



# Filière Systèmes industriels

## **Orientation Power and Control**

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## Conception of a small reflectometer to measure the reflectance spectrum of the iris

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# Conception of a small reflectometer to measure the reflectance spectrum of the iris

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### Abstract

Optical research is a domain which has made great progress. For example, at the beginning of February 2008, an artificial implant was fixed on the retina of a blind patient in Geneva, Switzerland. With this microelectronical device, this patient will be able to recover elementary vision.

But the research is still in development. The eye is a very complicated organ which will give further information through new optical and biological studies.

The aim of this project is to know more about the iris and its properties especially with the iris reflectance caused by the melanin. This pigment concentration in the different layers of the iris gives the commonly called "color of the eye".

An optical system has been developped around a spectrometer to measure the reflectance spectrum of the iris. This compact system uses a combination of leds for the illumination and a USB4000 spectrometer working with a spectral measurement software running on an Apple laptop.

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Chapter

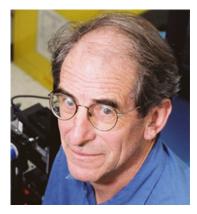
### Introduction

### 1.1 Location : SERI

This project has been done by Fabien Kuchler at the Schepens Eye Research Institute (SERI). This institute, situated at the West End of Boston, USA, is affiliated to the Harvard Medical School. It is a non-profit organization financed by public and private grants, foundations and endowments.



The supervisor of the work was Francois Delori, Senior Scientist at the SERI and Associate Professor of ophtalmology at the Harvard Medical School. He is mainly interested in light damage to the retina and the roles of macular pigment and of RPE lipofuscin in age-related macular degeneration (AMD).





### 1.2 Aim of the project

The aim of my work was the following :

- Construct a small reflectometer to measure the reflectance spectrum of the iris (to study melanin and light propagation in the iris).
- Measure reflectance spectra in a population of subjects and relate the results with skin pigmentation, eye color and fundus pigmentation.

In order to develop it, there were a couple of steps :

- Read literature about eye anatomy and biology.
- Investigate and compare different illumination systems.
- Select the optimal system and its optical components.
- Create and manufacture all the mechanical components.
- Mount and align all the components.
- Develop an electronic board for synchronization.
- Calibrate, test the system and start a wide study.

### 1.3 Possible contribution of the project

This system can be beneficial for biological research :

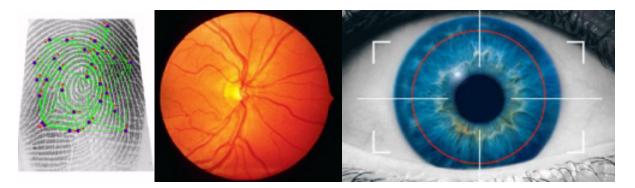
First, it will allow the study of the optical properties of the iris. The knowledge of the reflectance spectrum recorded with a spectral resolution better than 10 nm will allow to determine whether the concentration of melanin is the dominant modulator of the spectrum or whether other pigments (hemoglobin, lipofuscin) or other optical phenomena (scattering, interference) play a role. Of importance would be the change in the spectral shape between the blue and brown eyes and whether this change occurs gradually with increased concentration of melanin.

Second, the instrument will allow comparing iris pigmentation with skin pigmentation (measured with the same instrument but on the skin), and iris pigmentation with the pigmentation of the choroid (the pigmented blood layer behind the retina). This would be important for many epidemiology studies that often use "eye color" (iris color) to characterize the pigmentation of the eye. Results of such study could be used to develop a simple two-wavelength reflectance tests to classify eye color.

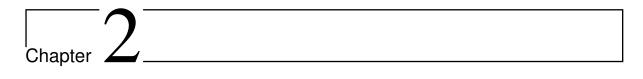
Third, through DNA samples obtained from the subjects, it should be possible to compare the iris reflectance spectra with the DNA, as well as other factors such as age, race, and sex.



Finally, the results of studies using the reflectometer may provide a new way of biometric identification (after fingerprints and retinal scans).







## Object of the study : the eye

### 2.1 Operating mode

The eye is the sight organ. It is composed of different parts :

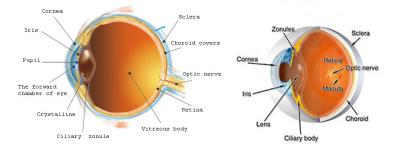


Figure 2.1: Anatomy of a human eye

- The cornea : it is the transparent window of the eye, providing also most of the refractive power.
- The iris : it plays the role of a diaphragm. It can vary the aperture to let more or less light pass through the pupil depending on the ambient lighting.
- The crystalline : it is the eye's lens. The image of an object is projected on the retina because of the cornea and the crystalline lens. The lens can change its shape in order to focus on the object.
- The retina : it is the photosensitive part in the eye. The projected image is then transmitted to the brain through the optic nerve.

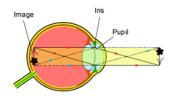


Figure 2.2: Image formation



### 2.2 The iris

As introduced in the previous section, the iris is an eye's component which can vary the pupil's aperture between 2 and 7 mm diameter. It is composed of four different layers :

- The anterior border layer : it consists of a dense packing of pigmented or nonpigmented cells similar in appearance to the cells present throughout the remainder of the stroma. Absence of cells produces the so called crypts in the border layer.
- The stroma : the iris stroma is a loose fibrocollagenous tissue composed of spindle shaped fibroblasts (stromal cells), blood vessels, nerves and macrophages (clump cells of Koganei) containing phagocytosed melanin pigment
- The dilator muscle: the dilator muscle layer is composed of the contractile processes of the myoepithelial cells of the inner layer of the posterior epithelium; it extends from the base of the iris to the sphincter muscle.
- The posterior pigmented epithelium : it is composed of two layers of cells which are densely pigmented with melanin.



Figure 2.3: Layers of the iris

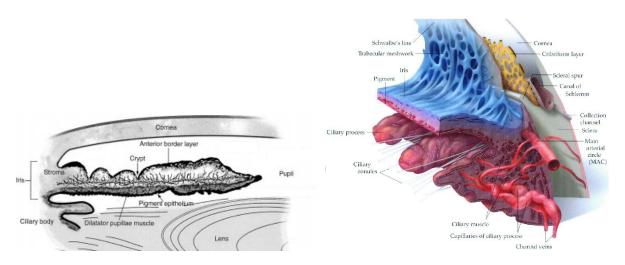


Figure 2.4: Structure of the iris



The color of the iris is in part determined by the concentration of melanin in the anterior layers and in the stroma. Blue eyes indicate that there is little melanin and dark brown eyes indicates lot of melanin ; grey and green are the intermediate colors.

The melanin in the iris has different functions :

- It decreseases the light diffusion and makes a better retinal image quality.
- It helps control the illumination of the retina (together with pupil constriction).

The iris is very interesting for identification. Each human iris is different, even with twins. It is easily accessible without physical contact and there is a wide variability of patterns.

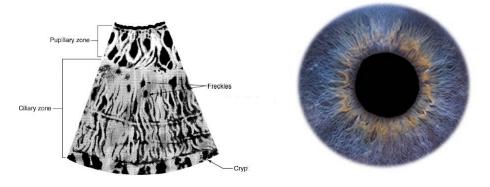


Figure 2.5: Iris's pattern

It is possible to see on the pictures that the structure of the iris is not totally homogeneous. It will have a consequence for the measurement system : a surface of the iris (ideally a ring) will be considered and integrated.



### 2.3 Melanin absorption

Different previous studies have shown that generally, the absorbance of melanin is more important in short visible wavelength and quite low in the near-infrared (NIR) wavelength.

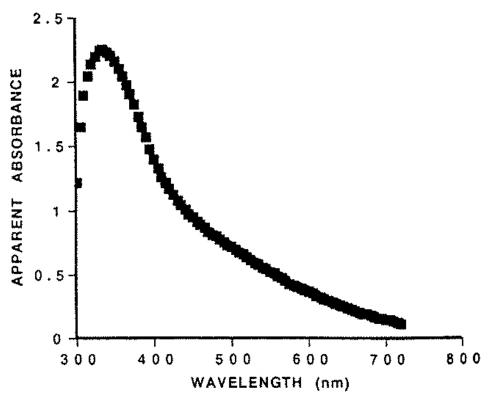


Figure 2.6: Melanin absorbance

Consequently, the illumination of the system should have more intensity in short wavelength than the rest in order to get enough signal in the spectrometer.



# Chapter 3

## System design

### 3.1 Design considerations

Basically, the reflectometer system must consist of a light source to illuminate the iris from 400 to 1000nm and a spectrometer to measure the reflected light at different wavelengths.

The illumination must not be unpleasant for the subject and it should respect the safety limits for eye exposure. Furthermore, the illumination should be rich is short wavelength where the reflectance is expected to be low (high melanin absorption).

Finally the system must contain an imaging system of the iris so that the reflectometer can be aligned to the iris.



Figure 3.1: System

To measure the reflectance of the iris one must only measure the reflectance in a ring-shaped area that corresponds with the pigmented part of the iris, and thus avoiding the white of the eye (sclera) and the central reflections within the pupil of the iris (see Chapter 4.3). Two systems were investigated to accomplish this:

- One in which the iris is illuminated only in a ring-shaped area and the reflected light is sampled over the entire iris, and
- another where the iris is uniformly illuminated but the reflected light is only detected from a

Both systems were investigated on an optical bench.



### 3.2 Ring-shaped illumination

To produce a ring-shaped illumination of the iris, I imaged an aperture on the iris as shown in the diagram. The aperture consisted in a central dark area surrounded by an open ring.

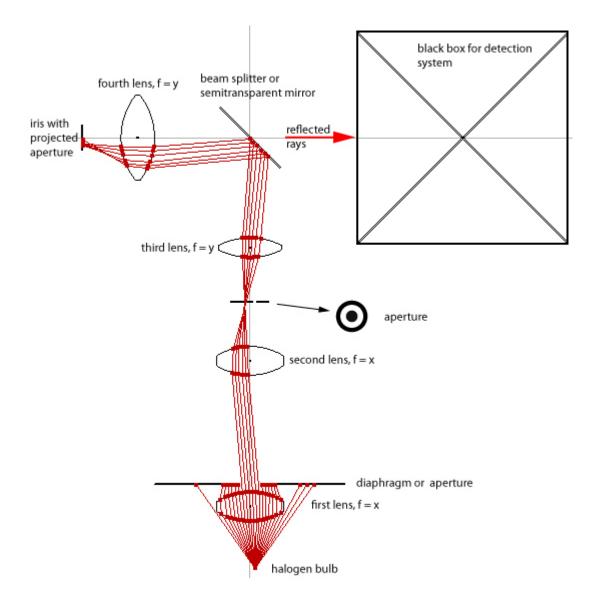


Figure 3.2: Halogen optical way

On this configuration, the rays of the transparent part of the aperture (light ring) are projected on the iris on a one to one (1:1) scale. Each pair of lenses has the same focal length.



The aperture is itself illuminated with a focused beam from a tungsten halogen lamp. It was a OSRAM HLX 64640 Xenophot bulb working at 24V with 150 W power. Its spectrum is the following :

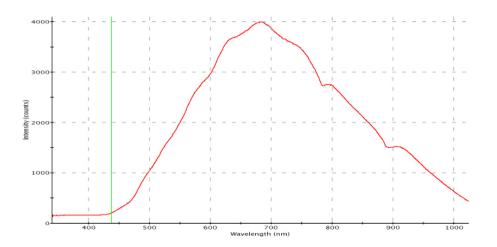


Figure 3.3: Halogen spectrum

The maximum of the spectrum is situated at 700nm and there is little intensity in short wavelengths (400-550nm).

As described in chapter 2.3, the iris absorbs a lot the light intensity in short wavelengths (300-500nm) however the illumination has a weak signal in those values. It is probable that the detected signal will be very low in short wavelengths which is a disadvantage.

On the optical table, the system of figure 3.2 has been constructed like this :

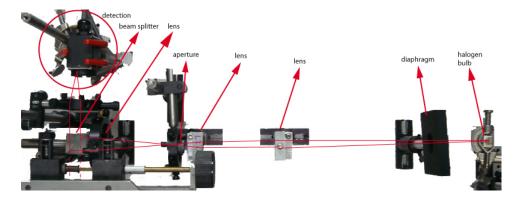


Figure 3.4: Halogen system on the optical table

The diaphragm is placed directly after the light source to avoid rays going everywhere in the room and especially on the detection camera.



Here is an example of the image we detect with the CCD camera :

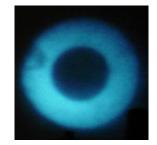


Figure 3.5: Aperture image on a sheet of paper, simulating the iris

The image of the aperture is projected and focused on a white paper sheet. The result is not perfect due to less than optimal optical components. Thus such approach is feasible with some optimization of the optical set-up. However, the physical size of the illumination system and the less than optimal spectrum of the illumination light are major disadvantages.

## 3.3 Ring-shaped detection : uniform illumination with LED's

The use of LED to illuminate the iris uniformly was further invastigated. The ringshaped aperture will therefore be placed in the detection system. The idea is to place the LED close to the eye (at about 50 to 70 mm).

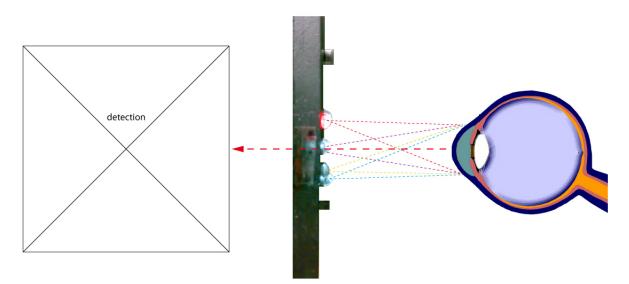
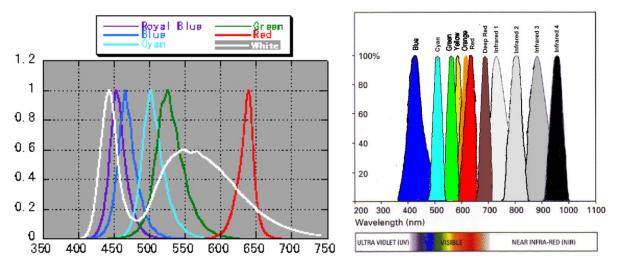


Figure 3.6: Uniform illumination with leds



Leds become more and more popular for different types of illumination. They don't consume a lot of energy and don't need a high voltage source. The main problem is that its spectrum is not continuous. As a consequence, if it is needed to have a wide spectrum, a combination of them should be used.



Here is an image of different leds' colors with their wavelength :

Figure 3.7: Spectra of leds

As the base of the illumination, a white warm led has been used. Its spectrum is the following :

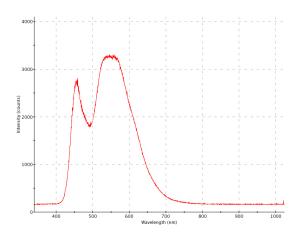


Figure 3.8: Spectrum of the white SS5W4UCCC led

Its intensity in short and mid wavelengths is better than the halogen bulb but a wider spectrum in long wavelengths (>600nm) is needed : one red led and three NIR (near infra-red) leds have been added.



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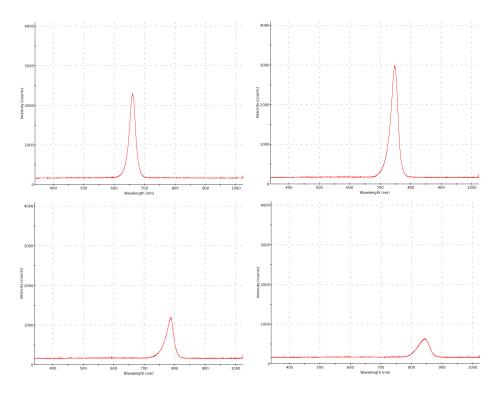


Figure 3.9: Spectra of the red 650nm, IR1 750nm, IR2 780nm and IR3 840nm leds

The combination of all those leds gives a "continuous" spectrum which can be used for the system :

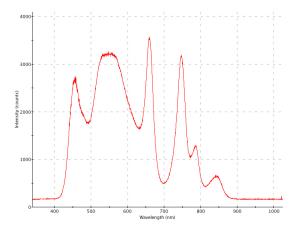
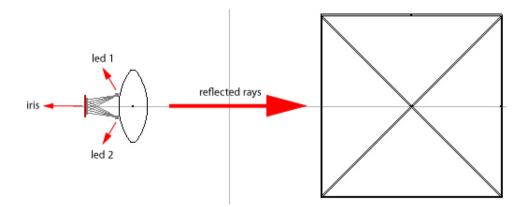


Figure 3.10: Reference spectrum of the illumination

The illumination used for the final tests was composed of eight leds. Four of them were white ones and the others centered at 650, 750, 780 and 840nm.

Of course it is not perfect but basically, the spectrum is wide and can be used for the detection. An advantage of using multiple LED's is that one can adjust the relative intensity of each LED, obtaining therefore a wide variety of spectra for the illumination.



The system would become immediately very compact which is a great advantage :

Figure 3.11: Direct illumination with the leds

All the leds are placed on a circle and slightly inclined to the center in order to have an homogeneous projection on the iris. The distance between the LED's and the iris was 50-70mm. Shorter distance would be unconfortable for the subject.

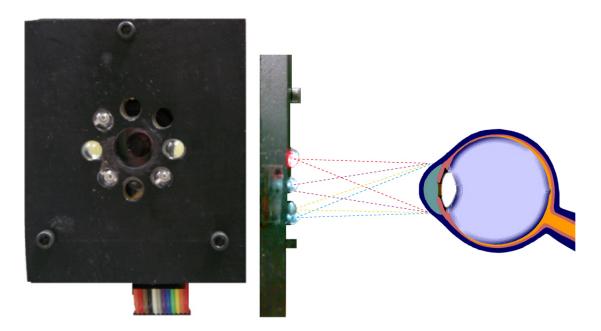


Figure 3.12: Leds illumination

The reflected rays pass through the central hole in direction of the detection system.



An advantage of this approach is that it allows to control for the reflection of the cornea and the lens. These reflections are called Purkinje's images and shown for two LED's in the following illustration:

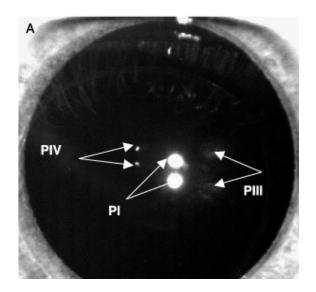


Figure 3.13: Purkinje's images

The first image (PI) is the brightest and is caused by the external layer of the cornea. The second one is difficult to see and is caused by the internal layer of the cornea. The third and fourth images (PIII and PIV) are caused respectively by the external and internal layer of the crystalline lens :

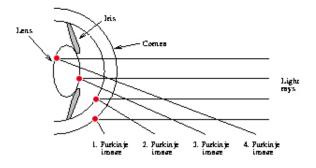


Figure 3.14: The four Purkinje's reflections

The signal received by the detector can be modified by these reflections if they are not situated in the pupil (noise in the baseline and varying intensity). A geometrical configuration must be calculated.



The distance between the leds and the eye can be chosen (the focal length of the first system's lens can be adapted in consequence, cf. chapter 4.2). Then the leds must be placed inside a cercle whose diameter can easily be calculated.

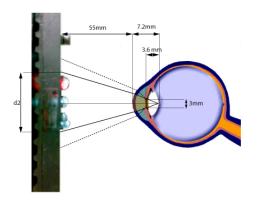


Figure 3.15: Leds' placement

If the refraction index between the different layers is fixed to 1 (ideal case, continuous line on figure 3.18), the  $d_2$  diameter can be found directly through the Thales theorem :

$$\frac{d_2}{55+7.2} = \frac{3}{3.6} \tag{3.1}$$

That means that the illumination must be placed inside a cercle of diameter 52.33mm if one wants the first Purkinje image inside the pupil.

In the system, the distance between opposite leds is a little less than 30mm as shown in the following picture so the condition for the first Purkinje image is respected :



Figure 3.16: Distance between the opposite leds

Moreover, if the real refraction indexes are considered, the angle will be bigger (discontinuous line on figure 3.18)

It was experimented with a design of the detection system and with a method to obtain an image of the illumated iris. The latter was accomplished with a CCD camera and a movable mirror. Here are two pictures of the system at the beginning. Only one led was used :

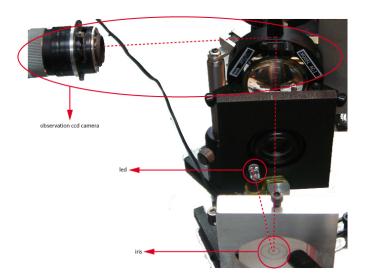


Figure 3.17: Led configuration

On the back view, it is possible to see the image on the monitor and the detection optical way :

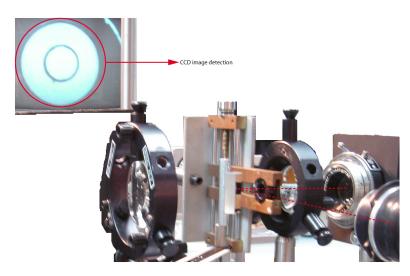


Figure 3.18: Back view and image of the target with a led

As mentionned before, with this system, the aperture is placed on the detection optical way. That means that an image of the target (iris) has to be virtually created and a diaphragm must be placed in this position. But the quality of the image is good. There is no blurred edge and the image can quite easily be adjusted (focus).

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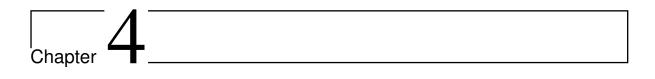
### 3.4 Synthesis

On one hand, the halogen system has the great advantage that the aperture is directly projected on the iris and that the spectrum is continuous. But its real disadvantages is the length of the optical way (about 50cm on the optical table) and the low spectral content at short wavelengths.

On the other hand, the leds illumination is very compact. Its spectrum can be more or less wide with a good combination of 8 leds but they must be geometrically well placed to avoid the Purkinje's phenomenon. The disadvantage would be that the aperture must be placed on the detection optical way.

As mentionned in the subject of the work, the system will be used for a wide study in a population of subjects and should be as light and compact as possible to facilitate its transport. Moreover, it should correspond to a product synonymous of new technology which doesn't consume a lot of energy. For all these reasons, the leds illumination has finally been chosen for the practical realization.





### Detection

### 4.1 Spectrometer

The base of the detection (and of the system) is the OceanOptics USB4000 spectrometer. Here is an image of the inside construction :

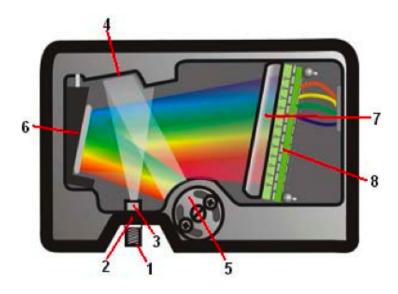


Figure 4.1: Inside view of the OceanOptics USB4000 spectrometer

Basically, the light enters the optical bench through the connector (1) and the slit (2). Then It goes onto the collimating mirror (4) and is focused towards the grating (5). The grating diffracts the light and directs the rays onto the focusing mirror (6) which focuses the light on the CCD detector (8). This detector converts the optical signal into a digital signal which is then transmitted to a software running on the Apple laptop.

In the case of the spectrometer used in the system, the slit is the  $200\mu m \cdot 1mm$ .

Finally, an external trigger can be used in the spectrometer through a 10-pin accessory connector situated next to the SMA connector (1). A pulse of 3V on a pin will engage the signal integration during a fixed time (40ms).



#### 4.2 Optical way

First of all, it was decided that the iris must not be too close to the illumination and the first lens. The subject should feel comfortable. In order to respect this constraint, the first lens used had a focal length of 75mm. When we substract the width of the illumination stand, there is still a space of about 50mm between the eye of the subject and the system.

The detection must be chosen in consequence between the first lens and the spectrometer.

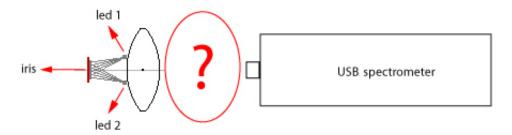


Figure 4.2: Optical way between first lens and spectrometer in the image plane of the iris formed by the first lens

The demagnification of the iris image gives rise to a physical constraint. The diameter of an iris is about 12mm and the entrance slit of the spectrometer is 1mm high. The demagnification must at least be equal to 12. But the ring-shaped aperture must be located in a plane conjugate to the iris. A second lens is used to form such image with some demagnification. The demagnification can not be too high because it is difficult to manufacturer the ring-shaped aperture if it is very small.

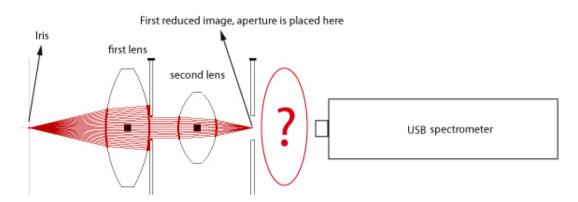


Figure 4.3: First reduced image

The second lens chosen has a focal length of 35mm. The image created is reduced by a factor  $m = \frac{75}{35} = 2.14$ 



The position of the second lens is such that it projects the image of the field stop of the first lens to infinity. Thus, a third and final lens must be used to image this aperture on the entrance slit of the spectrometer. That lens must have a very short focal length.

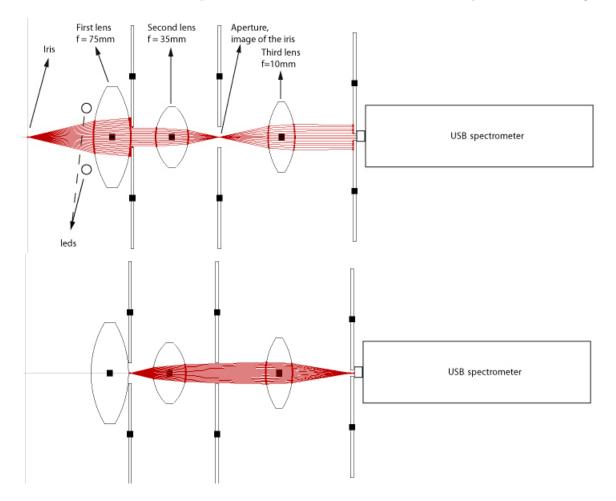
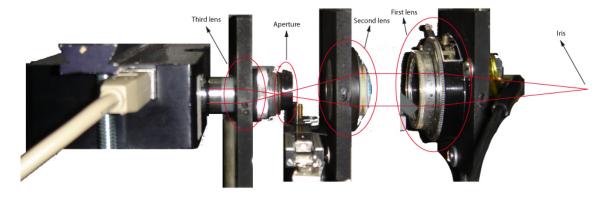


Figure 4.4: Optical configuration

Once the optical lenses calculated, many types of them have been tried. At the beginning, simple lenses were used but the quality of the image was bad. Spherical aberration was present. After a couple of tests, some higher quality elements have been chosen :

- The first lens is a Novar-Anastigmat camera objective with a focal length of 75mm with an included adjustable diaphragm.
- The second lens is a high quality doublet taken from a telescope with a focal length of 35mm.
- The third lens is a Unitron 88658 microscope objective with a 10x magnification, 0.3 NA numerical aperture and a 170 T.L. tube length.





The detection configuration was done like this :

Figure 4.5: Detection optical way

In parallel to the detection system, a CCD camera has been used in order to observe the iris, to have a control of the iris focus and to allow the alignment of the instrument. The optical path can be changed with a mirror which can move and project the rays onto the CCD camera.

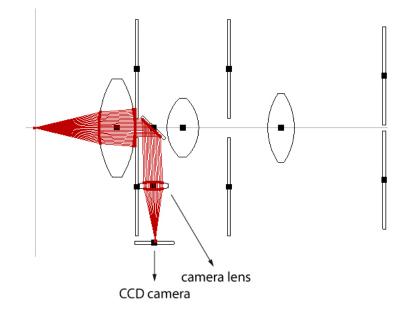
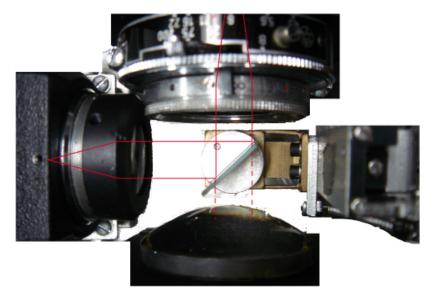


Figure 4.6: Optical way with the mirror





Here is an image of the real system with the rays focused on the CCD camera :

Figure 4.7: Image focused on the CCD camera

The mirror can move up and down. When it is in high position, the rays are projected on the CCD camera and the image created is then transmitted to a LCD screen placed on the side of the system. When it is in bottom position, the rays pass through the other lenses to the spectrometer. An operator will control the iris focus and the spectral acquisition through an electronic device.

### 4.3 Ring-shaped apertures

As introduced in chapter 3.3, an aperture has to be placed in the detection optical path. The light detected by the spectrometer must come only from the pigmented part of the iris.

In the position of the first image, a multiple aperture system has been placed. Basically, it is composed of 5 different apertures :



Figure 4.8: Detection optical way



- The empty aperture is used for calibration, optical adjustment.
- The plain black aperture is used for black reference signal acquisition.
- The different rings (Small, Medium, Large) can be used for the iris detection. It is a transparency ring of 0.75mm width with different diameters. The diameters of the central dark area are 2.75, 3.25 an 3.75mm.

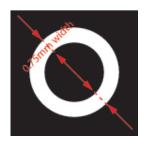


Figure 4.9: Aperture

### 4.4 Ancilliary systems

A couple of elements have been added during the development.

First, a led has been fixed in the illumination as a fixation led. The eye moved too much during a measurement.



Figure 4.10: Fixation led

Then, the operator must have his hand free for the adjustment of the focus on the screen. A foot pedal has been added for the acquisition. A pression on the foot pedal will make the solenoid work for the mirror movement.





Figure 4.11: Foot pedal

Finally, the signal integration must start once the mirror is completely out of the optical detection path. Two solutions have been tried.

First, a photosensitive sensor has been used :

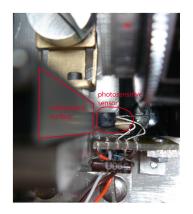


Figure 4.12: Photosensitive sensor

The problem was that the voltage didn't change perfectly between the two different states.

The other solution was a microswitch placed at the bottom of the mirror stand. The mirror stand pushes on the microswitch at the end of the movement.

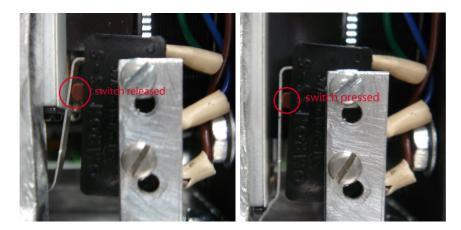


Figure 4.13: Microswitch released (left) and pressed (right)





Figure 4.14: Target lighted on by a led



Figure 4.15: Calibration with the target





Figure 4.16: Labsphere reflectance standard

This standard is not useful for all day measurement (must take care of it due to its high price).

The second one is a more practical standard. It simply a white paperboard whose reflectance in function of the wavelength can be calculated (see Chapter 6):



Figure 4.17: Paperboard reflectance standard



### 4.5 Synthesis

Different optical detections have been tried but simple lenses can't be used because of their spherical aberration. Some high quality lenses were used in the system. The first lens is a camera objective, the second one is a doublet and the third one is a microscope objective.

A multiple aperture is used in order to change the ring of light which best fits the subject's eye. An empty and a plain black aperture are available for the adjustment and white/dark calibration.

Finally, a parallel CCD camera can be used for the iris focus. An operator can watch the image of the iris on an LCD screen placed on the side of the system. This screen shows the image created on the camera when the mirror is placed in the top position. Once the focus is done, the operator can acquire the signal with the spectrometer through a solenoid that moves the mirror out of the observation light path. The LED's are then flashed to a high level current.

The system before the electronic development was the following :

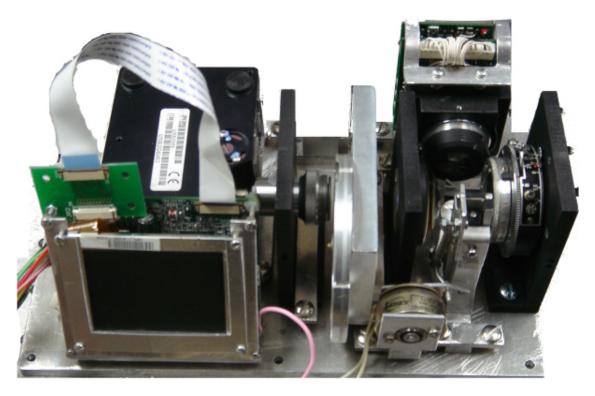
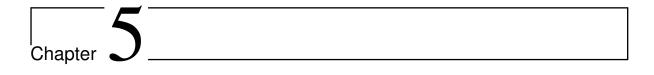


Figure 4.18: Optical system





## Electronics

### 5.1 Electronic elements

Starting from the optical system, we need to give power and to send different signals among the elements. Basically, the following picture resumes all the connections that are needed :

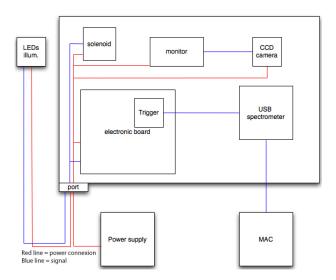
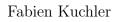


Figure 5.1: Electronic diagram

- The power supply must give power to the electronic board, the solenoid, the leds illumination, the LCD monitor and the CCD camera.
- The electronic board makes the mirror move through the solenoid.
- The image of the CCD camera is sent to the monitor.
- The trigger pulse from the electronic board is sent to the spectrometer.
- The USB spectrometer sends the analogical signal to the software running on the Apple laptop.
- The leds are switched on/off with different intensities by the electronic board.





### 5.2 Operating mode

Basically, there are three modes of operation which can be selected with a switch :

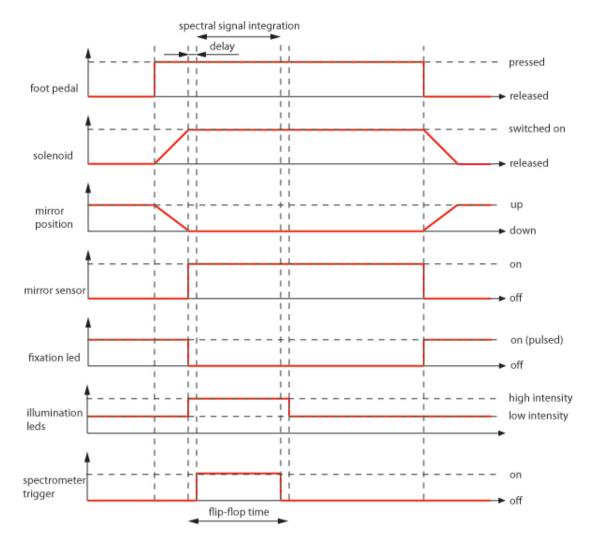
- In the off mode, the system is supplied but no element works. It can be used for dark calibration.
- In the white calibration mode, the illumination works at full intensity. Neither the foot pedal nor the pulsed fixation led are connected.
- In the acquisition mode, the foot pedal is connected, the illumination works at low intensity, the pulsed led is connected and everything is ready for a subject's spectral acquisition.

This sequential diagram shows the different steps of this mode :

Figure 5.2: Operating sequence

The subject fixes the pulsed red led (1). The operator focuses the system on the iris (2). Once it is done, he presses on the foot pedal (3). It makes the solenoid and the mirror stand move(4). The illumination is still at low intensity. When the mirror is completely down, the microswitch sends a signal to the electronic board. It lights on the leds at full intensity and stops the pulsed red led (5). After a short delay, the spectrometer sends the signal on the software during a fixed integration time (6).





In real time, the signal of the different elements are the following :

Figure 5.3: Time sequence

In order to do that, some transistors, switches and a flip-flop have been used on the electronic board. The different circuits are described in the next section.

#### 5.3 Electronic circuits

As the LCD screen, the CCD camera, the solenoid and the different leds worked at 12V, two transformators at this voltage were used. The first one supplies the solenoid only because it uses a high current (1A). The second one is used for all the other elements.

As described in the previous section, there are two illumination states : high and low intensity. The intensity must change once the microswitch is pressed. In order to make this variation, a timer and transistors were used.



Here are the different parts of the electronics diagram for each component :

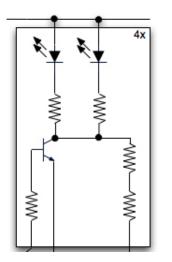


Figure 5.4: Led circuit with transistor

The led current (and its intensity) changes when the base of the transistor has a voltage coming from the LM555 timer.

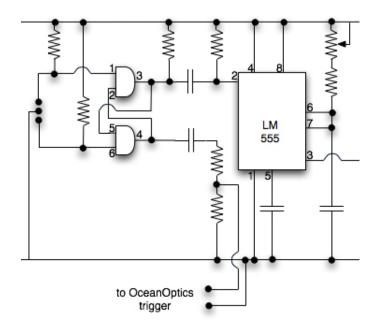


Figure 5.5: Flip-flop signal for the trigger and the timer

The timer gets the signal of a pulse generated by a flip-flop. Once the sensor is pressed, the flip-flop changes its state and a pulse is sended to the trigger and the timer.



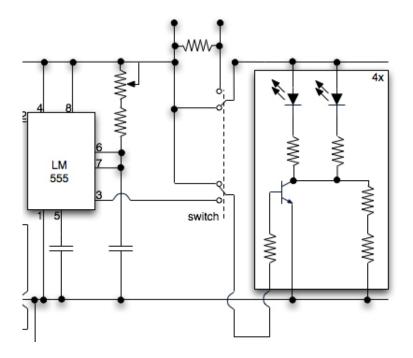


Figure 5.6: Main switch with 3 positions

The main switch has 3 positions : in the diagram position, the leds are in high mode, the timer doesn't has an influence on the leds. In the opposite connexion, the leds are in low mode, waiting for the timer signal ( output number 3) to start the transistors. In intermediate position, there are no connexions and the leds are powered off.



Figure 5.7: Circuit for the red led which indicate that the system is powered on

When the system is turned on, a led is lighted on by the 12V voltage.



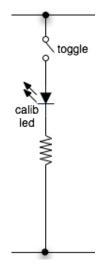


Figure 5.8: Circuit for the calibration target lighted by a led

For the calibration, the target can be lighted on with a toggle switch. Finally, all these elements have been fixed on an electronic board :

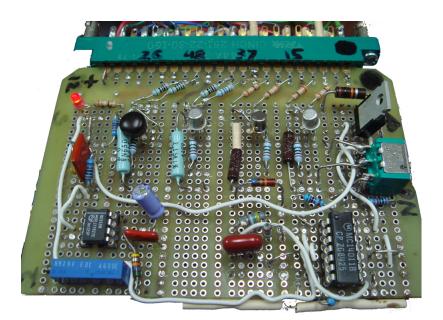


Figure 5.9: Electronic board



## Chapter 6

## Calibration and test

#### 6.1 Safety

If the system is used for a wide study on subjects, the security of the eye has to be ensured because an organ will control the safety level before approving the faisability of the study . A program called OcLC (Ocular Light Calculator) has been developped on the excel platform by Francois Delori.

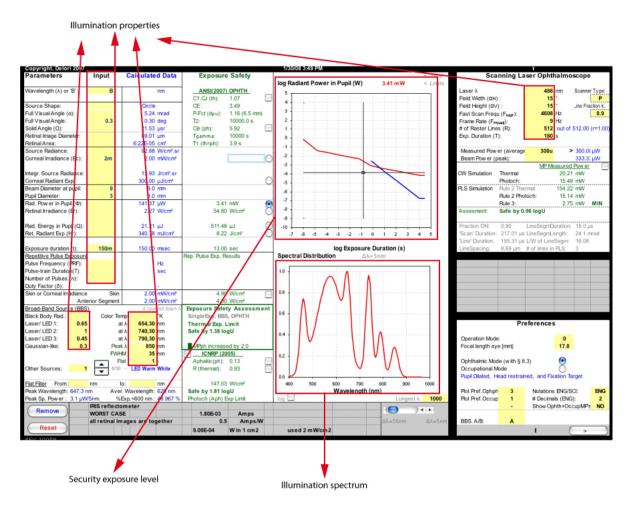


Figure 6.1: Light calculator printscreen



The illumination can be completely detailed in the different entries like the visual angle, the irradiance, the time exposure, the spectral width of the different leds and so

The spectrum of the illumination is reconstructed as well as the radiant power and exposure duration graph. On this last plot, the crossing of the black lines gives the operating point (the actual exposure of the reflectometer). The red and the blue line are the safety limits. The system is thus safe.

#### 6.2 Calibration

on.

For the signal processing, three calibration spectra must be acquired :

• White calibration with a high reflectance standard (>98% at all wavelengths) and the leds illumination as light source

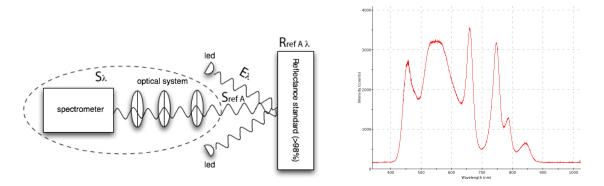


Figure 6.2: White calibration with the reflectance standard

• White calibration with a white paperboard and the leds illumination as light source

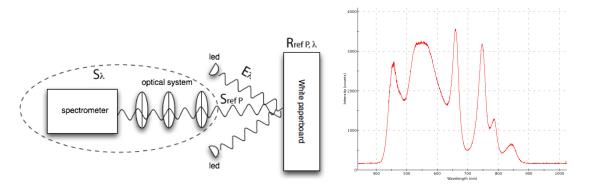
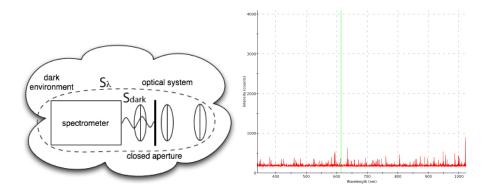


Figure 6.3: White calibration with the paperboard





• Dark calibration with the aperture closed in a dark room

Figure 6.4: Dark calibration with closed aperture

Then, a measurement can be done on a subject :

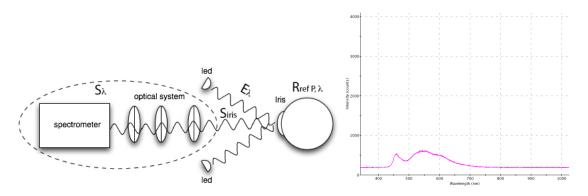


Figure 6.5: Iris measurement

The  $E_{\lambda}$  represents the light intensity in function of the wavelength  $\lambda$ .  $S_{\lambda}$  is the sensitivity of the spectrometer with the optical elements,  $R_{ref}$  correspond to the reflectance of each case and S is the signal acquired by the spectrometer.  $N_{\lambda}$  in the following equations is the dark noise.

The different signals can be found using these relations :

$$S_{iris} = E_{\lambda} \cdot R_{iris,\lambda} \cdot S_{\lambda} + N_{\lambda} \tag{6.1}$$

$$S_{dark} = 0 \cdot 0 \cdot S_{\lambda} + N_{\lambda} = N_{\lambda} \tag{6.2}$$

$$(S_{iris} - S_{dark}) = E_{\lambda} \cdot R_{iris,\lambda} \cdot S_{\lambda}$$
(6.3)

 $S_{iris}$  is the spectrum acquired by the spectrometer and  $R_{iris,\lambda}$  is the reflectance that must be calculated (figures 6.3 and 6.4)



$$S_{ref} = E_{\lambda} \cdot R_{ref,\lambda} \cdot S_{\lambda} + N_{\lambda} \tag{6.4}$$

$$(S_{refA} - S_{dark}) = E_{\lambda} \cdot R_{refA,\lambda} \cdot S_{\lambda}$$
(6.5)

$$(S_{refP} - S_{dark}) = E_{\lambda} \cdot R_{refP,\lambda} \cdot S_{\lambda}$$
(6.6)

 $S_{refA}$  is the spectrum acquired by the spectrometer when the iris is replaced by the white reflectance standard which has a reflectance of  $R_{refA,\lambda} = 98\%$  (figure 6.1).

 $S_{refP}$  is the spectrum acquired by the spectrometer when the iris is replaced by the white paperboard (figure 6.2). Its reflectance  $R_{refP,\lambda}$  can be calculated in dividing equation 6.6 by equation 6.5.

$$R_{refP,\lambda} = R_{refA,\lambda} \cdot \frac{(S_{refP} - S_{dark})}{(S_{refA} - S_{dark})}$$
(6.7)

Finally, in dividing equation 6.3 by equation 6.6,  $R_{iris,\lambda}$  becomes :

$$R_{iris,\lambda} = R_{refP,\lambda} \cdot \frac{(S_{iris} - S_{dark})}{(S_{refP} - S_{dark})}$$
  
$$= R_{refA,\lambda} \cdot \frac{(S_{iris} - S_{dark})}{(S_{refA} - S_{dark})}$$
  
$$= 98\% \cdot \frac{(S_{iris} - S_{dark})}{(S_{refA} - S_{dark})}$$
(6.8)

#### 6.3 Results

Two subjects were tested with the reflectometer. Integration duration was adjusted to avoid signal saturation.

The spectra of the reflected light are shown below for the reference, Francois's iris (FD) and Fabien's iris (FK). These spectra are normalised to a 100ms exposure :

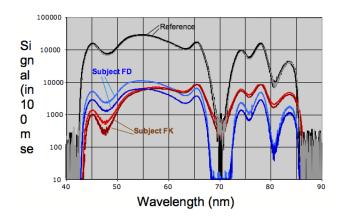


Figure 6.6: Normalised spectra in log scale



Then, the iris reflectance spectra are obtained by dividing the iris signal by the reference signal. The reflectance spectra obtained are the following :

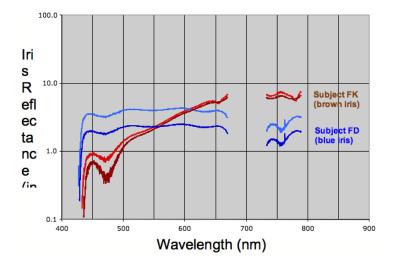


Figure 6.7: Iris reflectance

First of all, the data at around 700nm have been deleted because it was only noise (the illumination has no intensity at 700nm).

A interesting result is that although the reflectance at short wavelength is lower for the brown iris than for the blue iris, the reflectance in the NIR is higher for the brown iris than for the blue iris. This behavior was also observed for the reflectance of the fundus (retina+choroid). It may be that the melanosomes scatter light increasing thereby the reflectance when the absorption of melanin is low (in the NIR).

Here, the data are compared with the reflectance of the finger skin :

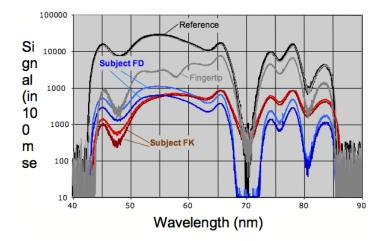


Figure 6.8: Iris and fingertip skin reflectance



As expected, the graph is similar, with a maxima in long wavelengths and the spectral bands of hemoglobin at 540 and 575 nm.

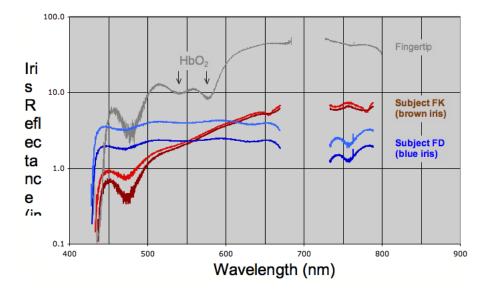


Figure 6.9: Iris and skin reflectance

#### 6.4 System improvement

Finally, some imperfections are present on the graph. For example at the 480nm wavelength, the decreasing signal is due to a non linearity of the illumination. A better alignement of the leds could give a more constant signal.

After several tests, it appears that several things have to be improved :

- The weak signal at 700nm must be completed by adding a led. The signal treatment at this wavelength gives a false noise signal.
- The leds alignement must be more precise. When the illumination is slightly tilted, the signal should change similarly at all wavelengths and it doesn't.
- The apertures are too small. They should be optimized.
- The optical way should be controlled. Apparently, the image detected is a slit of the ring-shaped image and not the entire surface.
- The delay between the leds lighting at full intensity and the beginning of the signal integration must be modified. Sometimes the integration is not synchronized.



#### | Chapter

## Conclusion

#### 7.1 Project

The aim of the work was to construct a small reflectometer to measure the reflectance spectum of the iris and to make a study on a population of subjects.

The second part of the project hasn't been reached. It seems that I was unfortunate. The leds that I had ordered were not shipped at once and finally arrived very late. During the last months, the two spectrometers became out of order and it took time to receive a new one. The LCD screen stopped working, too. I had to change it and modify the enclosure of the system.

Because of these technical problems, the system was operational only during the last week of my work... Then we were able to make measurements.

The optical conception is efficient. The simulations done with the playOptics software were close to reality and the practical construction became easier.

The electronic part is well conceived. A couple of things have been added or modified during the last weeks to facilitate the use of the system. The different signals are working.

The first signal treatment is working. Now it could be interesting to start the biological part of this work to get some results. Eventually, a small software could be useful for an automatic calculation of the iris reflectance with the different signals acquired during the calibration and the iris measurements.

#### 7.2 Personal comments

Even if we didn't start the study on a population of subjects, I really enjoyed working on this project. The conception is achieved and it was a great experience for me to start from an existing optical bench and to make the optical and electronical developments around it. Moreover, this system can be used for optical research but, if the results are satisfying, it could eventually be used for an industrial application in security or biometric identification.

Then, I appreciated working with Mr Francois DELORI. He was available and his long experience has helped me all along these 6 months. His different approach of optical engineering made me think about the theorical calculations and the practical realization. He sometimes simply tried solutions while I was deeply calculating how it could be real-



ized.

It was very different from the optic laboratory of the HES. At the Schepens Eye institute, we didn't have all the material at disposal. We had to try the optical system with the lenses we had, think if it was really necessary to order different elements and find other simple solutions. The same approach was done for the electronics. We had to order leds or other components only if the result was not satisfying with the material we had ("That's life !" as Francois likes to say).

This approach often leads to more ingenious solutions.

#### 7.3 Supervision

The project has been supervised and this report accepted and approved by Mr Francois DELORI.

Boston, March 4, 2008

Francois DELORI

Fabien KUCHLER



## Chapter **C**

## Acknowledgment

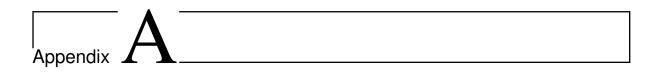
This diploma work is the result of a collaboration between the HES-SO Valais in Sion and the Schepens Eye Research Institute in Boston. It couldn't have been done without the help of many people that I would like to thank :

- Mr Francois DELORI from the SERI who invited me to Boston in order to do my diploma work, who supervised the work and helped me during these 6 months.
- Mr Martial GEISER from the HES-SO Valais in Sion who found me the post in Boston and supervised my work from Sion.
- Mrs Karine AMOS and Mrs Laurie KOCH from the MOVE office of the HES-SO Valais in Sion and Mrs Megan PORCARO from the human ressources of the SERI who helped me for all the administrative tasks.
- All the collaborators of the SERI and especially Mr Frank ROGERS who developped the electronic board.
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- [5] Reflectance standard, *http://labsphere.com*
- [6] Histology, http://www.missionforvisionusa.org/anatomy/2005/10/iris-histology.html
- [7] Purkinje images, http://www.journalofvision.org/6/10/5/article.aspx
- [8] Melanin absorbance, http://www.cl.cam.ac.uk/jgd1000/melanin.html
- [9] All others images, *http://images.google.com*
- [10] All optical simulations realised with *playOptics software*, 2004, Jared Schiffman, *http://www.jarfish.com*
- [11] All spectra acquired with *SpectraSuite software*, OceanOptics *http://www.oceanoptics.com*
- [12] Light safety level printscreen from Ocular Light Calculator, v.OcLC\_08.81, Francois Delori



### Lenses

Specifications of the lenses :

Lens 1 : Novar-Anastigmat camera objective lens, focal length = 75mm, f number = 1:4.5.



Figure A.1: Camera objective lens

Lens 2 : telescope doublet lens, focal length = 35mm.



Figure A.2: Doublet lens

Lens 3 : Unitron 88658 microscope objective lens, 10x magnification, 0.3 NA numerical aperture and 170 T.L. tube length.



Figure A.3: Microscope objective lens



Light source



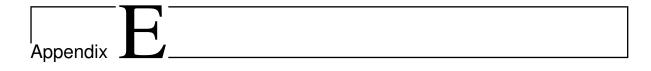
Enclosure



# Appendix D

Reflectance standard





## Electronics

Electronic diagram :

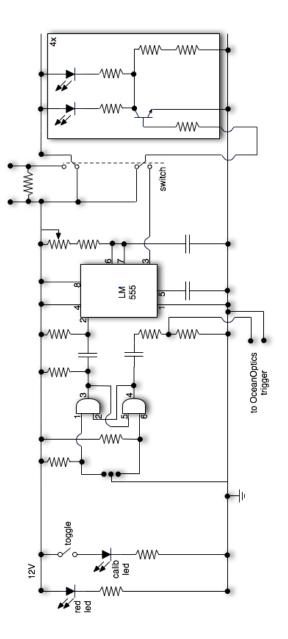
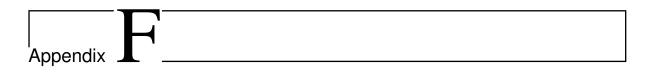


Figure E.1: Electronic diagram





## Workshop and material

Material used for machining and electronics development :



Figure F.1: 465 Oscilloscope



Figure F.2: D-612T voltage source





Figure F.3: Driller



Figure F.4: Driller and milling machine



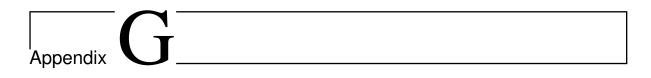


Figure F.5: Circular saw



Figure F.6: Alu plate cutter





## System pictures

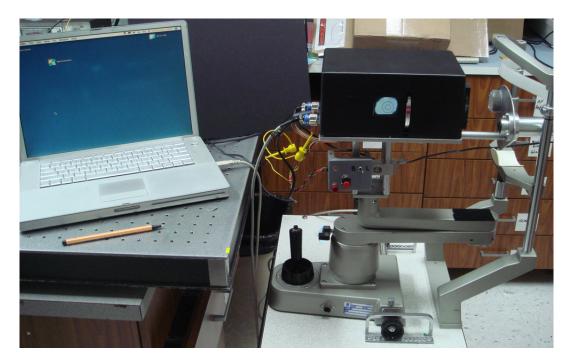


Figure G.1: System

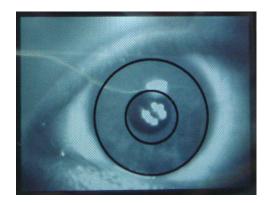


Figure G.2: Iris focus screen



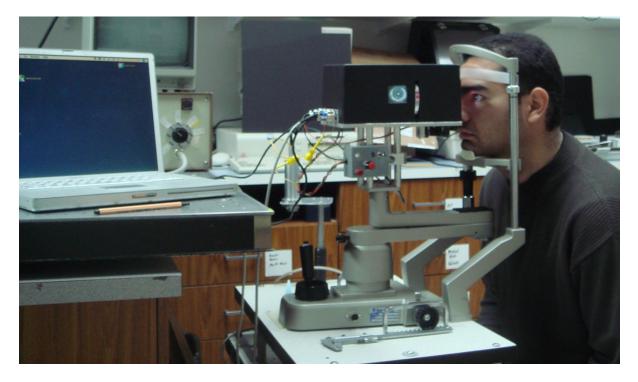


Figure G.3: Subject measurement

