

# Title of the document

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## Abstract

The report starts with a Samenvatting (Abstract). The abstract should be self-contained, i.e. a reader should be able to fully understand it without any prior knowledge about the research. Also, in the abstract there should be no references to (figures, tables, formulas etc. in) the remainder of the report, nor to the literature. The abstract tells the reader:

- (1) which research question has been studied,
- (2) what the research method/approach was,
- (3) which results have been obtained, and
- (4) what the main conclusions were.

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## 1 List of symbols

bla bla bla

bbb

## 2 Introduction

Microscopes are used extensively in natural sciences. They enable us to image small objects and structures which cannot be resolved by the human eye. The use of microscopes, could for example, aid in Studies of biological cells, molecule structures or object classification. To correctly conduct such microscopy studies, it is vital to know what the possibilities and limits are of the particular microscope in combination with image improvement techniques.

This experiment will focus on a Leica DM EP microscope in combination with a Color CCD and to what extent this set-up can be used to measure the size of small objects and find the birefringence of an unknown crystal. Furthermore, the possibilities of digital image improvement will be investigated.

After calibrating the pixels and finding the resolving power with the aid of respectively a microscopic ruler and a resolution target, images are made of a human hair, an optical glass fibre, starch particles, an unknown birefringent crystal and a biological sample of a fungus. To find the size of the human hair, optical fibre and starch particles, pixels are to be counted by a computer. By focussing on the different coloured layers of the crystal, it is possible to find the thickness of each layer and subsequently calculate the birefringence. Finally, some python algorithms are implemented on the image of the fungus to investigate improvements on contrast, colours and corrections.

In section 2 the theory regarding the experiment will be described, followed by the the experimental method in section 3. The results and discussion can be found in section 4. Lastly will be the conclusions in section 5.

### 3 Theory

In the Theorie (Theory) chapter, you describe all (and only!) the theory needed to understand and interpret the experiments in the remainder of the report. Explain to the reader why a piece of theory is relevant for your research. Equations should be numbered. If an equation cannot be assumed to be generally known by the readers (see General Hint 2 for the level of the audience), you should provide a reference to an accessible textbook or article (so not to the RP manual, lecture notes, Wikipedia etc.). In general, try to avoid referring to websites, online data or Wikipedia.

## 4 Experimental method

This experiment consists of four parts. First the calibration of the microscope. Secondly size measurements on respectively a human hair, an optical glass fibre and starch particles. Thirdly determining the birefringence of an unknown crystal and finally the computerized improvement of an image of a biological fungus sample. The different experimental methods will be treated separately.

The microscope that is used in all experiments is a Leica DM EP microscope. Its manual can be found in appendix A. This microscope is used in combination with 4 $\times$ , 10 $\times$ , 40 $\times$  Hi Plan POL objectives with respectfully a 0.10, 0.22 and 0.65 numerical aperture. A color ccd camera in combination with NI Vision Assistant software is used to acquire digital images.

### 4.1 Calibration

A microscopic ruler is used to measure the length that corresponds to one pixel in an image. This is achieved by focussing on a 1 mm, 100 division ruler and measuring the distance between two focussed, distant division lines and comparing the number of pixels to the physical length. The NI software is used to find the exact location of these two lines and subsequently find the perpendicular projection (see Figure ??). This procedure is repeated for all three objectives.

With the aid of a 1951 USAF resolution target, the resolving power of each objective can be found. First taking a focussed grayscale image on the target and then taking a perpendicular intensity profile for each well defined three-bar structure (see Figure ??). The visibility can subsequently be calculated with equation ?? and the corresponding spatial frequency. This is repeated for all three objectives.

### 4.2 Microscopic size measurements

All microscopic size measurements are made by measuring pixels and comparing this to the corresponding pixel length. This is done for images with the 40 $\times$  objective since this gives the smallest error.

#### 4.2.1 Human hair and optical glass fibre

Measuring the thickness of the human hair and optical glass fibre is done with the aid of the NI Vision software. First finding the two straight lines of the outer edges and subsequently measuring the perpendicular distance between the two (see Figure ??).

#### 4.2.2 Starch particles

In order to measure the size of individual starch particles, a small amount of starch is mixed with oil. Images are taken at different locations in the mixture, such that approximately 30 particles are focussed in the ...

## 5 Results and discussion

In the Resultaten en discussie (Results and discussion) chapter, you present your results, generally in the form of graphs, and you discuss them. A single small table (maximum 10 rows x 5 columns) is acceptable, but large tables should be in an appendix. In deviation from what many students believe, it is not desirable to separate the presentation and the discussion of results from each other. In professional literature, this is most often done together.

- You should introduce each graph:

- Why has this graph been included in the report. (What do we want to learn from this graph?).

- Why have you plotted this Y-axis variable as a function of this X-axis variable (which theoretical/expected relationship is tested/demonstrated in this graph)?

- Then you tell the reader what (according to you) he/she should see in the graph, limiting yourself to conclusions that are relatively indisputable. The more speculative conclusions should be in the next chapter.



## 6 Conclusions

In the Conclusions (Conclusions) chapter

- You give a clear and concise answer to the research question that was formulated in the Introduction
- You discuss to what extent, and why, your findings do (not) agree with theory/expectations/earlier work, you discuss more speculative conclusions, and you may do suggestions for further (improved/extended) research. The Conclusions should be self-contained and understandable for readers that have only read the introduction (and have not read the rest of your report, do not know the literature, do not know the experimental setup and have not read the RP manual). In the Conclusions chapter, you may not make references to graphs, tables, equations etc. in the remainder of the report.

## A Leica MP EM microscope manual

# Leica DM EP

## The Microscope for Teaching and Research

- **Advanced performance in a teaching polarizing microscope:**
  - Standard and advanced conoscopy modules
  - Polarizer with notch markings
  - 4-position objective turret, centerable
  - Sturdy, compact design
- **Convenience that makes work easy:**
  - Easy-to-access control functions
  - Ergonomic viewing angle
  - Accurate angular measurement with verniers on the rotating stage



Developed for college teaching and research use: the Leica DM EP.

### **Accurate and versatile for teaching**

The Leica DM EP is the ideal polarizing microscope for university and other instructional use, offering a standard and an advanced Bertrand lens module for unsurpassed ease of operation. With a wide range of accessories and Leica's renowned optics, the Leica DM EP is exceptional not only for its compact, durable design, but also for its efficiency and ease of operation.

### **Designed for optical brilliance and long life illumination**

The standard Köhler field diaphragm and magnetically fixed blue filter provide vivid, pin-sharp images. The 2,000-hour, 35-watt halogen lamp saves hundreds of dollars in replacement bulb cost over the life of the microscope. An illuminated intensity control system reminds the user to switch off the lamp after finishing work to increase the lamp's service life and save energy.



Maximum ease of use and high optical brilliance are the outstanding features of the Leica DM EP.

# Modular, Customized Configurations – Microscopes Designed for You

- **Flexibility that gives the freedom you need:**

- Wide selection of POL objectives

- **Compatibility that knows no bounds:**

- Fully compatible components across Leica's polarizing microscope product line
- Wide selection of analyzers, polarizers, and compensators
- Full wave & quarter wave plates are available
- Wide selection of POL observation tubes

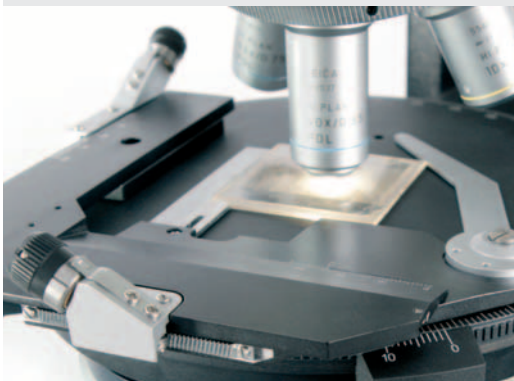


The result of combining maximum precision and optimum ergonomic design – the 360° analyzer.

## **Flexibility – Designed for you**

Flexible to the last detail. All Leica polarizing microscope components can be configured for all microscopes in the polarizing line. For example, you can choose from over twenty POL objectives for the Leica DM4500 P, DM2500 P or DM EP. The optical possibilities are unlimited. You will enjoy the benefits provided by this complete system when using the new 360° analyzer, the 360° polarizer or even with full wave plates. All components can be used for classroom teaching, everyday routine work, and research.

Leica's entire line of DIN standard compensators can be used in all Leica polarizing microscopes, as can the attachable mechanical stage for accurate sample positioning. This always ensures flexible interchange and replacement of parts.



Flexibility is key. All of Leica's rotating stage polarizing microscopes feature attachable, interchangeable mechanical stages.

# Technical Data

	Leica DM EP	Leica DM2500 P	Leica DM4500 P
• <b>Objective turret</b>	4x (M25), centerable	5x (M25), centerable	6x (M25), centerable, absolute encoded
• <b>Objectives</b>	HI Plan POL N Plan POL  Immersion objectives	HI Plan POL N Plan POL PL Fluotar POL Immersion objectives	HI Plan POL N Plan POL PL Fluotar POL Immersion objectives
• <b>Usable field of view</b>	20 mm	25 mm	25 mm
• <b>Contrast method Changeover Color reproduction</b>	Manual	Manual	Motorized CCIC: Constant Color Intensity Control
<b>Transmitted light</b>	Polarization contrast Orthoscopy Conoscopy Brightfield Phase contrast	Polarization contrast Orthoscopy Conoscopy Brightfield Phase contrast DIC	Polarization contrast Orthoscopy Conoscopy Brightfield Phase contrast DIC
<b>Incident light</b>	Darkfield Polarization contrast Brightfield	Darkfield Polarization contrast Brightfield Darkfield* DIC Fluorescence	Darkfield Polarization contrast Brightfield Darkfield* DIC Fluorescence
• <b>Conoscopy</b>	Bertrand lens cube in new IL axis Bertrand lens module (AB module) Advanced conoscopy module	Bertrand lens cube Bertrand lens module (AB module) Advanced conoscopy module	Fully integrated conoscopy beam path User guidance with display feedback
• <b>Transmitted light axis Illumination Operation</b>	12 V 35 W halogen lamp Manual User guidance with CDA	12 V 100 W halogen lamp Manual User guidance with CDA	12 V 100 W halogen lamp Motorized Integrated illumination manager
• <b>Incident light axis</b>	Manual User guidance with CDA	Manual User guidance with CDA	Motorized Integrated illumination manager, round and rectangular field diaphragms for ocular or camera observation
• <b>Condensers</b>	Manual changeover User guidance with CDA	Manual changeover User guidance with CDA	Motorized changeover of condenser head, 7x condenser disc, polarizer
• <b>Focus drive</b>	Manual, 2-gear gearbox	Manual, height-adjustable, Focus stop, 2 or 3-gear gearbox	Manual, 2-gear gearbox

\* on request

Even testen of dit appendix systeem een beetje werkt.