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The effect of fish consumption on macula structure and function of healthy individuals: an OCT and mfERG study

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Abstract. – OBJECTIVE: The study was designed to investigate the potential effect of fish consumption on macular structure and function of healthy individuals.

PATIENTS AND METHODS: The participants were Greek, who used to consume less than one portion of fish per week since their childhood. All participants underwent body mass index (BMI) measurements and ophthalmological examination. At their first examination, they were asked to consume at least 2 portions of fish per week over a period of 8 weeks, after which all the measurements were repeated.

RESULTS: Eighteen healthy individuals (36 eyes) participated in this study. The central macular thickness was reduced, while the amplitudes in the foveal and parafoveal area were increased after the fish consumption. However, all measurements remained within the normal range at both visits.

CONCLUSIONS: Regular fish consumption could enhance the structural and functional status of the macula.

Key Words:
Body mass index, Central macular thickness, Electroretinography, Fish consumption, Macular responses, Polyunsaturated fatty acids.

Introduction

The fovea is the central part of the macula, exhibiting the highest concentration of light-sensitive cone photoreceptors, and being responsible for detailed central and color vision. The macula houses three carotenoid pigments, lutein, zeaxanthin, and meso-zeaxanthin, which are collectively referred to as macular pigment[1]. Increasing evidence suggests that macular pigment eliminates the adverse impact of glare disability, light scatter and chromatic aberration, improving contrast sensitivity, while it exhibits antioxidant and anti-inflammatory properties[1,2]. The adaptation our daily dietary to foods enhancing macular pigment could protect the macula from oxidative stress and degenerative lesions[1-4].

Eggs, colored fruits, and vegetables (such as spinach and kale) are the basic dietary sources of lutein and zeaxanthin, whereas meso-zeaxanthin is derived from the isomerization of lutein[1,2]. Fish is a major dietary source of long-chain n-3 polyunsaturated fatty acids (n-3 PUFAs), predominantly of eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA), but it also contains high protein, selenium, zinc, vitamin A and D concentrations. Long chain n-3 and n-6 PUFAs are important in membrane structure and function, including retinal and neural membranes[5]. The plasma levels of omega-3 long-chain PUFAs contained in fish, especially docosapentaenoic acid (DPA) and EPA, have been significantly and positively correlated with macular pigment optical density (MPOD)[6]. DHA represents approximately 50-60% of the total fatty acid content within the outer segments of rod photoreceptors, regulating their renewal and differentiation, as well as the regeneration of rhodopsin. Moreover, DHA exhibits anti-angiogenetic and anti-inflammatory properties, while it prevents the selective apoptotic death of photoreceptors[7]. Indeed, the regular consumption of DHA, EPA, and

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fish has been associated with a 35-45% lower risk of visually-significant Age-Related Macular Degeneration (AMD)[6,8]. Chong et al[9] revealed the lower risk of both early and advanced AMD in people eating fish at least twice a week.

The implication of fish's PUFA in membranes of photoreceptors and visual signal transduction as well as the possible protecting effect against AMD were the motives to investigate the impact of fish consumption in visual performance of healthy individuals.

Patients and Methods

Participants

Potential participants were recruited for this study and were all living in the Attica area, Greece. They were eligible to take part if they met the following criteria: 1. Age between 30 and 65 years; 2. Body mass index (BMI) (i.e., weight in kg/ height in m^2) between 24.0 and 31.0 kg/m^2; 3. Fish consumption less than one portion (i.e. 150 g of cooked fish) per week since their childhood. Exclusion criteria included: 1. Pregnancy; 2. Weight loss diet; 3. Reception of dietary supplements; 4. Treatment for any medical disorder; and 5. Any eye disease or eye surgical procedure. All participants underwent anthropometric measurements and ophthalmological examination. At their 1st examination, they were asked to consume at least 1 portion (500 g) of fish (Greek gilthead) twice a week over a period of 8 weeks, after which all the measurements were repeated. Fish was given to the participants for free, and they came to Harokopeio University to get it. Fish was prepared, cooked and consumed at Harokopeio facilities. No adverse report was reported by the participants. Besides the fish consumption, the participants were not asked to limit or modify their daily diet.

The study was performed in accordance with the tenets of the Declaration of Helsinki and the protocol used was approved by the Ethics Committee of the General Hospital of Athens G. Gennimatas. Informed consent was obtained from all individual participants included in the study.

Anthropometric Measurements

Anthropometry was carried out on both visits. Weight was measured to the nearest 0.1 kg using a digital scale and height to the nearest 0.1 cm using a stadiometer with head in horizontal Frankfurt plane. Both measurements were taken with the subject in light clothing and without shoes. Body mass index was then calculated as weight (kg) divided by height squared (m^2). The anthropometric measurements were performed in the Department of Science of Nutrition and Dietetics of Harokopeio University.

Ophthalmological Examination

All participants submitted to ocular examination based on visual acuity evaluation (use of Snellen chart), optical coherence tomography (OCT) retinal thickness measurements, and multifocal electroretinography (mfERG) records at 1st visit and 2nd (8 weeks after the fish consumption) visit. All measurements were carried out in the 1st Department of Ophthalmology in the General Hospital of Athens G. Gennimatas. We studied both eyes (36 eyes) of eighteen healthy individuals. The BCVA in each visit was calculated as the mean best-corrected visual acuity (BCVA) of both eyes.

The *OCT examination* was performed by the same well-trained and experienced operator, using Heidelberg Spectralis (Spectralis HRA+OCT, Heidelberg Engineering Inc., München, Germany). During the procedure on the Spectralis OCT, subjects were asked to fixate on an internal fixation target to increase the chance of a well-centered scan at the fovea. The subjects were not repositioned nor the instrument realigned during the whole scanning procedure, in order to keep the measurement conditions as constant as possible. Before the examination, the pupils were dilated with drops containing 0.5% tropicamide and 2.5% phenylephrine. The central macular thickness (CMT) in the Spectralis OCT was calculated as the distance between the first signal from the vitreoretinal interface and the signal from the outer border of the RPE. To analyze CRT, a software algorithm of the Spectralis OCT interpolating thickness of the area between the scans was used. It provides a circular map analysis in which the average thickness is displayed as a color code or numeric values in nine ETDRS (Early Treatment Diabetes Retinopathy Study). The ETDRS map consists of three concentric rings with diameters 1, 3, and 6 mm, known from Stratus OCT. CMT was measured for both eyes at the 1st and 2nd visit, and the mean value of both eyes (in each visit) was used in statistical analysis.

The mfERG records were carried out according to the guidelines for a basic mfERG by the International Society for Clinical Electrophysiology of Vision ISCEV10, based on the EP-1000 model (Tomey, Nagoya, Japan). The pupils of the

Table I. The data of retinal responses amplitudes (mfD, in nV/deg2) and latencies (mfT, in ms) at 1st and 2nd visit (after fish consumption), as recorded by mfERG.

	Mean	SD	*p*
mf1D1 (nV/deg^2)	225.667	129.943	<0.001
mf1D2 (nV/deg^2)	267.806	118.448	
mf1T1 (ms)	41.592	0.6196	0.292
mf1T2 (ms)	41.653	0.6328	
mf2D1 (nV/deg^2)	57.639	36.332	<0.001
mf2D2 (nV/deg^2)	88.250	39.192	
mf2T1 (ms)	39.903	22.614	<0.001
mf2T2 (ms)	39.617	20.972	

SD = standard deviation; mf1D1 = amplitude in ring 1 at 1st visit; mf1D2 = amplitude in ring 1 at 2nd visit; mf1T1 = latency in ring 1 at 1st visit; mf1T2 = latency in ring 1 at 2nd visit; mf2D1 = amplitude in ring 2 at 1st visit; mf2D2 = amplitude in ring 2 at 2nd visit; mf2T1 = latency in ring 2 at 1st visit; mf2T2 = latency in ring 2 at 2nd visit.

patients were dilated with tropicamide 0.5% and phenylephrine 5% and the eyes were optically corrected for near vision so that the patients could see clearly the small fixation spot in the center of the stimulus matrix. For the signal acquisition, a bipolar contact lens was used, in which the active and the reference electrodes were incorporated in the contact lens. The ground electrode was attached to the earlobe. The fellow eye was closed and the duration of the data acquisition was 8 minutes, divided into eight sessions of 60 seconds. Retinal irritation was performed by an array of 61 hexagonal elements that were displayed on a cathode ray tube color monitor, driven at a frame of 75 Hz. These hexagons were scaled in size (central hexagons are smaller than peripheral) to produce approximately the same signal intensity in all areas of the retina and each of their elements has a 50% chance of being illuminated every time the frame changes. Each stimulus element flickered between black and white at a rate of 75 Hz, controlled by a predetermined m-sequence. The fixation point was located at the center (in a 20°-25° radius) and within a field of 40°-50° diameter, in order to include the blind spot.

Applying the ring pattern, the mfERG stimuli location and anatomic areas corresponded roughly as follows: ring 1 to the fovea (0°-2°) and ring 2 to the parafovea (2°-7°). The responses were summed with increased eccentricity from the fovea and the amplitude of the summed responses was divided by the total area of the hexagons in that ring. Cross-correlation between the m-sequence for a particular area and the single raw trace recording was performed. The stretch factor that was used was equal to 10.5, which is the most widely used figure. These averages give a more accurate view of the relative response densities of each group. The response amplitude per unit area or response density (nV/deg^2) is the measure of expression and it appears the maximum value in the fovea. The mfERG recordings, consisting of the retinal response density (nV/deg^2) and P1 latency (ms), were evaluated in ring 1 and 2 of both eyes at the 1st and 2nd visit. The mean value of amplitude and latency for both eyes (in each visit) was used in statistical analysis.

Statistical Analysis

The statistical program IBM SPSS Statistics 22.0 (IBM Corp. IBM SPSS Statistics for Windows, Armonk, NY, USA) was used for the analysis. Descriptive analysis of all parameters was first carried out, while non-parametric analysis Kolmogorov-Smirnov was used to check the normal distribution. The paired two-tailed *t*-test and the non-parametric Wilcoxon matched-pairs signed-rank test were used to calculate the significance of the mean differences between baseline values and those after the fish consumption. Bivariate correlation techniques were used to assess the relation between age, anthropometric and ocular measurements. Linear mixed-effects models were used to detect any relation between the central macular thickness and the mfERG recordings (amplitude). A *p*-value less than 0.05 was considered to indicate statistical significance.

Results

Anthropometric Measurements and Blood Profile

Eighteen healthy individuals (36 eyes) with mean age 45.3±8.7 years, including 8 males and 10 females, participated in this study. The mean

Table II. Association of the amplitudes between the retinal responses in the foveal area at 2nd visit (mf1D2) and the baseline amplitude (mf1D1), the baseline central macular thickness (CMT1) and the central macular thickness after the fish consumption (CMT2).

Explanatory variables	b coefficients	*p*-value	95% CI
mf1D1	0.513	0.028	0.065-0.960
CMT1	-1.299	0.088	-2.820-0.223
CMT2	1.818	0.103	-0.417-4.053
Significance of regression equation	F (3,14) = 3.963, p = 0.031		
R^2	0.459		

CI = confidence interval; CMT1 = central macular thickness at 1st visit; CMT2 = central macular thickness at 2nd visit; mf1D1 = amplitude in ring 1 at 1st visit; mf1D2 = amplitude in ring 1 at 2nd visit.

BMI, as calculated in the 1st visit and 2nd visit, was 29.1±3.6 kg/m^2 and 29.4±3.7 kg/m^2, respectively. No statistically significant difference was detected between the two measurements of BMI ($p = 0.112$).

Visual Acuity Measurements

No significant differences in BCVA were observed between the two visits (1st visit: 0.03±0.03 logMAR, 2nd visit: 0.03±0.03 logMAR, Wilcoxon matched-pairs signed-rank test: $p = 1.000$). The visual acuity was significantly, positively and strongly correlated with age at both visits (Spearman's correlations, 1st visit: r = 0.865, $p < 0.001$, 2nd visit: r=0.865, $p < 0.001$).

Evaluation of the Central Macular Thickness

The values (median ± SD) of CMT, as recorded by OCT for both eyes, seem to be reduced after 8 weeks of fish consumption (1st visit: CMT1 = 192.0±13.3 μm and 2nd visit: CMT2 = 188.4±9.2 μm). A significant decrease in central macular thickness was found after the fish diet, but the CMT values remained within normal range (paired 2-tailed *t*-test: t(17) = 3.078, $p = 0.007$). Also, a significant, strong, and positive correlation of CMT values between the two visits was noted (Pearson correlation, r = 0.969, $p < 0.001$). The central macular thickness at both visits were significantly, positively and moderately correlated with amplitude in ring 1 at 1st visit (Pearson correlation, CMT1-mf1D1: r = 0.487, $p = 0.041$, CMT2-mf1D1: r = 0.511, $p = 0.030$).

Records of mfERG

The amplitudes of mfERG responses, which were elevated after the fish consumption, as well as the latencies of the responses for both rings (foveal and parafoveal area), are displayed in Table I. The improved amplitudes of retinal responses were assessed to be statistically significant for both retinal areas (foveal and parafoveal area) after the fish consumption (paired 2-tailed *t*-test, ring 1: t(17) = -15.537, $p < 0.001$, ring 2: t(17) = -24.333, $p < 0.001$). On the other hand, the differences in latencies were not found to be significant between the two visits (paired 2-tailed *t*-test, ring 1: t (17) = -1.087, $p = 0.292$, ring 2: t(17) = 0.808, $p = 0.430$). A significant, moderate, and positive correlation of amplitude in ring 1 values between the two visits was found (Pearson correlation, r = 0.574, $p = 0.013$). Besides the observed alterations in retinal responses, all the values in both rings and at both visits were within normal range.

A moderately significant equitation was found in Model 1, where the outcome variable was the amplitude in the foveal area after the fish consumption and the explanatory variables were the amplitude in the foveal area at the 1st visit (mf1D2) and the central macular thickness at both visits (CMT1, CMT2). However, only mf1D2 was estimated to be a significant predictor (Table II).

Discussion

Assessing the impact of fish consumption on macular morphology and function, we noted that the CMT was reduced after the fish consumption, but it remained within normal range. Moreover, the values of CMT in both visits were strongly correlated, while they both related to amplitude in ring 1 at 1st visit. Although the retinal responses were within normal range at both visits, the amplitudes in the foveal and parafoveal area were increased after the fish consumption. The amplitudes at both visits were significantly correlated, while the amplitude in the foveal area after the fish diet was associated with the corresponding one at the 1st visit and the CMT at both visits. Our results are consistent with previous studies. It has been already referred that macular pigment optical density is positively correlated with the plasma levels of omega-3 long-chain

PUFAs contained in fish[6]. High dietary intakes of n3 LC-PUFAs seem to reduce the risk of for late AMD (neovascular and/or atrophic) by 38%, according to the meta-analysis performed by Chong et al[9]. The relative risk of AMD among women who consumed at least one portion of fish per week was 0.58, compared to those who consumed less fish[10]. Merle et al[11] associated that plasma total n3 PUFAs with a reduced risk for late AMD (OR = 0.62), including both atrophic (OR = 0.50, $p = 0.01$) and neovascular (OR = 0.64, $p = 0.06$) types.

The Blue Mountains Eye Study[12] and the EUREYE study[13] elucidated that the consumption of at least 1 serving of fish per week reduced the risk of late and neovascular AMD. Although omega-3 fatty acids contained in fish protect against advanced AMD, they were not found to prevent pigment abnormalities and large drusen formation[14,15]. The LUNA study supported the augmentation of MPOD at 0.5°, following the consumption of lutein, zeaxanthin, and antioxidants (vitamin C and E, zinc, selenium) for 6 months[16]. The improvement of retinal responses after fish consumption in our study is consistent with the observations of the abovementioned studies, which highlighted the beneficial effects of fish on MPOD and their protection against AMD.

Adams et al[17] assessed abdominal obesity as a risk factor for both early and advanced AMD in males, resulting in 13% and 75% increase in the odds ratio, respectively. Moreover, MPOD was negatively associated with BMI and the percentage of body fat in males. The same study noted that the increase in both body fat and fat consumption was related to decline of serum levels of lutein and zeaxanthin, for both sexes. Nevertheless, the dietary fat was not correlated with MPOD[18]. A recent study[19] elucidated no association of MPOD and obesity in the South-Indian population. In our study, no statistically significant difference was detected between the two measurements of BMI before and after the fish consumption.

A limitation of our study is the small number of participants and the short time of follow-up. Moreover, we only asked them to consume at least 2 portions of fish (gilthead seabream) without limiting their daily diet. This favored the variance in daily food consumption. The advantages of our study include the limits in participants' age, which excluded age-related macular degenerations, and the measurements of BMI. Excluding medical or ocular disorders, we managed to study the macular status of healthy individuals consuming fish. This is the first study in the literature which evaluates the impact of fish consumption on macular structure and function of healthy individuals, based on OCT and mfERG recordings.

Conclusions

This is the first study in the literature which evaluates the impact of fish consumption on macular structure and function of healthy individuals, based on OCT and mfERG records. Although the number of the participants was small in our study, we concluded that the CMT was reduced, while the amplitudes of retinal responses were increased after the fish consumption. However, both macular thickness and retinal responses were within normal range. Furthermore, a significant correlation was found between the CMT and the amplitudes at the 1st visit, which disappeared after the fish consumption. Although more studies are needed to investigate the role of fish in the visual performance of healthy individuals, regular fish consumption seems to enhance the structural and functional status of the macula.

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Conflict of Interest

The authors alone are responsible for the content and writing of this paper and have no conflict of interest to declare.

References

1) Loskutova E, Nolan J, Howard A, Beatty S. Macular pigment and its contribution to vision. Nutrients 2013; 5: 1962-1969.
2) Nolan JM, Loughman J, Akkali MC, Stack J, Scanlon G, Davison P, Beatty S. The impact of macular pigment augmentation on visual performance in normal subjects: COMPASS. Vision Res 2011; 51: 459-469.
3) Nowak JZ. Oxidative stress, polyunsaturated fatty acids-derived oxidation products and bisretinoids as potential inducers of CNS diseases: focus on age-related macular degeneration. Pharmacol Rep 2013; 65: 288-304.

4) Barker FM 2nd, Snodderly DM, Johnson EJ, Schalch W, Koepcke W, Gerss J, Neuringer M. Nutritional manipulation of primate retinas, V: effects of lutein, zeaxanthin, and n-3 fatty acids on retinal sensitivity to blue-light-induced damage. Invest Ophthalmol Vis Sci 2011; 52: 3934-3942.

5) Li D, Hu X. Fish and its multiple human health effects in times of threat to sustainability and affordability: are there alternatives? Asia Pac J Clin Nutr 2009; 18: 553-563.

6) Delyfer MN, Buaud B, Korobelnik JF, Rougier MB, Schalch W, Etheve S, Vaysse C, Combe N, Goff ML, Wolf-Schnurrbusch UE, Wolf S, Barberger-Gateau P, Delcourt C. Association of macular pigment density with plasma ω-3 fatty acids: the PIMAVOSA study. Invest Ophthalmol Vis Sci 2012; 53: 1204-1210.

7) Queroues G, Forte R, Souied EH. Retina and omega-3. J Nutr Metab 2011; 2011: 748361.

8) Seddon JM, George S, Rosner B. Cigarette smoking, fish consumption, omega-3 fatty acid intake, and associations with age-related macular degeneration: the US twin study of age-related macular degeneration. Arch Ophthalmol 2006; 124: 995-1001.

9) Chong EW, Kreis AJ, Wong TY, Simpson JA, Guymer RH. Dietary omega-3 fatty acid and fish intake in the primary prevention of age-related macular degeneration: a systematic review and meta-analysis. Arch Ophthalmol 2008; 126: 826-833.

10) Christen WG, Schaumberg DA, Glynn RJ, Buring JE. Dietary ω-3 fatty acid and fish intake and incident age-related macular degeneration in women. Arch Ophthalmol 2011; 129: 921-929.

11) Merle BM, Delyfer MN, Korobelnik JF, Rougier MB, Malet F, Féart C, Le Goff M, Peuchant E, Letenneur L, Dartigues JF, Colin J, Barberger-Gateau P, Delcourt C. High concentrations of plasma n3 fatty acids are associated with decreased riskfor late age-related macular degeneration. J Nutr 2013; 143: 505-511.

12) Joachim N, Mitchell P, Burlutsky G, Kifley A, Wang JJ. The incidence and progression of age-related macular degeneration over 15 years: the Blue Mountains Eye Study. Ophthalmology 2015; 122: 2482-2489.

13) Augood C, Chakravarthy U, Young I, Vioque J, de Jong PT, Bentham G, Rahu M, Seland J, Soubrane G, Tomazzoli L, Topouzis F, Vingerling JR, Fletcher AE. Oily fish consumption, dietary docosahexaenoic acid and eicosapentaenoic acid intakes, and associations with neovascular age-related macular degeneration. Am J Clin Nutr 2008; 88: 398-406.

14) Amirul Islam FM, Chong EW, Hodge AM, Guymer RH, Aung KZ, Makeyeva GA, Baird PN, Hopper JL, English DR, Giles GG, Robman LD. Dietary patterns and their associations with age-related macular degeneration: the Melbourne collaborative cohort study. Ophthalmology 2014; 121: 1428-1434.

15) Swenor BK, Bressler S, Caulfield L, West SK. The impact of fish and shellfish consumption on age-related macular degeneration. Ophthalmology 2010; 117: 2395-2401.

16) Trieschmann M, Beatty S, Nolan JM, Hense HW, Heimes B, Austermann U, Fobker M, Pauleikhoff D. Changes in macular pigment optical density and serum concentrations of its constituent carotenoids following supplemental lutein and zeaxanthin: the LUNA study. Exp Eye Res 2007; 84: 718-728.

17) Adams MK, Simpson JA, Aung KZ, Makeyeva GA, Giles GG, English DR, Hopper J, Guymer RH, Baird PN, Robman LD. Abdominal obesity and age-related macular degeneration. Am J Epidemiol 2011; 173: 1246-1255.

18) Nolan J, O'Donovan O, Kavanagh H, Stack J, Harrison M, Muldoon A, Mellerio J, Beatty S. Macular pigment and percentage of body fat. Invest Ophthalmol Vis Sci 2004; 45: 3940-3950.

19) Gupta A, Raman R, Biswas S, Rajan R, Kulothungan V, Sharma T. Association between various types of obesity and macular pigment optical density. Eye (Lond) 2012; 26: 259-266.