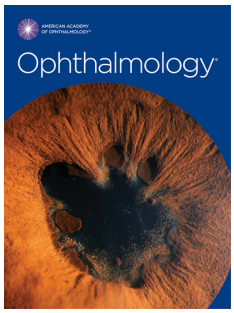
Journal Pre-proof



Placebo effect and its determinants in ocular hypotensive therapy: meta-analysis and multiple meta-regression analysis

Sooyeon Choe, PharmD, Young Kook Kim, MD, PhD, Woosuk Chung, Donghyun Ko, Marvin Lee, MD, PhD, Sung Ryul Shim, PhD, Ahnul Ha, MD

PII: S0161-6420(23)00430-X

DOI: https://doi.org/10.1016/j.ophtha.2023.06.012

Reference: OPHTHA 12443

To appear in: *Ophthalmology*

Received Date: 1 March 2023

Revised Date: 18 May 2023

Accepted Date: 13 June 2023

Please cite this article as: Choe S, Kim YK, Chung W, Ko D, Lee M, Shim SR, Ha A, Placebo effect and its determinants in ocular hypotensive therapy: meta-analysis and multiple meta-regression analysis *Ophthalmology* (2023), doi: https://doi.org/10.1016/j.ophtha.2023.06.012.

Placebo effect and its determinants in ocular hypotensive therapy: meta-analysis and multiple meta-regression analysis

Sooyeon Choe,[1,2,3] PharmD, Young Kook Kim,[1,2,4†] MD, PhD, Woosuk Chung,[5] Donghyun Ko,[5] Marvin Lee, [6] MD, PhD, Sung Ryul Shim,[7] PhD, Ahnul Ha, [8,9†] MD

Affiliations:
1. Department of Ophthalmology, Seoul National University Hospital, Seoul, Korea
2. Department of Ophthalmology, Seoul National University College of Medicine, Seoul, Korea
3. Department of Ophthalmology, Chungnam National University Hospital, Daejeon, Korea
4. EyeLight Data Science Laboratory, Seoul National University College of Medicine, Seoul, Korea
5. Department of Medicine, Seoul National University College of Medicine, Seoul, Korea
6. Allbarun eye clinic, Seoul, Korea
7. Department of Health and Medical Informatics, Kyungnam University College of Health Sciences, Changwon, Korea
8. Department of Ophthalmology, Jeju National University Hospital, Jeju-si, Korea
9. Department of Ophthalmology, Jeju National University School of Medicine, Jeju-si, Korea

† These two authors contributed equally to the study as co-corresponding authors.

Corresponding author (primary): Young Kook Kim
Address for reprints:
Department of Ophthalmology, Seoul National University College of Medicine

Financial Support: National Research Foundation of Korea (NRF) grant (no. NRF-2022R1F1A1064186). The funder had no role in the initiation or design of the study, collection of samples, analysis, interpretation of data, paper writing or submission for publication. The study and researchers are independent of the funder.

Running head: Placebo effect in ocular hypotensive therapy

Keywords: Placebo; Glaucoma; Systematic Review; Meta-analysis; Meta-regression analysis

Disclosures: There are no financial conflicts of interest to disclose.

Data Availability Statement: The data described in the article, codebook, and analytic code will be made available upon request.

This article contains additional online-only material. The following should appear online-only: Appendices 1-4, Figure S3, S4, S7-S9, and Tables S1, S4-S12.

ABBREVIATIONS

AICc = Akaike's information criterion corrected; **ES** = effect size; **IOP =** intraocular pressure; **NMA** = network meta-analysis; **OAG** = open-angle glaucoma; **OHT** = ocular hypertension; **PRISMA** = Preferred Reporting Items for Systematic Reviews and Meta-Analyses; **RCTs** = randomized clinical trials; ROB = risk of bias; **SIDE** = Separating Indirect from Direct Evidence

ABSTRACT

Topic: The placebo effect, and its potential determinants, in ocular hypotensive therapy.

Clinical Relevance: The placebo effect has been studied and documented within a wide clinical context. It remains unclear whether placebo is effective in glaucoma treatment or, if so, which factors are determinative of effect size.

Methods: Randomized controlled trials (RCTs) of topical ocular hypotensive therapy for patients with open-angle glaucoma (OAG) or ocular hypertension (OHT), conducted until June 2, 2022, were included. First, a perceived placebo effect was measured as the overall intraocular pressure (IOP; mmHg) change from the baseline. It was evaluated in terms of the effect size (ES; mean difference between the baseline and the endpoint) and then was compared with the ES as obtained from the untreated control in order to obtain true placebo effect. The primary outcome was ES based on four weeks of treatment. Meta-analysis-based statistical pooling was performed where appropriate, and 95% CIs were used for comparison. Potential placebo effect determinants were scrutinized using a multiple meta-regression model (PROSPERO: CRD42022348098).

Results: A total of 40 RCTs (7,829 eyes) with 33 placebo groups (2,055 eyes) along with 7 untreated groups (1,184 eyes) were included. Placebo was determined to be effective in lowering IOP (ES –1.30 mmHg, 95% CI, –1.75 to –0.84). This effect was superior to the effect calculated for the untreated controls by –2.27 mmHg (95% CI, –3.52 to –1.01). According to the multiple meta-regression model, the active treatment ES was a significant factor to prediction the amount of placebo effect. Placebo additionally lowered IOP by –0.45 mmHg per –1 mmHg of active treatment effect. Add-on study design and larger sample size were also associated with greater amount of placebo effect. No publication bias was evident in either a funnel plot or the Begg and Mazumbar adjusted rank correlation test result (P=0.24).

Conclusion: This meta-analysis indicates that placebo is effective in lowering IOP and is superior to the effect observed for the untreated controls. However, caution is required in interpreting the results, due to the small number of untreated-controlled trials and potential bias from the lack of direct comparison between the placebo and untreated arms.

INTRODUCTION

The placebo effect is the beneficial consequence of patients' positive expectation of their health status.[1] Variations in the ways and extents to which patients trust treatments and experience their symptoms are the main causative factors for the placebo effect; therefore, subjective health assessments generally are susceptible to such an effect.[2] However, there is mounting evidence that the placebo effect is manifested not only in subjective but also objective measures. Kirchhof et al. showed that the placebo effect can induce an immunosuppressive response, which is to say, reduced T-cell proliferation, in renal transplantation patients.[3] Also, Kemeny et al. demonstrated that employment of a placebo bronchodilator reduced bronchial hyperreactivity in patients with asthma.[4]

Glaucoma treatment currently is based on lowering of intraocular pressure (IOP), which is the only proven method to slow disease progression.[5] Thus, the importance of robust evidence on the efficacy as well as safety of IOP-lowering agents has been emphasized. Although randomized controlled trials (RCTs) with a standard treatment (e.g., timolol in glaucoma RCTs) instead of a placebo as a control (i.e., reference) have been widely conducted due to ethical concerns, use of a placebo group nonetheless is known as the most rigorous standard for evaluation of novel medical therapy efficacy.[6] Nevertheless, the placebo effect in IOP-lowering medications has not been quantified, meaning that objective and non-biased conclusions on the effectiveness of ocular hypotensive therapy for glaucoma have remained elusive. Placebo effect-size estimation may also impact upon clinical trials' sample-size calculations as well as health economics.[6]

Most studies have defined the placebo effect as a change from baseline, though distinguishing this change from the natural fluctuation of disease or regression to the mean is difficult.[7-9] This fact complicates placebo effect measurement, rendering it even more challenging. Direct comparison of the effects of active treatment, placebo and no-treatment could overcome this obstacle. And whereas RCTs typically focus on 1-at-a-time pairwise comparisons, network meta-analysis (NMA), an extended version of standard pairwise meta-analysis, enables the comparison of treatments that have never been subjected to direct comparison.[10, 11] Thus motivated, we conducted (1) a conventional meta-analysis, (2) an NMA and (3) a multiple meta-regression analysis in order to

99 determine [1] the clinical effects of placebo in lowering IOP, [2] the extent of IOP change as a placebo
100 effect relative to the effect observed in an untreated control, and [3] the possible factors impacting on
101 the effect of a placebo.

METHODS

The current study was conducted in compliance with the Declaration of Helsinki. Because no human subjects were included, this study was considered exempt by the institutional review board. Individual patient-level consent was not required. This study was conducted in accordance with the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA).[12] The study protocol was prospectively registered with an online open-access systematic-review-protocol database (PROSPERO; no. CRD42022348098).

Inclusion and Exclusion Criteria

RCTs meeting all of the following criteria were included in the present analyses: (1) open-angle glaucoma (OAG), ocular hypertension (OHT) had been included in the study population; (2) at least one type of IOP-lowering ocular hypotensive eye drop had been used as a treatment group; (3) placebo-controlled trials, or trials with an untreated control group as a comparator, had been conducted; (4) the treatment duration had been at least four weeks; (5) IOP had been reported as an outcome variable.

The following were the exclusion criteria: (1) studies had not been conducted with adult humans; (2) studies had entailed narrative or systematic reviews, commentaries or case reports; (3) studies had involved either secondary or angle-closure glaucoma; (4) studies had used glaucoma medications without approval from the United States Food and Drug Administration (either FDA or USFDA) or those that had not been intended for lowering of IOP, (5) studies had compared two active drugs but also used a placebo drug for matching of the number of instillation frequencies.

Literature Search

A systematic literature search was performed on June 2, 2022 using PubMed, EMBAS, Scopus, the Cochrane search engine, the World Health Organization International Clinical Trials Registry Platform, and Clinical-Trials.gov. Development of our search strategies was carried out with the assistance of an academic librarian who had expertise in systematic review, and were based on established terminology that included Medical Subject Headings as well as EMBASE search terms. The keywords were

combinations of *glaucoma, open-angle glaucoma, ocular hypertension, treatment, placebo, untreated control*, and *RCT*. We hand-searched reference lists of published articles to find additional relevant studies. Two reviewers (S.C. and W.C.) searched the literature independently and performed further cross-checking of the reference lists. The full search strategies are available in **Appendix 1** (available at www.aaojournal.org).

Study Selection

To identify relevant articles, the titles and abstracts of retrieved papers were exported to Endnote (version X9; Thomson Reuters), where duplicates were removed. Then, two investigators (S.C. and W.C.) assessed titles and abstracts independently for eligibility and retrieved the corresponding full-text articles in such cases. The same 2 investigators then independently assessed those articles for final eligibility. Any inconsistencies were resolved by discussion/consensus or were adjudicated by a third investigator (Y.K.K.).

Data Extraction

The following data were extracted from each study: name of first author; year of study commencement; publication year; geographic area; types of treatment drug; types of placebo drug; types of glaucoma; study sponsors (industry or public); study settings (multicenter or single center); study analysis protocol (intention-to-treat or per-protocol); study blinding; sample size (treatment, placebo and untreated control); age; sex (male, female); follow-up duration; IOP (mmHg) at baseline and targeted period (4 weeks after treatment); average IOP changes during the entire follow-up period.

We extracted means and standard deviations for continuous outcomes. If standard deviations had not been provided, we calculated them based on standard errors, confidence intervals (CIs) or other measures.[13-15] In papers representing results only graphically, we extracted the graphs' numerical values using Adobe Acrobat's XI measuring tool (Adobe Systems Incorporated).[16, 17] In cases where data were unavailable, we supplied it from other studies that had the same cohort or population source, or we calculated it manually where possible. Data were extracted independently and in a masked manner by two investigators (S.C and W.C.) and were entered into Microsoft Access 2016 (Microsoft Corporation, Redmond, WA, USA) in electronic format. Conflicting data entries were

identified by algorithm.

Primary Outcome

We used, as our primary outcome, mean IOP change from the baseline following the 4-week treatment. For all of the comparisons, a negative value was taken as the sign that the mean IOP had been lowered after the intervention. The perceived placebo effect was determined as the effect size (ES: mean difference [MD] between baseline and endpoint), which was compared with the ES that had been obtained from an untreated control group or active treatment group. We repeated the NMA for average change in IOP over the entire follow-up, taking into account the longer-term alterations in IOP as a sensitivity analysis.

Quality Assessment

We assessed study quality by the revised tool used to assess risk of bias (ROB) in randomized trials (RoB 2).[18] The following five bias domains were evaluated: (1) randomization processes, (2) deviations from intended interventions, (3) missing outcome data, (4) measurement of the outcome, and (5) selection of the reported result for parallel study design. For cross-over study designs, bias incurred from "period and carryover effects" was additionally assessed. Each domain was graded as either low ROB, some concerns, or high ROB.

Statistical Models for Meta-Analysis

Study-specific ESs were combined so as to estimate the pooled ES (with 95% CI) based on a random effects model. Inter-study heterogeneity was quantified using the I^2 statistic representative of the percentage of inter-study variation attributable to heterogeneity. To determine the source of between-study heterogeneity that might have made an ES estimate less precise, we performed subgroup analyses by random effects model for between-subgroup differences.[19] We looked at subgroups differing by any of 7 potential sources: (1) type of active treatment, (2) type of placebo, (3) type of glaucoma, (4) study setting (i.e., multicenter or single center), (5) type of sponsor, (6) geographic area, and (7) whether the study follows an add-on design, which involves evaluating the impact of an additional drug when administered alongside an existing medication.

In order to compare the pooled ES (with 95% CI) between the placebo group and an untreated control group, NMA was performed using the active treatment group as a link. The treatment groups were classified according to the active ingredient of each drug (i.e., beta-blockers, carbonic anhydrase inhibitors, prostaglandin analogues, and alpha-adrenergic agonists), and when a mixture of various ingredients was used, or when laser treatment was added, the relevant treatment groups were classified as "Others".

Cross-study heterogeneity of effect estimates as well as study heterogeneity effects on pooled ES of the NMA was assessed using, respectively, *Q* statistics and I^2 statistic.[20, 21] Inconsistency (i.e., nonagreement between direct/indirect intervention effects)[10] was evaluated using Separating Indirect from Direct Evidence (SIDE) and the back-calculation method (i.e., node-splitting).[22] For assessment of the confidence of NMA estimates, we used a semiautomated web application (Confidence in Network Meta-analysis; Institute of Social and Preventive Medicine).[23, 24]

Meta-regression determines if any linear association exists between variables and a comparative treatment effect, as well as the direction of that association.[25] The statistical models for meta-regression that were employed are provided in **Appendix 2** (available at www.aaojournal.org), and the definitions for the covariates included in the meta-regression are provided in **Table S1** (available at www.aaojournal.org). All of the 95% CIs and *P*-values were two-sided, and $P < 0.05$ was considered to represent statistical significance. All statistical analyses were performed with R 4.0.4 software (The R Foundation for Statistical Computing).

Publication bias was evaluated graphically, by funnel plot,[26] and quantitative assessment of publication bias was conducted by the Begg and Mazumbar adjusted rank correlation test, which evaluates funnel plot asymmetry by examining the correlation between the ESs and their variances.[27]

RESULTS

Study Selection and Appraisal

A total of 3,788 studies were retrieved from the systematic research. After removal of duplications and reading of the abstracts, 226 studies remained. After a thorough full-text review, 40 trials (7,829 eyes in total) meeting the inclusion and exclusion criteria were selected (**Figure 1;** references to these RCTs are presented in Appendix 3, available at www.aaojournal.org). These included 37 trials of parallel design and 3 trials of cross-over design. In total, 33 placebo groups (2,055 eyes) and 7 untreated controls (1,184 eyes) were available for our final analyses.

Among the 33 placebo-controlled trials, 16 (48%) used vehicle as the placebo, while others used non-vehicle or eye drops of unknown type. Twenty-four (24) trials were of multicenter design. Eleven (11) trials used beta-blockers, 9 used carbonic anhydrase inhibitors, 7 used prostaglandin analogues, and 5 used alpha adrenergic agonists, as the active treatment. Fourteen (14) trials had enrolled both OAG and OHT patients, while 14 and 11 had enrolled OAG and OHT, respectively. Twenty-two (22) trials had been conducted in North America, 9 in Europe, 3 in Asia, and 3 on more than one continent. Five (5) trials compared the additional IOP-lowering effect of a treatment drug with placebo drug as an adjunctive therapy. The characteristics of the included trials are shown in **Table 2**, and the data for the IOP changes between the baseline and the endpoint are provided in **Table 3**.

Most of the trials that were included in this analysis were evaluated as moderate ROB (**Appendix 4**, available at www.aaojournal.org). Two trials that were assessed as high ROB had an issue with deviations from the intended interventions.[28, 29] Detailed measures taken to minimize the regression to the mean effect in each study are presented in **Table S4** (available at www.aaojournal.org). We found no conclusive evidence of any systemic difference between the placebo- and untreated-controlled trials.

Perceived Efficacy of Placebo

Among the 33 placebo-controlled trials, the ES for IOP lowering was –1.30 mmHg (95% CI, –1.75 mmHg to –0.84 mmHg; **Figure 2**). Meanwhile, the ES for IOP lowering was -0.16 mmHg (95% CI, –0.31 mmHg to –0.01 mmHg; **Figure S3** available at www.aaojournal.org) among the 7 RCTs with untreated control.

The size of the box representing the point estimate for each study in the forest plot is in proportion to the contribution of that study's weight estimate to the summary estimate. The colors of the boxes indicate the following: blue (significant decrease in IOP), green (non-significant), and orange (significant increase in IOP). The horizontal lines indicate the 95% CIs. The diamond denotes the pooled effect, and the lateral tips of the diamond indicate the associated CIs. The drapery plot in **Figure S4** (available at www.aaojournal.org) shows the different meta-analytic results by *P* value functions (*P* value = 0.1, 0.05 and 0.01).

In subgroup analyses, the geographic area of the trials had a significant effect on between-study heterogeneity (*Q* = 0.74, *P* = 0.040). The ES in Asian trials was the greatest (-2.09; 95% CI, -2.81 to -1.37), followed by European (-1.38; 95% CI, -2.01 to -0.75) and North American (-1.09; 95% CI, -1.83 to -0.35) trials. The remaining 6 potential sources of heterogeneity (i.e., type of active treatment, type of placebo, type of glaucoma, study setting, type of sponsor, and study design) did not show any significant effect on between-study heterogeneity (**Table S5**, available at www.aaojournal.org).

Placebo Effect over Untreated Control

The NMA compared the pooled ES of the placebo group to that of the untreated control group. **Figure 5** shows a network plot of eligible studies (7 arms of 40 trials, with 40 pairwise comparisons). The ES in the placebo group was significantly greater than the ES observed in the untreated controls by –2.27 mmHg (95% CI, –3.52 to –1.01; **Figure 6**). A net league table showing head-to-head comparisons is provided in the form of **Table S6** (available at www.aaojournal.org).

In the NMA model, the between-design inconsistency was not significant (*P* = 0.404), and the Q value was low (*Q* = 0.70) when assuming a full design-by-treatment random-effects model (**Table S7**, available at www.aaojournal.org). There was no significant inconsistency between the direct and indirect evidence (all *P* values > 0.05; **Table S8** [available at www.aaojournal.org]).

Determinants of Placebo Effect

To determine the predictors of the placebo effect, multiple meta-regression analyses were performed, applying the 10 covariates described in **Table S1** (available at www.aaojournal.org). The best-fitted model was the combination of active treatment effect, study design, participant number, and active

drug type (Akaike's information criterion corrected [AICc] = 98.7). The model-averaged predictor importance plot displays the averaged importance of each predictor across all of the models (**Figure S7**, available at www.aaojournal.org). Active treatment (model importance = 1.00) was the most important predictor, followed by study design (model importance = 0.95) and number of participants (model importance = 0.81).

The meta-regression model showed that the placebo effect increased significantly with increased ES from active treatment (P = 0.01). Specifically, placebo additionally lowered IOP –0.45 mmHg per –1 mmHg of active treatment effect. The bubble plot in **Figure S8** (available at www.aaojournal.org) shows a positive association between the placebo group ESs and the active treatment effect (estimated amount of residual heterogeneity $\tau^2_{\text{unexplained}}$ = 1.26, R^2 = 20.6%).

Sensitivity Analyses

We noted that the conclusions on the primary outcome were not altered substantially after accounting for the longer-term IOP changes. The data for the average IOP change over the entire follow-up period are provided in **Table S9** (available at www.aaojournal.org). The ES in the placebo group was significantly greater than that observed in the untreated controls, by –1.99 mmHg (95% CI, –3.03 to –0.94; **Figure S9**). A net league table showing head-to-head comparisons is provided in **Table S10** (available at www.aaojournal.org). The results for assessment of network heterogeneity and consistency are depicted in **Tables S11-S12** (available at www.aaojournal.org).

Publication Bias

Figure 10 is a funnel plot depicting publication bias. The studies included in the analysis show symmetric patterns that may not be indicative of publication bias. Likewise, there was no evidence of publication bias by Begg and Mazumbar adjusted rank correlation test (T = 1.20, P = 0.24).

DISCUSSION

Although there is a large literature on placebo effects, the present study is the first to systematically analyze published data on the effect of placebo in ocular hypotensive therapy. For lowering of IOP, there are a variety of pharmacological treatment options (beta-blockers, carbonic anhydrase inhibitors, prostaglandin analogues, alpha-adrenergic agonists), most of which have been tested formally via randomized placebo-controlled trial. Thus, this meta-analysis was an excellent opportunity to explore the placebo effect with its determinants and to compare its magnitude relative to active or no-treatment in ocular hypotensive therapy. There are three key findings: (1) placebo is effective for lowering of IOP; (2) the effect is superior to non-treatment, and (3) the major determinants of its IOP-lowering effect are the strength of active treatment, whether the study followed an add-on design, and sample size.

Demonstrating placebo efficacy is logically parallel to demonstrating drug efficacy. Just as a patient's improvement after taking a drug does not prove that drug's efficacy, the same is true for placebo administration.[30] In order to determine the true placebo effect, therefore, other non-specific effects such as natural disease remission, regression to the mean, and other, time-dependent variables should be identified, and this can be accomplished by inclusion of an untreated control group in clinical trials.[9] In the present study, the important comparison between placebo and non-treatment in NMA showed placebo to have been superior to the untreated control (by –2.27 mmHg). This finding supports the hypothesis that placebo can have a true clinical effect in ocular hypotensive therapy.

Subjective assessments generally are considered to be more prone than objective ones to the placebo effect, and this is reflected in the exceptionally high positive placebo response rates seen in pain and psychiatric disorder studies.[31, 32] Since molecular events and neural network changes that underly placebo effects are mediated by expectancies or anticipated future outcomes, psychological variables are more likely to be affected by placebo effects. Expectancies and placebo effects, however, are not limited to subjective outcomes. In Parkinson's disease for example, placebo-induced release of endogenous dopamine along with associated neuronal changes have been well documented.[33, 34] A

blood pressure experiment of crossover design noted significant effects after placebo treatment of 1-month duration.[35] In this study, clinic and ambulatory blood pressures during the no-treatment period did not differ from the baseline, indicating that the objective outcome changes after placebo administration had been due to an actual placebo effect.

Placebo effects are associated with the release of dopamine,[34, 36] endogenous opioids,[37, 38] endocannabinoids,[39] oxytocin,[40] and vasopressin.[41] Dopamine is the predominant neurotransmitter for the retina,[42] and the role of dopaminergic system in IOP regulation has been studied. Dopamine has been shown to result in dose-dependent decrease of IOP in both rabbits and mice.[43, 44] The mechanism of action entails dose-dependent and parallel dopamine-induced changes in ciliary blood flow and aqueous production.[45] Opioids also have an IOP-lowering property.[46] The mechanism by which opioids reduce IOP involves the central diencephalic control centers via relaxation of the ocular muscles and the facilitation or inhibition of aqueous humor drainage and production.[47] It has been suggested that the nitric-oxide-releasing activity of the μ3 opioid receptor subtype may mediate this process.[48] Certainly, the underlying molecular mechanisms associated with the placebo effects of ocular hypotensive therapy should be further investigated.

In our results, ES of placebo increased in line with the ES of active treatment and also in add-on design studies. Participants in a trial of a more effective drug may have greater expectations about the effect, which may lead to a greater placebo effect. In add-on design RCTs measuring additive effect, participants are aware that they will receive at least one active treatment, which fact reduces uncertainty about drug efficacy, thereby leading potentially to a greater placebo effect. Additionally, the number of study participants was positively correlated with the placebo effect, as is consistent with a former study investigating the determinants of placebo effect.[7] This could have been due to the need for larger sample sizes to demonstrate statistical differences between placebo and active treatment groups in cases of a larger placebo effect.

The current study has several limitations that should be considered when interpreting its results. First, the degree of blinding remains a challenge in investigations of true placebo effect. In RCTs,

patients typically are informed that they will receive either the real treatment or a placebo. This leads to uncertainty among patients regarding what they actually received. The effects of placebo are significantly affected by patients' beliefs and expectations respecting treatment.[49] Thus, placebo effects in double-blind placebo-controlled trials are likely to be reduced relative to the effects of placebo treatment in clinical practice. Second, a relatively small number of studies included an untreated control arm, and none were specifically designed to address the comparison with placebo. Indirect comparisons are more at risk of bias than are direct ones. Although we conducted thorough examinations to determine whether there were any systematic differences between the placebo and untreated groups in attempts to mitigate the effects of regression to the mean, the results should be interpreted with caution. Third, the effects of placebo on glaucoma progression could not be investigated since the data were insufficiently granular for meaningful analyses. Given that the ultimate goal of IOP lowering is halting of glaucoma deterioration, there remains a specific need for information on how placebo treatment might affect structural and functional glaucoma-related parameters. Fourth, like many meta-analyses, we used study-level variables in our regression analysis, and so the analytical sensitivity may be lower than optimal. There might be many other, more important factors affecting the placebo ES that were not measured or considered in the current study.

In conclusion, this meta-analysis suggests that placebo is clinically effective for lowering of IOP in eyes with glaucoma or OHT. The amount of placebo effect was influenced by the effect of the strength of active treatment, whether the study followed an add-on design, and the sample size. This fact should be considered when evaluating the efficacy of proposed IOP-lowering medication as well as in health-economics analyses and sample-size calculations.

REFERENCES

1. Kaptchuk TJ, Miller FG. Placebo Effects in Medicine. *N Engl J Med*. 2015;373:8-9.
2. Colloca L, Barsky AJ. Placebo and Nocebo Effects. *N Engl J Med*. 2020;382:554-561.
3. Kirchhof J, Petrakova L, Brinkhoff A, et al. Learned immunosuppressive placebo responses in renal transplant patients. *Proc Natl Acad Sci U S A*. 2018;115:4223-4227.
4. Kemeny ME, Rosenwasser LJ, Panettieri RA, Rose RM, Berg-Smith SM, Kline JN. Placebo response in asthma: a robust and objective phenomenon. *J Allergy Clin Immunol*. 2007;119:1375-81.
5. Weinreb RN, Aung T, Medeiros FA. The pathophysiology and treatment of glaucoma: a review. *JAMA*. 2014;311:1901-11.
6. Castro M. Placebo versus best-available-therapy control group in clinical trials for pharmacologic therapies: which is better? *Proc Am Thorac Soc*. 2007;4:570-3.
7. Hróbjartsson A, Gøtzsche PC. Is the placebo powerless? An analysis of clinical trials comparing placebo with no treatment. *N Engl J Med*. 2001;344:1594-1602.
8. Hrobjartsson A. What are the main methodological problems in the estimation of placebo effects? *J Clin Epidemiol*. 2002;55:430-5.
9. Ernst E, Resch KL. Concept of true and perceived placebo effects. *BMJ*. 1995;311:551-3.
10. Cipriani A, Higgins JP, Geddes JR, Salanti G. Conceptual and technical challenges in network meta-analysis. *Ann Intern Med*. 2013;159:130-7.
11. Ha A, Jang M, Shim SR, Kim CY, Chang IB, Kim YK. Interventions for Glaucoma Medication Adherence Improvement: A Network Meta-analysis of Randomized Controlled Trials. *Ophthalmology*. 2022;129:1294-1304.
12. Page MJ, McKenzie JE, Bossuyt PM, et al. The PRISMA 2020 statement: an updated guideline for reporting systematic reviews. *Int J Surg.* 2021;88:105906.
13. Lipsey MW, Wilson DB. *Practical meta-analysis*. SAGE publications, Inc; 2001.
14. Higgins JP, Deeks JJ. Selecting studies and collecting data. *Cochrane handbook for systematic reviews of interventions: Cochrane book series*. 2008:151-185.
15. Higgins J, Deeks JJ, Altman DG. Special topics in statistics. Cochrane handbook for systematic reviews of interventions: Cochrane book series. 2008;
16. Shim SR, Lim YH, Hong M, Song G, Han HW. Statistical Data Extraction and Validation from Graph for Data Integration and Meta-analysis. *The Korea Journal of BigData*. 2021;6:61-70.
17. Durg S, Dhadde SB, Vandal R, Shivakumar BS, Charan CS. Withania somnifera (Ashwagandha) in neurobehavioural disorders induced by brain oxidative stress in rodents: a systematic review and

meta-analysis. *J Pharm Pharmacol*. 2015;67:879-99.

18. Sterne JAC, Savovic J, Page MJ, et al. RoB 2: a revised tool for assessing risk of bias in randomised trials. *BMJ*. 2019;366:l4898.

19. Borenstein M, Higgins JP. Meta-analysis and subgroups. *Prev Sci*. 2013;14:134-43.

20. Higgins JP, Thompson SG, Deeks JJ, Altman DG. Measuring inconsistency in meta-analyses. *BMJ*. 2003;327:557-560.

21. Cochran WG. The comparison of percentages in matched samples. *Biometrika*. 1950;37:256-266.

22. Dias S, Welton NJ, Caldwell D, Ades AE. Checking consistency in mixed treatment comparison meta-analysis. *Stat Med.* 2010;29:932-944.

23. Nikolakopoulou A, Higgins JP, Papakonstantinou T, et al. CINeMA: an approach for assessing confidence in the results of a network meta-analysis. *PLoS Med*. 2020;17:e1003082.

24. Papakonstantinou T, Nikolakopoulou A, Higgins JP, Egger M, Salanti G. Cinema: software for semiautomated assessment of the confidence in the results of network meta-analysis. *Campbell Syst Rev.* 2020;16:e1080.

25. Baker WL, White CM, Cappelleri JC, et al. Understanding heterogeneity in meta-analysis: the role of meta-regression. *Int J Clin Pract*. 2009;63:1426-34.

26. Egger M, Davey Smith G, Schneider M, Minder C. Bias in meta-analysis detected by a simple, graphical test. *BMJ*. 1997;315:629-34.

27. Begg CB, Mazumdar M. Operating characteristics of a rank correlation test for publication bias. *Biometrics*. 1994;50:1088-1101.

28. Epstein DL, Krug JH, Jr., Hertzmark E, Remis LL, Edelstein DJ. A long-term clinical trial of timolol therapy versus no treatment in the management of glaucoma suspects. *Ophthalmology*. 1989;96:1460-7.

29. Schulzer M, Drance SM, Douglas GR. A comparison of treated and untreated glaucoma suspects. *Ophthalmology*. 1991;98:301-7.

30. Miller FG, Rosenstein DL. The nature and power of the placebo effect. *J Clin Epidemiol*. 2006;59:331-5.

31. Kirsch I. Antidepressants and the placebo effect. *Z Psychol*. 2014;222:128-134

32. Hróbjartsson A, Copenhagen P. placebo is better than no treatment for subjective continuous outcomes and for treatment of pain. *N Engl J Med*. 2001;344:1594-602.

33. de la Fuente-Fernandez R, Ruth TJ, Sossi V, Schulzer M, Calne DB, Stoessl AJ. Expectation and dopamine release: mechanism of the placebo effect in Parkinson's disease. *SCIENCE.*

2001;293:1164-6.

34. Benedetti F, Colloca L, Torre E, et al. Placebo-responsive Parkinson patients show decreased activity in single neurons of subthalamic nucleus. *Nat Neurosci*. 2004;7:587-8.

35. Asmar R, Safar M, Queneau P. Evaluation of the placebo effect and reproducibility of blood pressure measurement in hypertension. *Am J Hypertens*. 2001;14:546-52.

36. Lidstone SC, Schulzer M, Dinelle K, et al. Effects of expectation on placebo-induced dopamine release in Parkinson disease. *Arch Gen Psychiatry*. 2010;67:857-65.

37. Amanzio M, Benedetti F. Neuropharmacological dissection of placebo analgesia: expectation-activated opioid systems versus conditioning-activated specific subsystems. *J Neurosci*. 1999;19:484-494.

38. Eippert F, Bingel U, Schoell ED, et al. Activation of the opioidergic descending pain control system underlies placebo analgesia. *NEURON.* 2009;63:533-543.

39. Benedetti F, Amanzio M, Rosato R, Blanchard C. Nonopioid placebo analgesia is mediated by CB1 cannabinoid receptors. *Nat Med*. 2011;17:1228-30.

40. Kessner S, Sprenger C, Wrobel N, Wiech K, Bingel U. Effect of oxytocin on placebo analgesia: a randomized study. *JAMA*. 2013;310:1733-5.

41. Colloca L, Pine DS, Ernst M, Miller FG, Grillon C. Vasopressin Boosts Placebo Analgesic Effects in Women: A Randomized Trial. *Biol Psychiatry*. 2016;79:794-802.

42. Djamgoz MB, Wagner HJ. Localization and function of dopamine in the adult vertebrate retina. *Neurochem Int*. 1992;20:139-91.

43. Shannon RP, Mead A, Sears ML. The effect of dopamine on the intraocular pressure and pupil of the rabbit eye. *Invest Ophthalmol*. 1976;15:371-80.

44. Bucolo C, Leggio GM, Maltese A, Castorina A, D'Agata V, Drago F. Dopamine-(3) receptor modulates intraocular pressure: implications for glaucoma. *Biochem Pharmacol*. 2012;83:680-6.

45. Reitsamer HA, Kiel JW. Effects of dopamine on ciliary blood flow, aqueous production, and intraocular pressure in rabbits. *Invest Ophthalmol Vis Sci*. 2002;43:2697-2703.

46. Drago F, Panissidi G, Bellomio F, Belle AD, Aguglia E, Gorgone G. Effects of opiates and opioids on intraocular pressure of rabbits and humans. *Clin Exp Pharmacol Physiol*. 1985;12:107-113.

47. Cunningham AJ, Barry P. Intraocular pressure--physiology and implications for anaesthetic management. *Can Anaesth Soc J*. 1986;33:195-208.

48. Dortch-Carnes J, Russell KR. Morphine-induced reduction of intraocular pressure and pupil diameter: role of nitric oxide. *Pharmacology*. 2006;77:17-24.

460 49. Pollo A, Amanzio M, Arslanian A, Casadio C, Maggi G, Benedetti F. Response expectancies in
461 placebo analgesia and their clinical relevance. *Pain*. 2001;93:77-84.

FIGURE LEGENDS

Figure 1. Flow Diagram Showing Selection Process for Inclusion of Studies in Analysis

Figure 2. Forest Plot of Intraocular Pressure-Lowering Effect of Placebo

The size of the box representing the point estimate for each study in the forest plot is in proportion to the contribution of that study's weight estimate to the summary estimate. The colors of the boxes indicate the following: blue (significant decrease in IOP), green (non-significant), and orange (significant increase in IOP). The horizontal lines indicate the 95% CIs. The diamond denotes the pooled effect, and the lateral tips of the diamond indicate the associated CIs. MD = mean difference; CI = confidence interval

Figure 5. Network Map for Network Meta-Analysis

The node size corresponds to the number of participants assigned to each treatment. Treatments with direct comparisons are linked with a line. The line thickness corresponds to the number of trials evaluating the comparison.

Figure 6. Forest Plot for Network Meta-Analysis

The size of the box representing the point estimate for each study in the forest plot is in proportion to the contribution of that study's weight estimate to the summary estimate. Blue boxes indicate the IOP-lowering effect with statistical significance, and orange boxes indicate elevated IOP with statistical significance. The horizontal lines indicate the 95% CIs. MD = mean difference; CI = confidence interval

Figure 10. Funnel Plots

Funnel plots for (A) placebo-controlled and (B) untreated-controlled trials. The funnel plots show the effect size of each study (expressed as the mean difference) on the x-axis, and the standard error (from large to small) on the y-axis. To facilitate interpretation, the plots include the idealized funnel-shape one would expect the studies to follow. The vertical line in the middle of the funnel shows the average effect size.

Table 2. Characteristics of Studies Included in Meta-analysis

Ref ID	Study	Sample Size (n)	Type of Glaucoma	Age (yrs)	Sex (% Female)	Follow-up (mon)	Active Drug	Placebo	Type of RCT	Study Setting	Study Sponsor	Geographic Area	Add-on Study Design	Study Blinding
Placebo Group														
1	Feghali et al., 1985	19	OAG, OHT	55.5 (14.3)	36.8	1.5	Betaxolol	NR	Parallel-designed RCT	NR	Pharma company	North America	No	Yes
2	Kass et al., 1989	124	OHT	58.2 (8.4)	61.3	60	Timolol	Diluent	Parallel-designed RCT	Multi center	Pharma company	North America	No	Yes
3	Alm et al., 1993	60	OAG	NR	NR	1	Latanoprost	NR	Parallel-designed RCT	Multi center	Pharma company	NR	No	Yes
4	Wilkerson et al., 1993	48	OAG, OHT	62.6 (NR)	52.1	1	Dorzolamide	Vehicle	Parallel-designed RCT	Multi center	NR	North America	No	Yes
5	Robin et al., 1995	174	OAG	64.5 (NR)	55.2	3	Apraclonidine	Vehicle	Parallel-designed RCT	Multi center	Pharma company	North America	Yes	Yes
6	Schwartz et al., 1995	37	OHT	60.1 (3.3)	48.6	18	Timolol	NR	Parallel-designed RCT	Single center	Pharma company	North America	No	Yes
7	Strahlman et al., 1996a	333	OAG, OHT	60.5 (11.7)	52.3	1.5	Dorzolamide	Vehicle	Parallel-designed RCT	Multi center	Pharma company	Mixed	No	Yes
8	Strahlman et al., 1996b	246	OAG, OHT	60.2 (10.7)	48.4	6	Dorzolamide	Vehicle	Parallel-designed RCT	Multi center	Pharma company	North America	Yes	Yes
9	Derick et al., 1997	186	OAG, OHT	58.9 (12.7)	49.5	1	Brimonidine	Vehicle	Parallel-designed RCT	Multi center	Pharma company	North America	No	Yes
10	FDA trial (c-97-40), 1999	256	OAG, OHT	65.9 (10.7)	55.9	1	Betaxolol Timolol Levobetaxolol	Vehicle	Parallel-designed RCT	Multi center	NR	NR	No	Yes
11	Harris et al., 1999	29	OAG, OHT	NR	NR	1	Dorzolamide	NR	Parallel-designed RCT	NR	Public grant	North America	No	Yes
12	Netland et al., 1999	24	OHT	60 (13)	83.3	1	Timolol	Vehicle	Cross over-designed RCT	NR	Pharma company	North America	No	Yes
13	Toris et al., 1999	56	OHT	53.6 (12.4)	75	1	Brimonidine	Vehicle	Parallel-designed RCT	Single center	NR	North America	No	Yes
14	Carlsson et al., 2000	31	OHT	56.1 (13.7)	NR	2	Brimonidine	Saline	Parallel-designed RCT	Single center	Pharma company	North America	No	Yes
15	Sall et al., 2000	409	OAG	62.9 (12.8)	54.5	3	Brinzolamide Dorzolamide	Vehicle	Parallel-designed RCT	Multi center	Pharma company	North America	No	Yes
16	Shin et al., 2000	108	OAG	61.8 (12.6)	53.7	3	Brinzolamide	NR	Parallel-designed RCT	Multi center	Pharma company	North America	Yes	Yes
17	Heijl and Bengtsson 2000/2001	90	OHT	62.8 (NR)	59.1	120	Timolol	NR	Parallel-designed RCT	Single center	Pharma company	Europe	No	Yes
18	Stewart et al., 2001	41	OAG	61.1 (11.4)	68.3	2	Unoprostone	Hypotears	Parallel-designed RCT	Single center	Pharma company	North America	Yes	Yes
19	Bergstrand et al., 2002	87	OAG	73 (NR)	NR	1.5	Dorzolamide	NR	Parallel-designed RCT	Multi center	Pharma company	Europe	No	Yes
20	Gandolfi et al., 2003	32	NTG	61 (10)	37.5	1	Brimonidine	NR	Cross over-designed RCT	Single center	NR	Europe	No	No
21	Kamal et al., 2003	356	OHT	65.7 (9.6)	20.5	60	Betaxolol	NR	Parallel-designed RCT	Single center	Pharma company	Europe	No	Yes

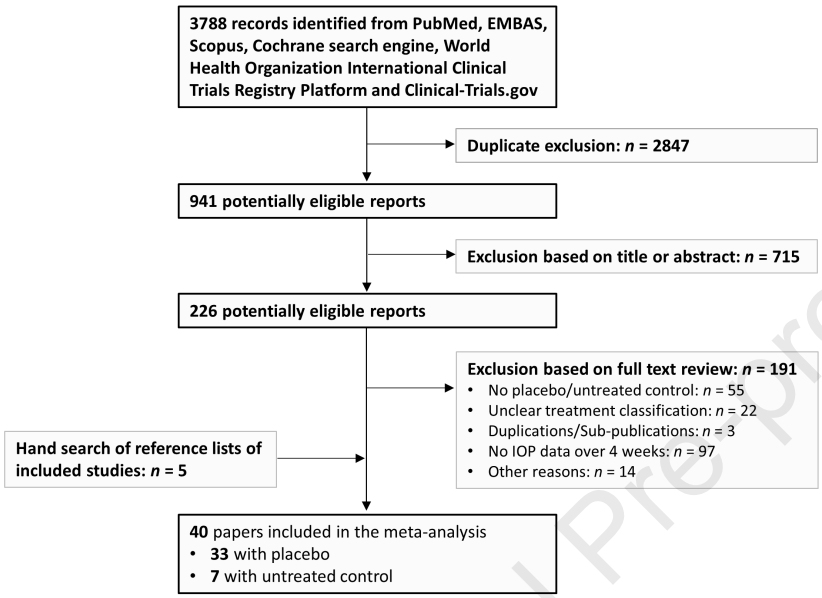
22	Toris et al., 2004	58	OAG, OHT	57.7 (2.0)	69	1	Unoprostone	Vehicle	Parallel-designed RCT	NR	Pharma company	North America	No	Yes
23	Arcieri et al., 2005	80	OAG, OHT	66.1 (13.7)	46.3	6	Unoprostone Bimatoprost Latanoprost Travoprost	Lubricant drop	Parallel-designed RCT	Multi center	Pharma company	Others	No	No
24	Miglior et al., 2005	1077	OHT	57.0 (10.3)	54.4	60	Dorzolamide	Vehicle	Parallel-designed RCT	Multi center	Pharma company	Europe	No	Yes
25	Harris et al., 2009	12	Healthy	27 (6)	50	1	Latanoprost	Artificial tears	Cross over-designed RCT	Single center	Public grant	North America	No	Yes
26	Craven et al., 2010	218	OAG, OHT	64.9 (11.1)	60.1	1	Bimatoprost	Vehicle	Parallel-designed RCT	Multi center	Pharma company	North America	No	Yes
27	Goldberg et al., 2012	153	OAG	65.7 (NR)	62.1	3	Brinzolamide	Vehicle	Parallel-designed RCT	Multi center	Pharma company	Mixed	Yes	Yes
28	Tanihara et al., 2013	210	OAG, OHT	59.5 (15.0)	58.6	2	Ripasudil	NR	Parallel-designed RCT	Multi center	Pharma company	Asia	No	Yes
29	Garway-Heath et al., 2015	516	OAG	65.5 (10.5)	47.1	24	Latanoprost	Vehicle	Parallel-designed RCT	Multi center	Pharma company	Europe	No	Yes
30	NCT03310580, 2018	40	NTG	62.4 (15.9)	72.5	1	Netarsudil	Vehicle	Parallel-designed RCT	NR	Pharma company	North America	No	Yes
31	Aihara et al., 2019a	91	OAG, OHT	65.3 (11.8)	58.2	1	Omidenepag isopropyl	NR	Parallel-designed RCT	Multi center	Pharma company	North America	No	No
32	Aihara et al., 2019b	63	OAG, OHT	66.2 (11.0)	58.7	1	Latanoprost Omidenepag isopropyl	NR	Parallel-designed RCT	Multi center	Pharma company	Asia	No	Yes
33	Araie et al., 2021	215	OAG, OHT	63.6 (13.3)	57.2	1	Netarsudil	Vehicle	Parallel-designed RCT	Multi center	Pharma company	Asia	No	Yes
Untreated Group														
34	Holmin et al., 1988	20	OAG	68.1 (NR)	66.7	36	Timolol Pilocarpine Acetazolamide	no placebo	Parallel-designed RCT	Multi center	NR	Europe	No	No
35	Epstein et al., 1989	107	OAG	59.5 (11.5)	56.1	NR	Timolol	no placebo	Parallel-designed RCT	Single center	Pharma company	North America	No	No
36	Schulzer et al., 1991	143	OHT	60.3 (10.5)	53.1	72	Timolol	no placebo	Parallel-designed RCT	NR	Pharma company	North America	No	No
37	Ravalico et al., 1994	49	OHT	61.4 (10.9)	46.2	24	Levobunolol	no placebo	Parallel-designed RCT	NR	NR	Europe	No	NR
38	CNTGS Group, 1998	140	NTG	65.8 (10.0)	68.6	NR	NR	no placebo	Parallel-designed RCT	Multi center	Public grant	Mixed	No	No
39	Heijl et al., 2002	255	OAG	68.1 (4.9)	66	minimum of 48	Betaxolol	no placebo	Parallel-designed RCT	Multi center	Public grant	Europe	No	No
40	Kass et al., 2002	1636	OHT	56.1 (9.2)	56.9	72	NR	no placebo	Parallel-designed RCT	Multi center	Mixed	North America	No	No

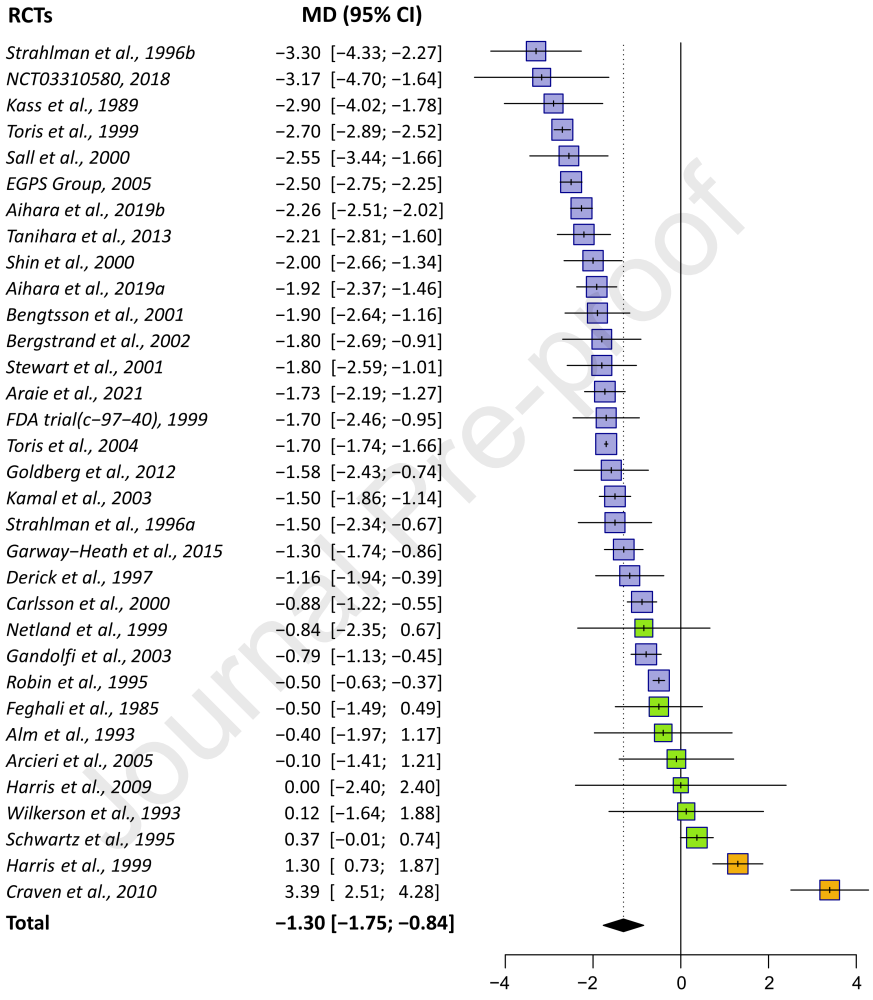
NR = not reported; NTG = normal-tension glaucoma; OAG = open-angle glaucoma; OHT = ocular hypertension; RCT = Randomized Controlled Trial

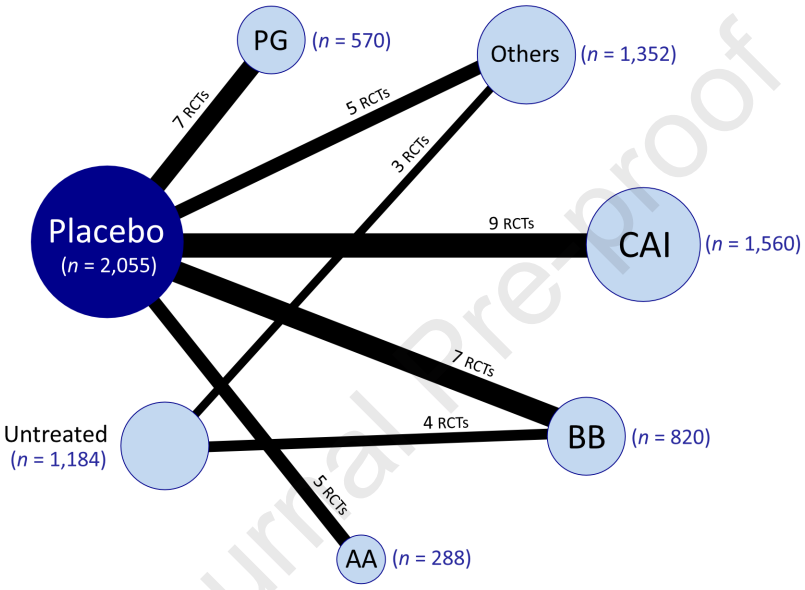
Table 3. Intraocular Pressures of Treatment and Control Groups

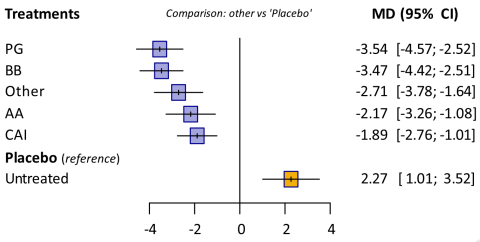
Ref ID	Study, Year	Treatment Group		Placebo or Untreated Group	
		Mean Change of IOP (mmHg)	SD of Change of IOP (mmHg)	Mean Change of IOP (mmHg)	SD of Change of IOP (mmHg)
Placebo Group					
1	Feghali et al., 1985	-4.2	6.5	-0.5	1.6
2	Kass et al., 1989	-5.4	4.4	-2.9	4.5
3	Alm et al., 1993	-5.7	3.6	-0.4	3.1
4	Wilkerson et al., 1993	-3.8	3.9	0.1	4.0
5	Robin et al., 1995	-1.3	0.6	-0.5	0.6
6	Schwartz et al., 1995	-4.4	0.8	0.4	0.9
7	Strahlman et al., 1996a	-3.8	4.3	-1.5	3.9
8	Strahlman et al., 1996b	-2.4	4.0	-3.3	3.5
9	Derick et al., 1997	-2.9	2.2	-1.2	2.7
10	FDA trial(c-97-40), 1999	-4.5	2.6	-1.7	2.5
11	Harris et al., 1999	-2.2	0.7	1.3	1.0
12	Netland et al., 1999	-4.2	2.7	-0.8	2.7
13	Toris et al., 1999	-5.0	0.7	-2.7	0.5
14	Carlsson et al., 2000	-4.7	0.8	-0.9	0.6
15	Sall et al., 2000	-4.2	3.2	-2.6	3.4
16	Shin et al., 2000	-3.8	2.6	-2.0	2.5
17	Heijl and Bengtsson, 2000/2001	-6.9	2.6	-1.9	2.5
18	Stewart et al., 2001	-2.9	1.9	-1.8	2.2
19	Bergstrand et al., 2002	-4.8	5.3	-1.8	3.0
20	Gandolfi et al., 2003	-3.1	1.9	-0.8	0.7
21	Kamal et al., 2003	-4.6	2.3	-1.5	2.4
22	Toris et al., 2004	-4.8	0.6	-1.7	0.1
23	Arcieri et al., 2005	-5.2	2.2	-0.1	2.7
24	Miglior et al., 2005	-3.7	2.5	-2.5	3.0
25	Harris et al., 2009	-4.0	2.6	0.0	3.0
26	Craven et al., 2010	-1.0	2.7	3.4	3.9
27	Goldberg et al., 2012	-3.0	2.8	-1.6	3.8
28	Tanihara et al., 2013	-3.5	2.3	-2.2	2.3
29	Garway-Heath et al., 2015	-4.0	3.4	-1.3	3.6
30	NCT03310580, 2018	-5.2	2.7	-3.2	2.7
31	Aihara et al., 2019a	-5.8	3.2	-1.9	4.1
32	Aihara et al., 2019b	-5.0	3.2	-2.3	2.4
33	Araie et al., 2021	-4.6	2.1	-1.7	1.8
Untreated Group					
34	Holmin et al., 1988	-4.0	2.3	1.2	3.7
35	Epstein et al., 1989	-4.0	1.2	-0.6	1.4
36	Schulzer et al., 1991	-6.8	3.5	-1.1	3.2
37	Ravalico et al., 1994	-8.7	1.8	-0.6	2.3
38	CNTGS Group, 1998	-6.3	2.5	-0.1	2.2
39	Heijl et al., 2002	-5.1	3.4	-0.1	1.9
40	Kass et al., 2002	-4.8	2.4	-0.1	2.8

IOP = intraocular pressure; SD = standard deviation

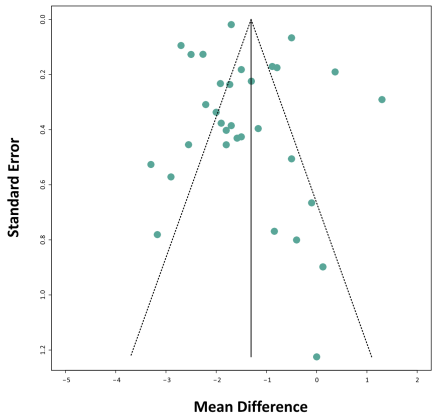




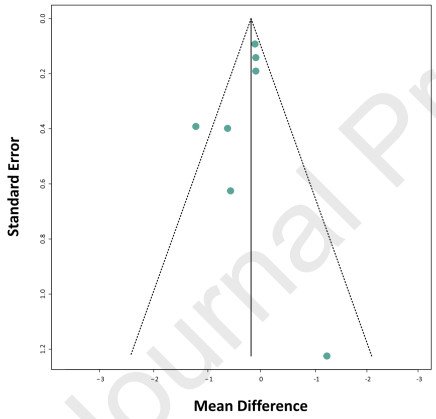




A



B



PRÉCIS

In this randomized controlled trial meta-analysis, the placebo effect in ocular hypotensive therapy was quantified systematically. The placebo was effective in lowering IOP, and superior to the effect observed for the untreated controls.