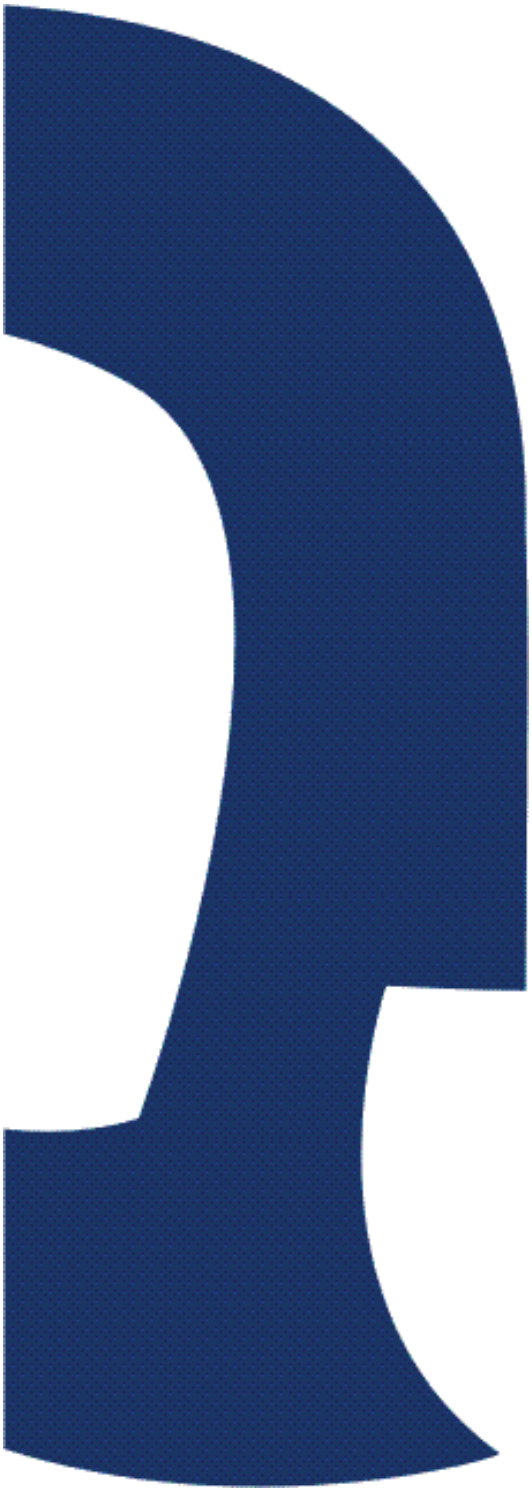

FACULTAD DE CIENCIAS DEL MAR

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Standardization of strobilation in *Phyllorhiza punctata*

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Final degree Project for obtaining the Degree in Marine Sciences

Standardization of strobilation in *Phyllorhiza punctata*

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1. ABSTRACT

In recent years many large aquariums have added a variety of jellyfish to their collections, and as a result the market for these individuals has substantially grown, leading to an increase in their price. This has led to a rising interest in the reproductive biology of such creatures, and in the convenience of studying aquariums in order to reproduce and maintain jellyfish species, so as to achieve autonomy and independence from distributors, therefore reducing facilities and maintenance costs.

Phyllorhiza punctata, (and *Aurelia spp.)* is one of the most sought after species in the market, since it is highly ornate, easy to maintain and safe to keep. The present study has been carried out in Biodomo facilities, in Parque de las Ciencias, in Granada, which are destined to exhibit the rich biodiversity of tropical areas, while preserving the environment, carrying out research and promoting education and environmental awareness.

The objective of this final degree project is to present a protocol for designing and maintaining *P. punctata* facilities, as for the reproduction of the species. To assist its reproduction a strobilation induction experiment has been performed, utilizing an iodine compound with three different concentrations.

KEYWORDS: *Phyllorhiza punctata,* Reproductive cycle, Strobilation induction, Iodine.

2. INTRODUCTION

At present, it is easy to find several species of jellyfish in most aquariums, mainly because of advances in the study of their biological cycles. Recording their living conditions inside tanks and standardizing their reproduction methods has allowed most centres to breed them and feature their beauty and uniqueness to millions of visitors, in addition to enabling thorough research in laboratories.

Phyllorhiza punctata von Lendenfeld, 1884, also known as 'white spotted jellyfish' belongs to the Mastigiidae family, Rhizostomeae order, and was initially described from specimens harvested in Port Jackson, east Australia (Mayer, 1910) (Fig. 1). *P. punctata* has an average diameter of 45-50 cm, with a maximum of 62 cm diameter reported (CABI, 2019).

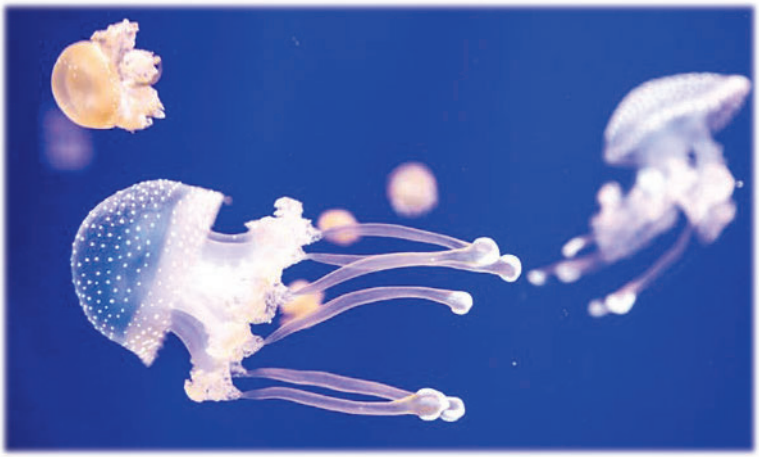

Figure 1. *Phyllorhiza punctata*

This tropical species can usually be found in coastal areas, such as estuaries, lakes and bays in Australia, the Philippines and Japan (Kramp, 1961). Since the middle of the 20th century, reports of its distribution area have increased globally, including Mexico (Ocaña-Luna et al, 2010), Puerto Rico (García, 1990), Brazil (de Souza et al, 2007), Hawaii (DeFelice et al, 2001) and China (Dong et al, 2019), among other places. Isolated specimens of *P. punctata* have been found in the Mediterranean Sea since 1965 along the Israeli coast (Galil et al, 1990; Cevik et al, 2011), while the first established breeding population was documented in 2005 and 2006 in Vlyho Bay, Greece (Abed-Navandi and Kikinger, 2007).

Another factor of importance in the study of *P. punctata* reproductive cycle is cataloguing it as an invading species in several countries (CONABIO, 2020; Martin, 2000; Perry, 2005). Knowledge of the reproductive biology of *P. punctata* can help prevent, eradicate, control, contain and mitigate its invasive behaviour and therefore avoid environmental and economic damage to fishing companies, as they feed on eggs and larvae from fish, crabs and shrimps. They could also obstruct fishing nets and damage fishing ships, resulting in the closure of fishing areas (CABI, 2019).

Several studies confirm that aquaculture and the increase in maritime traffic are some of the main foreign species introduction vectors (DeFelice et al, 2001; Graham et al, 2003). Artificial structures in docks and fish farming can lead to the emergence of exotic species, due to the fact that they become a viable habitat for them to proliferate (Ruiz et al, 2000; Ruesink et al, 2005; Forrest, Gardner and Taylor, 2009).

Breeding jellyfish in captivity, in confined enclosures, is quite complex. Satisfying their nutritional needs and reproducing their habitat is also challenging. Pharmaceutical and cosmetic industries have also taken interest in their potential applications and carried out extensive research on jellyfish, since some derived chemical compounds have

haemolytic, cytotoxic, antimicrobial, cardiovascular, carcinopreventive, inmunostimulatory, antioxidant and insecticidal effects (Ayed et al, 2012; Leone et al, 2013). We can also add that advances in knowledge, considering these measures of 'in situ' breeding can be of great relevance in preserving these species in their natural habitats.

The objective of this study is to describe the breeding of *P. punctate,* taking as a reference the experiments performed in Biodomo facilities, in Parque de las Ciencias, in Granada. In addition to the description of the facilities were the species is kept and its maintenance, feeding is also described, and an experiment has been performed to record the influence of the changes in concentration of the iodine compound we use in the strobilation process.

2.1. Diet

Both corals and jellyfish belong to the cnidarian phylum, therefore they share some characteristics. Species such as *P. punctata* contain symbiotic single-cell dinoflagellates of the *Symbiodinium* genus called zooxanthellae (Raspor, 2016), although populations from northern Gulf of México and eastern Florida in the U.S. lack zooxanthellae (Graham et al, 2003). During photosynthesis, zooxanthellae turn the externally available inorganic carbon and nitrogen into organic forms and transfer some of them to the host, predominantly in the form of carbohydrates (Muscatine and Cernichichiari, 1969), amino-acids (Wang and Douglas, 1998) and fatty acids (Papina et al, 2003). The contribution of photosynthesis' final products to the nutrition of the host in these cases was seen as substantial. The relation between zooxathellae and cnidarian hosts has been more thoroughly studied by betonic cnidarians (Muscatine, 1990) than by mobile cnidarians (Balderston and Claus, 1969; Muscatine and Marian, 1982).

Previous studies claim that certain corals can temporarily change their feeding habits, from autotrophic to heterotrophic modes of nutrition (Anthony and Fabricius, 2000). There is scientific evidence of *P. punctata* jellyfish's ability to use heterotrophic forms to compensate its photosynthetic activity in populations that do not contain zooxathellae (Graham et al, 2003), but no studies have been found on populations that contain them. In the event that these populations are not able to compensate their photosynthetic activity through heterotrophic nutrition, photosynthesis will be essential for the host's metabolic maintenance and zooxanthellae will be a physiological requirement for survival, as it is in the case of *Cassiopea sp,* which is not able to supply its metabolic needs under diminished photosynthetic rates (Mortillaro et al, 2009).

Jellyfish feeding in captivity varies according the species under care, since it is difficult to find living zooplankton and farming it is expensive. In consequence, it is required to provide a diet rich in the necessary nutrients for each species, and in this case, one which complements the input received from symbiosis.

2.2. Temperature and other physico-chemical parameters

As *P. punctata* is a species that is usually found in estuarine environments (Kramp, 1961), it can live in a wide range of salinities and temperatures. Previous studies show how temperature plays an important role in its maintenance, proliferation and strobilation. Despite its tolerance, *P. punctata* is best bred under high temperature and salinity conditions, during spring and summer days when temperature rises and rainfall decreases (Garcia, 1990; Rippingale and Kelly, 1995; Verity et al, 2011; Gueroun et all, 2015; Miranda, 2016). In addition, *P. punctata* can only strobilate from 20°C upwards, with the highest strobilation rates at a range of 23 to 24 °C (Jufri et al, 2001).

2.3. Lighting

Lighting is of vital importance for the maintenance of jellyfish that have zooxanthellae since, as mentioned above, their photosynthetic activity complements their heterotrophic nutrition. In addition, given the importance of symbiotic zooxanthellae for polyp strobilation, the input from lighting in their polyp stage is necessary, as zooxanthellae improve the ephyra's chances of survival (Astorga et al, 2012; Sugiura, 1965)

Zooxanthellae can adapt to different temperature and light intensities by changing their number, size, and the amount of chlorophyll they contain (Muller-Parker, 1985; Klueter et al, 2017). According to the jellyfish management protocol from Asociación de Zoológicos y Acuarios (AZA), the light spectrum used should contain wavelengths from 400-500 microns (AZA, 2013).

2.4. Strobilation induction

In Scyphozoa class, the transformation from polyp to jellyfish occurs as a strobilation, which is the polyps' transversal fission and consequent formation of discoid ephyra (Sukhoputova and Kraus, 2017). There are two different types of strobilation: polydisc, where a polyp produces many ephyrae, and monodisc, where a polyp produces a single ephyra (Helm, 2018). *P. punctata* strobilates as a monodisc (Rippingale and Kelly, 1995).

The strobilation process has been widely studied in several jellyfish species, and inducting factors were recognized. Environmental factors such as temperature, lighting, salinity and the presence of symbiotic zooxanthelae, and even the presence of strobilating polyps can induce strobilation (Rahat and Adar, 1980; Purcell et al, 1999; Xing, Y et al, 2019; Hoffman and Kremer, 1981; Olman and Webb, 1974). In laboratory conditions, the feeding frequency, as well as changes in pH and substances such as thyroxin, iodine and hydrogen peroxide can also induce strobilation (Calder, 1974; Spangenberg, 1971; Spangenberg, 1967; Helm, 2018; Sukhoputova and Kraus, 2017).

The onset of strobilation is identified by the appearance of irregularities in the tentacles' base (Fig. 2d, 2e), along with a change in colour (Jufri et al, 2001), until the ephyra is released (Fig. 2g). As the process continues, tentacles retract and are completely reabsorbed (Fig. 2f).

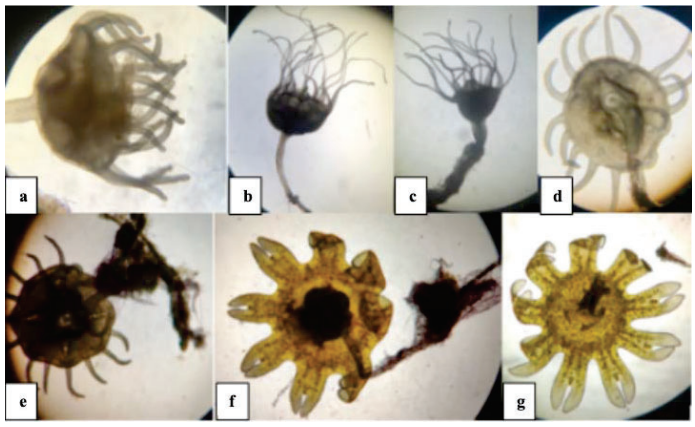

Figure 2. Strobilation process of *P. punctata,* photographed along 7 days until its end. (a-c) Polyp at the onset of strobilation.(d-e) Tentacles retraction and appearance of irregularities in their base.(f) Pulsating movements start (g) Ephyra is completely released.

3. MATERIAL AND METHODS

3.1. Installation

Jellyfish require excellent water quality for their development (Raskoff et al, 2003). In our case, sea water is brought in tank trucks every three days from the coast of Almuñécar to our work centre in Biodomo de Granada. Upon arrival, water density is adjusted to 1026 kg/m^3 from its original 1031 kg/m^3 density. After adjusting it, it is kept in a tank for 24 hours and its temperature is risen to 24°C. Water is mechanically and biologically filtered using perlon wool, sand filters and bioballs, after which it is sterilized with UV light.

For adult jellyfish a 770 litres kreisel tank is used (Fig. 3a). An appropriate water current is crucial for their health (Raskoff et al, 2003), for which this tank is ideal, as it allows the water to flow in a laminar flow, which causes the jellyfish to constantly move and keeps them from damage from bumping against the walls. 30% of the water is renewed every 48 hours to maintain its quality.

Ephyrae are kept in 22 litre kreisel tanks (Fig. 3b), but unlike fully fledged jellyfish, no water flow is used. Instead, constant bubbling causes their movement. Water is not renewed, so food loss in the filtration system is prevented.

Polyps are kept in 7 litre rectangular plastic tanks, with no water flow or aeration. 20% of the water is renewed every 24 hours.

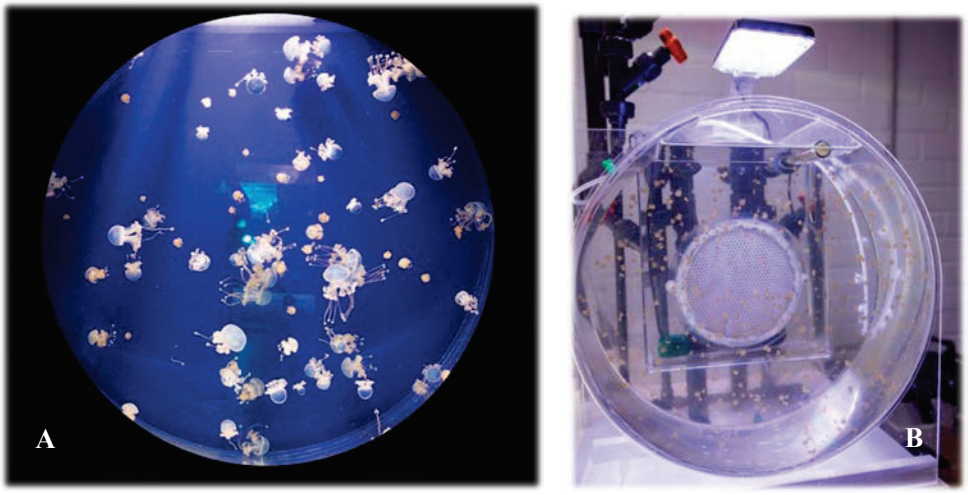

Figure 3. *P. punctata* kreisel tank. (2a) Adult; (2b) Ephyrae and juveniles

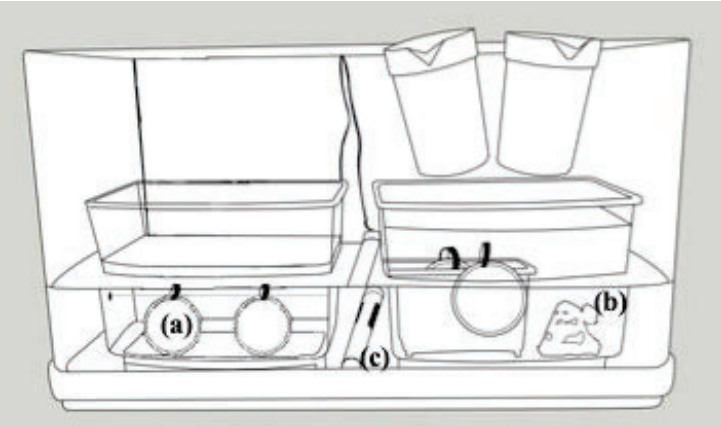

Figure 4. Polyps accommodation. (a) Polyps adhered to petri dish; (b) Polyps on a stone; (c) Water heater

3.2. Diets

For *P. punctata* different types of food are used, which complement the input received from lighting. This depends on their stage of development (Table 1). In adult jellyfish, the main food source is artemia nauplii, enriched with INVE Easy DHA Selco® (rich in n-3 HUFA, DHA and vitamin C) or phytoplankton for 48 hours from hatching (Fig. 5). This is administered twice a day in a 625±50 nauplii/mL concentration.

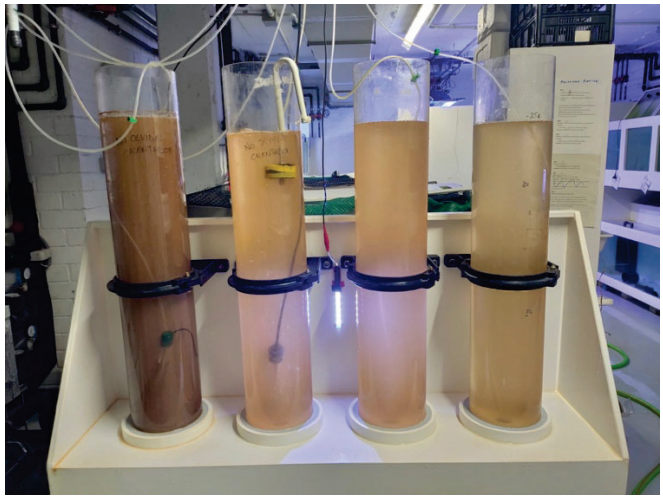

Figure 5. Artemia farming.

Artemia is a genus of brachiopod crustaceans which can be found in brackish water. Their morphology has barely evolved since the Triassic period. Their larvae are known as nauplii. They are used in aquaculture and fishkeeping because of their great nutritional value, mainly as a source of protein for fish, invertebrates, amphibians and even reptiles.

They are commercialized as dehydrated cysts, either decapsulated (in which the chorion or external layer is removed) or non-decapsulated. In our case, they are obtained by a process of decapsulation using innovative Sep-art technology patented by Ocean Nutrition, where cysts are originally magnetized and are decapsulated with a magnetic cannon, which helps separate the capsule from the nauplius (Ocean Nutrition, 2014). Other methods of decapsulation use sodium hypochlorite (NaClO) (Sorgeloos et al, 1977). Even if the second method is valid, NaClO traces may remain and affect the nauplii if they are not correctly neutralized with sodium thiosulfate (Wilmer, 2008).

Table 1. Type, amount and frequency of feeding depending on the stage of development.

PHASE	FOOD TYPE	FREQUENCY	AMOUNT	LIGHTING
Polyp	*Artemia* nauplii 0h	Once a day	*Ad libitum*	Daily 8 h on/16 h off
	Rotifers	Once every 48 hours	*Ad libitum*	
Ephyrae	*Artemia* nauplii 0–24 h	Once a day	100 mL/kreisel	Daily 12 h on/12 h off
Jellyfish	*Artemia* nauplii 48 h	Twice a day	300 mL/kreisel	Daily 12 h on/12 h off
	Minced mussel	Once a day	1g per jellyfish	

In order to improve the nutrition of adult jellyfish and reduce expenses, the last daily intake is complemented with minced mussels (Duarte, unpublished).

Ephyrae are fed once a day with nauplii that has just hatched, adding progressively 24 hour nauplii as they increase in size.

Most researches carried out in other jellyfish species indicate that feeding is a conclusive factor in strobilation (Sukhoputova and Kraus, 2017). Taking these studies as reference, polyps are fed once a day with artemia that has just hatched and with rotifers every 2 days.

3.3. Temperature and other physico-chemical parameters

Established parameters for jellyfish, ephyrae and polyps:

Parameter	Value
Density	1026 kg/m^3
Temperature	24 ºC
pH	8.45
NO_2	0 mg/L
NH_3/NH_4	0 mg/L
NO_3	0-7 mg/L

3.4. Ligting

We used two types of lights. A 150 W, 13000 °K HQI lamp for the adult jellyfish tank; a 70 W, 13000 °K HQI lamp for the ephyrae, and a 13.5 W, 13000 °K LED light for the polyps. The photoperiod used was 12h on/12h off for the adult jellyfish and ephyrae, and 8h on/16h off for the polyps.

4. STROBILATION INDUCTION

In the bibliography there are studies with different results on the polyps' preference for different substrata (Holst and Jarms, 2006; Brewer, 1984), such as plastic, glass or natural shells, which do not provide specific insight. We have therefore opted for sanded petri dishes, to increase porosity and ease adherence.

To obtain polyps, we proceeded to place the dishes in the kreisel tank filtration system. Once the polyps adhered to the dishes, they were moved to their own *ad hoc* prepared containers (Fig. 4). The petri dishes were kept suspended, away from the bottom and vertically oriented, to avoid dirt apposition and for better cleaning.

4.1. Strobilation experiment using an iodine compound of different concentrations

We induced strobilation through addition of Reef Dip™, an elemental iodine compound used in coral, anemone and polyps disinfection. Polyps of *P. punctata* were preconditioned for strobilation in sea water over a period of 4 months.

In order to ensure a high level of accuracy throughout the assessment in which the only study variable was the Reef Dip™ concentration, 15 polyps were used in each replica, obtained from the same petri dish. A different amount of compound was added to each polyp tank (0.5 mL, 2mL and 3.5mL), to assess the differences obtained under the same temperature, salinity, lighting and water volume (7L) conditions, which remained constant in each tank. These values were chosen from data provided by an experienced aquarium, where 2 ml of compound was used in 7 litres of sea water.

A tank was filled with water at 24 °C, where the 9 replicas were placed, so as to obtain a constant and equally-distributed temperature (Fig. 6). A light meter was used to ensure that the lighting levels were equal in all cases (2500 lux) (Fig. 7). A litre of water was renewed daily, adding the proportional amount of compound to each experiment. Polyps were fed with the same amount of artemia nauplii in each replica. The study was tripled for each concentration, taking a control study as a reference.

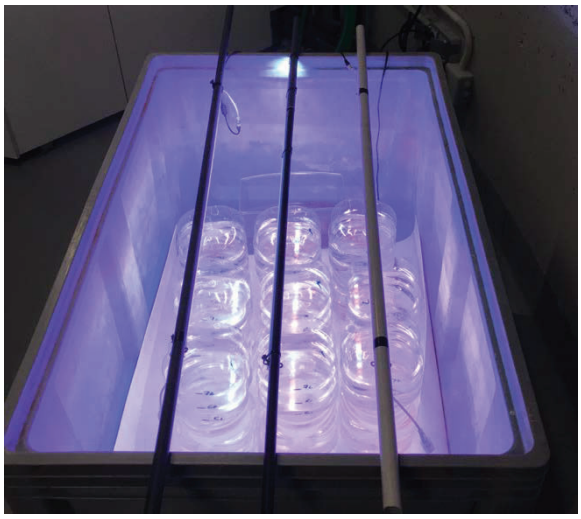

Figure 6. Installation for the polyps strobilation.

Figure 7. Luxmeter.

5. RESULTS

The quantitative range of activity of the iodine compound Reef Dip™ was determined through experimental groups of 15 organisms, at dilutions of 70 µL/L, 280 µL/L and 500 µL/L. In table 2 the number of ephyrae obtained in each concentration of Reef Dip™ is shown, as well as the time for the strobilation onset and ending. On the basis of constant parameters we can observe that the strobilation rate for the three concentrations is the same, except in sample C3, in which only 10 polyps underwent strobilation. However, the time for the strobilation onset and ending varies depending on the compound concentration.

Table 2. Strobilation induction in 9 groups of 15 polyps at different concentrations of Reef DipTM.

ReeF Dip (µL/L)		NUMBER OF POLYPS	NUMBER OF EPHYRAE	STROBILATION ONSET	STROBILATION END
70 µL/L	A1	15	15	5º day	8º day
	A2	15	15	5º day	7º day
	A3	15	15	5º day	9º day
280 µL/L	B1	15	15	6º day	13º day
	B2	15	15	5º day	13º day
	B3	15	15	6º day	12º day
500 µL/L	C1	15	15	7º day	13º day
	C2	15	15	6º day	11º day
	C3	15	10	6º day	14º day

There was an evident variation in the results, in the case of the higher concentration, where one of the replicas didn't reach the expected values, for which two graphs are shown (Fig. 8a and 8b). In the first graph the value obtained from replica C3 was included in the concentration average of 280 µL/L, and in the second it was not included, as it was likely that this result was incorrect and that some unidentified factor influenced its value. In figure 8b we can observe that in studies carried out at a concentration of 70 µL/L of Reef Dip™, polyps complete the strobilation cycle more quickly. At a concentration of 280 µL/L of Reef Dip™, the process of strobilation begins practically at the same time, but advances gradually, taking longer to end. However, in studies at higher concentrations (500 µL/L) the strobilation process advances exponentially, ending a little earlier than in the case mentioned before.

The data gathered from all the experiments was grouped by concentration and was represented as the average strobilation percentage for each point in time. Strobilation did not occur in the control study during the experiment.

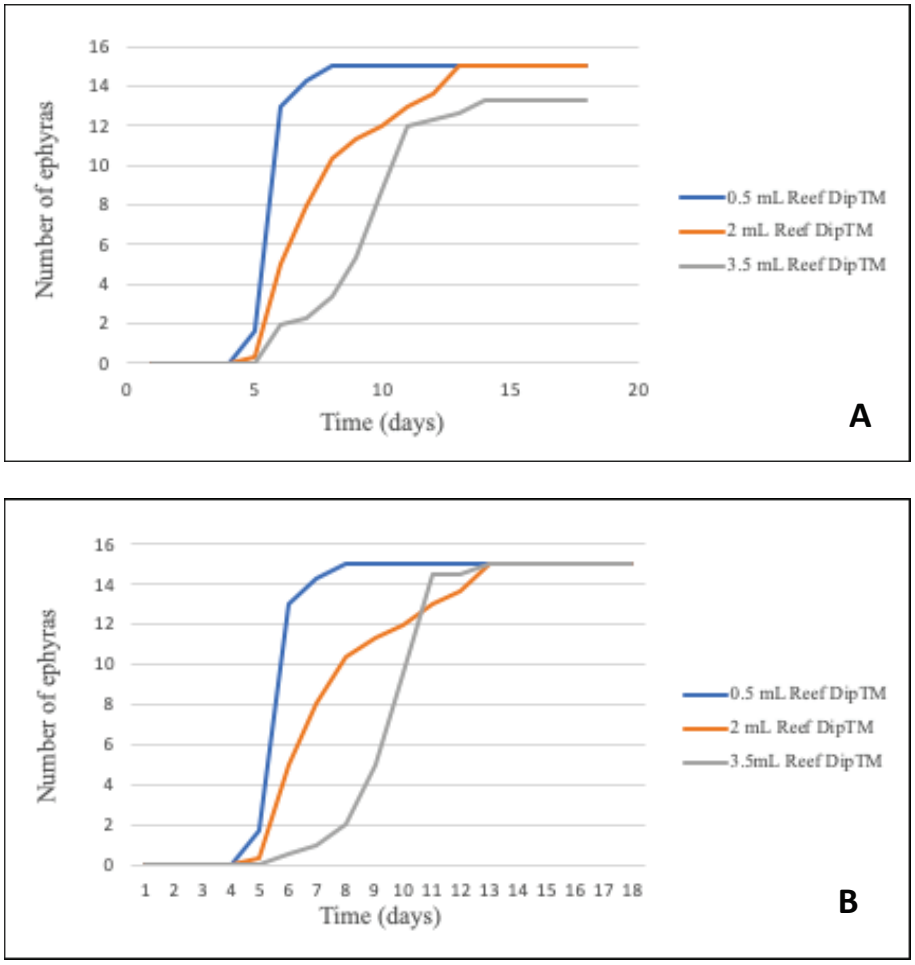

Figure 8. Strobilation time as a function of Reef Dip^{TM} concentration. Values are averages of all the replicas for each iodine concentration. (A) Results using all of the data obtained. (B) Results discarding data from replica C3.

Figure 9 shows an increase in strobilation up to 25%, 50%, 75% and 100%, with an increase in Reef Dip^{TM} concentration. Each bar represents the average of the times the selected strobilation rate for each iodine compound concentration was reached. Standard deviation was included.

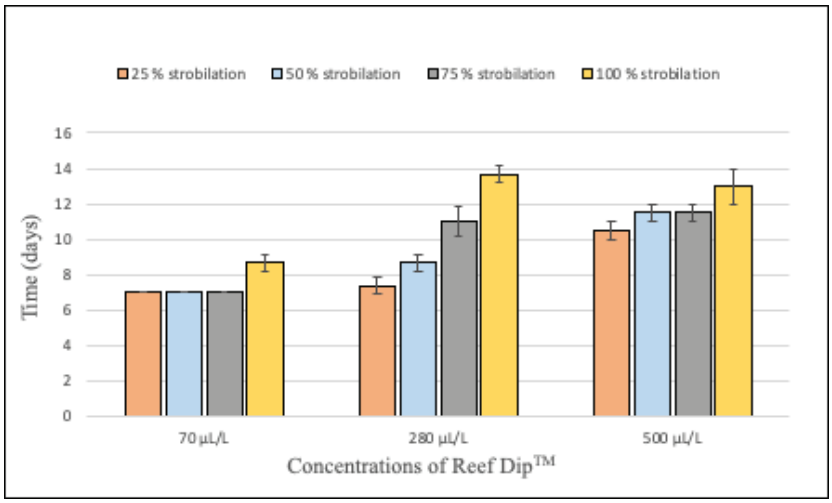

Figure 9. Time used to reach 25%, 50% 75% and 100% of strobilation as a function of Reef Dip™ concentration.

6. DISCUSSION

Thanks to the studies of the environmental factors that interfere in polyps' strobilation in other jellyfish species we currently know that iodine is a key factor (Spangenberg, 1967). Paspaleff (1938) y Spangenberg (1967) proved that *Aurelia aurita* polyps did not strobilate in 'synthetic' sea water, subjecting them to several different combinations of temperature, salinity, pH and nutrition; but ephyrae appeared when polyps were exposed to a potassium iodide solution in sea water.

In our study we can see how the addition of the iodine compound to the water acts as a precursor for strobilation. There were only differences in activity as a function of the concentrations used, in terms of the onset and ending times of the strobilation process. It would be interesting to observe in later studies the effect of these different concentrations on the development, growth and survival of polyps and ephyrae, since schedule limitations during confinement have limited the scope of the experiment with post-strobilation monitoring of the specimens.

In the same way, it would be convenient to study if the polyps induced through this compound lose their ability to answer to this stimulus over a few months, as Spangenberg (1967) claims happens with *Aurelia* polyps when temperature is used as the inducting stimulus for stobilation.

According to Spangenberg's work (1971), polyps present a higher inorganic iodine concentration in the segmented part of the strobilating polyp. This accumulated iodine can be released to the environment when strobilation ends, which could explain why the

presence of strobilating polyps induces the strobilation of the rest (Olmon and Webb, 1974).

As per previous studies in which iodine was used as a precursor for strobilation (Spangenberg, 1971), we have observed that *A. aurita* polyps take from 11 to 14 days to complete 100% of the strobilation process, while in our case, depending on the compound concentration, 100% of the process was completed between the 7^{th} and the 13^{th} day. Spangenberg uses dilutions of I_2 at $1{:}10^7$ and of KI y I^2 at $1{:}10^8$. The iodine concentrations we refer to in this study are relative values. Having used a commercial product, the absolute value of which we do not know since its information was restricted by its manufacturer, has impeded us from comparing our results with those obtained in the reference material.

There are still many questions unanswered about the life cycle and biochemical processes implicated in strobilation, since we lack basic information about many species. Obtaining such information will not only improve our understanding and enable us to hold them in captivity, but can also prove to be of great environmental value to understand the precursor processes for strobilation, which can be used to control jellyfish '*blooms*'.

7. CONCLUSION

This study of the standardization of strobilation of the *P. punctata* jellyfish has led to the following conclusions:

1. Standardized conditions of temperature, lighting, diet, tank characteristics and physicochemical parameters of the water used, previously described and put into practice in Biodomo, in Granada, have proven to be very adequate for the successful breeding of *P. punctata,* since its incorporation to the center's collection in 2018.
2. Multiple ecological signals can trigger the onset of strobilation, from environmental factors such as temperature, lighting, salinity, the presence of symbiotic zooxathellae and the presence of strobilating polyps, to laboratory induced factors like the addition of iodine to the water.
3. The iodine compound used in concentrations of 70 µL/L, 280 µL/L y 500 µL/L is equally effective to trigger strobilation, even if it differs in terms of onset and ending times.
4. In later studies, it would be interesting to observe the effects of using different concentrations of Reef Dip™ for the development, growth and survival of polyps and ephyrae, as well as their tolerance to such concentrations.

5. Having a sound knowledge of the reproductive biology of *P. punctata* is of great social, economic and ecological interest since, in addition to enabling an aquarium's maintenance, it can help prevent, eradicate, control, contain and mitigate their invasive behaviour. Also, *P. punctata,* like some other species that have pharmaceutical and cosmetic applications, could prove to be of great interest to these industries. This knowledge can lead to the development of powerful antagonists involved in the inhibition of strobilation, which can be used to control jellyfish '*blooms*'.

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9. APPENDIX

APPEDIX A

PROTOCOLO CULTIVO ARTEMIA. CONDICIONES ÓPTIMAS DE ECLOSIÓN.

1. Clean the tank using detergent, rinse and next desinfect using hypochlorite solution.
2. Use filtered seawater. Salinity:25-30 ppt; pH:8.0-8.5
3. Incubate between 2 and 3 g of cyst per liter of water. Apply vigorous aeration from the tank bottom.
4. Maintain water temperatura at 28ºC to 30ºC. Do not exceed 30ºC, optimal hatching temperature 29ºC. Provide continuous artificial or natural light (min. 2,000 lux at the water surface).
5. Once hatching is completed run the tank contents through the SEPARATOR or CysTM, collecting the outflowing *Artemia* in a submerged net.
6. After 24 hours of hatching harvest and rinse the nauplii

APPENDIX B

PLANTILLA PARA TOMA DE DATOS

DÍA 1	A1	A2	A3	B1	B2	B3	C1	C2	C3	0*
Densidad	1026 kg/m³	1026	1026	1026	1026	1026	1026	1026	1026	1026
Nº éfiras	0	0	0	0	0	0	0	0	0	0
pH	8,45	8,45	8,45	8,45	8,45	8,45	8,45	8,45	8,45	8,45
Temp.	24 °C	24°C	24 °C	24 °C	24 °C	24 °C	24 °C	24 °C	24 °C	24 °C
NO_2	0	0	0	0	0	0	0	0	0	0
NH_3/NH_4	0	0	0	0	0	0	0	0	0	0
NO_3	0	0	0	0	0	0	0	0	0	0

DÍA 2	A1	A2	A3	B1	B2	B3	C1	C2	C3	0*
Densidad	1026	1026	1026	1026	1026	1026	1026	1026	1026	1026
Nº éfiras	0	0	0	0	0	0	0	0	0	0
pH	8,45	8,45	8,45	8,45	8,45	8,45	8,45	8,45	8,45	8,45
Temp.	24°C	24°C	24°C	24°C	24°C	24°C	24°C	24°C	24°C	24°C
NO_2	0	0	0	0	0	0	0	0	0	0
NH_3/NH_4	0	0	0	0	0	0	0	0	0	0
NO_3	0	0	0	0	0	0	0	0	0	0

DÍA 3	A1	A2	A3	B1	B2	B3	C1	C2	C3	0*
Densidad	1026	1026	1026	1026	1026	1026	1026	1026	1026	1026
Nº éfiras	0	0	0	0	0	0	0	0	0	0
pH	8,45	8,45	8,45	8,45	8,45	8,45	8,45	8,45	8,45	8,45
Temp.	24°C	24°C	24°C	24°C	24°C	24°C	24°C	24°C	24°C	24°C
NO_2	0	0	0	0	0	0	0	0	0	0
NH_3/NH_4	0	0	0	0	0	0	0	0	0	0
NO_3	0-7	0-7	0-7	0-7	0-7	0-7	0-7	0-7	0-7	0-7

DÍA 4	A1	A2	A3	B1	B2	B3	C1	C2	C3	0*
Densidad	1026	1026	1026	1026	1026	1026	1026	1026	1026	1026
Nº éfiras	0	0	0	0	0	0	0	0	0	0
pH	8,45	8,45	8,45	8,45	8,45	8,45	8,45	8,45	8,45	8,45
Temp.	24°C	24°C	24°C	24°C	24°C	24°C	24°C	24°C	24°C	24°C
NO_2	0	0	0	0	0	0	0	0	0	0
NH_3/NH_4	0	0	0	0	0	0	0	0	0	0
NO_3	0-7	0-7	0-7	0-7	0-7	0-7	0-7	0-7	0-7	0-7

DÍA 5	A1	A2	A3	B1	B2	B3	C1	C2	C3	0*
Densidad	1026	1026	1026	1026	1026	1026	1026	1026	1026	1026
Nº éfiras	1	2	2	0	1	0	0	0	0	0
pH	8,45	8,45	8,45	8,45	8,45	8,45	8,45	8,45	8,45	8,45
Temp.	24°C	24°C	24°C	24°C	24°C	24°C	24°C	24°C	24°C	24°C
NO_2	0	0	0	0	0	0	0	0	0	0
NH_3/NH_4	0	0	0	0	0	0	0	0	0	0
NO_3	0-7	0-7	0-7	0-7	0-7	0-7	0-7	0-7	0-7	0-7

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DÍA 6	A1	A2	A3	B1	B2	B3	C1	C2	C3	0*
Densidad	1026	1026	1026	1026	1026	1026	1026	1026	1026	1026
Nº éfiras	13	13	13	2	7	6	0	1	5	0
pH	8,45	8,45	8,45	8,45	8,45	8,45	8,45	8,45	8,45	8,45
Temp.	24ºC	24ºC	24ºC	24ºC	24ºC	24ºC	24ºC	24ºC	24ºC	24ºC
NO_2	0	0	0	0	0	0	0	0	0	0
NH_3/NH_4	0	0	0	0	0	0	0	0	0	0
NO_3	0-7	0-7	0-7	0-7	0-7	0-7	0-7	0-7	0-7	0-7

DÍA 7	A1	A2	A3	B1	B2	B3	C1	C2	C3	0*
Densidad	1026	1026	1026	1026	1026	1026	1026	1026	1026	1026
Nº éfiras	14	15	14	6	7	11	1	1	5	
pH	8,45	8,45	8,45	8,45	8,45	8,45	8,45	8,45	8,45	8,45
Temp.	24ºC	24ºC	24ºC	24ºC	24ºC	24ºC	24ºC	24ºC	24ºC	24ºC
NO_2	0	0	0	0	0	0	0	0	0	0
NH_3/NH_4	0	0	0	0	0	0	0	0	0	0
NO_3	0-7	0-7	0-7	0-7	0-7	0-7	0-7	0-7	0-7	0-7

DÍA 8	A1	A2	A3	B1	B2	B3	C1	C2	C3	0*
Densidad	1026	1026	1026	1026	1026	1026	1026	1026	1026	1026
Nº éfiras	15	15	15	9	11	11	3	1	6	0
pH	8,45	8,45	8,45	8,45	8,45	8,45	8,45	8,45	8,45	8,45
Temp.	24ºC	24ºC	24ºC	24ºC	24ºC	24ºC	24ºC	24ºC	24ºC	24ºC
NO_2	0	0	0	0	0	0	0	0	0	0
NH_3/NH_4	0	0	0	0	0	0	0	0	0	0
NO_3	0-7	0-7	0-7	0-7	0-7	0-7	0-7	0-7	0-7	0-7

DÍA 9	A1	A2	A3	B1	B2	B3	C1	C2	C3	0*
Densidad	1026	1026	1026	1026	1026	1026	1026	1026	1026	1026
Nº éfiras	15	15	15	11	11	12	3	7	6	0
pH	8,45	8,45	8,45	8,45	8,45	8,45	8,45	8,45	8,45	8,45
Temp.	24ºC	24ºC	24ºC	24ºC	24ºC	24ºC	24ºC	24ºC	24ºC	24ºC
NO_2	0	0	0	0	0	0	0	0	0	0
NH_3/NH_4	0	0	0	0	0	0	0	0	0	0
NO_3	0-7	0-7	0-7	0-7	0-7	0-7	0-7	0-7	0-7	0-7

DÍA 10	A1	A2	A3	B1	B2	B3	C1	C2	C3	0*
Densidad	1026	1026	1026	1026	1026	1026	1026	1026	1026	1026
Nº éfiras	15	15	15	11	13	12	7	12	7	0
pH	8,45	8,45	8,45	8,45	8,45	8,45	8,45	8,45	8,45	8,45
Temp.	24ºC	24ºC	24ºC	24ºC	24ºC	24ºC	24ºC	24ºC	24ºC	24ºC
NO_2	0	0	0	0	0	0	0	0	0	0
NH_3/NH_4	0	0	0	0	0	0	0	0	0	0
NO_3	0-7	0-7	0-7	0-7	0-7	0-7	0-7	0-7	0-7	0-7

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DÍA 11	A1	A2	A3	B1	B2	B3	C1	C2	C3	0*
Densidad	1026	1026	1026	1026	1026	1026	1026	1026	1026	1026
Nº éfiras	15	15	15	13	13	13	14	15	7	0
pH	8,45	8,45	8,45	8,45	8,45	8,45	8,45	8,45	8,45	8,45
Temp.	24ºc	24ºc	24ºc	24ºc	24ºc	24ºc	24ºc	24ºc	24ºc	24ºc
NO_2	0	0	0	0	0	0	0	0	0	0
NH_3/NH_4	0	0	0	0	0	0	0	0	0	0
NO_3	0-7	0-7	0-7	0-7	0-7	0-7	0-7	0-7	0-7	0-7

DÍA 12	A1	A2	A3	B1	B2	B3	C1	C2	C3	0*
Densidad	1026	1026	1026	1026	1026	1026	1026	1026	1026	1026
Nº éfiras	15	15	15	13	13	15	14	15	8	0
pH	8,45	8,45	8,45	8,45	8,45	8,45	8,45	8,45	8,45	8,45
Temp.	24ºc	24ºc	24ºc	24ºc	24ºc	24ºc	24ºc	24ºc	24ºc	24ºc
NO_2	0	0	0	0	0	0	0	0	0	0
NH_3/NH_4	0	0	0	0	0	0	0	0	0	0
NO_3	0-7	0-7	0-7	0-7	0-7	0-7	0-7	0-7	0-7	0-7

DÍA 13	A1	A2	A3	B1	B2	B3	C1	C2	C3	0*
Densidad	1026	1026	1026	1026	1026	1026	1026	1026	1026	1026
Nº éfiras	15	15	15	15	15	15	15	15	8	0
pH	8,45	8,45	8,45	8,45	8,45	8,45	8,45	8,45	8,45	8,45
Temp.	24ºc	24ºc	24ºc	24ºc	24ºc	24ºc	24ºc	24ºc	24ºc	24ºc
NO_2	0	0	0	0	0	0	0	0	0	0
NH_3/NH_4	0	0	0	0	0	0	0	0	0	0
NO_3	0-7	0-7	0-7	0-7	0-7	0-7	0-7	0-7	0-7	0-7

DÍA 14	A1	A2	A3	B1	B2	B3	C1	C2	C3	0*
Densidad	1026	1026	1026	1026	1026	1026	1026	1026	1026	1026
Nº éfiras	15	15	15	15	15	15	15	15	10	0
pH	8,45	8,45	8,45	8,45	8,45	8,45	8,45	8,45	8,45	8,45
Temp.	24ºc	24ºc	24ºc	24ºc	24ºc	24ºc	24ºc	24ºc	24ºc	24ºc
NO_2	0	0	0	0	0	0	0	0	0	0
NH_3/NH_4	0	0	0	0	0	0	0	0	0	0
NO_3	0-7	0-7	0-7	0-7	0-7	0-7	0-7	0-7	0-7	0-7

DÍA 15	A1	A2	A3	B1	B2	B3	C1	C2	C3	0*
Densidad	1026	1026	1026	1026	1026	1026	1026	1026	1026	1026
Nº éfiras	15	15	15	15	15	15	15	15	10	0
pH	8,45	8,45	8,45	8,45	8,45	8,45	8,45	8,45	8,45	8,45
Temp.	24ºc	24ºc	24ºc	24ºc	24ºc	24ºc	24ºc	24ºc	24ºc	24ºc
NO_2	0	0	0	0	0	0	0	0	0	0
NH_3/NH_4	0	0	0	0	0	0	0	0	0	0
NO_3	0-7	0-7	0-7	0-7	0-7	0-7	0-7	0-7	0-7	0-7

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DÍA 16	A1	A2	A3	B1	B2	B3	C1	C2	C3	0*
Densidad	1026	1026	1026	1026	1026	1026	1026	1026	1026	1026
Nº éfiras	15	15	15	15	15	15	15	15	10	0
pH	8,45	8,45	8,45	8,45	8,45	8,45	8,45	8,45	8,45	8,45
Temp.	24ºC	24ºC	24ºC	24ºC	24ºC	24ºC	24ºC	24ºC	24ºC	24ºC
NO_2	0	0	0	0	0	0	0	0	0	0
NH_3/NH_4	0	0	0	0	0	0	0	0	0	0
NO_3	0-7	0-7	0-7	0-7	0-7	0-7	0-7	0-7	0-7	0-7

DÍA 17	A1	A2	A3	B1	B2	B3	C1	C2	C3	0*
Densidad	1026	1026	1026	1026	1026	1026	1026	1026	1026	1026
Nº éfiras	15	15	15	15	15	15	15	15	10	0
pH	8,45	8,45	8,45	8,45	8,45	8,45	8,45	8,45	8,45	8,45
Temp.	24ºC	24ºC	24ºC	24ºC	24ºC	24ºC	24ºC	24ºC	24ºC	24ºC
NO_2	0	0	0	0	0	0	0	0	0	0
NH_3/NH_4	0	0	0	0	0	0	0	0	0	0
NO_3	0-7	0-7	0-7	0-7	0-7	0-7	0-7	0-7	0-7	0-7

DÍA 18	A1	A2	A3	B1	B2	B3	C1	C2	C3	0*
Densidad	1026	1026	1026	1026	1026	1026	1026	1026	1026	1026
Nº éfiras	15	15	15	15	15	15	15	15	10	0
pH	8,45	8,45	8,45	8,45	8,45	8,45	8,45	8,45	8,45	8,45
Temp.	24ºC	24ºC	24ºC	24ºC	24ºC	24ºC	24ºC	24ºC	24ºC	24ºC
NO_2	0	0	0	0	0	0	0	0	0	0
NH_3/NH_4	0	0	0	0	0	0	0	0	0	0
NO_3	0-7	0-7	0-7	0-7	0-7	0-7	0-7	0-7	0-7	0-7

0* ESTUDIO CONTROL

Ángela Castrillón Rodríguez

Valoración personal de la realización del TFT

El nivel de integración e implicación en el Biodomo ha sido muy bueno desde el primer día. Hace 4 años me brindaron la oportunidad de formar parte del equipo aun careciendo de la titulación. Me enseñaron mucho acerca del manejo y cuidado de todas las especies que se encuentran en él. Además, me han dado la oportunidad de hacer uso de los recursos del centro para poder finalizar mis estudios por lo que estoy infinitamente agradecida.

En general, en la realización del Trabajo de fin de Grado todo han sido aspectos positivos, tanto el aprendizaje como el desarrollo del trabajo. Con la llegada del Covid la parte última del trabajo se vió afectada puesto que sólo podía acceder al centro los días y las horas que por protocolo me tocaba asistir a mi puesto de trabajo. De no ser así, me hubiese gustado haber podido recoger más datos y haber seguido con la investigación para aportar más resultados del experimento y por tanto más discusiones.

Agradezco mucho la ayuda de mis tutores por los consejos dados y lo que me han ayudado tanto a enfocar el TFG como a desarrollar el contenido y la metodología, cuestiones que no se aprenden en la carrera y que son tan importantes en el desarrollo profesional. Gracias también al Parque de las Ciencias, a Rainforest y a mis compañeros por permitir la realización del estudio ya que este TFG le ha dado un valor especial a mi trabajo rutinario como 'acuarista' que me ha aportado mucho en lo personal y en lo profesional abriéndome puertas para hacer un seguimiento de muchas de las actividades y eventualmente, a plantearme la posibilidad de escribir algún artículo o desarrollar algún estudio con mayor profundidad aprovechando la oportunidad de mi trabajo con multitud de especies de interés científico, ecológico y económico