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MICROBIOLOGICAL DIAGNOSIS OF SEA TURTLES IN THE COASTS OF BAJA CALIFORNIA SUR, MEXICO

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"Sabiduría como meta, patria como destino"

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Index

ABSTRACT

Currently, five of the seven species of sea turtles inhabit the coasts of Baja California Sur, as it is considered one of the areas with the greatest biodiversity in the world. These reptiles, belonging to the Testudines order, unify a healthy ecosystem providing the perfect balance in the first links of the food web, thanks to their robustness and their high vulnerability to disturbances in the ocean. Being cosmopolitan species and carrying out long migrations, they are exposed to various pathogens, such as bacteria or parasites, which alter their immunologic system, causing their death.

This study is based on the assessment of the microbial biota of the oral, cloacal and, in some cases, eye areas, and on the observation of the morphology of the different blood cells. These analyses were carried out by means of a morphological and biochemical classification of the different haematological and bacteriological samples obtained. The collection took place between August and November 2016 and 2017, specifically from Reserva de la Biosfera El Vizcaíno and Golfo de Ulloa.

Due to their role in ecology and the scarcity of existing studies on this subject, it would be helpful to focus on microbiological analysis to improve the fluidity of the characterization of the health status of sea turtles. In order to achieve a complete and effective conservation of this type of reptiles on the Pacific Ocean coasts.

1. INTRODUCTION

The west coast of the state of Baja California Sur (México) has been considered a great privilege, as five of the world's seven species of sea turtles inhabit the area, widely distributed throughout the Pacific Ocean. Several bays and lagoon systems provide an important growth habitat for these reptiles, which is why it is valued as an excellent feeding and nesting area (García-Martínez & Nichols, 2001; Labrada et al., 2010, Reséndiz et al., 2017).

Sea turtles belong to the Reptilia class, specifically to the Testudines order, since they have a series of characteristics that identify them, such as having on the upper part ribs fused to the shell and on the lower part a plastron, giving them the perfect hydrodynamic capacity for excellent swimming. They also have aerial respiration and a scaly body (Frazier, 2001). They are basic elements for conservation, helping to maintain a correct balance in the first links of the food web (Menéndez-Macías, 2015).

Females return to their region of origin to reproduce, a concept known as philopatry (Meylan & Meylan, 2000; Fitz-Simmons et al., 2000; Camacho et al., 2013). They occupy niches in various geographical regions, directly exposing themselves to threats that harm their state of mind (Bolten, 2003).

They are called ancestral species in danger of extinction, due to extrinsic factors, such as human consumption, changes in physical factors (water temperature, habitats, pollutants) or some infectious diseases generated by microorganisms, such as parasites or bacteria, generally (Aguirre et al., 2002). For this reason, it is of vital importance to characterize their most exhaustive physical condition (Parra-Gaxiola, 2012).

Like any living being, these are endowed with a series of bacteria that, under stressful conditions, act in a harmful way to the organism. The immune system is altered, increasing the degree of suffering from any harmful disease caused by an excess of pathogens (Aguirre et al., 2002). *Aeromonas spp., Pseudomonas spp., Staphylococcus aureus, Acinetobacter spp*. are considered opportunistic microorganisms, as they can penetrate damaged or highly damaged tissues, proliferate, and subsequently cause major diseases (Glazebrook & Campbell, 1990; Zavala- Norzagaray et al., 2015).

Bacteria are prokaryotic organisms divided into two levels, according to the type of organization they present, Grampositive and Gramnegative. One of the most commonly used mechanisms for the classification of these is the Gram stain, which provides a preview of the type, since some properties can be correlated with the cell envelope. Grampositive bacteria have a cell wall about 20-80 nm thick, as the outer layer of the cell. However, Gramnegative bacteria have a slightly thinner cell wall, >10 nm, but harbour an additional external membrane with a large number of pores and appendages. Such differences in the cell envelope give it different properties, in particular the responses to extrinsic factors such as heat, UV radiation and antibiotics (Mai-Prochnow et al., 2016). Most of the adjacent damage and even mortality in living organisms is related to the presence of Gramnegative bacteria, such as *Pseudomonas spp.* (Munson & Evans, 2012).

One of the most useful tools to know and diagnose the health status of a sea turtle in free life is hematology and blood biochemistry. These clinical studies may facilitate their

management, since they are the first to reflect changes in the organism, or possible alterations, due to a foreign factor, as well as help to specify a possible systemic bacterial infection (Aguirre & Balazs, 2000; Herbst, 2000). The interpretation of the blood cells morphology, for example eosinophils, lymphocytes, thrombocytes or erythrocytes, is considered to be the first factor to be analyzed in the hematological field, since in order to follow a pathological identification guide the condition of these cells must be taken into account (Casal & Orós, 2007; Sykes & Klaphake, 2008).

As for human consumption, coastal inhabitants, in this case in the Pacific, are probably unaware of what contamination or adverse effects, such as pathogenic microorganisms, the turtle meat they eat has been exposed to (Aguilar-González et al., 2014). Species such as *Salmonella, Mycobacterium, Vibrio* and *E. coli* have been classified as harmful to humans (Santoro et al., 2006).

This study is based on the assessment of the state of health of the species that inhabit the coasts of the state of Baja California Sur, focusing on the microbial activity of the cloacal and oral zones and in some cases, on the eye area. The samples were collected between August and November 2016 and 2017, specifically from La Reserva de la Biosfera El Vizcaíno and Golfo de Ulloa.

As a general objective, it is proposed to know the microbial biota of the different species of sea turtles and to determine how the survival of the populations that live around the coasts of the Western Pacific is affected. On the other hand, the specific objectives are: to distinguish the different bacteria that exist in the different areas or sampling zones, to check the genus of bacteria by means of biochemical tests and to differentiate blood cells by means of hematological techniques.

2. DATA AND METHODS

2.1 Geographic situation

The state of Baja California Sur has a length of 900 km and is located at about 28 ° 00 ° N and 22 ° 52' N, and between 109 ° 25 ° W and 115 ° 05' W. In terms of length, it has the longest coast in all of Mexico (approximately 2222 km), including many islands of volcanic origin (Thomson et al., 1979).

Baja California Sur is catalogued as one of the most productive areas in the world. Along the coast, there are some lagoons belonging to the Pacific Ocean, among them: Guerrero Negro, Laguna Ojo de Liebre, Laguna San Ignacio and Bahía Magdalena (Golfo de Ulloa). The different sampling areas are shown below:

Laguna Ojo de Liebre (LOL)

Located northeast of Baja California Sur, on the Pacific coast, extending from 27° 35' to 27° 55' N and 113° 58' to 114° 10' W. This area of irregular and hyperhaline bathymetry is part of La Reserva de la Biosfera El Vizcaino. It has a dry climate, with maximum water temperatures ranging between 20º and 26ºC in summer and minimum temperatures

of 12 to 20ºC in winter. Its waters are shallow, presenting channels with a depth of up to 20 m.
This lagoon is home to a wide variety of species of benthic macroalgae (Águila-Ramírez, 1998).

Laguna Guerrero Negro (LGN)

It is located on the Pacific Ocean coast, between 27º 35'-27º 52' N, 113º 58'-114º 10' W in the north and south of BCS and BC, and is part of the Laguna Ojo de Liebre complex, belonging to La Reserva de la Biosfera El Vizcaino (Figure 1).

It is a macromarsh lagoon with a length of 13 km and a reduced channel that leads to the bay of Vizcaíno. Its waters are practically shallow, as the depth is mainly between 2 and 12 m (Lluch et al., 1993).

Golfo de Ulloa (GU)

Located on the coast of the State of Baja California Sur, approximately between 25º- 27º North latitude and between 112º- 114º West longitude, from Cabo San Lázaro to the south of Punta Abreojos (Lluch-Belda, 2000). It is characterized by a strong influence of seasonal upsurges, becoming an important area with high primary productivity, with a large sum of consumers.
Located between the municipalities of Comondú and Mulegé, with a coastline extension of approximately 325 km (González-Rodríguez et al., 2011).
Sea turtles, specifically *Caretta caretta*, haunt the waters of the Golfo de Ulloa for most of their growth stage, as they frequent the waters of the Golfo de California to feed after nesting in Japan (Seminoff et al., 2006).

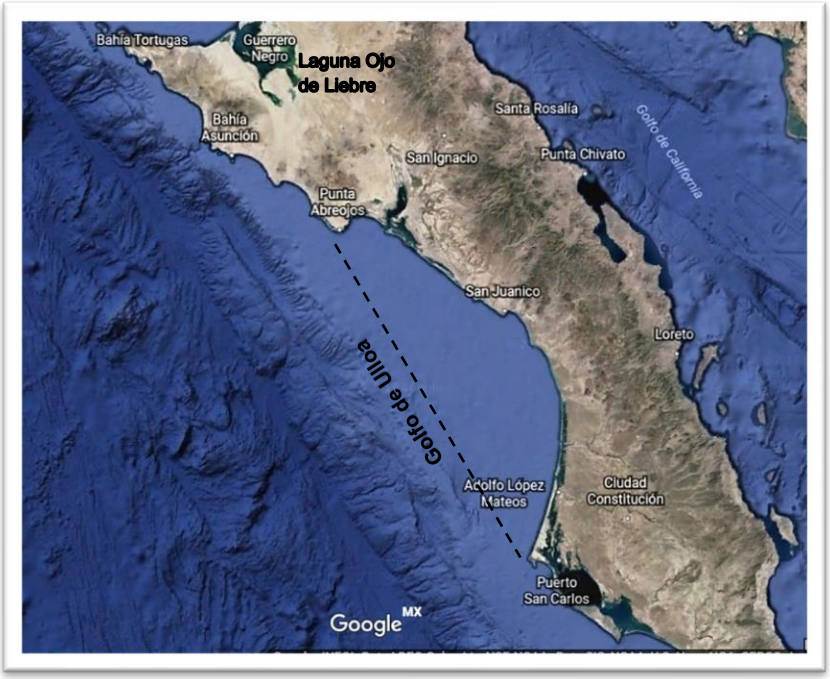

Figure 1. Location of the study area. Laguna Guerro Negro, Laguna Ojo de Liebre and Golfo de Ulloa.

2.2 Methodology

Field work

Monthly monitoring was carried out. Depending on the area of study, different methods were used. In the Golfo de Ulloa, being open sea, it is more difficult to capture the animal, so the monitoring technique called "rodeo" is used. When observing that the turtle is on the surface horizontally thermally regulating, the monitors are thrown into the sea to capture the specimen, before it is immersed again.

The other two methods are netting and encirclement, the first being carried out using a net 100 to 120 m long. The difference is given by the art, one is anchored to some buoys, remaining in that place a margin of time, which is checked every hour, not too much to avoid drowning; while the other, a circle is made in the form of a fence. These two techniques are carried out in Laguna Ojo de Liebre and Laguna Guerrero Negro.

After being captured, the specimen is subjected to a series of morphometric measurements, both straight and curved, and then recorded in the field guide. Measures such as: CCL (long curved shell), ACC (wide curved shell), LRC (long straight shell), ARC (wide straight shell), LP (length of plastron) and LC (length of tail). Curved measurements were made using a tape measure, while straight measurements were made with a calipers. Finally, the weight in kilograms was determined.
In addition, it should be noted that the temperature of the plastron, shell and left groin was taken from each individual. At the end of the characterization of the physical state, both front flippers were marked with Inconel metal plates.

Sterile clinical swabs were used to collect bacteriological samples from the mouth, cloaca and in some cases the eye area. Subsequently, they were deposited in tubes with a gel agar media with and without carbon, where the samples were stored, avoiding possible contamination from the collection to the laboratory.

To carry out this hematological analysis, blood collection needles and tubes from the Vacutainer system were used. Blood collection is performed by finding the cervical venous sinus of the turtle as described by Keller et al. (2006). Later, it was collected in tubes, two per turtle, one of which had anticoagulant (lithium heparin) and the other without anticoagulant. The samples were kept in refrigeration until they were processed in the oceanography laboratory of Universidad Autonoma de Baja California Sur (UABCS).

It should be noted that the collection of skin, shield and esophageal lavage was also carried out.

The collection of all previous samples was allowed, thanks to the OFFICE NUM. SPGA/DGVS/05533/16 and OFFICE NUM. SPGA/DGVS/07287/17.

Laboratory work

A total of 89 samples (swabs) of the species *Caretta caretta* and *Chelonia mydas*, commonly known as Loggerhead turtle and Green turtle respectively, were used for microbiological growth. In a previously sterilized area, the standard media, Blood agar and Dextrose agar were sown with the swabs from the biological cloacal, buccal and eye samples, using the cross-strain method. Later, they were incubated at a temperature of 35ºC for 24 hours. They were checked after 12 hours to see if there was growth, if it was scarce, it was left until 24 hrs. In some media it was left up to 48 hrs because no growth was observed.
After that time, the growth of several colonies began to differ. The colonies were smeared to determine whether they were Grampositive or Gramnegative bacteria. This was followed by a new replanting in specific TCBS agar media (thiosulphate citrate bile sucrose), which is special for Vibrio isolation, MacConkey agar, specific for differentiation of Enterobacteriaceae and several Gramnegative bacilli. The use of lamb's blood agar (COS), among others, to completely isolate them, and thus make the observation under the bacteria microscope easier and more accurate.
Once the colonies were isolated in the selective media, they were smeared again and classified by Gram stain, where they were later identified by means of biochemical tests (sucrose, oxidase, lysine, ornithine, lactose, indol, mannitol, sodium chloride at 0, 1, 6, 7 and 10%), using the API® 20 NE Microbial Identification Kit system, which gave results with a higher accuracy range.

On the other hand, in order to perform the hematological part of the study, smears were taken from the 15 blood samples obtained, which were stained using Giemsa's technique and the Dip Quick technique. Like the bacteriological samples, these were also examined under the microscope to observe the morphology and make a previous classification of the type of blood cells.

3. RESULTS

3.1 Bacteriology

In this study, there were many Grampositive cocobacils and bacilli in the cloacal samples from the different areas cited in the study area, although Grampositive cocobacillus and Gramnegative bacilli were also observed in smaller numbers (Figure 5; Figure 6).
For the evaluation of isolated growth, the morphology was considered, which ranged from irregular to circular; the colour, such as orange, green or even coloured vires of the media, due to the fact that bacteria change the pH of the media. Also, the two types of consistency, membranous and creamy, were evaluated, which, in some samples, acquired pastel colors.
As for the surface, they ranged from smooth to rough, with some elevation in some cases. Specifically, sample No. T53 was one of the samples that was replanted in several selective media, as it had a different morphology and texture than the rest (Table II; Figure 2; Figure 3).
Some media were contaminated, as shown in Figure 4, after the smears were taken, possibly due to poor sterilization of the working area or failure to properly seal the petri dishes, exposing them to transmission.

Table 1. Colony growth in standard culture media of sample T53.

Sterilized swabs	Study area	Specie	Type of bacterial colony
Sewer	GU	*C.caretta*	Colonies with creamy texture, dotted and irregular shapes, with a pale yellow colour. Turn a dark reddish color.
Mouth	GU	*C.caretta*	Uniform colonies and several circular, whitish colognes with a creamy texture. Petal edges and smooth surface. Turn a dark reddish color.

Table 2. Growth of buccal and cloacal swabs of the T53 sample in selective growing media.

Sample	Study area	Buccal swabs	Cloacal swabs	Selective growing media	Type of bacterial colony
			X	Mc Conkey	Low growth of irregular colonies.
		X	X	MSA2 (Saline mannitol agar)	Pointed colonies with a certain elevation. Membranous texture and a rough surface. Presented colored vire
			X	COS (Sheep Blood Agar)	Rough punctiform growth, with small elevations.
T53	GU		X	ASE	Rounded and pointed colonies with a membranous texture
			X	EAM (Eosin Methyl Blue)	Irregular colonies with a creamy texture. Violet color.
			X	TCBS (Thiosulphate citrate bile sucrose)	Growth of irregular colonies with a membranous consistency. It presented colored veins.

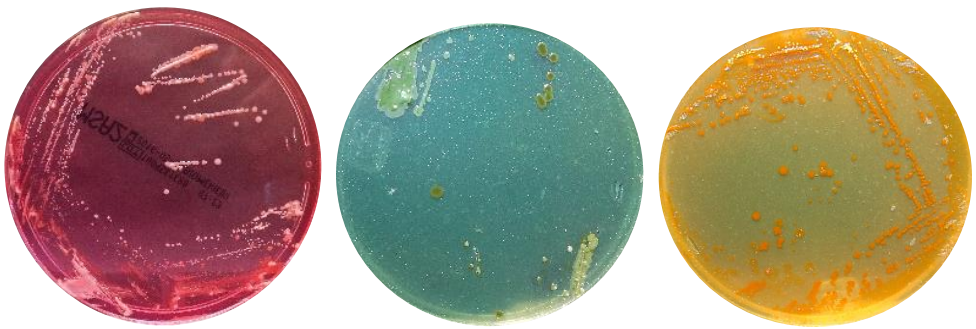

Figure 2. Selective growing media of MSA2, EAM and TCBS from sample T53. Genre *Pseudomonas.*

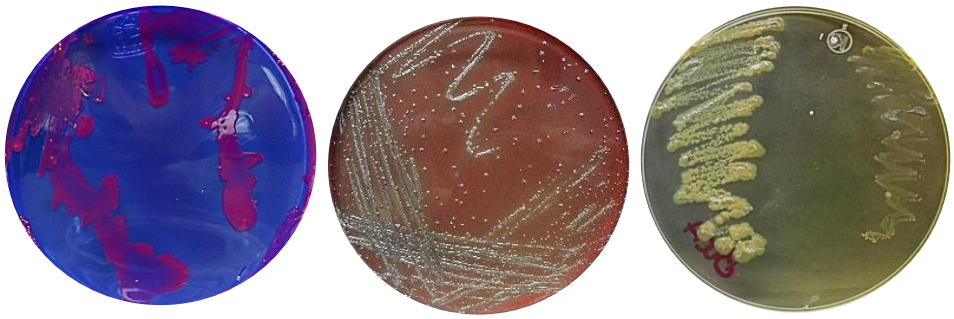

Figure 3. Selective growing media of MSA2, EAM and TCBS from sample T53. Family Bacillaceae.

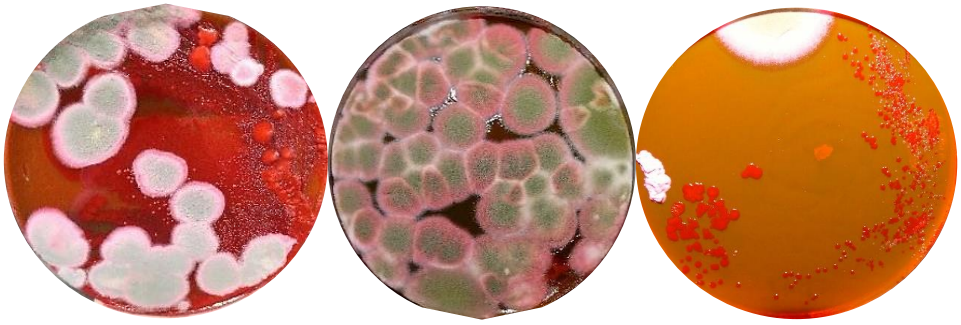

Figure 4. Samples replanted in selective media, contaminated by fungi.

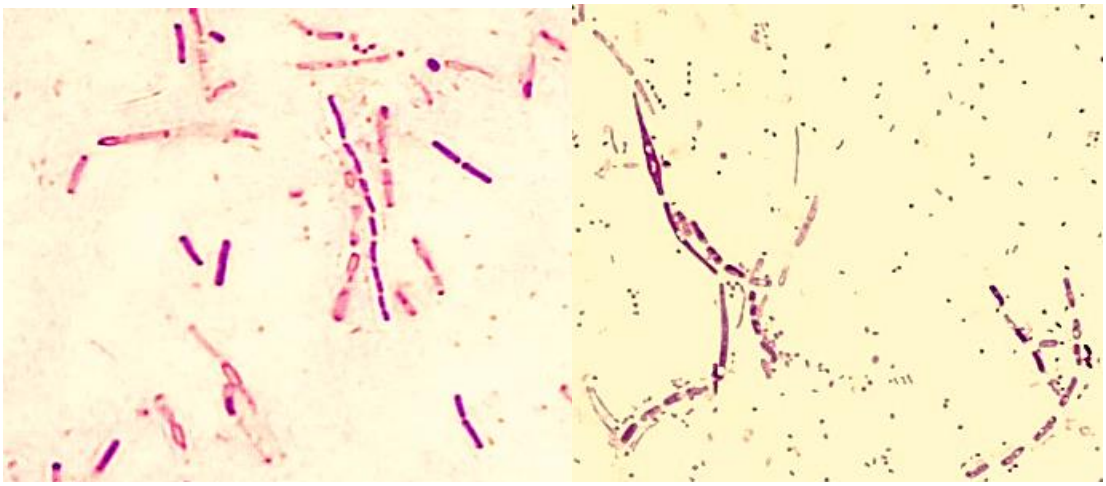

Figure 5. Grampositive Streptobacillus, by microscopic observation of Gram-stained smears (100X).

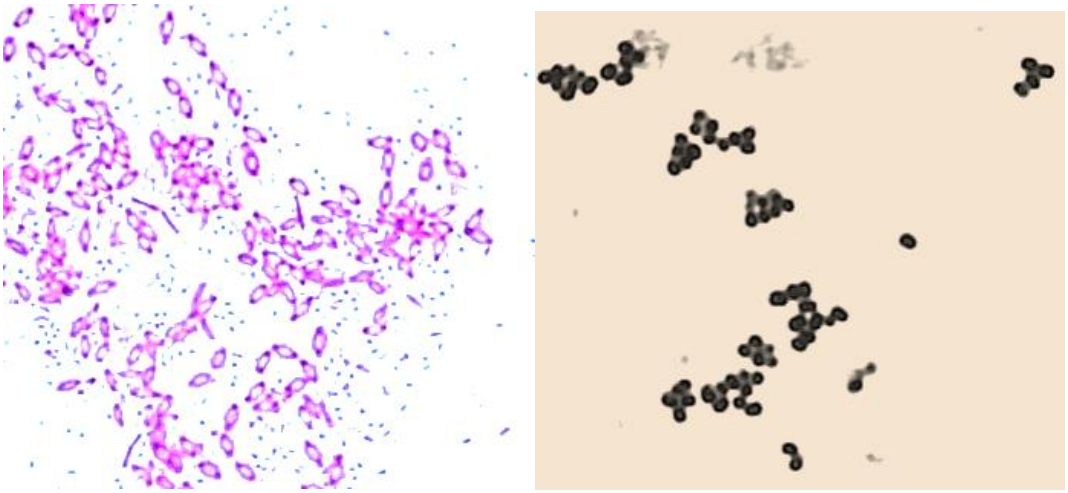

Figure 6. Differentiation of Grampositive endospores and Grampositive staphylococci, respectively, with a staining performed using the Gram technique (100X).

The biochemical tests were performed using the API 20E test battery. For this purpose, the working area was sterilized and a sample of each of the colonies of bacteria was taken from the reseeded cultures and placed in tubes with a 5% sodium chloride solution until the sample of bacteria in the solution was completely diluted. Each of the 20 microtubes contained in the strip was inoculated with a Pasteur pipette. The strips were then labelled and incubated at 37°C for 18 and 24 hours. Finally, the reactions were read using the reading table included in the API20E test manual (Figure 7).

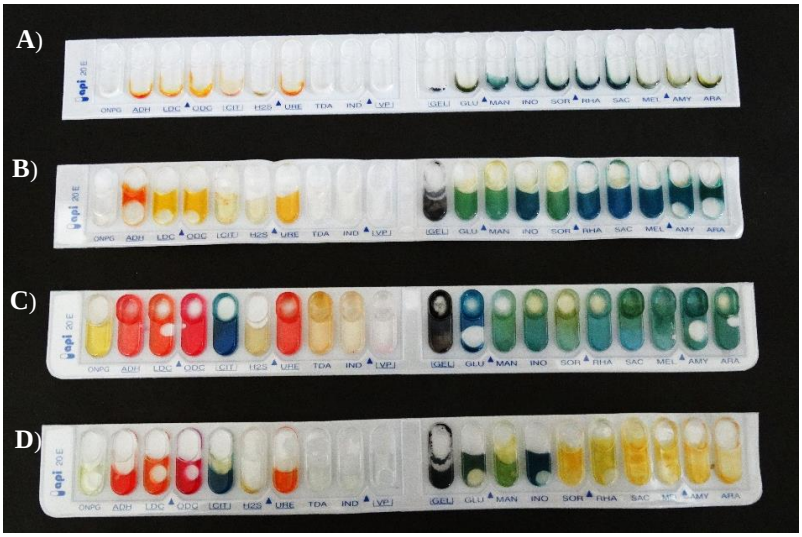

Figure 7. Biochemical tests on sample T53. A) Standard series; B) Negative series; C) Partially positive series, as only color change was observed on the left side; D) Positive series.

3.2 Hematology

After observing the morphology of the blood cells in the samples, three types of leukocytes or white blood cells were differentiated: eosinophils, heterophils and lymphocytes; in addition to erythrocytes or red blood cells, which were seen in all samples as shown in Figure 8, and some thrombocytes, known as platelets.

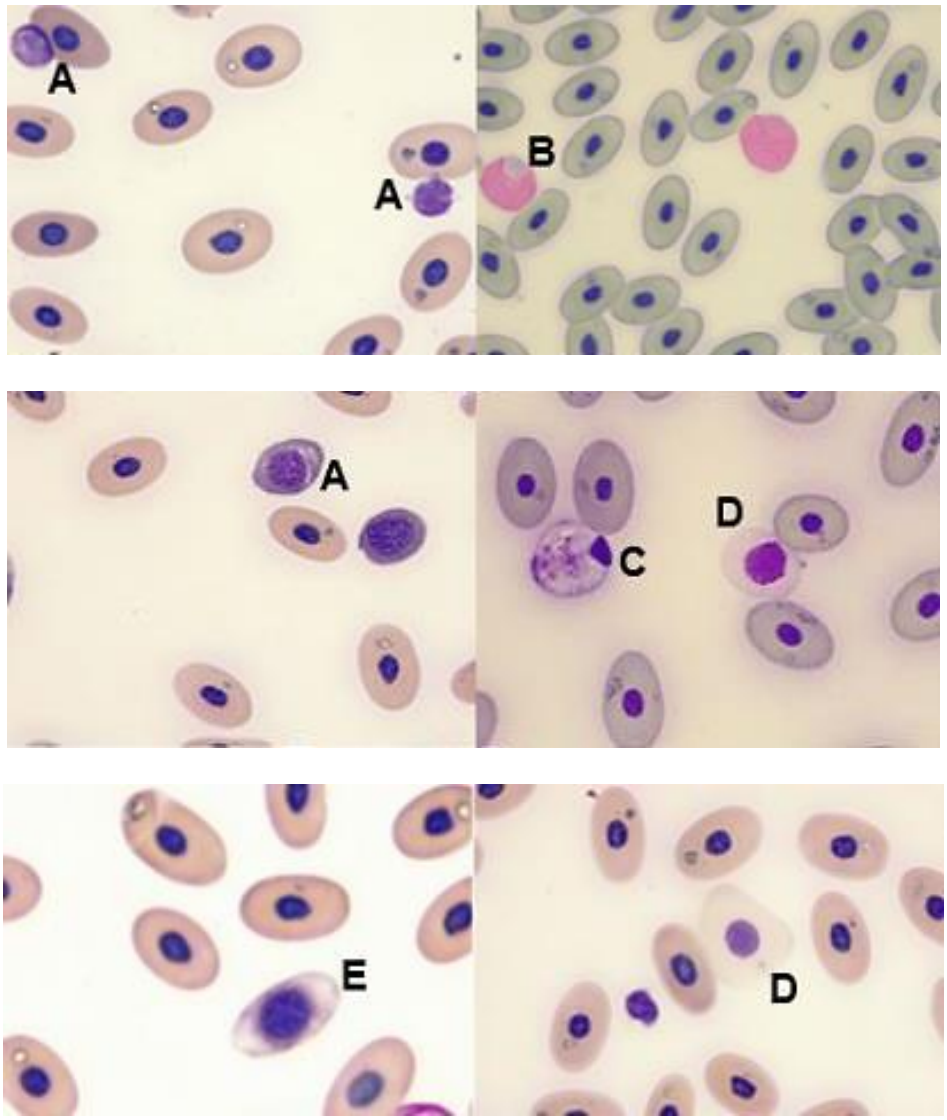

Figure 8. Blood cells observed in *Caretta caretta* captured in the Golfo de Ulloa: A) Lymphocyte; B) Eosinophil; C) Heterophyll; D) Thrombocytes or platelets; E) Immature erythrocyte. Giemsa stain (100X).

Erythrocytes were present in some samples being mature and, in some cases, immature. They presented an elliptical shape with a uniformly distributed cytoplasm. The cytoplasm was pale pink in color and had a blue-violet core.

Also, thrombocytes or platelets were found, but in smaller numbers, with an irregular shape and a violet oval central nucleus, with a grouped chromatin. The cytoplasm was a

pale, almost transparent color, which differentiates them from lymphocytes. They are characterized by adhering to the rest of the blood cells.

As for the white series, the eosinophils found were observed as cells with a defined and indefinite rounded shape, with dark pink internal granules. The nucleus is a fainter colour, with an eccentric location and an oval shape. In addition, they had a cytoplasm with an abundance of large granulations. Another type of leukocytes were lymphocytes, which are characterized by shaping adjacent cells, where the nucleus and cytoplasm are in almost the same proportion, with a well-defined contour. The chromatin is distributed on the outside of the cell acquiring a pale purple color, while the nucleus takes on a bluish purple color. The last cell type belonging to the white line were heterophils, which are the most common granulocytes in the blood of reptiles. They presented a strongly purple eccentric nucleus and with an irregular morphology, while the cytoplasm was of a very clear color, as can be seen in figure 8.

4. DISCUSSION

First, before performing the biochemical tests, it was important to observe them under a microscope, as this is considered to be the previous guide to follow in order to evaluate the type of bacteria that these animals possess. The predominance of Grampositive bacteria is evident in the samples, an aspect totally contradictory to the study carried out in 2015 by Nájera & Salinas on the bacterial flora of *Lepidochelys olivacea*, where they identified a large number of Gramnegative bacteria in the cloacal area, which under stressful conditions can cause infectious diseases.

The release of bacterial endospores, present in some of the samples, indicates an immediate response of the immune system to adverse conditions, such as sudden changes in water temperature, a factor with greater relevance in the area, or high levels of radiation. Endospores may remain inactive for a long period of time, until the environmental conditions are right to germinate, and to preserve the genetic material of the cell (De Hoon et al., 2010). This characteristic is possessed by certain Grampositive sporulated bacilli and cocobacilli, which were observed in this study, where the exact identification of the genus could not be further developed. This fact could verify that the physiological system of turtles was not in good condition due to several factors, which can deprive their proper functioning.

The bacteriological results obtained through biochemical tests in *Chelonia mydas* and *Caretta caretta*, affirm the existence of *Pseudomonas* in the cloacal zone, a criterion that agrees with the description of another study carried out in Mexico (Gámez-Vivaldo et al., 2009). This genus of Gramnegative bacteria is presented as a group of microorganisms with high levels of antibiotic resistance (Atrih & Foster, 2002). The conditions in the area are practically similar to our study area, so it is possible that the proliferation of these microorganisms is favored by the same factor.
To take this information as relevant, it must be related to various supplementary factors that condition the stability of their immune system, such as the environment in which they live or whether they have an adjacent pathology, either epidermal or intrinsic, since bacteria are altered by some factor, are not harmful to the animal. As defended by Serrano

et al. (2012), where they found that the mortality of *Lepidochelys kempii* was associated with the action of *Vibrio alginolyticus, Aeromonas hydrophila* and *Pseudomonas sp*., since this type of bacteria spread to other organs through skin lesions.

This study characterized samples belonging to the Bacillaceae family, which includes especially opportunistic bacteria, similar to Pseudomonadaceae (Warwick et al., 2013). The difference between these families is at the organizational level, where only Bacillaceae comprises Grampositive and Gramnegative bacteria, and some have the ability to release endospores. One example is *Bacillus spp.* which was found as a normal bacterial microbiota in Hawaiian and Australian sea turtles (Zavala-Norzagaray et al., 2015), or specifically in the intestinal microbiota, as described by Abdelrhman et al. (2016) in the loggerhead turtle, *Caretta caretta.*

It should also be noted that bacteria are normally found in coastal waters, such as *Vibrio spp*., which include malignant and benign species (Eiler et al., 2006).

On the other hand, the morphological and structural characteristics, in terms of the first evaluation of blood cells, are similar to the characterization of the species *Caretta caretta* carried out by Casal & Orós (2007) in the Canary Islands (Spain); Lara-Uc et al. (2011) in the morphological study of *Chelonia mydas* in Yucatan (México) and Prieto-Torres et al. 2012 on the same species in Venezuela.
The lymphocytes and thrombocytes or platelets observed by Sykes and Klaphake (2008) in reptiles are morphologically similar to those obtained, the lack of cytoplasmic color appears identically in the results of this study, while erythrocytes appear without any nuclear deformity, a difference cited by Martínez-Silvestre et al. (2011) where amorphous nuclei are differentiated in erythrocytes with abnormalities.
Heterophils and eosinophils may cause confusion when classifying, since their morphological characteristics are similar; but they differ in the type of granulation, as explained by Fei-Yan et al. (2011) in *Caretta caretta* and Martínez-Silvestre et al. (2011) in reptiles.

Hematological values are altered by various factors, like age, maturity, size, etc., as discussed by Casal et al. (2009). The number of basophils was low, which is normal for healthy specimens (Fei-Yan et al., 2011). The frequency of red blood cells can vary according to the species and are directly related to the presence of alterations in the organism, such as anemia, malnutrition, environmental contaminants, etc. (Martínez-Silvestre et al., 2011) These sea turtle species possess heterophils with different cytochemical characteristics, showing different enzymatic species in them (Casal & Óros, 2007). Large eosinophils with a granular content are represented as a response to an inflammatory stimulus, as cited by Work et al. (1998).

This type of results gives us a slight preliminary idea of what the blood structure is like, and then a biochemical analysis is performed, which includes both the differential white blood cell count, a parameter that can show stress levels due to causes such as migratory displacement or the technique used to capture it (Montilla et al., 2014); or how the hematocrit is performed.

In addition, these emblematic beings, thanks to being flag species, benefit from the ease of attracting public attention, being this a means to make any rational subject aware of the great damage that is unconsciously produced to the oceans, and consequently, to the habitats of many animals (Caro & O'Doherty, 1999). Therefore, thanks to their role within biology, sea turtles are considered a basic tool for reflecting environmental disturbances and for achieving favorable environmental education (Aguirre et al., 2002).

5. CONCLUSION

In this microbiological diagnosis, there were a greater number of Grampositive cocobacils and bacilli of different shapes and sizes. The conclusive biochemical tests indicated that the Bacillaceae family and the genus Pseudomonas were present which could be used for environmental resistance control.

By characterizing the cell morphology 6 types of blood cells were differentiated, which were healthy, without any anomaly; and a shortage of basophils.

These results are not sufficient to conclude a complete characterization of the state of health of sea turtles. Therefore, this microbiological diagnosis is intended to continue in order to conclude with relevant results or parameters that will serve as a basis to support future projects.

The implementation of a monitoring program for these species could be a stroke of luck, since studying the health of the populations, could monitor the health of the ecosystems they inhabit. To preserve and improve the health of the Western Pacific Ocean, presented as one of the richest in the world.

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VALORACIÓN PERSONAL (PERSONAL ASSESSMENT)

1. Actividades desarrolladas

Para llevar a cabo el presente Trabajo de Fin de Grado, se realizaron una serie de tareas o actividades que engloban tanto un análisis experimental como bibliográfico. A continuación, se describe brevemente dichas actividades:

1.1. Trabajo de campo

Se efectuaron salidas de campo mensuales, desde agosto hasta noviembre, en varias áreas de estudio diferente (Golfo de Ulloa, Laguna Ojo de Liebre y Laguna Guerrero Negro). Tras la captura de los ejemplares por varios tipos de arte, y como parte principal de este trabajo, se recolectaron muestras bacteriológicas y hematológicas, siendo las muestras de piel, escudo y lavados esofágicos, añadidos complementarios.

Por otro lado, también se obtuvieron datos físicos, tales como morfometría, sexo y peso de cada uno de los animales capturados.

1.2. Trabajo de laboratorio

Una vez obtenidas las muestras, se procesaron en el laboratorio de oceanografía de la Universidad Autónoma de Baja California Sur (UABCS).
Este análisis exhaustivo conllevó una serie de técnicas de cultivo bacteriológico, tinción de Gram y observación microscópica (procedimientos expresados detalladamente en el apartado "*Material and methods*").

1.3. Búsqueda bibliográfica

Con el fin de redactar este documento, fue necesaria una recopilación bibliográfica sobre el tema. Esta tarea predominó durante los seis meses de la temporalización del trabajo, desde la previa familiarización con conceptos básicos hasta la realización de una comparación con otros estudios ya publicados sobre este ámbito.

2. Formación recibida

Previo al comienzo de mis prácticas externas, recibí una pequeña formación en el campo de la microbiología y de la zoología, concretamente en quelonios, por medio de las asignaturas de Microbiología y Tortugas Marinas, cursadas en la UABCS. Ambas fueron impartidas por mi tutora, que respalda tanto mis prácticas externas como mi trabajo fin de grado. En estas asignaturas, obtuve un aprendizaje sobre los conceptos básicos que posteriormente, tendría que utilizar en este estudio; por ejemplo, los diferentes métodos de tinción, conceptos de bacteriología, morfometría y patrones a seguir en el diagnóstico sobre la salud de los quelonios, etc.

Por otro lado, la formación tomada también incluyó el manejo de un microscopio digital con una cámara de 1,3 MPixel integrada, que me permitió la observación simultánea, para

posteriormente, obtener imágenes fijas de las distintas formas, colores y tamaños de los tipos de bacterias que existían en las muestras.

3. Nivel de integración e implicación con el personal

El nivel de implicación e integración fue bastante bueno, ya que no existió ningún problema que dificultase mi aprendizaje, ni la finalización de mí TFG. El grupo de trabajo actuó de manera correcta y dinámica, en la preparación previa del material, en el laboratorio y en las salidas de campo.

4. Aspectos positivos y negativos relacionados con el desarrollo

Dentro de los aspectos positivos, de carácter general, puedo destacar la comprensibilidad, por parte de los integrantes del proyecto, a la hora de brindar toda la formación necesaria en este ámbito, la atención y disponibilidad prestada en todo momento, especialmente, por parte de mi tutora, y la implicación empática recibida por compañeros del proyecto y personas externas pertenecientes al departamento.
En cuanto a los aspectos negativos, puedo destacar la falta de tiempo disponible para poder realizar unas prácticas más extensas, con un mayor contenido de información en la línea de trabajo clínica.

5. Valoración personal del aprendizaje conseguido

Podría afirmar que, en el ámbito personal, he adquirido una gran variedad de conocimientos empíricos, aprendiendo lo que realmente conlleva la realización de un estudio científico de investigación, donde es necesario un esfuerzo de adaptación y comprensión, en el que el trabajo en grupo se debe valorar notablemente dentro de éste. También, cómo realizar un manejo adecuado de los quelonios en su hábitat natural, cómo hacer una óptima recolección, administración y procesamiento de mis propias muestras y como enfocar mis resultados hacia un fin de preservación ecológica de los ecosistemas marinos.

Con esto, he aprendido una gran cantidad de lenguaje científico, útil para la realización de trabajos o actividades posteriores, y una visión diferente sobre la rama microbiológica y hematológica; información personalmente enriquecedora que me ayudará a visualizar mejor mis prioridades de cara al futuro.