University of Applied Sciences





# **FINAL REPORT**

Detecting bones under water using a forensic light source

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# Final report

# A study of detecting bones under water using a forensic light source

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#### Dear reader,

In front of you, you find the final report I made during my gradutation internship at Loci Forensics B.V. during the period of August 29<sup>th</sup> until April 3<sup>th</sup>. In this report my research, in which I try to find a reliable method to detect bones under water, will be discussed.

This report is written on behalf of the studies Forensic Research at Saxion University of Applied Sciences in Enschede, The Netherlands.

The period I had at Loci Forensics B.V. was an amazing opportunity in which I have learned to develop my research skills. For that, I want to give out a special thanks to Martin Eversdijk and René Gelderman of Loci Forensics B.V. They gave me the chance and space to learn on a level I could never have asked for and were always there when I needed. I also want to thank the Arrest Team of the Amsterdam Police Department for their help during my experiment at the Vinkeveense Plassen in Utrecht. Without them I could not have conducted this experiment. Besides, I want to thank the people I worked with during my internship, social as well as work related. Last, I would like to thank Ton Lemmens for the supervision from Saxion University of Applied Sciences.

I hope you enjoy reading my research!

Kind regards,

**Romy Regterschot** 

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# Nederlandse samenvatting

Door het slechte zicht onder water in Nederland zijn botten nauwelijks te herkennen, of te herkennen als botten onder water. Dit slechte zicht wordt veroorzaakt door verschillende factoren zoals lichtinval, de in het water aanwezige deeltjes en het ecosysteem van het water. Omdat de botten niet worden herkend of gedetecteerd, worden stoffelijke resten niet ontdekt. Het doel van dit onderzoek was om bot(delen) beter zichtbaar te maken onder water, zodat deze makkelijker te vinden zijn voor duikers. Dit gebeurt me behulp van een forensische lichtbron (FLS).

De FLS wordt gebruikt voor het visualiseren van biologisch bewijs zoals speeksel, sperma, bloed en urine. De FLS kan het contrast tussen het biologische bewijs en de achtergrond vergroten of het bewijs fluorescentie geven. Waar vlekken niet zichtbaar zijn met het blote oog, wordt de FLS gebruikt om het contrast tussen het bewijs en de achtergrond te vergroten. Fluorescentie treedt op wanneer een FLS licht uitzendt naar het biologische bewijs. Het biologische bewijs absorbeert het licht op een bepaalde golflengte en zendt vervolgens de geabsorbeerde energie opnieuw uit als licht op een langere golflengte. Botten fluoresceren wanneer bepaalde golflengten van licht erop worden uitgezonden. Fluorescentie van het bot neemt af met de tijd, wat kan worden gerelateerd aan de afbraak van collageen.

In dit onderzoek worden verschillende variabelen getest om tot een methode te komen die helpt bij het zoeken naar botten onder water. Een van de variabelen is de golflengte van het licht waarmee de botten worden aangestraald. Er worden ook verschillende filters gebruikt om een duidelijker zicht te krijgen op het mogelijke gebruik van filters bij het detecteren van botten. Daarnaast worden verschillende degradaties van botten gebruikt, evenals verschillende afstanden tussen de forensische lichtbron en de botten. Als laatst wordt een reeks mogelijke vals positieven getest.

De golflengte die de bot(delen) doet fluoresceren is 390 nm. Wanneer de botten werden belicht met licht met golflengtes van 365, 405 of 455 nm werd geen fluorescentie waargenomen. Op kleinere afstand is het licht dat op de botten valt meer gebundeld dan op grotere afstand. Met de onderzochte afstanden kon geen conclusie worden getrokken over de optimale afstand tussen de FLS en de botten omdat er geen significante afname van fluorescentie werd waargenomen. Het gebruik van een FLS bij het verlichten van bot(delen) met een golflengte van 365, 390, 405 of 455 nm is het best wanneer er geen gebruikt gemaakt wordt van filters. Verder wordt er geen zichtbaar verschil in fluorescentie waargenomen tussen de verschillende degradatieniveaus. De mogelijke vals positieven fluoresceren niet.

Om tot een meer betrouwbare uitkomst van het onderzoek te komen moet er een vervolgonderzoek worden uitgevoerd. Tijdens dit vervolgonderzoek kunnen de in dit onderzoek gebruikte variabelen opnieuw getest worden. Daarnaast is het waardevol om deze variabelen uit te breiden en te veranderen.

Sleutelwoorden: Forensische lichtbron; fluorescentie; botweefsel

# English summary

Due to the bad under water visibility in the Netherlands, bones can hardly be detected or recognized as bones under water. This bad visibility is caused by various factor such as incidence of light, the particles present in the water and the ecosystem of the water. Due to the bones not being recognized or detected, mortal remains are not discovered. The purpose of this research was to make bone(parts) more visible under water so divers can find them easier. This is done with the use of a forensic light source (FLS).

The FLS is used for visualizing biological evidence such as saliva, semen, blood and urine. The FLS can either increase the contrast between the biological evidence and the background or make the evidence fluorescence. Where stains are not visible to the naked eye, the FLS is used to increase the contrast between the evidence and background. Fluorescence occurs when a FLS emits light to the biological evidence. The biological evidence absorbs the light at a particular wavelength and subsequently reemits the absorbed energy as light at a longer wavelength. Bones fluoresce when certain wavelengths of lights are emitted to them. Fluorescence of the bone decreases with time, which can be related to the degradation of collagen.

In this research, different variables are tested in order to create a method that will help search for bones under water. One of the variables is wavelength of the light of which the bones are emitted with. Different filters are also used to create a more clear sight of the possible use of filters when detecting bones. Different degradations of bones are used as well as different distances between the forensic light source and the bones. At last, a range of possible false positives are tested.

The wavelength which makes the bone(parts) fluoresce in this research is 390 nm. When the bones were illuminated with light with wavelengths of 365, 405 or 455 nm, no fluorescence was observed. No conclusion regarding distance between the forensic light source and the bones could be made with the distances examined because no significant decrease of fluorescence was observed. Using a forensic light source when illuminating bone(parts) with a wavelength of 365, 390, 405 or 455 nm is best when no yellow, orange or red filter is used but when observed with the use of the naked eye. No visible difference in fluorescence is observed between the different degradation levels and the possible false positives did not fluoresce.

In order to get a more reliable outcome, a follow up research must be done. In this follow up research the variables tested in this research should be tested again. When these are tested again, the variables can be changed and expanded.

Keywords: Forensic light source; fluorescence; bone tissue

# 1. Introduction

#### 1.1 Inducement

In the Vinkeveense Plassen at Utrecht, a diver is doing his weekly training with his training group. When the diver hits the bottom of the water, he feels around with his hands over the surface of the bottom and got a hold of something. When looking closely to the object he found, he realizes it is something that may be a skull. He takes it up to the surface of the water and realizes that he was right. He immediately contacted the local Police Force to come and investigate. When the Police arrived, they confirmed that the finding of the diver is indeed a human skull. The Police has reason to believe that the rest of the remains could be found in the Vinkeveense Plassen as well and send a diver to the place that the skull was found. However, the visibility under water of the Vinkeveense Plassen is nil and it is therefore almost impossible to find the rest of the remains. For the diver who is searching for the rest of the remains, it is difficult to detect bones in the water because the field of view here is not optimal. When using a light source which only uses white light, bones are as visible as all of the other things in the water. Although it is of great importance that the bones are found, none of the other remains are found that day.

It is of great importance that the rest of the remains are found. Not only for the family of the deceased who wants to get their family member back and give the deceased a proper burial, but also for the forensics in order to be able to conduct a fully reliable research of what happened. Next to that, it can be a traumatizing experience for the person who finds the remains of the deceased in the water.

#### 1.2 Problem analysis

Due to the bad under water visibility in the Netherlands, bones can hardly be detected or recognized as bones under water. This bad visibility is caused by various factor such as incidence of light, the particles present in the water and the ecosystem of the water. Due to the bones not being recognized or detected, mortal remains are not discovered.

#### 1.3 Purpose and research questions

The purpose of this research was to develop a method that makes bone(parts) more visible under water when using a forensic light source. This is done in order to make the bones better detectable for divers which can create more quantity and deficiency in the detection of the bones. In order to achieve this, different experiments were conducted using multiple variables. To make this research reliable, the following main- and subquestions are answered (table 1.1):

| Main question:  | To what extent can a forensic light source make bone(parts) of a full grown        |
|-----------------|--|
|                 | human being visible in natural water in the Netherlands?                           |
| Sub question 1: | At what wavelength of UV-light are the bone(parts) most visible?                   |
| Sub question 2: | What does the distance between the bone(parts) and the forensic light source do    |
|                 | to the degree to which bone(parts) are made visible by the forensic light source?  |
| Sub question 3: | What can filters on the forensic light source contribute to making the bone(parts) |
|                 | more visible?  |

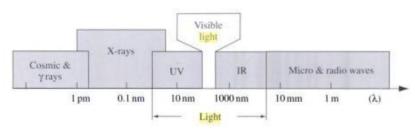
| Sub question 4: | What difference can be observed in the degree of fluorescence of the bone(parts) |
|-----------------|--|
|                 | when using the forensic light source when bone(parts) of different degradation   |
|                 | levels are used?   |
| Sub question 5: | Do other components in the water light up when using the forensic light source?  |
|                 | If yes, which?   |

Table 1.1: Main- and sub questions of this research

# 2. Theoretical background

#### 2.1 Forensic light source and barrier filters

Light is a form of electromagnetic energy and is regarded as a wave phenomenon [1]. Light consists of three sub parts: Ultra violet (UV), visible and Infra-red (IR). Those three sub parts are all part of the electromagnetic spectrum (figure 2.1) [2] [3]. Visible light has a wavelength of approximately 400 to 700 nm, which includes the range of human visibility. The visible light spectrum is based on increasing wavelength and decreasing energy [3]. A single electromagnetic ray of 450 nm is seen as blue light, a ray of 550 nm as green or yellow light and a ray of 650 nm as red or orange light. The combination of electromagnetic rays between 400 and 700 nm is seen as white light [2] [3].



#### Figure 2.1: The electromagnetic spectrum [3]

The term forensic light source (FLS) is commonly used to refer to an illumination system adapted to crime scene examination [2], such as laser and high-intensity filtered light sources. A FLS without a laser is called an alternate light source (ALS) [4]. The FLS is used for visualizing biological evidence such as saliva, semen, blood and urine [5]. The FLS can either increase the contrast between the biological evidence and the background or make the evidence fluorescence. Where stains are not visible to the naked eye, the FLS is used to increase the contrast between the evidence and background [4]. However, this research will focus on fluorescence of the evidence. When light strikes matter, it can absorb the light partly or entirely or the light passes through the matter. If the matter absorbs the light (partly), energy is transferred to the molecule. Every molecule has energy levels and can go to a higher energy level by the absorption of a particular quantum of light [1]. Fluorescence occurs when a FLS emits light to the biological evidence (matter). The biological evidence absorbs the light at a particular wavelength and subsequently re-emits the absorbed energy as light at a longer wavelength (figure 2.2) [4].

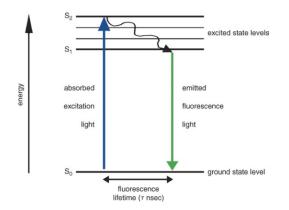


Figure 2.2: Jablonski Diagram. This figure shows the energy states of a molecule. The ground state of a molecule is equal to S0, the excited states are in between S1 and S2 [6].

The human eye has the highest sensitivity for green and yellow light, which is around 550 nm (figure 2.3) [2] [3]. However, the re-emitted light will not always be of that particular wavelength. In order to properly see the re-emitted light, barrier filters are used. These filters ensure that reflected excitation light is blocked and that the fluorescence of the object of interest is transmitted. The object of interest is seen as a bright image against a dark background, when the barrier filter is placed in front of the eye [7]. A barrier filter is selected by determining the wavelength, and thus the colour of the light, that the object of interest emits. The colour of the barrier filter is complementary to the light emitted [2]. To select the colour of the filter, the colour wheel can be used. Colours that are opposite on the wheel are complementary (figure 2.4) [2].

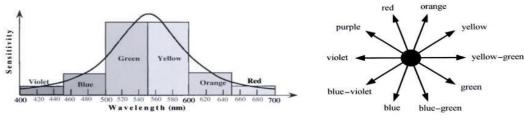


Figure 2.3: The spectral sensitivity of the human eye [2]



#### 2.2 Bones

#### 2.2.1 Molecular structure of bone

Bone is composed of organic material, inorganic or mineral material and water. The wet weight of the bone consist of approximately 65 to 70 percent inorganic material [8] [9] [10] [11]. The purpose of the inorganic matrix is to store most of the ions of the human body (99% of calcium, 85% phosphorus and 40 to 60% of the body's magnesium and sodium) [8]. These ions contribute in giving the bone most of its stiffness by forming crystalline structures of hydroxyapatite that surround and are within the collagen fibres [8] [9]. These crystalline structures are a form of calcium phosphate [9].

The organic component of bone consists for 90 percent of collagen, which is the most occurring protein in the human body [9]. Collagen is responsible for giving bone its flexibility. The balance of collagen and minerals gives the bone its flexibility and stiffness requirements [8]. The other 10 percent of the organic component of bone consists non-collagenous proteins such as fibronectin, osteopontin, osteocalcin and bone sialoprotein. It also consists proteoglycans such as decorin en biglycan [8].

#### 2.2.2 Degradation of bone

The hard tissue of the human body degrades when a human being deceases [10]. This research is concerned with bone degradation under water. However, not much is known about this. In order to conduct this research, bone degradation in (wet) soil is used as a reference because it takes humidity in consideration.

Both the mineral and the organic components of bone degrade. This happens in three principal mechanisms: the chemical dissolution of bone mineral, attack from soil-dwelling micro-organisms and the chemical breakdown of collagen [10].

#### Chemical dissolution of bone mineral

Water in the soil causes the chemical dissolution of bone. Dissolution of bone mineral may occur because bone is slightly soluble in water. This may occur when bone is in contact with damp soil with the external surfaces as well as the pore spaces in the bone. Once the dissolution has started, a loop is initiated: when the bone porosity increases, more water is able to touch the inner and outer surface of the bone, which means a greater dissolution of bone mineral is possible [10].

#### Microbial degradation

Microbial degradation is caused by soil-dwelling micro-organisms, such as bacteria. The bacteria attack collagen fibrils by removing the hydroxyapatite surrounding the fibril, then they break down the collagen which increased the bone porosity. The difference with chemical dissolution of bone mineral is that the pores in microbial degradation are smaller and demineralised zones are often surrounded by zones of increased mineral density. Microbial degradation begins on the inside of the bones, where chemical dissolution of bone mineral includes both the inside and the outside [10].

#### Collagen hydrolysis

Collagen hydrolysis is a mechanism that brakes down collagen by hydrolysis. This mechanism does not affect the microscopic structure of bone as is only causes an increased number of very small pores. Loss of collagen is only occurring over thousands of years [10].

#### 2.3 Fluorescence of bone

Bones fluoresce when certain wavelengths of lights are emitted to them (<u>figure 2.5</u>). It is said that the proteins in the collagen of the bone are largely responsible for the fluorescent properties of the bone [12], while other scientists say that it is still inconclusive which part of the bone causes fluorescence [13]. Fluorescence of the bone decreases with time, which can be related to the degradation of collagen [12].

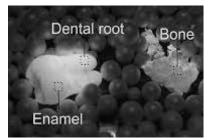


Figure 2.5: Fluorescence of bone and dental root

# 3. Research strategy

#### 3.1 Purpose

The purpose of this research was to develop a method that makes bone(parts) more visible under water when using a forensic light source. This is done in order to make the bones better detectable for divers which can create more quantity and deficiency in the detection of the bones.

#### 3.2 Substantiation

In order to develop a method that makes bone(parts) (more) visible under water, experiments will be done. The experiments will be conducted in two different stages. The experiments will consider the following variables in order to answer the following sub questions (table 3.1):

| Wavelength of the       | At what wavelength of UV-light are the bone(parts) most visible?        |
|-------------------------|---|
| forensic light source   |   |
| Distance between light  | What does the distance between the bone(parts) and the forensic light   |
| source and the bone     | source do to the degree to which bone(parts) are made visible by the    |
|                         | forensic light source?  |
| Filters in combination  | What can filters on the forensic light source contribute to making the  |
| with the forensic light | bone(parts) more visible?   |
| source                  |   |
| Bone(parts) with        | What difference can be observed in the degree of fluorescence of the    |
| different levels of     | bone(parts) when using the forensic light source when bone(parts) of    |
| degradation             | different degradation levels are used?                                  |
| False positives (coral  | Do other components in the water light up when using the forensic light |
| stone, white plastic,   | source? If yes, which?  |
| shells, fish bone)      |   |

Table 3.1: Variables and their connected sub questions

Wavelength of the forensic light source is an important variable because this will determine whether the bone excites to a higher state and therefore if it will fluoresce or not. The filters contribute to make fluorescence more visible when fluorescence is not visible enough to the naked eye. Different levels of degradation are important to discover the degradation of fluorescence when bone degrades. The distance between the light source and the bone is of great importance because the amount of light that reaches the bone and therefore the degree of fluorescence decreases at a bigger distance. The knowledge of the existence of false positives is meaningful because this creates awareness that not all of the fluorescent objects have to be bones.

#### 3.2.1 Stage 1: Experiments in a swimming pool with tap water

The experiments of stage one are conducted in a 259x162x62 (LxWxH) cm Bestway Steel Pro swimming pool. The swimming pool was filled with approximately 1900 litres of tap water and covered with sheeting to create a dark setting similar to nature water. Experiment 1A was conducted at depths of 50, 75, 100, 125 and 150 cm depth. These depths are chosen in order to explore if depths with differences of 25 cm show difference in fluorescence of the bones. The bones used in the experiments are the ribs of a pig. Bones of a pig are used because of the close similarity in composition with human bones.

The bones used in the experiments are obtained at the local supermarket. The false positives used were obtained in a local fishing store and were chosen because these materials often occur in natural water. The forensic light source, filters, white board and magnets were already owned by Loci Forensics B.V.. The camera and swimming pool were obtained at web shops such as Bol.com and Amazon.

In order to capture the results of stage 1, the AKASO V50 Elite 8x Slow Motion Video Action Camera was used. This camera was placed on a tripod in the water at a distance of approximately 20 cm from the bones. The results were captures by taking a picture in .JPEG format at every difference in variables. The light source used at stage 1 was not waterproof which meant a setup had to be created to use this light source. For this setup PVC pipes are used and put together in a T-shape to make a setup that also allows the light source to vent and not become overheated. In order to enable the light to reach the bones, a 'window' is made in the PVC pipes with glass glued to the pipe.

Experiment 1A is executed in order to find out if the bones will light up under different conditions. Experiment 1B is executed in order to find out if other components that are in the water will fluoresce. All of the bones are marked with different levels of degradation.

#### Experiment 1A:

Materials:

- Bones 3x (with degradation levels of 0, 1, 2, 3 and 4 weeks);
- Forensic light source with wavelengths of 365, 390, 405 and 455 nm (Lumatec Superlite M05);
- Swimming pool;
- Tap water;
- Filters (orange, red and yellow);
- Sheeting (400x300 cm);
- AKASO V50 Elite 8x Slow Motion Video Action Camera;
- Tripod;
- Whiteboard;
- Magnets;

#### Execution:

- 1. Fill the swimming pool with tap water
- 2. Measure 50, 75, 100, 125 and 150 cm horizontal in the pool and mark these lengths
- 3. Place three bones of each level of degradation on a whiteboard using magnets
- 4. Place the camera on the tripod and place it in the water at ~ 20 cm from the bones (figure 3.1)

- 5. Cover the pool with sheeting
- Illuminate the bones with the forensic light source with a wavelength of 365 nm at a distance
  50 cm and photograph the result
- 7. Repeat step 5 with the orange filter placed in front of the camera, followed by the yellow and red filter
- 8. Repeat step 5 and 6 with wavelength 390, 405 and 455 nm
- 9. Repeat step 3 to 7 with 75, 100, 125 and 150 horizontal distance between light source and bone



Figure 3.1: Setup of the camera in front of the bones

#### Experiment 1B:

#### Materials:

- Forensic light source with wavelengths of 365, 390, 405 and 455 nm (Lumatec Superlite M05);
- Swimming pool;
- Tap water;
- Filters (orange, red, yellow);
- Sheeting (400x300 cm);
- AKASO V50 Elite 8x Slow Motion Video Action Camera;
- False positives.

#### Execution:

- 1. Fill the pool with tap water and the false positives
- 2. Cover the pool with sheeting
- 3. Illuminate the false positives with the forensic light source with a wavelength of 365 nm at 50 cm and photograph the result
- 4. Repeat step 3 with the orange filter placed in front of the camera, followed by the yellow and red filter
- 5. Repeat step 3 and 4 with wavelength 390, 405 and 455 nm

#### 3.2.2 Stage 2: Experiment in natural water

The experiment of stage two is conducted in the Vinkeveense Plassen at Utrecht. The experiment was comparable to experiment 1A in order to find out if the results are valid. However, in this experiment no filters are used because of the difficulty in use. In this experiment, a different light source is used because the Höfftech light source is waterproof. This light source combines light between wavelengths of 365 to 395 nm.

The experiment in stage 2 is conducted at depths of 2, 4, 6, 8 and 10 m. These depths are chosen in order to explore the incidence of light and if the light source works under this incidences of light. These depths create an situations close to reality as well. The bones used in this experiment are the ribs of a pig. Bones of a pig are used because of the close similarity in composition with human bones. The bones used in the experiments are obtained at the local supermarket. The forensic light source was obtained at a local web shop and the depth meter was owned by the Arrest Team of the Amsterdam Police Department.

At experiment 2, two series of diving from 2 to 10 meter were done. The diver was attached to a string which made communication between the diver and the person on the quay. Both series consisted of the diver swimming from the quay to 2 metres and put the bones on the bottom of the lake and got them on camera. Then the diver picked up the bones and swam to 4 metres to do the same. The same was done with depths of 6, 8 and 10 metres.

In order to capture the results of stage 2, the AKASO V50 Elite 8x Slow Motion Video Action Camera was used. This camera was attached to a handgrip and taken under the water. The camera filmed during the complete trip under the water from 2 to 10 metres in a .MOV file. The results were obtained by pausing the video and capturing pictures out of the .MOV file.

#### Experiment 2:

#### Materials:

- Bones 3x (with degradation levels of 0 and 1 week);
- Forensic light source (Höfftech 365/395 nm);
- Natural water such as a lake;
- AKASO V50 Elite 8x Slow Motion Video Action Camera;
- Depth meter.

#### Execution:

- 1. Dive in the water and go to a depth of 2 metres using a depth meter
- 2. Place three bones of each level of degradation in the water
- 3. Illuminate the bones with the forensic light source with a wavelength of 365-395 nm and photograph the result

4. Repeat step 2 and 3 at a depth of 4, 6, 8 and 10 metres

#### 3.3 Hypotheses

This research will have 3 hypotheses because of the 3 experiments that are conducted.

Hypothesis 1A: The bones will light up best when illuminated with a wavelength of 390 nm without a filter. The distance between the bones and the forensic light source that will make the bone illuminate best is 50 cm. Bones with a degradation level of 0 weeks will fluoresce best.

Hypothesis 1B: Only the fish bone cause fluorescence at a wavelength of 390 nm without a filter.

Hypothesis 2: The bones will light up best when illuminated with a wavelength of 365-395 nm without a filter. Bones with a level of degradation of 0 weeks will light up best at a depth of 10 meters.

## 4. Methods

#### 4.1 Desk research

Desk research is used to gain known information. In this section of literature research, books, articles, websites and other references will be used. In order to find the used literature, the internet is used. The Saxion Library and scientific sites are used to find the information gained. The main goal of using desk research is to get background information about the subject and to help to answer the subquestions that are cited in chapter 1.3.

#### 4.2 Experiments

Experiments are conducted in order to gain unknown information. The experiments will provide information about the wavelength that is best to emit bones, which depth is optimal, if there are false positives etcetera. The main goal of using experiments is to gain information that will help answer the sub-questions and eventually the main question. The experiments that are conducted, as well as the materials used, are stated in chapter 3.2.

#### 4.3 Data analysis

Data analysis is done to understand the information that is gained by conducting the experiment that are cited in chapter 3. Data analysis is done by observing the results and determining if fluorescence was present. The main goal of the data analysis was to transform the gained information into answers on the main question and thus into a method that is optimal to find bones under water with the use of a forensic light source.

# 5. Results

The purpose of this research was to develop a method that makes bone(parts) more visible under water when using a forensic light source. This is done in order to make the bones better detectable for divers which can create more quantity and deficiency in the detection of the bones. These experiments are performed in order to figure out if bone fluoresces and in which conditions as well as to figure out if the chosen possible false positives fluoresce. Fluorescence of the bone is visible when the bone gives light. <u>Figure 5.1</u> gives an example of a bone (in the middle) fluorescing. In this chapter the results of the conducted experiments are presented.



Figure 5.1: Example of a bone fluorescing

5.1 Experiment 1A: Bones in a swimming pool

#### 5.1.1 Bones that have not been in water

At a depth of 50 cm, one bone that has not been in water fluoresces without a filter when illuminated by light with a wavelength of 390. When a yellow, orange or red filter is used, only the one bone illuminated by light with a wavelength of 390 fluoresce (<u>table 5.1</u>). The fact that only one bone fluoresces may be the result of the light not reaching the bones or too low light intensity. Bones illuminated a wavelength of 365, 405 or 455 nm do not fluoresce.

|        | No filter | Yellow filter | Orange filter | Red filter |
|--------|-----------|---------------|---------------|------------|
| 365 nm |           |               | 1.99          | 1          |
| 390 nm |           | T.J.J         |               |            |
| 405 nm |           | 3/1           |               |            |
| 455 nm | TA        | 775           | 3/1           | 111        |

Table 5.1: Images of bones that have not been in water illuminated by a FLS at 50 cm depth

Bones that have not been in water that are illuminated at a depth of 75 cm show exact the same results as bones that have not been in water at a depth of 50 cm (table 5.2).

|        | No filter | Yellow filter | Orange filter            | Red filter |
|--------|-----------|---------------|--------------------------|------------|
| 365 nm | T         | T             |                          |            |
| 390 nm |           | ŢŢŢ           | $\left( \right) \right)$ | 1.1        |
| 405 nm | 1/1       | 3/1           | 7/1                      | 111        |
| 455 nm |           |               | E / J                    |            |

Table 5.2: Images of bones that have not been in water illuminated by a FLS at 75 cm depth

Bones that have not been in water that are illuminated at a depth of 100 cm show exact the same results as bones that have not been in water at a depth of 50 and 75 cm (table 5.3).

|        | No filter | Yellow filter | Orange filter | Red filter                                   |
|--------|-----------|---------------|---------------|--|
| 365 nm | T         | T             |               | Bones not visible<br>due to<br>underexposure |
| 390 nm | 1.77      | 1.77          | 1.)           | 111  |
| 405 nm | 1/1       |               |               | 11   |
| 455 nm |           |               | E/A           | 1/1  |

Table 5.3: Images of bones that have not been in water illuminated by a FLS at 100 cm depth

Bones that have not been in water that are illuminated at a depth of 125 cm show exact the same results as bones that have not been in water at a depth of 50, 75 and 100 cm (table 5.4).

|        | No filter | Yellow filter | Orange filter | Red filter                                   |
|--------|-----------|---------------|---------------|--|
| 365 nm |           |               |               | Bones not visible<br>due to<br>underexposure |
| 390 nm |           | 1.77          | []]           | 1.1  |
| 405 nm | 7/1       | 3/1           | 1/1           | 111  |
| 455 nm |           | 1             |               | <b>X</b> ///                                 |

Table 5.4: Images of bones that have not been in water illuminated by a FLS at 125 cm depth

At a depth of 150 cm one bone that has not been in water fluoresces without a filter when illuminated by light with a wavelength of 390. When a yellow, orange or red filter is used when illuminated by light with a wavelength of 390 nm, only one bone fluoresces as well. Bones illuminated a wavelength of 365, 405 or 455 nm do not fluoresce (table 5.5).

|        | No filter | Yellow filter | Orange filter | Red filter                                   |
|--------|-----------|---------------|---------------|--|
| 365 nm | 477       |               |               | Bones not visible<br>due to<br>underexposure |
| 390 nm |           |               | 1/            | $(\mathcal{I})$                              |
| 405 nm | 7/1       | 1/1           | 1/1           | Bones not visible<br>due to<br>underexposure |
| 455 nm |           |               | X/M           | Bones not visible<br>due to<br>underexposure |

Table 5.5: Images of bones that have not been in water illuminated by a FLS at 150 cm depth

#### 5.1.2 One week old bones

At a depth of 50 cm bones that have been in water for one week fluoresce without a filter when illuminated by light with a wavelength of 390. When a yellow, orange or red filter is used when illuminated by light with a wavelength of 390 nm, only one bone fluoresces. The fact that only one

bone fluoresces may be the result of the light not reaching the bones or too low light intensity. Bones illuminated a wavelength of 365, 405 or 455 nm do not fluoresce (table 5.6).

|        | No filter | Yellow filter | Orange filter | Red filter                                   |
|--------|-----------|---------------|---------------|--|
| 365 nm | ANE       | TH            |               | Bones not visible<br>due to<br>underexposure |
| 390 nm | 1-27      | 1.77          | 1.77          | •/ /   |
| 405 nm | 1         | 11            |               | Bones not visible<br>due to<br>underexposure |
| 455 nm | N         | T             | 1.1.1         | 1 II   |

Table 5.6: Images of bones that have in water for one week illuminated by a FLS at 50 cm depth

At a depth of 75 cm bones that have been in water for one week fluoresce without a filter, with a yellow filter and an orange filter when illuminated by light with a wavelength of 390. When a red filter is used when illuminated by light with a wavelength of 390 nm, only one bones fluoresces. The fact that only one of the bones fluoresces may be the result of the light not reaching the bones or too low light intensity. Bones illuminated a wavelength of 365, 405 or 455 nm do not fluoresce (table 5.7).

|        | No filter | Yellow filter | Orange filter | Red filter                                   |
|--------|-----------|---------------|---------------|--|
| 365 nm | TIE       |               |               | Bones not visible<br>due to<br>underexposure |
| 390 nm | 111       | 1.77          | 1.77          | · <b>j</b> )                                 |
| 405 nm |           | 11            |               | Bones not visible<br>due to<br>underexposure |
| 455 nm | <b></b>   | TR            | T             |  |

Table 5.7: Images of bones that have in water for one week illuminated by a FLS at 75 cm depth

At a depth of 100 cm bones that have been in water for one week fluoresce without a filter when illuminated by light with a wavelength of 390. When a yellow or orange filter is used when illuminated by light with a wavelength of 390 nm the bones show a light fluorescence. When a red filter is used only one of the bones fluoresces (<u>table 5.8</u>). Bones illuminated a wavelength of 365, 405 or 455 nm do not fluoresce.

|        | No filter | Yellow filter | Orange filter                                | Red filter                                   |
|--------|-----------|---------------|--|--|
| 365 nm | 711       |               | Bones not visible<br>due to<br>underexposure | Bones not visible<br>due to<br>underexposure |
| 390 nm | 1.11      | 1.11          | <i>())</i>                                   | $\langle I \rangle$                          |
| 405 nm |           |               |  | Bones not visible<br>due to<br>underexposure |
| 455 nm |           | T             | T  | Bones not visible<br>due to<br>underexposure |

Table 5.8: Images of bones that have in water for one week illuminated by a FLS at 100 cm depth

At a depth of 125 cm bones that have been in water for one week fluoresce without a filter when illuminated by light with a wavelength of 390. When a yellow filter is used the bones show a light fluorescence. When an orange or red filter is used when illuminated by light with a wavelength of 390 nm, only one bone fluoresces. Bones illuminated a wavelength of 365, 405 or 455 nm do not fluoresce (table 5.9).

|        | No filter | Yellow filter | Orange filter                                | Red filter                                   |
|--------|-----------|---------------|--|--|
| 365 nm |           |               | Bones not visible<br>due to<br>underexposure | Bones not visible<br>due to<br>underexposure |
| 390 nm | 111       | 1.11          | 1.77   |  |
| 405 nm | 11        |               | Bones not visible<br>due to<br>underexposure | Bones not visible<br>due to<br>underexposure |
| 455 nm | 0.00      | T             |  | Bones not visible<br>due to<br>underexposure |

Table 5.9: Images of bones that have in water for one week illuminated by a FLS at 125 cm depth

At a depth of 150 cm one of the bones that has been in water for one week fluoresces without a filter, with a yellow filter and with an orange filter when illuminated by light with a wavelength of 390. However, when a yellow or orange filter is used, only one of the bones fluoresces. When a red filter is used, none of the three bones fluoresces. Bones illuminated a wavelength of 365, 405 or 455 nm do not fluoresce (table 5.10).

|        | No filter | Yellow filter | Orange filter                                | Red filter                                   |
|--------|-----------|---------------|--|--|
| 365 nm |           |               | Bones not visible<br>due to<br>underexposure | Bones not visible<br>due to<br>underexposure |
| 390 nm |           | 577           | 11   | Bones not visible<br>due to<br>underexposure |
| 405 nm | 71        | 1.13          | Bones not visible<br>due to<br>underexposure | Bones not visible<br>due to<br>underexposure |
| 455 nm | 0.00      | TI            |  | Bones not visible<br>due to<br>underexposure |

Table 5.10: Images of bones that have in water for one week illuminated by a FLS at 150 cm depth

#### 5.1.3 Two week old bones

At a depth of 50 cm, bones that have been in water for two weeks fluoresce without a filter when illuminated by light with a wavelength of 390. When a yellow, orange or red filter is used, fluorescence

is barely visible to non-existent (<u>table 5.11</u>). Bones illuminated a wavelength of 365, 405 or 455 nm do not fluoresce.

|        | No filter   | Yellow filter | Orange filter | Red filter                                   |
|--------|-------------|---------------|---------------|--|
| 365 nm | ••••<br>1#1 | 100           | 7-1           | Bones not visible<br>due to<br>underexposure |
| 390 nm | (b)         |               |               |  |
| 405 nm | 101         | 100           | 191           | 101  |
| 455 nm |             | 101           | 101           | 1.21   |

Table 5.11: Images of bones that have in water for two weeks illuminated by a FLS at 50 cm depth

At a depth of 75 cm, bones that have been in water for two weeks fluoresce without a filter when illuminated by light with a wavelength of 390. When a yellow, orange or red filter is used, fluorescence is barely visible to non-existent. Bones illuminated a wavelength of 365, 405 or 455 nm do not fluoresce (table 5.12).

|        | No filter | Yellow filter | Orange filter | Red filter                                   |
|--------|-----------|---------------|---------------|--|
| 365 nm | 101       | 101           | 701           | Bones not visible<br>due to<br>underexposure |
| 390 nm |           | **            |               |  |
| 405 nm | 121       | 100           | 1.01          | Bones not visible<br>due to<br>underexposure |
| 455 nm | 000       | 101           | 1-1           | Bones not visible<br>due to<br>underexposure |

Table 5.12: Images of bones that have in water for two weeks illuminated by a FLS at 75 cm depth

At a depth of 100 cm, bones that have been in water for two weeks fluoresce without a filter when illuminated by light with a wavelength of 390. When a yellow, orange or red filter is used, fluorescence

is barely visible to non-existent. When illuminated by light with wavelengths of 365, 405 or 455 nm no fluorescence was observed (table 5.13).

|        | No filter                                   | Yellow filter | Orange filter                                | Red filter                                   |
|--------|---|---------------|--|--|
| 365 nm |   | 121           | Bones not visible<br>due to<br>underexposure | Bones not visible<br>due to<br>underexposure |
| 390 nm |   | **            |  |  |
| 405 nm | Bones not visible<br>due to<br>overexposure | 121           | 101  | Bones not visible<br>due to<br>underexposure |
| 455 nm | Bones not visible<br>due to<br>overexposure | 101           |  | Bones not visible<br>due to<br>underexposure |

Table 5.13: Images of bones that have in water for two weeks illuminated by a FLS at 100 cm depth

At a depth of 125 cm, bones that have been in water for two weeks fluoresce without a filter when illuminated by light with a wavelength of 390. When a yellow, orange or red filter is used, fluorescence is barely visible to non-existent. When illuminated by light with wavelengths of 365, 405 or 455 nm no fluorescence was observed either with or without a filter (<u>table 5.14</u>).

|        | No filter                                   | Yellow filter                                | Orange filter                                | Red filter                                   |
|--------|---|--|--|--|
| 365 nm |   | Bones not visible<br>due to<br>underexposure | Bones not visible<br>due to<br>underexposure | Bones not visible<br>due to<br>underexposure |
| 390 nm |   |  | - F - )                                      |  |
| 405 nm | Bones not visible<br>due to<br>overexposure | 100  |  | Bones not visible<br>due to<br>underexposure |
| 455 nm | Bones not visible<br>due to<br>overexposure | 101  | Bones not visible<br>due to<br>underexposure | Bones not visible<br>due to<br>underexposure |

Table 5.14: Images of bones that have in water for two weeks illuminated by a FLS at 125 cm depth

At a depth of 150 cm, bones that have been in water for two weeks fluoresce without a filter when illuminated by light with a wavelength of 390. When a yellow, orange or red filter is used, fluorescence

is barely visible to non-existent. When illuminated by light with wavelengths of 365, 405 or 455 nm no fluorescence was observed (table 5.15).

|        | No filter                                   | Yellow filter                                | Orange filter                                | Red filter                                   |
|--------|---|--|--|--|
| 365 nm |   | Bones not visible<br>due to<br>underexposure | Bones not visible<br>due to<br>underexposure | Bones not visible<br>due to<br>underexposure |
| 390 nm |   |  |  |  |
| 405 nm | •••<br>/ ~ /                                | <b>•••</b> •                                 | Bones not visible<br>due to<br>underexposure | Bones not visible<br>due to<br>underexposure |
| 455 nm | Bones not visible<br>due to<br>overexposure | 101  | Bones not visible<br>due to<br>underexposure | Bones not visible<br>due to<br>underexposure |

Table 5.15: Images of bones that have in water for two weeks illuminated by a FLS at 150 cm depth

#### 5.1.4 Three week old bones

At a depth of 50 cm, two of the bones that have been in water for three weeks fluoresce without a filter when illuminated by light with a wavelength of 390. The reason why only two bones fluoresce is that the other bone is affected or not illuminated enough to fluoresce. When a yellow, orange or red filter is used, fluorescence is barely visible to non-existent. When illuminated by light with wavelengths of 365, 405 or 455 nm no fluorescence was observed (table 5.16).

|        | No filter | Yellow filter | Orange filter | Red filter                                   |
|--------|-----------|---------------|---------------|--|
| 365 nm |           | 1 6 4         | 164           | Bones not visible<br>due to<br>underexposure |
| 390 nm |           | A A A         | AA            | A a  |
| 405 nm |           |               |               | 2 .  |
| 455 nm |           | 124           |               |  |

Table 5.16: Images of bones that have in water for three weeks illuminated by a FLS at 50 cm depth

At a depth of 75 cm, the bones that have been in water for three weeks fluoresce without a filter when illuminated by light with a wavelength of 390. When a yellow, orange or red filter is used when illuminated by light with a wavelength of 390 nm, fluorescence is barely visible tot non-existent. When illuminated by light with wavelengths of 365, 405 or 455 nm no fluorescence was observed. When illuminated by light with wavelengths of 365, 405 or 455 nm no fluorescence was observed (table 5.17).

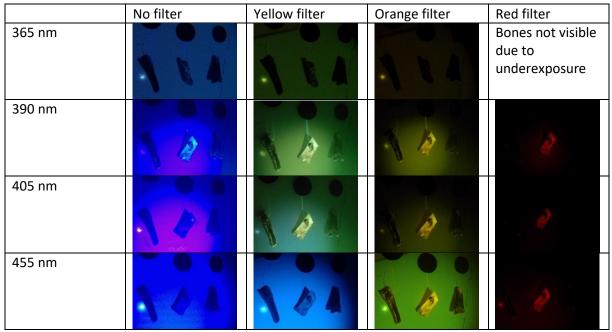


Table 5.17: Images of bones that have in water for three weeks illuminated by a FLS at 75 cm depth

At a depth of 100 cm, the bones that have been in water for three weeks fluoresce without a filter when illuminated by light with a wavelength of 390. When a yellow, orange or red filter is used when illuminated by light with a wavelength of 390 nm, fluorescence is barely visible to non-existent. When illuminated by light with wavelengths of 365, 405 or 455 nm no fluorescence was observed (table 5.18).

|        | No filter | Yellow filter | Orange filter | Red filter                                   |
|--------|-----------|---------------|---------------|--|
| 365 nm | 1 6 4     |               |               | Bones not visible<br>due to<br>underexposure |
| 390 nm | 1 2 1     |               |               | e et   |
| 405 nm | 104       |               |               | e <sup>\$</sup>                              |
| 455 nm | 104       | 104           | 144           | ð  |

Table 5.18: Images of bones that have in water for three weeks illuminated by a FLS at 100 cm depth

At a depth of 125 cm, two of the bones that have been in water for three weeks fluoresce without a filter when illuminated by light with a wavelength of 390. The reason why only two bones fluoresce is that the other bone is affected or not illuminated enough to fluoresce. When a yellow, orange or red filter is used when illuminated by light with a wavelength of 390 nm, fluorescence is barely visible to non-existent. When illuminated by light with wavelengths of 365, 405 or 455 nm no fluorescence was observed (table 5.19).

|        | No filter | Yellow filter | Orange filter                                | Red filter                                   |
|--------|-----------|---------------|--|--|
| 365 nm |           |               | Bones not visible<br>due to<br>underexposure | Bones not visible<br>due to<br>underexposure |
| 390 nm | 1 de la   |               | 10 10  | ¢  |
| 405 nm | 104       |               |  | ليه  |
| 455 nm |           | 104           | R AN   | e a  |

Table 5.19: Images of bones that have in water for three weeks illuminated by a FLS at 125 cm depth

At a depth of 150 cm, two of the bones that have been in water for three weeks fluoresce without a filter when illuminated by light with a wavelength of 390. The reason why only two bones fluoresce is that the other bone is affected or not illuminated enough to fluoresce. When a yellow, orange or red filter is used when illuminated by light with a wavelength of 390 nm, fluorescence is barely visible tot non-existent. When illuminated by light with wavelengths of 365, 405 or 455 nm no fluorescence was observed (table 5.20).

|        | No filter | Yellow filter | Orange filter                                | Red filter                                   |
|--------|-----------|---------------|--|--|
| 365 nm |           |               | Bones not visible<br>due to<br>underexposure | Bones not visible<br>due to<br>underexposure |
| 390 nm | 100       |               | 140  | e)   |
| 405 nm | 100       | N A A         | 45   | e.   |
| 455 nm |           | TOA           |  | Bones not visible<br>due to<br>underexposure |

Table 5.20: Images of bones that have in water for three weeks illuminated by a FLS at 150 cm depth

#### 5.1.5 Four week old bones

At a depth of 50 cm, two of the bones that have been in water for four weeks fluoresce without a filter when illuminated by light with a wavelength of 390. The reason why only two bones fluoresce is that the other bone is not illuminated enough to fluoresce. When a yellow or orange filter is used when illuminated by light with a wavelength of 390 nm, fluorescence is barely visible to non-existent. When the red filter is used the bones do not fluoresce. When illuminated by light with wavelengths of 365, 405 or 455 nm no fluorescence was observed (table 5.21).

|        | No filter                                   | Yellow filter | Orange filter | Red filter                                   |
|--------|---|---------------|---------------|--|
| 365 nm | 3.71  | 7.91          | 2-71          | Bones not visible<br>due to<br>underexposure |
| 390 nm | 4 10 1                                      |               | A COP         | a stra                                       |
| 405 nm | Bones not visible<br>due to<br>overexposure |               | 8-1           | e- 1-  |
| 455 nm | Bones not visible<br>due to<br>overexposure | 7-11          | 7-70          | ** /   |

Table 5.21: Images of bones that have in water for four weeks illuminated by a FLS at 50 cm depth

At a depth of 75 cm, the bones that have been in water for four weeks fluoresce without a filter when illuminated by light with a wavelength of 390. When a yellow or orange filter is used when illuminated

by light with a wavelength of 390 nm, fluorescence is barely visible to non-existent. When the red filter is used the bones do not fluoresce. When illuminated by light with wavelengths of 365, 405 or 455 nm no fluorescence was observed (table 5.22).

|        | No filter                                   | Yellow filter | Orange filter | Red filter                                   |
|--------|---|---------------|---------------|--|
| 365 nm | 3.98  | 3-31          | 1             | Bones not visible<br>due to<br>underexposure |
| 390 nm | 1000  |               |               | 6 N. T                                       |
| 405 nm | 2-7X  | 1999          | e-1           | ***  |
| 455 nm | Bones not visible<br>due to<br>overexposure | 7-11          | 7-71          | Bones not visible<br>due to<br>underexposure |

Table 5.22: Images of bones that have in water for four weeks illuminated by a FLS at 75 cm depth

At a depth of 100 cm, only one of the bones that have been in water for four weeks fluoresce without a filter when illuminated by light with a wavelength of 390. The reason why only one of the bones fluoresces is that the other bone is not illuminated enough to fluoresce or the bones are affected. When a yellow, orange or red filter is used when illuminated by light with a wavelength of 390 nm, fluorescence is barely visible to non-existent. Illumination by light with a wavelength of 365, 405 or 455 nm shows no fluorescence (table 5.23).

|        | No filter                                   | Yellow filter | Orange filter                                | Red filter                                   |
|--------|---|---------------|--|--|
| 365 nm | 371   |               | Bones not visible<br>due to<br>underexposure | Bones not visible<br>due to<br>underexposure |
| 390 nm |   |               |  | 8 A - 1                                      |
| 405 nm | Bones not visible<br>due to<br>overexposure |               |  | Bones not visible<br>due to<br>underexposure |
| 455 nm | Bones not visible<br>due to<br>overexposure | 771           | 771  | Bones not visible<br>due to<br>underexposure |

Table 5.23: Images of bones that have in water for four weeks illuminated by a FLS at 100 cm depth

At a depth of 125 cm, one of the bones that have been in water for four weeks fluoresces without a filter when illuminated by light with a wavelength of 390. When a yellow, orange or red filter is used when illuminated by light with a wavelength of 390 nm, fluorescence is barely visible to non-existent. Illumination by light with a wavelength of 365, 405 or 455 nm shows no fluorescence (<u>table 5.24</u>).

|        | No filter                                   | Yellow filter | Orange filter                                | Red filter                                   |
|--------|---|---------------|--|--|
| 365 nm |   |               | Bones not visible<br>due to<br>underexposure | Bones not visible<br>due to<br>underexposure |
| 390 nm |   |               | PF   | 8 × 3  |
| 405 nm | Bones not visible<br>due to<br>overexposure |               | e / 1  | Bones not visible<br>due to<br>underexposure |
| 455 nm | Bones not visible<br>due to<br>overexposure | 771           |  | Bones not visible<br>due to<br>underexposure |

Table 5.24: Images of bones that have in water for four weeks illuminated by a FLS at 125 cm depth

At a depth of 150 cm, the bones that have been in water for four weeks fluoresce without a filter when illuminated by light with a wavelength of 390. When a yellow, orange or red filter is used when illuminated by light with a wavelength of 390 nm, fluorescence is barely visible to non-existent. Illumination by light with a wavelength of 365, 405 or 455 nm shows no fluorescence (<u>table 5.25</u>).

|        | No filter                                   | Yellow filter                                | Orange filter                                | Red filter                                   |
|--------|---|--|--|--|
| 365 nm |   | Bones not visible<br>due to<br>underexposure | Bones not visible<br>due to<br>underexposure | Bones not visible<br>due to<br>underexposure |
| 390 nm |   |  |  | *  |
| 405 nm | Bones not visible<br>due to<br>overexposure | 7-7-1  |  | Bones not visible<br>due to<br>underexposure |
| 455 nm | 171   | 7-11   | Bones not visible<br>due to<br>underexposure | Bones not visible<br>due to<br>underexposure |

Table 5.25: Images of bones that have in water for four weeks illuminated by a FLS at 150 cm depth

#### 5.2 Experiment 1B: Bones in natural water

This experiment was conducted at the Vinkeveense Plassen in Utrecht. The FLS used in this experiment has light with wavelengths between 365 and 395 nm. The reason that only light with this wavelengths is used is that only this FLS was waterproof. In order to execute this experiment, a diver from the Arrest Team of the Amsterdam Police Department dived to depths of 2, 4, 6, 8 and 10 meters and deposited bones of 0 and 1 week(s) at these depths.

Due to the incidence of light in the Vinkeveense Plassen, bones at a depth of 2, 4 and 6 meters were visible with the human eye without using a light source. Bones that have not been in water and bones that have been in water show the same results at a depth of both 8 and 10 meters (figure 5.2 till 5.5). Using a FLS with a wavelength of 365-395 nm gives no fluorescence when bones that have not been in water and bones that have been in water for one week are illuminated.

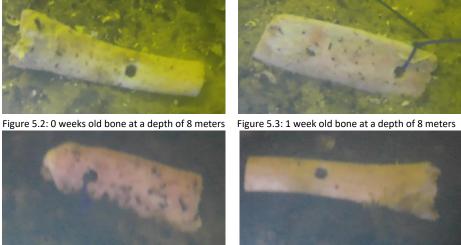


Figure 5.4: 0 weeks old bone at a depth of 10 meters Figure 5.5: 1 week old bone at a depth of 10 meters

#### 5.3 Experiment 2: False positives

For this experiment fishbone, shells, white coral, pebble stone and white plastic are used as possible false positives. <u>Table 5.26</u> shows the possible false positives when illuminated with different wavelengths and different uses of filters. When illuminated by light with a wavelength of 365, 390, 405 or 455 nm none of the possible false positives fluoresce. The tie rib which fluoresces at 365 nm when no filter was used, was not one of the false positives.

|        | No filter | Yellow filter | Orange filter  | Red filter  |
|--------|-----------|---------------|--|---|
| 365 nm | D         | 9             | Valse positives not<br>visible due to<br>underexposure | Valse positives<br>not visible due<br>to<br>underexposure |
| 390 nm |           | C             | Valse positives not<br>visible due to<br>underexposure | Valse positives<br>not visible due<br>to<br>underexposure |
| 405 nm |           |               |  | Valse positives<br>not visible due<br>to<br>underexposure |
| 455 nm |           |               |  | Valse positives<br>not visible due<br>to<br>underexposure |

Table 5.26: Images of possible false positives illuminated by a FLS at 50 cm depth

## 6. Discussion

The aim of this research was to develop a method that makes bone(parts) more visible under water when using a forensic light source. The method was not completely developed because it did not work on all of the bones and therefore the aim is achieved only partly. Three hypotheses were made before starting the research:

*Hypothesis 1A:* The bones will light up best when illuminated with a wavelength of 390 nm without a filter. The distance between the bones and the forensic light source that will make the bone illuminate best is 50 cm. Bones with a degradation level of 0 weeks will fluoresce best.

Hypothesis 1B: Only the fish bone cause fluorescence at a wavelength of 390 nm without a filter.

*Hypothesis 2:* The bones will light up best when illuminated with a wavelength of 365-395 nm without a filter. Bones with a level of degradation of 0 weeks will light up best at a depth of 10 meters.

The part about the wavelength is correct at hypothesis 1A. The other parts are inconclusive. Hypothesis 1B is not correct because fish bone does not fluoresce at a wavelength of 390 nm. Hypothesis 2 is not correct because bones do not fluoresce at a combined wavelength of 365-395 nm and therefore the other part of the hypothesis is inconclusive.

During this research a few problems stood in the way of conducting the research. At first, the plan was to use forensic light sources with wavelengths of 365, 365-390 390, 405 and 455 nm that were waterproof. In that way a situation would have been created that was most similar to when light sources are used to find bones under water. Because of COVID-19, some of those forensic light sources stated before were not delivered and therefore could not be used. Because of this, an adjustment had to be made and a different setup had to be created. Therefore, forensic light sources that are not waterproof are used. In order to use those non waterproof light sources, PVC pipes are used and put together in a T-shape to make a setup that also allows the light source to vent and not become overheated. In order to enable the light to reach the bones, a 'window' is made in the PVC pipes with glass glued to the pipe. This window may have caused a different refraction of light compared to when the forensic light source was put directly into the water when illuminating the bones.

When conducting the research it has occurred that when illuminating the bones under water, the intensity of the forensic light source changed at a few points, causing the bones to not be illuminated at all points. This may be the cause of only one or two bones fluorescing at the pictures taken. Another cause for the light to not illuminate all of the bones at some times is that the research illuminated three bones at the same time. When the bones were illuminated with a distance with the light source of 50 for an example, some of the bones were not always illuminated because of the width of the light beam. This could have caused that no fluorescence was observed.

The images taken of the research are not too reliable as well. The images are taken with an under water camera. However, the movement of the water when taking the pictures caused pictures that were out of focus at some points. Over- and underexposure was something that often happened. Underexposure happened when the light source was too far from the bones so that the bones could

not be illuminated. Overexposure happened when the light source was too close to the bones and no picture could be taken because of that.

The same three bones should have been used to conduct experiment 1A completely. They should have been used to do the experiment every week to get a reliable view of how the bones degrade during time. However, because of the forensic light sources not available at the beginning, different bones had to be used for most of the time, which may have altered the results of the research. Every bone altered differently, some of them were completely altered by mud for an example after being under the water for a few weeks. This may have caused that fluorescence is absence, while it could have been present when not altered. Also, pig bones were used instead of full grown human bones. Pig bones are very similar in structure compared to human bones [14].

Fluorescence was not equally visible at every point. Which means that there is no guarantee that the fluorescence was present at every point the human eye implemented. The same goes for when the human eye did not see fluorescence of the bones, it could have been there. At some points it was difficult to distinguish fluorescence with the altered colour of the light through the different filters.

At last, the overall planning could have been made better and therefore the research could have been conducted more gradually. The planning of the research could have been handled better as well. Experiment 2 was conducted before conducting experiment 1A and 1B. If experiment 1A was conducted before experiment 2, a light source with a wavelength that makes bones fluoresce could have been used in order to find out if the light source also makes bones fluoresce in natural water.

# 7. Conclusion

The purpose of this research was to develop a method that makes bone(parts) more visible under water when using a forensic light source. In order to conclude if a forensic light source can make bone(parts) of a full grown human being visible in natural water in the Netherlands, the variables had to be examined and experimented with. The conclusion states the conclusions of the sub questions at first and at last the overall conclusion is given.

Light with wavelengths of 365, 390, 405 and 455 nm were used to examine if bone would fluoresce. When observing the results of when the bones were illuminated with light with wavelengths of 365, 405 or 455 nm, no fluorescence was observed. The only wavelength which makes the bone(parts) fluoresce in this research is 390 nm.

At second, the distance between the bone(parts) and the forensic light source was examined. In this research, no conclusion regarding distance between the forensic light source and the bones could be made with the distances examined because no significant decrease of fluorescence was observed.

Different filters were used to examine if filters can make fluorescence noticeable. When observing the results regarding the usage of yellow, orange or red filters a small difference is noticeable in comparison with when no filter is used. It is observed that the usage of the filters did not make the fluorescence of the bones as visible as when no filter was used. The same goes for when no fluorescence was observed with without a filter; no fluorescence was visible when a filter was used. In conclusion, using a forensic light source when illuminating bone(parts) with a wavelength of 365, 390, 405 or 455 nm is best when no yellow, orange or red filter is used but when observed with the use of the naked eye.

At second last, the difference in the degree of fluorescence is examined when different degradation levels of the bone(parts) are used. The bone(parts) have been in water for 0, 1, 2, 3 or 4 weeks before illuminated with the forensic light source. The bones that have been in water showed the same amount of fluorescence as bones that were not in water. This research shows no visible difference in fluorescence between the degradations.

Lastly, different false positives were examined. Pebble stone, white plastic, shells and fish bone were used. However, none of these materials fluoresced when illuminated with light of 365, 390, 405 or 455 nm.

A forensic light source is able to make bones parts of a pig visible in water when using light with a wavelength of 390 nm when no filters are used. The distance between the light source and the bones as well as the different degradations show no difference in fluorescence. Using a forensic light source can certainly make detection of bones under water easier.

# 8. Recommendations

In order to get a more reliable outcome a follow up research must be done. In this follow up research the variables tested in this research should be done again. In order to make the research more complete a wider span of wavelengths could be tested. This research only used wavelengths of 365, 390, 405 and 455 nm but higher wavelengths and wavelengths in between the tested wavelengths could be tested in order to find out if more wavelengths would work for bones.

This research only conducts degradation levels up to four weeks. When finding bones under water chances are that the bones are in the water for a longer time than that. It is use that bones with a degradation level up to years or even decades are tested. As stated, only bones of a pig are used during this research. In order to find out if human bones will fluoresce as well as pig bones, human bones should be tested as well. Human bones do not only degrade when a human being has deceased, but also when a person ages. In addition to testing bones with different degradation levels after death, bones of people with different ages should be tested. More than three bones should be tested so that a significant method with an acceptable success rate can be developed.

In this research, the bones were tested in natural water but with a forensic light source with wavelengths that did not make the bones fluoresce. In order to create a successful method, the bones should be tested in natural waters. Distinction in the type of water should be made. There could be a difference between salt and sweet water in the degradation of the bone. The same goes for water with different vegetation types, different animals living in the water, different soil in the water etcetera.

The forensic light source should be placed directly into the water when conducting a follow up research in order to create a setup that is closest to reality when divers are searching for bone(parts). More false positives should be tested because, as seen in this research, more materials fluoresce, just not the ones in the scope of this research. It would also be useful to use measurement machines such as a spectrometer for light in order to measure the light.

In order to create a more close to the reality outcome of the research, the set up should be changed. The camera could be placed on top of the light source to create a vision field more realistic to what a diver is able to see. When testing in natural water with actual divers, the camera should be placed on the head of the diver in order to see what the diver actually sees. With that, when diving in natural water, bones can placed beforehand without the diver knowing where to find the bones in order for him to actual search for the bones. Next to that, when testing in a swimming pool or similar situation, the bones can be placed in soil with different possible false positives.

At last, the outcome of the research should be put into statistics to test if the method that will be developed is reliable. When the outcome of the statistics gives an outcome that gives a high level or reliability, the method could be practised.

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