

Hematological parameters in *Oreochromis niloticus* cultivated in tropical conditions in experimental farm Santo Domingo of University of Amazonia, Florencia, Caquetá, Colombia

Parámetros hematológicos en *Oreochromis niloticus* cultivada in condiciones tropicales en la Granja Experimental Santo Domingo de la Universidad de la Amazonia, Florencia- Caquetá, Colombia

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Abstract— The hematological parameters were evaluated in the *Oreochromis niloticus* species, weighing between 120 and 375 g, where chemical standards of water were included (T °, pH and OD), morphometry and haematological diagnosis, carried out at the Santo Domingo Experimental Farm from the University of the Amazonia, Florencia - Caquetá-Colombia. A directed and completely random sampling of 32 individuals was used, the blood was extracted by cardiac puncture and placed in Eppendorf tubes with EDTA. Blood smears were stained with Ema staining color for its laboratory analysis. The InfoStat was applied as a statistical method for the analysis of the variables to be measured. A total of leukocytes of $1.11 \times 10^5 / \text{mm}^3$ was found, predominating neutrophils with 49.3% followed by lymphocytes with 38.8%, monocytes with 5.9%, eosinophils with 4.3%, basophils with 1.7 % and thrombocytes with 63.8% as an independent series. For the red series, an average of erythrocytes of $1.5 \times 10^6 / \text{mm}^3$, hemoglobin of 7.9 g / dL; hematocrit of 23.3%, plasma proteins 3.3 g / dL, mean corpuscular volume 165 (μ^3), mean corpuscular hemoglobin 55 (uug) and mean corpuscular hemoglobin concentration of 33.3%. In conclusion, it was determined that some individuals presented Leukopenia with Lymphopenia and thrombocytosis. In addition, two individuals presented Normochromic Microcytic Anemia (AMN) and 19 individuals presented Normochromic Normocytic Anemia (ANN). The analysis of Total Plasma Proteins (PPT) determined that the high mortality rate was not due to nutritional deficiency. There was a positive similarity between the VCM and HCM, and negatively with the total number of erythrocytes.

Index Terms—*Oreochromis niloticus*, Leukogram, Eritogram, Anemia, Ema staining color.

Resumen— Se evaluaron los parámetros hematológicos en la especie *Oreochromis niloticus*, con peso entre los 120 y 375 g, donde se incluyeron estándares químicos del agua (T°, pH y OD), la morfometría y el diagnóstico hematológico, realizados en la Granja Experimental Santo Domingo de la Universidad de la Amazonia, Florencia- Caquetá-Colombia. Se utilizó un muestreo dirigido y completamente al azar de 32 individuos, la sangre fue extraída por punción cardíaca y colocada en tubos Eppendorf con EDTA. Los frotis sanguíneos se tiñeron con coloración Ema color tinción para su análisis en laboratorio. Se aplicó el InfoStat como método estadístico para el análisis de las variables a medir. Se encontró un total de leucocitos de $1,11 \times 10^5 / \text{mm}^3$, predominando los neutrófilos con 49,3% seguido de linfocitos con 38,8%, monocitos con 5,9%, eosinófilos con 4,3%, basófilos con 1,7 % y trombocitos con 63,8 % como serie independiente. Para la serie roja se encontró un promedio de eritrocitos de $1,5 \times 10^6 / \text{mm}^3$, hemoglobina de 7.9 g/dL; hematocrito de 23.3%, proteínas plasmáticas 3,3 g/dL, volumen corpuscular medio 165 (μ^3), hemoglobina corpuscular media 55 (uug) y concentración de hemoglobina corpuscular media de 33.3%. Como conclusión se determinó que algunos individuos presentaron Leucopenia con Linfopenia y trombocitosis. Además, dos individuos presentaron Anemia Microcítica Normocrómica (AMN) y 19 individuos presentaron Anemia Normocítica Normocrómica (ANN). El análisis de Proteínas Totales del Plasma (PPT) determinó que la alta tasa de mortalidad no fue por deficiencia nutricional. Se

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presentó una similitud positiva entre el VCM y HCM, y de forma negativa con el número total de eritrocitos.

Palabras claves— Oreochromis niloticus, Leucograma, Eritrograma, Anemia, tinción Ema color.

I. INTRODUCTION

Tilapia is the common name of a group of species belonging to the family Cichlidae, of which, the genus *Oreochromis* represents the main source of income and therefore is categorized as fish of greater commercial relevance in aquaculture; Therefore, at the global level, they appear as the third most important group, after carp and salmon, and their cultivation presents one of the fastest growing activities worldwide [1] - [2].

However, the research that has been conducted in areas such as ichthyophysiology and ichthyopathology of species of productive and economic importance (such as Tilapia) under tropical conditions are very scarce, this has brought serious consequences in the national fish sector as it locates it at a certain disadvantage compared to other countries, in which, research in this area has some relevance and correlation with its fishing production [3]. Likewise, from the productive point of view, it is gradually becoming an alternative source of protein of great relevance for the most vulnerable population, but at the same time it guarantees income and a formal employment to these people, which is verified in the gradual increase of fish culture of 12% per year (average) between the years of 1990 and 2011, data that allow establishing tilapia as the second species of commercial importance [4] - [5] - [6].

However, like any production, fish farming demands "precise, effective and low-cost tools" that allow a diagnosis and constant monitoring of production indices; for this, the complete and detailed study of the chemical and haematological parameters compose an accessible and important strategy that acts as a bioindicator for the detection of pathologies that come to harm in fish production [4] - [7], since fish are susceptible to problems of nutritional imbalance, anoxic conditions, stress effects, chemical products, intoxication, genetic variability, attack and invasion of pathogens (bacterial, parasitic, viral), and environmental changes in populations that live in a natural state or in confinement [4] - [8].

That is why a study of blood physiology is transcendental, since changes in the intrinsic and extrinsic character of the environment are rapidly and efficiently reflected in the blood [9]. Therefore, it becomes essential, in a first scenario, to identify and establish the "normal" haematological parameters for the tropics, which will support the examination of the species, such as Nile Tilapia (*Oreochromis niloticus*), in order to look for an improvement in the quality and production of species for the region [3] - [10] - [11].

In the framework of the study of blood physiology, it is essential to evaluate standard parameters that indicate in a timely and accurate manner the pathological phenomena that develop in the individual; For example, red blood cells, also called erythrocytes, are the most abundant blood cells in most

ichthyofauna. Its main function lies in the transport of oxygen to tissues and CO₂ to the lungs for proper elimination. On the other hand, white blood cells or leukocytes are widely used as an indicator of health in fish because they are the units of defense against pathogens and are involved "in the immunological regulation" of the organism. Likewise, thrombocytes act in the coagulation processes [12]. A blood count can be very beneficial for the diagnosis of anemia and stress states, in addition to knowing and evaluating the conditions of the aquatic environment in which they are found, in order to determine a possible bacterial septicemia. A characteristic to be taken into account is that erythrocytes and thrombocytes in fish have a characteristic of being nucleated, a feature that does not occur in mammals [9] - [13].

Thus, this study projected to present a hematological analysis in individuals of *Oreochromis niloticus*, in order to establish their blood values under tropical conditions, as well as the possible pathologies present in individuals.

II. METODOLOGY

A. Study area

The study was conducted at the Santo Domingo fish station experimental farm of the University of Amazonia, located between the geographical coordinates 1 ° 36'26 "of Latitude North, and 75 ° 46 '1.63" of West longitude, 7 km from the City of Florencia on the road that connects with the municipality of Morelia. The region is classified as a tropical humid forest, it has an average annual rainfall of 3600 mm³, an average relative humidity between 80% and 85% and an average annual air temperature of 28 ° C. The fish were cultivated under optimal conditions of oxygen (4.40 mg / L), temperature (26.5 ° C), pH (5.99-7.1) and feeding (4% biomass / daily); Later, they were transferred to the animal physiology laboratory of the University of Amazonia (FA-UA) in Florencia-Caquetá, where the samples were taken and analyzed respectively [14].

B. Sampling

To minimize the stress of the fish at the time of sampling and change of environment, they were anesthetized with Eugenol in relation to 1mg / 20L of water. The extraction of the blood was done using the cardiac puncture methodology used by [4], [13], [15] using disposable syringes of 1 cc, with hypodermic needles and collected in Eppendorf tubes with EDTA solution for subsequent hematological analysis.

C. Morphometric parameters

A Ubermann digital caliper and a millimeter ruler were used for measurements of total length, standard length, head width, body width, body height and head length for all individuals; In addition, in order to determine the weight, an Electronic Scale was used [16].

D. Hematological Analysis

To perform the blood smears the methodology proposed by [17] was used, a direct total cell count was performed by the Hemocytometer method using a dilution of Natt-Herrick diluent for three minutes and placed in a Neubauer chamber, which allows to stain the cells differentially to visualize them. The direct differential cellular study in the blood smear was performed in an optical microscope with a built-in digital camera. The blood smears were stained with Hemacolor staining as [4] stipulates it. With regard to the indirect count of leukocytes in the blood smear, the parameters were evaluated according to the methodology proposed by [3].

E. Statistic analysis

For the analysis of the information obtained from the 32 fish, a guided sampling and descriptive statistical methods were used to analyze the variables to be measured: hematocrit, hemoglobin; erythrocytes, MCV, HCM, CHCM, leukocytes; (heterophiles, lymphocytes, eosinophils, monocytes) and thrombocytes used by [9], [18] likewise, the statistical software InfoStat was used.

III. RESULTS AND DISCUSSION

The individuals studied had an average weight of 208.75 ± 66.73 and an average total length of 22.14 ± 1.98 presented in Table I coinciding with the values recorded by [4], which in turn were similar with those reported by [19].

TABLE I
MORPHOMETRIC DATA OF THE O. NILOTICUS INDIVIDUALS.

	Weight (gr)	Total length (cm)	Standard length	Length of the head	Width of the head	Width of the body	Height of the body
Average	208,75	22,14	19,11	53,47	32,28	28,22	73,48
Máximo	375	26,4	49,6	64,93	39,82	35,27	90,45
Mínimo	120	18,7	14,6	30,51	22,6	23,75	60,62
Standard deviation	66,73	1,98	5,93	6,82	4,08	2,88	7,33

The analysis of conglomerates by the Ward method and the Euclidean distance made for the Hematological parameters in Nilotic Tilapia (*Oreochromis niloticus*) individuals cultivated in Santo Domingo farm, Florencia- Caquetá, showed that with a cofenetic distance of 0.54 for morphometric analyzes

evidenced the formation of four groups; group number one (1) with 15 individuals who presented the highest values, group two (2) and three (3) with average values; while group number four (4) presented the lowest values (Figure 1).

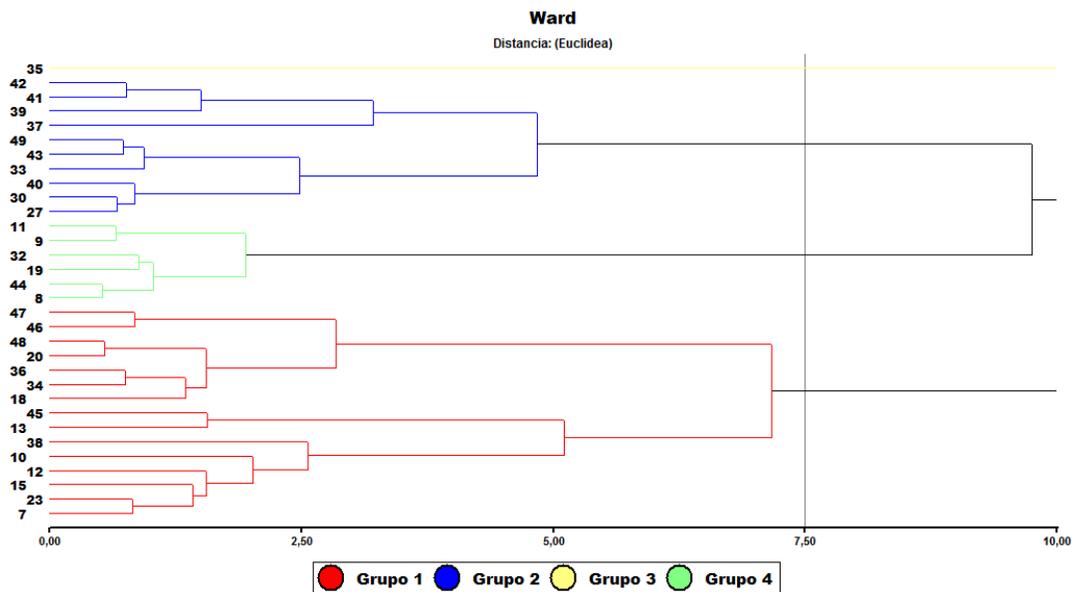


Fig 1. The analysis of conglomerates by the Ward method and the Euclidean distance made for the Hematological parameters in Nilotic Tilapia (*Oreochromis niloticus*) individuals cultivated in Santo Domingo farm, Florencia- Caquetá.

The main component analysis (MCA) performed on the correlation matrix for the Hematological parameters in individuals of Nile Tilapia (*Oreochromis niloticus*) cultivated

in Santo Domingo farm, Florencia- Caquetá showed that the first two main components (MP) contribute the 86 % of the total variability, the first 2 components showed characteristic roots

greater than the unit capturing 86% of the variability. When analyzing the coefficients of the vectors associated with the first two main components, it was found that the variables of greatest importance in their order were: Total length, weight and height of the body.

According to Figure 2 the first main component (MP1) with a contribution of 68.0% of the variability organized 16

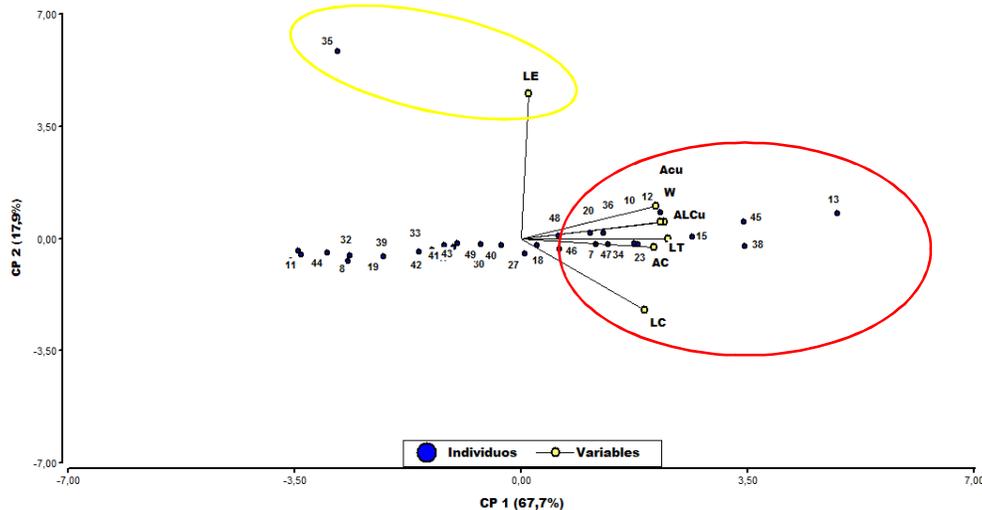


Fig 2. Main components analysis (MCA) carried out for the Hematological parameters in individuals of Nile Tilapia (*Oreochromis niloticus*) cultivated in Santo Domingo farm, Florencia-Caquetá.

LEUCOGRAM

On the other hand, with the thirty-two blood samples analyzed, the presence of erythrocytes, lymphocytes, monocytes, heterophils, eosinophils, thrombocytes (Figure 4) and basophils was evidenced, as has been reported in other studies conducted for this species [20], [4] predominating heterophiles with an average percentage of 49.3% (Appendix 2), indicating a certain degree of abnormality since it is more common to observe lymphocyte predominance. However, some studies such as that of [21] coincide in the value of leukocytes for the species *Oreochromis niloticus* and *Cyprinus carpio*. However, low values for *O. niloticus* were found in the studies of [4] and [22], also indicating that the high percentage of these leukocytes may be due to low water quality and high content of circulating organic matter, by stress conditions induced by low oxygen concentrations or in response to parasitism and infection, including exposure to various pollutants as stipulated by [23].

Regarding the percentage of heterophiles, in a study developed by [22] for the species *Oreochromis aureus* obtained an average between 1% and 7% of heterophiles, however [4] found a maximum average of 27%; this indicates that for the fish Salmonidae these cells are the second most numerous population, being located behind the lymphocytes (corresponding about 20% of the surrounding leukocytes) demonstrating that this cell is the leukocyte type with the greatest tendency to variation [24].

individuals with the highest values of weight, total length, head length, head width, body width and body height to the positive end of such component; while the second main component (MP2) with a contribution of 18% organized the standard length with an individual to the positive end of such component (Figure 2).

Morphologically speaking, in the present work we found cells of medium and oval size, with notorious cytoplasm, slightly basophilic and fine granulations; the nucleus is eccentric, reniform, similar to the data reported by [25]. White cells or so-called leukocytes play an important role in immune function, and the higher the number of these in blood may indicate a better defense response of the body, since in high-density production systems are constantly exposed to high bacterial loads and a poor condition of the body of water [4] and [26] therefore, when performing the respective analysis of the values of this type of cell in our investigation, we obtained an average of $1.11 \text{ cel} \times 10^5 / \text{mm}^3$, which according to [4] is not indicative of a leukocytosis, nor leukopenia with lymphopenia, resulting for these authors $1.21 \pm 0.07 \times 10^5 / \text{mm}^3$, which indicates similarity between both values.

For [4] the leukocyte average corresponded to $2.18 \times 10^5 / \text{mm}^3$, a value that is within the range for this species (tilapia). Likewise [10] obtained an average of $0.082 \pm 0.075 \text{ leukocytes} \times 10^5 / \text{mm}^3$, a very low value compared to ours. On the other hand, in a study developed by [13] the individuals of *Chirostoma estor estor* presented a number of less than 20,000 cells per mm^3 due (possibly) to chronic stress, which indicates a leukopenia with lymphopenia.

Taking into account the values of leukocytes in the present investigation, some authors stipulate that due to the constant situations to which fish face, for example, stress, leukocyte values are not high to determine leukocytosis ($1.11 \text{ cel} \times 10^5 / \text{mm}^3$), or low to describe as Leukopenia disease with

lymphopenia, however, remains controversial because of the few studies implemented to this type of species. Although it should be noted that probably the animals are constantly with the greatest contribution of nutrients by the daily biomass implemented, therefore, they are likely to show a defensive response to this situation exerted by the environment [27] - [13].

Lymphocytes are present in immune responses or stimulus immunological, for example in the inflammatory process of fish; when performing the analysis of these cells in our research, we found characteristics such as: large, dark purple nucleus, covering almost the entire cell, it is irregular and presenting chromatin (Figure 4). The little cytoplasm that is illustrated in light blue; data similar to those reported by [4] and [24]. The lymphocyte average for this investigation was 38.84%, which is within the range established for the species *O. niloticus*, however compared to other studies conducted for the same species is relatively low, for example see [3] - [4] obtained a percentage of lymphocytes of 52.8%, which were predominant; in relation to Salmonid fish, these correspond to a domain of 70-80% of these cells according to [24] and for [28] the predominance of circulating lymphocytes in *Oreochromis aureus* was 53-54%.

In other studies carried out for species such as *Astronotus ocellatus*, a predominance of lymphocytes was found, which is consistent with that found in other teleost fish [29] Another case in particular is reported for rainbow trout (*Oncorhynchus mykiss*) where lymphocytes are the type of leukocyte more frequent (89-98%) as stipulated [17].

The primordial functions of eosinophils are associated with the antiparasitic response, modulation of allergic reactions and phagocytosis as described [17] - [4] and [30], in our investigation we found lobed cells, the eccentric and lobed nucleus occupying a fourth part of the whole cell, and the cytoplasm has large granules (orange, purple) coinciding with [17] and according to [3], these cells are very rare for this species, i. e. they only reach a number lower than 5% of the total leukocytes, for example the average of eosinophils in this investigation was 4.3%, a high value for such species.

According to the authors mentioned above, their representation only reached 2.5% as the maximum value for the countdown of white ones; however for [23] these cells were not observed and stipulate that the presence of these granulocytes may be related to diseases such as eosinophilia, since under normal conditions (healthy fish) they are scarce or absent. Similar data reported by [31] show that this type of cells was not observed in work performed in *O. niloticus*, and for other species such as *Ictalurus punctatus*, these values coincide with our study [9].

The basophils presented an average percentage of 1.71% in this investigation, coinciding with the data of the species *Pterophyllum scalare* [29], however, other authors identified these cells with a lower percentage than the one found, for example [29] obtained 0.0 ± 0.75 , [4] with parasitized tilapia obtained 0.0 ± 0.44 , [4] found 0.43 ± 3.18 and [3] with $0.35 \pm$

0.11% . But other studies carried out by [31] and [28] did not report the presence of these cells for *O. niloticus*, and that according to [17] the basophils that are observed are in very low amount in circulating blood representing <1% of total leukocytes; this may be due to problems of exposure to toxic substances, indicating a decrease in the level of nonspecific immunity for fish as reported [22].

Monocytes are part of the nonspecific immune response and phagocytosis [17] and [4]. The morphological characteristics found in the present investigation highlight that these cells are large, irregular and with abundant cytoplasm; the nucleus is large and kidney shaped, in addition it is eccentric, lobed, blue-violet, occupying much of the cell cytoplasm, this in turn is irregular, pale gray, contains small granules, coinciding with the data reported by [25], [17] y [22]. In this research an average of 5.9% of monocytes was presented, which according to [32] explains that they comprise less than 10% of the total production of white blood cells, indicating normality of the results and coinciding in works carried out by [23] and [3] with an average percentage of 0-8.0% for this species; likewise [22] identified Monocytes with a total percentage of 2-13%. [4] determined a percentage of 1.48-17.31% and [33] reports similar values for adult *O. niloticus* before and after acclimation for seven days to males and females (3.37%, and 5.04 % respectively).

The main function of thrombocytes is to control the loss of fluids from superficial wounds of fish and at the same time to interact with the defense system through phagocytic and bacterial activity [4] - [34] and [17]. In our study, we found a high percentage of thrombocytes (63.8%), which indicates thrombocytosis possibly due to the high level of stress associated with the medium [35].

According to [23] they observed small thrombocytes that measure $4.95 \times 5.87 \mu\text{m}$, elongated, and grouped, with a dense nucleus of purple color, occupying a large part of the cell, little cytoplasm, showing elongations at their ends; coinciding with authors [3].

For other species according to [17] in *Salmo Salar*, [25] *Rhandia quelen*, [36] in *Salminus affinis* and [13] in *Chirostoma estor estor*, such thrombocytes presented an average of 14.45%, a value that is within the range indicating normality (Appendix 2) ; Besides, other authors obtained similar percentages, see [3] with (14.53%), [32] with (10.5%), [4] with (26%) and [37] with (5%) being this last one the lowest.

For *Oreochromis aureus* they found a higher value ranging from 55 to 74% [22], for other species *Ictalurus punctatus* was 20%, *Micropterus salmoides* was close to 19%, in *Morone saxatilis* with 54.9% (according to a study developed by [9] - [13]) identified a percentage of 14-62% in *Chirostoma estor estor*. Therefore, in relation to the above, thrombocytes in *O. niloticus* have been described as the most abundant blood cells after erythrocytes [30] and [37].

According to [4] they include thrombocytes in the differential count of leukocytes, for the investigation it was taken as an

independent series, coinciding with other authors such as [36]-[38], since the reagent of Natt and Herrick allowed in a same dilution to identify leukocytes and thrombocytes for their total count, noting that when blood is collected with anticoagulant, the thrombocytes exhibit an oval shape as obtained in this investigation

The main components analysis (MCA) performed on the correlation matrix for the Hematological parameters in individuals of Nile Tilapia (*Oreochromis niloticus*) cultivated in Santo Domingo farm, Florencia- Caquetá showed that the first three main components (MC) contribute the 85 % of the

total variability, the first 4 components showed characteristic roots greater than the unit capturing 99% of the variability. When analyzing the coefficients of the vectors associated with the first two main components, it was found that the variables of greatest importance in their order were: Hto, Hb and VCM. According to Figure 3, the first main component (MC1) with a contribution of 49.7% of the variability organized 10 individuals that presented the highest values of GB, Hb, Hto, VCM, HCM and CHCM; while the second main component (MC2) with a contribution of 20.8% Organized the GR with 3 individuals who presented the highest values for these variables (Figure 3).

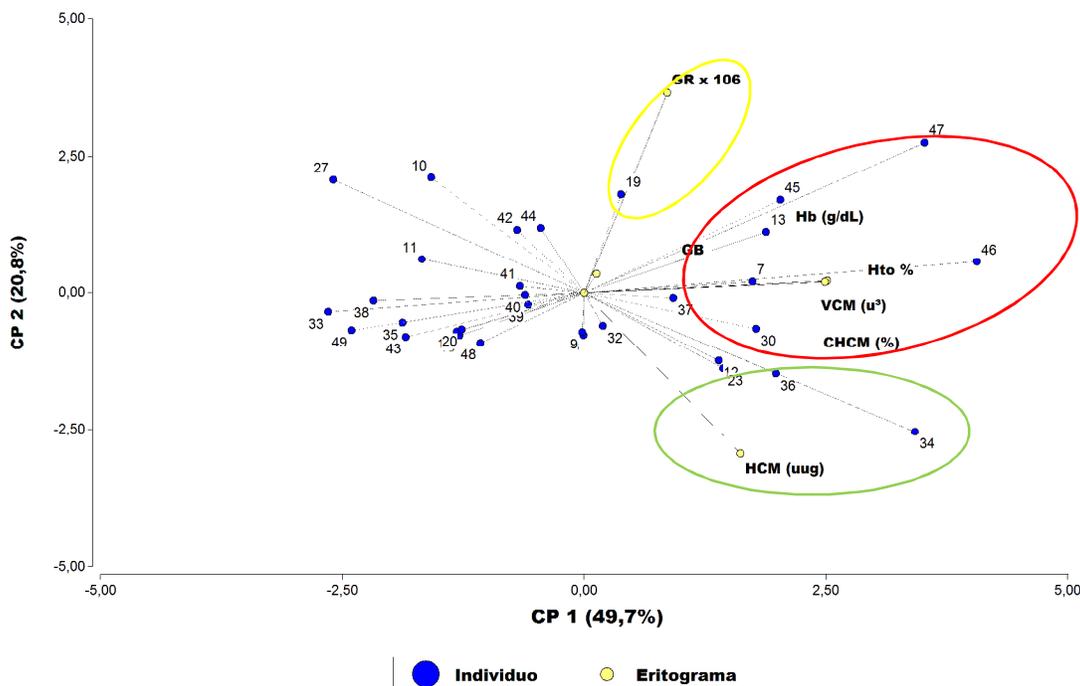


Fig 3. Analysis of main components (MCA) carried out for the Hematological parameters in individuals of Nile Tilapia (*Oreochromis niloticus*) cultivated in Santo Domingo farm, Florencia- Caquetá.

ERYTHROGRAM

The erythrocytes allow the transport of oxygen bound to hemoglobin [17] and [39] which, when they are mature, are the only ones that must be "found in the peripheral blood", because the presence of these cells, in a low state of maturity indicate damage in the production of red blood cells. In the present study characteristics were described that correspond to mature erythrocytes, i. e. the nucleus is dark, oval and central. The cytoplasm is light, oval and without granules, coinciding with that reported by [27] obtaining an average of 1.5 celx106 / mm3 indicating normality, concordant with [39] for a larger species such as *Arapaima gigas* and similar values for the species *O. niloticus* according to [4] and [3]; also for [35] *O. niloticus* showed a higher average in erythrocytes 6.93 ± 8.28 celx106 / mm3, determining values of 256.7 celx106 / mm3 for the hybrid *O. urolepishormnorum* x *Oreochromis mossambicus*.

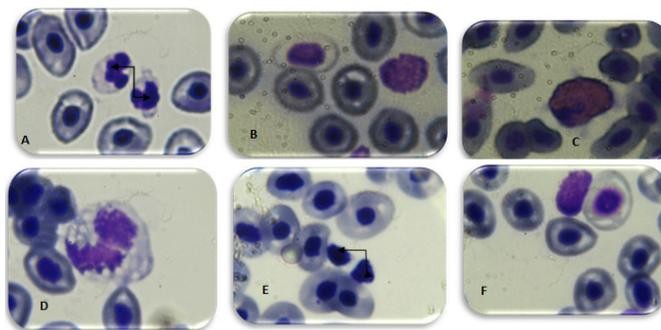


Fig 4. *Oreochromis niloticus* blood cells. A) red blood cells and neutrophils. B and C.) Eosinophil D) Monocyte. E) Fusiform thrombocyte. F) Elongated lymphocyte.

The concentration of hematocrit obtained in this study shows an average percentage of 23.3%, indicating normality (Appendix 1); finding similar values for the species from 27% -43% in works done by [3], also for [31] the average percentage was 31.85%, for [10] it was 29.1%, see [4] 33.56% and [20] of 24.93%. On the other hand, in a study conducted in the species

O. aureus, see [22] obtained an average of 26%; for other species the reports obtained for *Colossoma macropomum* date from 30%, 66-36% and 74% for [7] 22.7- 25.5% for the blanquillo *Sorubim cuspicaudus*, [38] in the species *Salminus affines* 36 was obtained, 2%, see [36], in *Salmo trutta L.* the average corresponds to 34%, in *Morone saxatilis* 31.19%, in *Ictalurus punctatus* 30.64%, in *Micropterus salmoides* 28.26% according to the data obtained by the study of [9], and 33.95% in *Christoma estor estor* according to [13].

The hematocrits are indicators of physiological and aquatic environment alterations as indicated [3], showing that this value is higher in freshwater fish than in marine water fish, the latter presenting a greater number of red blood cells, which are also smaller [13]; and it is noteworthy that these values vary from one species to another, and even in the same species according to the concentration of dissolved oxygen, activity, gonadal stage, age, gender, physiological status of the fish, water quality, photoperiod and diet as [3] reports it.

For our study, this average percentage is within the range (Appendix 1), but it is a lower value as mentioned above; for this reason, some authors have established that in the lotic environments it is common to see that the hematocrit is greater, while in lentic environments a lower value is observed in terms of percentage [36]. However, [9] establishes that the hematocrit may increase if the individual is in a phase of "moderate malnutrition", but decreases in "severe malnutrition"; bacteria can also be responsible for the decrease in hematocrit, for example individuals of *Oreochromis niloticus* were infected with *E. tarda* and *Oreochromis aureus* were infected with *Corynebacterium sp.* And this tendency was evidenced, which is due to an alteration of hematopoiesis by bacteria or their products [20]. On the other hand, if a hematocrit is present with a higher percentage than the one estimated, it should be determined if there is a polycythemia and if it is accompanied by high hemoglobin values due to dehydration, hypoxia, stress effects or in sexually mature males. However, if there is a reduction in the hematocrit, it usually indicates a type of anemia resulting from possible inflammatory, renal or splenic diseases, nutritional disorders, toxins, traumas, skin ulcerations, parasitism, bacterial or viral septicemia [4].

Regarding the concentration of hemoglobin, an average of 7.9 g / dl was obtained, a value within the range for the species (Appendix 1), however, similar values were found for *O. niloticus* in studies of [32] with an average of 5.8 g / dL in non-parasitized species and 6.54 g / dL in parasitized species, on the other hand [40] found a hemoglobin concentration value of 102.2 g / dl, [41] of 4.05 -10.43 g / dl, [31] of 10.52 g / dl, [42] of 8.2 g / dl, [43] of 6.1 g / dl, [37] of 9.1 g / dl dl, for [49] of 4.25 - 4.63 g / dl, [4] obtained 9.32-9.94 g / dL, for [3] 5.59-11.61 g / dl and for other species such as *Oreochromis nilótica* x *O. mossambicus* (red tilapia), [44] found 6 g / dl and [22] 6.2 g / dl for *O. aureus* (*Oreochromis nilotic* x *O. mossambicus* x *O. aureus*).

For other species, hemoglobin concentration values of 9.94-11.17 g / dL have been obtained in *Colossoma macropomum* [7], 4.8 g / 100ml for *Christoma estor estor* [13] 9.4- 10.5 g / dl

for tilefish *Sorubim cuspicaudus* [38], 12.53 g / dL in *Salminus affinis* [36], for [9] values ranged between 3.6-16.2 gr / 100 ml in *Salmo trutta L.*, for *Morone saxatilis* species was 7.6 g / dl, for *Micropterus salmoides* it was 5.10 g / 100 ml and for [3] the species *Colossoma bidens* presented 10.40 g / dl, *Prochilodus scrofa* (9.70 g / dl) and *C. macropomum* x *C. bidens* (12.7 g / dl).

The concentration of hemoglobin is used as a physiological indicator of contamination and organic stress dysfunction as reported [7], [34] and [45] thus allowing the transport of oxygen according to [17], although the hematimetric indices vary from one species to another, and even in the same species according to the concentration of dissolved oxygen, activity, gonadal stage and physiological state of the fish [13] - [37], is usually high in large fish and air breathing (lentic habitats), however, for small fish (which are less active), without air breathing (usually presented in lotic habitats), the values are lower as stated [9].

The concentration of total proteins showed an average of 3.3 gr / dL, value within the range for the species (Appendix 1) which indicates normality, finding similar values for the *O. niloticus* species of 3, 6 g / dL by [31], 3.1 g / dL for [46] 3.86 and 3.20 g / dL for [4]. However, some authors found a very small value for the species *O. niloticus* with 0.433 g / dL according to [47], and 4.3 g / dL according to [32]. For [9] Total Plasma Proteins, decrease in early phases, therefore, the period of malnutrition is prolonged since the PPT increases, also the Hemoglobin and the Micro hematocrit decrease, presenting in this period a defined anemic syndrome.

In other species the reports indicate that in *Morone saxatilis* it was 4.6 gr / L according to [9] and for *Piaractus brachypomus* it was 27.2 g / L in broodstock with PPT concentration in blood, which was relevant due to the affectation by the level of food, being greater the more food is available [48].

The VCM obtained an average of 165 (u3), which is similar to that reported by [4] with 165.76 (u³) and similar values were found for [43] with a volume of 66.38 (u3) in males and 132.23 (u3) in females, [33] with 148.80 (u3), for [31] with 175.46 (u3), [37] found 191.2 (u3), for [42] it was 174.59 (u3), [3] 200.47 (u3), for [49] was 143.52 (u3), according to [41] for non-parasitized species it was 153.3 (u3) and 135 (u3) in parasitized species; on the other hand [32] obtained 226.24 (u3) for *O. aureus*, [22] for another species such as *Colossoma macropomum* obtained 390.82 (u3), in turn [7] in *Astronotus ocellatus* obtained 93.9 (u3), [38] in *Chirostoma estor estor* obtained 183 (u3), and in *Salminus affinis* 163.8 (u3) according to [36].

These values are related to the cell size of erythrocytes, and are used to classify them as normocytic, microcytic or macrocytic [13] and [34], this hematological index relates to respiratory function and the middle corpuscular concentration of hemoglobin.

Although our results are apparently within the range established for the species *O. niloticus*, they are very low values

compared to other authors. However, the type of anemia presented can be established according to the parameters of red, countdown of red ones, blood cell or red cell and hemoglobin or hemoglobin concentration. In the present study it was found that 16 of the 32 individuals presented Normochromic Normocytic Anemia which according to [24] is generated as a result of hemorrhages in the intestine, liver, pyloric and ocular caecum, product of endothelial damage or when severe or chronic blood loss occurs, the net loss of iron can cause anemia due to deficiency of this mineral. They also explain that it has been associated with environmental stresses (increase in population density or stress). This type of disease is very unique in the species *Oreochromis aureus* [20]. On the other hand, two individuals presented Normochromic Microcytic Anemia with a VCM of 61 u3 and 78 u3 which, compared with the values of [4], is very low for fish with a weight between 150 gr and 250 gr, because the range ranges from 69.57 to 459.46 u3.

In HCM or middle corpuscular hemoglobin an average of 55 (uug) was obtained, which is within the range according to appendix 1, similarly was found similar values as stipulated [33] with 47.6 (uug) in males and 21, 27 (Uug) in females. Likewise for [43] the value obtained was 35 (uug), 50.50 (uug) for [3] with 53.9 (uug), [42] with 50.87 (uug), for [4] 40 , 74 (uug) see [31] with 52.93 (uug) for [41] with 10.69 (uug) for [49], for the species *O.aureus* it was 54.4 (uug) according to [22] and for other species such as *Colossoma macropomum* the value was close to 122.46 (uug) according to [7] in *Salminus affinis* individuals the HCM value 58.5 (uug) according to [36] for [29] in the *Astronotus ocellatus* species obtained 31.07 (uug) and for *Sorubim cuspicaudus* in a study developed by [38] such value was 28.4 (uug).

The percentage value of MCHC is related to the distribution of hemoglobin within the erythrocyte [34] indicating a slightly lower concentration or irregular distribution of Hb (hypochromia and anisocromia) [13] In this study we obtained a percentage of 33.3%, which indicates that it is within the established range shown in Appendix 1, finding similarity in different studies, for example for [43], [37], [31], [41], [42],

[49] and [4] obtained values of 24.65, 27.8, 35.24, 41.14, 28.3, 38.18 and 29.14 respectively for the species *O. niloticus*. In the case of [33] obtained a total of 27.38 for males and 32.75 for females. On the other hand, as [32] reported the percentage of CHCM for non-parasitized tilapia species was 29.7, while for parasitized species it is 24.8. In other species such as *Colossoma macropomum* the value was 33.32 [7] for *Salminus affinis* was 35.5 [36], for *Astronotus ocellatus* was 33.1 [29], for *Sorubim cuspicaudus* 42.9 [38] and in *Chirostoma estor estor* was 14.9 [13].

An element of utmost importance to take into account is the correlation between the Average Corpuscular Volume and the Average Corpuscular Hemoglobin, since they are directly proportional, i. e. if one increases the other, too, however, there is a negative correlation with the Erythrocytes, since they differ with respect to the value of VCM and HCM [4].

The main components analysis (MCA) performed on the correlation matrix for the Hematological parameters in individuals of Nile Tilapia (*Oreochromis niloticus*) cultivated in Santo Domingo farm, Florencia- Caquetá showed that the first four main components (MC) contribute 84 % of the total variability, the first 3 components showed characteristic roots greater than the unit capturing 71% of the variability. When analyzing the coefficients of the vectors associated with the first two main components, it was found that the variables of greatest importance in their order were: EOS and BASO.

According to Figure 5, the first main component (MP1) with a contribution of 35.2% of the variability organized 9 individuals who presented the highest values of HETE (%) to the positive end of such component; at the negative end it organizes 7 individuals with the variables MONO (%), BASO (%), LYM (%) and EOS (%). While the second main component (MP2) with a contribution of 26.5% organized the GB and PLT with 5 individuals who presented the highest values for these variables, while PT to the negative end of such component with 12 individuals (Figure 5).

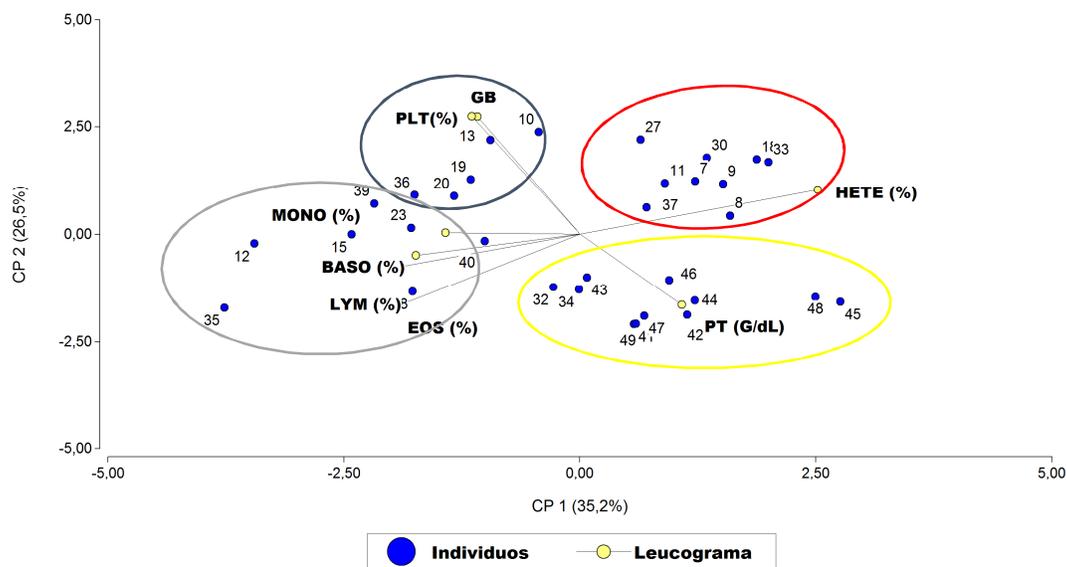


Fig 5. Main components analysis (MCA) carried out for the Hematological parameters in individuals of Nile Tilapia (*Oreochromis niloticus*) cultivated in Santo Domingo farm, Florencia-Caquetá.

IV. CONCLUSION

Once the blood studies were carried out, it was possible to show that two individuals had a Microcytoma Normochromic anemia, since their value of VCM is below the range, in addition to that 16 individuals presented Normochromic anemia. These situations can be rooted in overpopulation in the environment, which generates a decrease in oxygen levels (apoxia) and therefore stress generates a lack of appetite in individuals, which triggers anemia. On the other hand, when performing the analysis of the leucogram, some fish presented a leukopenia with lymphopenia and thrombocytosis, which indicates an immune system response to a pathogenic agent, which is why it is believed to be a bacterial septicemia.

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BIOGRAPHIES



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Engineering in 2000, from the Unified National Corporation "CUN"; her undergraduate degree was held in the city of Bogotá at the INCCA University in Colombia, which is a pioneer of the systems engineers in Colombia; in the year 2003; He graduated as a Systems Engineer. And later, the Specialization in High Speed Networks and Distributions at the INCCA University of Colombia and obtained the degree on December 5, 2008. Works in the Government of Caquetá in the Secretariat of Planning and ICT. He has also served as a teacher in the bipartisan program in the Ministries of Education and the Ministry of Defense in the "Educational Project Military Forces of Colombia", Educational Center for Youth and Adults named: PADRE LUIS DE RIZ - INFORPH OF COLOMBIA. Diploma in culture of peace and conflict, for later publication in the magazine TRASHUMANTE (American Journal of Social History) of the University of Antioquia and the Division of Social Sciences and Humanities of the Autonomous Metropolitan University (UAM), Cuajimalpa Unit (Mexico D.C).



Alba Miriam Poche Ceballos. She was born in the municipality of Belalcazar Páez cauca Colombia in 1986, academic bachelor of the educational institution Barrios Unidos del Sur. She got a degree in veterinary medicine and Zootechnics at the University of Amazonia in 2011, specialist in veterinary clinical laboratory in 2013, currently a professor of the subject clinical

laboratory, Medical surgeon in Project. She does strengthening actions to reduce canine and feline overpopulation in the department of Caquetá, analyst in clinical laboratory of small animals, linked in 2016 to the BIDA research group as a researcher in the area of analysis and clinical diagnosis in different species of animals. She has 3 scientific publications in national and international indexed journals in the area of small animals and wildlife. In addition, she participates in the co-authorship of 2 clinical diagnostic research articles.

Appendix II: Total and differential count of white blood cells

N° of individuals	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	References
neutrophils %	60	63	65	42	56	31	48	36	65	51	46	39	60	60	49	66	6,77-46,64
Lymphocytes %	30	27	24	57	35	49	39	48	28	32	37	46	29	33	34	28	48,39-75,47
eosinophils %	1	4	1	0	2	7	3	5	2	8	6	5	3	2	5	2	1,67-9,0
monocytes %	6	4	6	1	7	8	9	6	5	7	9	7	7	5	8	3	1,48-17,31
basophils %	3	2	4	0	0	5	1	5	0	2	2	3	1	0	4	1	0,43-3,18
Total	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	
Thrombocytes %	64	62	63	107	77	84	104	88	82	98	96	66	99	84	33	55	13,53-56,8

N° of individuals	17	18	19	20	21	22	23	24	25	26	27	28	29	30	31	32	References
N° of individuals	47	29	38	51	37	39	42	46	51	46	49	60	52	49	56	49	6,77-46,64
neutrophils %	39	48	49	42	48	45	47	43	38	42	43	34	36	38	39	36	48,39-75,47
Lymphocytes %	6	9	4	3	8	6	5	5	5	5	3	3	5	6	1	6	1,67-9,0
eosinophils %	8	8	8	4	4	8	3	4	5	7	5	2	7	6	3	8	1,48-17,31
monocytes %	0	6	1	0	3	2	3	2	1	0	0	1	0	1	1	1	0,43-3,18
basophils %	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	
Total	28	48	83	84	61	95	69	39	27	44	23	27	44	40	32	35	13,53-56,8