



FEDERAL INSTITUTE OF CEARÁ
IFCE *CAMPUS* FORTALEZA
GRADUATE PROGRAM IN TELECOMMUNICATIONS ENGINEERING

ODILON LINHARES CARVALHO MENDES

**AUTOMATIC SEGMENTATION OF MACULAR HOLES IN OPTICAL
COHERENCE TOMOGRAPHY IMAGES**

FORTALEZA / CE
2020

ODILON LINHARES CARVALHO MENDES

AUTOMATIC SEGMENTATION OF MACULAR HOLES IN OPTICAL COHERENCE
TOMOGRAPHY IMAGES

Completion of course work presented to the Graduate Program in Telecommunications Engineering at the Federal Institute of Education, Science and Technology of Ceará (IFCE) - *Campus* Fortaleza, as a partial requirement to obtain the Master's Degree in Telecommunications Engineering. Concentration area: Engineering IV.

Advisor: Prof. Dr. Auzuir Ripardo de Alexandria.

Co-advisor: Dr. Abrahão da Rocha Lucena.

FORTALEZA / CE

2020

Dados Internacionais de Catalogação na Publicação
Instituto Federal do Ceará - IFCE
Sistema de Bibliotecas - SIBI
Ficha catalográfica elaborada pelo SIBI/IFCE, com os dados fornecidos pelo(a) autor(a)

- M538a Mendes, Odilon Linhares Carvalho.
 Automatic segmentation of macular holes in optical coherence tomography images / Odilon Linhares Carvalho Mendes. - 2020.
 124 f. : il. color.
- Dissertação (Mestrado) - Instituto Federal do Ceará, Mestrado em Engenharia de Telecomunicações, Campus Fortaleza, 2020.
 Orientação: Prof. Dr. Auzuir Ripardo de Alexandria.
 Coorientação: Prof. Dr. Abrahão da Rocha Lucena.
1. Computer Vision (CV). 2. Macular Hole (MH). 3. Optical Coherence Tomography (OCT). 4. Segmentation. I. Título.

CDD 621.382

ODILON LINHARES CARVALHO MENDES

AUTOMATIC SEGMENTATION OF MACULAR HOLES IN OPTICAL COHERENCE
TOMOGRAPHY IMAGES

Completion of course work presented to the Graduate Program in Telecommunications Engineering at the Federal Institute of Education, Science and Technology of Ceará (IFCE) - *Campus Fortaleza*, as a partial requirement to obtain the Master's Degree in Telecommunications Engineering. Concentration area: Engineering IV.

Approved in: ____ / ____ / ____.

EXAMINATION BOARD

Prof. Dr. Auzuir Ripardo de Alexandria (Advisor)
Instituto Federal do Ceará (IFCE) – Campus Fortaleza

Prof. Dr. Abrahão da Rocha Lucena (Co-advisor)
Escola Cearense de Oftalmologia

Prof. Dr. Tarique da Silveira Cavalcante
Instituto Federal do Ceará (IFCE) – Campus Fortaleza

Prof. Dr. Francisco José Alves de Aquino
Instituto Federal do Ceará (IFCE) – Campus Fortaleza

Prof. Dr. Daniel da Rocha Lucena
Escola Cearense de Oftalmologia

ACKNOWLEDGMENTS

After completing a challenging undergraduate course in Mechatronics Engineering, I never thought I was going to do two master's degrees at the same time. IFCE and ITA welcomed me to this challenge. Looking back, I can now see that, after all the effort, all the week and weekend classes, and all the days and nights of study, I must make the due acknowledgments.

I would first like to thank God, for having lit my way, where I met wonderful people and learned more than I expected. I would also like to thank my parents, Chagas and Elisabete, who have always supported and encouraged me along this difficult journey, giving me the strength to continue fighting for my goals. I could not have made it without their support and comprehension!

I am grateful to the team of professors from the PPGET, who went to great lengths to pass on their knowledge, and especially to my advisor, Prof. Dr. Auzuir Ripardo, for all his support, dedication and time invested in reviewing this work and providing valuable feedback. My special thanks also go to IFCE and the LEM laboratory, places that I will certainly remember.

Furthermore, this work could not have been done without the help of *Escola Cearense de Oftalmologia*, especially Dr. Abrahão Lucena and Dr. Daniel Lucena, who in addition to providing knowledge, provided important data for this work.

I also thank the State Government of Ceará, for sponsoring my studies and research and for believing that education is the basis for the construction of a great State.

“Technology is a gift of God. After the gift of life it is perhaps the greatest of God's gifts. It is the mother of civilizations, of arts and of sciences.”

Freeman Dyson

RESUMO

A retina faz parte do sistema ocular responsável pela visão. Em sua região central encontra-se a mácula, que permite uma visão detalhada. Uma doença macular distinta, menos observada, é o Buraco Macular (BM). É uma condição que causa diminuição da visão e ocorre devido ao uso excessivo da fóvea, miopia alta, trauma no olho e severa exposição ao sol, na qual um buraco altera a estrutura da retina. O tratamento ou uma possível cirurgia depende do tamanho e forma do BM. A identificação de BM em imagens de Tomografia de Coerência Óptica (OCT) pode ser analisada por meio dos limites superiores da Membrana Limitante Interna (MLI) e do Epitélio Pigmentar da Retina (EPR). A segmentação manual delonga tempo e se torna desvantajosa. A segmentação automática é rápida e de baixo custo computacional, sendo interessante para auxiliar médicos especialistas. Sendo assim, o principal objetivo deste trabalho é desenvolver um algoritmo que segmenta automaticamente a camada MLI e a área do BM em imagens OCT. Outros objetivos são: aquisição automática das medidas do BM e segmentação da camada MLI para imagens sem BM, provando que o algoritmo também pode ser usado em Mácula Normal (MN). A base de dados é pública e possui 102 imagens OCT com BM e 206 imagens OCT com MN. Técnicas de pré-processamento são usadas para remover ruídos e melhorar a qualidade da imagem. A segmentação da camada MLI é realizada através de um conjunto de técnicas envolvendo Distância Euclidiana e Algoritmos de Inundação e Seguimento de borda. A média e o desvio padrão (DP) do erro absoluto (EA) em pixels são de $2,72 \pm 1,91$ e o tempo médio de processamento (TMP) por imagem é 0,24s. Na segmentação da área do BM, o mesmo conjunto de técnicas é utilizado. Os resultados são: acurácia de 99,37%, Interseção sobre União (IsU) de 78,61%, Coeficiente de Similaridade Dice (DSC) de 86,98%, Coeficiente de Correlação de Matthews (MCC) de 87% e TMP por imagem de 19,95s. Para as medições do BM, a média e DP do EA para todas as medidas é menor que $107 \mu\text{m}$. A média e DP do EA em pixels para a camada MLI com MN são de $3,38 \pm 1,89$ e o TMP por imagem é de 8,27s. Para todas as aplicações, o método proposto alcançou resultados satisfatórios. A segmentação automática do BM e a extração de suas medidas serve como contribuição para auxiliar no diagnóstico médico dessa patologia.

Palavras-chave: Visão Computacional (CV). Buraco Macular (BM). Tomografia de Coerência Óptica (OCT). Segmentação.

ABSTRACT

The retina is a part of the ocular system responsible for vision. In the central region of the retina is the macula that enables detailed view. There is a distinct but uncommon macular disease called Macular Hole (MH). It causes a condition of low vision related to the weakening of the fovea, high myopia, eye trauma and severe exposure to the sun; a MH alters the natural retinal structure. The treatment or a possible surgery depends of the size and shape of the MH. An MH can be identified in Optical Coherence Tomography (OCT) images through the top boundaries of the Internal Limiting Membrane (ILM) and the Retinal Pigment Epithelium (RPE). Manual segmentation of OCT images is time consuming whereas automatic segmentation is fast and has a low computational cost, and consequently of interest to specialists. Thus, the main objective of this work is to develop an algorithm that automatically segments the ILM boundary layer and the area of the MH in OCT images. Other objectives that were also pursued included the automatic acquisition of MH measurements, and segmentation of the ILM boundary layer for images without MH, in order to demonstrate that the algorithm can also been used for Normal Macula (NM). The database used in this work is public and has 102 OCT images with MH and 206 OCT images with NM. Preprocessing techniques were applied to remove noise and enhance image quality. The ILM boundary layer segmentation was performed through a set of techniques involving shortest distance from a point to a curve (Euclidean Distance), Flood Fill and Border Following algorithms. The mean and standard deviation (SD) of the unsigned error (UE) in pixels were 2.72 ± 1.91 and the medium processing time (MPT) per image was 0.24s. The same set of techniques was used to segment the MH area. The results were: accuracy of 99.37%, Intersection over Union (IoU) of 78.61%, Dice Similarity Coefficient (DSC) of 86.98%, Matthews Correlation Coefficient (MCC) of 87% and MPT per image of 19.95s. The mean and SD of UE for all the MH measures were less than 107 μm . The mean and SD of UE in pixels for the ILM boundary layer segmentation with NM were 3.38 ± 1.89 . The MPT per image was 8.27s. The proposed method reached satisfactory results for all applications made. The automatic segmentation of MH and the extraction of its measures is a significant contribution to aid the medical diagnosis of this pathology.

Keywords: Computer Vision (CV). Macular Hole (MH). Optical Coherence Tomography (OCT). Segmentation.

LIST OF FIGURES

Figure 1: Eye anatomy	18
Figure 2: OCT images with (a) macular hole and (b) normal macular	19
Figure 3: Some metrics of MH pathology	20
Figure 4: 2D view of a 3D SD-OCT image of the retina (left) and a B-scan image (top right). One vertical A-scan of the B-scan is shown in the plot (bottom right)	21
Figure 5: Standard OCT plan based on a low time-coherence Michelson interferometer. The intensity IE at the interferometer exit depends on the sample response $h(x, z)$ convolved with the source coherence function $rSource(z)$. LS = low time-coherence light source; PC = personal computer	27
Figure 6: Pixel coordinates	28
Figure 7: Effects of (a) sampling and (b) quantization	29
Figure 8: Point and local operations	30
Figure 9: Frequency domain operation on an OCT image. (a) Input image, (b) magnitude spectrum and (c) output filtered image	31
Figure 10: Gaussian Filter with different kernels applied to an OCT image	32
Figure 11: Example of histogram equalization. (a) Original OCT image, (b) whitened image and (c) darkened image	34
Figure 12: (a) Original OCT image, (b) histogram with cut-off point T (red) and (c) thresholding image segmentation	35
Figure 13: A series of dilations and erosions	36
Figure 14: Growing-and-merging method. (a) Input image, (b) super pixels result, (c) clustering result and (d) merged result	38
Figure 15: Edge detection in a binary OCT image	39
Figure 16: Flood Fill algorithm	41
Figure 17: Surroundings among (b) connected components (c) and borders	43
Figure 18: Shortest distance from a point to a curve	44
Figure 19: Intersection of images	46
Figure 20: The best and most effective keywords	48
Figure 21: Magenta color denotes the classic shortest path segmentation and green color denotes the proposed algorithm, length-adaptive segmentation	49
Figure 22: Top images show measures of MH. Bottom images show the formation of a MH volume	49

Figure 23: MH volume formed through segmentation of ILM and RPE	50
Figure 24: Flowchart of the process	55
Figure 25: Difference of brightness levels between images	58
Figure 26: OCT image with an area marked in red to measure the brightness level.....	58
Figure 27: (a) Original image and (b) Gaussian Blur filtered image.....	59
Figure 28: Segmentation results for different cutoff values; (a) 40, (b) 45, and (c) 50.....	60
Figure 29: (a) Gaussian Blur filtered image and (b) thresholded image	60
Figure 30: Threshold and segmentation (a) without brightness adjustment and Gaussian Blur filter, (b) without brightness adjustment, (c) without Gaussian Blur filter and (d) with both of them.....	61
Figure 31: (a) Thresholded image and (b) image after a series of erosions and dilations..	62
Figure 32: Segmentation (a) without and (b) with erosion and dilation application	63
Figure 33: Seed point (cross mark) in an OCT image	64
Figure 34: (a) A series of erosions and dilations and (b) Flood Fill application.....	64
Figure 35: (a) ILM boundary layer segmentation on the original image, and (b) on the background	65
Figure 36: (a) Euclidean distance to find the shortest points (white) through the seeds (cross marks), (b) three regions and the seed points to apply the Flood Fill to, (c) MH area in black, (d) MH area after inversion of colors.....	66
Figure 37: MHI.....	67
Figure 38: Seed points used to find the extreme points of the MH area	67
Figure 39: Seed points of extreme areas in (b) and (c) to obtain the minimum diameter ..	68
Figure 40: Measures of the MH in (a) background and (b) original image.....	68
Figure 41: Examples of discarded images	70
Figure 42: Prediction (wider line) and GT (thinnest line) contours on the same background	71
Figure 43: Boxplots of mean and SD of UE (pixel) and medium processing time per image (s) for the tests of ILM boundary layer segmentation with MH pathology.....	72
Figure 44: MH areas for (a) prediction and (b) GT	73
Figure 45: Confusion matrix of MH area segmentation for (a) related areas and (b) unrelated areas	73
Figure 46: Boxplots of accuracy, IoU, DSC, MCC (%) and medium processing time per image (s) for the evaluation of MH area segmentation	74

Figure 47: Bland-Altman graphs for the mean and SD of signed error of MH measurements (μm). Top left (top diameter), top right (bottom diameter), bottom left (height), bottom right (minimum diameter)	76
Figure 48: Confusion matrix of MHI	77
Figure 49: ILM segmentation for an NM (a) original image and (b) background	78
Figure 50: Boxplots of mean and standard deviation (pixel) and processing time (s) for the evaluations of ILM boundary layer segmentation with NM	78
Figure 51: Contour segmentation (left) and confusion matrix of pixels (right) show a discrepancy between the prediction (red) and ground-truth (yellow)	83
Figure 52: Macular Edema examples. MH29 and MH37, respectively	104
Figure 53: Pseudo MH examples. From left to right, top to bottom: MH15, MH28, MH56, MH61, MH90 and MH100	105
Figure 54: Lamellar MH examples. From left to right, top to bottom: MH1, MH22, MH65, MH79, MH94, MH95, MH96, MH97 and MH101	106
Figure 55: Epiretinal Membrane example, MH92.....	107
Figure 56: Vitreomacular Traction Syndrome examples. From left to right, top to bottom: MH4, MH10, MH25 and MH67.....	108
Figure 57: MH with Retinal Detachment, MH11	108
Figure 58: Normal Macula examples. From left to right, top to bottom: MH21, MH26 and MH82.....	109
Figure 59: A lamellar macular hole plus a macular edema. From left to right: MH17 and MH38.....	110
Figure 60: An epiretinal membrane plus a macular pseudo hole. From left to right, top to bottom: MH30, MH47, MH57, MH84, MH85 and MH93	110
Figure 61: A macular edema, a lamellar macular hole and an epiretinal membrane. MH35	111
Figure 62: A macular edema plus a macular pseudo hole. MH 36.....	111
Figure 63: An epiretinal membrane plus a macular edema. MH91.....	111

LIST OF TABLES

Table 1: Confusion matrix for MH area segmentation	45
Table 2: Comparison between papers	53
Table 3: Papers directly linked to segmentation of MH volume/area or contour	54
Table 4: Brightness level adjustment.....	58
Table 5: Mean and SD of UE (pixel) and medium processing time per image (s) of ILM boundary layer segmentation with MH pathology	72
Table 6: Results of evaluation metrics in percentage for MH area segmentation	75
Table 7: Mean and SD of UE (μm) for measurements of MH	75
Table 8: Evaluation metric results for MHI classification in percentage	77
Table 9: Mean and SD of UE (pixel) and medium processing time per image (s) for ILM boundary layer segmentation with NM	79
Table 10: Mean and SD of UE comparison (pixel) and medium processing time per image (s) of ILM boundary layer segmentation with MH pathology	79
Table 11: IoU, DSC and MCC comparison in percentage of MH area segmentation.....	81
Table 12: Mean and SD of UE (pixel) for ILM boundary layer segmentation with NM...	82
Table 13: Individual results for each OCT image with an MH pathology	112
Table 14: Measurements of MH ground-truth (μm)	114
Table 15: Measurements of MH prediction (μm)	116
Table 16: Unsigned error of MH measurements (μm).....	118
Table 17: Individual results for each OCT image with an NM	120

LIST OF ACRONYMS

AMD:	Age-related Macular Degeneration
BMO:	Bruch's Membrane Opening
CAD:	Computer-Aided Diagnosis
CNN:	Convolutional Neural Network
CNV:	Choroidal Neovascularization
CSR:	Central Serous Retinopathy
CV:	Computer Vision
DME:	Diabetic Macular Edema
DR:	Diabetic Retinopathy
DSC:	Dice Similarity Coefficient
EFB:	Set of techniques: Euclidean Distance; Flood Fill; Border Following
ERM:	Epiretinal Membrane
FH:	Full-thickness Holes
FN:	False Negative
FP:	False Positive
GDM:	Geodesic Distance Method
GT:	Ground Truth
IDE:	Integrated development environment
IFCE:	<i>Instituto Federal de Educação, Ciência e Tecnologia do Ceará</i>
ILM:	Internal Limiting Membrane
IoU:	Intersection over Union
LCI:	Low time-Coherence Interferometry
LEM:	<i>Laboratório de Ensaios Mecânicos</i>
LMH:	Lamellar Macular Hole
LSTM:	Long Short Term Memory
MCC:	Matthews Correlation Coefficient
ME:	Macular Edema
MH:	Macular Hole
MHI:	Macular Hole Index
MPH:	Macular Pseudo Hole
NM:	Normal Macular
MPT:	Medium Processing Time

OCT:	Optical Coherence Tomography
PC:	Pearson Correlation
PH:	Pseudo Holes
PPGET:	<i>Programa de Pós-Graduação em Engenharia de Telecomunicações</i>
PVD:	Posterior Vitreous Detachment
RD:	Retinal Detachment
RP:	Retinitis Pigmentosa
RPE:	Retinal Pigment Epithelium
SD:	Standard Deviation
SD-OCT:	Spectral Domain-Optical Coherence Tomography
SE:	Signed Error
SS-OCTA:	Swept-Source-OCT Angiography
TD-OCT:	Time Domain-Optical Coherence Tomography
TN:	True Negative
TP:	True Positive
UE:	Unsigned Error
VMT:	Vitreomacular Traction

CONTENTS

1	INTRODUCTION	18
1.1	Motivation.....	22
1.2	Justification	23
1.3	Objectives	24
1.3.1	Specific objectives	24
1.4	Scientific Production.....	24
1.5	Text Organization	25
2	THEORETICAL BACKGROUND	26
2.1	Optical Coherence Tomography	26
2.2	Digital image.....	28
2.3	Sampling and Quantization.....	29
2.4	Image preprocessing	29
2.4.1	Spatial domain operation	30
2.4.2	Frequency domain operation.....	30
2.4.3	Noise removal	31
2.4.4	Contrast Enhancement	32
2.5	Image segmentation	33
2.5.1	Thresholding-Based segmentation.....	35
2.5.2	Region-Based segmentation.....	37
2.5.3	Edge-Based segmentation	38
2.5.4	Gradient-Based segmentation	39
2.5.5	Classification-Based segmentation	40
2.6	Flood Fill algorithm	41
2.7	Border Following algorithm	42
2.8	Evaluation metrics	43
2.8.1	Distance-Based segmentation evaluation.....	44

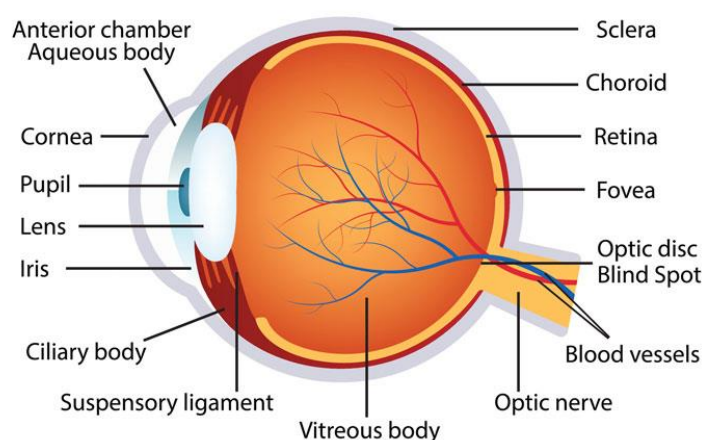
2.8.2	Area-Based segmentation evaluation.....	45
2.9	Final considerations of the chapter	46
3	STATE OF THE ART.....	48
3.1	Review of MH segmentation in OCT images.....	48
3.2	Review of other segmentations in OCT images	50
3.3	Final considerations of the chapter	52
4	METHODOLOGY	55
4.1	Integrated development environment	56
4.2	Image database.....	56
4.3	Ground-Truth	57
4.4	Brightness adjustment normalization.....	57
4.5	Gaussian Blur Filter	59
4.6	Thresholding	59
4.7	Erosion and Dilation	62
4.8	Flood Fill algorithm	63
4.9	ILM boundary layer segmentation with MH	64
4.10	Area-based segmentation of MH.....	65
4.11	Measurements of the macular hole.....	66
4.12	Final considerations of the chapter.....	68
5	RESULTS AND DISCUSSIONS	70
5.1	Results for ILM boundary layer segmentation with MH pathology.....	70
5.2	Results for MH area segmentation.....	72
5.3	Results for measurements of macular holes.....	75
5.4	Results for ILM boundary layer segmentation without pathology	77
5.5	Discussion of ILM boundary layer segmentation with MH pathology	79
5.6	Discussions of MH area segmentation.....	80
5.7	Discussions of measurements of macular holes	81

5.8	Discussions of ILM boundary layer segmentation without any pathology	81
5.9	Limitations of the proposed method	82
6	CONCLUSIONS	84
6.1	Contributions	85
6.2	Future Prospects.....	85
	REFERENCES	87
	Appendix A – Algorithm for the proposed method.....	102
	Appendix B – MH images excluded from the work.....	104
	Appendix C - Individual results for each OCT image with MH pathology	112
	Appendix D - Measurements of MH ground-truth (μm).....	114
	Appendix E - Measurements of MH prediction (μm)	116
	Appendix F - Unsigned error of MH measurements (μm)	118
	Appendix G - Individual results for each OCT image with NM	120

1 INTRODUCTION

The human eye has a diameter varying from 2.0 to 2.5 cm (FORSYTH; PONCE, 2002). The reflected light from objects enters the eye through the pupil, a frontal opening in the iris. Then it passes through the lens until it reaches the retina, which is the inner posterior layer of the eye (GONZALEZ; WOODS; EDDINS, 2004). A drawing of the human eye is shown in detail in Figure 1.

Figure 1: Eye anatomy



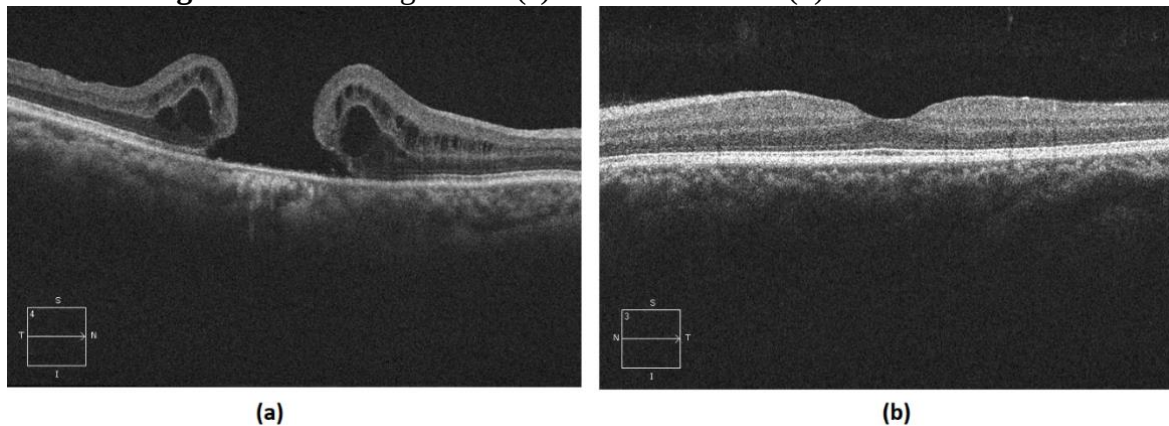
Source: Eye (2020).

The retina is the part of the ocular system responsible for vision. It captures the images and sends them to the brain through electrical impulses along the optic nerve. In the central region of the retina is the macula that is responsible for sharp vision (HASSAN *et al.*, 2016). Some macular pathologies are Central Serous Retinopathy (CSR), Age-related Macular Degeneration (AMD) and Macular Edema (ME) (TRABELSI; ZGHAL; SLOKOM, 2016; HOOD *et al.*, 2007; VAN DIJK *et al.*, 2010). Another less commonly encountered macular disease is called Macular Hole (MH) (ZHANG *et al.*, 2015) that can be seen in Figure 2.

A macular hole is a condition of low vision related to the weakening of the fovea, high myopia, eye trauma and severe exposure to the sun, in which a hole alters the natural structure of the retina (FAGHILI *et al.*, 2014). Age is the most common cause of MH. With advancing age, the vitreous begins to shrink and pull away from the retina. Usually the vitreous pulls away with no problems. However, sometimes the vitreous can stick to the

retina. This causes the macula to stretch and a hole to form. A MH can also form when the macula swells due to other eye diseases, or it can be caused by an eye injury (WHAT, 2019).

Figure 2: OCT images with (a) macular hole and (b) normal macular



Source: Adapted from OCTID (2018).

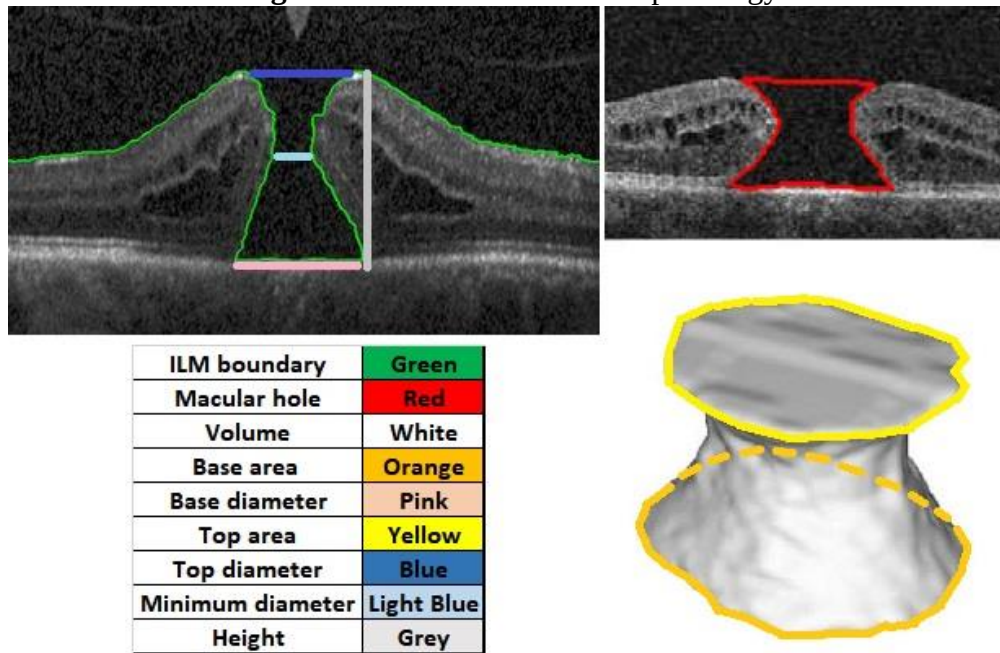
The MH pathology affects about 1 in 500 people over 40 years (THAPA *et al.*, 2013), and for those over 55 years the number rises to 1.65 per 500 people (LUCKIE; HERIOT, 1995). Today people live longer due to the advances made in medical and consequently there has been an increase in the number of retinal diseases (KLEIN; KLEIN; LINTON, 1992; VELEZ-MONTOYA *et al.*, 2014).

If a patient has a macula hole in only one eye, he/she will not usually complain until a significant decrease in vision occurs. However, if the presence of the hole is bilateral, the complaints will come sooner. Among the symptoms commonly reported are loss of distant vision, difficulty in reading and gray spot in the central region of vision. Furthermore, some people report image distortion (BURACO, 2020).

One of the main treatments is the introduction of a special gas into the eye, in order to relieve the traction forces that are acting on the macula with the aim to close the MH. The sooner the operation is performed the greater the recovery of vision. The gas helps to close the MH; however, the patient should remain in a looking down position for 3-5 days. An individual who undergoes this operation is prohibited to travel by plane or to move to higher altitudes, as the gas can expand and increase the pressure of the eye causing serious complications including blindness. As the MH closes the vision of the patient tends to improve, and there is a chance that it will return to normal; however, this largely depends on how soon the surgery is performed after the discovery of the disease. The complications inherent to this surgery are cataracts for those who have not yet operated them, infection, retinal detachment and bleeding (BURACO, 2020).

The treatment depends on the size and shape of the MH. Some important metrics are volume, areas, diameters and height. The measurement of these metrics are important to assess if it is possible to close the hole (CHHABLANI *et al.*, 2015; MADI; MASRI; STEEL, 2016; STEEL *et al.*, 2016). The identification of MH in OCT images is possible through the top boundaries of the Internal Limiting Membrane (ILM) and the Retinal Pigment Epithelium (RPE) (PHILIPPAKIS *et al.*, 2018; FABRITIUS *et al.*, 2009; DE MOURA *et al.*, 2018). Some important metrics of this pathology are shown in Figure 3.

Figure 3: Some metrics of MH pathology

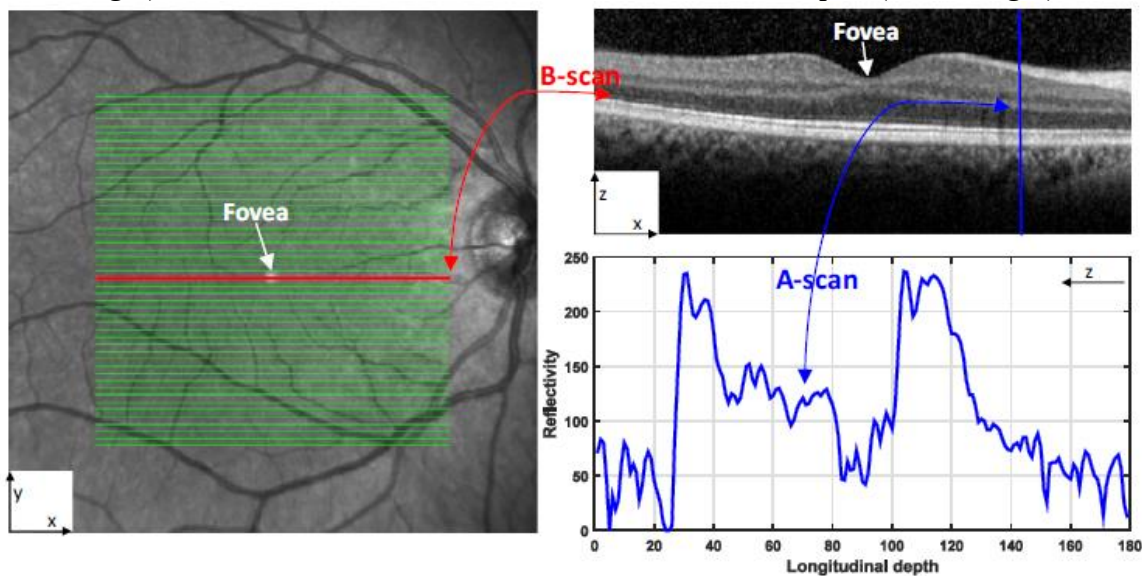


Source: Adapted from Keller *et al.* (2016); Xu *et al.* (2013).

A MH can be a Full-thickness Hole (FH), a Pseudo Hole (PH) (ALTAWHEEL; IP, 2003) and a Lamellar Macular Hole (LMH). A LMH is a vitreoretinal disorder characterized by an irregular foveal contour, a break in the inner fovea, dehiscence of the inner foveal retina from the outer retina, and the absence of a full-thickness foveal defect with intact foveal photoreceptors. The pathogenesis is only partially understood (FRISINA; PILOTTO; MIDENA, 2019), and there is a different form of treatment for each situation. The size of the MH is the major risk factor for complications in surgery (IP *et al.*, 2002), and it determines whether the approach will be just a treatment with enzymatic vitreolysis or surgery (TADAYONI *et al.*, 2012; HALLER *et al.*, 2015). Surgery has achieved good results in restoring patient's visual acuity (FREEMAN *et al.*, 1997; PARK *et al.*, 1999; HARITOGLOU *et al.*, 2001).

Optical Coherence Tomography (OCT) is a way of mapping and determining the shape of retinal sections without physical contact and has become a powerful tool to diagnose pathologies (SUI *et al.*, 2017; GONG *et al.*, 2015; TANG; ZHENG; CAO, 2017). This tool produces high quality retinal images. Although, it was first introduced in 1991 (HUANG *et al.*, 1991; COSCAS; CUNHA-VAZ; SOUBRANE, 2010; TRANOS *et al.*, 2004), it only appeared commercially in 1996 (SCHUMAN *et al.*, 1996). Now it is standard practice for medical retinal analysis (SHUMAN; JOEL, 2004). Various A-scans (1D) build a B-scan (2D) image (DREXLER *et al.*, 2003), and an aggregation of B-scans constructs a 3D structure (RICCO *et al.*, 2009; XU *et al.*, 2010; HU; CHEN; YI, 2019). These structures are shown in Figure 4.

Figure 4: 2D view of a 3D SD-OCT image of the retina (left) and a B-scan image (top right). One vertical A-scan of the B-scan is shown in the plot (bottom right)



Source: Hassan *et al.* (2016).

OCT images may have speckle noise (SCHMITT; XIANG; YUNG, 1999; GRZYWACZ *et al.*, 2010; LINDENMAIER *et al.*, 2013), which may hinder the segmentation and compromise the results (DUAN *et al.*, 2017; CHEN *et al.*, 2019; LV *et al.*, 2018). Shadows caused by retinal blood vessels and some pathologies on retinal structures challenge the segmentation even more, and in this case problems related to defining the contour incorrectly are more likely to occur (SADDA *et al.*, 2006; KREBS *et al.*, 2008).

There are two ways to remove stains from OCT images. The first is a parallel method that preserves the resolution, improves contrast, but changes the physical structure (HUGHES; SPRING; PODOLEANU, 2010; DESJARDINS *et al.*, 2006; IFTIMIA; BOUMA; TEARNEY, 2003); and the second way is a serial method that preserves the

physical structure, but is more time consuming (JØRGENSEN *et al.*, 2007; SZKULMOWSKI *et al.*, 2012).

Automatic segmentation and diagnosis of pathologies using OCT is not an easy task. More than one pathology may affect the results negatively. The same pathology may have different characteristics, and MH is one example. The guaranty of an identical comparison between regions is not possible in some studies using more than one dataset from different devices (HO *et al.*, 2009; MYLONAS *et al.*, 2009).

This field of study is directly linked to computer vision (CV), which ensures visual information for a given application. CV is a branch of artificial intelligence and automated systems can acquire information and perform tasks (MENDES; BORILLE, 2019). This technology has made significant advances in several areas: medicine, industry, science, and military among others (ANDREOPOULOS; TSOTSOS, 2013). CV obtains information through image analysis. In order to process an image, it must first be converted into a digital image (pixel coordinates), which may vary in grayscale or be a mix of primary colors (ROSENFELD, 1988). There are some levels of vision analysis: low-level vision, gathering and processing of images; intermediate-level vision, segmentation and classification; and high-level vision, artificial intelligence that learns tasks automatically and generates results (DE SOUZA ALVES *et al.*, 2018).

Countless preprocessing techniques can be applied to images, and the same can be said for segmentation. Using medical images to make diagnoses has become both popular and reliable. Millions of images are produced by hospitals each year. They come from different sources like magnetic, tomography, or ultrasound. These images have features that allow specialists to diagnose pathologies (AYACHE, 1995).

1.1 Motivation

Although manual segmentation of OCT devices is carried out by specialists (SALARIAN *et al.*, 2015), this technique is time consuming which makes it disadvantageous for studies of segmentation and classification (HU *et al.*, 2019; GIRISH; KOTHARI; RAJAN, 2016; WANG; WANG; LI, 2017; FERNÁNDEZ; SALINAS; PULIAFITO, 2005; VERMEER *et al.*, 2011). On the other hand, manual segmentation is still the major segmentation technique for OCT images due to the lack of reliable automated methods (GEORGE; JIJI, 2019). Thus,

a fast and low computational cost segmentation technique would be of great interest to help specialists to analyze retinal layers.

Despite the challenges encountered in segmentation techniques for MH pathologies, the motivation of this work is strongly based on the importance of automated methods to segment OCT images in order to help medical diagnosis. Furthermore, the advances in research involving this area can contribute to the acceptance of this technology in medical practices. This work may be a source of study for researchers, clinicians and engineers who want to delve into MH segmentation techniques and bring about improvements in retinal image analysis (DE MUL; DE BONT; BERG, 2007; MARRUGO *et al.*, 2012). Different parameters acquired by OCT images contribute to patient diagnoses as well as to academic researchers (VERCELLIN *et al.*, 2018; MONTUORO *et al.*, 2017). Computer-aided diagnosis (CAD) systems can help health professionals in their conclusions, since CAD systems provide information beyond the images (PETRICK *et al.*, 2013; RAMZAN *et al.*, 2018; GERENDAS *et al.*, 2018).

1.2 Justification

The importance of understanding the clinical case and all its variables is fundamental for the development of this work. Health and technology are areas that complement each other. Health provides the necessary elements for the methodological basis of new technologies. While technology benefits health with improved and robust equipment and procedures that improve the quality of life for society as a whole.

Early diagnoses are essential for treatment to go ahead without the need for surgery. However, the evolution of the disease is asymptomatic, and usually the patient only begins to complain when the problem is at an advanced stage. Unfortunately, a late diagnosis can lead to blindness (MENDES *et al.*, 2019).

Most research involving retinal problems is concerned with the more common pathologies, such as CRS, AMD, and ME, leaving the MH problem aside, despite the fact that, MH affects about 1 in 500 people over 40 years old (THAPA *et al.*, 2013), and for those over 55 years the rate is 1.65 per 500 people (LUCKIE; HERIOT, 1995).

One reason to carry out this work at IFCE is the fact that this institute produces relevant works in the field of biomedical engineering and there are qualified professionals to provide all the necessary support. Some examples are: de Moraes *et al.* (2020) developed

a computational process which relies on the evaluation of heart rate variability as a prognostic marker in the stratification of clinical heart conditions; Sousa (2020) developed a system to simulate the medical examination procedures in the nasal cavity for training and study, using the 3D human anatomy atlas and computed tomography as a reference; de Alexandria *et al.* (2014) created a pSnakes method that was effective to segment images of the left ventricle; and da Silva Felix *et al.* (2012) developed a new method of automatic initialization for active contours, which is a method applied to tomography images of the lungs of healthy volunteers and ill patients.

However, the main justification for this work is the need to improve the segmentation of the MH pathology. This work can help physicians in making diagnostic decisions (MENDES *et al.*, 2019). MH measurements are fundamental to decide if the case requires only clinical treatment or if surgery is necessary.

1.3 Objectives

The main objective of this work is the development of an algorithm that automatically segments the ILM boundary layer and the area of a MH pathology in OCT images.

1.3.1 Specific objectives

- The use of image preprocessing in order to remove noise and enhance quality;
- Apply distance-based segmentation and area-based segmentation methods;
- Evaluate the algorithm through distance-based segmentation and area-based segmentation evaluation metrics;
- Automatic acquisition of MH measurements;
- Segment the ILM boundary layer in images without MH and its validation, so that the algorithm can also be used in normal cases.

1.4 Scientific Production

During the development of this work, the following scientific research was published:

Mendes, O. L. C., Lucena, A. R., Lucena, D. R., Cavalcante, T. S. and Alexandria, A. R. (2019) Automatic Segmentation of Macular Holes in Optical Coherence Tomography Images: A review. *Journal of Artificial Intelligence and Systems*, 1, 163185. <https://doi.org/10.33969/AIS.2019.11010>.

1.5 Text Organization

The work is divided into six sections. Section 2 describes the theoretical background. Section 3 introduces the state of the art methodologies. Section 4 details the proposed methodology. Section 5 discusses the results and compares them with the state-of-the-art methods. Lastly, Section 6, provides the conclusion, summaries the results of the proposed method and presents some future prospects.

2 THEORETICAL BACKGROUND

The application of digital imaging in the field of medicine is consolidated. However, medical images can be affected by noise, blurriness and may suffer due to poor contrast and sharpness, which may result in a false diagnosis. Suitable algorithms for image processing are necessary in order to obtain better results. Computer vision techniques are important for this and are fundamental to help physicians make better decisions.

This Section provides the theoretical background necessary to follow the methodology clearly. The texts contain brief but important information and they are based on classic references from different technical areas. The principles described here are applicable to a wide range of CV tasks, but the scope of this work is focused on the preprocessing and segmentation of OCT images.

2.1 Optical Coherence Tomography

Tomographic techniques generate slice images of 3D objects. Optical tomographic techniques are important because they can provide non-invasive diagnostic images. There are two fundamental optical tomography techniques: diffuse optical tomography (DOT), and optical diffraction tomography (ODT). OCT is based on ODT that uses single scattered light and produces tomographic images by the Fourier diffraction projection theorem (BORN; WOLF, 1999). Standard diffraction tomographic methods can also be used for imaging with diffuse-photon density waves (LI *et al.*, 1997).

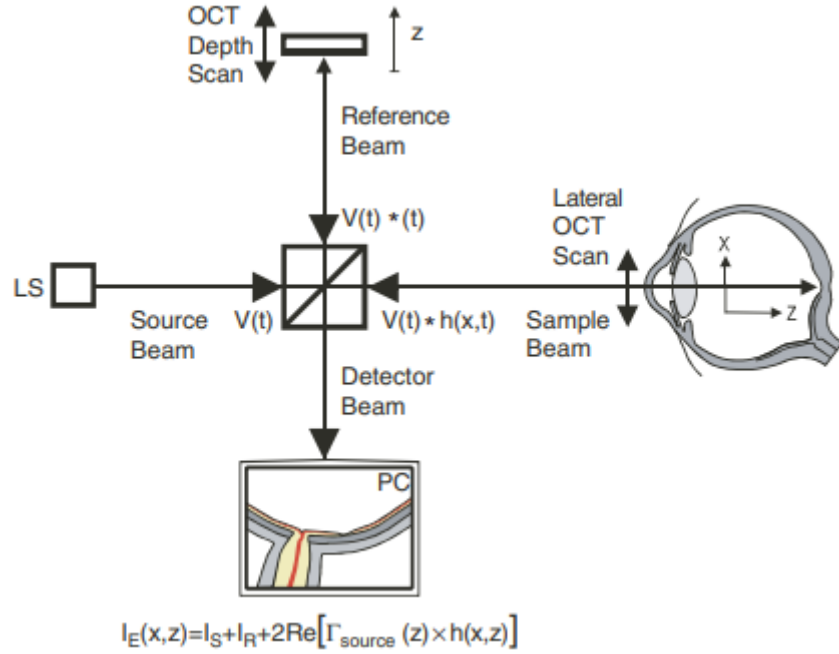
OCT uses ballistic and near-ballistic photons. Laterally adjacent depth-scans are used to obtain a 2D map of reflection sites in a sample. Figure 5 shows a standard OCT plan. A low time-coherence light source is used in a standard Michelson interferometer. OCT synthesizes cross-sectional images from a series of adjacent low time-coherence interferometry (LCI) depth-scans that measure absolute distances. LCI is based on the occurrence of fringes if the optical path lengths of reference and sample beams coincide within the ‘coherence gate’, which is of the size of the round trip coherence length l_c :

$$l_c = \frac{2 \ln 2}{\pi} \frac{\bar{\lambda}^2}{\Delta \lambda}, \quad (1)$$

where $\bar{\lambda}$ is the mean wavelength and λ the spectral width (FERCHER *et al.*, 2003).

Figure 5: Standard OCT plan based on a low time-coherence Michelson interferometer.

The intensity I_E at the interferometer exit depends on the sample response $h(x, z)$ convolved with the source coherence function $r_{source}(z)$. *LS* = low time-coherence light source; *PC* = personal computer



Source: Fercher *et al.* (2003).

In the Time Domain-OCT (TD-OCT) depth information of the retina is obtained after a longitudinal translation in time of a reference arm. Spectral Domain-OCT (SD-OCT) measures the interferometric signal detected as a function of optical frequencies, allowing for imaging speeds 50 times faster than TD-OCT, and providing a greater number of images per unit area (KOIZUMI *et al.*, 2008). This increase in resolution and scanning speed allows high density raster scanning of retinal tissue while minimizing eye motion artefacts (FORTE *et al.*, 2009). The Swept-Source-OCT Angiography (SS-OCTA) provides treated and untreated images. Angiography technology demonstrates unprecedented detail of the microvasculature (SCHAAL *et al.*, 2017). Cirrus HD-OCT, 3D-OCT 100 and 2000, RS-3000, DRI OCT-1 and Spectralis OCT are examples of SD-OCT devices; Stratus OCT is an example of TD-OCT (WALDSTEIN *et al.*, 2015; XU *et al.*, 2019); and PLEX Elite 9000 is an example of SS-OCTA (SCHAAL *et al.*, 2017).

The images of OCT devices are easier to segment than those from ultrasound or magnetic-resonance imaging (UNIVERSITY, 2019), as shown by Alexandria *et al.* (2011) who demonstrated the difficulty of segmenting ultrasound image. These OCT images are necessary before any decision is made to perform surgery or not (TAKAHASHI;

NAGAOKA; YOSHIDA, 2016; GUPTA; GUPTA; DOGRA, 2004; KONG *et al.*, 2018) because they provide the information necessary to analyze retinal diseases (LU *et al.*, 2017; JOUSSEN *et al.*, 2007). The advantages of OCT are: fast image acquisition (POTSAID *et al.*, 2008), excellent sensitivity to very low light (WOJTKOWSKI; MACIEJ, 2010) and high resolution for millimeter structures (SAWADZKI *et al.*, 2008).

2.2 Digital image

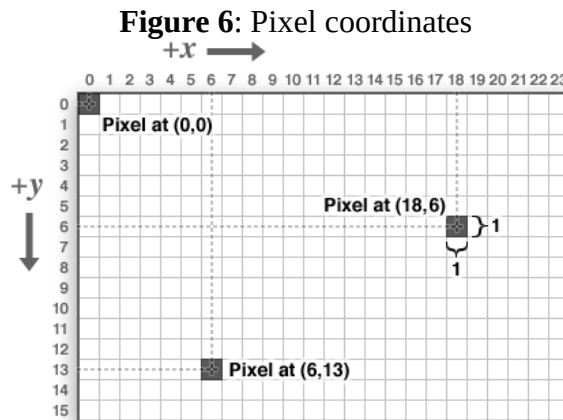
The images seen by human eye can be compared to continuous functions as they can not be separated in parts. Computers are not capable of processing continuous images, so the image is represented as two-dimensional arrays of pixels, making it a digital image. The luminous intensity can be decomposed according to:

$$f(x, y) = i(x, y).r(x, y), \quad (2)$$

where i is the lighting component and r is the reflectance component. These arrangements are functions (x, y) , where x and y are the spatial coordinates and f is the luminous intensity of the pixel under consideration (RENCZ; RYERSON, 1999; RUSS, 1997). An image with three primary sources of color (red, green and blue), called RGB, can be represented by: (GONZALEZ; WOODS, 2002)

$$f(x, y) = f_R(x, y) + f_G(x, y) + f_B(x, y). \quad (3)$$

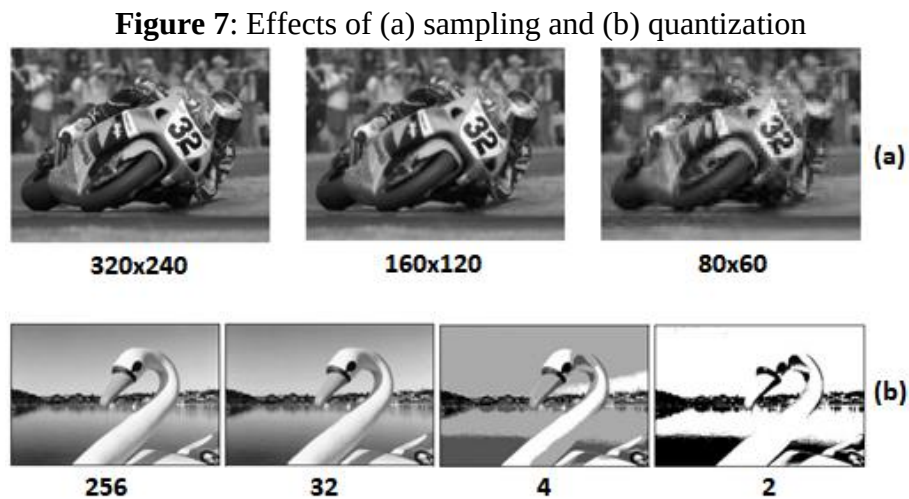
The location of a pixel in a 2D image is represented in Figure 6. The y coordinate pointing down and x to the right represent the convention used in image scanning.



Source: Coordinate (2020).

2.3 Sampling and Quantization

Sampling and quantization are characteristics responsible for image quality. The sampling rate is responsible for the spatial resolution of the digitized image, while the quantization level determines the number of gray levels in the digitized image. The magnitude of a sampled image is expressed as a digital value in image processing. The conversion between continuous values of the image function and its digital equivalent is called quantization. The number of quantization levels should be high enough for human perception of fine shading in the image. The occurrence of false contours is the main problem in images quantized with insufficient levels of brightness. The effects of sampling and quantization are shown in Figure 7.



Source: Adapted from Jacques Junior (2020).

2.4 Image preprocessing

Before starting the preprocessing techniques, an image must be acquired. Such images are obtained through a device (usually a camera) that captures the reflected light on the surface of an object. After being captured by a digitization process, the image needs to be represented in an appropriate way for computational treatment. Images can be represented in two or more dimensions. After that, preprocessing techniques can be applied through an algorithm.

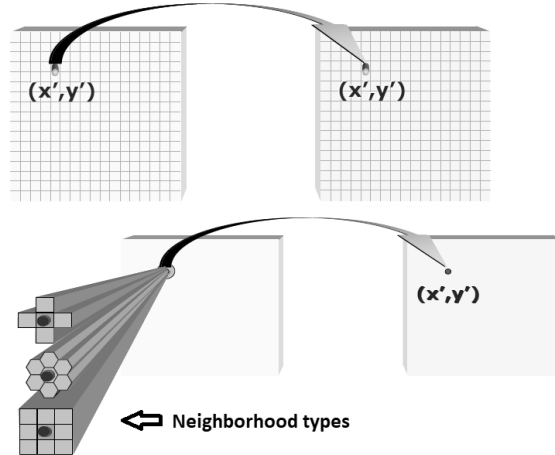
The purpose of preprocessing is to improve the image quality, correcting defects and highlighting important details for the desired application. This step is important to provide satisfactory results in segmentation. Various preprocessing techniques can be used, such as:

brightness and contrast adjustment, noise reduction, correction of irregular lighting, edge enhancement, among others (HASSAN *et al.*, 2016; TRABELSI; ZGHAL; SLOKOM, 2016).

2.4.1 Spatial domain operation

In the spatial domain, the pixels are manipulated. This operation can be classified into point (point-to-point) and local. In the point operations, each output pixel depends only on its corresponding input pixel. It includes gray-level transformation and histogram equalization. In local operations, the value of an output pixel depends on its corresponding input pixel and its neighborhood. It includes image smoothing and image sharpening (HASSAN *et al.*, 2016). Examples of point and local operations are represented in Figure 8.

Figure 8: Point and local operations



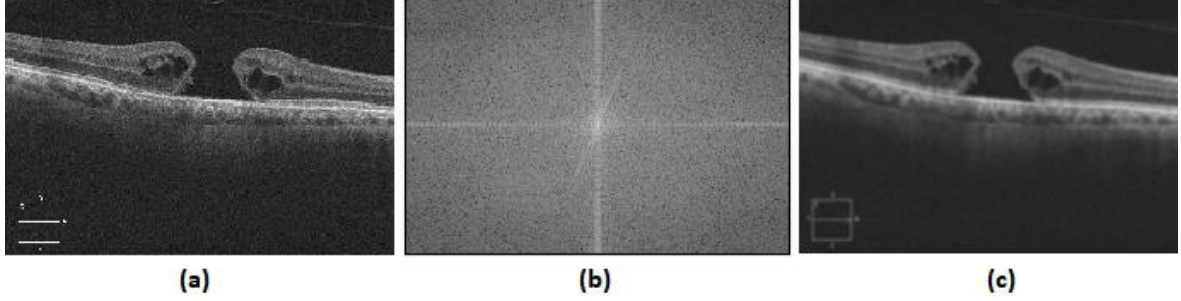
Source: Adapted from de Queiroz and Gomes (2006).

2.4.2 Frequency domain operation

In frequency domain, the Fourier Transform of an image is performed. The result is multiplied by a filter, and then the Inverse Fourier Transform is taken to obtain the original enhanced image. This process includes low-pass and high-pass filters and homomorphic filtering. In low-pass filtering, high frequency components are eliminated and low frequency components are retained. It can remove the sharp transitions associated with noise and preserve the information. In high-pass filtering, the inverse occurs. Homomorphic filters are used to extract illumination and reflectance components from the image (VASUKI;

KANIMOZHI; DEVI, 2017). An example of a frequency domain operation on an OCT image is shown in Figure 9.

Figure 9: Frequency domain operation on an OCT image. (a) Input image, (b) magnitude spectrum and (c) output filtered image



Source: Adapted from OCTID (2018).

2.4.3 Noise removal

Images captured by a device are subject to different types of noise. Depending on the application, the noise must be removed after image digitalization. This process does not increase the inbuilt information content of the image, but can highlight the features of interest for the next step, the segmentation.

2.4.3.1 Linear Filter

A Linear Filter is a basic method to obtain the average pixel intensity using neighbor pixel intensity values. The filtering processing is described by:

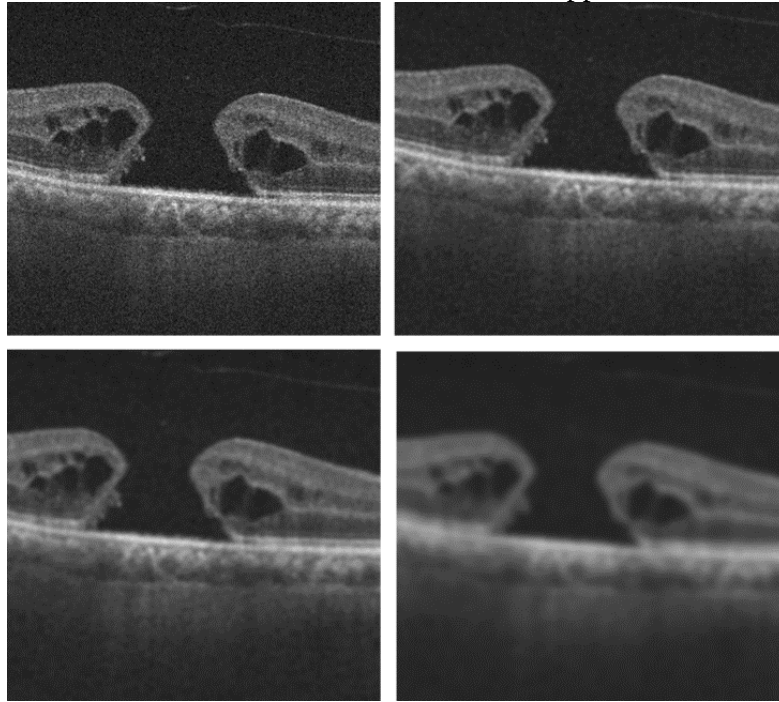
$$f'(x, y) = \sum_{i=-M}^{+M} \sum_{j=-N}^{+N} w_{i,j} f(x + i, y + j), \quad (4)$$

where $f(x, y)$ is the pixel intensity of the x and y coordinates, $M \times N$ is the size of the image and w is the weight of the filter. The values of $M \times N$ and w are chosen depending on the degree of filtration required. The filter size is a trade-off between noise reduction and loss of image detail.

Some examples of linear filters are: Gaussian Filter, High and Low Pass Filter and Wiener Filter. The Gaussian Filter can remove Gaussian noise and it performs well in edge detection (HUSSEIN *et al.*, 2011). Examples of Gaussian Filter applied to an OCT image with different kernels (cut-off frequencies) are shown in Figure 10, where the top left image

is the original. The High/Low Pass Filter lets high/low frequency pass and attenuates the components with frequencies lower/higher than the cut-off frequency (GRYCUK *et al.*, 2014). The Wiener Filter is an optimal linear filter minimizing the overall mean square error during the process of inverse filtering and noise smoothing. It can be used to reduce Gaussian noise (VASAVADA; TIWARI, 2014).

Figure 10: Gaussian Filter with different kernels applied to an OCT image



Source: Adapted from OCTID (2018).

A Standard Median Filter is applied to each pixel without considering the contamination status of an image. Thus, corrupted pixels are filtered, resulting in the loss of some image information. A Hybrid Median Filter can be applied for easy removal of impulse noise without losing the edges. It preserves the edges better than a Median Filter. In the Adaptive Median Filter, corrupted and uncorrupted pixels are discriminated, with noisy pixels replaced by their median value and uncorrupted pixels left unchanged. The filter performs better with low noise densities. Better noise removal is also obtained at higher noise densities after increasing the window size (HE; ZHENG; SUN, 2016).

2.4.4 Contrast Enhancement

Contrast is the difference in luminance that makes an object distinguishable in an image. Contrast enhancing aims to increase the difference of the intensity values among

pixels so that they can be easily distinguished by human vision or computer vision. Most contrast enhancing techniques use the image histogram, which is a plot showing the occurrence of intensity values in images (JAIN, 1989).

2.4.4.1 Histogram equalization

A Histogram is a graphical representation of the number of pixels for each shade of gray or color present in an image. Brightness and contrast can be adjusted through histogram equalization. Clear/Dark images have histograms with high concentrations of high/low brightness pixels. The variation in contrast is associated with the degree of dispersion of the histogram. Some point operations use the histogram as a decision parameter to provide results for the pixels of the processed image. Histogram equalization can be described by:

$$j' = \sum_{i=l}^j H(i) / \sum_{i=l}^L H(i), \quad (5)$$

where H represents the original histogram. The minimum and maximum intensity are l and L , respectively. The intensity value in the histogram is i , while j and j' stand for the intensity value in the original histogram and its corresponding intensity value in the equalized histogram, respectively (HE; ZHENG; SUN, 2016). The histogram equalization applied in an OCT image can be seen in Figure 11.

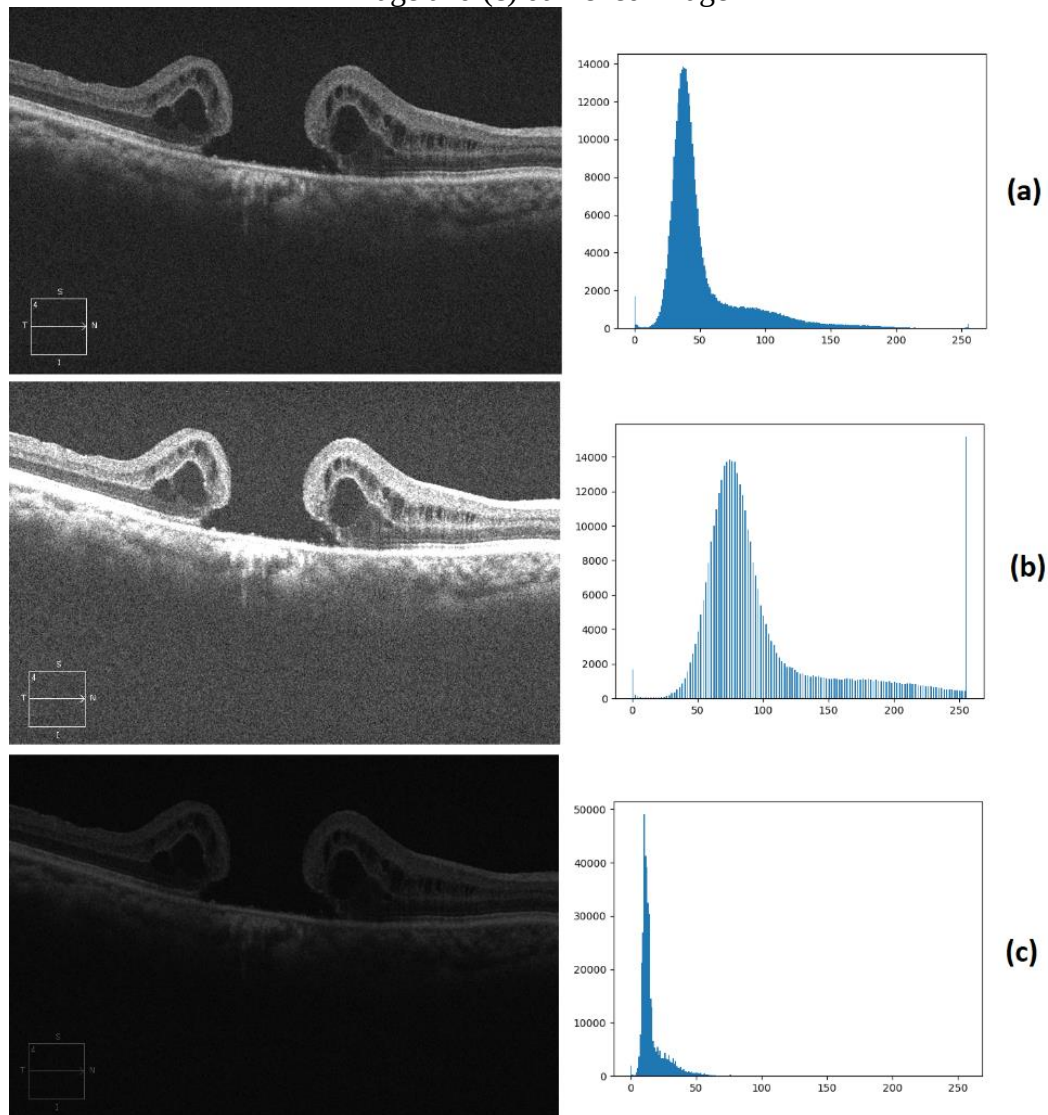
2.5 Image segmentation

Segmentation is the process of separating one or more objects of interest from the rest of the image. This rest of the image is then normally called background. A general segmentation technique is defined by:

$$\bigcup_{i=1}^n S_i = F \quad \text{with} \quad S_i \cap S_j = \emptyset, \quad i \neq j, \quad (6)$$

where F is the set of all the pixels and $P()$ is defined as a group of connected pixels; then the segmentation is a partitioning of the set F into a set of connected subsets or regions ($S_1, S_2, \dots, S_n$). The uniformity predicate $P(S_i) = \text{true}$ for all regions (S_i) and $P(S_i \cup S_j) = \text{false}$, when S_i is adjacent to S_j (HOROWITZ, 1974).

Figure 11: Example of histogram equalization. (a) Original OCT image, (b) whitened image and (c) darkened image



Source: Adapted from OCTID (2018).

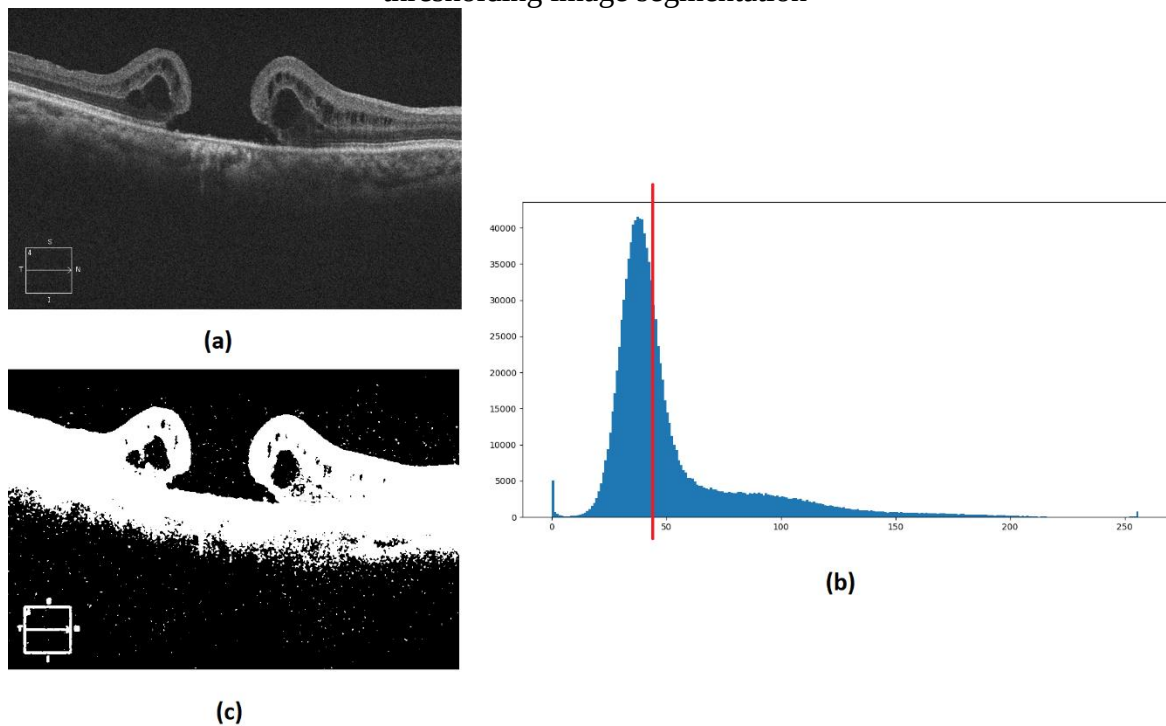
There are many types of image segmentation techniques, but there is no single method which can be considered good for all images, nor are all methods equally good for a particular type of image. Moreover, a technique for a certain class of image may not be applied to another class of image. The literature on color image segmentation is limited compared with gray tone images (PAL; PAL, 1993).

There are many applications for segmentation, from the detection of a MH in an OCT image to the identification of an airport from remote sensing data. Image segmentation techniques are classified into five main categories: Thresholding-Based segmentation, Region-Based segmentation, Edge-Based segmentation, Gradient-Based segmentation and Classification-Based segmentation (HE; ZHENG; SUN, 2016).

2.5.1 Thresholding-Based segmentation

In thresholding-based segmentation, the image histogram is partitioned into two classes using a single value or multiple classes using multiple values based on its characteristics. For the case of a single value, a constant T is defined within of the shades of the gray range of the image as the cutoff point. If the pixel intensity is less than T ($I_{i,j} < T$), it becomes the background/object. If the pixel intensity is greater than T ($I_{i,j} > T$), it becomes the object/background. Threshold techniques are based on an image-space region. The threshold method applied to an OCT image can be seen in Figure 12.

Figure 12: (a) Original OCT image, (b) histogram with cut-off point T (red) and (c) thresholding image segmentation



Source: Adapted from OCTID (2018).

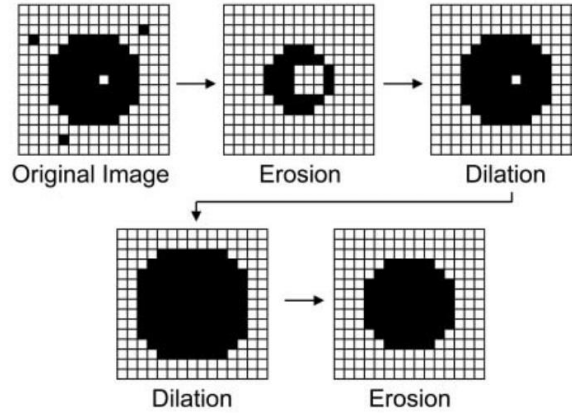
To obtain the image (c) from the original OCT image, a constant T equal to 45 was defined (cutoff point). The pixels with intensity less than T become the background (black). The pixels with intensity greater than T become the object (white).

Threshold segmentation techniques can be divided into classes. A local technique is based on the local properties of the pixel and its neighborhood. A global technique segments an image based on the information obtained globally. Split, merge and growing techniques use both the notions of homogeneity and geometrical proximity in order to obtain good segmentation results (AL-AMRI *et al.*, 2010).

2.5.1.1 Erosion and Dilation

Defects may appear after a threshold operation. These defects are pixels misclassified into the background or object. Given this, dilation and erosion are proposed for the post processing of images (HE; ZHENG; SUN, 2016). Dilation adds pixels to the boundaries of objects, while erosion removes pixels from object boundaries. The number of pixels added to or removed from the objects in an image depends on the size and shape of the structuring element used for the preprocessing. Dilation and erosion techniques are represented in Figure 13.

Figure 13: A series of dilations and erosions



Source: Ikebe; Asai (2005).

Dilation is used for expanding an element A by using the structuring element B , and is defined by:

$$A \oplus B = \{Z | [(\hat{B})_z \cap A] \subset A\}. \quad (7)$$

This equation is based on obtaining the reflection of B from its origin and shifting this reflection by z . The dilation of A by B is the set of all displacements z , such that $\hat{B}$ and A overlap by at least one element.

Erosion is used for shrinking element A by using element B . Erosion for Sets A and B in Z^2 , is defined by:

$$A \ominus B = \{Z | [(B)_z \subseteq A]\}. \quad (8)$$

This equation indicates that the erosion of A by B is the set of all points z such that B , translated by z , is combined in A .

2.5.2 Region-Based segmentation

Region-Based segmentation methods are based on continuity. The segmented image is partitioned into homogeneous areas of connected pixels through the application of homogeneity criteria among candidate sets of pixels. Each pixel in a region is similar with respect to some characteristics or computed property such as color, intensity or texture (WANJARI; KALASKAR; DHORE, 2015).

Given a set of image pixels I and a homogeneity predicate $P(\cdot)$, it is needed to find a partition S of the image I into a set of n regions R_i , defined by:

$$\bigcup_{i=1}^n R_i = I. \quad (9)$$

The homogeneity predicate and partitioning of the image have the properties that any region satisfies the predicate:

$$P(R_i) = \text{True}, \quad (10)$$

for all i , and any two adjacent regions cannot be merged into a single region that satisfies the predicate:

$$P(R_i \cup R_j) = \text{False}. \quad (11)$$

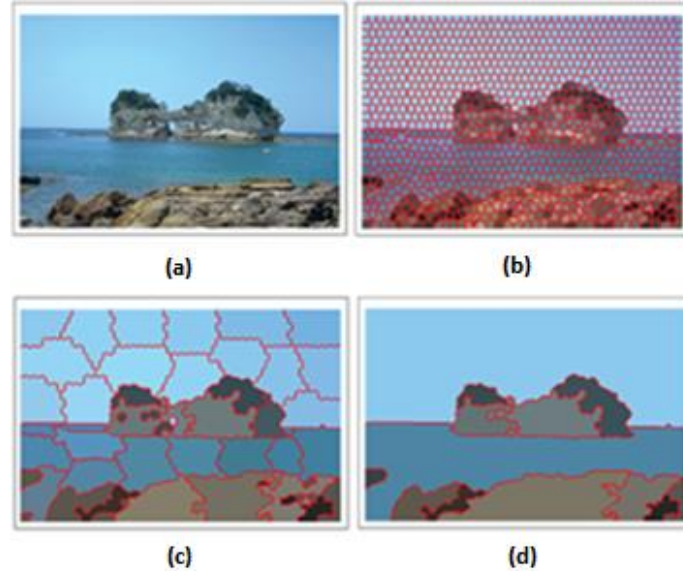
The homogeneity predicate $P(\cdot)$ defines the conformity of all points in the region R_i to the region model (JAIN; KASTURI; SCHUNCK, 1995).

There are two region-based segmentation techniques: growing-and-merging and splitting-and-merging (NAVON; MILLER; AVERBUCH, 2005). In the growing-and-merging method, a set of pixels (seeds) are chosen as the growing regions. Neighboring pixels are iteratively merged into the region if they have similar characteristics to that region: intensity and texture. The growing procedure is repeated with another pixel that has not been merged into any regions, until all pixels have been merged into regions. Figure 14 demonstrates the growing-and-merging method.

In the splitting-and-merging method, the whole image is taken as a unique region, then it is continuously divided until no more splits are possible. Thus, adjacent regions are merged until no more merging is possible. A problem is that the position and orientation of

the image lead to a final blocky segmentation and a regular division leads to over segmentation by splitting (VICENTE; KOLMOGOROV; ROTHER, 2008).

Figure 14: Growing-and-merging method. (a) Input image, (b) super pixels result, (c) clustering result and (d) merged result



Source: Adapted from Hsu and Ding (2013).

2.5.3 Edge-Based segmentation

Edge is a discontinuity in gray level values and can be detected only when a brightness difference presents itself between the two regions. Edges can also be understood as local changes in the image intensity that occur on the boundary between two regions (CHENG *et al.*, 2001).

Edge detection techniques are classified into two categories: sequential and parallel. In sequential techniques, the decision whether a pixel is an edge pixel or not is dependent on the result of the detector of some previously examined pixels. In parallel methods, the decision is based on the point under consideration and some of its neighboring points. It can be applied to every point in the image simultaneously (FU; MUI, 1981). Edge-based segmentation algorithm may be divided into some steps:

1. read input image;
2. apply horizontal mask G_x and vertical mask G_y to the input image;
3. apply different edge detection algorithms and find the gradient;
4. construct separate images for G_x and G_y ;

5. combine the results to find the absolute magnitude of the gradient, described by:

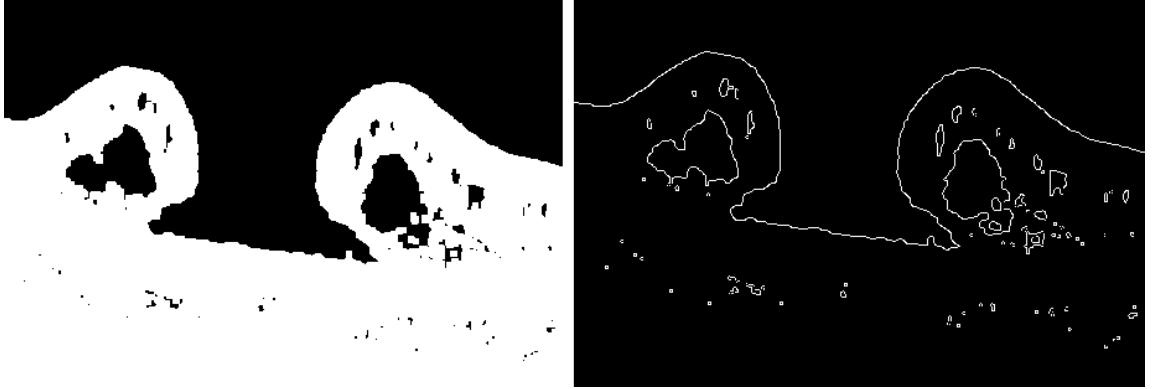
$$|G| = \sqrt{G_x^2 + G_y^2}; \quad (12)$$

6. the absolute magnitude is the output slope magnitude image.

For some slope magnitude images, the values of pixels are too small or too large. To improve visibility of those images, scaling up or down, with the appropriate factor has to be done.

There are different types of edge: step, line, ramp and roof edges. In step edge, the image intensity abruptly changes from one value on one side of the discontinuity to a different value on the other side. In line edge, image intensity abruptly changes value but then returns to the starting value within a short distance. Step and line edges are rare in real images because sharp discontinuities rarely exist in real signals. Ramp edge is the reality of step edge and roof edge is the reality of line edge (MUTHUKRISHNAN; RADHA, 2011). An example of edge detection applied to a binary OCT image is shown in Figure 15.

Figure 15: Edge detection in a binary OCT image



Source: Adapted from OCTID (2018).

Through edge detection, important features such as corners, lines and curves can be extracted from the contours. Higher-level CV algorithms, like pattern recognition, use these features.

2.5.4 Gradient-Based segmentation

In gradient-based segmentation, the gradient of images is computed using convolute gradient operators, and a threshold t is set to distinguish the effective edge whose gradient

is larger than t . The threshold can usually be selected from the cumulative of gradient histograms according to the scheme that 5 to 10% of pixels with the largest gradient can be chosen as edges (JAIN, 1989). For a given image I , the gradient magnitude in each pixel can be determined by:

$$|\vec{G}_j| = \left[\left(\frac{\partial I_j}{\partial x} \right)^2 + \left(\frac{\partial I_j}{\partial y} \right)^2 \right]^{1/2}, \quad (13)$$

where j denotes the color (or luminance) channel under consideration. For any point on a borderline defined by the segmentation, the projection of the gradient vector $\vec{G}$ onto the orthogonal direction of the line ($\hat{n}_l$) defines an accuracy metric for the borderline (PEREIRA *et al.*, 2019). Such projection is given by:

$$G_{\perp,l,j} = \vec{G}_{l,j} \cdot \hat{n}_l. \quad (14)$$

An example is the Watershed algorithm. It can be explained analogously to a hydrological problem, where a relief is flooded progressively from water sources positioned in the valleys. Thus, water would start to accumulate in the lower parts of this relief first, forming several hydrographic basins in different places until they reach higher levels, where water from several basins would come together: flat areas. These will be the zones where the basins that were formed would come together, creating regions. While the flood occurs, each time a region is on the verge of merging with another. A dividing line is created to prevent waters from different regions from touching each other. At the end of the process, each region will have formed a segment of the image, and the dividing lines will be properly contoured.

2.5.5 Classification-Based segmentation

In this technique, each pixel is regarded as an independent observer whose variables are generally obtained through image features like, color, shape, and texture. Therefore, a matrix that contains every pixel as an observer is obtained as the input of the classification. Each observer is then classified into a class (object and background), according to its variables using a learning model (DU; SUN, 2006). A set of images successfully segmented manually is provided as the training set. Coefficients of the learning model are obtained so

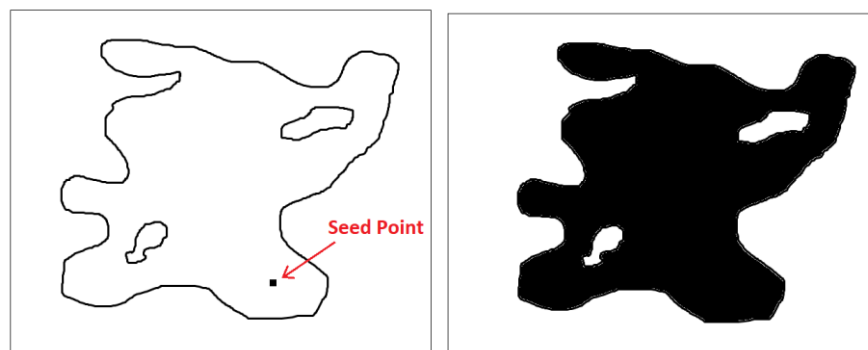
that the testing image set can be classified using the same model with the acquired coefficients.

2.6 Flood Fill algorithm

Flood filling operations are widely used in medical image processing. Almost all the image processing operations of medical scans produce a binary form of an original image in some stage. The binary forms of medical images are normally produced by simple segmentation techniques such as threshold. They contain foreground objects surrounded by background regions.

The Flood Fill algorithm takes three parameters: a starting point or seed point, a target color, and a replacement color. The algorithm looks for all nodes in the array that are connected to the starting point by a path of the target color and changes them to the replacement color. The Flood Fill algorithm application is shown in Figure 16.

Figure 16: Flood Fill algorithm



Source: Author.

The Flood Fill algorithm is most commonly used to fill the holes in the given input image. For binary images, it basically changes the background pixels to foreground pixels until it reaches the object boundaries. For grayscale images, it makes the dark areas surrounded by lighter areas to the same intensity levels. In binary images, the boundaries of the objects need to be specified by connectivity. The starting point for filling can also be specified (GONZALEZ; WOODS, 2002).

The starting point is important, as the algorithm will go from pixel to pixel until it finds the boundaries that will stop it. There are two methods for proceeding to neighboring pixels (kernel): 4-connected and 8-connected methods. The 4-connected method detects and fills the holes one by one in pixel-wise manner. This approach sometimes leads to partial

filling in narrow holes while processing from inside the holes. In the 8-connected operation, the diagonal pixels are included. Hence, it would correctly fill any form of hole (HEARN *et al.*, 2004).

Each pixel will be analyzed (by the algorithm) in order to decide whether to change its color or not. Each time neighboring pixels are analyzed and their colors are changed, the algorithm fills in the spaces until it finds boundaries that make it stop the process.

The Flood Fill algorithm for cases of grayscale analyses is represented by:

$$src(x', y') - loDiff \leq src(x, y) \leq src(x', y') + upDiff, \quad (15)$$

where $src(x, y)$ is the coordinate value of the pixel under evaluation; $src(x', y')$ is the value of one of the pixel neighbors that is already known to belong to the connected component; $loDiff$ and $upDiff$ are the maximal lower and the maximal upper brightness differences between the currently observed pixel and one of its neighbors belonging to the component, or a seed pixel being added to the component, respectively. The equation is general and contributes to the understanding of the binarized method. Summarizing, a Flood Fill algorithm may be divided into the following steps:

1. definition of the position of the starting point (seed point);
2. choice of the neighboring pixels method (kernel);
3. decision of a replacement color and a target color;
4. definition of the path and directions that will be taken by the seed point;
5. analyze each pixel found and define whether or not to replace its color;
6. repeat step 5 until the algorithm has covered all the pixels within the boundaries.

2.7 Border Following algorithm

The Border Following algorithm can extract the topological structure of a given binary image. It has a large variety of applications, including image recognition, image analysis, and image data compression. It derives a sequence of coordinates from the border between forward (1-pixels) and background (0-pixels) regions.

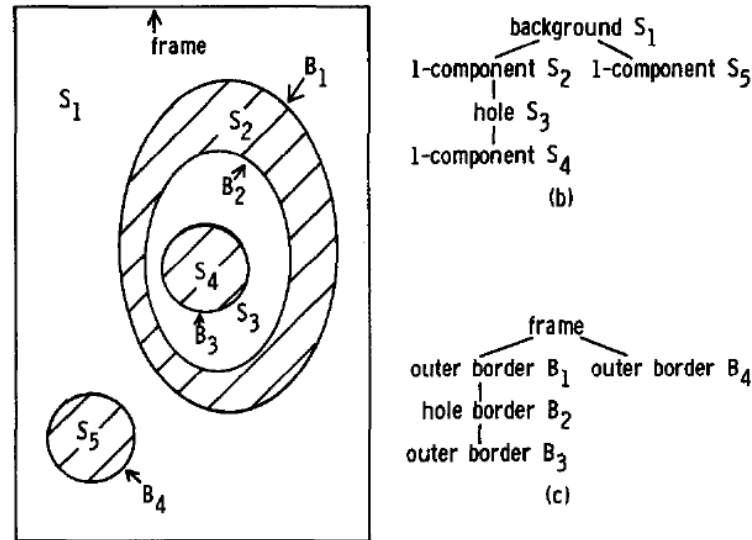
The algorithm scans an input binary image from top to bottom and left to right seeking the starting point, interrupting the raster scan when a pixel (i, j) is found satisfying the condition of either an outer border or a hole border. Then, an identifiable number is

assigned to the newly found border, and is called the sequential number of the border and is denoted by NBD (SUZUKI, 1985).

The algorithm follows the found border from the starting point, marking the pixels. If the current following border is between the 0-component which contains the pixel $(p, q + 1)$ and the 1-component which contains the pixel (p, q) , the value of the pixel (p, q) is changed to NBD. Otherwise, set the value of the pixel (p, q) to NBD unless (p, q) is on an already followed border (SUZUKI, 1985).

This memorized border should be either the parent border of the newly found border or a border that shares a common parent with the newly found border. The parent border of an outer border between a 1-component S_1 and the 0-component S_2 , which surrounds S_1 directly is defined as: the hole border between S_2 and the 1-component which surrounds S_2 directly, if S_2 is a hole; the frame of the picture, if S_2 is the background (SUZUKI, 1985). The Border Following algorithm is explained in Figure 17. After following and marking all the borders, the raster scan is resumed. When the scan reaches the lower right corner of the image, the algorithm stops.

Figure 17: Surroundings among (b) connected components (c) and borders



Source: Suzuki (1985).

2.8 Evaluation metrics

The performance of the segmentation was evaluated by comparing the ground-truth (GT) segmented manually by one or more specialists. The highlighted pixels of prediction

are compared with the GT ones. The two types of segmentation, distance-based and area-based, have different forms and metrics of evaluation.

2.8.1 Distance-Based segmentation evaluation

In the case of distance-based segmentation, the Euclidean Distance (ED) is used to compute the mean and standard deviation (SD) of signed and unsigned error (SE and UE) between the shortest pixels of the automatic segmentation contour (prediction) and the contour of the GT. The Euclidean Distance is the straight-line distance between two points in space. In image analysis the space is two-dimensional and the ED is expressed by:

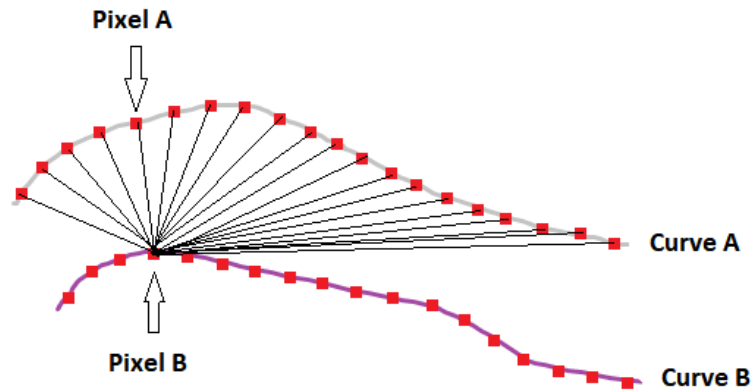
$$ED = \sqrt{(X_{seg} - X_{gt})^2 + (Y_{seg} - Y_{gt})^2}, \quad (16)$$

where X_{seg} and Y_{seg} are the coordinates of each pixel in the automatic segmented image, and X_{gt} and Y_{gt} are the coordinates of each pixel in the GT image.

2.8.1.1 Shortest distance from a point to a curve

To analyze the ILM boundary layer segmentation the Euclidean Distance based on the shortest distance from a point to a curve must be known. It is represented in Figure 18.

Figure 18: Shortest distance from a point to a curve



Source: Author.

For each point of the curve B (prediction), the ED calculation is performed with all points of curve A (ground-truth). Then, the shortest point of A will be chosen to calculate

the distance error at that local point of B . The process is carried out from point to point of curve B with all points of curve A until all the points of curve B are performed. At the end of the process, there will be a shortest distance error for each point of B related to curve A . Then, the calculation of the main and SD error between the two curves, prediction and GT, can be performed.

2.8.2 Area-Based segmentation evaluation

In the case of the area-based segmentation, true positive (TP), true negative (TN), false positive (FP), and false negative (FN) are commonly used. Positive and negative refer to pixels belonging to the MH area and background according to the comparison between prediction and GT, respectively. The confusion matrix for the MH area segmentation is represented in Table 1.

Table 1: Confusion matrix for MH area segmentation

		Ground-truth	
		MH	Non-MH
Prediction	MH	TP	FP
	Non-MH	FN	TN

Source: Author.

Some adopted evaluation metrics are: Accuracy, Intersection over Union (IoU) or Jaccard Index, Dice Similarity Coefficient (DSC) or F1 score and Matthews Correlation Coefficient (MCC). Accuracy simply calculates the percentage of pixels in the image correctly classified, but it can provide misleading results. There may be class imbalance, as TP and TN pixels can dominate the image. The amount of FP and FN pixels neglected can affect the reliability of the result (METRICS, 2020). Accuracy is defined by:

$$Accuracy = \frac{TP + TN}{TP + FP + TN + FN}. \quad (17)$$

The IoU is the area of overlap between prediction and GT divided by the area of union between them. The IoU calculation is described by:

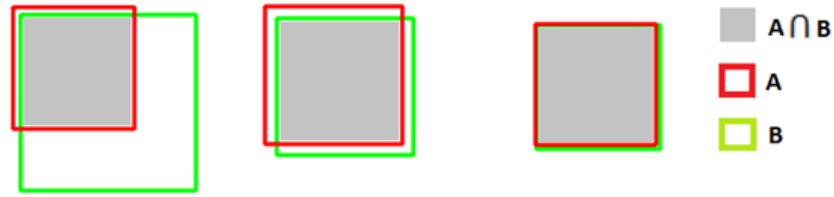
$$IoU = \frac{A \cap B}{A + B - A \cap B}. \quad (18)$$

The DSC is similar to IoU. The area of overlap images is multiplied by 2 and divided by all the pixels in both images (METRICS, 2020). The DSC is expressed by:

$$DSC = \frac{2 \cdot (A \cap B)}{A + B}. \quad (19)$$

An example of the intersection of images is shown in Figure 19 to help understand the equations. Where A represents the prediction and B the ground-truth.

Figure 19: Intersection of images



Source: Author.

In MCC, the higher the correlation between the predicted and GT values, the better the prediction. This metric is also perfectly symmetric, no class is more important than another (MATTHEWS, 2019). The MCC calculation is:

$$MCC = \frac{(TP \cdot TN) - (FP \cdot FN)}{\sqrt{(TP + FP) \cdot (TP + FN) \cdot (TN + FP) \cdot (TN + FN)}}. \quad (20)$$

The MCC produces a high score only if the prediction obtained good results in all of the four confusion matrix parameters (TP , FP , TN and FN). Accuracy and IoU are the most popular adopted evaluation metrics in area-based segmentation. However, Chicco and Jurman (2020) show that they can inflate the results.

2.9 Final considerations of the chapter

Preprocessing and segmentation techniques improve the visual quality of the desired object in the image helping the physicians with their decision making. Preprocessing improves the image quality, correcting defects and highlighting important details for the desired application. Various preprocessing techniques can be used, such as: brightness and contrast adjustment, noise reduction, correction of irregular lighting, edge enhancement etc.

Segmentation is a process of separating one or more objects of interest from the rest of the image. There are many types of techniques, which are classified into five main categories: Thresholding-Based segmentation, Region-Based segmentation, Edge-Based segmentation, Gradient-Based segmentation and Classification-Based segmentation.

Two algorithms that are of significant importance are: Flood Fill and Border Following. The Flood Fill changes the background pixels to foreground pixels until it reaches the object boundaries. The Border Following can extract the topological structure of a given binary image.

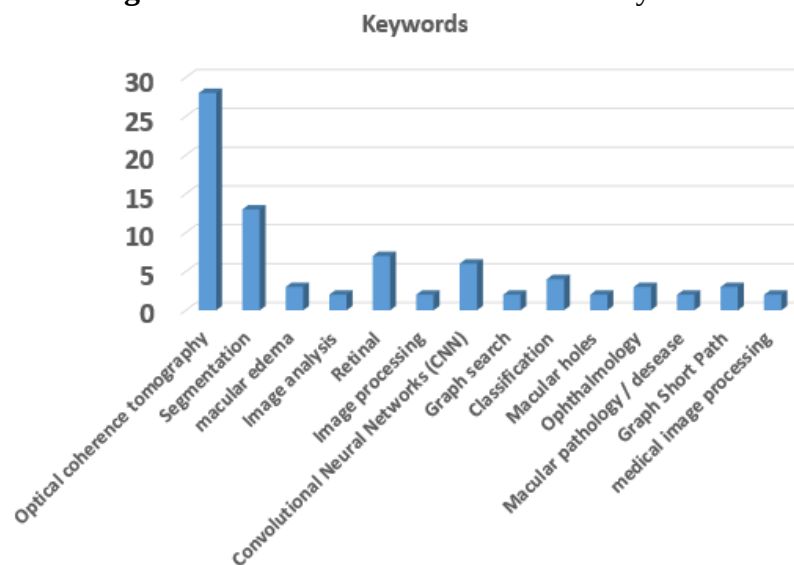
There are two types of segmentation, distance-based and area-based that have different forms and metrics of evaluation. In the case of the distance-based segmentation, the Euclidean Distance is used to obtain the mean distance between the contour prediction and the contour of GT. In case of the area-based segmentation TP , TN , FP and FN are commonly used. The evaluation metrics commonly adopted are: Accuracy, IoU, DSC and MCC.

The theoretical background of this chapter aimed to explain to the reader which techniques are used in the development of this work. This section involved fundamentals that range from the basic understanding of the formation of a digital image to the validation metrics for contour and area segmentations. A good theoretical basis facilitates the understanding of the methodology.

3 STATE OF THE ART

A limited amount of works have been made in this field of study. Therefore, this work can contribute to encourage more research into approaches on automatic segmentation of MH contour and area in OCT images. During the research, many keywords appeared and the most relevant of them for this field are plotted on the graph in Figure 20. This section analyzes the most notable works and explore the most recent and important approaches for automated segmentation in OCT images.

Figure 20: The best and most effective keywords



Source: Author.

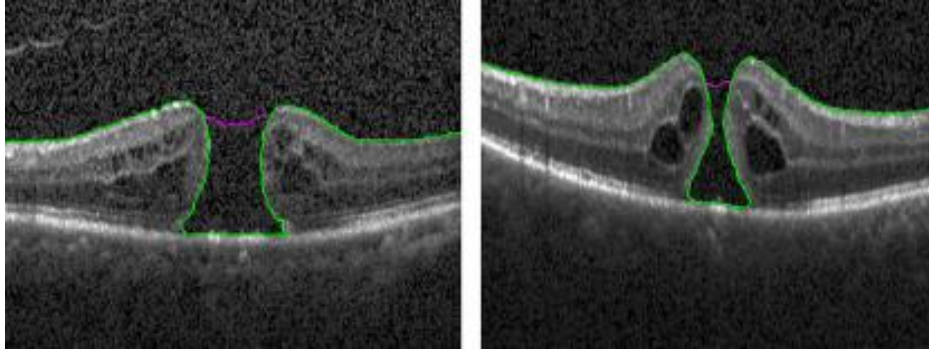
Each work discussed here was divided in respect to its application: automated segmentation of MH volume (set of areas) or contour from OCT images (3 papers) and automated segmentation of boundary layers with other pathologies in OCT images that have similar approaches to MH segmentation (23 papers). Although only three papers are directly linked to segmentation of MH volumes/areas or contours, the other works are supportive as their techniques can be used for MH segmentation. No papers that were directly linked to the segmentation of the MH pathology were found after 2017.

3.1 Review of MH segmentation in OCT images

Keller *et al.* (2016) created an algorithm to segment the MH boundary layer. The Mean and SD distance error between the GT and the predicted segmentation were

performed. A comparison of the shortest path and length-adaptive segmentation was made and can be seen in Figure 21.

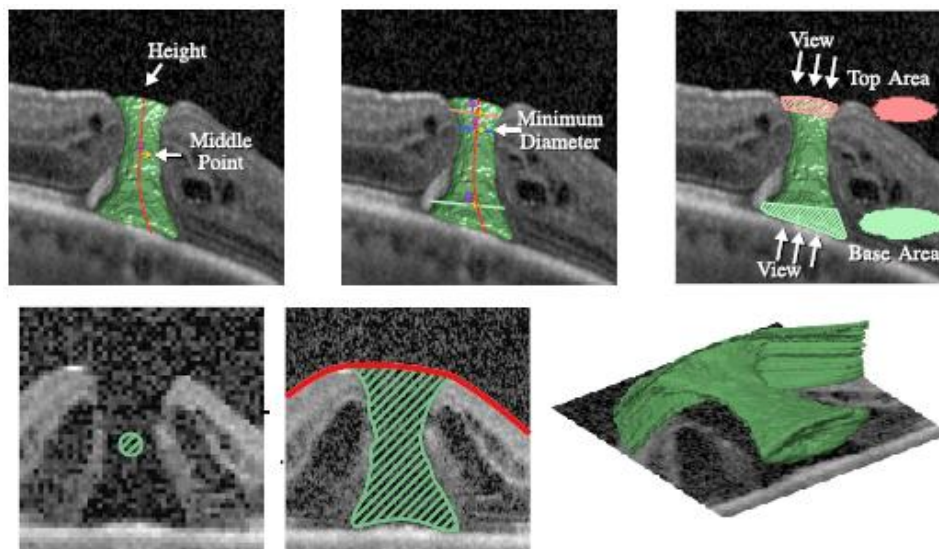
Figure 21: Magenta color denotes the classic shortest path segmentation and green color denotes the proposed algorithm, length-adaptive segmentation



Source: Keller *et al.* (2016).

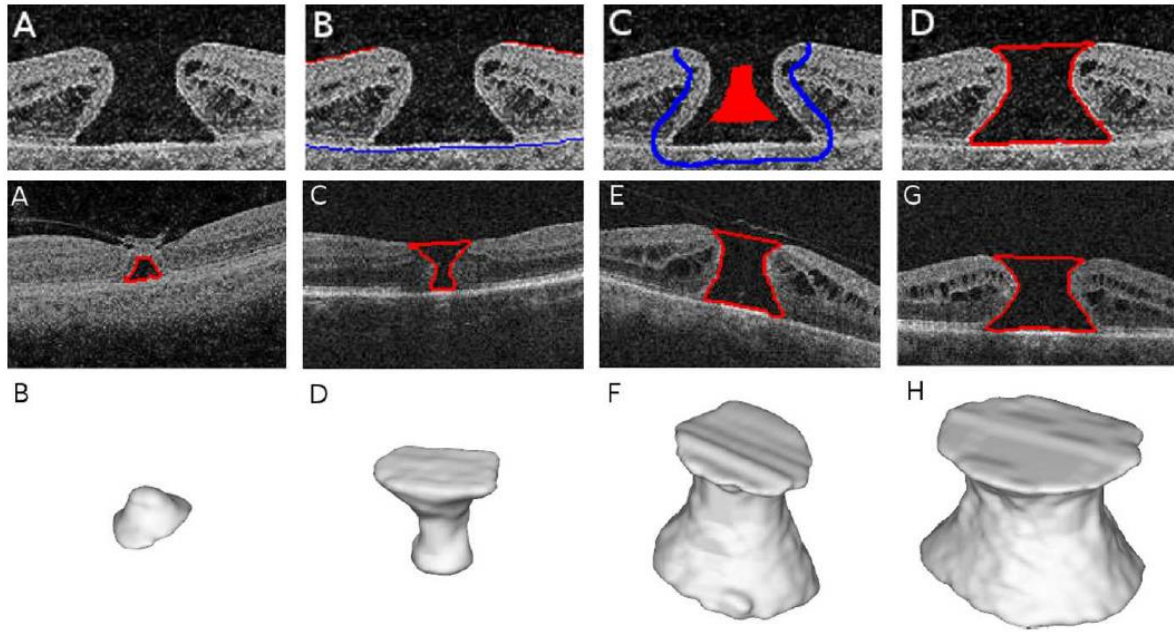
Nasrulloh *et al.* (2017) developed a technique to extract measurements from the automated segmentation of the volume of MH. A set of images of the same eye is needed to assemble the three-dimensional mosaic, forming the MH volume. Due to the type of segmentation, the validation metrics were accuracy, sensitivity, IoU and DSC. The measures of the MH and a 3D surface are shown in Figure 22.

Figure 22: Top images show measures of MH. Bottom images show the formation of a MH volume



Source: Nasrulloh *et al.* (2017).

Xu *et al.* (2013) suggested a method to segment the MH volume through the segmentation of two boundary layers, ILM and RPE. Again, a set of images of the same eye is needed. The red line in is the ILM segmentation and the blue line is the RPE segmentation, both are shown in Figure 23.

Figure 23: MH volume formed through segmentation of ILM and RPE

Source: Xu *et al.* (2013).

3.2 Review of other segmentations in OCT images

The next works were not dedicated to segmenting MH pathology, but similar boundary layers and areas in OCT images. Miri *et al.* (2017) proposed an algorithm to perform the automatic segmentation of the ILM boundary layer within the optic nerve head. Although similar to the work of Keller *et al.* (2016), Miri *et al.* (2017) segmented the ILM boundary layer with a different deformation, which is similar to MH. Zhang *et al.* (2015) proposed a technique to segment Cystoid Macular Edema (CME) in the presence of a MH pathology. The two pathologies were segmented in this work, however only CME was validated with evaluation metrics.

Sui *et al.* (2017) developed an algorithm to segment two choroidal boundary layers. A deep learning method were used and a total of 23,972 OCT images were necessary. Hu *et al.* (2019) suggested a segmentation technique of five retinal boundary layers from normal macular using the same techniques as Sui *et al.* (2017).

Hussain, Bhuiyan and Ramamohanarao (2015) created a method to segment the ILM and the Bruch's Membrane Opening (BMO) from NM. The validation metrics were accuracy, precision and IoU of the Disc area. Hussain *et al.* (2017) created a method to identify four boundary layers in the presence of AMD pathology with drusen and OCT-

defined atrophy using the same techniques as Hussain, Bhuiyan and Ramamohanarao (2015). However, different from them, the validation was based on contour, not on area.

Novosel *et al.* (2017) developed a method to segment eight boundary layers from images with CSR and AMD. Mean and SD of SE and UE were evaluated. Stankiewicz *et al.* (2017) suggested the segmentation of ERM. The validation metrics were the same as Novosel *et al.* (2017).

Naz, Akram and Khan (2017) suggested the automatic segmentation of six boundary layers from OCT images with Diabetic Macular Edema (DME) using two techniques: kernel regression with graph theory and dynamic programming, and structure tensor. Hassan, Ahmed and Li, (2018) proposed an algorithm to segment six boundary layers from OCT images with three conditions: ME, CSR and NM. The accuracy was obtained through a classification algorithm.

Chiu *et al.* (2012) proposed the segmentation of three retinal boundary layers from images with AMD, RPE and drusen. This work was one of the most compared and studied papers by researchers in this field. Tian *et al.* (2015) developed an automatic algorithm called OCTRIMA 3D (OCT Retinal IMage Analysis 3D) that segments OCT volume data in the macular region quickly and accurately. The images have three conditions: NM, DME and dry AMD.

Dodo *et al.* (2019) created an algorithm to segment nine layer areas from images with NM. Mean and SD of DSC were evaluated. González-López *et al.* (2019) developed an automatic segmentation method for four boundary layers also with NM. IoU and DSC were chosen to evaluate it.

Xiang *et al.* (2018) created a method to segment the greatest number of layers in OCT images with CSR pathology, a total of twelve layers. The validation was performed by the Mean and SD of unsigned and signed surface distance errors. Lang *et al.* (2017) created a method to segment multiple boundary layers and area layers in retinitis pigmentosa (RP). Mean and SD of the boundary layer errors and thickness layer errors were evaluated.

Rossant *et al.* (2015) suggested a segmentation method of multiple boundary layers for RP subjects using different active contour approaches. This work extracted specific measures of the pathology. Duan *et al.* (2015) created a technique to segment retinal layers using the Region Growing method. There was no information concerning the database and the results were only qualitative. However, a visual comparison was made between the proposed work and three state-of-the-art methods. It was possible conclude empirically that the proposed method had the best automated segmentation technique.

Niu *et al.* (2014) proposed a technique to segment six retinal boundary layers using dual-gradient and spatial correlation smoothness constraints. The proposed algorithm was applied to OCT images with NM and AMD conditions. Gopinath, Rangrej and Sivaswamy (2017) proposed an algorithm using deep learning, consisting of a combination of Convolutional Neural Network (CNN) and Long Short Term Memory (LSTM). The CNN was used to extract layers of interest from the image and extract the edges, while the LSTM was used to trace the boundary layers. This model was trained on a mixture of NM and AMD cases using minimal data.

El Tanboly *et al.* (2016) suggested the segmentation of the largest number of retinal layers, twelve, considering healthy and diseased subjects. Duan *et al.* (2017) created an algorithm of automatic segmentation based on the Geodesic Distance Method (GDM) for both two and three dimensional OCT images. It is capable of segmenting nine boundary layers with NM and AMD pathologies.

Maurya *et al.* (2020) developed a method to detect the diabetic macular edema (DME) through the segmentation of three layers of the retina. The method consists of gradient information-based segmentation.

To compare and divide the twenty-six papers from sections 3.1 and 3.2 some attributes were necessary. Comparisons between the works, dividing them in two main objectives, contour and area/volume segmentation, are shown in Table 2. The same comparison, but between the papers directly linked to segmentation of MH volume/area or contour are shown in Table 3.

Table 2 shows that there are few published works specifically for MH segmentation. Furthermore, there is a trend to use mean and standard deviation metrics for contour segmentation and a variety of other metrics for area segmentation. The results are mostly analyzed using pixel or μm as measurements. Finally, all the works mentioned here are relatively recent and were carried out as of 2012.

3.3 Final considerations of the chapter

This section analyzed the most significant works exploring the latest and most important approaches for automatic segmentation of OCT images. Despite the limited number of works directly linked to MH segmentation, this literary review also addressed works that segment layers and boundary layers using OCT images, but with other

pathologies. The variety of methodologies seen in this search served as knowledge and a basis for new possibilities, such as the methodology used in this work, which involves a set of techniques acting together.

Table 2: Comparison between papers

Objective		Authors	Metric	Result
Contour segmentation	ILM boundary (MH)	Keller <i>et al.</i> (2016)	Mean $\pm$ SD	3.4 ± 5.74 (pixel)
	ILM boundary	Miri <i>et al.</i> (2017)	Mean $\pm$ SD	6.17 ± 3.19 (μm)
	9 boundaries	Duan <i>et al.</i> (2017)	Mean $\pm$ SD	0.683 ± 0.09 (μm)
	2 boundaries	Sui <i>et al.</i> (2017)	Mean $\pm$ SD	8 ± 7.9 (pixel)
	ILM boundary	Hu <i>et al.</i> (2019)	Mean $\pm$ SD	0.82 ± 0.1 (pixel)
	ILM boundary	Hussain <i>et al.</i> (2017)	Mean $\pm$ SD	1.07 ± 0.17 (pixel)
	3 boundaries	Chiu <i>et al.</i> (2012)	Mean $\pm$ SD	4.2 ± 2.8 (μm)
	ILM boundary	Novosel <i>et al.</i> (2017)	Mean $\pm$ SD	14 ± 21.3 (μm)
	ILM boundary	Naz <i>et al.</i> (2017)	Mean $\pm$ SD	0.0054 ± 0.0014 (μm)
	ILM boundary	González-López <i>et al.</i> (2019)	Mean $\pm$ SD	1.26 ± 0.86 (pixel)
	ILM boundary	Stankiewicz <i>et al.</i> (2017)	Mean $\pm$ SD	26.55 ± 8.81 (μm)
	ILM boundary	Gopinath <i>et al.</i> (2017)	Mean $\pm$ SD	1.49 ± 0.47 (pixel)
	ILM boundary	Lang <i>et al.</i> (2017)	Mean $\pm$ SD	2.52 ± 0.69 (μm)
	ILM boundary	Niu <i>et al.</i> (2014)	Mean $\pm$ SD	2.77 ± 2.10 (μm)
	3 boundaries	Maurya <i>et al.</i> (2020)	Mean	18.84 (pixel)
	ILM boundary	Tian <i>et al.</i> (2015)	Mean	0.72 (pixel)
Area segmentation	MH volume	Nasrulloh <i>et al.</i> (2017)	Accuracy	99.19 ± 0.56 (%)
	CME area	Zhang <i>et al.</i> (2015)	Accuracy	99.7 ± 0.1 (%)
	MH volume	Xu <i>et al.</i> (2013)	Accuracy	96 (%)
	9 volumes	Duan <i>et al.</i> (2017)	UE	0.6596 (μm)
	Disc area	Hussain <i>et al.</i> (2015)	Mean $\pm$ SD	61.87 ± 105.66 (μm)
	12 layers	ElTanboly <i>et al.</i> (2016)	DSC	0.763 ± 0.1598 (pixel)
	11 layers	Xiang <i>et al.</i> (2018)	DSC	92.73 ± 14.21 (μm)
	Fovea location	Hassan <i>et al.</i> (2018)	Accuracy	97.5 (%)
	RNFL layer	Dodo <i>et al.</i> (2019)	DSC	0.9292 ± 0.034 (pixel)
	Multiple layers	Duan <i>et al.</i> (2015)	Visual	Empirical results
	RNFL layer	Lang <i>et al.</i> (2017)	Mean $\pm$ SD	6.59 ± 2.8 (μm)
	RNFL layer	Niu <i>et al.</i> (2014)	Mean $\pm$ SD	2.15 ± 0.21 (μm)
	Retinal layers	Rossant <i>et al.</i> (2015)	Mean $\pm$ SD	Many measures

Source: Author.

Table 3: Papers directly linked to segmentation of MH volume/area or contour

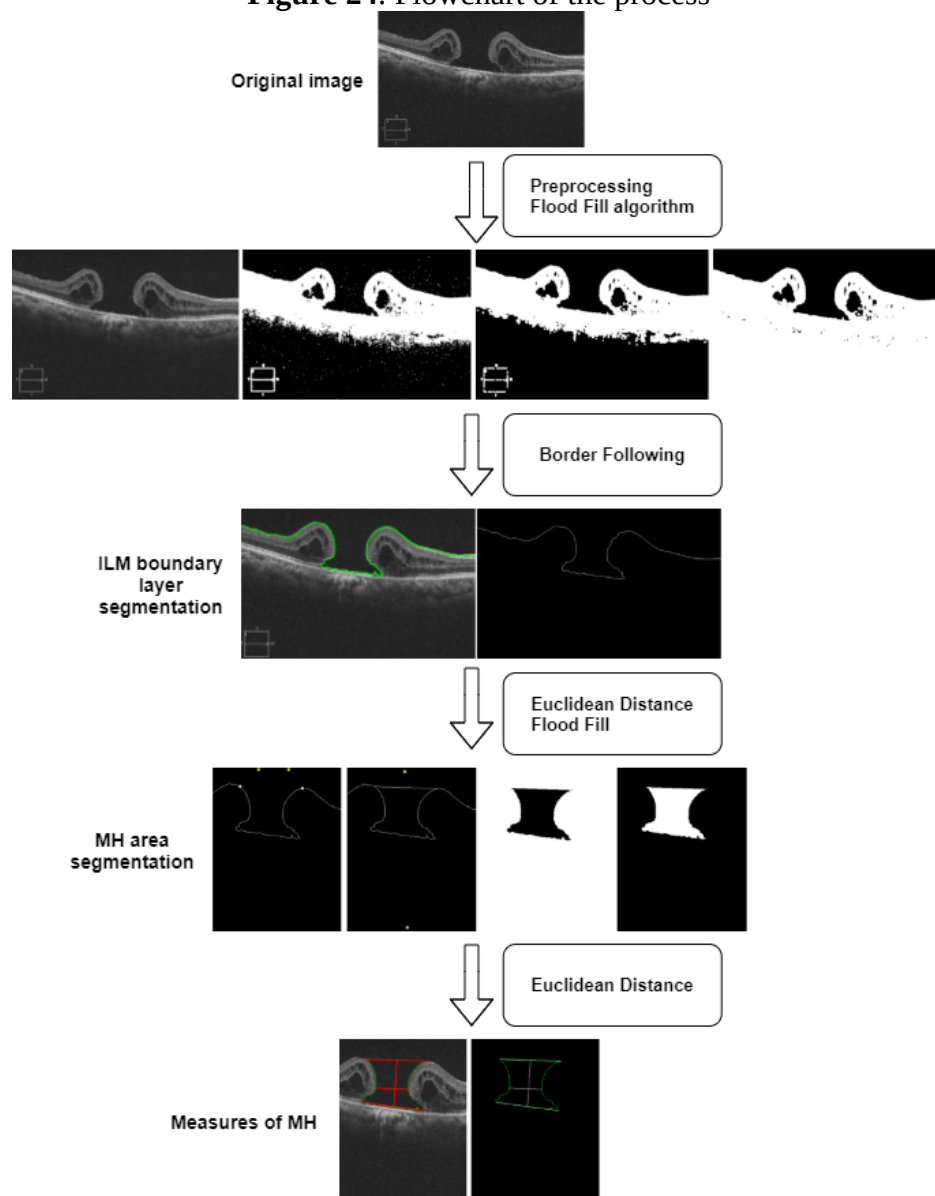
Objective		Authors	Metric	Result
Contour segmentation	ILM boundary (MH)	Keller <i>et al.</i> (2016)	Mean $\pm$ SD	3.4 $\pm$ 5.74 (pixel)
	MH volume	Nasrulloh <i>et al.</i> (2017)	Accuracy	99.19 $\pm$ 0.56 (%)
Area segmentation	MH volume	Xu <i>et al.</i> (2013)	Accuracy	96 (%)

Source: Author.

4 METHODOLOGY

In this Section, techniques for preprocessing and segmenting MH in OCT images are performed. Two types of segmentation are implemented: distance-based segmentation and area-based segmentation. This chapter is divided under the following subtitles: integrated development environment, image database, ground-truth, brightness adjustment normalization, Gaussian Blur filter, thresholding, erosion and dilation, Flood Fill algorithm, ILM boundary layer segmentation with MH (Border Following), Area-Based segmentation of MH and Measurements of the macular holes (shortest distance from a point to a curve and Flood Fill). A flowchart of the process is shown in Figure 24.

Figure 24: Flowchart of the process



Source: Author.

The proposed methodology is a set of techniques, including the shortest distance from a point to a curve (Euclidean Distance), Flood Fill and Border Following algorithms that work together. They are applied throughout the development of the proposed work. Details of the algorithm used in the methodology are given in Appendix A.

The differential of this set of techniques is the mathematical simplicity involved and the ease of application. It is capable of performing the same function as a more complex technique, a machine learning method for example, with lower computational cost. Shortest distance from a point to a curve, Flood Fill and Border Following algorithms are consolidated tools that can be implemented in any programming language.

4.1 Integrated development environment

The proposed work was implemented using Python language (version 3.7) in the PyCharm Integrated Development Environment (IDE). The computer is equipped with Windows 10 platform with an Intel Core i7-3537U CPU at 2.5 GHz and 8 GB RAM.

Other programming environments could be used, such as Matlab, Scilab or Octave. However, Python was chosen due to the number of libraries that make the development of the algorithm less complex. The Python libraries used for the development of the proposed work were: cv2, numpy, imutils, skimage, math, matplotlib, warnings, imageio, PIL, sklearn, seaborn, pandas and time. These libraries are commonly used for image processing and mathematical calculations.

4.2 Image database

Data was acquired from OCT images captured by different devices. Images from public databases are commonly used for research in various areas. Researchers study and compare these databases in order to improve the development of technologies. The OCT images used in this work all came from a public source called Optical Coherence Tomography Image Retinal Database (OCTID).

The high-resolution JPEG images were obtained from a raster scan protocol with a 2 mm scan length and 512x1024 pixel resolution (1 pixel $\sim$ 3.9 μ m). These images were captured using a Cirrus HD-OCT machine (Carl Zeiss Meditec, Inc., Dublin, CA) at Sankara

Nethralaya (SN) Eye Hospital, Chennai, India. The images were then resized to a 500x750 pixel size (GHOLAMI *et al.*, 2020; OCTID, 2019).

The OCTID was the only publicly available database found with cases of MH pathology. It contains more than 500 high-resolution images categorized into different pathological conditions: NM, MH, AMD, CSR, and Diabetic Retinopathy (DR). For this work, only images with NM and MH were necessary. The dataset has a total of 102 B-scan OCT images with MH and 206 B-scan OCT images with NM.

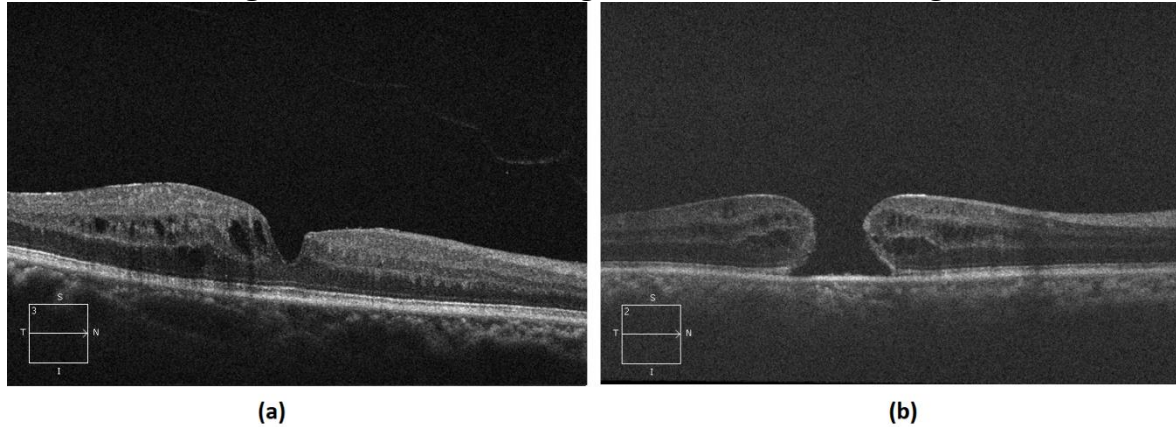
4.3 Ground-Truth

The OCTID image database has no ground-truth to make comparisons and validate the algorithms used. Thus, a specialist from *Escola Cearense de Oftalmologia* (Fortaleza, Brazil) contributed to the work by creating the GT of the ILM contour segmentations from both images with MH and NM, and the GT of MH area segmentations. In addition, MH measurements were also extracted for further evaluation. A Samsung Galaxy Note 10.1 tablet with a S-Pen was used to carry out the manual segmentations.

All the images with NM were used to evaluate whether the algorithm was able to segment the ILM boundary layer and so could be used in other situations. Images that were not a full-thickness MH were excluded by the specialist. Among the 37 discarded images were conditions related to Macular Edema (ME), Macular Pseudo Hole (MPH), Lamellar Macular Hole (LMH), Epiretinal Membrane (ERM), Vitreomacular Traction (VMT) syndrome and MH with Retinal Detachment (RD). There were also NM images in the MH image dataset. A total of 65 MH images were used. Appendix B describes the images, belonging to the OCTID image database that were excluded from this work.

4.4 Brightness adjustment normalization

The database has images taken with different settings. Consequently, the images had varying levels of brightness, which hindered the development processes. Therefore, all images were adjusted to normalize brightness by taking the brightness of one image as a reference for the others. Different brightness levels of the images from the same database are shown in Figure 25.

Figure 25: Difference of brightness levels between images

Source: Adapted from OCTID (2018).

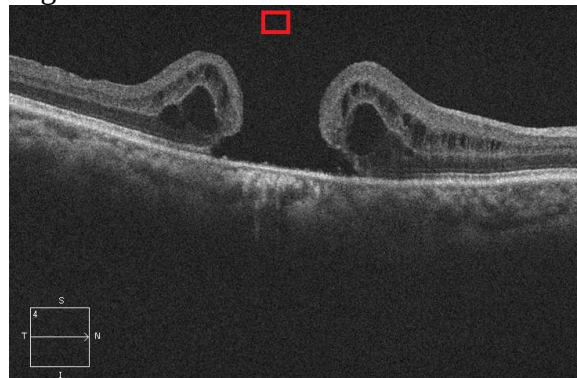
The images must be in the gray scale mode. A small area of the reference image is used to measure its brightness level. For the normalization, the tested image must be between the required brightness level. Table 4 gives the parameters for the normalization of the brightness levels.

Table 4: Brightness level adjustment

Brightness Level (BL)	Alpha
$BL \geq 0.23$	0.65
$0.15 \leq BL < 0.23$	0.85
$0.11 < BL < 0.15$	1.00
$0.08 < BL \leq 0.11$	1.30
$BL \leq 0.08$	1.60

Source: Author.

Alpha is responsible for adjusting the contrast. When BL is between 0.11 and 0.15, no adjustment is necessary. If it is outside these limits, the adjustment is made by adding or removing brightness from the image. An example of OCT image with a red area used to measure the brightness level is shown in Figure 26.

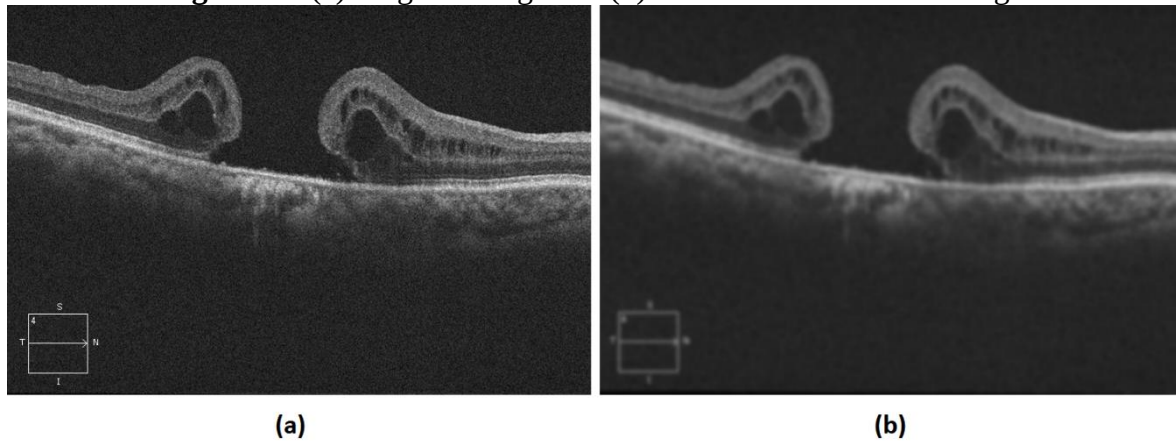
Figure 26: OCT image with an area marked in red to measure the brightness level

Source: Adapted from OCTID (2018).

4.5 Gaussian Blur Filter

OCT images have noise, especially Gaussian noise. Smoothing (blurring) techniques help reduce it. In terms of image processing, any sharp edges in images are smoothed, while minimizing any access of blurring. The kernel used is set to a 11x11 pixel size. The larger the kernel, the smoother the image becomes. In the case of OCT images, there must be a trade-off between smoothing and eliminating noise. If it gets too smooth, the algorithm loses the ability to segment the boundary layers correctly. A comparison between an original and a filtered image is shown in Figure 27.

Figure 27: (a) Original image and (b) Gaussian Blur filtered image



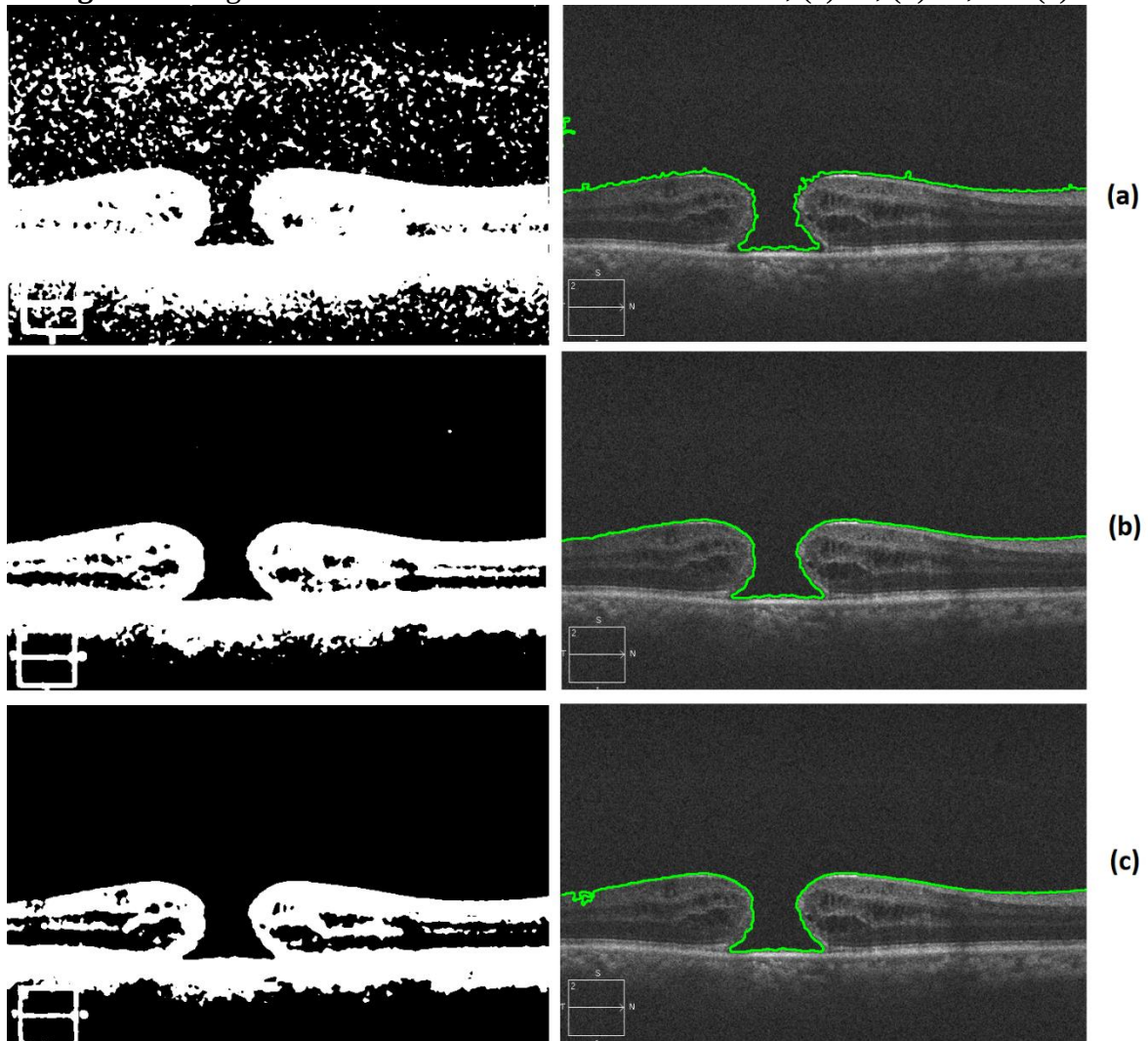
Source: Adapted from OCTID (2018).

4.6 Thresholding

In CV applications, the separation of regions with the objects of interest from the background regions is an useful task. Thresholding often provides an easy and convenient way to perform this segmentation based on different intensities in the foreground and background regions of an image. There are thresholding techniques for color images (not part of the scope of this work) and for gray scale images.

In the case of gray scale images, the output is a binary image representing the segmentation. The same threshold value of 45 is applied to all pixels, ranging from 0 to 255 in shades of gray. This cutoff was chosen as it best suits a good segmentation for a pattern of images processed following the applied methodology. Some ILM contour segmentations are shown in Figure 28 after different thresholding cutoff values were applied; the best result was for a 45 cutoff point.

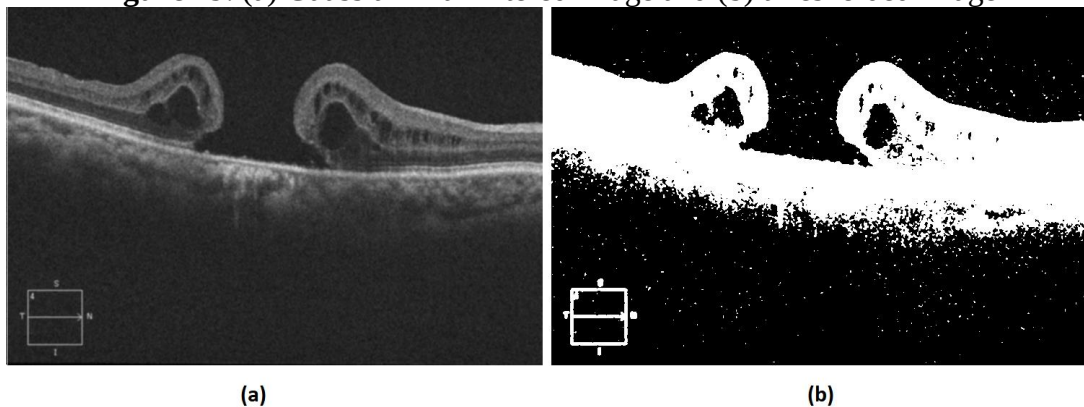
Figure 28: Segmentation results for different cutoff values; (a) 40, (b) 45, and (c) 50



Source: Author.

Each pixel in the image is compared with the threshold. If the intensity of the pixel is higher than the threshold, the pixel is set to white in the output. If it is less than the threshold, it is set to black. The thresholding technique is shown in Figure 29.

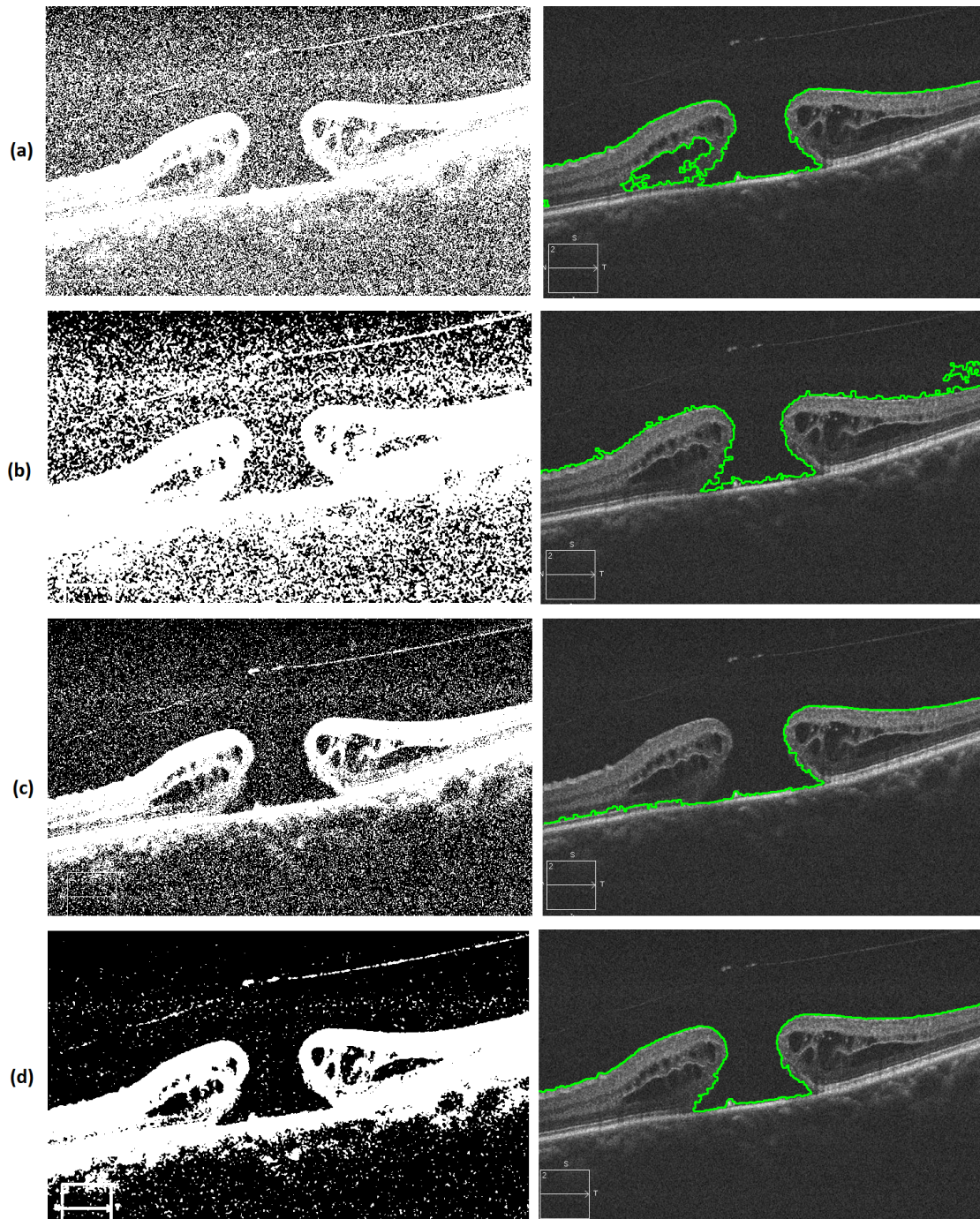
Figure 29: (a) Gaussian Blur filtered image and (b) thresholded image



Source: Adapted from OCTID (2018).

Both the brightness adjustment and the Gaussian Blur filter are important for thresholding. The challenge of the algorithm is to find a compromise solution between these two parameters, because their responses have an impact on thresholding and consequently on segmentation. The influences of brightness adjustment and Gaussian Blur filter on the thresholding process and segmentation are demonstrated in Figure 30.

Figure 30: Threshold and segmentation (a) without brightness adjustment and Gaussian Blur filter, (b) without brightness adjustment, (c) without Gaussian Blur filter and (d) with both of them



Source: Author.

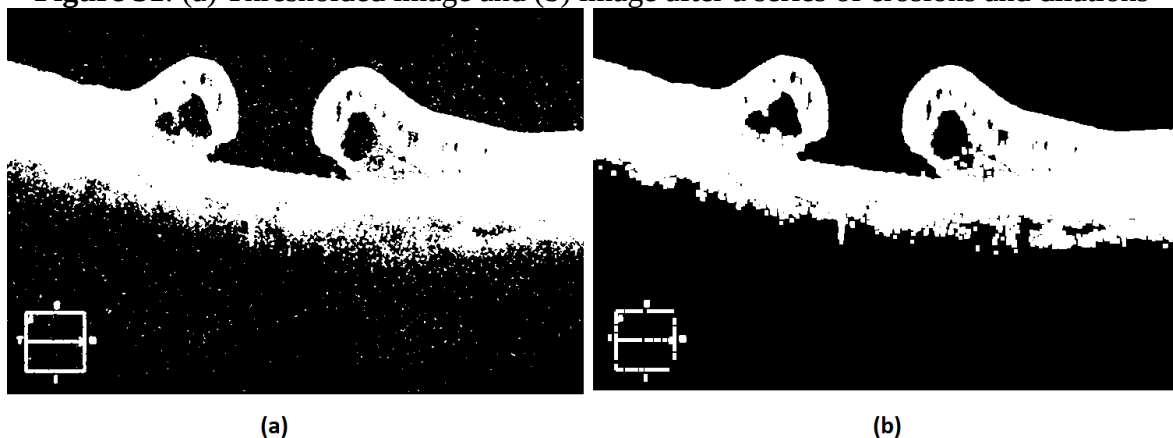
The thickness of the contour in these images has been enlarged in order to be able to observe the quality of the results more easily. When the algorithm compares the prediction and GT, the contour has a width of only one pixel.

4.7 Erosion and Dilation

Erosion and dilation are morphological operations. Dilation adds pixels to the boundaries of the object, while erosion removes pixels from the object boundary. The number of pixels added to or removed from the object depends on the size and shape of the structuring element used to process the image. It needs two inputs: the first one is the original image (after thresholding), and the second one is the structuring element or kernel, which decides the nature of the operation. The kernel used in both dilation and erosion is 3x3. Two erosions and two dilations are applied alternately. This approach increases the effectiveness of the technique and ensures the original contours of the desired object for segmentation.

The value of each pixel in the output image is based on a comparison of the corresponding pixel in the input image with its neighbors. In this application, dilation fills small holes and erosion removes islands and small objects, so that only substantive objects remain. These steps remove small white objects, preserving the contours of the boundaries that are essential to perform a satisfactory segmentation. An image after a series of two erosions and two dilations is demonstrated in Figure 31.

Figure 31: (a) Thresholded image and (b) image after a series of erosions and dilations

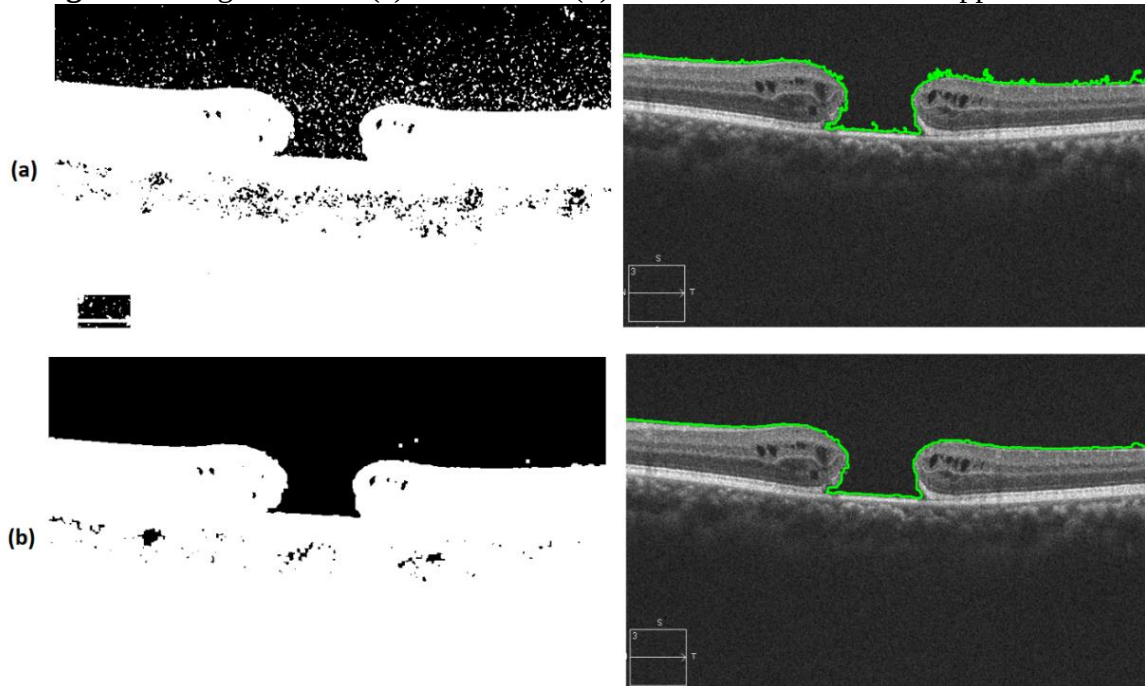


Source: Author.

Erosion and dilation are necessary, as they remove white objects that may interfere with the segmentation. However, there is a trade-off between noise removal and contour quality. The more erosion and dilation processes applied, the more the segmentation will

present errors. Therefore, only two alternate applications of erosion and dilation are applied. The effect of these techniques is shown in Figure 32.

Figure 32: Segmentation (a) without and (b) with erosion and dilation application



Source: Author.

4.8 Flood Fill algorithm

The Flood Fill algorithm is the first step in the set of techniques applied. Principally it is used to determine a bounded area connected to a given node in a multi-dimensional array. The first step to use this technique is to choose the image that will go through the Flood Fill process. The image is binarized through a thresholding process.

A specified seed point is chosen (x, y) and reassign all pixel values until reaches the object boundary. The seed point with coordinates $x = 550$ and $y = 470$ (cross mark) is shown in Figure 33.

The chosen coordinates are at the position close to the bottom of the image because the objective of this step is to separate the image into two regions (black and white). After the series of erosion and dilation have been performed, the bottom half of the image is a black space, as can be seen in Figure 31(b). So, the objective is make that space white. Thereby, the ILM boundary layer is well defined, separated by the forward and background regions, so that it can be segmented. This operation changes the background pixels to foreground pixels, and stops when it reaches the object boundaries.

Figure 33: Seed point (cross mark) in an OCT image



Source: Adapted from OCTID (2018).

The chosen kernel or neighboring pixels uses the 8-connected method. This kernel includes the diagonal pixels. Hence, it would correctly fill any format. The color for the fill is white, the forward color. The algorithm travels in all directions, analyzing each pixel found and defining whether or not to replace its color. This process is executed until the whole area within the boundaries have been covered. The result of the Flood Fill application can be seen in Figure 34.

Figure 34: (a) A series of erosions and dilations and (b) Flood Fill application



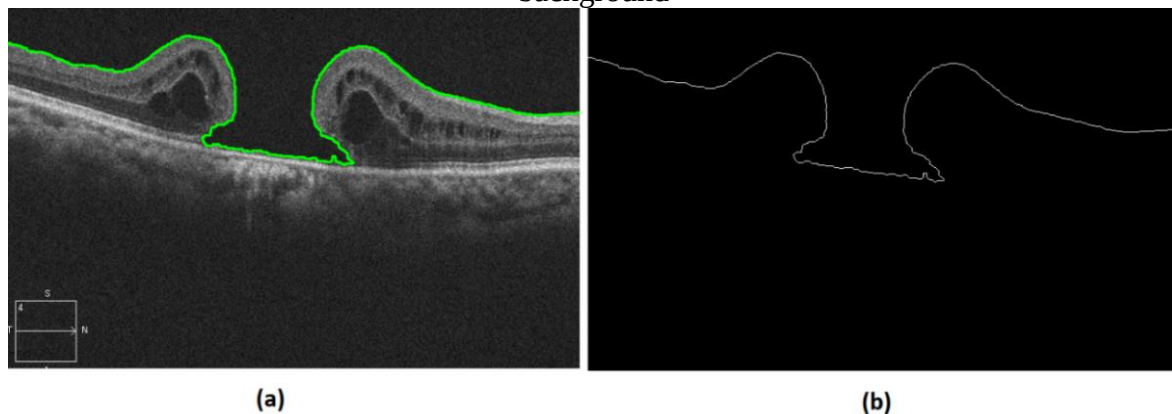
Source: Author.

4.9 ILM boundary layer segmentation with MH

After the Flood Fill application, the ILM boundary layer is differentiated, therefore being segmented. This segmentation step is performed by finding the largest contour between the white object and the background; this is done because the binary image has small black holes that also form small contours.

Then, the second step of the set of techniques is performed, that is the Border Following algorithm proposed by Suzuki (1985). Three components are necessary: the first one is the source image, the binarized image after the Flood Fill algorithm; the second one is the contour retrieval mode, the approach retrieves only the largest contour, and the side and bottom cuts in the image eliminate unwanted parts of the contour; the third one is the contour approximation method, it compresses horizontal, vertical, and diagonal segments and leaves only their end points. The contour method generates an array of (x, y) coordinates from the largest contour related to the black background. The result of the preprocessing and segmentation methods is the ILM boundary layer segmentation that is represented in Figure 35.

Figure 35: (a) ILM boundary layer segmentation on the original image, and (b) on the background



Source: Adapted from OCTID (2018).

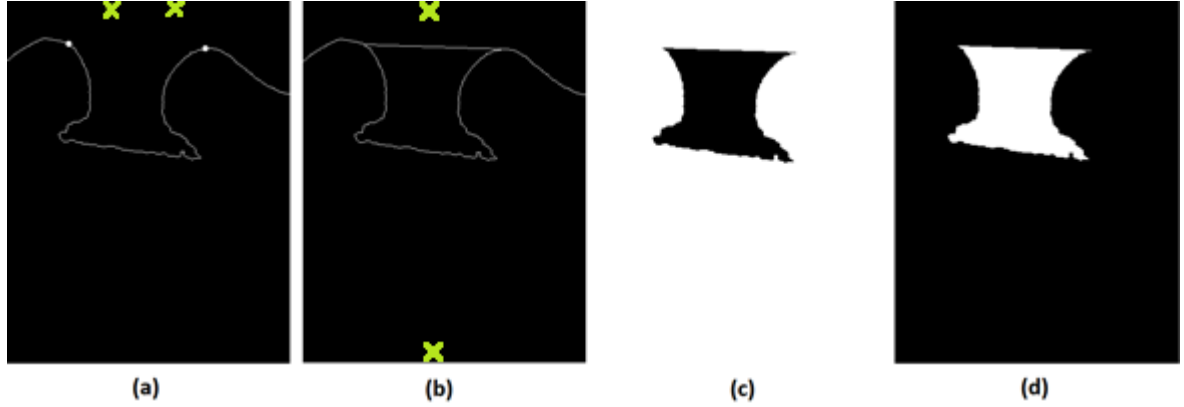
4.10 Area-based segmentation of MH

In this work, both contour and area segmentations were performed. The segmentation of a MH area is obtained by applying the third step of the set of techniques. Here, the extreme top left and top right pixels of the macular hole must be located. The shortest distance from a point to a curve is used to find these pixels. The objective is to find the shortest pixel of the curve related to each of two seed pixels fixed on the top left and top right at a certain distance from the center top of the image. These pixels will be the top left and top right points of a MH.

Having found these extreme pixels, the next step is to draw a straight line between them, closing the hole. Now with the closed area, the image can be separated into three regions, a bottom region, a top region and the one bounded by the macular hole. The Flood

Fill technique is again applied to the upper and lower delimited regions of the image, leaving only the area of the macular hole in black. Finally, the colors of the pixels are inverted, so the MH area becomes white and background becomes black. The process to obtain the MH area is summarized in Figure 36.

Figure 36: (a) Euclidean distance to find the shortest points (white) through the seeds (cross marks), (b) three regions and the seed points to apply the Flood Fill to, (c) MH area in black, (d) MH area after inversion of colors



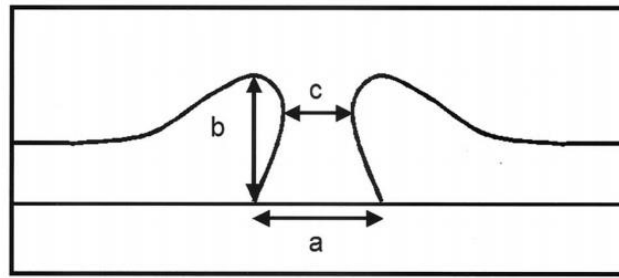
Source: Author.

4.11 Measurements of the macular hole

In addition to the important segmentation process, macular hole measurements are fundamental for the surgeon to decide whether or not a hole closure surgery is feasible. The relationship between some measures determine whether the hole can be closed or not and whether the pathology is developing to a worse scenario or not.

An important metric to decide a hole closure surgery is the Macular Hole Index (MHI). The MHI is defined as the ratio of the hole height to the base diameter, which refers to the perpendicular and horizontal dimensions of the hole visualized from cross-sectional OCT images (KUSUHARA *et al.*, 2004). Patients are classified into two groups. If the MHI is greater than 0.5 the surgery can be performed without problems, however, if the MHI is less than 0.5 the possibility of closing the macular hole is less, so the surgery is less likely to be successful. In such cases, for the closure it is decided to use, in addition to the conventional technique, the addition of a miniotic membrane in the MH is performed, to assist in the closure. Figure 37 shows the MHI and it is calculated as:

$$MHI = b/a. \quad (21)$$

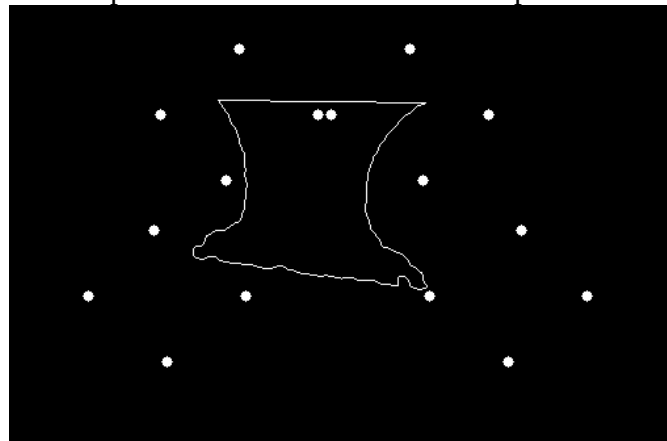
Figure 37: MHI

Source: Kusuhara *et al.* (2004).

Independent of the MHI, the most important measures for specialists at the time of analysis are: top diameter, base diameter, minimum diameter and height. To obtain these measures, some steps are necessary.

The first step is to obtain the contour of the MH area. The Border Following algorithm that is applied to find the ILM boundary layer contour is also applied here.

Four seeds are used to find each of the extreme points of the MH area contour (top left, top right, bottom left and bottom right). The pixel on the MH area contour that is shortest to the relationship between the four seed points will be the desired extreme point. These measurements of a shortest point to a curve are calculated using the Euclidean Distance. Once again, the set of techniques is used. Figure 38 shows the sixteen seed points used to find the four extreme points of the MH area.

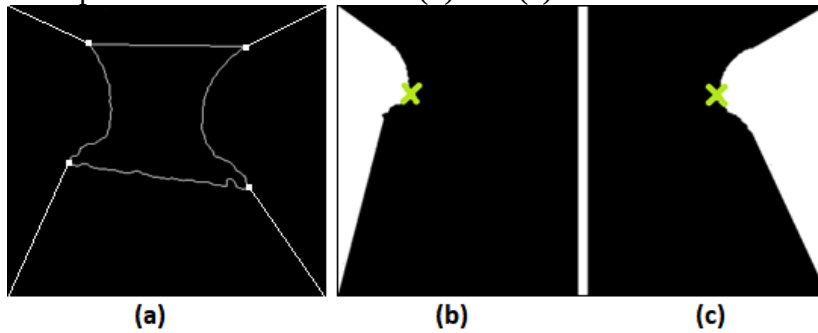
Figure 38: Seed points used to find the extreme points of the MH area

Source: Author.

The top diameter is the distance between the top left and top right pixels of the MH area previously obtained. The bottom diameter is obtained in the same way as the top diameter, through the shortest distance using the Euclidean Distance method. The height is then determined using the midpoint of the bottom diameter and the midpoint of the top diameter and tracing a line between them.

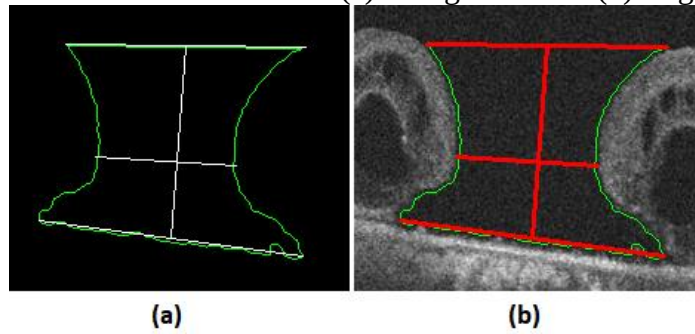
The minimum diameter is obtained through the shortest distance from a point to a curve and Flood Fill algorithms. A straight line is drawn from the left pixel of the top diameter to the top left pixel of the image and another straight line is drawn from the left pixel of the bottom diameter to the bottom left pixel of the image, creating an area on the left side, which is filled through the Flood Fill. The most extreme right pixel in this area is the left pixel of the minimum diameter. The same process is carried out on the right side, finding the right pixel of the minimum diameter. Once the pixels are found, a line is drawn between them; this line is the minimum diameter. The process to find the minimum diameter is shown in Figure 39 and all the measures of a MH are represented in Figure 40.

Figure 39: Seed points of extreme areas in (b) and (c) to obtain the minimum diameter



Source: Author.

Figure 40: Measures of the MH in (a) background and (b) original image



Source: Author.

The methodology of this work is extensive and includes a variety of different techniques. The algorithm of the proposed methodology can be seen in detail in Appendix A.

4.12 Final considerations of the chapter

The proposed work was implemented using Python language. The database used is a public database called Optical Coherence Tomography Image Database (OCTID). The preprocessing steps performed before the segmentation were: brightness adjustment

normalization, Gaussian Blur filter, threshold, erosion and dilation. ILM boundary layer segmentation with MH pathology, area-based segmentation of MH, and measures of MH. The ILM boundary layer segmentation with NM was taken after the preprocessing techniques.

The proposed methodology is a set of techniques that work together: shortest distance from a point to a curve (Euclidean Distance), Flood Fill and Border Following algorithms. During all stages of the development of the proposed work, the set of techniques is applied.

Flood fill is an algorithm that is mainly used to determine a bounded area connected to a given node in a multi-dimensional array. The Border Following finds the border between a white object and a black background. The objective of the Euclidean Distance in this work is to find the shortest distance between a seed point and a desired pixel of a contour or area.

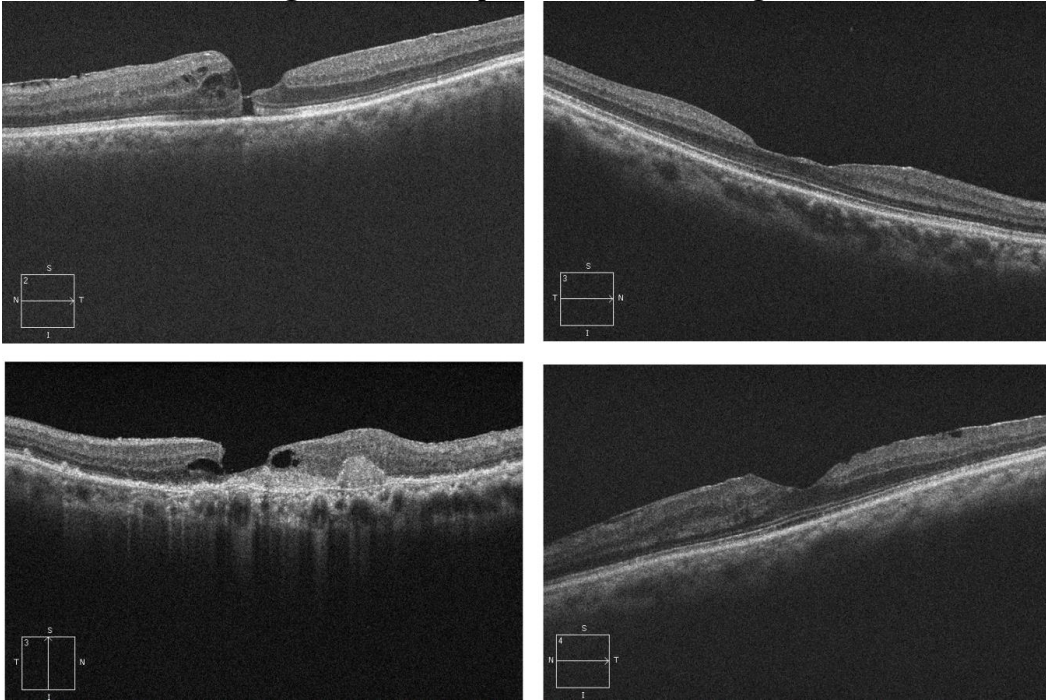
The differential of this set of techniques is the mathematical simplicity involved and the ease of application. These techniques together can perform the same function as a more complex technique, such as a machine learning method, with lower computational costs. The shortest distance from a point to a curve (Euclidean Distance), Flood Fill and Border Following algorithms are consolidated tools, which can be implemented in any programming language.

5 RESULTS AND DISCUSSIONS

In this section, the results from the OCT images are presented and discussed in four parts. The first is the evaluation of ILM boundary layer segmentation in OCT images with MH pathology. The second evaluates the segmentation of the macular hole area. The third, is the evaluation of the MH measurements. The fourth part, evaluates the ILM boundary layer segmentation using OCT images without any pathology (NM cases), proving that the algorithm can also be used in other situations.

The OCTID has 102 OCT images with MH, of which 65 were used. The 37 discarded images contained examples of ME, MPH, LMH, ERM, VMT syndrome, MH with RD and also NM. Figure 41 shows some examples of the discarded images. All excluded images can be seen in Appendix B. Although these classes of holes are in the database, they do not fall within the scope of this study, as they are not interesting for physicians in terms of surgical analysis. Regarding the cases of NM, all 206 OCT images were used in this work.

Figure 41: Examples of discarded images



Source: Adapted from OCTID (2018).

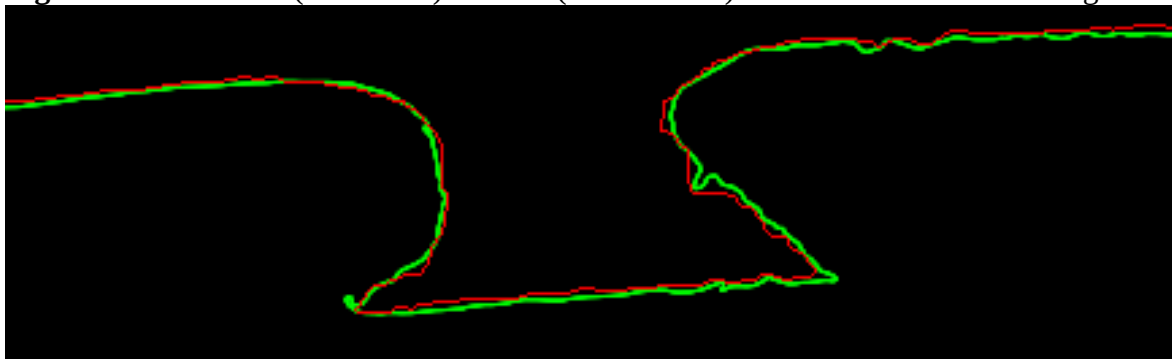
5.1 Results for ILM boundary layer segmentation with MH pathology

During the development of this work, a lot of time was spent to find the main techniques applied to contour and area segmentations in OCT images. Although, there are

some studies in this field, the majority were for other types of pathologies. There were few works related to macular hole segmentation.

The segmentation of the ILM boundary layer with MH pathology is part of the first step of the algorithm. A good segmentation of the MH area is only possible if this previous step has satisfactory results. As already mentioned, this step is part of the distance-based segmentation. The distance error of the shortest points to the curve has to be calculated by applying the shortest distance from a point to a curve (Euclidean Distance). An example of the prediction and ground-truth contours on the same background are shown in Figure 42.

Figure 42: Prediction (wider line) and GT (thinnest line) contours on the same background



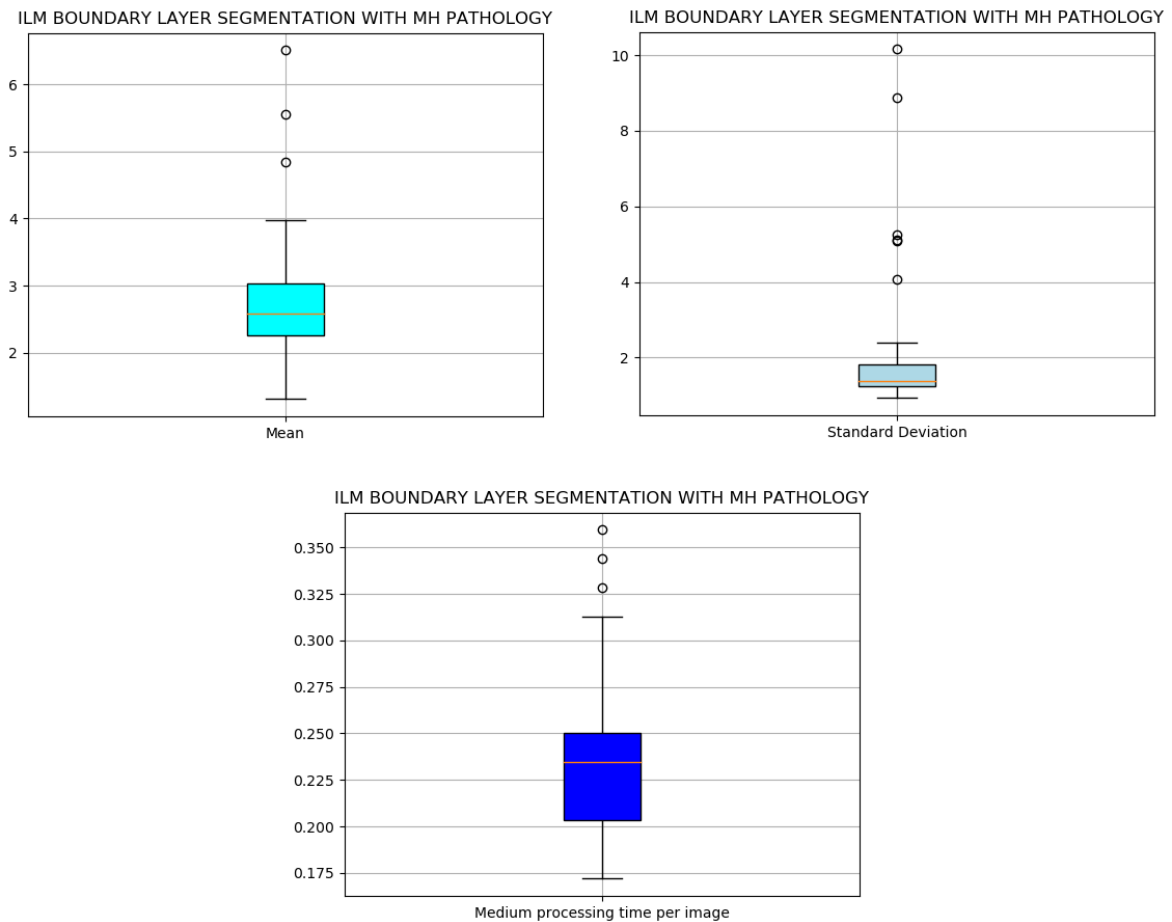
Source: Author.

The evaluation metrics used are mean and standard deviation of the unsigned error. Other works use these metrics for this type of segmentation. Boxplots with results are shown in Figure 42. The boxplot is important because it identifies where 50% of the most likely values, the median and the extreme values are located. This shows whether the algorithm is working in a standard way for all OCT images.

Outliers are the individual points distant from the median box. There are only a few outliers in the boxplots of Figure 43. This may have been due to some points outside the curve that were not completely eliminated by the preprocessing techniques. The prediction segmentation is compared with the GT generated by a specialist and the results are shown in Table 5.

The proposed method is important for the scientific community who are involved in research in the area of ILM boundary layer segmentation of OCT images with MH pathology. The results are satisfactory and better than the state-of-the-art method as seen in Section 5.5.

Figure 43: Boxplots of mean and SD of UE (pixel) and medium processing time per image (s) for the tests of ILM boundary layer segmentation with MH pathology



Source: Author.

Table 5: Mean and SD of UE (pixel) and medium processing time per image (s) of ILM boundary layer segmentation with MH pathology

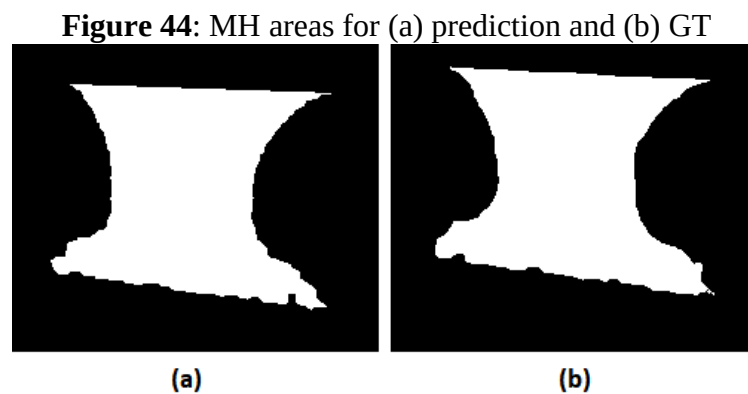
Method	Mean and SD	Proces. Time per image (s)	Quant. images
Proposed method	2.72 ± 1.91	0.24	65

Source: Author.

5.2 Results for MH area segmentation

Macular hole area segmentation is important as it can provide the shape and the measures of the hole. The different features in the shapes can be used in other applications, such as classification algorithms. Some of these features are the measures of diameters and height. Diagnostic imaging exams use this type of segmentation for various tasks in the search for abnormalities and to make calculations in order to decide whether or not to have surgery.

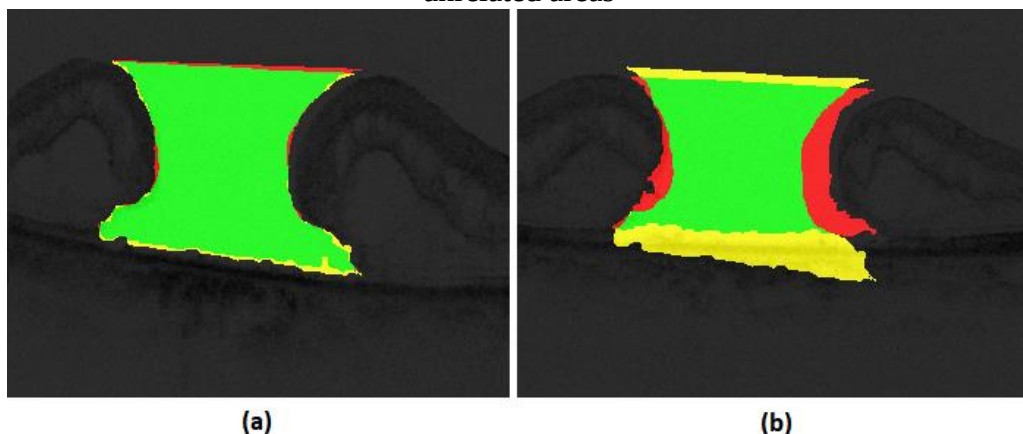
The MH area segmentation is only possible after a correct ILM boundary layer segmentation. This type of segmentation reinforces the effectiveness of the algorithm, since other evaluation metrics, such as accuracy, Intersection over Union (IoU) or Jaccard Index, Dice Similarity Coefficient (DSC) or F1 score and Matthews Correlation Coefficient (MCC) can be applied. In addition, a confusion matrix of pixels that allows a qualitative evaluation of the segmentation to be generated. Examples of prediction and ground-truth for the area are shown in Figure 44.



Source: Author.

The qualitative information given by the confusion matrix is another way to observe how well the algorithm is working. The confusion matrix for these related and unrelated areas is represented in Figure 45. The evaluation is performed by analyzing pixel by pixel of the predicted and ground truth MH areas.

Figure 45: Confusion matrix of MH area segmentation for (a) related areas and (b) unrelated areas

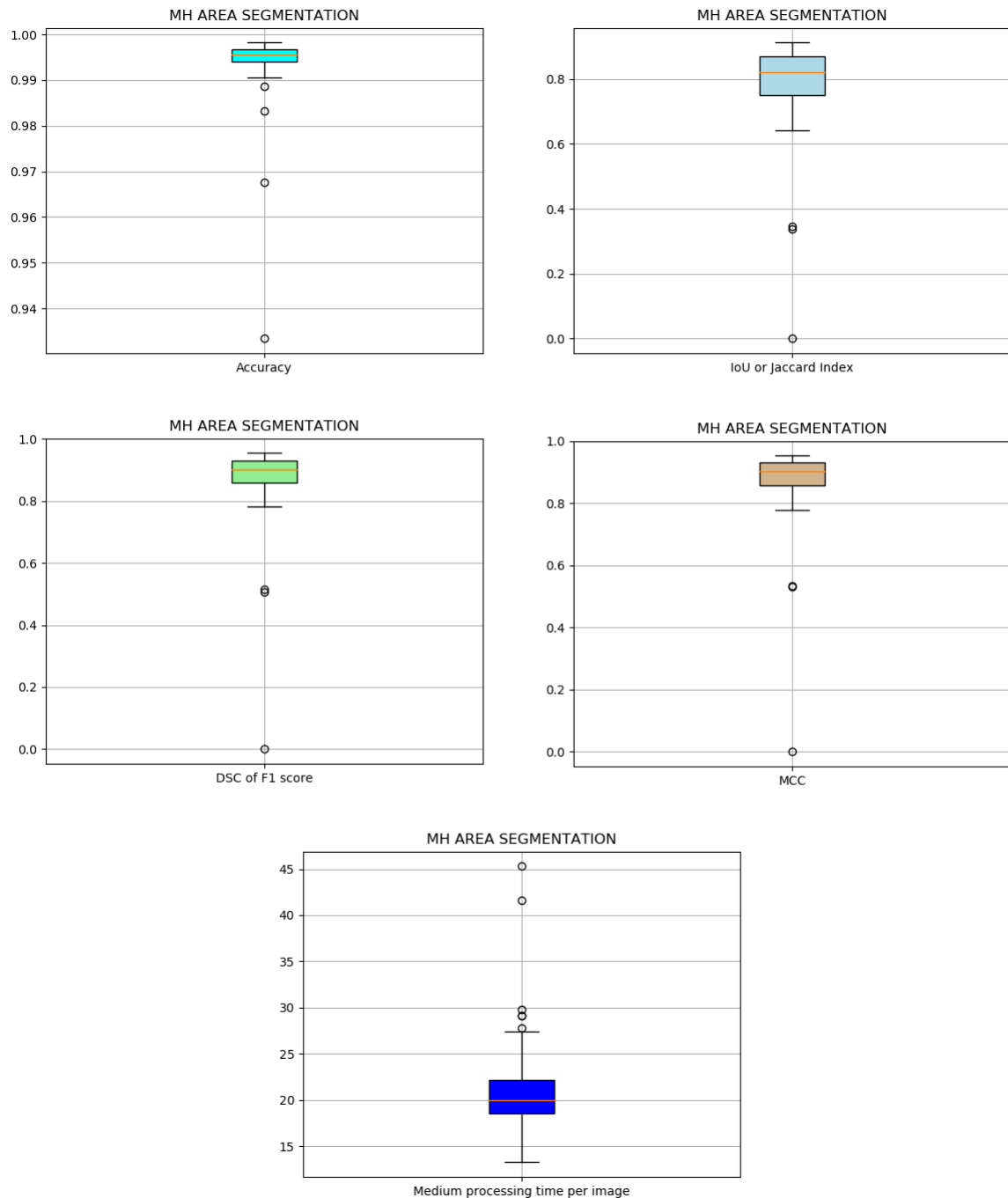


Source: Author.

The green color (*TP*) occurs when the automatic area segmentation is correctly predicted. The red color (*FP*) occurs when the automatic area segmentation is incorrectly

predicted. The shades of gray (TN) occur when the background is correctly predicted. The yellow color (FN) occurs when the GT is incorrectly predicted. Boxplots with the results of the evaluated metrics for all MH images are shown in Figure 46.

Figure 46: Boxplots of accuracy, IoU, DSC, MCC (%) and medium processing time per image (s) for the evaluation of MH area segmentation



Source: Author.

As already mentioned, the boxplot is important because it identifies where 50% of the most likely values, the median and the extreme values are located. There are few outliers

in the boxplots. Outliers are acceptable, since the algorithm deals with various formats of area. These variations can generate more discrepancies in some results. Overall, the results are satisfactory. The prediction segmentation is compared with the GT generated by a specialist and the results of the evaluation metrics are shown in Table 6.

Table 6: Results of evaluation metrics in percentage for MH area segmentation

	Accuracy	IoU	DSC	MCC	Proces. Time per image (s)
Proposed method	99.37	78.61	86.98	87	19.95

Source: Author.

There is an image with zero in the evaluation for IoU, DSC and MCC, which means that the algorithm was probably unable to segment the MH area for that image. Even so, the lowest accuracy value, which is probably of the image with failed area segmentation, has a value of 0.925. This high value explains why accuracy is not a robust form of assessment for segmenting areas in images. The large amount of *TN* disguise the fact that the algorithm did not perform well in area segmentation. Therefore, for comparison with the state of the art in Section 5.6, accuracy was not taken into account.

5.3 Results for measurements of macular holes

Macular hole measurements are of fundamental importance for the specialists. These professionals need to take these measurements manually to calculate the possibilities of surgery. Automatic methods for these measurements have been little explored by the scientific community. Despite the objective of this work was the contour and area segmentation of MH, an extension of the algorithm was performed in order to obtain these measurements.

The measurement results were obtained by comparing the algorithm results (prediction) and GT, which are the measurements made by the physician. The mean and SD of UE in micrometers for each measure prediction compared to the GT is shown in Table 7.

Table 7: Mean and SD of UE (μm) for measurements of MH

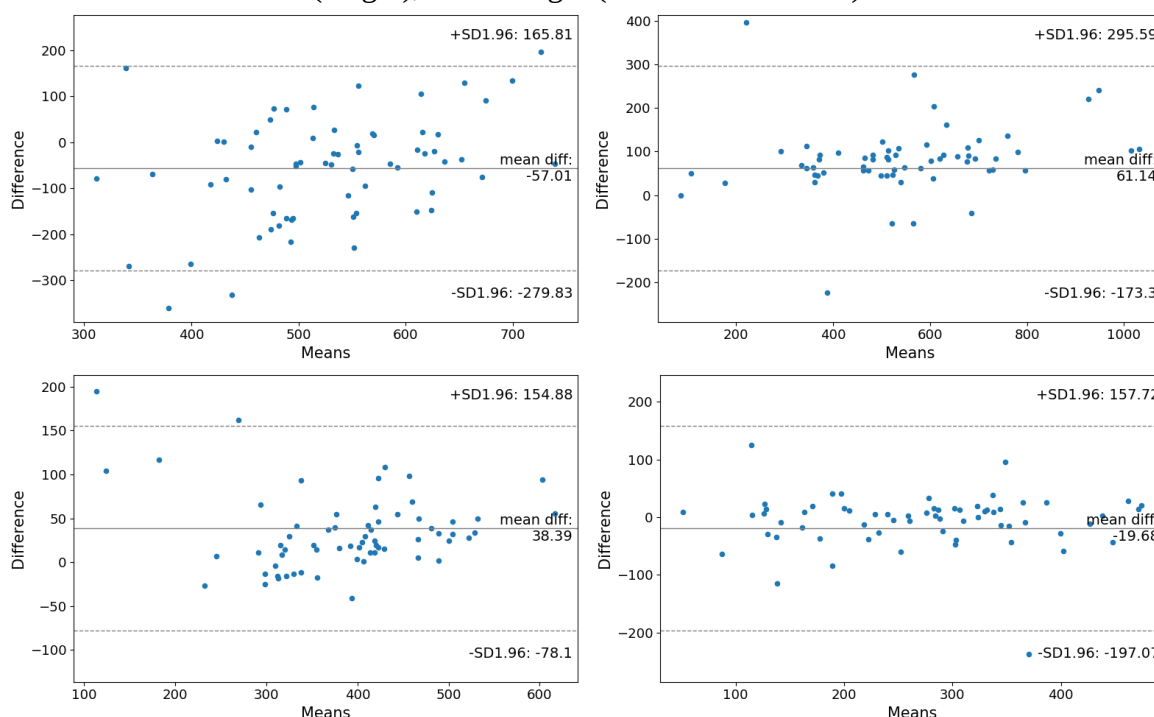
	Top diameter	Bottom diameter	Height	Minimum diameter
Proposed method	95.2 $\pm$ 79.2	106.8 $\pm$ 83.2	44.6 $\pm$ 55.4	41.9 $\pm$ 83

Source: Author.

The Bland-Altman method was used to calculate the mean and SD error of the unsigned errors in micrometers for each measurement. The Bland-Altman graph shows the

mean error on the horizontal axis and the standard deviation on the vertical axis. Other important information is also shown. There are three horizontal lines inside the graph, the center line is the mean error, and the other two are the maximum deviations given as references. The Bland-Altman graphs for the MH measurement errors are shown in Figure 47.

Figure 47: Bland-Altman graphs for the mean and SD of signed error of MH measurements (μm). Top left (top diameter), top right (bottom diameter), bottom left (height), bottom right (minimum diameter)



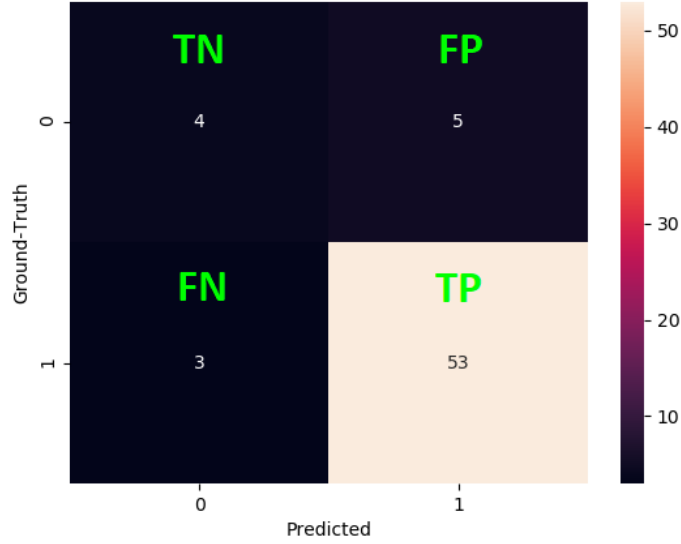
Source: Author.

An important metric to decide on a hole closure surgery is the Macular Hole Index, which is defined as the ratio of the height to the base diameter. MHI patients are classified into two classes. If the index is greater than 0.5 the surgery is possible to be performed without problems, however, if the index is less than 0.5 the possibility of closing the macular hole is less, so the surgery is less likely to work effectively. However, if closure is decided upon then in addition to the conventional technique, a miniotic membrane is applied in the MH, to assist in the closure.

In order to generate a classification result by comparing the MHI of the GT measures and the MHI of the predictive measures, values of MHI above 0.5 receive the number 1, which will identify the possibility to perform a hole closure surgery, otherwise, the value 0 will be assigned, informing that surgery is not possible.

The confusion matrix in Figure 48 shows where the algorithm went right and wrong more clearly. Knowing the trend of the results of the proposed method is important. In order to evaluate the MHI, two metrics are used: accuracy and IoU. Table 8 shows the evaluation metric results. Section 5.7 discusses these results.

Figure 48: Confusion matrix of MHI



Source: Author.

Table 8: Evaluation metric results for MHI classification in percentage

	Accuracy	IoU
Proposed method	87.69	92.98

Source: Author.

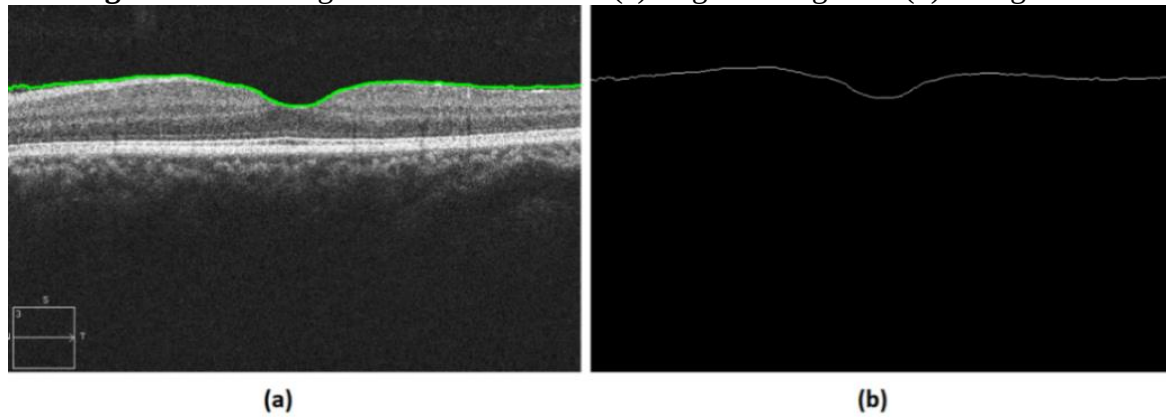
5.4 Results for ILM boundary layer segmentation without pathology

The proposed method achieved promising results for ILM boundary layer segmentation with MH pathology. Thus, it was decided to test the algorithm in OCT images without any pathology. As expected, the results were also satisfactory. Figure 49 shows the results for an NM.

The evaluation metrics used are mean and standard deviation of the unsigned error. Other works use these metrics for this type of segmentation. Boxplots with results are shown in Figure 50.

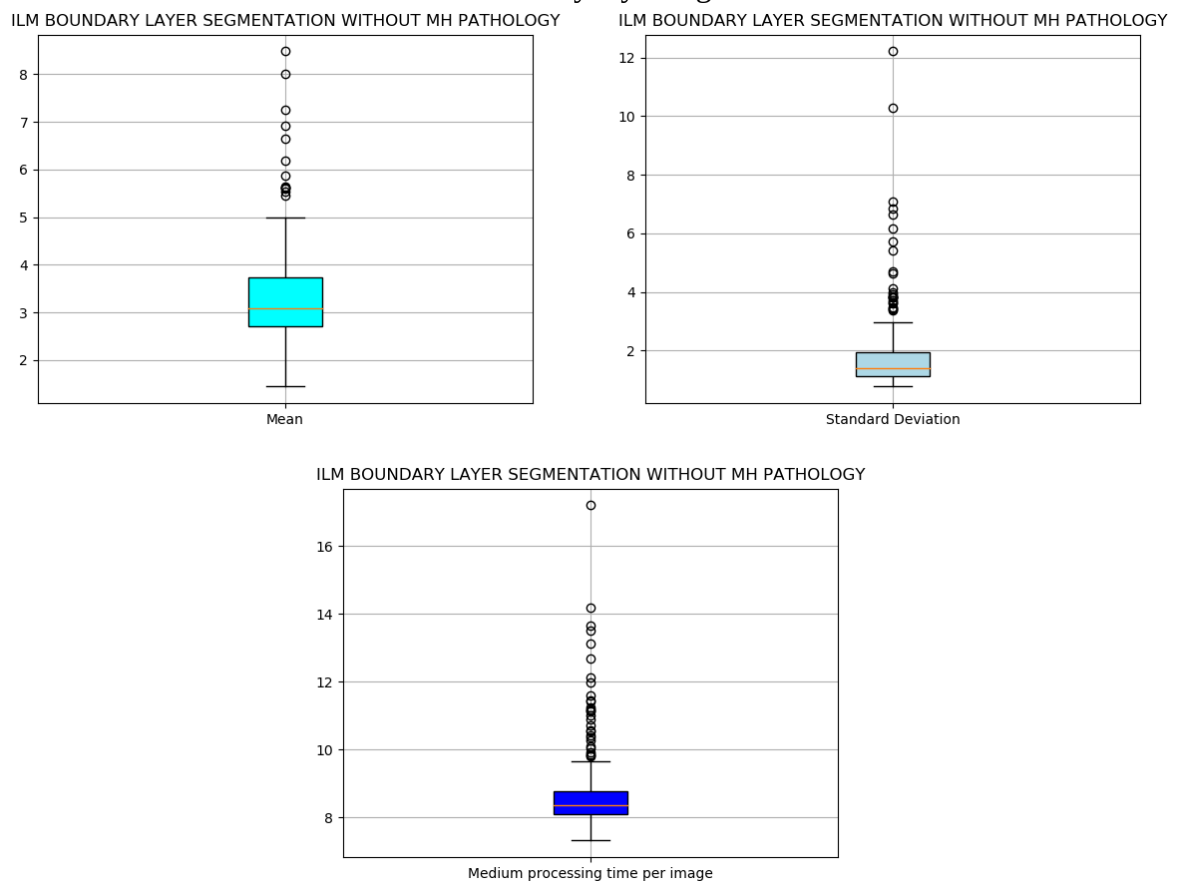
Considering the 206 NM images, there were only a few outliers in the boxplots. This may have been due to some points outside the curve that were not completely eliminated by the preprocessing techniques.

Figure 49: ILM segmentation for an NM (a) original image and (b) background



Source: Adapted from OCTID (2018).

Figure 50: Boxplots of mean and standard deviation (pixel) and processing time (s) for the evaluations of ILM boundary layer segmentation with NM



Source: Author.

The capacity of the ILM boundary layer segmentation without any pathology paves the way for a new stage of the algorithm to classify OCT images into the conditions of NM or MH. This can be developed in a future work.

The prediction segmentation is compared with the GT generated by a specialist and the results are shown in Table 9. These results are competitive with the state-of-the-art

method, as can be seen in Section 5.8. The medium processing time per image in the proposed method was 8.27s (minimum and maximum processing time per image, respectively: 7.27s and 17.32s).

Table 9: Mean and SD of UE (pixel) and medium processing time per image (s) for ILM boundary layer segmentation with NM

Method	Mean and SD	Proces. Time per image (s)
Proposed method	3.38 ± 1.89	8.27

Source: Author.

5.5 Discussion of ILM boundary layer segmentation with MH pathology

The proposed work is compared with a state-of-the-art method in order to validate and prove its effectiveness. A comparison of mean and SD of UE in pixels for the two methods is described in Table 10, as well the comparison between the medium processing time per image and the number of images used for both methods. Keller *et al.* (2016) used a private database, while the proposed method used a public one. However, the images are similar and have the same characteristics, and thus do not affect the veracity of the comparison.

Table 10: Mean and SD of UE comparison (pixel) and medium processing time per image (s) of ILM boundary layer segmentation with MH pathology

Method	Mean and SD	Proces. Time per image (s)	Quant. images
Keller <i>et al.</i> (2016)	3.4 ± 5.74	2.65	50
Proposed method	2.72 ± 1.91	0.24	65

Source: Author.

The mean and SD distance between each pixel location of the GT and the shortest point of the prediction contour segmentation was performed for both methods. The mean and SD of UE was better in the proposed method. In the state-of-the-art method, the SD was greater than the mean; possibly this was because the algorithm confused objects outside the contour, and these objects appear due to a failure in the noise removal techniques. This was one of the difficulties found during the development of the proposed algorithm and motivated us to spend more time processing images before the segmentation.

The proposed method had a medium processing time per image of 0.24s (minimum and maximum processing time per image, respectively: 0.17s and 0.37s). The number of pixels that go through the mathematical operation of finding the shortest distance to a curve

can influence the variation in the processing time per image. That is, the longer the contour, the longer the processing time.

Although the proposed method uses more preprocessing techniques than the state-of-the-art method being compared, the processing time is shorter. Moreover, the computer used by Keller *et al.* (2016) (Intel Core i7-4930K 3.4 GHz CPU) has a slight processing advantage over the one used in this work.

Another advantage of the proposed work is that MH measurements were made. Keller *et al.* (2016) mentioned that the data obtained could be used to estimate parameters such as macular hole height and base width. However, they did not perform any such calculations.

Therefore, the proposed work has significant importance to the scientific community in ILM boundary layer segmentation of OCT images with MH pathology due to the satisfactory results. These results are better than the state-of-the-art method with which it was compared. In addition, in the literature, as far as the author know, there is only one work in ILM segmentation of OCT images that is directly focused on MH pathology.

5.6 Discussions of MH area segmentation

Only one work providing a similar segmentation of MH was found in the literature. However, this work approach a volumetric segmentation, requiring a larger number of images. These images must be of the same retina in order to form the volumetric segmentation. This is a favorable factor, since all images contain the same settings. However, there is no public database providing images of this type, especially for MH pathology.

Therefore, a comparison between this state-of-the-art method and the proposed one, requires some clarification. First of all the comparison is possible because to perform a volumetric segmentation it is necessary to perform several segmentations of area and then join them. However, the number of images is considerably greater for volumetric segmentations. The proposed work used only a public database that does not contain a large number of images. Thus, the processing time cannot be compared. Although the databases are different the images are similar and have the same characteristics, which does not affect the veracity of the comparison.

A comparison of IoU, and DSC for the methods is described in Table 11. The MCC metric was used for the proposed method, however, it was not used for the one being

compared. Pixel accuracy was not compared because even if the result was bad the accuracy remains high due to the number of TN pixels.

The methods achieved equivalent results. The MCC method, despite being considered one of the most appropriate and fair metrics to evaluate the effectiveness of an area segmentation algorithm, was not used by the compared state-of-the-art method. The medium processing time per image in the proposed method was of 19.95s (minimum and maximum processing time per image, respectively: 12.7s and 45.5s). The size of the MH area can influence the variation in processing time per image.

Table 11: IoU, DSC and MCC comparison in percentage of MH area segmentation

Method	IoU	DSC
Nasrulloh <i>et al.</i> (2017)	76.34 ± 10.31	89.16 ± 7.55
Proposed method	78.61	86.98

Source: Author.

5.7 Discussions of measurements of macular holes

The results for the mean and SD of UE for the MH measurements show satisfactory values. This proves that in addition to performing two types of segmentation, the algorithm is also able to take measurements effectively. No measurement mean error was greater than $107 \mu\text{m}$. The coherence of the results for the Bland-Altman graphics is also a good tool to analyze the capacity of the algorithm to stay within the standards results for all images.

Another important parameter is the Macular Hole Index. These results were satisfactory and showed that the algorithm can also be evaluated through an equation that relates the ability of the algorithm to perform a simple classification, without the need for more robust techniques, such as machine learning applications.

5.8 Discussions of ILM boundary layer segmentation without any pathology

The effectiveness of the algorithm for a ILM boundary layer segmentation without any pathology was demonstrated when compared with some state-of-the-art methods. A comparison of the results among these techniques is shown in Table 12.

Although an accurate comparison is extremely complicated, given each state-of-the-art method was validated under different image conditions and settings, the results, at a pixel-wise level, achieved by the proposed methodology demonstrate a competitive performance

in relation to the other works. Furthermore, the proposed method does not require training or the use of a large number of OCT images, and its implementation is simpler.

Table 12: Mean and SD of UE (pixel) for ILM boundary layer segmentation with NM

Method	Mean and SD
Duan <i>et al.</i> (2017)	0.23 ± 0.06
Hu <i>et al.</i> (2019)	0.82
Hussain <i>et al.</i> (2017)	0.97 ± 0.24
	1.07 ± 0.17
González-López <i>et al.</i> (2019)	1.27 ± 0.82
Stankiewicz <i>et al.</i> (2017)	14.74 ± 3.66
	8.5 ± 2.82
Gopinath <i>et al.</i> (2017)	1.11 ± 0.3
	1.49 ± 0.47
Lang <i>et al.</i> (2017)	0.65 ± 0.18
Niu <i>et al.</i> (2014)	0.71 ± 0.54
Tian <i>et al.</i> (2015)	0.71
Proposed method	3.38 ± 1.89

Source: Author.

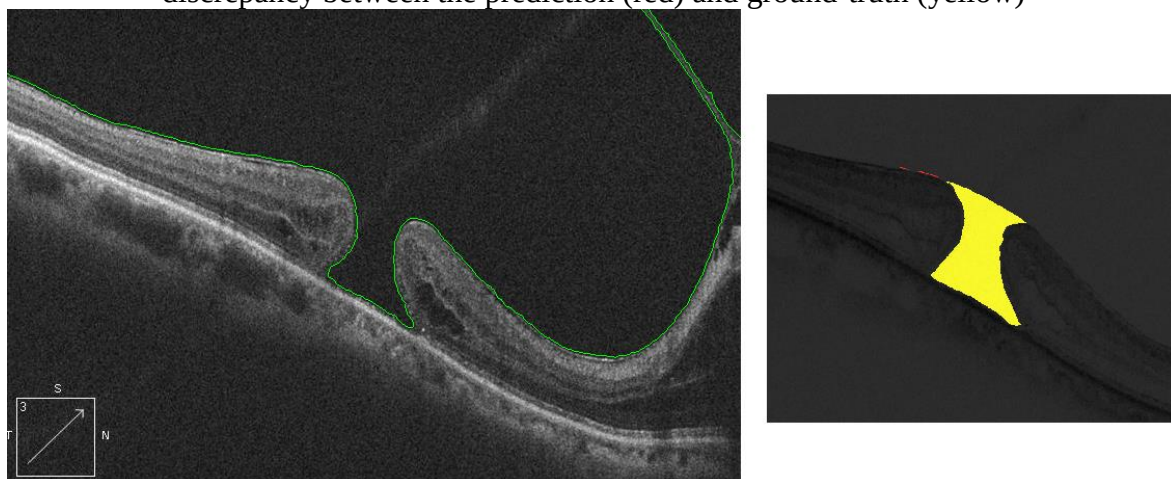
The capacity of ILM boundary layer segmentation with and without MH pathology paves the way for a new stage of this algorithm, which can be developed in future works to classify OCT images for NM or MH.

5.9 Limitations of the proposed method

The results of the proposed work were satisfactory, reaching both the main objective and the specific objectives defined in the introduction through the validation techniques adopted. However, there are also some limitations. As an example, in image MH 14, the algorithm was unable to complete the segmentation, which compromised the macular hole measurements. Also the segmentation error of image MH 14 can be seen in Figure 51.

The explanation for the error is that the contour of the ILM boundary layer is quite curved making the execution of the shortest distance from a point to a curve (trying to find the two points at the top of the image to close the area) to find points not consistent with the closing of the MH.

Figure 51: Contour segmentation (left) and confusion matrix of pixels (right) show a discrepancy between the prediction (red) and ground-truth (yellow)



Source: Author.

6 CONCLUSIONS

The main objective of this work, which was the development of an algorithm that automatically segmented the ILM boundary layer and the area of the MH pathology in OCT images, was reached. In addition, other objectives were also achieved: the automatic acquisition of MH measurements; and segmenting the ILM boundary layer for images with NM and validating the results, proving that the algorithm can also be used for NM.

The differential of the proposed work, is the set of techniques, the mathematical simplicity involved and the ease of application. The proposed model is capable of performing the same function as a more complex technique, a machine learning method for example, with lower computational cost. The shortest distance from a point to a curve (Euclidean Distance), Flood Fill and Border Following algorithms used here are consolidated tools, which can be implemented in any programming language.

The segmentation of an ILM boundary layer with a MH pathology is part of the first step of the algorithm. Then it is necessary to calculate the distance error of the shortest pixels to the GT contour, with the Euclidean Distance. The proposed work was compared with a state-of-the-art method in order to validate and prove its effectiveness. The proposed method had better results.

Macular hole area segmentation is important as this gives the shape and measurements of the hole. The proposed method and the compared one achieved equivalent results.

The algorithm also obtained the measurements of MH. These results were evaluated by comparing the algorithm results (prediction) and the ground-truth. The results were satisfactory, thus proving that in addition to performing two types of segmentation the algorithm is also capable of taking measurements effectively.

The algorithm was tested in OCT images without any pathology. As expected, the results were also satisfactory. All the OCT images with NM in the database were used. The proposed method was compared with nine other methods. The results achieved by the proposed methodology demonstrated a competitive performance in relation to the other works.

The proposed work also performed two types of segmentation (contour and area) in images with MH pathology. In addition it extracted important measures of MH, and the

algorithm was also applied to NM images. The proposed work achieved satisfactory results for all applications.

6.1 Contributions

The main contribution of this work is the importance of this automatic method to segment OCT images and help the specialists in this area. Furthermore, advances in research involving this area can contribute to the acceptance of automatic techniques in medical practices. This work may be a study source for researchers, clinicians and engineers wishing to delve into MH segmentation techniques.

The proposed method can help specialists to make accurate diagnostic decisions. The measurements of MH are fundamental to know if the case requires only treatment or if is possible to perform surgery.

Although most research involving retinal problems is concerned with the more common pathologies, such as CRS, AMD, and ME, Macular Holes affect a large portion of the population over forty years of age. OCT images can provide different parameters and thus contribute to patient diagnoses as well as to academic research. A fast and low cost segmentation technique is interesting to help these experts with retinal layer analyses.

The differential of this set of techniques is the mathematical simplicity involved and the ease of application. It is capable of performing the same function as a more complex technique with a lower computational cost. Furthermore, these techniques can be implemented in any programming language.

6.2 Future Prospects

Further research is necessary to find new approaches or to improve the existing ones. A closer relationship between the medical community and researchers strengthens such improvements. Future prospects that will encourage the use of MH segmentation systems as proposed here in daily medical practices, are:

- application of the approach to other pathologies;
- development of a classification algorithm for MH and NM conditions;

- creation of a publicly database of OCT images with MH pathology containing ground-truth of the contour and area, besides feature information to help researchers;
- application of an embedded system in a mobile device;
- use of machine and deep learning techniques to improve the algorithms;
- application of the approach to slices of B-scan images from the same MH, allowing 3D segmentation representation.

REFERENCES

AL-AMRI, Salem Saleh et al. Image segmentation by using threshold techniques. **arXiv preprint arXiv:1005.4020**, 2010.

ALEXANDRIA, Auzuir Ripardo et al. Método de contornos ativos pSnakes aplicado em imagens de ecocardiograma de eixo curto para cálculo de fração de ejeção. **Revista Brasileira de Engenharia Biomédica**, v. 27, n. 3, p. 147-62, 2011.

ALTAWHEEL, Michael; IP, Michael. Macular hole: improved understanding of pathogenesis, staging, and management based on optical coherence tomography. In: **Seminars in ophthalmology**. Taylor & Francis, 2003. p. 58-66.

ANDREOPOULOS, Alexander; TSOTSOS, John K. 50 years of object recognition: Directions forward. **Computer vision and image understanding**, v. 117, n. 8, p. 827-891, 2013.

AYACHE, Nicholas. Medical computer vision, virtual reality and robotics. **Image and Vision Computing**, v. 13, n. 4, p. 295-313, 1995.

BORN, Max; WOLF, Emil. Principles of Optics, 7th (expanded) edition. **United Kingdom: Press Syndicate of the University of Cambridge**, v. 461, 1999.

BREIMAN, Leo. Random forests. **Machine learning**, v. 45, n. 1, p. 5-32, 2001.

BURACO macular. **Instituto de Retina**. 2020. Available in: <<https://www.institutoderetina.com.br/home/buraco-macular/#:~:text=Buraco%20de%20M%C3%A1cula%20%C3%A9%20uma,vis%C3%A3o%2C%20levando%20a%20redu%C3%A7%C3%A3o%20visual>>. Access in 6.10.2020.

CHEN, Huaiguang et al. Speckle reduction based on fractional-order filtering and boosted singular value shrinkage for optical coherence tomography image. **Biomedical Signal Processing and Control**, v. 52, p. 281-292, 2019.

CHENG, Heng-Da et al. Color image segmentation: advances and prospects. **Pattern recognition**, v. 34, n. 12, p. 2259-2281, 2001.

CHICCO, Davide; JURMAN, Giuseppe. The advantages of the Matthews correlation coefficient (MCC) over F1 score and accuracy in binary classification evaluation. **BMC genomics**, v. 21, n. 1, p. 6, 2020.

CHIU, Stephanie J. et al. Validated automatic segmentation of AMD pathology including drusen and geographic atrophy in SD-OCT images. **Investigative ophthalmology & visual science**, v. 53, n. 1, p. 53-61, 2012.

CHHABLANI, Jay et al. Role of macular hole angle in macular hole closure. **British Journal of Ophthalmology**, v. 99, n. 12, p. 1634-1638, 2015.

COORDINATE System and Units. **Adafruit**. 2020. Available in: <<https://learn.adafruit.com/adafruit-gfx-graphics-library/coordinate-system-and-units>>. Access in 03.19.2020.

COSCAS, Gabriel; CUNHA-VAZ, José; SOUBRANE, Gisèle. Macular edema: definition and basic concepts. In: **Macular Edema**. Karger Publishers, 2010. p. 1-9.

DA SILVA FELIX, John Hebert et al. AUTOIN: method of automatic initialization of active contours applied to lungs in CT images. **IEEE Latin America Transactions**, v. 10, n. 4, p. 1954-1960, 2012.

DE ALEXANDRIA, Auzuir Ripardo et al. pSnakes: A new radial active contour model and its application in the segmentation of the left ventricle from echocardiographic images. **Computer methods and programs in biomedicine**, v. 116, n. 3, p. 260-273, 2014.

DE BUSTROS, Serge et al. Vitrectomy for idiopathic epiretinal membranes causing macular pucker. **British Journal of Ophthalmology**, v. 72, n. 9, p. 692-695, 1988.

DE MORAES, Jermana Lopes et al. Stratification of cardiopathies using photoplethysmographic signals. **Informatics in Medicine Unlocked**, v. 20, p. 100417, 2020.

DE MOURA, Joaquim et al. Automatic characterization of the serous retinal detachment associated with the subretinal fluid presence in optical coherence tomography images. **Procedia Computer Science**, v. 126, p. 244-253, 2018.

DE MUL, Marleen; DE BONT, Antoinette; BERG, Marc. IT-supported skill-mix change and standardisation in integrated eyecare: lessons from two screening projects in The Netherlands. **International journal of integrated care**, v. 7, n. 2, 2007.

DE QUEIROZ, José Eustáquio Rangel; GOMES, Herman Martins. Introdução ao processamento digital de imagens. **Rita**, v. 13, n. 2, p. 11-42, 2006.

DE SOUZA ALVES, Thamiris et al. From knowledge based vision systems to cognitive vision systems: a review. **Procedia Computer Science**, v. 126, p. 1855-1864, 2018.

DESJARDINS, A. E. et al. Speckle reduction in OCT using massively-parallel detection and frequency-domain ranging. **Optics express**, v. 14, n. 11, p. 4736-4745, 2006.

DODO, Bashir I. et al. Retinal oct segmentation using fuzzy region competition and level set methods. In: **2019 IEEE 32nd International Symposium on Computer-Based Medical Systems (CBMS)**. IEEE, 2019. p. 93-98.

DREXLER, Wolfgang et al. Enhanced visualization of macular pathology with the use of ultrahigh-resolution optical coherence tomography. **Archives of ophthalmology**, v. 121, n. 5, p. 695-706, 2003.

DU, Cheng-Jin; SUN, Da-Wen. Learning techniques used in computer vision for food quality evaluation: a review. **Journal of food engineering**, v. 72, n. 1, p. 39-55, 2006.

DUAN, Jinming et al. Optical coherence tomography image segmentation. In: **2015 IEEE International Conference on Image Processing (ICIP)**. IEEE, 2015. p. 4278-4282.

DUAN, Jinming et al. Automated segmentation of retinal layers from optical coherence tomography images using geodesic distance. **Pattern Recognition**, v. 72, p. 158-175, 2017.

EL TANBOLY, Ahmed et al. A novel automatic segmentation of healthy and diseased retinal layers from OCT scans. In: **2016 IEEE International Conference on Image Processing (ICIP)**. IEEE, 2016. p. 116-120.

EYE conditions. **Florida Eye Clinic**. 2020. Available in: <<https://www.floridaeyeclinic.com/eye-conditions/eye-conditions-2/>>. Access in 03.19.2020.

FABRITIUS, Tapio et al. Automated segmentation of the macula by optical coherence tomography. **Optics express**, v. 17, n. 18, p. 15659-15669, 2009.

FAGHIHI, Hooshang et al. Spontaneous closure of traumatic macular holes. **Canadian Journal of Ophthalmology**, v. 49, n. 4, p. 395-398, 2014.

FANG, Leyuan et al. Iterative fusion convolutional neural networks for classification of optical coherence tomography images. **Journal of Visual Communication and Image Representation**, v. 59, p. 327-333, 2019.

FERNÁNDEZ, Delia Cabrera; SALINAS, Harry M.; PULIAFITO, Carmen A. Automated detection of retinal layer structures on optical coherence tomography images. **Optics express**, v. 13, n. 25, p. 10200-10216, 2005.

FOOS, Robert Y.; WHEELER, Noel C. Vitreoretinal juncture: synchysis senilis and posterior vitreous detachment. **Ophthalmology**, v. 89, n. 12, p. 1502-1512, 1982.

FORSYTH, David A.; PONCE, Jean. **Computer vision: a modern approach**. Prentice Hall Professional Technical Reference, 2002.

FORTE, R. et al. Comparison of time domain Stratus OCT and spectral domain SLO/OCT for assessment of macular thickness and volume. **Eye**, v. 23, n. 11, p. 2071-2078, 2009.

FREEMAN, William R. et al. Vitrectomy for the treatment of full-thickness stage 3 or 4 macular holes: results of a multicentered randomized clinical trial. **Archives of Ophthalmology**, v. 115, n. 1, p. 11-21, 1997.

FRISINA, Rino; PILOTTO, Elisabetta; MIDENA, Edoardo. Lamellar macular hole: state of the art. **Ophthalmic research**, v. 61, n. 2, p. 73-82, 2019.

FU, King-Sun; MUI, J. K. A survey on image segmentation. **Pattern recognition**, v. 13, n. 1, p. 3-16, 1981.

GEORGE, Neetha; JIJ, C. V. Two stage contour evolution for automatic segmentation of choroid and cornea in OCT images. **Biocybernetics and Biomedical Engineering**, v. 39, n. 3, p. 686-696, 2019.

GERENDAS, Bianca S. et al. Correlation between morphological characteristics in spectral-domain-optical coherence tomography, different functional tests and a patient's subjective handicap in acute central serous chorioretinopathy. **Acta ophthalmologica**, v. 96, n. 7, p. e776-e782, 2018.

GHOLAMI, Peyman et al. OCTID: Optical coherence tomography image database. **Computers & Electrical Engineering**, v. 81, p. 106532, 2020.

GIANCARDIO, Luca et al. Exudate-based diabetic macular edema detection in fundus images using publicly available datasets. **Medical image analysis**, v. 16, n. 1, p. 216-226, 2012.

GIRISH, G. N.; KOTHARI, Abhishek R.; RAJAN, Jeny. Automated segmentation of intra-retinal cysts from optical coherence tomography scans using marker controlled watershed

transform. In: **2016 38th Annual International Conference of the IEEE Engineering in Medicine and Biology Society (EMBC)**. IEEE, 2016. p. 1292-1295.

GONG, Guanghua; ZHANG, Hongming; YAO, Minyu. Speckle noise reduction algorithm with total variation regularization in optical coherence tomography. **Optics express**, v. 23, n. 19, p. 24699-24712, 2015.

GONZALEZ, Rafael C.; WOODS, Richard Eugene; EDDINS, Steven L. **Digital image processing using MATLAB**. Pearson Education India, 2004.

GONZÁLEZ-LÓPEZ, A. et al. Robust segmentation of retinal layers in optical coherence tomography images based on a multistage active contour model. **Heliyon**, v. 5, n. 2, p. e01271, 2019.

GOPINATH, Karthik; RANGREJ, Samrudhdi B.; SIVASWAMY, Jayanthi. A deep learning framework for segmentation of retinal layers from OCT images. In: **2017 4th IAPR Asian Conference on Pattern Recognition (ACPR)**. IEEE, 2017. p. 888-893.

GRYCUK, Rafał et al. Improved digital image segmentation based on stereo vision and mean shift algorithm. In: **International Conference on Parallel Processing and Applied Mathematics**. Springer, Berlin, Heidelberg, 2013. p. 433-443.

GRZYWACZ, Norberto Mauricio et al. Statistics of optical coherence tomography data from human retina. **IEEE transactions on medical imaging**, v. 29, n. 6, p. 1224-1237, 2010.

GUPTA, Vishali; GUPTA, Amod; DOGRA, Mangat R. **Atlas of optical coherence tomography of macular diseases**. CRC Press, 2004.

HALLER, Julia A. et al. Efficacy of intravitreal ocriplasmin for treatment of vitreomacular adhesion: subgroup analyses from two randomized trials. **Ophthalmology**, v. 122, n. 1, p. 117-122, 2015.

HAOUCHINE, Belkacem et al. Diagnosis of macular pseudoholes and lamellar macular holes by optical coherence tomography. **American journal of ophthalmology**, v. 138, n. 5, p. 732-739, 2004.

HARITOGLOU, Christos et al. Macular changes after peeling of the internal limiting membrane in macular hole surgery. **American journal of ophthalmology**, v. 132, n. 3, p. 363-368, 2001.

HASSAN, Bilal et al. Structure tensor based automated detection of macular edema and central serous retinopathy using optical coherence tomography images. **JOSA A**, v. 33, n. 4, p. 455-463, 2016.

HASSAN, Bilal; AHMED, Ramsha; LI, Bo. Automated foveal detection in OCT scans. In: **2018 IEEE International Symposium on Signal Processing and Information Technology (ISSPIT)**. IEEE, 2018. p. 419-422.

HE, H.-J.; ZHENG, C.; SUN, D.-W. Image segmentation techniques. In: **Computer Vision Technology for Food Quality Evaluation**. Academic Press, 2016. p. 45-63.

HEARN, Donald et al. **Computer graphics with OpenGL**. Upper Saddle River, NJ: Pearson Prentice Hall,, 2004.

HO, Joseph et al. Assessment of artifacts and reproducibility across spectral-and time-domain optical coherence tomography devices. **Ophthalmology**, v. 116, n. 10, p. 1960-1970, 2009.

HOOD, Donald C. et al. Structure versus function in glaucoma: an application of a linear model. **Investigative ophthalmology & visual science**, v. 48, n. 8, p. 3662-3668, 2007.

HOROWITZ, Steven L. Picture segmentation by a directed split-and-merge procedure. In: **IJCPR**. 1974. p. 424-433.

HSU, Chi-Yu; DING, Jian-Jiun. Efficient image segmentation algorithm using SLIC superpixels and boundary-focused region merging. In: **2013 9th International Conference on Information, Communications & Signal Processing**. IEEE, 2013. p. 1-5.

HU, Junjie; CHEN, Yuanyuan; YI, Zhang. Automated segmentation of macular edema in OCT using deep neural networks. **Medical image analysis**, v. 55, p. 216-227, 2019.

HU, Kai et al. Automatic segmentation of retinal layer boundaries in OCT images using multiscale convolutional neural network and graph search. **Neurocomputing**, v. 365, p. 302-313, 2019.

HUANG, David et al. Optical coherence tomography. **science**, v. 254, n. 5035, p. 1178-1181, 1991.

HUGHES, Michael; SPRING, Marika; PODOLEANU, Adrian. Speckle noise reduction in optical coherence tomography of paint layers. **Applied optics**, v. 49, n. 1, p. 99-107, 2010.

HUSSAIN, Md Akter; BHUIYAN, Alauddin; RAMAMOHANARAO, Kotagiri. Disc segmentation and BMO-MRW measurement from SD-OCT image using graph search and tracing of three bench mark reference layers of retina. In: **2015 IEEE International Conference on Image Processing (ICIP)**. IEEE, 2015. p. 4087-4091.

HUSSAIN, Md Akter et al. Automatic identification of pathology-distorted retinal layer boundaries using SD-OCT imaging. **IEEE Transactions on Biomedical Engineering**, v. 64, n. 7, p. 1638-1649, 2016.

HUSSEIN, Walid B. et al. A novel edge detection method with application to the fat content prediction in marbled meat. **Pattern recognition**, v. 44, n. 12, p. 2959-2970, 2011.

IFTIMIA, Nicusor; BOUMA, Brett E.; TEARNEY, Guillermo J. Speckle reduction in optical coherence tomography by "path length encoded" angular compounding. **Journal of biomedical optics**, v. 8, n. 2, p. 260-263, 2003.

IKEBE, Masayuki; ASAI, Tetsuya. A Digital Vision Chip for Early Feature extraction with rotated template-matching CA. **Journal of Robotics and Mechatronics**, v. 17, n. 4, p. 372, 2005.

IP, Michael S. et al. Anatomical outcomes of surgery for idiopathic macular hole as determined by optical coherence tomography. **Archives of Ophthalmology**, v. 120, n. 1, p. 29-35, 2002.

JAIN, Anil K. **Fundamentals of digital image processing**. Prentice-Hall, Inc., 1989.

JAIN, Ramesh; KASTURI, Rangachar; SCHUNCK, Brian G. **Machine vision**. New York: McGraw-hill, 1995.

JØRGENSEN, Thomas Martini et al. Enhancing the signal-to-noise ratio in ophthalmic optical coherence tomography by image registration—method and clinical examples. **Journal of biomedical optics**, v. 12, n. 4, p. 041208, 2007.

JOUSSEN, Antonia M. et al. (Ed.). **Retinal vascular disease**. Heidelberg: Springer, 2007.

JACQUES JUNIOR, J. C. S. **Processamento de imagens: fundamentos**. Available in: <https://www.inf.pucrs.br/~smusse/Simulacao/PDFs/aula_02_Fundamentos_PI.pdf>. Access in 3.25.2020.

- KELLER, Brenton et al. Length-adaptive graph search for automatic segmentation of pathological features in optical coherence tomography images. **Journal of biomedical optics**, v. 21, n. 7, p. 076015, 2016.
- KLEIN, Ronald; KLEIN, Barbara EK; LINTON, Kathryn LP. Prevalence of age-related maculopathy: the Beaver Dam Eye Study. **Ophthalmology**, v. 99, n. 6, p. 933-943, 1992.
- KOIZUMI, Hideki et al. Three-dimensional evaluation of vitreomacular traction and epiretinal membrane using spectral-domain optical coherence tomography. **American journal of ophthalmology**, v. 145, n. 3, p. 509-517. e1, 2008.
- KONG, Mingui et al. Measurable range of subfoveal choroidal thickness with conventional spectral domain optical coherence tomography. **Translational vision science & technology**, v. 7, n. 5, p. 16-16, 2018.
- KREBS, I. et al. Optical coherence tomography: limits of the retinal-mapping program in age-related macular degeneration. **British Journal of Ophthalmology**, v. 92, n. 7, p. 933-935, 2008.
- KUSUHARA, Sentaro et al. Prediction of postoperative visual outcome based on hole configuration by optical coherence tomography in eyes with idiopathic macular holes. **American journal of ophthalmology**, v. 138, n. 5, p. 709-716, 2004.
- LANG, Andrew et al. Improving graph-based OCT segmentation for severe pathology in Retinitis Pigmentosa patients. In: **Medical Imaging 2017: Biomedical Applications in Molecular, Structural, and Functional Imaging**. International Society for Optics and Photonics, 2017. p. 101371M.
- LEE, Cecilia S. et al. Deep-learning based, automated segmentation of macular edema in optical coherence tomography. **Biomedical optics express**, v. 8, n. 7, p. 3440-3448, 2017.
- LI, X. D. et al. Diffraction tomography for biochemical imaging with diffuse-photon density waves. **Optics Letters**, v. 22, n. 8, p. 573-575, 1997.
- LINDENMAIER, Andras A. et al. Texture analysis of optical coherence tomography speckle for characterizing biological tissues in vivo. **Optics letters**, v. 38, n. 8, p. 1280-1282, 2013.
- LU, Donghuan et al. Retinal fluid segmentation and detection in optical coherence tomography images using fully convolutional neural network. **arXiv preprint arXiv:1710.04778**, 2017.

LUCKIE, Alan; HERIOT, Wilson. Macular holes: pathogenesis, natural history and surgical outcomes. **Australian and New Zealand journal of ophthalmology**, v. 23, n. 2, p. 93-100, 1995.

LV, Hongli et al. Speckle noise reduction for optical coherence tomography based on adaptive 2D dictionary. **Laser Physics Letters**, v. 15, n. 5, p. 055401, 2018.

MADI, Haifa A.; MASRI, Ibrahim; STEEL, David H. Optimal management of idiopathic macular holes. **Clinical ophthalmology (Auckland, NZ)**, v. 10, p. 97, 2016.

MARGHERIO, Raymond R. et al. Surgical management of vitreomacular traction syndromes. **Ophthalmology**, v. 96, n. 9, p. 1437-1445, 1989.

MARRUGO, Andrés G. et al. Image analysis in modern ophthalmology: from acquisition to computer assisted diagnosis and telemedicine. In: **Optics, Photonics, and Digital Technologies for Multimedia Applications II**. International Society for Optics and Photonics, 2012. p. 84360C.

MATTHEWS Correlation Coefficient Is The Best Classification Metric You've Never Heard Of. **Towards Data Science**. 2019. Available in: <<https://towardsdatascience.com/the-best-classification-metric-youve-never-heard-of-the-matthews-correlation-coefficient-3bf50a2f3e9a>>. Access in 4.14.2020.

MAURYA, Prashant Kumar et al. Automated detection of diabetic macular edema involving cystoids and serous retinal detachment. **Optics & Laser Technology**, v. 127, p. 106157, 2020.

MENDES, Odilon Linhares C. et al. Automatic Segmentation of Macular Holes in Optical Coherence Tomography Images: A review. **Journal of Artificial Intelligence and Systems**, v. 1, n. 1, p. 163-185, 2020.

MENDES, Odilon Linhares Carvalho; BORILLE, Giovanna Miceli Ronzani. Computer vision systems in unmanned aerial vehicle: a review. **Journal of Mechatronics Engineering**, v. 2, n. 2, p. 11-22, 2019.

METRICS to Evaluate your Semantic Segmentation Model. **Towards Data Science**. 2019. Available in: <<https://towardsdatascience.com/metrics-to-evaluate-your-semantic-segmentation-model-6bcb99639aa2>>. Access in 4.14.2020.

MICHEL, R. G.; WILKINSON, C. P.; RICE, T. A. Retinal detachment. **St Louis: CV Mosby**, v. 117, 1990.

MIRI, Mohammad Saleh et al. Incorporation of gradient vector flow field in a multimodal graph-theoretic approach for segmenting the internal limiting membrane from glaucomatous optic nerve head-centered SD-OCT volumes. **Computerized Medical Imaging and Graphics**, v. 55, p. 87-94, 2017.

MONTUORO, Alessio et al. Joint retinal layer and fluid segmentation in OCT scans of eyes with severe macular edema using unsupervised representation and auto-context. **Biomedical optics express**, v. 8, n. 3, p. 1874-1888, 2017.

MUTHUKRISHNAN, R.; RADHA, Miyilsamy. Edge detection techniques for image segmentation. **International Journal of Computer Science & Information Technology**, v. 3, n. 6, p. 259, 2011.

MYLONAS, Georgos et al. Comparison of retinal thickness measurements and segmentation performance of four different spectral and time domain OCT devices in neovascular age-related macular degeneration. **British journal of ophthalmology**, v. 93, n. 11, p. 1453-1460, 2009.

NASRULLOH, Amar V. et al. Multi-scale segmentation and surface fitting for measuring 3-D macular holes. **IEEE transactions on medical imaging**, v. 37, n. 2, p. 580-589, 2017.

NAVON, Ety; MILLER, Ofer; AVERBUCH, Amir. Color image segmentation based on adaptive local thresholds. **Image and vision computing**, v. 23, n. 1, p. 69-85, 2005.

NIU, Sijie et al. Automated retinal layers segmentation in SD-OCT images using dual-gradient and spatial correlation smoothness constraint. **Computers in biology and medicine**, v. 54, p. 116-128, 2014.

NARENDRA, Patrenahalli M. A separable median filter for image noise smoothing. **IEEE Transactions on Pattern Analysis and Machine Intelligence**, n. 1, p. 20-29, 1981.

NAZ, Samra; AKRAM, M. Usman; KHAN, Shoab A. Automated segmentation of retinal layers from OCT images using structure tensor and kernel regression+ GTDP approach. In: **2017 1st International Conference on Next Generation Computing Applications (NextComp)**. IEEE, 2017. p. 98-102.

NOVOSEL, Jelena et al. Joint segmentation of retinal layers and focal lesions in 3-D OCT data of topologically disrupted retinas. **IEEE transactions on medical imaging**, v. 36, n. 6, p. 1276-1286, 2017.

OCTID, Optical Coherence Tomography Image Database. **Scholars Portal Dataverse**. 2018. Available in: <https://dataverse.scholarsportal.info/dataset.xhtml?persistentId=doi:10.5683/SP/MBMQGD>. Access in 10.25.19.

PAL, Nikhil R.; PAL, Sankar K. A review on image segmentation techniques. **Pattern recognition**, v. 26, n. 9, p. 1277-1294, 1993.

PARK, Donald W. et al. Macular hole surgery with internal-limiting membrane peeling and intravitreal air. **Ophthalmology**, v. 106, n. 7, p. 1392-1398, 1999.

PEREIRA, Pedro MM et al. Image Segmentation using Gradient-based Histogram Thresholding for Skin Lesion Delineation. In: **BIOIMAGING**. 2019. p. 84-91.

PETRICK, Nicholas et al. Evaluation of computer-aided detection and diagnosis systems a. **Medical physics**, v. 40, n. 8, p. 087001, 2013.

PHILIPPAKIS, E. et al. Measurement of full-thickness macular hole size using en face optical coherence tomography. **Eye**, v. 32, n. 3, p. 590-596, 2018.

POTSAID, Benjamin et al. Ultrahigh speed spectral/Fourier domain OCT ophthalmic imaging at 70,000 to 312,500 axial scans per second. **Optics express**, v. 16, n. 19, p. 15149-15169, 2008.

RAMZAN, Aneeqa et al. Automated glaucoma detection using retinal layers segmentation and optic cup-to-disc ratio in optical coherence tomography images. **IET Image Processing**, v. 13, n. 3, p. 409-420, 2018.

RENCZ, Andrew N.; RYERSON, Robert A. (Ed.). **Manual of remote sensing, remote sensing for the earth sciences**. John Wiley & Sons, 1999.

RICCO, Susanna et al. Correcting motion artifacts in retinal spectral domain optical coherence tomography via image registration. In: **International Conference on Medical Image Computing and Computer-Assisted Intervention**. Springer, Berlin, Heidelberg, 2009. p. 100-107.

ROSENFELD, Azriel. Computer vision: basic principles. **Proceedings of the IEEE**, v. 76, n. 8, p. 863-868, 1988.

ROSSANT, Florence et al. Parallel Double Snakes. Application to the segmentation of retinal layers in 2D-OCT for pathological subjects. **Pattern Recognition**, v. 48, n. 12, p. 3857-3870, 2015.

ROY, Abhijit Guha et al. ReLayNet: retinal layer and fluid segmentation of macular optical coherence tomography using fully convolutional networks. **Biomedical optics express**, v. 8, n. 8, p. 3627-3642, 2017.

RUSS, John C. The image processing handbook. **SCANNING-NEW YORK AND BADEN BADEN THEN MAHWAH-**, v. 19, p. 60-60, 1997.

SADDA, Srinivas R. et al. Errors in retinal thickness measurements obtained by optical coherence tomography. **Ophthalmology**, v. 113, n. 2, p. 285-293, 2006.

SALARIAN, Mahdi et al. Accurate segmentation of retina nerve fiber layer in OCT images. In: **2015 IEEE International Conference on Electro/Information Technology (EIT)**. IEEE, 2015. p. 653-656.

SCHAAL, Karen B. et al. Vascular abnormalities in diabetic retinopathy assessed with swept-source optical coherence tomography angiography widefield imaging. **Retina**, v. 39, n. 1, p. 79-87, 2019.

SCHMITT, Joseph M.; XIANG, S. H.; YUNG, Kin Man. Speckle in optical coherence tomography. **Journal of biomedical optics**, v. 4, n. 1, p. 95-105, 1999.

SCHUMAN, Joel S. et al. Reproducibility of nerve fiber layer thickness measurements using optical coherence tomography. **Ophthalmology**, v. 103, n. 11, p. 1889-1898, 1996.

SCIKIT-IMAGE. **Skeletonize**. Available in: https://scikit-image.org/docs/dev/auto_examples/edges/plot_skeleton.html>. Access in 3.25.2020.

SINGER, Daniel E. et al. Screening for diabetic retinopathy. **Annals of Internal Medicine**, v. 116, n. 8, p. 660-671, 1992.

SOUSA, F.O. Desenvolvimento de sistema de simulação de endoscopia nasal. **Master's Thesis (Master in Telecommunication Engineering)**. Instituto Federal do Ceará, Fortaleza, 2020.

STANKIEWICZ, Agnieszka et al. Novel full-automatic approach for segmentation of epiretinal membrane from 3D OCT images. In: **2017 Signal Processing: Algorithms, Architectures, Arrangements, and Applications (SPA)**. IEEE, 2017. p. 100-105.

STEEL, D. H. W. et al. Predicting macular hole closure with ocriplasmin based on spectral domain optical coherence tomography. **Eye**, v. 30, n. 5, p. 740-745, 2016.

SUI, Xiaodan et al. Choroid segmentation from optical coherence tomography with graph-edge weights learned from deep convolutional neural networks. **Neurocomputing**, v. 237, p. 332-341, 2017.

SUZUKI, Satoshi et al. Topological structural analysis of digitized binary images by border following. **Computer vision, graphics, and image processing**, v. 30, n. 1, p. 32-46, 1985.

SZKULMOWSKI, Maciej et al. Efficient reduction of speckle noise in Optical Coherence Tomography. **Optics express**, v. 20, n. 2, p. 1337-1359, 2012.

TADAYONI, Ramin et al. Decreased retinal sensitivity after internal limiting membrane peeling for macular hole surgery. **British Journal of Ophthalmology**, v. 96, n. 12, p. 1513-1516, 2012.

TAKAHASHI, Atsushi; NAGAOKA, Taiji; YOSHIDA, Akitoshi. Enhanced vitreous imaging optical coherence tomography in primary macular holes. **International ophthalmology**, v. 36, n. 3, p. 355-363, 2016.

TANG, Chang; ZHENG, Xiao; CAO, Lijuan. OCT despeckling via weighted nuclear norm constrained non-local low-rank representation. **Laser Physics Letters**, v. 14, n. 10, p. 106001, 2017.

THAPA, Suman S. et al. Prevalence and pattern of vitreo-retinal diseases in Nepal: the Bhaktapur glaucoma study. **BMC ophthalmology**, v. 13, n. 1, p. 9, 2013.

TRANOS, Paris G. et al. Macular edema. **Survey of ophthalmology**, v. 49, n. 5, p. 470-490, 2004.

TIAN, Jing et al. Real-time automatic segmentation of optical coherence tomography volume data of the macular region. **PloS one**, v. 10, n. 8, p. e0133908, 2015.

TRABELSI, Hedi; ZGHAL, Imen; SLOKOM, Nesrine. Segmentation of cystoids macular edema in optical cohenrence tomography. In: **2016 2nd International Conference on Advanced Technologies for Signal and Image Processing (ATSIP)**. IEEE, 2016. p. 303-306.

VAN DIJK, Hille W. et al. Decreased retinal ganglion cell layer thickness in patients with type 1 diabetes. **Investigative ophthalmology & visual science**, v. 51, n. 7, p. 3660-3665, 2010.

VASAVADA, Jesal; TIWARI, Shamik. Sobel-Fuzzy Technique to Enhance the Detection of Edges in Grayscale Images Using Auto-Thresholding. In: **Proceedings of the Second International Conference on Soft Computing for Problem Solving (SocProS 2012), December 28-30, 2012**. Springer, New Delhi, 2014. p. 617-627.

VASUKI, P.; KANIMOZHI, J.; DEVI, M. Balkis. A survey on image preprocessing techniques for diverse fields of medical imagery. In: **2017 IEEE International Conference on Electrical, Instrumentation and Communication Engineering (ICEICE)**. IEEE, 2017. p. 1-6.

VELEZ-MONTOYA, Raul et al. Current knowledge and trends in age-related macular degeneration: genetics, epidemiology, and prevention. **Retina**, v. 34, n. 3, p. 423-441, 2014.

VERCELLIN, Alice C. Verticchio et al. Diagnostic capability of three-dimensional macular parameters for glaucoma using optical coherence tomography volume scans. **Investigative ophthalmology & visual science**, v. 59, n. 12, p. 4998-5010, 2018.

VERMEER, K. A. et al. Automated segmentation by pixel classification of retinal layers in ophthalmic OCT images. **Biomedical optics express**, v. 2, n. 6, p. 1743-1756, 2011.

VICENTE, Sara; KOLMOGOROV, Vladimir; ROTHER, Carsten. Graph cut based image segmentation with connectivity priors. In: **2008 IEEE conference on computer vision and pattern recognition**. IEEE, 2008. p. 1-8.

UNIVERSITY of California, Davis. **VSRI Laboratory**. Available in: <<http://vsri.ucdavis.edu/research/retinal/oct>>. Access in 08.08.2019.

WALDSTEIN, Sebastian M. et al. Quantitative comparison of macular segmentation performance using identical retinal regions across multiple spectral-domain optical coherence tomography instruments. **British Journal of Ophthalmology**, v. 99, n. 6, p. 794-800, 2015.

WANG, Chuang; WANG, Ya Xing; LI, Yongmin. Automatic choroidal layer segmentation using markov random field and level set method. **IEEE journal of biomedical and health informatics**, v. 21, n. 6, p. 1694-1702, 2017.

WANJARI, Manish T.; KALASKAR, Keshao D.; DHORE, Dr Mahendra P. Document Image Segmentation using Region Based Methods. **International Journal of Computing Science and Information Technology**, ISSN, p. 2278-9669, 2015.

WHAT Is a Macular Hole? What Causes a Macular Hole? **American Academy of Ophthalmology**. 2019. Available in: <<https://www.aao.org/eye-health/diseases/what-is-macular-hole>>. Access in 6.26.2020.

WOJTKOWSKI, Maciej. High-speed optical coherence tomography: basics and applications. **Applied optics**, v. 49, n. 16, p. D30-D61, 2010.

XIANG, Dehui et al. Automatic retinal layer segmentation of OCT images with central serous retinopathy. **IEEE journal of biomedical and health informatics**, v. 23, n. 1, p. 283-295, 2018.

XU, Juan et al. 3D OCT eye movement correction based on particle filtering. In: **2010 Annual International Conference of the IEEE Engineering in Medicine and Biology**. IEEE, 2010. p. 53-56.

XU, David et al. A novel segmentation algorithm for volumetric analysis of macular hole boundaries identified with optical coherence tomography. **Investigative ophthalmology & visual science**, v. 54, n. 1, p. 163-169, 2013.

XU, Min et al. Texture preservation and speckle reduction in optical coherence tomography using the shearlet-based total variation algorithm. **Optics and Lasers in Engineering**, v. 122, p. 265-283, 2019.

YU, Kai et al. Shared-hole graph search with adaptive constraints for 3D optic nerve head optical coherence tomography image segmentation. **Biomedical optics express**, v. 9, n. 3, p. 962-983, 2018.

ZAWADZKI, Robert J. et al. Ultrahigh-resolution optical coherence tomography with monochromatic and chromatic aberration correction. **Optics Express**, v. 16, n. 11, p. 8126-8143, 2008.

ZHANG, Li et al. Automated segmentation of intraretinal cystoid macular edema for retinal 3D OCT images with macular hole. In: **2015 IEEE 12th International Symposium on Biomedical Imaging (ISBI)**. IEEE, 2015. p. 1494-1497.

Appendix A – Algorithm for the proposed method

- 1: Load Libraries (cv2, numpy, imutils, skimage, math, matplotlib, warnings, imageio, PIL, sklearn, seaborn, pandas and time);
- 2: Analyze each image at a time (“for” loop);
- 3: Start counting time process;
- 4: Brightness adjustment normalization (Table 3);
- 5: Gaussian Blur filter to remove noise and enhance quality of image (kernel 11x11 pixel size);
- 6: Thresholding to binarize the image (cutoff point of 45);
- 7: Series of two erosions and two dilations alternately (kernel 3x3 pixel size);
- 8: Flood Fill algorithm at inferior background of image (seed at 550x470 coordinate and kernel using the 8-connected method);
- 9: Border Following algorithm to find the ILM boundary layer segmentation with MH (contour retrieval mode chosen to get the longest contour; contour approximation method chosen compresses horizontal, vertical, and diagonal segments and leaves only their end points; side and bottom cuts in the image to eliminate unwanted parts of the contour);
- 10: Compare prediction and ground-truth contours and get mean and standard deviation of distance error through the shortest distance from a point to a curve (Euclidean Distance);
- 11: Algorithm stops here if the objective is to find the ILM boundary layer segmentation without MH pathology, otherwise, it continues;
- 12: Find the coordinates of the two top pixels of the curve to close the MH (shortest distance from a point to a curve);
- 13: Get MH area using Flood Fill algorithm at the inferior and superior areas of the image, isolating the macular hole;
- 14: Invert image colors to compare prediction and ground-truth through the confusion matrix of the pixels (evaluation metrics: accuracy, IoU, DSC and MCC);
- 15: Border Following algorithm to find the MH area contour (contour retrieval mode chosen to get the longest contour; contour approximation method chosen compresses horizontal, vertical, and diagonal segments and leaves only their end points);
- 16: Get the six extreme points of the contour (shortest distance from a point to a curve and Flood Fill algorithm; see section 4.11 for more details);

17: Evaluate the measurements of MH prediction through the comparison with the ground-truth measures (classification of images using the macular hole index evaluation to reinforce the validation; see section 5.3 for more details).

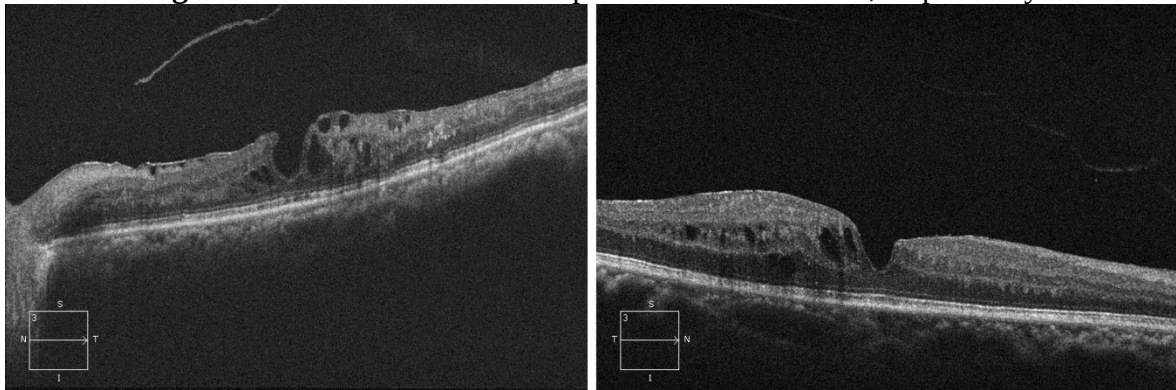
Appendix B – MH images excluded from the work

The specialist physician from *Escola Cearense de Oftalmologia* responsible for analyzing and generating the GT from the OCTID dataset excluded some MH images from the work. According to him, among the 37 discarded images are conditions relating to Macular Edema (ME), Macular Pseudo Hole (MPH), Lamellar Macular Hole (LMH), Epiretinal Membrane (ERM), Vitreomacular Traction (VMT) syndrome and MH with Retinal Detachment (RD). There are also some NM images in the MH image dataset. A total of 65 MH images were used. The images excluded from this work are described below.

Macular Edema

A Macular Edema (ME) is a complication of Diabetic Retinopathy and is the most common cause of vision loss and blindness (SINGER *et al.*, 1992). ME is defined as a swelling of the retina in diabetic patients due to the leakage of fluid within the central macula from microaneurysms (dilated small blood vessels) that form as the result of chronic damage due to elevated blood sugar levels (GIANCARDIO *et al.*, 2012). Images with ME can be seen in Figure 52.

Figure 52: Macular Edema examples. MH29 and MH37, respectively



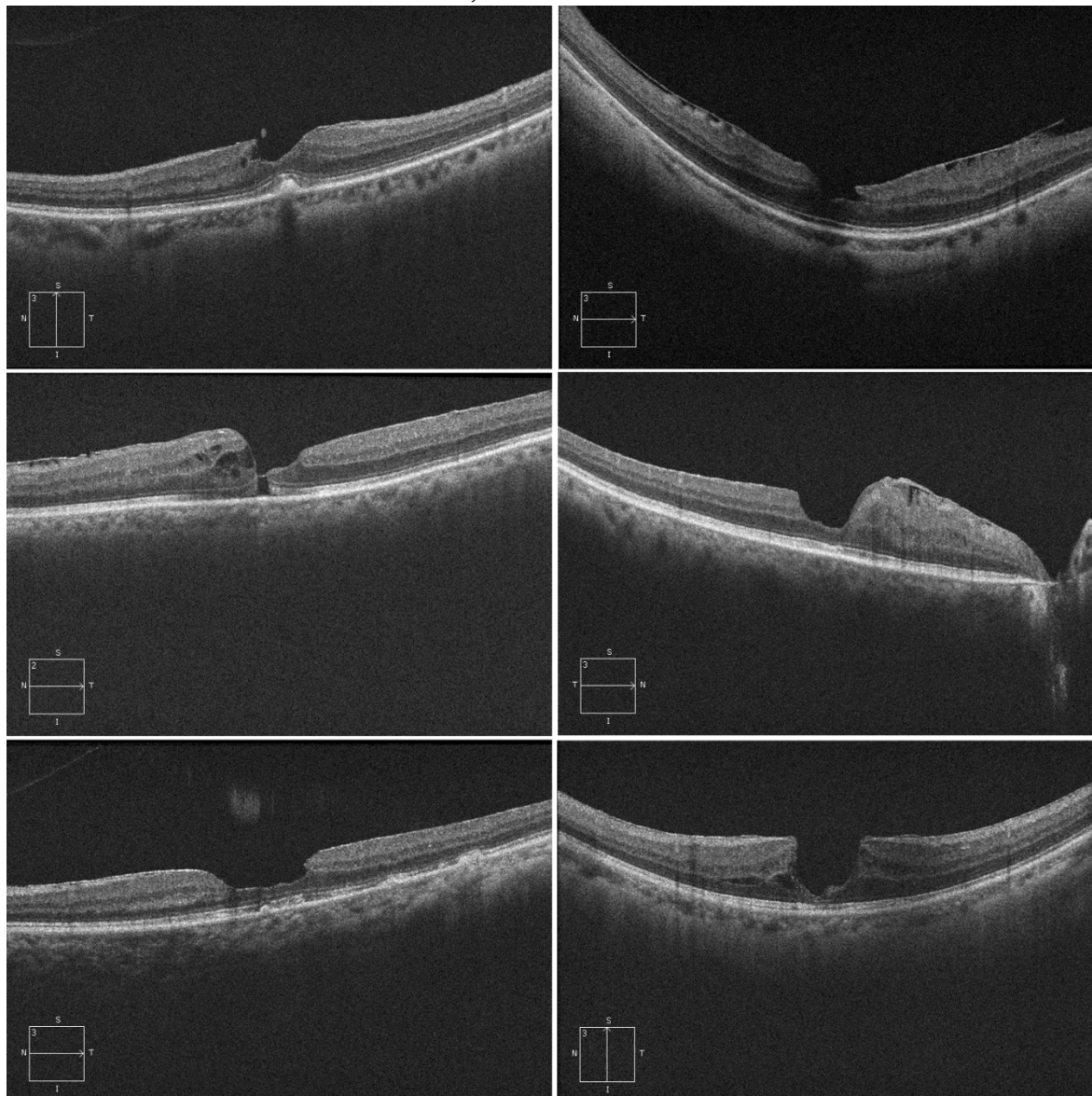
Source: Adapted from OCTID (2018).

Macular Pseudo Hole

A MPH is attributable to the centripetal contraction of an epiretinal membrane. ERM is always present in pseudo holes. It has the round and reddish appearance of a macula (HAOUCHINE *et al.*, 2004). It is called a pseudo hole because the ILM and ERM layers do

not touch each other, as in the case of a full-thickness MH. Images of MPH can be seen in Figure 53.

Figure 53: Pseudo MH examples. From left to right, top to bottom: MH15, MH28, MH56, MH61, MH90 and MH100

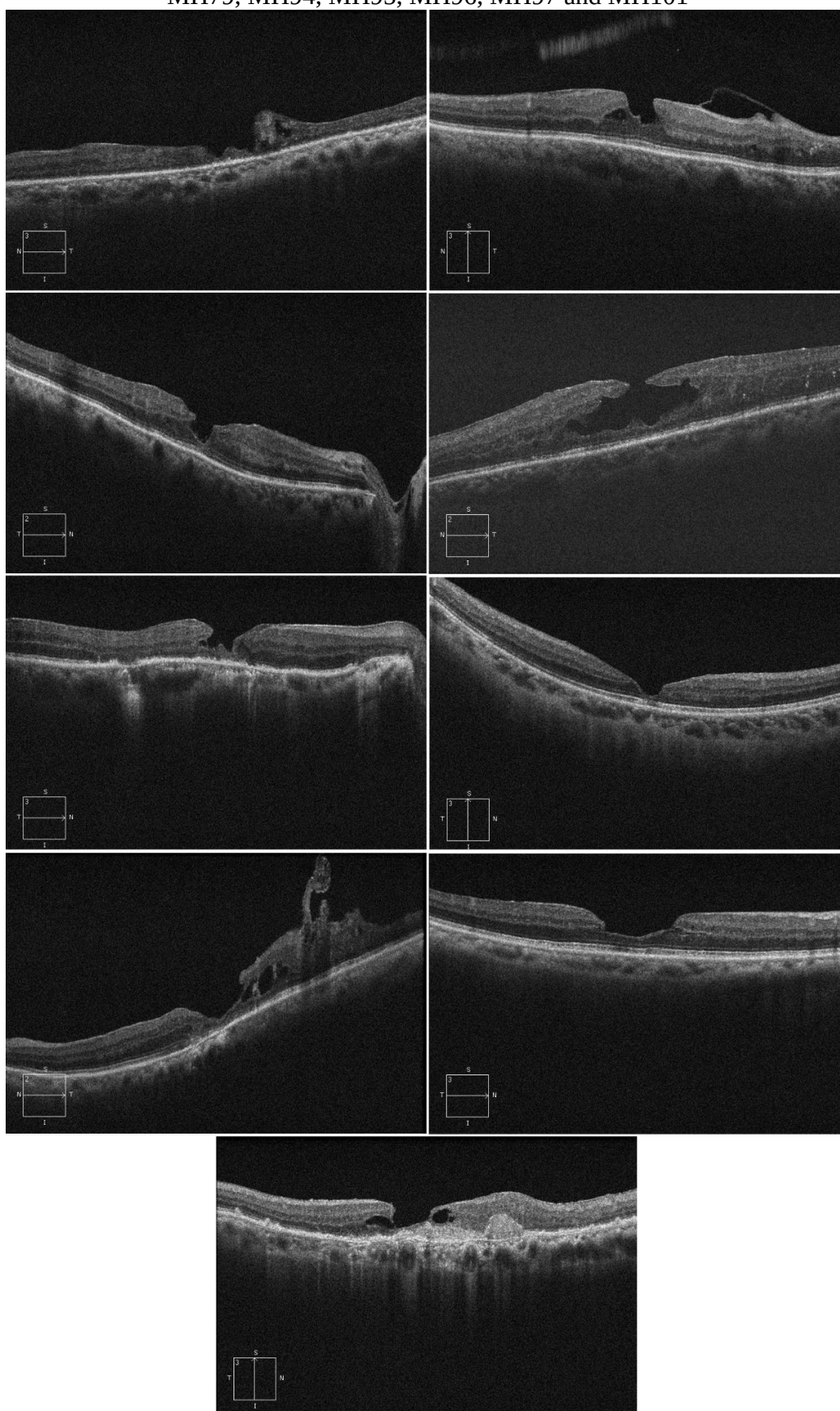


Source: Author.

Lamellar Macular Hole

A LMH usually results from an abortive process of MH formation or may be a complication of a chronic macular cystoid edema (HAOUCHINE *et al.*, 2004). As in the case of MPH, the ILM and RPE layers do not touch each other, as in the case of a full-thickness MH. Images of LMH can be seen in Figure 54.

Figure 54: Lamellar MH examples. From left to right, top to bottom: MH1, MH22, MH65, MH79, MH94, MH95, MH96, MH97 and MH101

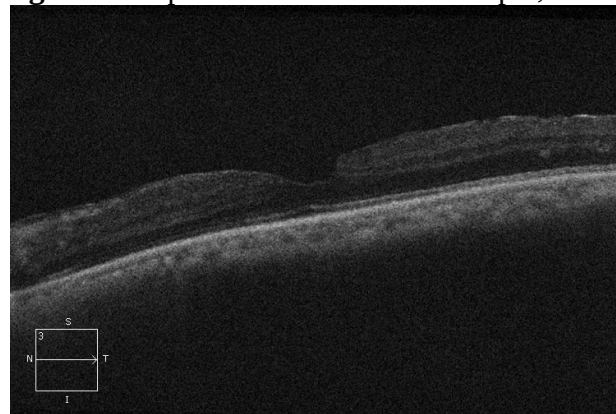


Source: Adapted from OCTID (2018).

Epiretinal Membrane

Epiretinal membranes may occur in the posterior pole of eyes without any associated retinal disease. Membranes may rarely be congenital or developmental, but more often they are acquired. Idiopathic epiretinal membranes can cause reduced vision and metamorphopsia by several mechanisms, including tissue covering and/or distorting the macula; low traction retinal detachment of the posterior pole; vascular leakage with intraretinal edema; and obstruction of the axoplasmic flow (DE BUSTROS *et al.*, 1988). An image of an epiretinal membrane can be seen in Figure 55.

Figure 55: Epiretinal Membrane example, MH92

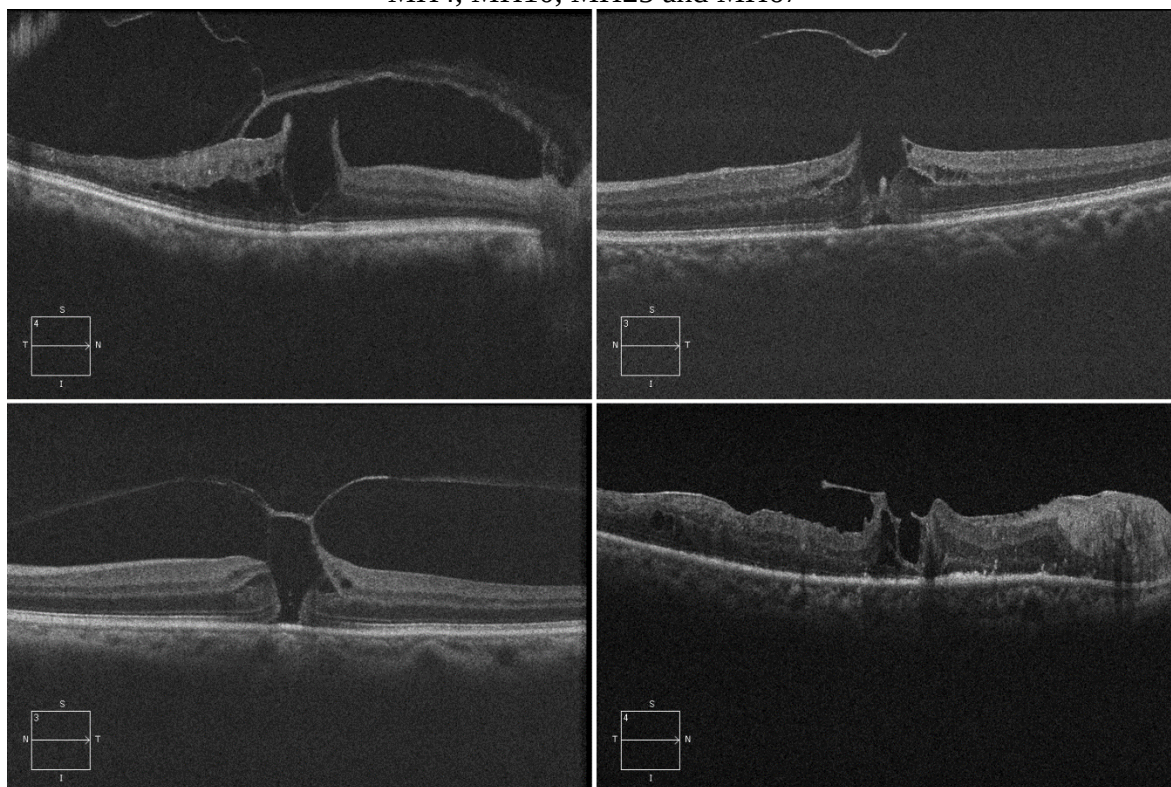


Source: OCTID (2018).

Vitreomacular Traction Syndrome

Posterior vitreous detachment occurs because of changes in the vitreous gel due to aging (FOOS; WHEELER, 1982). The posterior cortical vitreous begins to detach from the retina and a complete separation posterior to the equator rapidly ensues. In some cases, the separation may be incomplete, resulting in persistent vitreous traction on the posterior retina: vitreomacular traction syndrome (GASS, 1987). Macular traction, frequently accompanied by cystoid changes in the neurosensory retina of the macula, causes decreased visual acuity, metamorphopsia, photopsia, and micropsia (MARGHERIO *et al.*, 1989). Images of vitreomacular traction syndrome can be seen in Figure 56.

Figure 56: Vitreomacular Traction Syndrome examples. From left to right, top to bottom: MH4, MH10, MH25 and MH67



Source: Adapted from OCTID (2018).

MH with Retinal Detachment

A retinal detachment occurs when the retina is separated from the rest of the layers of the eye. This condition usually occurs after the development of a tear in the retina, allowing fluid to get under the retina forming a detachment (MICHEL; WILKINSON; RICE, 1990). An image of MH with retinal detachment can be seen in Figure 57.

Figure 57: MH with Retinal Detachment, MH11

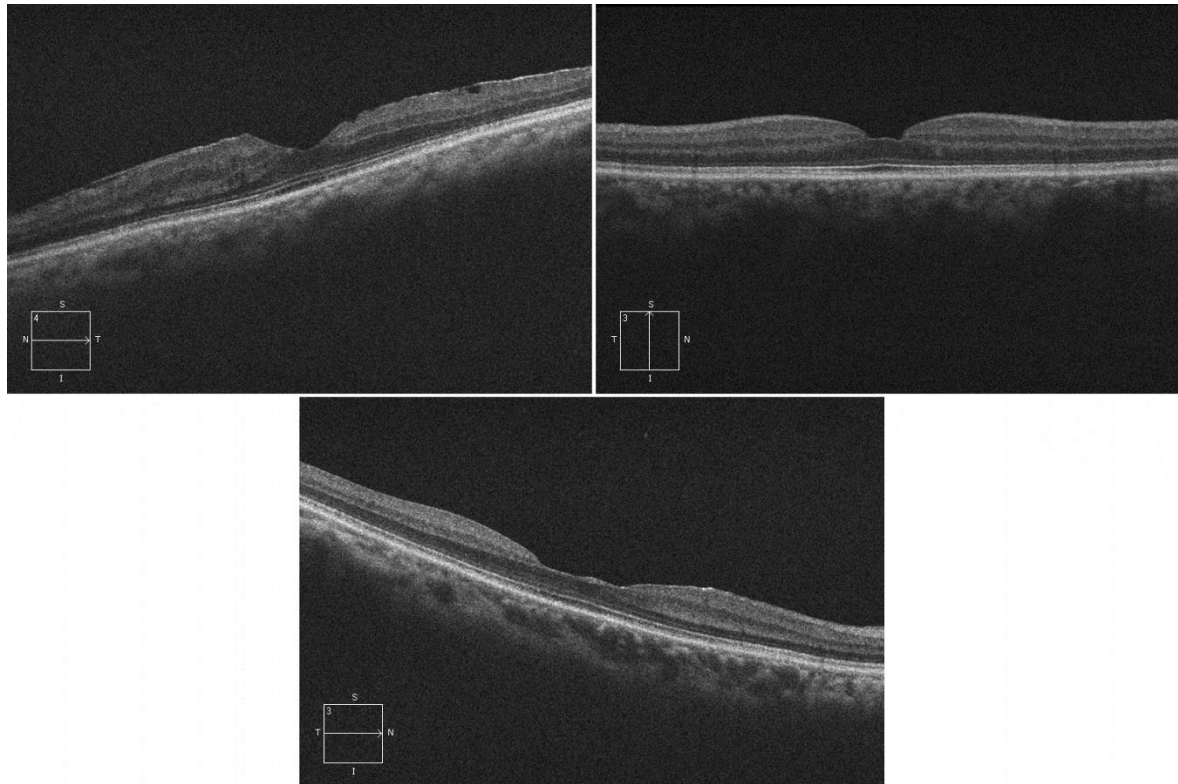


Source: OCTID (2018).

Normal Macula

In the normal macula, all layers are healthy. There is no presence of pathologies. Images of NM can be seen in Figure 58.

Figure 58: Normal Macula examples. From left to right, top to bottom: MH21, MH26 and MH82

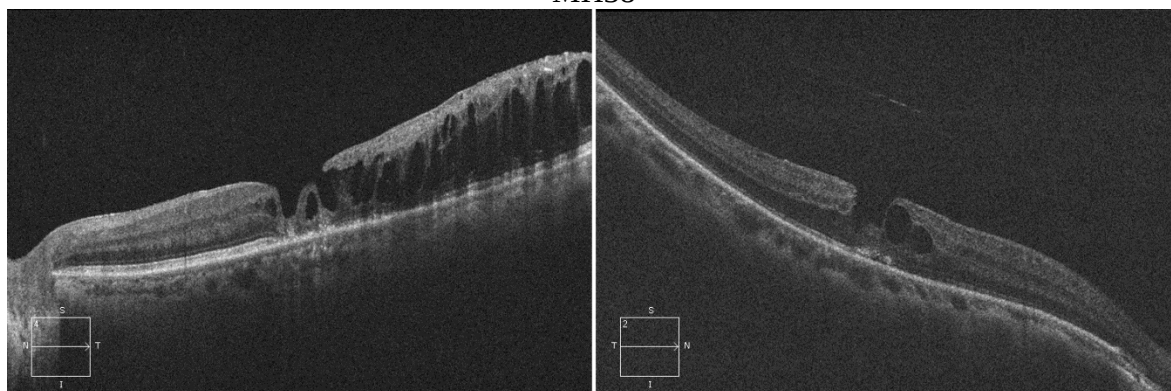


Source: Adapted from OCTID (2018).

Images with more than one condition

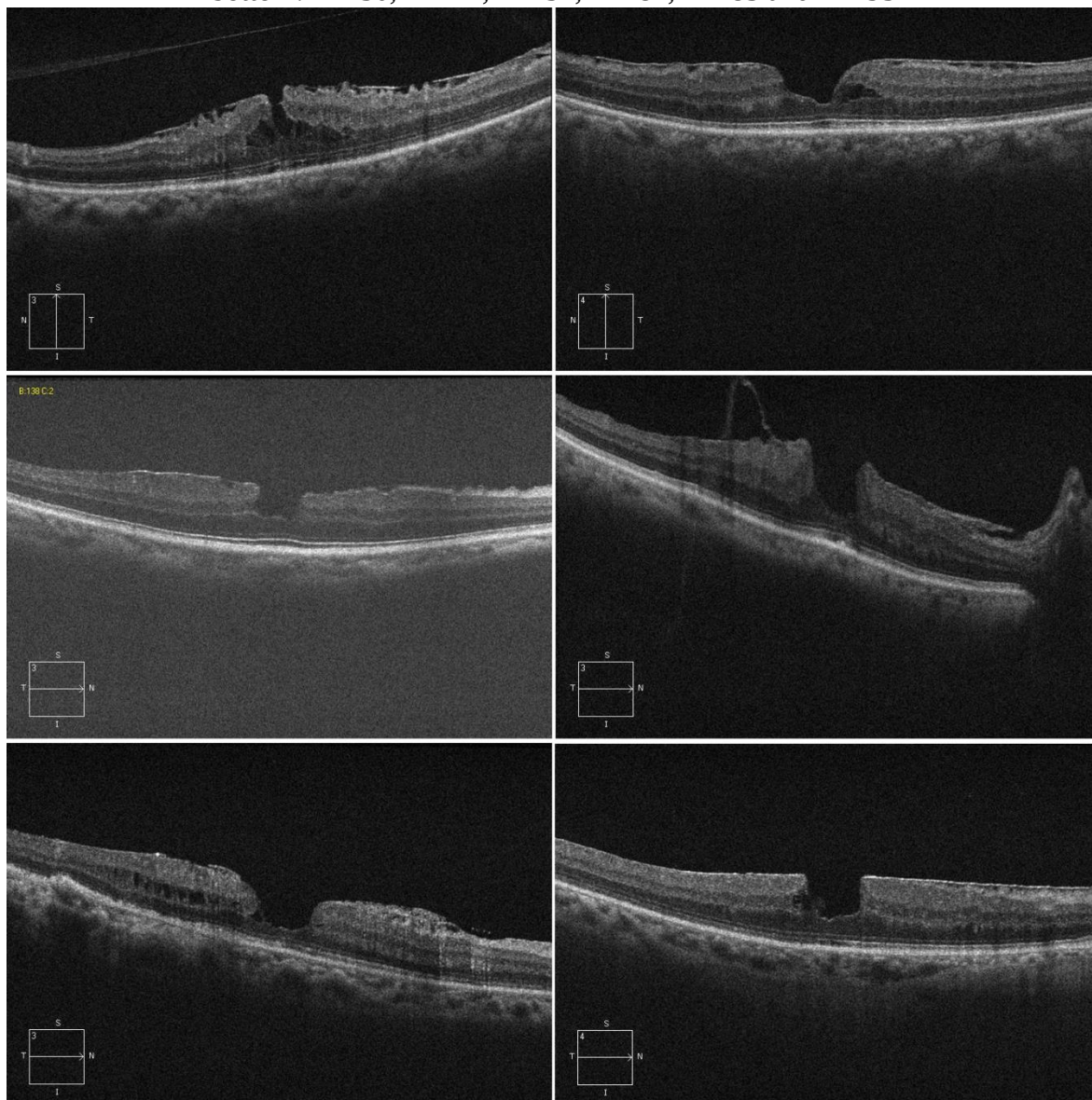
According to the specialist consulted for this work, the OCTID database also contains images with more than one condition, which can appear together. Figure 59 shows two cases of lamellar macular hole plus a macular edema. In the six images as of Figure 60 there are two conditions: epiretinal membrane plus macular pseudo hole. In Figure 61 there is one image with three conditions: macular edema, lamellar macular hole and epiretinal membrane. A macular edema plus a macular pseudo hole is shown in Figure 62. The last case contains an epiretinal membrane plus a macular edema seen in Figure 63.

Figure 59: A lamellar macular hole plus a macular edema. From left to right: MH17 and MH38



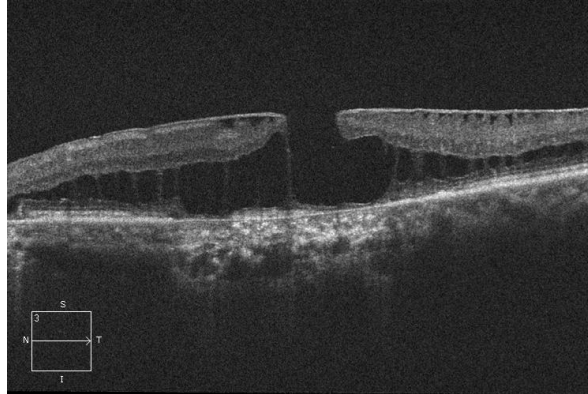
Source: Adapted from OCTID (2018).

Figure 60: An epiretinal membrane plus a macular pseudo hole. From left to right, top to bottom: MH30, MH47, MH57, MH84, MH85 and MH93



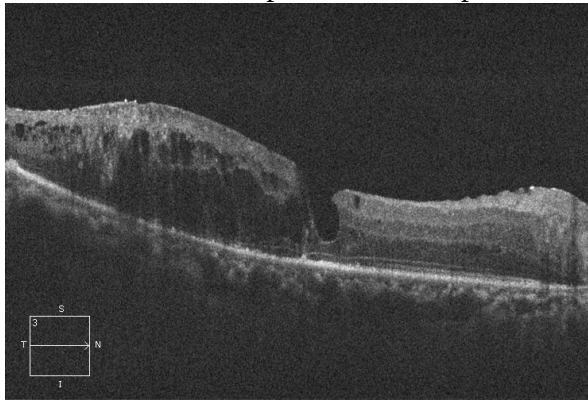
Source: Adapted from OCTID (2018).

Figure 61: A macular edema, a lamellar macular hole and an epiretinal membrane. MH35



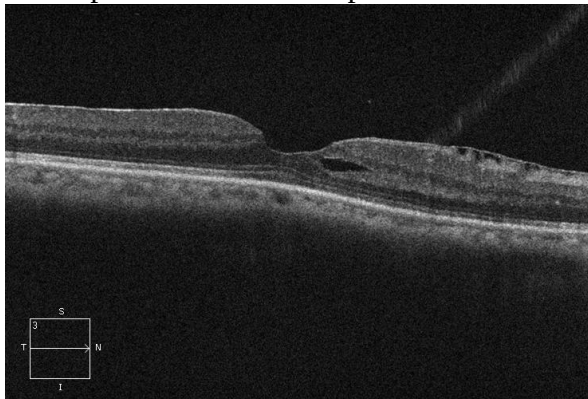
Source: OCTID (2018).

Figure 62: A macular edema plus a macular pseudo hole. MH 36



Source: OCTID (2018).

Figure 63: An epiretinal membrane plus a macular edema. MH91



Source: OCTID (2018).

This appendix shows and explains the reasons for the exclusion of these 37 images from the work by the specialist. Thus, only 65 images from the public OCTID database were used.

Appendix C - Individual results for each OCT image with MH pathology

Table 13 shows the individual results for each OCT image with an MH pathology. Mean and SD of UE are the results for the ILM boundary layer segmentation. Accuracy, IoU, DSC and MCC are the results for the MH area segmentation.

Table 13: Individual results for each OCT image with an MH pathology

MH	Mean (pixel) of UE	SD (pixel) of UE	Acc (%)	IoU (%)	DSC (%)	MCC (%)
2	2.5697	1.5789	0.9956	0.8873	0.9403	0.9382
3	2.9109	1.2836	0.9968	0.8753	0.9335	0.9332
5	2.5489	1.0258	0.9936	0.87	0.9305	0.9296
6	3.046	1.5116	0.9921	0.8206	0.9015	0.9021
7	3.7781	1.3676	0.9956	0.8268	0.9052	0.9042
8	3.1025	1.8116	0.9959	0.839	0.9124	0.9109
9	2.7983	1.6008	0.9967	0.8983	0.9464	0.9457
12	2.2951	1.2991	0.9967	0.7725	0.8717	0.87
13	1.7542	1.1806	0.997	0.8604	0.925	0.9236
14	3.0777	5.2502	0.9833	0	0	-0.0011
16	1.7263	1.2925	0.9982	0.7806	0.8768	0.8759
18	2.5652	1.1563	0.9956	0.8016	0.8899	0.8917
19	3.9771	5.0929	0.9951	0.7711	0.8708	0.8684
20	2.2497	1.5652	0.9981	0.8781	0.9351	0.9349
23	2.0385	0.9828	0.9958	0.7506	0.8575	0.8579
24	2.8657	1.2413	0.9945	0.8718	0.9315	0.9308
27	2.1724	1.1843	0.9977	0.8728	0.9321	0.9325
31	2.2692	1.408	0.9954	0.8027	0.8905	0.8884
32	1.309	0.9289	0.9977	0.8938	0.9439	0.9429
33	2.4782	1.964	0.9915	0.6986	0.8226	0.8317
34	2.5459	2.1216	0.9958	0.8247	0.9039	0.9018
39	2.8349	1.2541	0.9956	0.8174	0.8995	0.8995
40	3.4687	1.0616	0.9952	0.7765	0.8742	0.8734
41	1.9141	1.2414	0.9972	0.8417	0.9141	0.9128
42	1.7402	1.2111	0.9974	0.9037	0.9494	0.9486
43	2.5616	1.2041	0.9928	0.8631	0.9265	0.9254
44	1.8714	1.2462	0.9948	0.7907	0.8831	0.8839
45	2.5498	1.9302	0.9971	0.8198	0.901	0.9029
46	1.9612	1.11	0.9975	0.8741	0.9328	0.9322
48	1.8978	1.8413	0.9676	0.6431	0.7828	0.7773
49	2.4571	1.6119	0.9959	0.3467	0.5149	0.5309
50	1.9457	1.3365	0.9974	0.8994	0.947	0.9468
51	1.421	1.2887	0.9943	0.7325	0.8456	0.8442
52	2.3637	1.3034	0.9977	0.8898	0.9417	0.9413
53	1.4648	1.2084	0.9967	0.8047	0.8918	0.8917

54	2.9091	1.7243	0.996	0.8613	0.9255	0.9244
55	2.2263	1.9804	0.9959	0.8286	0.9063	0.9042
58	3.2278	1.6204	0.9923	0.7364	0.8482	0.8527
59	2.857	1.8368	0.998	0.8615	0.9256	0.9248
60	3.1701	1.9189	0.9932	0.8244	0.9038	0.9021
62	2.8677	1.0117	0.9967	0.7541	0.8598	0.8583
63	2.5344	1.8592	0.9966	0.8308	0.9076	0.9094
64	2.6044	1.2605	0.9949	0.7628	0.8654	0.8702
66	3.35	1.358	0.9938	0.7111	0.8312	0.8299
68	2.2804	1.1495	0.9939	0.7672	0.8682	0.8702
69	2.2191	1.0258	0.9948	0.8496	0.9187	0.9168
70	2.5972	1.8122	0.9949	0.7903	0.8828	0.8846
71	3.6146	2.0347	0.9887	0.6948	0.8199	0.8141
72	3.7547	4.0702	0.9951	0.6801	0.8096	0.8101
73	4.8499	5.1252	0.991	0.7285	0.8429	0.848
74	2.9653	1.5025	0.9912	0.7443	0.8534	0.8492
75	3.4624	1.4984	0.9937	0.7754	0.8735	0.8703
76	3.1928	1.1559	0.9953	0.8842	0.9386	0.9377
77	2.8841	1.321	0.9967	0.7603	0.8638	0.8637
78	5.5469	10.157	0.9334	0.3384	0.5057	0.5353
80	2.1463	1.5717	0.9955	0.8702	0.9306	0.9304
81	2.7589	1.3244	0.9962	0.8892	0.9414	0.9404
83	2.976	1.5493	0.9952	0.8753	0.9335	0.933
86	2.303	2.399	0.9965	0.9126	0.9543	0.9531
87	6.5068	8.8653	0.9923	0.7027	0.8254	0.8242
88	2.3178	1.2692	0.9958	0.8799	0.9361	0.9355
89	2.6627	1.3875	0.9972	0.8751	0.9334	0.9339
98	2.6004	1.7874	0.9906	0.7421	0.8519	0.851
99	2.589	1.3777	0.9968	0.8528	0.9206	0.9197
102	2.317	1.2453	0.9958	0.7157	0.8343	0.8322

Source: Author.

Appendix D - Measurements of MH ground-truth (μm)

Table 14 shows the individual measures for each OCT image with an MH pathology.

Table 14: Measurements of MH ground-truth (μm)

MH	Top diameter	Bottom diameter	Height	Minimum diameter
2	578.231	777.778	489.796	307.256
3	605.442	534.013	401.36	242.63
5	766.44	758.503	536.281	362.812
6	824.263	750.567	451.247	478.458
7	719.955	554.422	325.397	377.551
8	434.24	612.245	505.669	114.512
9	552.154	673.469	520.408	294.784
12	272.109	386.621	346.939	129.252
13	523.809	546.485	303.855	280.045
14	419.501	419.501	397.959	176.871
16	391.156	132.653	314.059	55.555
18	626.984	419.501	386.621	293.651
19	405.896	554.421	297.052	260.771
20	520.408	369.614	327.664	207.483
23	549.887	242.63	350.34	122.449
24	523.809	734.694	494.331	476.19
27	546.485	376.417	387.755	202.948
31	411.565	579.365	303.855	337.868
32	270.975	391.156	410.431	283.447
33	616.78	527.211	384.354	373.016
34	633.787	588.435	248.299	484.127
39	518.14	490.93	436.508	209.75
40	551.02	522.676	361.678	257.37
41	450.113	407.029	424.036	137.188
42	471.655	554.422	430.839	285.714
43	578.231	1068.027	556.689	290.249
44	570.295	520.408	422.902	218.821
45	471.655	342.404	416.1	134.921
46	371.882	495.465	469.388	158.73
48	513.605	1065.76	643.991	337.868
49	198.413	164.399	210.884	51.02
50	506.803	732.426	361.679	312.925
51	206.349	489.796	470.521	80.499
52	479.592	391.156	512.472	120.181
53	409.297	459.184	407.029	138.322
54	473.923	714.286	394.558	332.199
55	436.508	664.399	285.714	334.467
58	545.351	575.964	500	217.687

59	564.626	276.644	240.363	251.701
60	384.354	1037.415	545.351	180.272
62	399.093	191.61	332.199	116.78
63	404.762	401.36	433.106	210.884
64	497.732	502.267	445.578	167.8
66	470.521	377.551	291.383	286.848
68	534.014	641.723	429.705	222.222
69	502.267	1083.9	419.501	231.292
70	392.29	650.794	492.063	151.927
71	666.667	764.172	373.016	396.825
72	476.19	563.492	175.737	439.909
73	715.419	709.75	403.628	385.487
74	488.662	532.88	484.127	278.911
75	514.739	715.419	323.129	351.474
76	562.358	824.263	470.521	342.404
77	359.41	412.698	307.256	212.018
78	430.839	696.145	649.66	147.392
80	633.787	724.49	401.36	426.304
81	603.175	700.68	430.839	399.093
83	719.955	831.066	431.973	420.635
86	616.78	827.664	479.592	344.671
87	615.646	705.215	326.531	356.009
88	425.17	625.85	527.211	323.129
89	638.322	509.07	340.136	309.524
98	328.798	662.131	505.669	278.911
99	521.542	564.626	320.862	332.199
102	379.818	554.422	218.821	242.63

Source: Author.

Appendix E - Measurements of MH prediction (μm)

Table 15 shows the individual measures for each OCT image with an MH pathology.

Table 15: Measurements of MH prediction (μm)

MH	Top diameter	Bottom diameter	Height	Minimum diameter
2	562.6	694.4	487.9	313.4
3	629.4	489.1	397.7	237.9
5	631.8	700.5	508.5	371.2
6	627.9	694.2	388.5	464.4
7	589.7	472.7	305.8	352.7
8	530.4	550.4	472.5	143.9
9	475.8	581.3	488.8	261.1
12	350.8	339.5	364.2	122.5
13	549.7	500.4	322.2	272.4
14	258.3	23.7	27.6	52
16	572.6	82.7	329.7	47.1
18	604.5	327.6	349.2	280.8
19	570.7	467.8	285.7	257.9
20	544.9	301.7	312.9	192.1
23	698.1	671.8	188.3	591.7
24	452.7	651.6	425.8	448.5
27	519.9	314.4	371.9	241.2
31	577.8	516	319.9	325.4
32	604	346.2	393.8	322.9
33	494.3	435.9	291.5	432.1
34	708.7	481.3	241.1	464.3
39	508.8	433.5	421.2	168.4
40	558.2	440.7	347.6	263.4
41	460.5	355.2	413.4	145.6
42	448.7	524.1	414.1	283
43	559.7	827.1	507.2	275
44	679.3	475.8	393.4	245.6
45	523.5	242.1	393.5	121.2
46	464.3	430.4	464.1	195.8
48	440.7	963.3	588.9	352
49	559.3	492	16.1	504.1
50	554.7	624.1	342	299.9
51	476.4	553.9	374.6	195.4
52	523.5	328.3	488.1	154.7
53	577.7	362.3	406.1	114.8
54	521.3	553.4	355.4	313.4
55	666.9	705.9	310.6	324.5
58	566.8	483.7	461.6	177.1

59	620	500.4	123.9	489.7
60	601.4	816.2	511.7	161.1
62	553.8	164.2	344	113.2
63	507	288.7	396.2	198.9
64	448.5	445.2	399.3	159.1
66	632.2	347.2	305.1	289.9
68	685.6	562.5	410.7	282.6
69	548	978.1	408.5	226.2
70	472.1	535.6	442.8	170
71	561.8	637.9	414.3	300.8
72	631	440.7	71.7	436.8
73	762.7	507	349.4	413.4
74	604.2	597.9	375.7	302.7
75	609.9	638	336.8	337
76	608.6	768.3	416.3	333.4
77	567.4	331.5	311.4	225.1
78	429	997	555.3	231.3
80	670.6	633.3	382.8	469.2
81	619	611.6	406.3	374
83	629.6	732	390.2	432.8
86	636.7	691.2	453.5	360
87	657.2	429.2	261.3	317.8
88	422.4	587.4	481.1	323.7
89	621.6	423.4	310.6	294.2
98	397.8	577.7	407.2	325.6
99	579.1	462.2	312.4	375.9
102	569.1	496.4	245.7	248.2

Source: Author.

Appendix F - Unsigned error of MH measurements (μm)

Table 16 shows the individual unsigned error of measures for each OCT image with an MH pathology.

Table 16: Unsigned error of MH measurements (μm)

MH	Top diameter	Bottom diameter	Height	Minimum diameter
2	15.631	83.378	1.896	6.144
3	23.958	44.913	3.66	4.73
5	134.64	58.003	27.781	8.388
6	196.363	56.367	62.747	14.058
7	130.255	81.722	19.597	24.851
8	96.16	61.845	33.169	29.388
9	76.354	92.169	31.608	33.684
12	78.691	47.121	17.261	6.752
13	25.891	46.085	18.345	7.645
14	161.201	395.801	370.359	124.871
16	181.444	49.953	15.641	8.455
18	22.484	91.901	37.421	12.851
19	164.804	86.621	11.352	2.871
20	24.492	67.914	14.764	15.383
23	148.213	429.17	162.04	469.251
24	71.109	83.094	68.531	27.69
27	26.585	62.017	15.855	38.252
31	166.235	63.365	16.045	12.468
32	333.025	44.956	16.631	39.453
33	122.48	91.311	92.854	59.084
34	74.913	107.135	7.199	19.827
39	9.34	57.43	15.308	41.35
40	7.18	81.976	14.078	6.03
41	10.387	51.829	10.636	8.412
42	22.955	30.322	16.739	2.714
43	18.531	240.927	49.489	15.249
44	109.005	44.608	29.502	26.779
45	51.845	100.304	22.6	13.721
46	92.418	65.065	5.288	37.07
48	72.905	102.46	55.091	14.132
49	360.887	327.601	194.784	453.08
50	47.897	108.326	19.679	13.025
51	270.051	64.104	95.921	114.901
52	43.908	62.856	24.372	34.519
53	168.403	96.884	0.929	23.522
54	47.377	160.886	39.158	18.799

55	230.392	41.501	24.886	9.967
58	21.449	92.264	38.4	40.587
59	55.374	223.756	116.463	237.999
60	217.046	221.215	33.651	19.172
62	154.707	27.41	11.801	3.58
63	102.238	112.66	36.906	11.984
64	49.232	57.067	46.278	8.7
66	161.679	30.351	13.717	3.052
68	151.586	79.223	19.005	60.378
69	45.733	105.8	11.001	5.092
70	79.81	115.194	49.263	18.073
71	104.867	126.272	41.284	96.025
72	154.81	122.792	104.037	3.109
73	47.281	202.75	54.228	27.913
74	115.538	65.02	108.427	23.789
75	95.161	77.419	13.671	14.474
76	46.242	55.963	54.221	9.004
77	207.99	81.198	4.144	13.082
78	1.839	300.855	94.36	83.908
80	36.813	91.19	18.56	42.896
81	15.825	89.08	24.539	25.093
83	90.355	99.066	41.773	12.165
86	19.92	136.464	26.092	15.329
87	41.554	276.015	65.231	38.209
88	2.77	38.45	46.111	0.571
89	16.722	85.67	29.536	15.324
98	69.002	84.431	98.469	46.689
99	57.558	102.426	8.462	43.701
102	189.282	58.022	26.879	5.57

Source: Author.

Appendix G - Individual results for each OCT image with NM

Table 17 shows the individual results for each OCT image with an NM. Mean and SD of UE are the results for the ILM boundary layer segmentation.

Table 17: Individual results for each OCT image with an NM

MH	Mean (pixel) of AE	SD (pixel) of AE
1	3.5379	1.6638
2	2.7296	1.0163
3	3.0336	1.4521
4	3.1298	2.1881
5	2.5694	1.8867
6	5.4594	4.6235
7	4.844	4.1356
8	1.8752	0.9214
9	3.651	1.8988
10	3.4895	2.3273
11	2.696	1.1711
12	2.7762	1.1004
13	4.0207	2.5556
14	3.4162	2.0072
15	2.1856	1.3571
16	2.6236	0.8153
17	3.809	1.5218
18	1.7807	1.3629
19	2.9656	1.4233
20	2.9006	2.1749
21	2.4932	1.2366
22	2.7082	1.2529
23	2.0828	0.8365
24	4.3625	2.4001
25	2.8088	0.9913
26	8.4812	12.2086
27	5.8653	6.1787
28	1.8628	1.2777
29	3.847	1.8122
30	3.8213	2.0033
31	4.6572	2.6294
32	4.8741	3.8067
33	2.7965	0.9177
34	2.3132	1.1363
35	1.8643	1.1967
36	2.9109	1.1419

37	3.6633	0.9241
38	4.3157	2.641
39	3.5721	1.794
40	3.6513	2.68
41	3.0668	1.292
42	2.9734	2.1431
43	3.473	2.4077
44	3.5927	2.789
45	3.6254	1.3909
46	2.6966	1.1812
47	2.5193	0.8981
48	4.7827	4.7072
49	2.4848	1.3616
50	2.2043	1.0225
51	2.4538	0.7714
52	2.3601	1.1727
53	4.8668	1.893
54	2.4477	0.807
55	2.0913	0.9314
56	2.2265	1.2431
57	3.0033	1.2841
58	3.0336	1.5621
59	2.9297	2.626
60	3.4784	1.4271
61	5.6159	7.0962
62	2.7021	1.4764
63	2.4852	0.8889
64	2.5945	1.2316
65	2.4414	0.9539
66	4.1466	2.8281
67	3.1218	1.9206
68	4.6663	2.9479
69	2.7822	1.4741
70	2.9419	1.3228
71	2.7894	2.7319
72	2.8834	0.9169
73	3.0195	0.9169
74	2.5442	1.2763
75	2.8879	1.3488
76	3.3331	1.3367
77	3.1592	1.6583
78	3.4149	1.6128
79	3.4071	2.4313
80	4.1541	2.5712
81	2.7723	1.5275

82	3.9545	1.9243
83	2.8507	1.6972
84	2.6253	1.1619
85	3.0292	1.2335
86	2.7982	1.6381
87	3.3934	1.3538
88	2.9757	1.1083
89	2.8587	0.9318
90	2.3461	1.1354
91	2.5306	1.8108
92	2.0053	1.0255
93	2.6072	1.6183
94	3.5052	1.643
95	3.0379	1.5693
96	3.5513	1.8972
97	3.2446	1.3268
98	4.9075	3.4383
99	7.2559	5.7294
100	2.9922	1.6145
101	1.7389	1.0101
102	4.6856	3.3716
103	3.5691	1.406
104	2.8283	0.8852
105	3.1699	2.2721
106	2.3305	0.9502
107	5.5928	3.626
108	6.9215	6.6383
109	3.4986	1.2461
110	3.3659	1.519
111	3.2767	1.8984
112	2.2442	0.9154
113	2.5696	1.026
114	2.673	1.2277
115	2.7574	0.8664
116	2.4726	1.0275
117	1.4465	0.8369
118	4.6637	3.8501
119	2.5723	0.9944
120	3.6782	3.701
121	2.6479	0.9123
122	3.6556	1.2408
123	2.577	1.1508
124	2.8132	0.8731
125	2.6682	1.0208
126	2.42	1.0991

127	2.7262	1.6034
128	3.1079	1.577
129	3.6351	1.1399
130	3.57	1.1247
131	8.0088	10.2877
132	3.0616	1.9526
133	4.0542	1.5534
134	2.7556	1.0503
135	2.7057	1.817
136	4.1613	1.9597
137	3.317	1.0837
138	2.9817	1.0337
139	3.9067	1.2789
140	3.0466	1.0274
141	3.2182	1.0478
142	2.0003	1.1757
143	3.3664	1.0785
144	1.9151	1.2461
145	4.0586	1.9689
146	5.6248	3.9199
147	4.4623	1.7304
148	3.9996	1.6491
149	3.2306	1.4513
150	4.0584	1.451
151	2.8736	1.867
152	3.7401	2.9357
153	2.5466	1.7682
154	2.9799	1.229
155	2.7884	0.9256
156	2.2511	1.0519
157	2.904	1.917
158	3.6439	1.0745
159	2.3474	1.1508
160	2.6708	0.9209
161	4.5469	3.973
162	3.4389	2.7287
163	4.9928	2.404
164	3.6962	1.7306
165	3.4645	1.7116
166	3.1494	1.3289
167	3.1497	1.0867
168	2.9918	1.3077
169	3.4772	1.076
170	4.3843	1.9661
171	4.0757	1.6939

172	2.8803	1.3411
173	3.7378	1.556
174	2.9813	1.1865
175	4.1205	2.189
176	2.5326	1.3566
177	2.7134	1.1581
178	2.942	1.1196
179	3.5572	1.3382
180	5.5373	5.4179
181	4.7301	3.4845
182	4.9857	3.4158
183	6.6388	3.6613
184	3.1837	1.5201
185	4.1663	1.7336
186	2.9733	1.44
187	3.7459	1.7015
188	3.6839	1.403
189	5.6461	3.8061
190	4.1728	2.5036
191	3.9612	1.3348
192	3.258	1.1678
193	3.3494	1.7537
194	3.1283	1.0546
195	3.2413	1.6162
196	2.7342	1.2502
197	2.8341	1.3494
198	2.5544	0.9331
199	2.7287	0.9958
200	4.699	3.7978
201	6.1921	6.8607
202	2.927	1.4538
203	3.2936	1.2733
204	3.4187	1.1824
205	2.8859	1.1284
206	2.599	1.0984

Source: Author.