region / Country)
Base Station: Mindoro State University Victoria Campus
Sites of Implementation: Mindoro State University Victoria Campus/Mindoro State University Bongabong Campus

(4) Project Duration (number of months)	(5) Project Start Date	(6) Project End Date
36 months	January 2021	December 2023

(7) Major Accomplishments
a. Drafted MOA with UP-Diliman Institute of Biology (UP-IB), Provincial Government of Oriental Mindoro (PGORM), and Bureau of Fisheries and Aquatic Resources –MiMaRoPa (BFAR-MiMaRoPa); Palompon Institute of Technology (PIT) for the collaborative sampling of fish in Lake Danao; Lake Danao National Park Protected Area Management Board (LDNP-PAMB); informal linkages with Barangay Bayani, Naujan, and Brgy. Malabo, Victoria for being the landing sites of specimens gathered; formed informal linkage with the Naujan Lake National Park Protected Area Management Board (NLNP-PAMB) for being the local clearing house of the project as well as the final recipient of the conservation outcomes of the project;
b. Collected fish and tissue samples from Naujan Lake for the hot, dry month season (April); and fish and tissue samples from Teal Lake, Laguna Lake, and Lanao Lake;
c. Initiated involvement of 4 undergraduate students in water quality study.
d. Leveraged for the development of Center for Environmental Studies.
e. presented 5 papers and drafted 4 papers for publication.
f. Training of the Goby Team (8 people) on aquaculture protocol of big head carp as basis for the protocol development for goby.

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A. Actual accomplishment of the project (vis-a-vis the objectives)

OBJECTIVES	ACCOMPLISHMENTS		
	TARGET	ACTUAL	PERCENTAGE (%)
Administrative Preparations Staffing Contact of Linkages Purchase of equipment	Project staff hired Linkages contacted and MOU drafted Equipment delivered	 All project staff hired Five (5) MOU drafted and finalized	 100% 100% 80%
Discriminate white goby populations within the lake based on geometric morphometrics	Morphological segregation of Naujan Lake White Goby population	100% of the morphometric data processed 80% of the fish from other lakes collected	100% 80%

Collection of samples Sample processing Data Analysis			
Describe environmental parameters of Naujan lake related to occurrence of white goby Collection of samples Sample processing Data Analysis	Physico-chemical and biological description of selected sites within Naujan Lake	80% of the Naujan Lake plankton samples collected and analyzed 0% benthos collected 0% sediment collected	80% 0% 0%

C. Expected Outputs / IPs (Expected Outputs should be measurable.)

	EXPECTED OUTPUTS	ACTUAL OUTPUTS
Publications/Presentations	One (1) publication	Five (5) presentations done, four (4) papers drafted for publication
Patents/IP		
Products		
People Services		Training of nine (9) project staff on aquaculture protocol; assisted conduct of one (1) baby thesis; had the proposal for the enhance of the MCES laboratories approved, together with the development of Natural Food Laboratory and the Aquaculture Laboratory
Places and Partnerships	Two (2) Memorandum of Agreements signed	Five (5) Memorandum of agreements drafted and finalized
Policy		

(8) Risk Management Plan

OBJECTIVES	(9) RISKS AND ASSUMPTIONS	(10) RISK MANAGEMENT PLAN	(11) ACTIONS TO BE TAKEN
Discriminate white goby populations within the lake based on geometric morphometrics.	Population of White Goby in the lake is differentiated morphologically that are either affected by environment and genetics.	Factors are intervened that failure to determine which affects more may defeat the purpose of using the data for broodstock selection. Thus, morphological data would be used as the first basis for broodstock selection.	Molecular approach would be optimized to have a good baseline on both aquaculture and restocking trajectory
Describe environmental parameters of Naujan lake related to occurrence of white goby.	Environmental parameters are related to population differentiation.		
Identify the genetic profile of *G. giuris* from Naujan Lake relative to other lakes around the Philippines.	Population of white goby in the lake is homogenous.		
Identify the appropriate conditioning parameters for *G. giuris* broodstock.	Conditioning parameters of broodstocks are dependent on the in-situ conditions.	The attainment of the objectives is interdependent that the failure in the attainment of the first objective will affect the results of the later objectives. As such, risk management plan would entail running trial experimental set-ups using known parameters and needs from known commercialized aquaculture species (i.e. tilapia, dayungan) would be done.	Rigor would be taken in treatment design
Look at the effect of broodstock size on breeding performance and hatchling production of *G. giuris* in natural spawning and hormone-induced spawning.	Larger broodstock results to higher hatchling production but no data yet on the optimum parameters by which broodstock can perform.		

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Study the appropriate conditions for egg incubation of *G. giuris* eggs.	Egg incubation is maximized given optimum conditions but data on the optimum conditions has to be determined yet.		
Ascertain the effects of larval stocking density on growth, survival and fry production of *G. giuris*	Larval density is inversely proportional to growth and survival.		
Growth and survival of hatchery-produced *G. giuris* fry fed diets containing different protein levels	Optimized growth is attained with optimum protein levels, however, there is no data yet on the relationship of protein level and growth rate.		
Optimization of feeding rate and frequency of *G. giuris* fry reared in nylon net cage (hapa) under brackishwater and freshwater environments	Optimized growth is attained with optimized feeding, however, there is no data yet on feeding rate and growth rate.		

(12) Problems/Concerns

The implementation of molecular part of Study 1 is delayed due to the incompatible power supply and delay in the delivery of equipment by the supplier.

(13) Suggested solutions to the above concerns

Incompatible power supply - strong coordination with supply office is being done.
Delivery of equipment – follow-up is regular with the supplier.

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Coastal Resource Management

Description:

This project aims to educate the community on coastal management through continuous monitoring of the seas and aquatic resources. It serves as an ongoing activity for the university, ensuring that it remains a catalyst for promoting the conservation and sustainable utilization of marine resources. By actively engaging the community, the project not only disseminates knowledge about the importance of preserving coastal environments but also fosters a sense of responsibility among the residents. The university's commitment to this initiative highlights its role in advocating for environmental stewardship and sustainable practices, ensuring the health and longevity of marine ecosystems for future generations.





Activities:

ALWAN Training

Alwan Training is an innovative approach for community-based science that focuses on monitoring the coastal areas of Oriental Mindoro Island to sustain resources through continuous observation and surveillance of aquatic ecosystems. This method prioritizes community engagement and participation, allowing local residents to play an active role in environmental conservation efforts. The training is designed to be accessible and practical, requiring minimal specialized equipment, making it easily adoptable by community members.

By involving local residents as community scientists, Alwan Training empowers them to become agents of change, fostering a sense of ownership and responsibility for the conservation and preservation of their natural resources. This grassroots approach ensures that the knowledge and skills imparted through the training are relevant and sustainable, promoting long-term stewardship of the aquatic environments. The program not only enhances the community's capacity to monitor and protect their coastal resources but also strengthens their ability to respond to environmental challenges, contributing to the overall resilience and sustainability of the region's ecosystems. Through Alwan Training, the community is equipped with the tools and knowledge necessary to make informed decisions and take proactive measures in safeguarding their aquatic resources for future generations.


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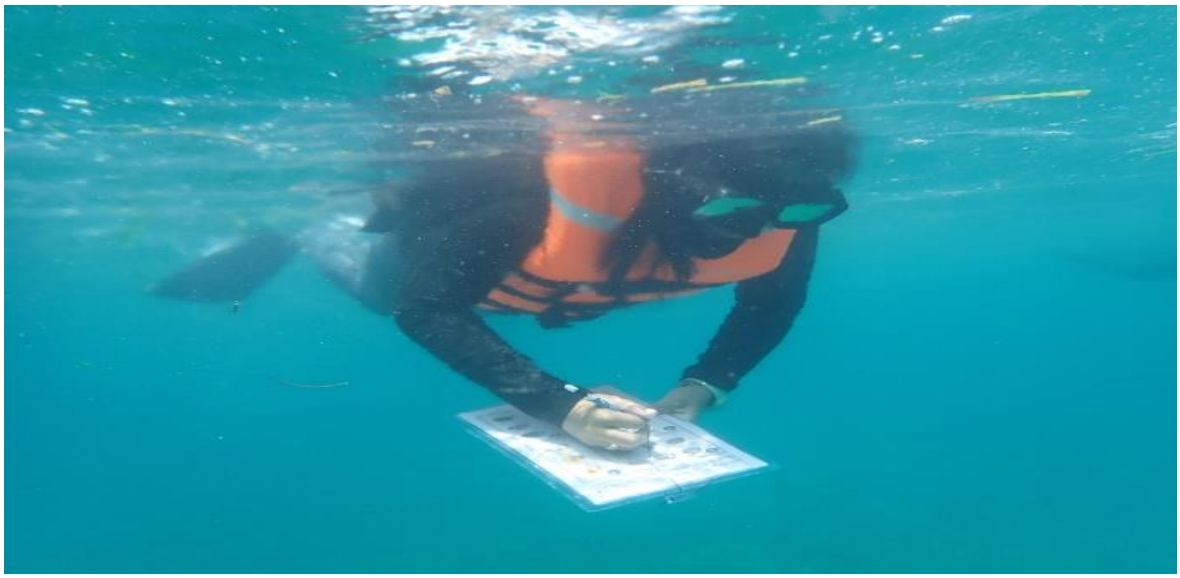

Coastal Clean-up Drive:

Integrating Alwan Training into a CRM system could amplify its impact, streamline processes, and boost participant engagement. It will allow for efficient tracking of community scientists' progress, help in disseminating training materials, and ensure timely communication.


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Phytoplankton in Naujan Lake

Description:

This project aims to develop specialized feed for aquatic organisms in aquaculture, with a particular focus on the nutrition and development of goby larvae. The goal is to ensure optimal growth and health of these larvae, supporting their life cycle from early stages. By producing high-quality, nutrient-rich food, the project seeks to enhance the overall productivity and sustainability of aquaculture practices. This initiative not only addresses the immediate nutritional needs of goby larvae but also contributes to the long-term success and viability of aquaculture operations, ensuring a stable food supply for the aquatic organisms involved

Table 2. Table representation of Dissolve oxygen recorded.

Dissolve Oxygen								
Date	A1	A2	A3	B1	B2	B3	MW Chlorella	Scenedesmus
08/17/23	51.2	44	26.2	31.9	34.2	32		
08/18/23	12.4	26.6	14.5	13.8	11	12.5		
08/19/23	12.2		11.3	11.2	11.8	11.1	15.9	15.3
08/20/23	7.2		8.3	8.6	8.8	8.6	8.5	8.6
08/21/23	5.4		4.6	4	4	3.8	4.3	4.3
08/22/23	1.5		2.4	3	3.3	3.3	3.7	4
08/23/23	4.1		5	3.5	3.1	9.6	10.2	9.3
08/24/23	4.2		3.7	3.8	3.7	3.7	4.4	3.9
08/25/23	5.2		4.3	3.9	3.8	3.6	3.7	4

08/26/23	4.9		4	4	3.6	4.2	4.6	5

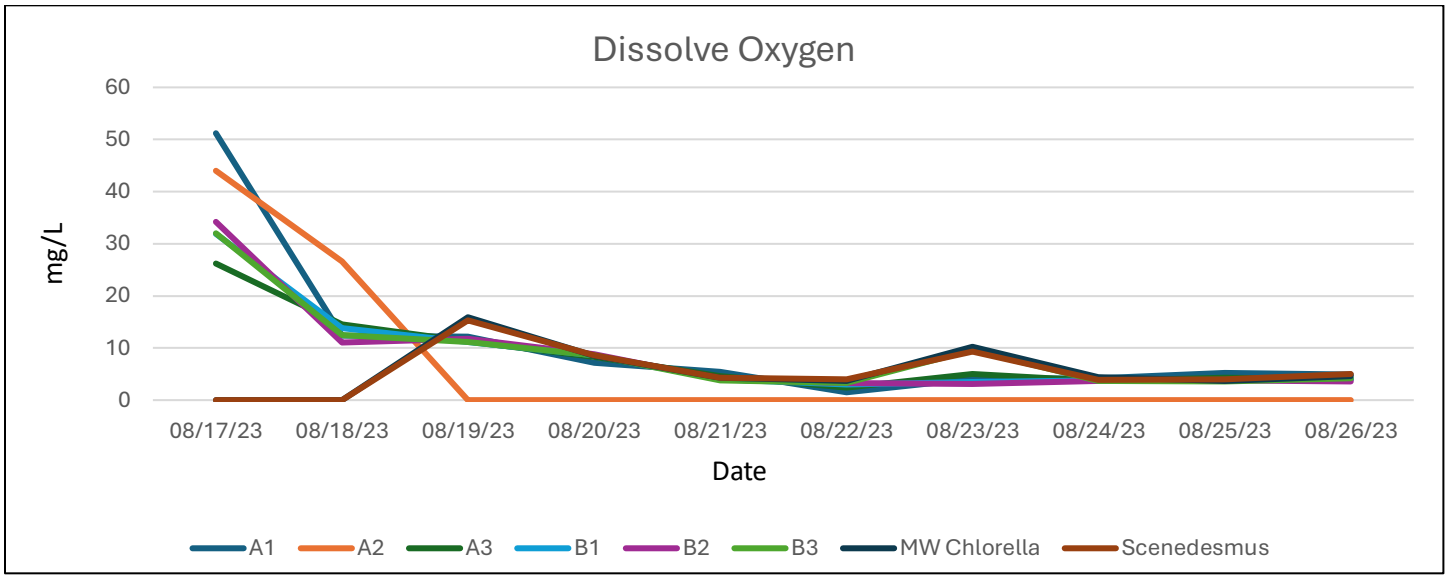


Figure 3. The graphs show the fluctuation of dissolve oxygen in the natural food culture due to daily power interruption.

Table 4. Hydrogen ion concentration recorded in the culture of Chlorella and Scenedesmus.

Date	pH							
	A1	A2	A3	B1	B2	B3	MW Chlorella	Scenedesmus
08/17/23	6.2	5.41	4.99	7.54	6.01	6.14		
08/18/23	5.33	5.36	5.2	7.78	6.2	6.56		
08/19/23	4.49		5.01	7.68	5.92	6.26	7.66	8.37
08/20/23	4.18		5.15	7.63	6.05	6.1	7.7	8.89
08/21/23	4.25		2.17	7.73	5.93	6.01	7.75	8.95
08/22/23	4.01		5.03	7.76	6.02	6.21	7.83	8.83
08/23/23	8.01		8	8.2	8.2	6.02	7.82	8.19
08/24/23	8.32		8.22	8.32	6.33	6.75	8.06	9.19
08/25/23	8.46		8.94	8.26	6.26	6.39	7.96	9.02
08/26/23	8.35		9.2	8.33	6.37	6.6	8	9.16

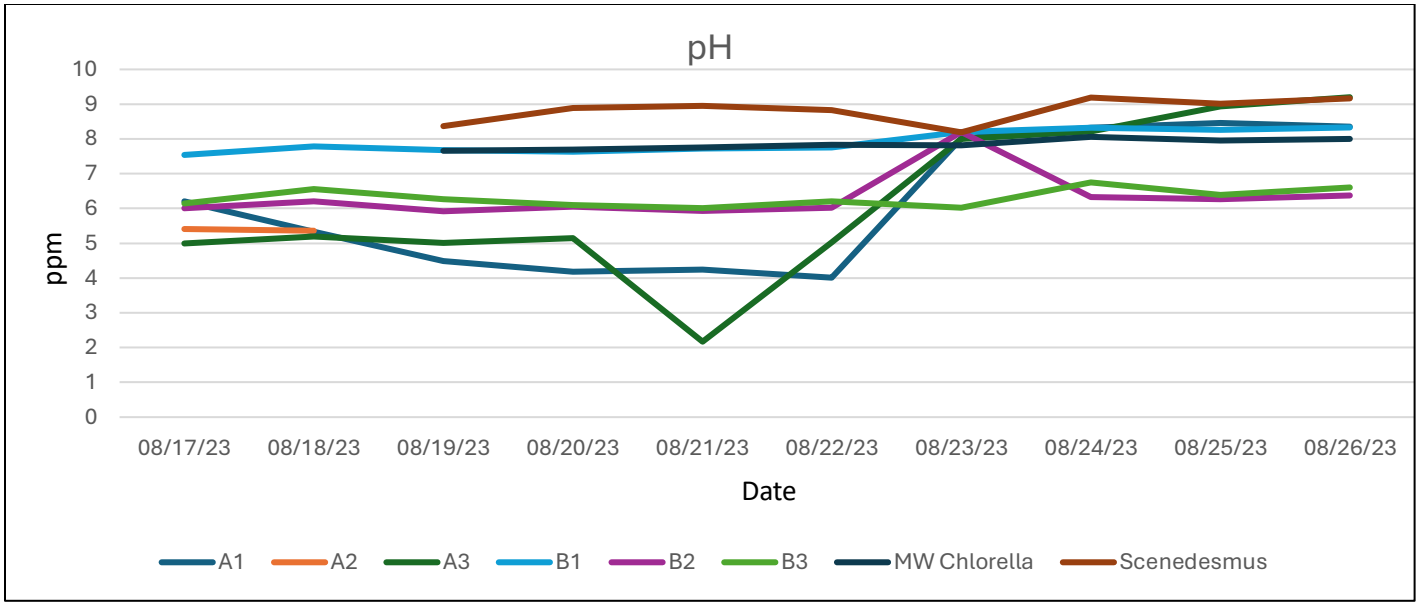


Figure 5. Graph representation of pH reading.

The dissolve oxygen recorded was ranging from 2 mg/L to 52 mg/L. According to *Limnological and Fisheries Survey of Naujan Lake* (Issue 2, pp. 61-74). Results of the chemical analysis of the water showed a considerable amount of dissolved oxygen ranging from 4.9 to 8.5 parts per million; pH ranges from 6.0 to 9.8; carbon dioxide ranges from 0.8 to 2.0 and alkalinity had an annual mean reading of 145.4 to 162 as calcium carbonate, from surface to bottom. Visibility test ranged from 1 to 2.5 meters. Phytoplankton identified consisted of Cyanophyta, Cholorophyta and Chrysophyta while Zooplankton was represented by rotifers, copepods and cyclop.

How to Convert Milligram/liter to Part/million (ppm)

1 mg/L = 1.001142303 part/million (ppm)
1 part/million (ppm) = 0.9988590004 mg/L

Example: convert 15 mg/L to part/million (ppm):
15 mg/L = 15 × 1.001142303 part/million (ppm) = 15.017134545 part/million (ppm)

From: 2 **milligram/liter**
To: 2.002284606 **part/million (ppm)**
Convert Clear
Result: 2 milligram/liter = 2.002284606 part/million (ppm)

From: 52 **milligram/liter**
To: 52.059399756 **part/million (ppm)**
Convert Clear
Result: 52 milligram/liter = 52.059399756 part/million (ppm)

The recorded dissolve oxygen in Natfood Laboratory converted into ppm.

Table 3. Graph representation of temperature recorded in Chlorella and Scenedesmus culture.

Date	Temperature							
	A1	A2	A3	B1	B2	B3	MW Chlorella	Scenedesmus
08/17/23	31.8	31.7	32.2	32.2	32.5	32.3		
08/18/23	31.5	31.7	32.1	32.1	32.5	32.5		
08/19/23	31		31.4	31.4	32	31.58	31.2	31.3
08/20/23	33.1		33.2	33.2	33.7	33.7	33.2	33.2
08/21/23	32.2		32.5	32.5	33.1	33.2	32.7	32.2
08/22/23	31.6		32.1	32.1	32.8	32.3	31.9	31.9
08/23/23	30.7		31	31	31.6	31.2	30.7	30.8
08/24/23	31.3		31.6	31.6	32.1	32.1	31.8	32.1
08/25/23	32.5		32.6	32.6	33.3	33.1	32.8	33.1
08/26/23	29.8		30.2	30.2	31.1	30.8	30.5	30.6

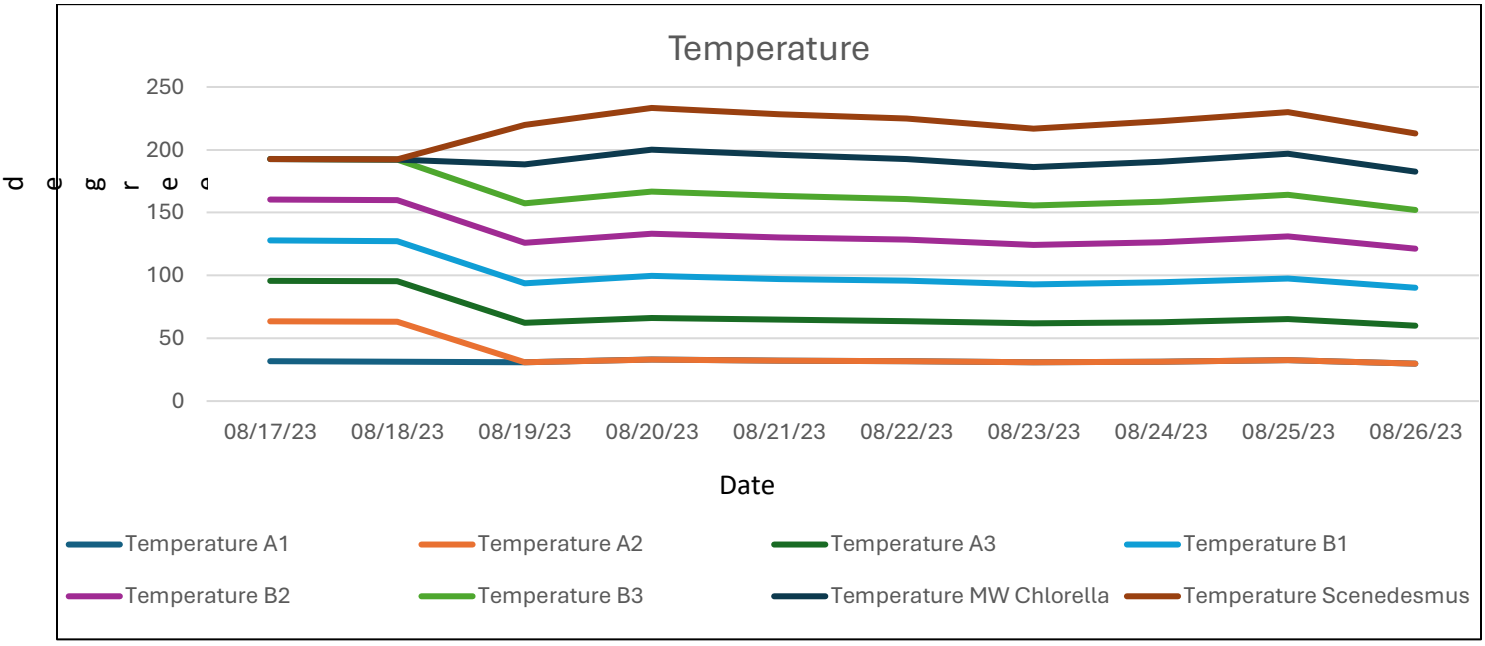
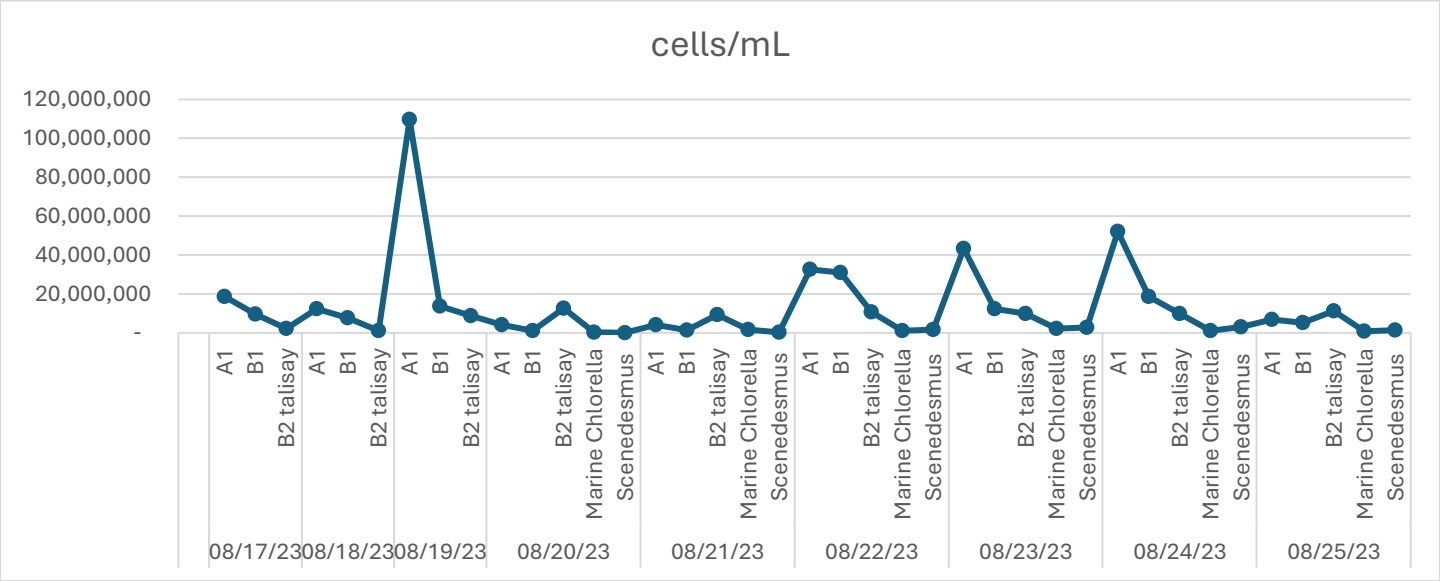


Figure 4. The temperature recorded ranges from 31-33°C.

A study from India, with the title *Evaluating the effect of various environmental factors on the growth of the marine microalgae, Chlorella vulgaris*, in line with their observation, a report has

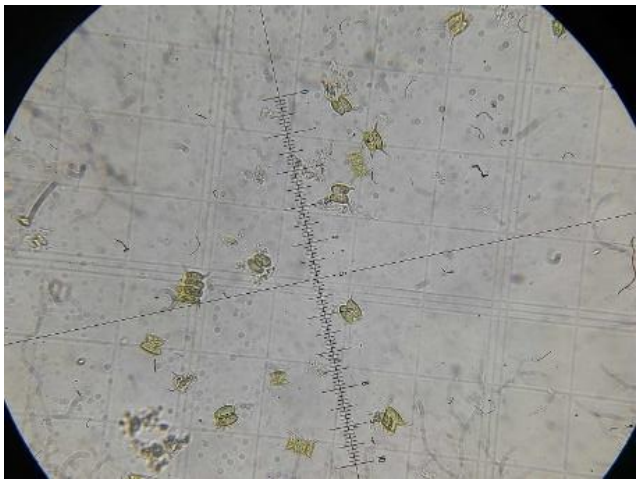


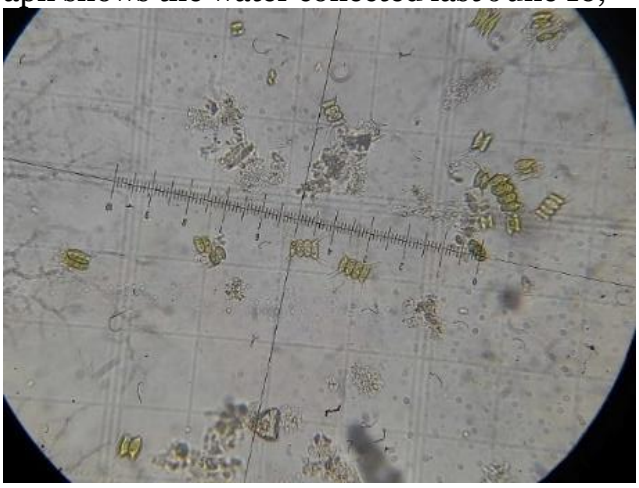
documented that the optimum temperature for freshwater Chlorella is also 25°C (Singh and Singh, 2015). Another study has revealed that C. vulgaris cells were able to tolerate a higher temperature of even 40°C (Sharma et al., 2012).

Table 5. Cell counting of natural food culture in the laboratory. Fresh and Marine water Chlorella and Scenedesmus culture.

		16 squares middle block													
Date	Sample	Upper chamber			Lower Chamber			Total	Upper & Lower 1	Upper & Lower 2	Upper & Lower 3	cells/mL (1)	cells/mL (2)	cells/mL (3)	Average
		1	2	3	1	2	3								
08/17/23	A1	211	2037	5417	208	1,441	1835	11,149	419	3,478	7,252	2,095,000	17,390,000	36,260,000	18,581,667
	B1	852	669	835	952	739	1685	5,732	1,804	1,408	2,520	9,020,000	7,040,000	12,600,000	9,553,333
	B2 talisay	303	181	240	289	162	95	1,270	592	343	335	2,960,000	1,715,000	1,675,000	2,116,667
08/18/23	A1	1136	848	1840	1104	992	1568	7,488	2,240	1,840	3,408	11,200,000	9,200,000	17,040,000	12,480,000
	B1	1024	352	1023	768	975	464	4,606	1,792	1,327	1,487	8,960,000	6,635,000	7,435,000	7,676,667
	B2 talisay	116	73	157	87	111	182	726	203	184	339	1,015,000	920,000	1,695,000	1,210,000
08/19/23	A1	54992	1255	1152	5,216	1920	1264	65,799	60,208	3,175	2,416	301,040,000	15,875,000	12,080,000	109,665,000
	B1	795	901	1978	1,661	1284	1699	8,318	2,456	2,185	3,677	12,280,000	10,925,000	18,385,000	13,863,333
	B2 talisay	809	123	1303	1,125	596	1331	5,287	1,934	719	2,634	9,670,000	3,595,000	13,170,000	8,811,667
08/20/23	A1	482	471	340	141	330	767	2,531	623	801	1,107	3,115,000	4,005,000	5,535,000	4,218,333
	B1	101	89	134	63	150	116	653	164	239	250	820,000	1,195,000	1,250,000	1,088,333
	B2 talisay	1176	1345	1201	1848	1151	909	7,630	3,024	2,496	2,110	15,120,000	12,480,000	10,550,000	12,716,667
	Marine Chlorella	12	20	19	71	52	69	243	83	72	88	415,000	360,000	440,000	405,000
	Scenedesmus	10	17	15	3	11	8	64	13	28	23	65,000	140,000	115,000	106,667
08/21/23	A1	223	287	608	369	385	587	2,459	592	672	1,195	2,960,000	3,360,000	5,975,000	4,098,333
	B1	148	131	111	128	136	107	761	276	267	218	1,380,000	1,335,000	1,090,000	1,268,333
	B2 talisay	987	1103	932	852	733	1009	5,616	1,839	1,836	1,941	9,195,000	9,180,000	9,705,000	9,360,000
	Marine Chlorella	204	171	380	114	76	133	1,078	318	247	513	1,590,000	1,235,000	2,565,000	1,796,667
	Scenedesmus	39	14	42	18	21	53	187	57	35	95	285,000	175,000	475,000	311,667
08/22/23	A1	1904	6840	3881	1760	1520	3624	19,529	3,664	8,360	7,505	18,320,000	41,800,000	37,525,000	32,548,333
	B1	1095	1818	3079	3,396	5819	3360	18,567	4,491	7,637	6,439	22,455,000	38,185,000	32,195,000	30,945,000
	B2 talisay	1184	720	901	1,232	1463	924	6,424	2,416	2,183	1,825	12,080,000	10,915,000	9,125,000	10,706,667
	Marine Chlorella	109	203	84	121	97	108	722	230	300	192	1,150,000	1,500,000	960,000	1,203,333
	Scenedesmus	215	184	138	184	163	160	1,044	399	347	298	1,995,000	1,735,000	1,490,000	1,740,000
08/23/23	A1	4103	4464	3833	2736	6605	4288	26,029	6,839	11,069	8,121	34,195,000	55,345,000	40,605,000	43,381,667
	B1	2310	1471	1634	667	856	529	7,467	2,977	2,327	2,163	14,885,000	11,635,000	10,815,000	12,445,000
	B2 talisay	885	989	1333	874	1142	766	5,989	1,759	2,131	2,099	8,795,000	10,655,000	10,495,000	9,981,667
	Marine Chlorella	151	353	415	241	98	116	1,374	392	451	531	1,960,000	2,255,000	2,655,000	2,290,000
	Scenedesmus	131	1145	57	81	120	78	1,612	212	1,265	135	1,060,000	6,325,000	675,000	2,686,667
08/24/23	A1	7168	6180	6393	4230	3215	4105	31,291	11,398	9,395	10,498	56,990,000	46,975,000	52,490,000	52,151,667
	B1	4556	923	765	3601	646	687	11,178	8,157	1,569	1,452	40,785,000	7,845,000	7,260,000	18,630,000
	B2 talisay	1274	883	941	1112	875	813	5,898	2,386	1,758	1,754	11,930,000	8,790,000	8,770,000	9,830,000
	Marine Chlorella	126	78	92	184	101	83	664	310	179	175	1,550,000	895,000	875,000	1,106,667
	Scenedesmus	1171	110	86	177	80	148	1,772	1,348	190	234	6,740,000	950,000	1,170,000	2,953,333
08/25/23	A1	645	541	648	787	593	961	4,175	1,432	1,134	1,609	7,160,000	5,670,000	8,045,000	6,958,333
	B1	456	323	473	851	259	711	3,073	1,307	582	1,184	6,535,000	2,910,000	5,920,000	5,121,667
	B2 talisay	825	944	813	1815	961	1399	6,757	2,640	1,905	2,212	13,200,000	9,525,000	11,060,000	11,261,667
	Marine Chlorella	115	62	81	99	74	84	515	214	136	165	1,070,000	680,000	825,000	858,333
	Scenedesmus	156	163	128	126	103	207	883	282	266	335	1,410,000	1,330,000	1,675,000	1,471,667

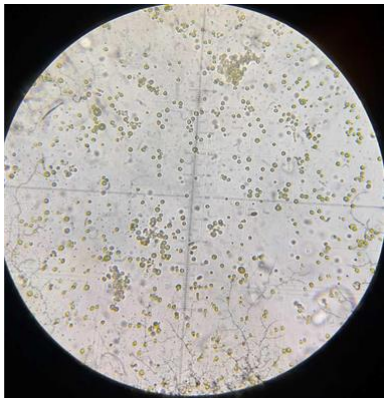
Figure 6. Cell density





The graph shows the water collected last June 16, 2023 labeled A1 has the concentrated chlorella cells with the density of 109,665,000 mg/L while the lowest recorded count was the water collected from Naujan Lake last June 28 mixed with Talisay leaves which labeled B2. August 17 was the 61st day of the culture. The A1 has the concentrated chlorella cells, a new culture was conducted last August 28 labeled as A1-1 which has 20 L of tapwater, 4 L of starter (from A1 carbouy culture) and 20 grams of NPK.

Scenedesmus culture



FW Chlorella culture

Reading of physico-chemical parameters

III. Documentation

Top of the head view of the mouth of Goby larvae

A documentation observed under microscope were in the larvae has rotifer on its mouth.

Gape of mouth under 10x magnification

Egg batch

Date Spawned	Date hatched	Date Collapsed	Egg batch	Estimated eggs	Feed	Concentration	Survival
01/13/2023		01/16/2023	1	10,000	MW Chlorella	500-600 cells/2uL	3 days
01/18/2023		01/21/2023	2	5,000	MW Chlorella	500-600 cells/2uL	3 days
01/20/2023		01/23/2023	3	10,000	MW Chlorella	500-600 cells/2uL	3 days
02/10/2023	02/13/2023	02/13/2023	4	10,000	MW Chlorella	2uL	3 days
02/17/2023		02/20/2023	5	3,000	MW Chlorella	2uL	3 days
03/09/2023	03/12/2023	03/15/2023	6	12,000	MW Chlorella	2uL	6 days
03/22/2023		03/25/2023	7	3,000	MW Chlorella	2uL	3 days
08/16/2023		0/18/2023	8	10,000	FW Chlorella	180,000,000 cells/mL	2 days

On the day of egg collected in the hatchery, green water was mixed with the larvae rearing container for conditioning.

(see files:Egg and Larvae Monitoring inside the 2023 files- Natfood folder)

IV. Phytoplankton species found in green water sample cultured in the Laboratory

- Chlorella -Its shape is spherical with a size that varies from 1 to 10 microns. These microalgae contain, in addition to chlorophyll, a significant number of intracellular proteins, carbohydrates, lipids, vitamin C, β-carotenes and B vitamins (B1, B2, B6 and B12), which is why it is commonly used for the preparation of food supplements, as well as for the production of cosmetics, clinical treatments and even for the detoxification of heavy metals in wastewater. (Conorado-Reyes et al, 2020)

Photo taken at the MBC Natfood Lab

- Scenedesmus - The colony has four cylindrical cells attenuated at poles. Each marginal cell has two stiff and sharp thorns. The cells dimensions are 10×2 µm. (Medard et al, 2018)

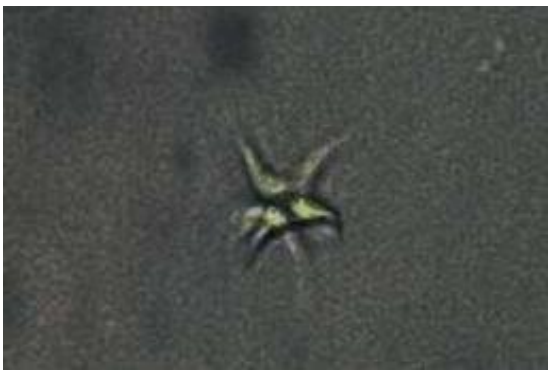


Photo taken at the MBC Natfood Lab

- Palmella - PALMELLOID: Colonial members in which “non – motile” cells remain embedded in an amorphous gelatinous or mucilaginous matrix. In this type neither the number, nor the shape and size of cells is constant. The cells are aggregated in a common mucilaginous envelope. All the cells are independent of each other and fulfil the function of an individual. Most normally flagellate or coccoid unicellular algae may enter (often temporary) palmella stage, a condition where the flagella are lost and the individuals undergo successive vegetative divisions while embedded in a common gelatinous matrix, named after the volvocalean (Chlorophyceae) genus Palmella. This term may be strictly applied to those algae where the cells will readily revert to a motile condition or may be expanded to include all algae where the palmelloid habit is more permanent. Example: Palmella, Microcystis. (Sahoo, D. & Baweja, P., 2015, General Caharacteristics of Algae, India, 3-29)

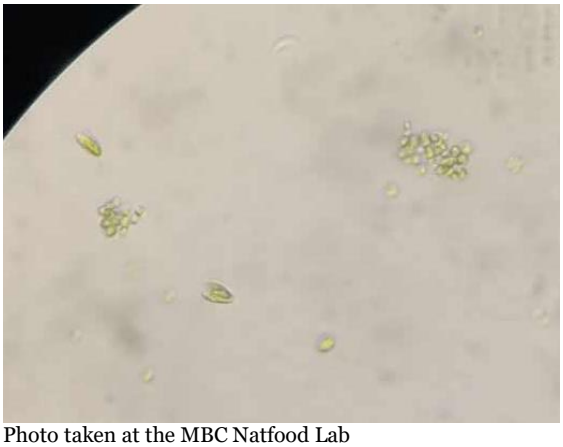


Photo taken at the MBC Natfood Lab

- Euglena - Euglena is a genus made up of single celled eukaryotic organisms ranging in size from 15 to 500 micrometers whose characteristic green color stems from the presence of chloroplasts which enables Euglena to produce energy from photosynthesis. *Euglena gracilis* is a promising alternative supplement because it’s a source of dietary protein, pro vitamins, lipids and the β-1,3-glucan paramylon, in fact, studies have demonstrated that microalgae biomass can be a potential substitute for soybean and fish meal (Aemiro et al. 2016, Gissibl et al. 2019). Most Euglena species have the ability to be both heterotrophic and

autotrophic. Heterotrophs can be feed on bacteria, microscopic eukaryotes, and dissolved organic compounds and autotrophs used their photosynthesizing chloroplasts. An interesting fact is that when light is not available most species of Euglena have the ability to become heterotrophic (Zakryś et al. 2017, Singleton 2018).

- Coelastrum – This was found in Scenedesmus culture in the natural food laboratory.

  The nonmotile greens are ubiquitous and widely distributed in aquatic habitats throughout the North American continent. Aquatic habitats where these algae are found include temporary pools of water, waterfalls, ponds and lakes (e.g., Ankistrodesmus, Coelastrum, and Scenedesmus), rivers (e.g., *Actinastrum* and Dictyosphaerium), slow flowing streams (e.g., Hydrodictyon), marshes, and estuaries. Nonmotile greens are primarily a component of the plankton community (e.g., Desmodesmus, *Golenkinia, Pediastrum*, and *Scenedesmus*). They also may be found growing attached to rocks in lakes (e.g., *Apatococcus* and *Desmococcus*), on leaf surfaces or on other algae (e.g., *Apiocyctis* and *Characium*), and/or on the sediment surface. In the temperate zone, they are most abundant in freshwater ecosystems during the summer, when light and temperature are near their seasonal maximum and nutrients become limiting (e.g., N and P). Reynolds (1984) characterized freshwater plankton from temperate latitudes using trophic status and showed that green algae may be important in phytoplankton dynamics from spring through to autumn. Happey-Wood (1988) reviewed the ecology of freshwater planktonic green algae.

Photo taken at the MBC Natfood Lab