Intravitreal Anti–Vascular Endothelial Growth Factor Therapy and Retinal Nerve Fiber Layer Loss in Eyes With Age-Related Macular Degeneration: A Meta-Analysis

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Purpose. Clinical study findings regarding the association between repeated injections of intravitreal anti–vascular endothelial growth factor (VEGF) and the risk of retinal nerve fiber layer (RNFL) thinning in patients with age-related macular degeneration (AMD) have been inconsistent. We investigated this association by using a meta-analysis.

Methods. In August 2015, we systematically reviewed PubMed, EMBASE, and the Cochrane Central Register of Controlled Trials. Two independent evaluators identified eligible articles by using predetermined selection criteria. Average RNFL thickness before and after intravitreal anti-VEGF injections was examined by using data obtained at baseline and at the last follow-up visit.

Results. Six studies on 288 eyes were ultimately included. The meta-analysis revealed that average RNFL thickness following repeated anti-VEGF injections was not significantly different from baseline (mean difference [MD] = −0.171, 95% confidence interval [CI]: −0.371 to 0.029, $P = 0.093$) or control group measurements (MD = −0.091, 95% CI: −0.517 to 0.335, $P = 0.674$). However, subgroup analyses by the methodologic quality of study revealed a significant RNFL thickness loss in two low-biased, controlled experimental studies (MD = −0.534, 95% CI: −0.783 to −0.286, $P = 0.001$), but not in four observational studies (MD = −0.038, 95% CI: −0.171 to 0.095, $P = 0.576$).

Conclusions. There was no association between anti-VEGF injections and RNFL thickness changes when all studies were examined together. However, when two low-biased, controlled clinical trials were separately examined, repeated anti-VEGF injection was associated with RNFL loss. Large-scale, prospective studies are needed to determine long-term effects of anti-VEGF treatments on the RNFL in AMD patients.

Keywords: age-related macular degeneration, anti–vascular endothelial growth factor, retinal nerve fiber layer

Age-related macular degeneration (AMD) is a major cause of severe visual impairment in elderly populations worldwide.[1,2] Because the prevalence of AMD increases as longevity increases, treating it has become more important. Treating exudative AMD with intravitreal anti–vascular endothelial growth factor (VEGF) agents has been a major breakthrough in macular therapeutics.[3–6] In fact, 90% of patients maintained visual acuity and 30% of patients reported an increase in visual acuity with anti-VEGF treatment.[6–9] However, because of a short efficacy duration, repeated anti-VEGF injections are needed to maintain the antiangiogenic effect that keeps this chronic condition under control.

Recent interest has arisen on the long-term safety of repeated anti-VEGF injections required for treating AMD. It is well known that VEGF is a neurotrophic factor present in the central nervous system and retina, including the retinal nerve fiber layer (RNFL).[10–12] Theoretically, chronic suppression of neurotrophic cytokines may result in harmful downstream effects on the RNFL. In addition, reports[13–16] have suggested that intravitreal anti-VEGF injections are associated with both transient and sustained intraocular pressure (IOP) elevations. These episodic IOP fluctuations could also lead to RNFL damage. Using an experimental animal model of glaucoma, Foxton et al.[17] have demonstrated that VEGF acts directly on retinal ganglion cells to promote cell survival and that VEGF blockade significantly exacerbates neuronal cell death. Martinez-de-la-Casa et al.[18] report that, in AMD patients, repeated intravitreal anti-VEGF injections result in RNFL deterioration because of anti-VEGF agent toxicity and induced IOP fluctuations. In contrast, Horsley et al.[19] and Entezari et al.[20] have not found that repeated intravitreal anti-VEGF injections lead to significant RNFL thickness changes in AMD patients. Additionally, Shin et al.[21] report that retinal disease itself could be a cause of RNFL loss rather than an effect of anti-VEFG treatment.

Although it is generally accepted that anti-VEGF agents are effective as long as they are frequently injected, some controversy remains over whether or not repeated anti-VEGF injections have adverse effects on RNFL thickness.

To the best of our knowledge, no previous studies have evaluated the long-term effects of anti-VEGF agents on the RNFL with a meta-analysis. Therefore, we performed a meta-analysis by using existing relevant studies to estimate the effect of repeated intravitreal anti-VEGF injections on RNFL thickness in patients with exudative AMD. We hope that our study results will help retina and glaucoma specialists forecast long-term RNFL changes and guide patients toward the correct treatment decisions, particularly for patients with both wet AMD and glaucoma.

Materials and Methods

A systematic review was performed according to the established guidelines for reporting systematic reviews.[22,23] No funds were used in the conduct and completion of this study.

Literature Search

The PubMed, EMBASE, and Cochrane Central Register of Controlled Trials databases were systematically searched from database inception dates through July 10, 2015. No language restrictions were placed on the search. The following keywords were used to search all databases: macular degeneration, VEGF or avastin or bevacizumab or lucentis or ranibizumab or eylea or aflibercept or macugen or pegaptanib, and retinal nerve or inner retina. Article titles and abstracts were examined to exclude clearly unrelated articles. The full text of the remaining papers was evaluated to determine relevancy. In addition, relevant studies cited in selected papers and published reviews were considered for inclusion in analyses. All literature searches were performed by two independent reviewers (HJS, HC).

Eligibility Criteria

Studies fulfilling the following inclusion criteria were included in our meta-analysis: (1) published study evaluating RNFL thickness in eyes treated with repeated intravitreal injection of any anti-VEGF agent (bevacizumab, ranibizumab, aflibercept, and pegaptanib) for exudative AMD; (2) clinical trial, prospective or retrospective cohort study, or case-control study; (3) satisfactory quality article that measured RNFL thickness by using optical coherence tomography (OCT) before the first injection and at the last follow-up visit; and (4) satisfactory OCT scan quality recorded in the article. Studies were excluded from the meta-analysis if any of the following were true: (1) study included eyes with a history that could potentially affect the RNFL (e.g., glaucoma, retinal disease other than AMD, intraocular laser procedures, and vitrectomy); (3) study subjects received three or fewer injections; (4) mean subject observation period was shorter than 12 months; and (5) sample size was smaller than 20 eyes.

Data Extraction

Two authors (HJS, HC) independently extracted the following data: first author name, publication year, study design, subjects' age, follow-up duration, sample size, type of anti-VEGF agent, number of injections, OCT type, average of overall peripapillary RNFL thickness (360° entire measurement), and temporal quadrant RNFL thickness. Discrepancies in data extraction were jointly reviewed until a consensus was reached with a third reviewer (HCK).

Quality Assessment

Study quality was independently evaluated by two authors (HJS, HC) using the Newcastle-Ottawa scales (NOS) for examining the quality of cohort studies.[24] The NOS assesses study quality by using the following three categories: selection, comparability, and outcomes. Nine points (stars) on the NOS reflect the highest study quality. From quality assessment standards used in previous meta-analyses,[25] a study awarded six or more stars was defined as a high-quality study in our meta-analysis. Any quality assessment disagreements were resolved after discussion with and re-evaluation by a third reviewer (HCK).

Main and Subgroup Analysis

We primarily assessed the cumulative effect of intravitreal anti-VEGF injections on the RNFL in injected eyes by comparing overall RNFL thickness measured at baseline to that measured at the last examination. The mean change in RNFL thickness was also evaluated in the fellow, untreated eye to serve as a control. Because retinal edema and/or scarring might have contained the nerve fiber layer and contributed to peripapillary RNFL thickness, the temporal quadrant was also examined to detect this effect. Eyes were also divided into subgroups by the number of anti-VEGF injections administered ($\geq$10 injections and $<$10 injections) and the study design (experimental study group [ESG] and observational study group [OSG]).

Statistical Analyses

Statistical analyses were performed by using Comprehensive Meta-Analysis version 2.0 (Biostat, Englewood, NJ, USA). Heterogeneity across studies was examined by using I^2, which measures the percentage of the total variation across studies.[26] Substantial heterogeneity was defined as an I^2 value greater than 50%.[27] In the absence of significant heterogeneity, a fixed-effect model was used. When there was statistical heterogeneity, a random-effects model was used to estimate differences between the injection and control groups. A sensitivity analysis was conducted to investigate the robustness of our conclusions by eliminating one study in the meta-analysis at a time to determine its effect on the pooled data. The publication bias was evaluated by using both Egger's regression intercept test and Begg-Mazumdar rank correlation test.[28] A funnel plot was built to assess this bias by using standard error and mean difference.[29,30] A two-tailed P value of <0.05 was considered significant for this meta-analysis.

Results

Literature Search

A flow diagram showing how relevant studies were identified is presented in Figure 1. We found a total of eight potentially relevant papers by focusing on RNFL changes in eyes treated with repeated intravitreal anti-VEGF injections for exudative AMD.[18–21,31–34] Among these eight publications, one non-randomized clinical trial was excluded because it had a small sample size (n = 18 eyes) and a short follow-up period (6 months)[20] and one prospective cohort study was excluded because only three anti-VEGF injections had been administered.[34] Therefore, six publications were ultimately included in the meta-analysis.

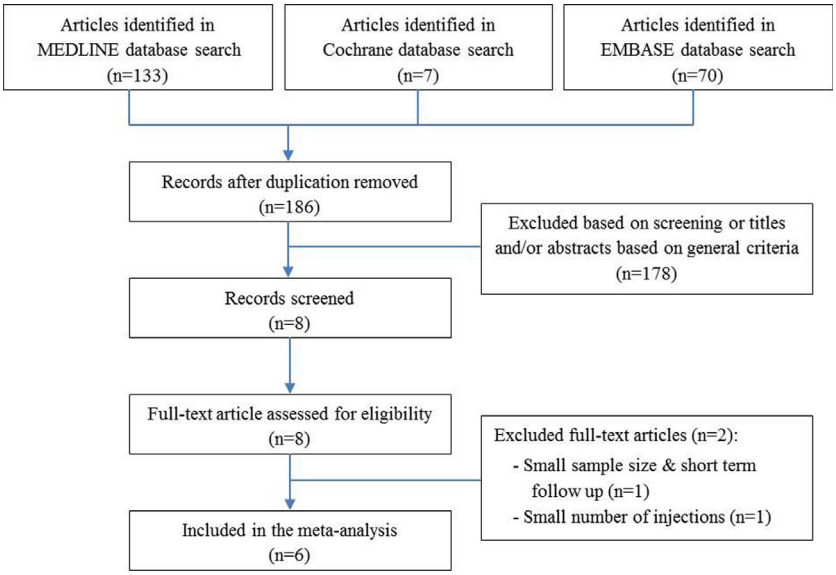

Figure 1. Flow diagram of the literature search procedure.

Study Characteristics and Quality Assessment

The characteristics of trials included in the meta-analysis are summarized in Table 1. The six studies included data from 288 eyes and were published between 2010 and 2015. Two of these studies were nonrandomized controlled clinical trials[18,32] and four were retrospective cohort studies.[19,21,31,33] In the two nonrandomized controlled clinical trials, the healthy fellow eyes that were not treated (no injections) served as the control group.[18,32] Of the four retrospective cohort studies, two had control groups that consisted of either age-matched AMD patients who did not receive intravitreal anti-VEGF thearpy[21] or randomly chosen eyes from age-matched healthy subjects.[33] The other two studies did not have a control group that was anti-VEGF treatment-naïve.[19,31] All studies treated the patients on the basis of a PRN ("as-needed") protocol. Of the 288 eyes, 176 eyes (61%) received ranibizumab alone, 69 eyes (24%) received bevacizumab alone, 34 eyes (12%) received ranibizumab and bevacizumab, and 9 eyes (3%) received ranibizumab and/or bevacizumab combined with pegaptanib. Measurements of RNFL thickness were carried out by using only high-quality images, defined as a signal strength of at least 7 in Stratus OCT images,[19,31] a quality score greater than 16 dB in Spectralis OCT images,[18,21,32] and a signal strength score of at least 7 in Cirrus OCT images.[33]

Table 2 shows RNFL measurements from and results of individual studies. Among the six included studies, four did not report a significant change in RNFL thickness after repeated anti-VEGF injections.[19,21,31,33] However, one study reported a significant reduction in RNFL thickness from baseline in the injection group after 12 months of follow-up[18] and another reported a significant reduction from baseline in RNFL thickness in both the injection and control (untreated fellow eyes) groups after 12 months of follow-up.[32] None of the patients in our meta-analysis required IOP-lowering medication or had a sustained increase in IOP or inflammation that required intervention.

Study quality was assessed by using the NOS star system (range, 0–9 stars), which revealed a mean quality score 7.8 stars for the six included studies (Table 1). All studies were awarded either three or four stars in the selection category because they all attempted to adjust for confounding factors by using restrictive inclusion criteria. For the comparability category, four studies[19,21,31,33] were awarded one star and two studies were awarded two stars[18,32] because they satisfied both comparability criteria. All included studies were awarded three stars for the outcome assessment category.

Meta-Analysis

Figure 2A shows the mean change from baseline in RNFL thickness in injected eyes for all six studies (n = 288 eyes, mean number of injections = 7.8, mean follow-up period = 20.4 months).[18,19,21,31–33] There was no significant change from the baseline in RNFL thickness after anti-VEGF injection (mean difference [MD] = –0.171, 95% confidence interval [CI]: –0.371 to 0.029, P = 0.093). Heterogeneity between studies was detected (I^2 = 62.5%), so we used the random-effect model analysis.

Figure 2B shows the comparison between mean RNFL thickness changes in the injection and untreated control eyes. Four of the six included studies had control groups that did not receive anti-VEGF injections.[18,21,32,33] These four studies had a total of 182 eyes that received an average of 6.6 injections. These eyes were followed up for a mean duration of 22.6 months. There was no significant difference in the change in RNFL thickness between injection and untreated control eyes (MD = –0.091, 95% CI: –0.517 to 0.335, P = 0.674). Heterogeneity between studies was detected (I^2 = 70.8%), so the random-effect model analysis was used.

A sensitivity analysis was performed by omitting one study at a time and reexamining pooled data from the remaining studies. This showed that no study had a significant effect on pooled data. Additionally, a publication bias was not observed in the selected studies (symmetric funnel plot, Begg's test: P = 0.85, Egger's test: P = 0.47; Fig. 3).

Subgroup Analysis by Study Design Type

Figure 4 shows the mean change from baseline in RNFL thickness in injected eyes for OSG and ESG studies. There was a significant difference between the change in RNFL thickness in OSG and ESG studies. The OSG studies included four retrospective cohort studies that examined a total of 217

Table 1. Characteristics and Quality of Studies Included in the Meta-Analysis

Study	Publication Time, y	Study Design	Country	No. of Patients	Mean Age, y	Anti-VEGF Type	Mean Injection Numbers	Follow-up Duration, mo	Control Group Without Anti-VEGF Injection	OCT Model	NOS Scale			
											Selection	Comparability	Outcome	Total Score
Horsley et al.[19]	2010	Retrospective, cohort	United States	41	79.2	R and/or B or P	16	27	No	Stratus	***	*	***	7
Martinez-de-la-Casa et al.[18]	2012	Nonrandomized controlled clinical trials	Spain	49	78.5	R	4.8	12	Fellow eyes	Spectralis	****	**	***	9
Sobacı et al.[31]	2013	Retrospective, cohort	Turkey	65	68.6	R or B	5.65	13.9	No	Stratus	***	*	***	7
Shin et al.[21]	2014	Retrospective, cohort	South Korea	82	70	R and/or B	5.7	23	Age-matched AMD patients	Spectralis	****	*	***	8
Parlak et al.[32]	2015	Nonrandomized controlled clinical trials	Germany	22	66.3	R	4.7	12	Fellow eyes	Spectralis	****	**	***	9
Demirel et al.[33]	2015	Retrospective, cohort	Turkey	29	73.9	R	13.9	39	Age-matched healthy subjects	Cirrus	***	*	***	7

B, bevacizumab; P, pegaptanib; R, ranibizumab; y, year; No., number; mo, month.

Table 2. The RNFL Measurements and Results of Individual Studies*

Study	Baseline				Last Follow-up				Change				Results
	Injection Group		Control Group		Injection Group		Control Group		Injection Group		Control Group		
	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	
Horsley et al.[19]	92.4	15.2	N/A		93.8	15.2	N/A		1.4	N/A	N/A		Injection group →
Martinez-de-la-Casa et al.[18]	105.7	12.2	101.8	11.6	100.2	11.0	100.5	108	−5.6	9.1	1.3	9.6	Injection groups ↓ / control group →
Sobacı et al.[31]	105.6	6.9	N/A		104.6	8.2	N/A		−1.0	N/A	N/A		Injection group →
Shin et al.[21]	98	11.7	98.8	9.0	97.5	12.1	98.2	9.8	0.47	3.22	0.62	2.68	Injection group → / control group →
Parlak et al.[32]	101.4	14.2	99.1	8.8	99.9	14.5	96.2	9.5	−1.5	3.9	−2.9	2.58	Injection group ↓ / control group ↓
Demirel et al.[33]	92.3	7.2	95.4	6.7	92.5	8.1	95.4	6.7	0.16	2.26	0	1.82	Injection group → / control group →

N/A, data not available; SD, standard deviation; ↓, significant decrease of RNFL thickness at last follow-up compared with baseline; → , no significant change of RNFL thickness at last follow-up compared with baseline.

* Unit: μm.

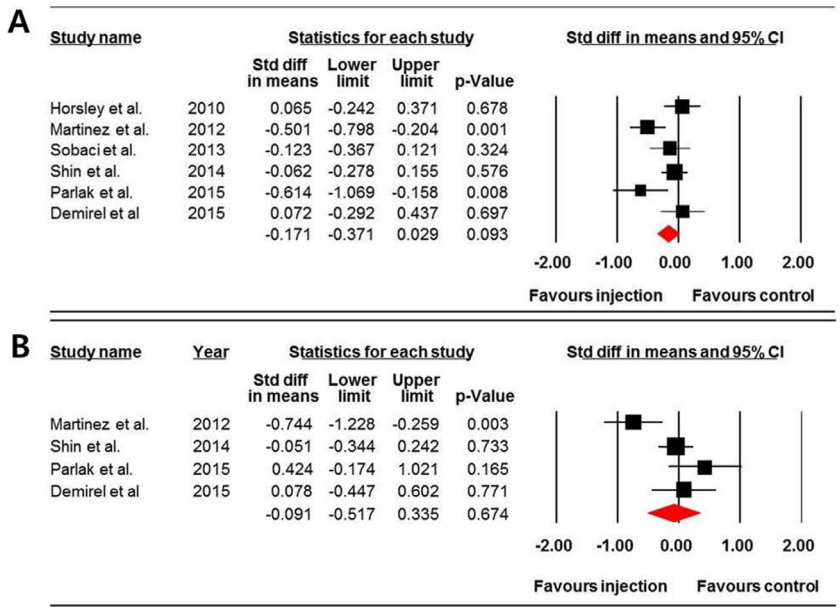

Figure 2. (**A**) Mean change from baseline in RNFL thickness after anti-VEGF injection in all included studies. (**B**) Comparison of mean RNFL thickness change between injection and untreated control eyes.

eyes.[19,21,31,33] The mean number of injections and the follow-up period were 7.9 injections and 22.6 months, respectively. There was no significant change from baseline in RNFL thickness following anti-VEGF injections in OSG studies (MD = –0.038, 95% CI: –0.171 to 0.095, $P = 0.576$). The two ESG studies were nonrandomized controlled clinical trials that examined a total of 71 eyes.[18,32] The mean number of injections and the follow-up period were 4.8 injections and 12 months, respectively. In contrast to the OSG studies, the ESG studies showed a significant reduction from baseline in

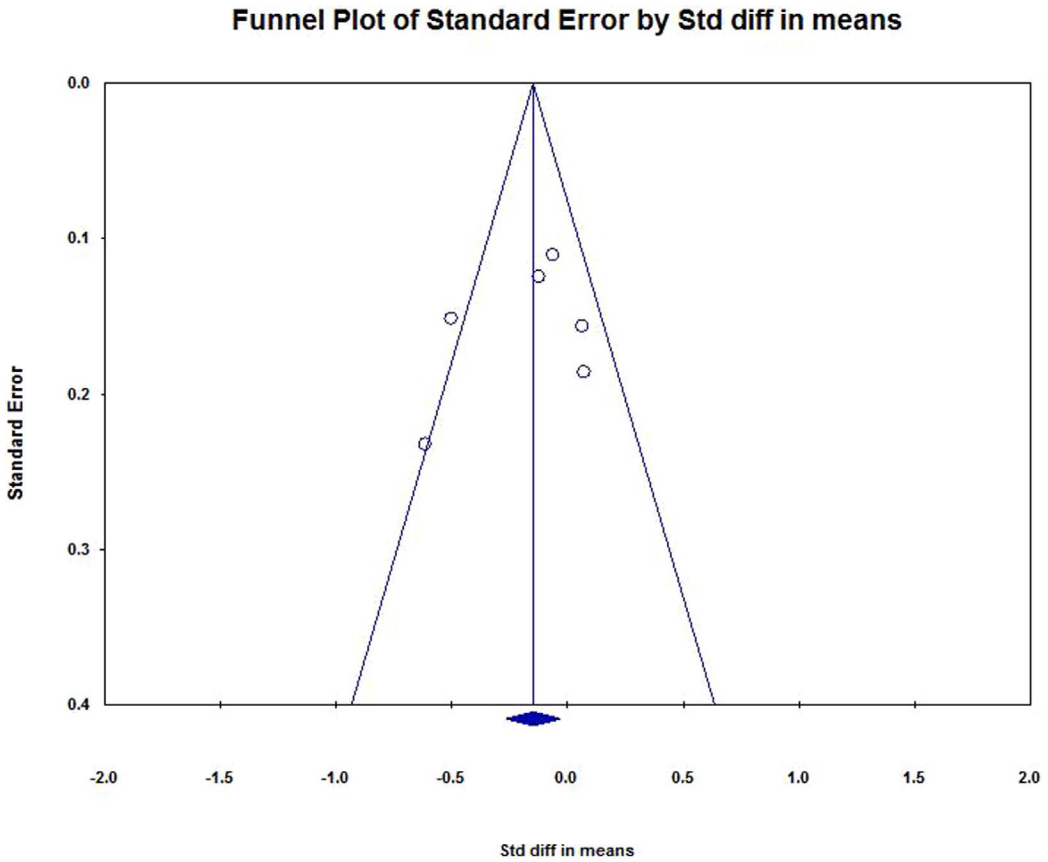

Figure 3. Funnel plot with respect to mean change from baseline in RNFL thickness after intravitreal anti-VEGF injections.

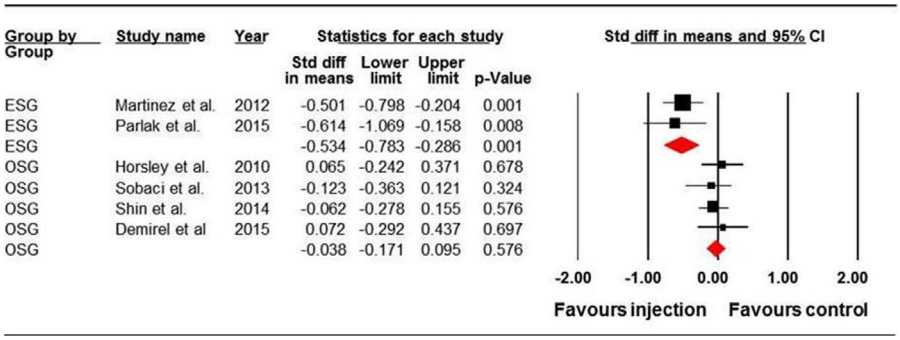

Figure 4. Subgroup analysis by study design (ESGs versus OSGs).

RNFL thickness (MD = –0.534, 95% CI: –0.783 to –0.286, P = 0.001).

Subgroup Analysis by Measurement Sector

Figure 5 shows the comparison between mean RNFL thickness changes in the entire measurement area and the temporal quadrant. Of the six included studies, two nonrandomized controlled clinical trials[18,32] and two retrospective cohort studies[21,31] examined changes in RNFL thickness in each quadrant and were included in this meta-analysis (n = 218 eyes, mean number of injections = 5.4, mean follow-up period = 16.7 months).[18,21,31,32] There was a significant change in overall RNFL thickness from baseline (MD = –0.263, 95% CI: –0.515 to –0.011, P = 0.041); this finding was largely due to the reduction in RNFL thickness observed in the two nonrandomized controlled clinical trials.[18,32] However, there was no significant change from baseline in the temporal quadrant RNFL thickness (MD = –0.107, 95% CI: –0.727 to –0.514, P = 0.736).

Subgroup Analysis by Number of Injections

Figure 6 shows the mean change from the baseline in RNFL thickness in injected eyes for both injection frequency groups (≥10 injections and <10 injections). The group that received ≥10 injections (n = 70 eyes, mean number of injections = 15.1, mean follow-up period = 32 months) came from two retrospective cohort studies.[19,33] There was no significant change from baseline in RNFL thickness (MD = –0.068, 95% CI: –0.242 to 0.378, P = 0.666). The group that received <10 injections (n = 218 eyes, mean number of injections = 5.4, mean follow-up period = 16.7 months) came from two nonrandomized controlled clinical trials[18,32] and two retrospective cohort studies.[21,31] There was a significant decrease in RNFL thickness from baseline in this group (MD = –0.250, 95% CI: –0.441 to –0.058, P = 0.011), largely because of a deceased RNFL thickness observed in the two nonrandomized controlled clinical trials.

Discussion

In a meta-analysis of all included studies, we did not identify reasons for significant concern over the effect of repeated anti-VEGF injection on the RNFL thickness when patients were carefully selected and anti-VEGF agents were used on the basis of PRN protocol (rather than monthly injections). However, we found a significant association between anti-VEGF injection and the risk for RNFL thinning in the low-biased, nonrandomized, controlled clinical trials reported by Martinez-de-la-Casa et al.[18] and Parlak et al.[32] when they were examined separately.

In the present meta-analysis, we investigated the association between anti-VEGF injections and the risk of RNFL loss by examining changes in RNFL thickness in injected eyes. Comparing RNFL changes in injected eyes to those in untreated fellow eyes may have resulted in an overlooking of RNFL thinning due to possible contralateral effects of intravitreal anti-VEGF. Intravitreal anti-VEGF injections can affect the fellow eye via the systemic circulation. Rouvas et al.[35] have treated a single eye in patients with bilateral AMD and observed clinical improvement in the contralateral eye. Similarly, Parlak et al.[32] have observed a significant RNFL thinning in both the injection eye and fellow eye after intravitreal anti-VEGF injections. In such cases, comparing injected eyes to fellow eyes may underestimate RNFL thinning in the injected eye. It is also known that RNFL thickness decreases with age at a rate of approximately 0.2 μm per year[36–38] and that the decrease in RNFL thickness was negligible during the follow-up period.[21] Therefore, comparing RNFL changes in the injected eye with age-matched healthy subjects was also less valuable. Thus, we included two

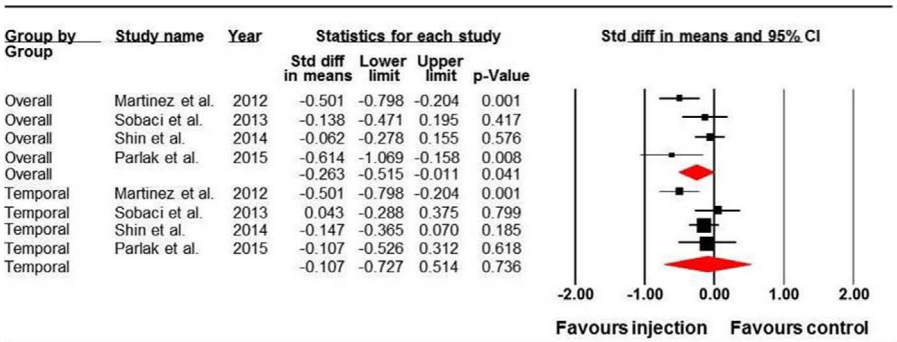

Figure 5. Subgroup analysis by measurement sector (overall versus temporal quadrant).

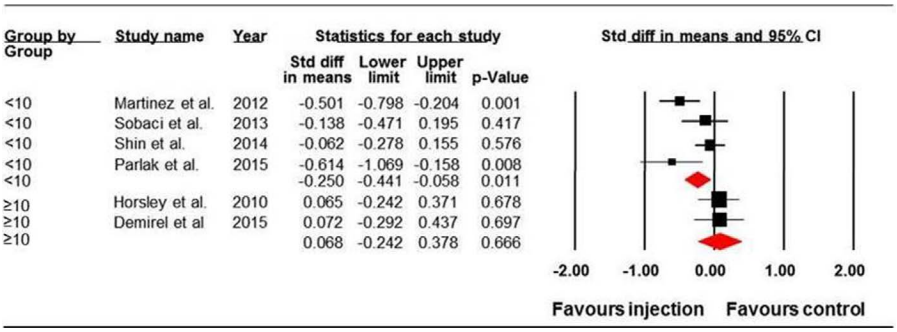

Figure 6. Subgroup analysis by the number of injections (<10 injections versus ≥10 injections).

retrospective cohort studies without untreated control groups,[19,31] but found no discrepancy in the meta-analysis result when comparing RNFL thickness changes in injected eyes with baseline analyses (Fig. 2A) and untreated control groups (Fig. 2B).

There was no randomized controlled trial included in our meta-analysis because most patients presenting to ophthalmic departments had vision problems due to macular edema secondary to exudative AMD. Clinically, it is not feasible or ethical to follow-up and observe patients in need of anti-VEGF therapy without providing proper treatment. Thus, it could be argued that nonrandomized controlled clinical trials provide the highest level of evidence in this kind of epidemiologic study.

We did find a discrepancy in the association between repeated anti-VEGF injections and the risk of RNFL thinning with different study design, which is related to the methodologic quality of research. Anti-VEGF injections were associated with an increased risk of RNFL loss in ESG studies, but not in OSG studies (Fig. 4). Experimental studies (e.g., randomized or nonrandomized controlled trials and prospective studies with predetermined eligibility criteria and outcome measures) provide a higher level of evidence than observational studies (e.g., case-control studies and cohort studies). In the included nonrandomized controlled trials, Martinez-de-la-Casa et al.[18] and Parlak et al.[32] observe a progressive RNFL thinning of −5.6 μm (mean number of injections = 4.8) and −1.5 μm (mean number of injections = 4.7), respectively, after 12 months of follow-up. Although the small sample size ($n = 71$ eyes) in the two ESG clinical trials was not large enough to sufficiently explain the hazardous effect of repeated anti-VEGF injections on the RNFL, this finding could raise caution in the era of growing interest for intravitreal anti-VEGF injections. Also, this is noteworthy for ophthalmologists, who should be cautious about administering multiple intravitreal injections.

To investigate the potential association between macular edema and changes in RNFL thickness, we compared changes in overall RNFL thickness and temporal RNFL thickness. Basically, individual studies included in our meta-analysis obtained OCT data by using strict criteria and attempted to exclude the effect of macular thickness changes on RNFL thickness. The temporal quadrant was selected because RNFL thickness in this region may be related to macular tomography changes. Alterations in the temporal RNFL thickness would be more pronounced than that in the overall RNFL thickness, if changes in overall RNFL thickness were associated with resolution of macular edema in response to anti-VEGF agents. In a subgroup meta-analysis of four of the included studies, there was no significant change from baseline in temporal RNFL thickness compared with overall RNFL thickness. Thus, we did not find a significant association between RNFL and macular thickness changes (Fig. 5). Additionally, in a subgroup analysis performed according to the number of injections, the subgroup receiving <10 injections exhibited a significant reduction in RNFL thickness, but the subgroup receiving ≥10 doses did not (Fig. 6). It should be noted that the RNFL reduction in the subgroup receiving <10 injections was dominated by two nonrandomized controlled clinical trials that reported a significant RNFL loss. Therefore, our meta-analysis did not address frequency-related side effects or cumulative doses associated with increased risk of RNFL loss.

The results of our meta-analysis should be interpreted with caution for several reasons. First, most studies targeted patients with AMD accompanied by mild to moderate macular edema. In fact, patients were excluded if they had poorly controlled AMD or macular edema that extended beyond the boundaries of RNFL measurements. Eyes that are resistant to therapy require more injections and may have different ganglion cell survival because of disease activity.[39] Thus, our meta-analysis results included only patients with mild to moderate exudation, which could have introduced a selection bias. Second, all 288 eyes included in the analyses were treated on an as-needed basis, and the degree of neurotrophic cytokine inhibition may differ from that induced by monthly injections. Because treatments occurred intermittently and included drug-free periods, the affected tissue may have had time to recover.[33] Therefore, our findings largely reflect the PRN protocol. Third, most studies used the standard anti-VEGF agent dose (1.25 mg/0.05 mL bevacizumab or 0.5 mg/0.05 mL ranibizumab). The results would likely have been different with the use of higher anti-VEGF doses as well as sustained-release formulations, which might affect the RNFL more drastically.[20,31] Finally, the examined studies included AMD patients with no other ocular pathology (e.g., glaucoma or optic neuropathy) and may only be clinically significant for patients with an initially normal nerve fiber layer thickness.

Patients at high risk for or who already have a compromised RNFL may be more susceptible to optic nerve damage from anti-VEGF injections. Park et al.[40] report significant RNFL thinning after repeated intravitreal anti-VEGF injections (for treatment of various retinal diseases) in patients with underlying glaucoma, but not in patients without glaucoma. Also, Kahook et al.[13] suggest that IOP elevations may be more likely to occur in patients with glaucoma because of a compromised outflow facility after intravitreal bevacizumab injections. Therefore, close postinjection surveillance may be warranted in these patients. The positive effect of anti-VEGF agents on visual function or maintenance in AMD patients without a compromised RNFL seems to outweigh potential adverse effects on the RNFL. However, in patients with glaucoma or considerable RNFL thinning who are scheduled to receive anti-VEGF agent injections, ophthalmologists should be vigilant about closely monitoring IOP and RNFL thickness after injections.

Although the usefulness of RNFL thickness measurements and changes in patients without glaucoma have not yet been determined, OCT measurements of RNFL thickness changes act as a surrogate marker of axonal health in clinical field and are used for the diagnosis and follow-up of neurodegenerative diseases such as optic neuritis[41] and multiple sclerosis.[42] Thus, OCT could become a viable tool for measuring the effect of anti-VEGF agents on RNFL thickness that may indirectly (via IOP fluctuations) or directly (via anti-VEGF agent neurotoxic effects) result. Additionally, different OCT systems were used in the studies included in our meta-analysis. The Cirrus OCT system generally uses a 3.46-mm-diameter scan circle to calculate circumpapillary RNFL thickness,[33] but the Stratus and Spectralis OCT systems use a 3.4-mm scan circle.[18,19,21,31,32] Although each study used different retinal segmentation algorithms and peripapillary targets for OCT measurements, the same techniques and procedures were applied within studies and both injection and control groups were examined with the same protocols. Therefore, the differences in OCT RNFL thickness measurements between studies were negligible.

The current study had several limitations. First, a relatively small number of studies were eligible to be included in our meta-analysis. Second, the follow-up period and injection intervals varied between studies, largely because of differences in retinal disease courses. Third, we did not evaluate serial RNFL thickness changes because individual studies did not provide sufficient data on RNFL changes or trends over time. Lastly, even though individual studies obtained OCT data under strict criteria and an association between RNFL and macular thickness changes was not found, it is possible that microscopic hydration of the RNFL layer secondary to macula edema masked the presence of true global RNFL thinning.

Despite these limitations, we believe that the results of this meta-analysis are clinically important and provide valuable preliminary data on this subject. In conclusion, we found conflicting results regarding the association between repeated anti-VEGF injection and RNFL loss. The presence/absence of a significant association was dependent upon study design, which is confounded with methodologic quality of the research. A meta-analysis that included only low-biased, controlled clinical trials provided possible evidence linking repeated anti-VEGF injection to an increased risk for RNFL loss. In this regard, additional well-designed, large-scale, prospective, longitudinal studies are needed to verify the results of this meta-analysis.

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