UNIVERSIDADE D COIMBRA FACULDADE DE MEDICINA

MESTRADO INTEGRADO EM MEDICINA – TRABALHO FINAL

BEATRIZ MARIA DA ROCHA SOUSA FERREIRA

Small Choroidal Melanocytic Lesions: Clinical Predictors of Growth and Malignant Progression

ARTIGO CIENTÍFICO

ÁREA CIENTÍFICA DE OFTALMOLOGIA

Trabalho realizado sob a orientação de:

PROFESSOR DOUTOR RUI DANIEL MATEUS BARREIROS PROENÇA

DOUTORA MARIA CRISTINA DIAS FERRÃO FONSECA

ABRIL/2019

Small Choroidal Melanocytic Lesions:
Clinical Predictors of Growth and Malignant Progression

Author: Beatriz Maria da Rocha Sousa Ferreira

Affiliation: Faculty of Medicine, University of Coimbra, Portugal

e-mail: bia.sousaferreira@gmail.com

CONTENTS

ABBREVIATIONS

BCVA – Best Corrected Visual Acuity

CHUC – Centro Hospital e Universitário de Coimbra

CI – Confidence Interval

COMS – *Collaborative Ocular Melanoma Study*

HR – Hazard Ratio

IBM SPSS® – International Business Machines, Statistical Package for the Social Sciences

LBD – Largest Basal Diameter

OCT – Optical Coherence Tomography

OD – Optic Disc

OORC – Onco-Ophthalmology Reference Center

OR – Odds Ratio

RPE – Retinal Pigment Epithelium

SE – Standard Error

SEER-NCI – Surveillance, Epidemiology and End Results Program of the National Cancer Institute

TFSOM – To Find Small Ocular Melanoma

TFSOM-HHD – To Find Small Ocular Melanoma Using Helpful Hints Daily

ABSTRACT

Introduction: Overlapping features between benign choroidal nevi and choroidal melanoma make the differential diagnosis quite complex. Previous studies show a relation between certain features of small choroidal melanocytic lesions and their progression to malignancy which would benefit from further investigation.

Objective: To evaluate small choroidal melanocytic lesions features that correlate significantly with a higher potential of growth and progression to malignancy.

Materials and Methods: Retrospective observational study including 58 patients with the initial diagnosis of small choroidal melanocytic lesions. Data regarding demographic, clinical and ultrasonographic features was collected from a pre-existing data base. Univariate and multivariate statistical analysis were used to identify statistically significant risk and protective factors. The main outcome was documented lesion growth.

Results: Mean patient age was 60 years and median follow-up time was 20.5 months. Mean thickness and largest basal diameter at initial assessment was 2.61 mm and 7.28 mm, respectively. 20 of the 58 (34.5%) patients had lesions that demonstrated growth. Univariate analysis identified seven statistically significant factors associated with lesion growth: presence of subretinal fluid ($p<0.001$; OR = 9.9), presence of orange pigment ($p<0.001$; OR=29), nearest lesion margin distance to the optic disc <3 mm ($p=0.015$; OR=4.024), nearest lesion margin distance to the fovea <3 mm ($p=0.027$; OR=3.578) and hollow echogenicity ($p<0.001$; OR=non estimable). Two significant protective factors against lesion growth were found: presence of an amelanotic halo surrounding the lesion ($p=0.001$; OR=0.053) and presence of overlying drusens ($p<0.001$; OR=0.102). In the multivariate analysis only 4 of the 6 features included maintained statistical significance, namely, presence of subretinal fluid ($p=0.044$; HR=5.071), presence of orange pigment ($p=0.003$; HR=24.899), presence of amelanotic halo ($p=0.026$; HR=0.085) and presence of overlying drusens ($p=0.008$; HR=0.155).

Discussion and Conclusion: The most significant predictive factor pointing to growth and malignant transformation was ultrasonographic low internal echogenicity, overcoming clinical factors found in previous studies. This feature should be carefully investigated in the initial evaluation of every small choroidal melanocytic lesion, along with the other features to estimate the associated risk. This will allow an early treatment and eventually a better prognosis.

Keywords: “Choroidal Melanocytic Lesion”, “Small Choroidal Melanoma”, “Growth”, “Malignancy”, “Choroidal Neoplasm”.

RESUMO

Introdução: Existe uma sobreposição entre as características clínicas dos nevus e do melanoma da coroideia que torna o seu diagnóstico diferencial bastante complexo. Estudos prévios mostraram uma relação entre determinadas características das pequenas lesões melanocíticas da coroideia e a sua progressão para a malignidade, o que motivam esta investigação adicional.

Objetivo: Avaliar as características clínicas e ecográficas de pequenas lesões melanocíticas da coroideia que se correlacionam significativamente com um maior potencial de crescimento e progressão para malignidade.

Materiais e Métodos: Estudo observacional retrospectivo que inclui 58 doentes com o diagnóstico de pequena lesão melanocítica da coroideia. Os dados referentes às características demográficas, clínicas e ecográficas foram coletados a partir de uma base de dados pré-existente. Foi efectuada uma análise estatística univariável e multivariável para identificar fatores de risco e fatores protetores com significado estatístico, tendo em conta o crescimento documentado das lesões.

Resultados: A idade média dos doentes foi de 60 anos e a mediana do tempo de seguimento de 20,5 meses. A espessura média e o maior diâmetro basal na avaliação inicial foram de 2,61 mm e 7,28 mm, respetivamente. Em 20 dos 58 (34,5%) doentes foi documentado crescimento da lesão. Sete fatores estatisticamente significativos associados ao crescimento da lesão na análise univariável foram: fluido subretiniano ($p<0,001$; OR=9,9), pigmento laranja ($p<0,001$; OR=29), distância da margem ao disco óptico <3 mm ($p=0,015$; OR=4,024), distância da margem à fóvea <3 mm ($p=0,027$; OR=3,578) e ecogenicidade interna de baixa refletividade ($p<0,001$; OR=não estimável). Foram encontrados dois fatores protetores: presença de halo amelanótico ($p=0,001$; OR=0,053) e de drusens sobrejacentes ($p<0,001$; OR=0,102). Na análise multivariável apenas 4 das 6 características incluídas mantiveram significado estatístico: fluido subretiniano ($p=0,044$; HR=5,071), pigmento laranja ($p=0,003$; HR=24,899), presença de halo amelanótico ($p=0,026$; HR=0,085) e presença de drusens sobrejacentes ($p=0,008$; HR=0,155).

Discussão e Conclusões: O fator preditivo mais significativo para crescimento e transformação maligna foi a ecogenidade interna de baixa refletividade, demonstrada na ecografia, ultrapassando os factores clínicos apontados em estudos prévios. A ecografia deve ser realizada cuidadosamente na observação inicial de todas as pequenas lesões melanocíticas da coroideia, assim como a avaliação de todas as outras características de forma a ser possível estimar o risco associado e permitir um tratamento precoce e um melhor prognóstico.

Palavras-Chave: "Lesão Melanocítica da Coroideia", "Pequeno Melanoma da Coroideia", "Crescimento", "Malignidade", “Neoplasia da Coroide”.

INTRODUCTION

Choroidal melanoma is the primary malignant intraocular tumor with the highest prevalence in adults. During a period of 40 years, melanoma of the choroid accounted for 67.3% of all ocular melanomas according to the Surveillance, Epidemiology, and End Results Program of the National Cancer Institute (SEER-NCI) database in the United States.[1] There are studies revealing an incidence rate of up to 8.6 per million population of uveal melanoma in some European countries[2] and a 5.2 per million population incidence rate in the United States.[1] However, the differential diagnosis of melanocytic lesions of the choroid is not always linear translating the difficulty in the distinction between a choroidal melanoma in its initial phase and a benign choroidal nevus.

Both small choroidal melanomas and choroidal nevi can have a similar appearance, emerging as a mass with variable pigmentation, shape and size, making the differential diagnosis very challenging. There is a remarkable size overlap between small choroidal melanomas and larger choroidal nevi, accounting for approximately 5 choroidal nevi for every melanoma with the thickness range between 2.5 to 3 mm and roughly 3 choroidal nevi for every melanoma with the largest basal diameter range between 7 to 8 mm. These findings translate the high probability of misclassification of benign versus malignant lesions when it is exclusively based on size.[3]

Several studies have identified multiple characteristics associated with a higher risk of growth and progression to malignancy, namely male gender, absence of alterations of the adjacent retinal pigment epithelium,[4] thickness >2 mm, largest basal diameter >5 mm, presence of orange lipofuscin pigment, absence of drusens, presence of subretinal fluid, justapapillary location, presence of symptoms,[4-10] ultrasonographic hollowness[5,7,8,10] and absence of an amelanotic halo surrounding the lesion.[5,7,10] It was also demonstrated that the combination of several risk factors implies a higher risk of tumor growth.[8-11] When there are no risk factors associated, the risk of transformation into melanoma is very low, around 1% to 4%. The risk of transformation progressively increases with the addition of more risk factors[8-11] and specific combinations of this risk factors imply a risk that can be as high as almost 100% of progression to malignancy according to some studies.[8]

According to this findings, the TFSOM mnemonic was proposed to help remember the lesion features that are risk factors for the progression to choroidal melanoma, which stands for “to find small ocular melanoma” that included thickness greater than 2 mm, subretinal fluid, symptoms, orange pigment and lesion margin 3 mm or less from the optic disc.[7,10] Posteriorly, the mnemonic was extended to “to find small ocular melanoma – using helpful hints daily”, TFSOM-HHD, including ultrasonographic hollowness, halo absence and drusens absence, respectively.[5,7,10]

However, there is still the need to refine all this information to make the elaboration of concise criteria and allow an adequate approach of all patients with melanocytic lesions of the choroid and an early identification of choroidal melanoma, which will lead to a more favorable prognosis of these patients.

In the present study our objective is to evaluate the demographic, clinical and imagological characteristics of a Portuguese population of patients diagnosed with small melanocytic lesions of the choroid and determine which ones correlate significantly to their potential for growth and progression to malignancy. This way we can further understand which patients have increased risk of transformation into a choroidal malignant lesion and should be monitored closely and indicated for treatment in an earlier stage of the disease.

MATERIALS AND METHODS

Study design

This observational retrospective study included a group of patients referred for a first visit to the Onco-Ophthalmology Reference Center at the Centro Hospital e Universitário de Coimbra (OORC-CHUC) with the diagnosis of small choroidal melanocytic lesions over a period of approximately 6 years, from March of 2013 to September of 2018.

All the clinical and demographic information regarding these patients, who were assessed in the multidisciplinary therapeutic decision-making consultation of the OORC-CHUC, was registered in a database, taking the appropriate measures to anonymize and prevent patient identification. This database is property of the Onco-Ophthalmology Reference Center and was provided for the purpose of this analysis. The study was approved by the Institutional Review Board of CHUC and followed the tenets of the Declaration of Helsinki.

The patients included in this study were evaluated in two or more visits, during the period of March of 2013 to March of 2019. Each patient was examined by at least two experienced ocular oncologists using indirect ophthalmoscopy and were evaluated by high-resolution ultrasonography, retinography, fundus autofluorescence and optical coherence tomography. Visual acuity was assessed using the Snellen visual acuity chart.

Ultrasonography

The 20 MHz probe of Aviso ultrasound platform (Quantel Medical™, Clermont-Ferrand, France) was used in all lesion measurements. The magnetic 20 MHz probe for posterior pole has a transducer frequency of 20 MHz, an angle of exploration of 50°, with a 24 to 26 mm focus and axial resolution of 100 μm and a lateral resolution of 250 μm. One specialist in ocular ultrasonography and one ocular oncologist measured the thickness of the lesion from the inner surface of the sclera to lesion apex and the largest basal diameter. Thickness was measured from two meridians, along the largest basal diameter and perpendicular to it. Representative digitized scans were prospectively stored at the time of each diagnostic and follow-up visit.

For the purpose of this study, lesion thickness and largest basal diameter from stored scans were re-measured, by consensus of two investigators. We used a mouse-driven cursor to manually mark the inner scleral surface of the lesion and its apex, perpendicular to the largest basal diameter, to measure the linear distance between these points.

Eligibility criteria

From all of the patients included in the database with the diagnosis of small choroidal melanocytic lesions, for this study were only selected cases with lesions ≥1.5 mm and <3.5 mm in thickness and ≥4 mm and ≤12 mm in largest basal diameter and with a minimum follow-up of 6 months, gathering the cohort of patients with lesions sufficiently large not to be considered benign nevi but small enough and demonstrating certain characteristics indicative of stability that led the specialists to believe that the adequate approach was periodic follow-up and not immediate therapeutic intervention. All patients who did not meet the size criteria or with an insufficient follow-up time were excluded. Only one choroidal melanocytic lesion per patient and per eye was included in this study. If there was more than one lesion in each patient or each eye, the largest lesion was considered.

The collected data from the center database regarding this cohort of patients was: demographic information (gender, date of birth and date of the first and following visits), visual acuity of the affected eye, presence of visual symptoms, presence of visual field defects, presence of flashes and/or floaters, affected eye (right or left eye), lesion location (nasal, temporal, superior, inferior or macular), lesion dimensions in the first and following visits (thickness and largest basal diameter, both in mm), nearest lesion margin distance to the optical disc (higher or lower than 3 mm), nearest lesion margin distance to the fovea (higher or lower than 3 mm), lesion features (presence of drusens, orange pigment and/or subretinal fluid), presence of adjacent retinal pigment epithelium fibrous metaplasia, presence of an amelanotic halo surrounding the lesion and demonstration of low reflectivity internal echoes in the ultrasonographic evaluation.

The only clinical outcome considered in this study was tumor growth. Tumor growth was defined as an increase in any of the lesion dimensions (thickness or largest basal diameter) compared to the initial observation.

Statistical analysis

Statistical analysis was performed using IBM SPSS® (International Business Machines, Statistical Package for the Social Sciences), version 22. Descriptive statistics for continuous variables was described as mean ± standard error or median and range, depending on the normality of the distribution, tested using Kolmogorov-Smirnov test. Inferential statistics for univariate analysis was performed using chi-square statistics, with the appropriate corrections according to Cochran's criteria. Estimates for relative risk were obtained by the determining odds-ratio. For multivariate analysis all criteria with a significant odds ratio were included in a logistic regression, separating risk from protective factors. All comparisons were performed at a significance level of 0.05.

RESULTS

According to the eligibility criteria, 58 patients were included in this study. 20 patients (34.5%) had small choroidal melanocytic lesions with documented growth (Fig. 1 and 2). The other 38 patients (65.52%) no growth was documented during the follow-up period.

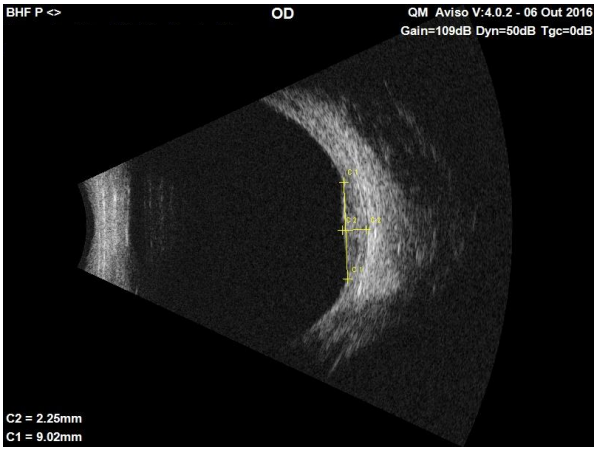

Fig. 1. Thickness and LBD evaluated by US (20 MHz probe).

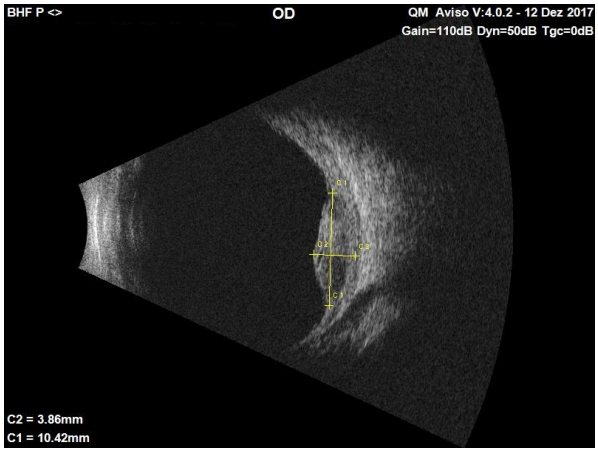

Fig. 2. Documented growth after 1 year of follow-up.

The mean age in the first assessment of this cohort was 60 years (SE=1.74; range: 29-84) and the mean follow-up time was 20.5 months (SE=1.82; range: 6-55.2 months). Regarding the initial dimensions of all lesions included in the study, the mean thickness was 2.61 mm (SE=0.09; range: 1.5-3.50) and the mean largest basal diameter was 7.28 mm (SE=0.25; range: 4.0-12.0). The distribution of the multiple variables of this study is summarized in Table I and II.

Table I. Clinical features: quantitative variables distribution (58 cases).

Feature	Mean	Median	SE	Min - Max	Significance*
Follow-up (months)	24.22	20.5	1.82	6.0 - 55.2	0.013
Patient's age (years)	60.07	60.0	1.74	29 - 84	0.200
First visit thickness (mm)	2.61	2.63	0.09	1.50 - 3.50	0.200
Last visit thickness (mm)	3.01	2.88	0.16	1.22 - 8.16	0.200
Thickness difference	0.45	0.14	0.14	-0.96 - 4.97	0.001
First visit LBD (mm)	7.28	7.27	0.25	4.00 -12.0	0.200
Last visit LBD (mm)	7.85	7.61	0.34	3.00 - 16.0	0.200
LBD difference	0.58	0.24	0.27	-3.87 - 8.97	0.006

SE - Standard Error; LBD - Largest Basal Diameter.
*Calculated by the Kolmogorov-Smirnov test of normality. Significance <0.05 represents a non-normal distribution.

Table II. Clinical features at the first evaluation (58 cases).

Feature		n	(%)
Gender	Female	39	(67)
	Male	19	(33)
Affected Eye	Right	33	(57)
	Left	25	(43)
Quadrant Location	Nasal	7	(12.1)
	Temporal	13	(22.4)
	Superior	20	(34.5)
	Inferior	10	(17.2)
	Macular	8	(13.8)
Visual Symptoms	Absent	23	(40)
	Present	35	(60)
BCVA	0.1	7	(12.1)
	0.2	4	(6.9)
	0.25	2	(3.4)
	0.3	6	(10.3)
	0.4	6	(10.3)
	0.5	4	(6.9)
	0.6	8	(10.3)
	0.8	11	(19)
	0.9	2	(3.4)
	1.0	8	(13.8)
Visual Field Defect	Absent	52	(90)
	Present	6	(10)
Flashes and/or Floaters	Absent	45	(78)
	Present	13	(22)
RPE Fibrous Metaplasia	Absent	33	(57)
	Present	25	(43)
Subretinal Fluid	Absent	41	(71)
	Present	17	(29)
Halo	Absent	38	(66)
	Present	20	(34)
Drusens	Absent	27	(47)
	Present	31	(53)
Orange Pigment	Absent	31	(53)
	Present	27	(47)
Hollow Echogenicity	Absent	29	(50)
	Present	29	(50)
Distance to Foveola	≥3mm	29	(50)
	<3mm	29	(50)
Distance to Optic Disc	≥3mm	33	(57)
	<3mm	25	(43)

BCVA - Best Corrected Visual Acuity; RPE - Retinal Pigment Epithelium.

The correlation between the categorical variables and the presence or not of growth is presented in Table III.

Table III. Univariate analysis of features present at initial assessment of 58 small choroidal melanocytic lesions and their association with growth into choroidal melanoma.

			Growth (n=20)		No Growth (n=38)			
Feature		**Total (n=58)**	**n**	**%**	**n**	**%**	**p-value**	**OR (95%CI)**
Gender	Female	39	13	33.3	26	66.7	0,792	-
	Male	19	7	36.8	12	63.2		
Affected Eye	Right	33	12	36.4	21	63.6	0.729	-
	Left	25	8	32	17	68		
Visual Symptoms	Absent	23	6	26.1	17	73.9	0.275	-
	Present	35	14	40	21	60		
Visual Field Defect	Absent	52	17	32.7	35	67.3	0.398	-
	Present	6	3	50	3	50		
Flashes and/or Floaters	Absent	45	15	33.3	30	66.7	0.732	-
	Present	13	5	38.5	8	61.5		
RPE Fibrous Metaplasia	Absent	33	12	36.4	21	63.6	0.729	-
	Present	25	8	32	17	68		
Subretinal Fluid	Absent[a]	41	8	19.5	33	80.5	<0.001	9.9 (2.7-36.3)
	Present	17	12	70.6	5	29.4		
Halo	Absent[a]	38	19	50	19	50	0.001	0.053 (0-0.4)
	Present	20	1	5	19	95		
Drusens	Absent[a]	27	16	59.3	11	40.7	<0.001	0.102 (0-0.4)
	Present	31	4	12.9	27	87.1		
Orange Pigment	Absent[a]	31	2	6.5	29	93.5	<0.001	29 (5.6-149)
	Present	27	18	66.7	9	33.3		
Hollow Echogenicity	Absent[a]	29	0	0	29	100	<0.001	Non Estimable
	Present	29	20	69	9	31		
Distance to Foveola	≥3mm[a]	29	14	48.3	15	51.7	0.027	3.578 (1.1-11.4)
	<3mm	29	6	20.7	23	79.3		
Distance to Optic Disc	≥3mm[a]	33	13	52	12	48	0.015	4.024 (1.3-12.7)
	<3mm	25	7	21.2	26	78.8		

RPE - Retinal Pigment Epithelium; OR - Odds Ratio; [a] Reference variable.

Statistically significant factors in the univariate analysis associated with lesion growth include presence of subretinal fluid (p<0.001; OR=9.9) (Fig. 3), presence of orange pigment (p<0.001; OR=29) (Fig. 4), nearest lesion margin distance to the optic disc of less than 3 mm (p=0.015; OR=4.024) (Fig. 5), nearest lesion margin distance to the fovea of less than 3 mm (p=0.027; OR=3.578) (Fig. 6) and hollow echogenicity (p<0.001) (Fig. 7 and 8). Odds ratio was not calculated for the hollow echogenicity (low internal reflectivity) due to the lack of events on feature absent in association with growth on this variable.

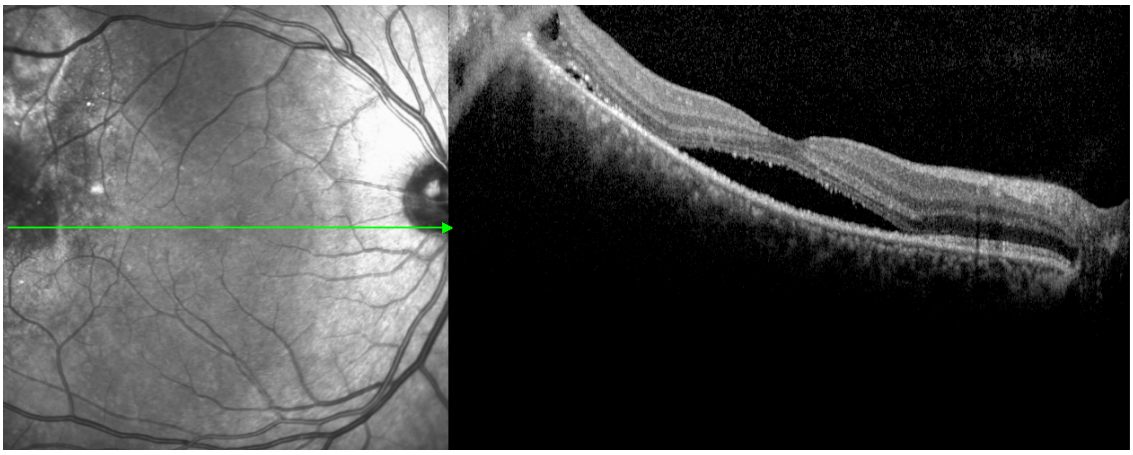

Fig. 3. Optical Coherence Tomography (OCT) demonstrating subretinal fluid.

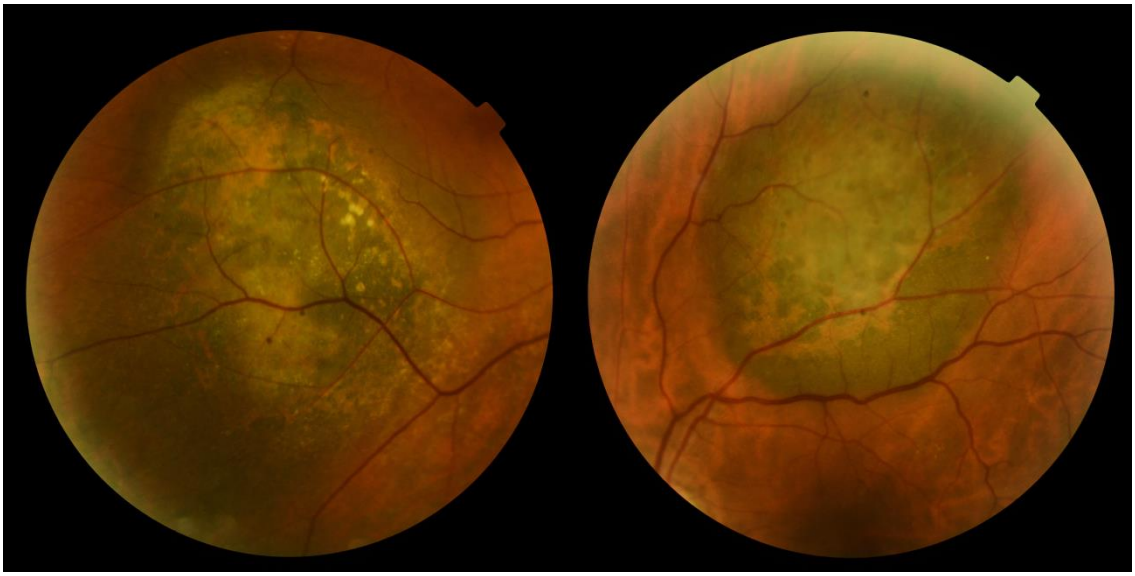

Fig. 4. Presence of orange pigment over the lesion.

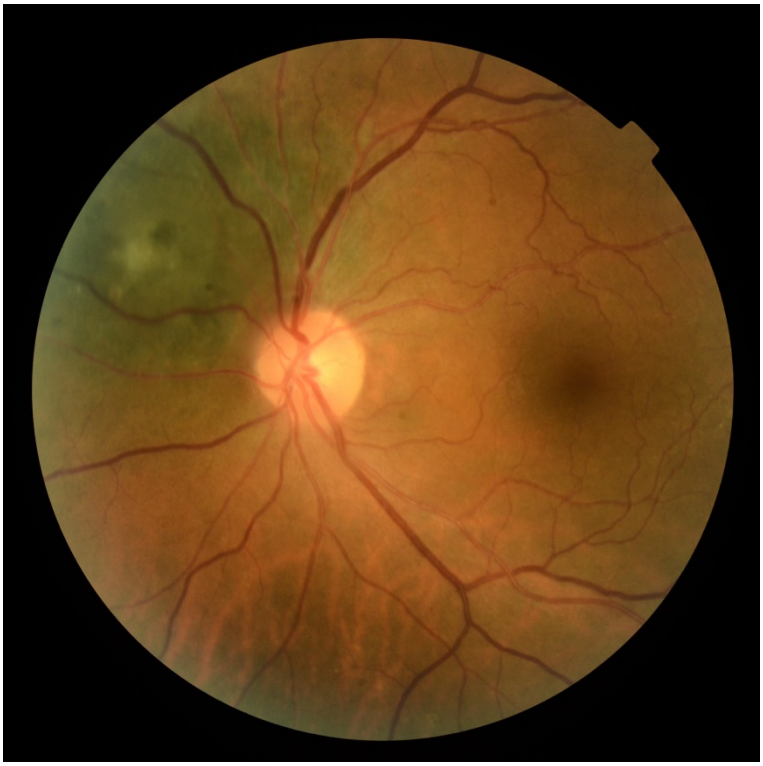

Fig. 5. Nearest lesion margin distance to the optic disc <3 mm.

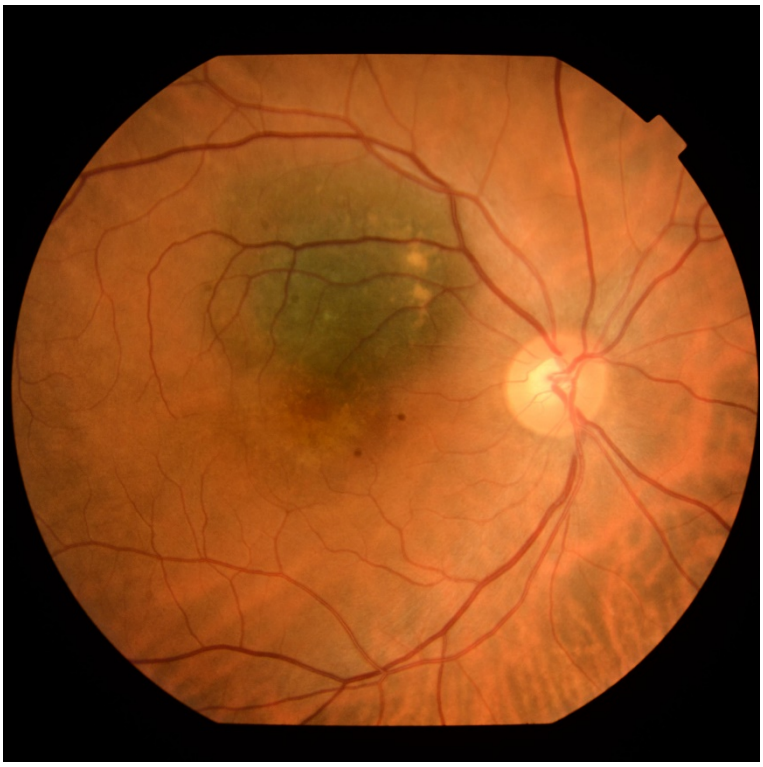

Fig. 6. Nearest lesion margin distance to the fovea <3 mm.

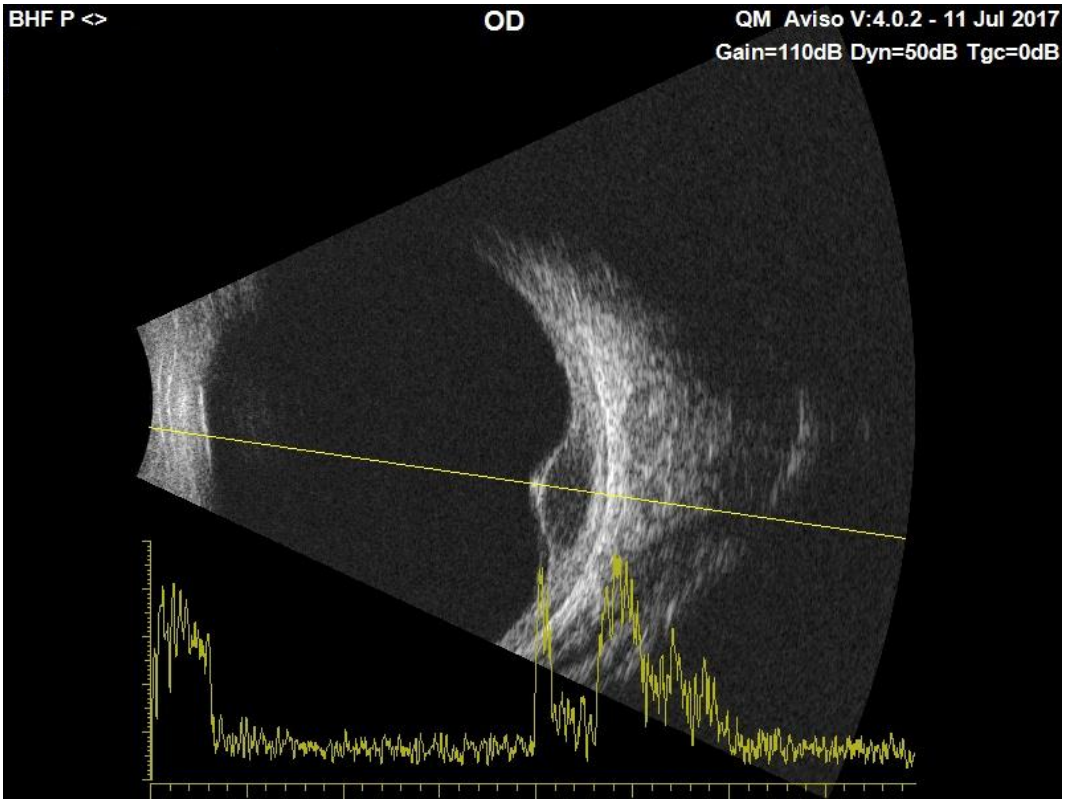

Fig. 7. Ultrasonography (20 MHz probe) demonstrating low internal reflectivity of the lesion.

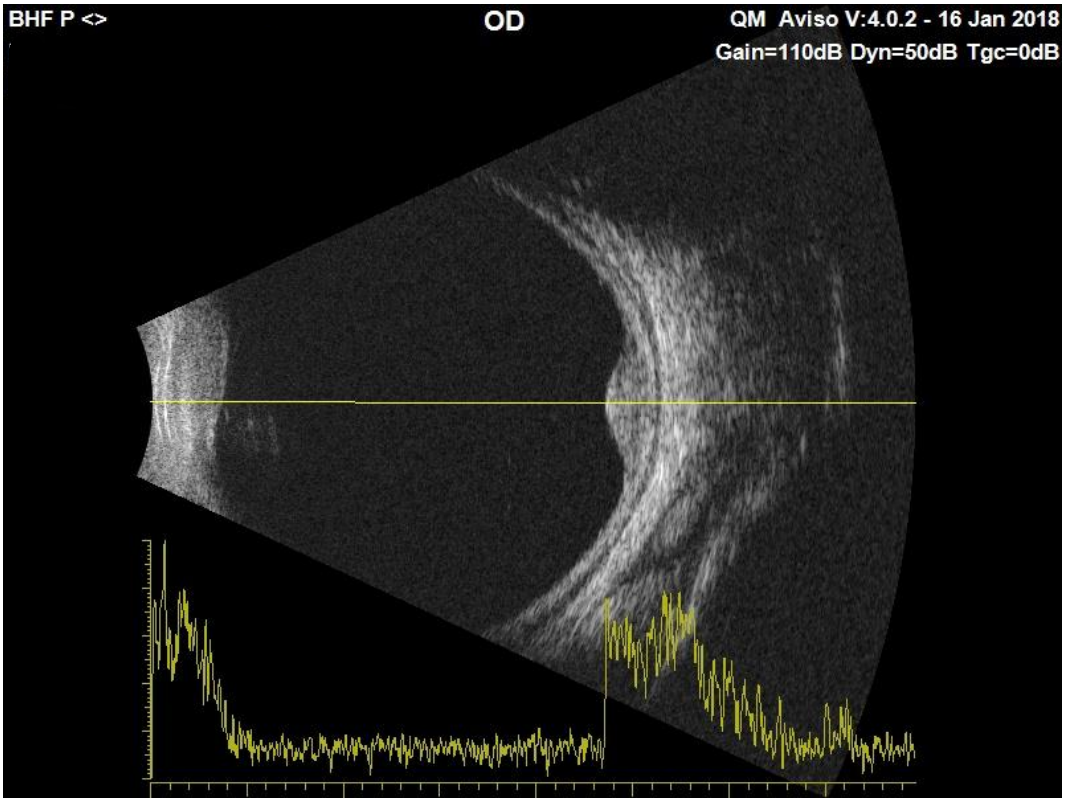

Fig. 8. Ultrasonography (20 MHz probe) demonstrating high internal reflectivity of the lesion.

There were two significant factors correlated with absence of lesion growth, specifically, presence of amelanotic halo surrounding the lesion (p=0.001; OR=0.053) and presence of overlying drusens (p<0.001; OR=0.102).

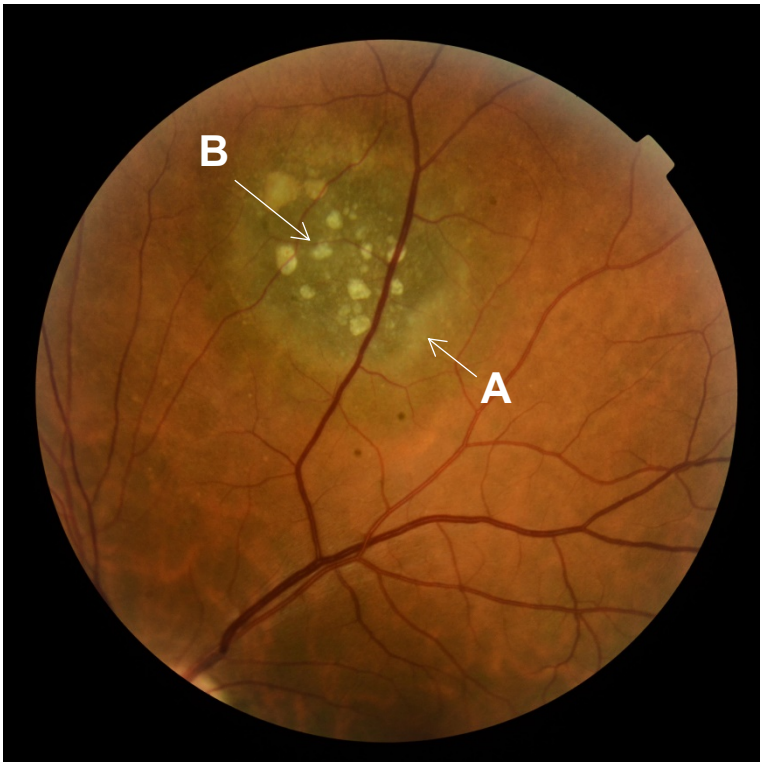

Fig. 9. Amelanotic halo surrounding the lesion (A) and presence of overlying drusens (B) were associated with absence of growth.

Gender (p=0.792), the affected eye (p=0.729), visual symptoms (p=0.275), visual field defects (p=0.398), flashes/floaters (p=0.732) and retinal pigment epithelium fibrous metaplasia (p=0.729) were not statistically significant factors associated with lesion growth.

In the subsequent multivariate analysis (Table IV), two of the five identified significant risk factors in the univariate analysis maintained statistical significance, namely, the presence of subretinal fluid (p=0.044; HR=5.071) and the presence of orange pigment (p=0.003; HR=24.899). The two significant protective factors found in the univariate analysis (amelanotic halo surrounding the lesion and overlying drusens) also kept their statistical significance in the multivariate analysis.

Table IV. Multivariate analysis of features present at initial assessment of 58 small choroidal melanocytic lesions and their association with growth.

	p - value	HR	95% CI
Risk Factors			
Subretinal Fluid (vs. absent)	0.044	5.071	1.04 - 24.66
Orange Pigment (vs. absent)	0.003	24.899	3.09 - 200.76
Margin <3 mm from OD (vs. ≥3 mm)	0.477	1.800	0.36 - 9.09
Margin <3 mm from fovea (vs. ≥3 mm)	0.362	0.403	0.06 - 2.85
Protective Factors			
Halo (vs. absent)	0.026	0.085	0.01 - 0.75
Drusens (vs. absent)	0.008	0.155	0.04 - 0.62

HR - Hazard Ratio; CI - Confidence Interval; OD - Optic Disc.

DISCUSSION

Choroidal melanoma is associated with a 5-year all-cause mortality of 16% in melanomas less than 3 mm of thickness, 32% in melanomas with 3 to 8 mm of thickness and 53% in melanomas with more than 8 mm of thickness.[12] These findings support the fact that an early detection of the tumor is crucial for a more favorable prognosis. Differential diagnosis of small choroidal melanoma with choroidal nevi and indeterminate choroidal melanocytic lesions is very challenging on account of the existing clinical similarity between their features. Differentiating a clearly benign nevus from a suspicious indeterminate choroidal melanocytic lesion that is more likely to have poorer outcome is of greater importance because choroidal nevus have a very low chance (1/8845) of progression to melanoma.[13]

Multiple studies have been published consisting in the analysis of factors related with a higher risk of growth and transformation into melanoma in small choroidal melanocytic lesions. However, there is a great disparity mainly regarding the inclusion criteria based on the lesion dimensions. This translates in a considerable variability of the results. Relatively to indeterminate choroidal melanocytic lesions specifically, the existing literature is not so extensive probably due to the lack of objective criteria for the classification of choroidal melanocytic lesions and therefore making this categorization process quite subjective.

The growth rate observed in our study is coincident with reported growth rates in previously published studies, specifically in the retrospective study conducted by Butler *et al.*[14] that included 293 “indeterminate pigmented choroidal tumors” (36% of the lesions demonstrated enlargement) and in the fifth report from the *Collaborative Ocular Melanoma Study (COMS)*[6] that included 188 “small choroidal melanomas” with 1 to 3 mm of thickness and 5 to 16 mm of largest basal diameter (31% of those lesions showed growth). Several other studies reported growth rates are quite diverse, representing the variance of inclusion criteria, population composition and study design, ranging from 2.4 to 18%.[4,5,7,15]

Since the publication of the study carried out by Shields *et al.* in 1995[15] introducing the TFSOM mnemonic to abbreviate the identified risk factors for growth of small choroidal melanocytic lesions, multiple other studies were executed in order to further comprehend the influence of this features on tumor growth and find additional information on this topic. The COMS study[6] found that larger initial tumor dimensions, presence of orange pigment, absence of areas of retinal pigmented epithelial changes adjacent to the tumor and absence of drusens were factors predictive of growth. In an analysis conducted by Singh *et al.*,[4] features associated with lesion growth included thickness ≥2 mm, proximity to the foveola of ≤3 mm, presence of subretinal fluid, symptoms and orange pigment and male gender. Later, a study including 2514 patients with small choroidal melanocytic lesions[5] added the presence

of ultrasonographic hollowness and absence of halo to the previously found risk factors. In a recent analysis,[7] using multimodal imaging for the identification of risk factors in choroidal nevus associated with the transformation into melanoma, showed a significant correlation with thickness >2 mm, ultrasonographic hollowness (both evaluated by ocular ultrasonography), presence of subretinal fluid (evaluated by OCT), presence of vision loss symptoms (evaluated by Snellen visual acuity), presence of orange pigment (evaluated by fundus autofluorescence) and diameter >5 mm (evaluated by fundus photography).

In the present study, after multivariate analysis, we identified five factors in the initial assessment that correlated significantly with the observed final outcome. Nevertheless, the most relevant risk factor found was ultrasonographic hollowness i.e. the presence of internal low reflectivity echoes in the lesion. Odds ratio for this variable could not be calculated due to the fact that all of the cases with documented growth had this feature present.

The most relevant clinical risk factor, associated with a 25 times higher risk of growth and progression to malignancy was the presence of orange pigment, followed by presence of subretinal fluid (5 times higher risk of growth). Two protective features of small choroidal melanocytic lesions against lesion growth were also recognized in this analysis. Firstly, the presence of an amelanotic halo surround the lesion, with a 12 times lower chance of growth, and secondly, the presence of overlying drusens with a 6.5 times lower risk of growth. These findings are consistent with previously published literature. These clinically evaluated features have been frequently identified as risk factors throughout several studies. The presence of orange lipofuscin pigment that is accumulated in granules in the retinal pigment epithelium, can exhibit a toxic effect making the retinal pigment epithelium more unstable and acting as a pro-inflamatory agent which can be associated with the higher rate of growth in small choroidal lesions that have this feature. Subretinal fluid consists in the accumulation of fluid between the neurosensory retina layer and the retinal pigment epithelium layer probably occurs due to the traction force generated by the rapid enlargement of the choroidal lesion posteriorly to the retina. The presence of an amelanotic halo is related to the atrophic degeneration of the surrounding retinal pigment epithelium and the presence of drusens correlates with a slower and chronic growth of the lesion, making both of these features associated with lesion stability and inactivity, reason why they revealed protective of transformation to malignancy.

In our study, data from the different analyzed features was collected resorting to multiple imagiologic complementary diagnostic exams. The evaluation of lesions dimensions and ultrasonographic hollowness was performed by ocular high-resolution ultrasonography using a 20 MHz B-mode probe. This has a superior capability in the detection of ultrasonographic hollowness in lesions with smaller thickness compared with the 10 MHz probe more

commonly used.[16] Even though several imaging modalities were used in the evaluation of the patients in this study, most of this risk factors can be observed in simpler imaging such as indirect ophthalmoscopy and ocular ultrasonography, which are more accessible universally and easier to apply in everyday practice. Ultrasonographic hollowness, an acoustically silent zone within the tumor, we believe that it is related with lesion growth because it is a feature present in the majority of choroidal melanomas.[17] It is probably related to the ultrasonographic features of different cells types in the lesion.

Of course there are limitations in this study. The cohort of patients gathered was referred to a specialized oncology center which induces a referral bias, since perhaps only the most worrisome lesions were referred. It is a retrospective study including a small number of cases with different follow-up times, resulting in great variability which probably affected the obtained results. Nonetheless, the present study does contribute with valuable information for the identification of small choroidal melanocytic lesions that have a greater potential for growth and transformation into a malignant choroidal tumor. Further studies with a prospective design and including a larger number of cases would be beneficial to surpass these limitations.

CONCLUSIONS

The observations found in the present study are in line with the previously available literature. According to our findings, clinical features predictive of growth of a small choroidal melanocytic lesion include the presence of orange pigment, presence of subretinal fluid, and absence of an amelanotic halo and overlying drusens.

However, the most significant predictive factor pointing to growth and malignant transformation was ultrasonographic low internal reflectivity, due to its presence in every lesion included with documented growth, overcoming clinical factors found in previous studies. This feature should be carefully investigated in the initial evaluation of every small choroidal melanocytic lesion in combination with the other identified features. This will allow an individualized estimation of the risk of growth and transformation into a malignant tumor according to each lesion's characteristics, an early treatment and eventually a better prognosis.

ACKNOWLEDGMENTS

I would like to express my gratitude to my supervisor and co-supervisor, Professor Dr. Rui Proença and Dr. Cristina Fonseca, respectively, for guiding me with such mastery and for all of the time and efforts invested in this project. Without them the accomplishment of this work would not have been possible.

I dedicate a very special thanks to Dr. João Casalta for the gentleness and valuable contribution to this project.

I would also like to thank my family and closest friends for the kindness and constant words of encouragement. A special thanks to Maria do Carmo and Jaime for their ideas and collaboration in improving the quality of this work.

REFERENCES

1. Aronow ME, Topham AK, Singh AD. Uveal Melanoma: 5-Year Update on Incidence, Treatment, and Survival (SEER 1973-2013). Ocul Oncol Pathol. 2018;4(3):145-51.
2. Virgili G, Gatta G, Ciccolallo L, Capocaccia R, Biggeri A, Crocetti E, et al. Incidence of uveal melanoma in Europe. Ophthalmology. 2007;114(12):2309-15.
3. Augsburger JJ, Correa ZM, Trichopoulos N, Shaikh A. Size overlap between benign melanocytic choroidal nevi and choroidal malignant melanomas. Invest Ophthalmol Vis Sci. 2008;49(7):2823-8.
4. Singh AD, Mokashi AA, Bena JF, Jacques R, Rundle PA, Rennie IG. Small choroidal melanocytic lesions: features predictive of growth. Ophthalmology. 2006;113(6):1032-9.
5. Shields CL, Furuta M, Berman EL, Zahler JD, Hoberman DM, Dinh DH, et al. Choroidal nevus transformation into melanoma: analysis of 2514 consecutive cases. Arch Ophthalmol. 2009;127(8):981-7.
6. Factors predictive of growth and treatment of small choroidal melanoma: COMS Report No. 5. The Collaborative Ocular Melanoma Study Group. Arch Ophthalmol. 1997;115(12):1537-44.
7. Shields CL, Dalvin LA, Ancona-Lezama D, Yu MD, Di Nicola M, Williams BK, et al. CHOROIDAL NEVUS IMAGING FEATURES IN 3,806 CASES AND RISK FACTORS FOR TRANSFORMATION INTO MELANOMA IN 2,355 CASES: The 2020 Taylor R. Smith and Victor T. Curtin Lecture. Retina. 2018.
8. Dalvin LA, Shields CL, Ancona-Lezama DA, Yu MD, Di Nicola M, Williams BK, et al. Combination of multimodal imaging features predictive of choroidal nevus transformation into melanoma. Br J Ophthalmol. 2018.
9. Shields CL, Demirci H, Materin MA, Marr BP, Mashayekhi A, Shields JA. Clinical factors in the identification of small choroidal melanoma. Can J Ophthalmol. 2004;39(4):351-7.
10. Rishi P, Koundanya VV, Shields CL. Using risk factors for detection and prognostication of uveal melanoma. Indian J Ophthalmol. 2015;63(2):110-6.
11. Shields CL, Cater J, Shields JA, Singh AD, Santos MC, Carvalho C. Combination of clinical factors predictive of growth of small choroidal melanocytic tumors. Arch Ophthalmol. 2000;118(3):360-4.

12. Diener-West M, Hawkins BS, Markowitz JA, Schachat AP. A review of mortality from choroidal melanoma. II. A meta-analysis of 5-year mortality rates following enucleation, 1966 through 1988. Arch Ophthalmol. 1992;110(2):245-50.

13. Singh AD, Kalyani P, Topham A. Estimating the risk of malignant transformation of a choroidal nevus. Ophthalmology. 2005;112(10):1784-9.

14. Butler P, Char DH, Zarbin M, Kroll S. Natural history of indeterminate pigmented choroidal tumors. Ophthalmology. 1994;101(4):710-6; discussion 7.

15. Shields CL, Shields JA, Kiratli H, De Potter P, Cater JR. Risk factors for growth and metastasis of small choroidal melanocytic lesions. Ophthalmology. 1995;102(9):1351-61.

16. Pineiro-Ces A, Rodriguez Alvarez MJ, Santiago M, Bande M, Pardo M, Capeans C, et al. Detecting ultrasonographic hollowness in small choroidal melanocytic tumors using 10 MHz and 20 MHz ultrasonography: a comparative study. Graefes Arch Clin Exp Ophthalmol. 2014;252(12):2005-11.

17. Singh P, Singh A. Choroidal melanoma. Oman J Ophthalmol. 2012;5(1):3-9.