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Information to support the exemption from the Landing obligation for high survivability of red sea bream (*Pagellus bogaraveo*) after its capture by artisanal longline fisheries "voracera"

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[redacted]

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1. INTRODUCTION AND BACKGROUND

In line with the new Common Fisheries Policy and, according to the Article 15 of Regulation 1380/2013 / EU, discards from European fisheries shall be landed. However, as it appears at Article 15 (paragraph 4b), species whose scientific evaluation indicates high survival rates and complete recovery after the fishing process, may be returned to the sea.

The red sea bream (*Pagellus bogaraveo*, figure 1) is exploited in the area of the Strait of Gibraltar by an artisanal fleet based in the port of Tarifa and, in a lesser extent, in Algeciras and Ceuta. This species is caught with a specific hook art called "voracera", which is set with the help of a stone or block of concrete (figure 2) and remains in the water arround 15 to 30 minutes.

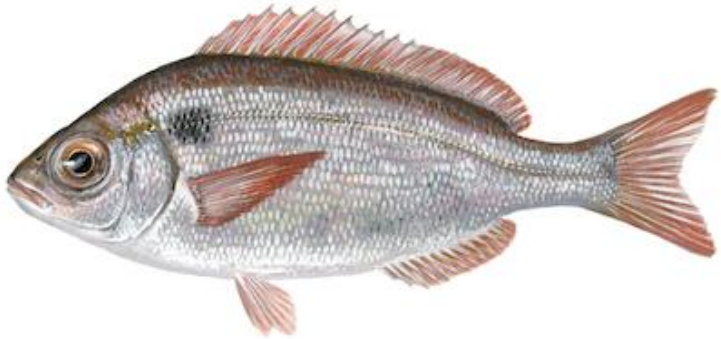

Figure 1. Red sea bream (Pagellus bogaraveo).

Figure 2. Concrete blocks used to set up the fishing gear called "Voracera", and lines of baited hooks fastened to the blocks.

The current legislation contemplates a mínimum first capture size of 33 cm, but smaller specimens under this size are usually captured. The main objective of the present study has been to evaluate the survival capacity of these specimens after have been fished with the "voracera" in the Strait of Gibraltar.

In order to evaluate the survival capacity of a species, an approach to its capacity of recovery from the fishing process is needed. It has been demonstrated that fishing is a stress process for fish (Olsen et al., 2012). The responses to stress in these organisms can be classified into primary, secondary and tertiary responses (Barton, 2002).

Primary response include the activation of the sympathetic nervous system, releasing catecholamine hormones from the chromaffin tissue (Reid et al., 1998), and the stimulation of the interrenal axis, which releases corticosteroid hormones (cortisol in teleost) to the circulatory system (Wendelaar Bonga, 1997).

The secondary response is defined as those actions produced by these hormones (Mommsen et al., 1999), summarized in the release of energy metabolites to plasma, increase in respiratory rate to favor the availability of oxygen, and increase in heart rate to mobilize these substrates throughout the body.

The tertiary response extends to the organism and population level (Wedemeyer et al., 1990), affecting the performance of the animal (growth, reproduction and behaviour), and being able even to lead to the death of the individual.

The recovery process after a stressful situation includes regain homeostasis body level, either by returning to basal state or by establishment of an allostatic condition (McEwen and Wingfield, 2003; Costas et al., 2011).

There have been numerous studies of stress responses in teleost fish (Iwama, 1998; Flik et al., 2006). Within the main metabolites released as result of secondary response, glucose and lactate are good indicators of stress in fish (Arjona et al., 2007).

In stress situations, the mobilization of glucose through the circulatory system facilitates its immediate use by those tissues that need an extra energy contribution.

On the other hand, lactate is considered a metabolic by-product of glycolysis during high energetic expenditure situations, where it occurs a state of hypoxia (Gladden, 2004).

The time required for a species to show signs of recovery varies according to the stressful agent, the environmental conditions and / or the size of the individual. In previous studies made with teleosts like gilt-head sea bream (*Sparus aurata*) or sole (*Solea senegalensis*) has been seen that secondary responses to a situation of acute stress return to baseline values between 4 and 6 hours after the stressful situation (Laiz-Carrion et al., 2005, Costas et al., 2011).

Therefore, acute stress as may be the fishing process, *a priori* will have an impact on the red sea bream (member of the sparid's family, such as the gilt-head sea bream) during the first 4-6 hours after its liberation.

If there are no signs of recovery after that time (normal movements and basal values of stress indicators in plasma), is likely that the limits of animal tolerance have been exceeded and the recovery could be endangered.

2. OBJECTIVES

Evaluate the survival capacity of non-commercial sizes (<33 cm in total length) of the red sea bream (*Pagellus bogaraveo*) after artisanal hook fishing in the Strait of Gibraltar.

This Main Objective contemplates two Specific Objectives:

1. Evaluation of the survival rates of individuals captured by commercial fisheries boats.

2. Analysis of physiological responses to stress caused by fishing to evaluate the recovery capacity of the captured animals.

3. MATERIAL AND METHODS

Three experiments were carried out: i) survival experiment on-board; ii) experiment to know the times of physiological recovery on-land; and iii) analysis of physiological responses to stress after commercial capture.

The inland experiment validate the results of the on board experience, allowing a temporary follow-up under controlled conditions.

3.1 On-board survival experiment

3.1.1 Vessel

A red sea bream capture campaign was carried out aboard the commercial ship "[redacted] "(number plate [redacted] with base port in Tarifa (Cádiz, Spain). This boat was chartered in 2010, it has [redacted] m length, [redacted] and a capacity for 5 crewmen. The shipping dates were on 7th, 8th, 10th and 13th of November 2017.

A total of 14 hauls of “voracera” (between 3 and 5 hauls per day) were made on the fishing grounds of "Bad stones" (latitude 35-36º 54-56 ', longitude 05º 48-49') and "Discoteca" (latitude 35-36º 55-56 ', longitude 05º 50-51') of the Strait of Gibraltar. The range of depths was from 128 to 247 m.

The time of permanence of the art in the water varied between 20 and 35 minutes, with an average of 10 minutes since the concrete block reached the bottom and the art were raised up to the surface. On the ship there were 4 tanks of more than 2000 L, which were pumped continuously with water from the surface of the sea.

3.1.2 Hauls and captures

12 valids hauls were made, in which a total of 102 red sea bream with a size below the commercial minimum (29.4 ± 0.2 cm total length, mean ± SEM, and a calculated weight of 378 ± 7 g).

To calculate the wet weight (in grams) of the animals was taken into account the total length (in centimeters) according to the Von-Bertalanffy equation, assuming the values of: a = 0.008 and b = 3.178. These values were obtained by the Spanish Oceanography Institute (IEO), in its headquarters in Cádiz (Spain), after years of studying the natural populations of this species.

3.1.3 Recovery in tanks

Of the 102 red sea bream captured, 66 were used for the survival experiment on-board. It was calculated an average time of 10 minutes since the art reached the bottom until the fish were hoisted on board.

Once they were embarked, the animals were immediately marked individually with a rubber label placed on the caudal peduncle and they were released into the recovery tanks.

The whole process lasted less than 30 seconds per animal, time from its release into the air until its release in the tanks. A single tank was used per haul, and the number of animals varied between 1 and 24 animals, with an average of 7 animals per haul.

3.1.4 Survival

The animals were kept in tanks for 5 hours, and the individuals deceased after that time were counted. The survival percentage was calculated for each set.

3.2 Experiment on-land to know physiological recovery times

3.2.1 Experimental conditions

To check out if the animals recover internal metabolic homeostasis after fishing, and to consider that they have recovered completely, an experiment was carried out under controlled conditions at the laboratory.

The experiment was carried out in the facilities of marine cultures of the IEO from Vigo (Spain), with red sea bream cultivated in captivity of almost 4 years old. 54 red sea breams of 25.3 ± 0.2 cm of total length, 23.5 ± 0.2 cm of fork length and 270.6 ± 6.0 g of wet weight (values represented as mean ± SEM) were used.

These animals were randomly distributed into groups of 3 animals, they were located in 18 flat-bottomed cubic tanks of 400L of capacity, and they were kept in open circuit regime for two weeks for its acclimatization.

The daily feeding was done with commercial feed and the cleaning and maintenance of the tanks were limited thus not to cause an additional stress to the fishes. The animals fasted 24 h before the experiment starts. The tanks were numbered randomly and divided into two groups: one control, and another experimental. The experimental treatment consisted in emulating the fishing process of the "voracera". For this purpose, a chase of the animals was carried out inside the tanks with hand nets for 10 minutes, estimated time of stay on the hook during commercial fishing.

This procedure has been previously tested by other research groups (Gesto et al., 2015) and is useful for assessing the level of stress and exhaustion in bone fish. The samples were taken at 0h, 5h and 24h after the stress process. 3 tanks were used for each experimental group and time. The time of 5 h was selected because the gilt-head sea bream, a member of the same family as the red sea bream, presents an appreciable recovery in its plasma levels of cortisol and energy metabolites 4 h after acute stress (Laiz-Carrion et al., 2005).

3.2.2 Sampling

For sampling, all 3 animals from the same tank were captured by hand nets at the same time and immediately transferred to a supplementary tank with 100 L of seawater and 2-phenoxyethanol at a concentration of 0.5 mL /L seawater (0.05%, v / v). After anesthesia, the animals were weighed and measured and a blood sample was extracted from the caudal peduncle with heparinized syringes.

After this process, the animals were returned to their original tanks in less than 4 minutes since its capture. The blood was centrifuged at 10000 g during 3 minutes at 4 ° C and the plasma was separated and frozen at -80 ° C until analysis. Almost all animals were recovered after the experiment, but a total of 6 individuals died in the process of recovery after sampling. It should be noted that 4 of the animals that died belonged to the control group, while the other 2 belonged to the group stressed, but from the times 5 h and 24 h post-stress.

3.3 Analysis of physiological responses to stress after commercial capture

On board the ship used for the survival experiment an analysis of physiological responses after fishing was made.

For this test, 36 animals from 7 different hauls were selected (varying between 2 and 7 animals per set, with an average of 5 animals per haul) which were marked individually with rubber bands in the caudal peduncle, and sampled immediately after capture (10 minutes of fight on the hook followed by 15 seconds of exposure to the air).

200 microliters of blood were taken from the caudal peduncle with heparinized syringes and the animals were released in 2000 L tanks for recovery.

A recovery tank was used for each haul. After 5 h, the animals were immediately captured using hand nets, wrapped in a damp cloth to avoid additional stress, and a new sample of 200 microliters of blood from the caudal peduncule was taken. The Individual marking allows to correlate the blood samples from 0 h to 5 h after fishing and recovery. The tubes with blood were centrifuged at 10000 g for 3 minutes at 4 ° C. The plasma was separated and frozen in liquid nitrogen and then stored at -80 ° C.

3.4 Variables analyzed

From blood plasma taken during the on-land on-board experiments cortisol, glucose, lactate, triglycerides, proteins and osmolality were analyzed, being these the main biomarkers of acute stress in bone fish.

Cortisol was analyzed using a commercial kit (Arbor Assay, MI, USA). Glucose, lactate and triglycerides were analyzed by commercial kits (Spinreact SA, Girona, Spain), as well as total proteins (Pierce BCA Protein Assay Kit, Thermo Scientific, IL, USA).

The osmolality was measured by a vapor pressure osmometer (Vapro 5520 Osmometer, Wescor, USA). All analyzes except osmolality were performed using a microplate reader (Bio-Tek Instruments, Winooski, VT, USA), and using the KCJunior Data Analysis program for Microsoft Windows XP.

3.5 Statistics

Normality and homoscedasticity were analyzed by the Shapiro-Wilk's and Levene's tests, respectively. The differences between groups for the physiological experiment on-land were calculated by means of a two-way ANOVA with the group (control and stress) and the time (0, 5 and 24 h) as variance factors.

For the experiment of physiological recovery of the boat the differences between groups were calculated by a repeated measures ANOVA with the haul number and the time (0 and 5 h) as variance factors. Logarithmic transformations of the data were made when it was necessary to meet the requirements of the ANOVA.

When the ANOVA results showed significant differences, the Tukey's test was used as *a posteriori* test.

The Student's t-test for paired samples was used to evaluate the differences between the physiological parameters of the experiment done on-board once it was established that the factor haul did not affect the dependent variables analyzed. Statistical significance was established in $p < 0.05$. All results are shown as mean ± SEM.

4. RESULTS AND DISCUSSION

4.1 Survival rates on-board

The survival rates of the two on-board experiments were calculated. As there were no significant differences between the survivals of the groups captured in each haul ($p < 0.05$, Student's t), we proceeded to unify the groups of each haul, constituting duplicates of the same sample (haul). Therefore, it made a total of 12 valid hauls during a 4-day campaign in the Strait of Gibraltar, with 102 animals captured.

The result of this study shows a figure of survival of 90.6 ± 6.2% after 5 h of recovery in on-board tanks. If we have in mind that the animals evaluated were in similar conditions, but not accurate, to those of the natural environment, this figure could be even higher in case of direct release to the ocean.

Although other factors should be taken into account, such as post-capture predation. However, the figures are very similar to those calculated in other discarded species such as the small-spotted catshark (*Scyliorhinus canicula*) captured by bottom trawling (Revill et al., 2005), being one of the species considered more resistant from the spanish south-Atlantic area (Barragán-Méndez, Ruiz-Jarabo et al., Article sent for publication).

Although survival rates were greater than 90%, it is important to confirm if the animals truly reach the recover after the recovery period or not.

Through a physiological approach can be established if the red sea bream captured arrived in exhaustion conditions until the no return point.

Therefore, to know the animals state after capture, and after recovery time is essential to be able to support the hypothesis that the red sea bream which remains alive after the capture would survive if they were released to the environment.

4.2. Evaluation of physiological recovery at the laboratory

4.2.1 Cortisol

Plasma cortisol is the main glucocorticoid of teleost fishes (Mommsen et al., 1999). The increase of it in the red sea bream subjected to a process of acute stress like the fishing process (figure 3) is necessary to mobilize energy reserves during the first moments.

Our results at the laboratory coincide with those of other fishes such as gilthead seabream (*S. aurata*) or sole (*S. senegalensis*) subjected to acute stress (Laiz-Carrion et al., 2005; Costas et al., 2011).

Although there is no significant differences in the stressed group between 0 and 5h, a decrease of a third part of the concentration of cortisol happened between one time and another.

In such a way, we can say that a period of 5 hours is enough for the red sea bream (*P. bogaraveo*) to start showing signs of recovery at the level of primary markers of stress. It worth to pointing out the high concentrations of cortisol present in plasma of animals from the control group (40.2 ± 7.7 ng Cortisol mL^{-1}).

Previous studies show that plasma cortisol in teleost fish in basal conditions oscillate around 20 ng mL-1 (Ellis et al., 2012; Louison et al., 2017), although there may be variations depending on the kind of analysis, the species studied, the population or the time of year.

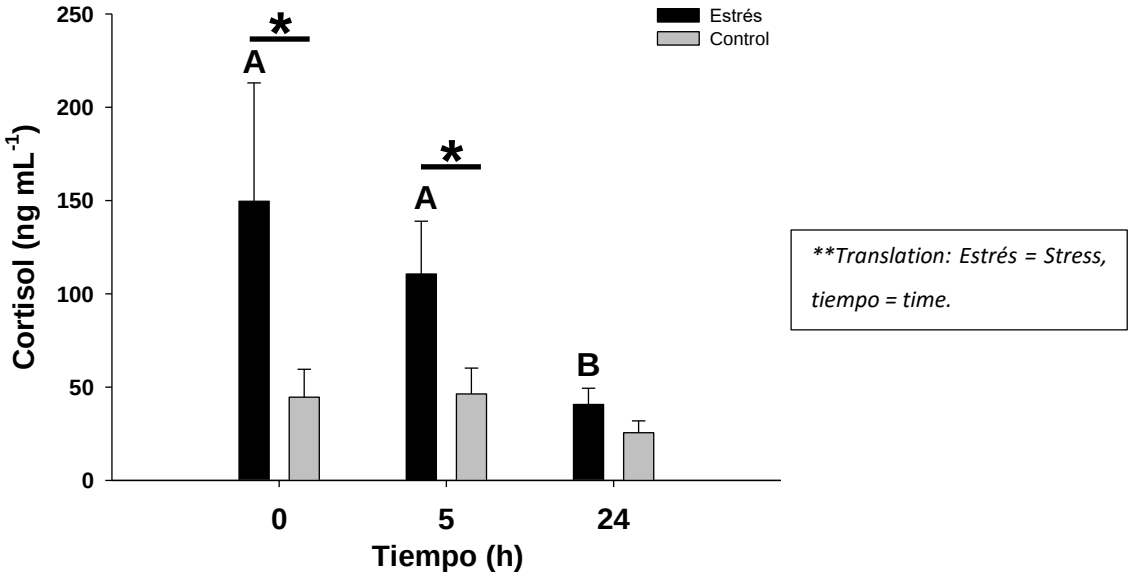

Figure 3. Plasma cortisol in Pagellus bogaraveo subjected to persecution (similar stress as that suffered by the fishing with a hook) for 10 minutes, with recovery afterwards and sampling at 0, 5 and 24 h after stress. Black bars indicate the stressed group, and gray bars indicate the control group. Different letters indicate significant differences between times for the stressed group; while the asterisk (*) indicates differences between the control group and the stressed group for a specific time ($p < 0.05$, two-way ANOVA followed by the Tukey's test). *Black columns indicate stressed group, grey ones control group.*

4.2.2 Energy metabolites and osmoregulatory parameters

The stress caused by the fishing simulation causes the release of energy reserves to the blood (Table 1). After a stressful situation, typical secondary responses promoted by that hormones release (such as cortisol) include mobilisation of glucose and, under anaerobic conditions, the appearance of lactate. These responses described coincide with our results. In time 0 h after stress, the red sea bream begins to release glucose to the blood, which shows its maximum concentration at 5 h after the start of recovery.

This late increase (no noticeable 10 minutes after acute stress) matches what has been described previously in the senegalese sole (Costas et al., 2011) and the small-spotted catshark (Barragán-Méndez, Ruiz-Jarabo et al., Article sent for publication), and it may be due to a very high glucose consumption in the first moments post-stress, preventing the increase in the concentration of plasma glucose until the rate of metabolic consumption decreases.

Nevertheless, the lactate concentration in the present study is significantly greater to that in the control group after 10 minutes of pursuit, which indicates an immediate activation of anaerobic metabolism in this species, in line with other teleosts (Frisch and Anderson, 2005, Costas et al., 2011).

Proteins and triglycerides (TAG) do not seem to vary in the red sea bream subjected to a process of acute stress, so it can be deduced that the main metabolites of consumption at short term are others (glucose, for example).

Parameter	Group	0 h	5 h	24 h
Glucose	**Stress**	3.97 ± 0.26 B	5.54 ± 0.39 A*	4.26 ± 0.14 B
(mmol L^{-1})	**Control**	3.18 ± 0.15	3.19 ± 0.12	3.46 ± 0.09
Lactate	**Stress**	2.80 ± 0.46 A*	2.30 ± 0.32 A	1.06 ± 0.12 B
(mmol L^{-1})	**Control**	1.07 ± 0.08	1.40 ± 0.12	1.24 ± 0.08
TAG	**Stress**	0.79 ± 0.09	0.64 ± 0.08	0.65 ± 0.07
(mmol L^{-1})	**Control**	0.75 ± 0.04	1.01 ± 0.18	0.74 ± 0.12
Proteins	**Stress**	28.1 ± 1.0	23.6 ± 0.8	24.6 ± 1.5
(mg dL^{-1})	**Control**	27.5 ± 1.4	25.7 ± 0.8	27.5 ± 1.3
Osmolality	**Stress**	291 ± 3 A*	260 ± 2 B	257 ± 2 B
(mOsmol kg^{-1})	**Control**	264 ± 4	267 ± 1	263 ± 1

Table 1. Plasma glucose, lactate, triglycerides (TAG), proteins and osmolality in Pagellus bogaraveo subjected to persecution (similar stress as that suffered by the fishing with a hook) for 10 minutes, with recovery afterwards and sampling at 0, 5 and 24 h after stress. Different letters indicate significant defferences between the control group and the stressed group for a specific time ($p < 0.05$, two-way ANOVA followed by the Tukey's test).

Regarding changes in the osmoregulatory system, the present study shows an increase in plasma osmolality in the group subjected to acute stress at time 0h (Table 1).

Due to the initial increase in energy expenditure produced by the process of stress, other systems are harmed and suffer imbalances that, once finished the process, they return to their basal state.

These kind of responses are normal in marine fish, osmoregulators, because they have an internal fluids' osmolality lower to that of the external environment. After a stressful situation, a series of responses such as vasodilation of blood vessels increasing the surface of contact between the blood and the external environment (hypertonic), which lead to a passive dehydration of the animal (McCormick et al., 2013). Our results show that the red sea bream is able to return to control levels after 5 h after acute stress.

4.2.3 Mortality at the laboratory

In the experiment carried out under controlled conditions, there was a certain mortality rate in non-stressed animals (control) and in those recovered after a acute stress situation.

The cause of this mortality can be the release of catecholamines and cortisol in the chromaffin and interrenal tissue as described in sea bass (*Dicentrarchus labrax*) (Rotllant et al., 2006), causing heart failure in healthy animals and / or not exhausted.

The high levels of cortisol (Figure 3) found in this species seem support this hypothesis. In this way, healthy animals that have not been subjected to physical fatigue during the process of capture, can suffer a cardiac arrest. However, the red sea bream that have exhausted a certain amount of energy fighting during its capture process (escaping from a net or trying to free himself from a hook, for example) are more likely to survive than those caught quickly.

4.3. Evaluation of physiological recovery after fishing

4.3.1 Cortisol

Plasma cortisol levels in red sea bream caught in the Strait of Gibraltar are shown in Figure 4. A significant reduction in cortisol concentration is observed after 5 h of recovery in the tanks on board. The concentrations described in this experiment coincide with those of the experiment on land ($p > 0.05$ Student's t), so that a similarity between both experimental conditions can be inferred.

It has been described that the fishing process raises cortisol levels in teleosts (Addis et al., 2012). In this way, the red sea bream captured in natural environment show signs of physiological recovery after 5 hours. In other studies made with teleosts captured in natural environment, there are no differences due to the fishing process or exposure to air (Methling et al., 2017). It can be because the animals in that experiment were not fully recovered, or the responses to fishing are species-dependent. Other studies show a increase in cortisol levels 24 h after suffering a peeling similar to that produced by purse-seine fishing (Olsen et al., 2012), which recover between 4 and 7 days after stress.

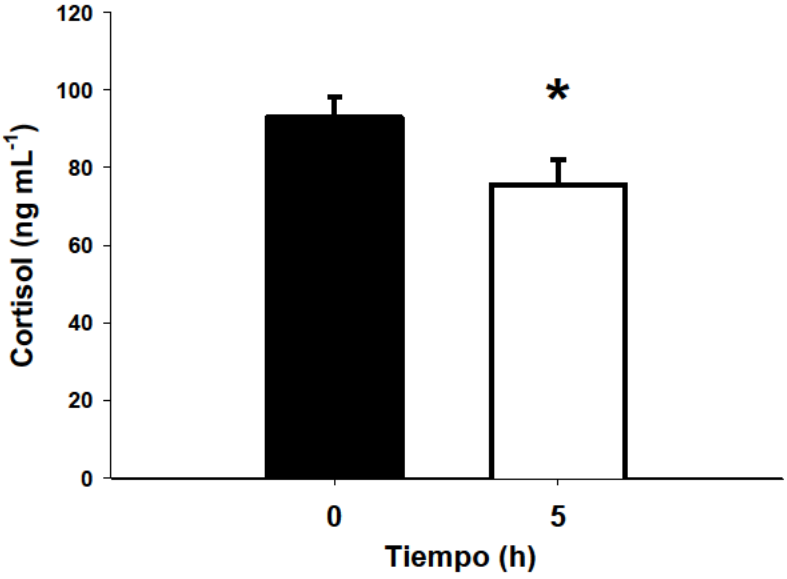

Figure 4. Plasma cortisol in *Pagellus bogaraveo* caught in the Strait of Gibraltar through the art of fishing "voracera". Samples taken just after the fishing (0 h) and after 5 hours of recovery in tanks on board (5 h). The asterisk (*) indicates significant differences between the two sampling times ($p<0.05$, Student's t for paired samples).

***Translation: Tiempo = time*

4.3.2 Energy metabolites and osmoregulatory parameters

The results of the experiment on-board the commercial fishing vessel indicate a physiological recovery of the animals after 5 h in the tanks (Table 2). The levels of glucose at time 0 h of the ship coincide with the levels of the group stressed at 0 h of the experiment on-land, although the glucose levels after 5 h on the ship are greater than those on-land at the same time. This difference may be due to processes such as feeding / fasting of the red sea bream in both experiments.

The levels of lactate from both experiments (ship and land) coincide at times 0 h and 5 h respectively, which reinforces the idea that both experimental approaches are comparable in terms of intensity and duration of the stress agent. Thus, we can talk about a recovery process in the red sea bream captured after a fishing with "voracera". Reinforcing this idea are the plasma osmolality data, which decreases in both experiments after 5 h of recovery.

The GAT and protein results indicate plasma differences between wild animals and those kept in captivity. In this way, the response of the first includes the mobilization of plasma proteins after capture (0 h), while the red sea bream experiments on-land did not include this response. These differences can be due to the different composition of the diets of both groups, as well as to the fact that the captive animals were fasting 24 h before the experiment, while the last ingestion of wild animals is unknown.

Table 2. Plasma glucose, lactate, triglycerides (TAG), proteins and osmolality in *Pagellus bogaraveo* caught in the Strait of Gibraltar through the fishing gear of "Voracera". Samples taken after fishing (0 h) and 5 hours after recovery in tanks on board. The asterisk (*) indicates significat differences between the sampling times (p<0.05, Student's t for paired samples)

Group	Glucose (mmol L^{-1})	Lactate (mmol L^{-1})	TAG (mmol L^{-1})	Proteins (mg dL^{-1})	Osmolality (mOsmol kg^{-1})
0 h	4.4 ± 0.1	3.2 ± 0.2	1.8 ± 0.2	26.7 ± 0.5	302 ± 3
5 h	8.8 ± 0.4 *	2.1 ± 0.2 *	1.6 ± 0.2	21.5 ± 0.4 *	291 ± 3 *

5. CONCLUSIONS

1) Red sea bream (*P. bogaraveo*) with a total length smaller than 33 cm, caught in the Strait of Gibraltar through the art of fishing called "voracera", present survival rates of 90.6 ± 6.2%.

2) The surviving animals manage to recover their basal homeostatic levels, being able to speak of an effective physiological recovery between 5 and 24 hours after the capture.

3) This study was carried out during the month of November of the year 2017, in certain environmental conditions (temperature, salinity, etc.), so that the conclusions have to take into account this limitation. However the Strait of Gibraltar does not have a great variation in these conditions throughout the year, so a similar survival and recovery rates are expected during other periods, although complementary studies should be carried out ensure.

6. REFERENCES

Addis, P., Secci, M., Locci, I., and Cau, A. C. 2012. Harvesting, handling practices and processing of bluefin tuna captured in the trap fishery: possible effects on the flesh quality. Collective Volume of Scientific Papers ICCAT, 67: 390-398.

Arjona, F. J., Vargas-Chacoff, L., Ruiz-Jarabo, I., Martin del Rio, M. P., and Mancera, J. M. 2007. Osmoregulatory response of Senegalese sole (*Solea senegalensis*) to changes in environmental salinity. Comp Biochem Physiol A Mol Integr Physiol, 148: 413-421.

Barton, B. A. 2002. Stress in fishes: a diversity of responses with particular reference to changes in circulating corticosteroids. Integrative and Comparative Biology, 42: 517-525.

Costas, B., Conceicao, L., Aragao, C., Martos, J. A., Ruiz-Jarabo, I., Mancera, J., and Afonso, A. 2011. Physiological responses of Senegalese sole (*Solea senegalensis* Kaup, 1858) after stress challenge: Effects on non-specific immune parameters, plasma free amino acids and energy metabolism. Aquaculture, 316: 68-76.

Ellis, T., Yildiz, H. Y., Lopez-Olmeda, J., Spedicato, M. T., Tort, L., Overli, O., and Martins, C. I. 2012. Cortisol and finfish welfare. Fish Physiol Biochem, 38: 163-188.

Frisch, A., and Anderson, T. 2005. Physiological stress responses of two species of coral trout (*Plectropomus leopardus* and *Plectropomus maculatus*). Comp Biochem Physiol A Mol Integr Physiol, 140: 317-327.

Gesto, M., Hernandez, J., Lopez-Patino, M. A., Soengas, J. L., and Miguez, J. M. 2015. Is gill cortisol concentration a good acute stress indicator in fish? A study in rainbow trout and zebrafish. Comp Biochem Physiol A Mol Integr Physiol, 188: 65-69.

Gladden, L. B. 2004. Lactate metabolism: a new paradigm for the third millennium. J Physiol, 558: 5-30.

Laiz-Carrion, R., Guerreiro, P. M., Fuentes, J., Canario, A. V. M., Martin Del Rio, M. P., and Mancera, J. M. 2005. Branchial osmoregulatory response to salinity in the gilthead sea bream, *Sparus auratus*. Journal of Experimental Zoology Part A Comparative Experimental Biology, 303A: 563-576.

Louison, M. J., Adhikari, S., Stein, J. A., and Suski, C. D. 2017. Hormonal responsiveness to stress is negatively associated with vulnerability to angling capture in fish. J Exp Biol, 220: 2529-2535.

McCormick, S. D., Farrell, A. P., and Brauner, C. J. 2013. Euryhaline fishes, Academic Press, Oxford, UK.

McEwen, B. S., and Wingfield, J. C. 2003. The concept of allostasis in biology and biomedicine. Hormones and Behavior, 43: 2-15.

Methling, C., Skov, P. V., and Madsen, N. 2017. Reflex impairment, physiological stress, and discard mortality of European plaice *Pleuronectes platessa* in an otter trawl fishery. ICES (International Council for the Exploration of the Sea) Journal of Marine Science, 496: 207-218.

Mommsen, T. P., Vijayan, M. M., and Moon, T. W. 1999. Cortisol in teleosts: dynamics, mechanisms of action, and metabolic regulation. Reviews in Fish Biology and Fisheries, 9: 211-268.

Olsen, R. E., Oppedal, F., Tenningen, M., and Vold, A. 2012. Physiological response and mortality caused by scale loss in Atlantic herring. Fisheries Research, 129: 21-27.

Reid, S. G., Bernier, N. J., and Perry, S. F. 1998. The adrenergic stress response in fish: control of catecholamine storage and release. Comparative Biochemistry and Physiology C, 120: 1-27.

Revill, A. S., Dulvy, N. K., and Holst, R. 2005. The survival of discarded lesser-spotted dogfish (*Scyliorhinus canicula*) in the Western English Channel beam trawl fishery. Fisheries Research, 71: 121-124.

Rotllant, J., Ruane, N. M., Dinis, M. T., Canario, A. V., and Power, D. M. 2006. Intra-adrenal interactions in fish: catecholamine stimulated cortisol release in sea bass (*Dicentrarchus labrax* L.). Comp Biochem Physiol A Mol Integr Physiol, 143: 375-381.

Wedemeyer, G. A., Barton, B. A., and McLeay, D. J. 1990. Stress and acclimation. *In* Methods of Fish Biology, pp. 451-489. Ed. by C. B. Schreck, and P. B. Moyle. American Fisheries Society, Bethesda MD.

Wendelaar Bonga, S. E. 1997. The stress response in fish. Physiological Reviews, 77: 591-625.