STUDY REPORT

Prospective, randomized, open label phase II study to assess efficacy and safety of Macugen® (pegaptanib 0.3 mg intravitreal injections) plus panretinal photocoagulation (PRP) and PRP (monotherapy) in the treatment of patients with high risk proliferative diabetic retinopathy.

PROTOCOL Nº:	CC-02-2009
EudraCT Number :	2012-001200-38
SPONSOR:	AIBILI, José Cunha-Vaz
REPORT VERSION Nº:	0
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Investigator(s) : José Cunha-Vaz, MD, PhD

According ICH-E3

Study Report

Study title:	Prospective, randomized, open label phase II study to assess efficacy and safety of Macugen® (pegaptanib 0.3 mg intravitreal injections) plus panretinal photocoagulation (PRP) and PRP (monotherapy) in the treatment of patients with high risk proliferative diabetic retinopathy.
Test drug/investigational product:	MACUGEN® (pegaptanib 0.3 mg intravitreal injections)
Indication studied:	Proliferative Diabetic Retinopathy
Study design:	Prospective, randomized, open label phase II study
Sponsor:	AIBILI – Association for Innovation and Biomedical Research on Light and Image
Protocol identification:	Protocol No. CC-02-2009 (EudraCT 2009-016760-36)
Development phase of study:	Phase II
Study initiation date:	24/03/2010 (Initiation Visit); 21/04/2010 (First Patient First Visit)
Early termination date:	31/10/2012 (Last Patient Last Visit)
Study completion date:	31/10/2012 (Last Patient Last Visit) ; 15/04/2013 (Close-Out Visit)
Principal Investigator:	José Cunha-Vaz, MD, PhD AIBILI – CEC (Center for Clinical Trials), Coimbra, Portugal
Company/Sponsor signatory:	AIBILI – 4C (Coimbra Coordinating Center for Clinical Research), Coimbra, Portugal
Statement:	This study was conducted in compliance with Good Clinical Practices, including the archiving of essential documents.
Report date(s):	30/12/2013
Earlier reports from the same study:	None.

Approval			
Name	**Function**	**Institution**	**Date**
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João Figueira	Study Physician	Review	2013/12/20

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2 Synopsis

2.1 Study Synopsis

Title of study: Prospective, randomized, open label, phase II study to assess efficacy and safety of Macugen® (pegaptanib 0.3 mg intravitreal injections) plus panretinal photocoagulation (PRP) and PRP (monotherapy) in the treatment of patients with high risk proliferative diabetic retinopathy (PDR).

Purpose and rationale: The purpose of this trial was to evaluate safety and to compare the efficacy of a combination of intravitreous injection of pegaptanib (0.3 mg) plus PRP versus PRP alone in the regression of retinal neovascularization in eyes with high-risk PDR:

- Group 1: The control eyes group that received the standard care (PRP according the Diabetic Retinopathy Study; DRS)
- Group 2: Eyes that received three intravitreous injections of pegaptanib (0.3 mg) plus PRP (2 weeks ± 1 week after the first injection). The PRP was performed by starting with DRS third ring, extending from the ora serrata to the mid-periphery, with coalescing spots followed (if needed) by the DRS second ring an finally DRS first ring.

Objectives:

Primary objective:

1. Regression of retinal neovascularization.

Secondary objectives:

1. Changes from baseline in Best-Corrected Visual Acuity (BCVA);
2. Changes from baseline in macular retinal thickness by Optical Coherent Tomography (OCT);
3. Changes from baseline in Visual Fields;
4. Recurrence of retinal neovascularization;
5. Number of treatments needed;
6. Additional focal or grid laser for DME;
7. Drug safety profile;
8. Need for vitrectomy due to occurrence of vitreous hemorrhage or retinal detachment.

Study rationale:

Panretinal photocoagulation (PRP) can cause regression of retinal neovascularization and reduce the risk of severe vision loss in people with proliferative diabetic retinopathy (PDR). However, this destructive treatment may be associated with side effects (e.g. pain, transient blurring, loss of

peripheral and/or night vision, increased risk of macular edema and central vision loss) and it is not always efficient in the regression of the neovascularization.

Vascular endothelial growth factor (VEGF) has been shown to play a role in retinal neovascularization and retinal vascular leakage related with PDR and diabetic macular edema.

Anti-VEGF treatments have been hypothesized as an adjunctive treatment for the management of retinal neovascularization and macular edema related with diabetic retinopathy (DR).

Anti-VEGF agents, such as Macugen®, combined with PRP are expected to control neovascularization without the need for photocoagulation of the posterior pole, around the macula, thus avoiding the major side effects of standard PRP (visual field loss).

A modification of panretinal photocoagulation (PRP) was recently proposed by Madeira et al., (2009) at the 2009 EURETINA Meeting. The described technique involves the progressive application of the DRS photocoagulation rings in a different sequence. First ring: corresponds to the DRS third ring, extruding from the ora serrata to the midperiphery. Second ring: corresponds to DRS second ring, extruding from the midperiphery towards the vortex veins. Third ring: corresponds to DRS first ring, and will only be performed if necessary. This technique resulted in less aggressive visual fields losses by achieving results with only most peripheral photocoagulation. The combination of intravitreal anti-VEGF treatment with pegaptanib, where a series of 3 injections are injected to reverse the neovascularization, while maintaining the macula dry will be completed by the more long term effect of the panretinal photocoagulation. This peripheral photocoagulation proposed is expected to eliminate the chronic VEGF stimulus by eliminating the chronic ischemic factor, while maintaining the visual fields useful for daily activities such as driving, etc.

Timelines:

Treatment duration:

Patients were assigned to one of the following treatment arms in a ratio of 1:1, i.e.:

- Group 1: The control eyes group receiving the standard care (PRP), that can be repeated during the 12 months of follow-up according to the Diabetic Retinopathy Study (DRS) protocol.
- Group 2: Trial subjects receiving a combination treatment of pegaptanib (0.3 mg) intravitreal injections at weeks 0, 6 and 12, plus PRP (2 weeks ± 1 week after the 1st injection). The PRP was performed in the first session in the most peripheral ring of photocoagulations – the DRS third ring. For the next period of follow-up, if neovascularization regression was not completed and/or if there was a recurrence, subjects received additional combination treatments, extending photocoagulations from the ora serrata to the mid-periphery, including

DRS second ring, and finally DRS first ring. The intravitreal injections had a minimum interval of 6 weeks, and the PRP had a minimum interval of 12 weeks.

Follow-up duration: One year.

Study design:

This is a prospective, randomized, open label, phase II study to assess efficacy and safety of Macugen® (pegaptanib 0.3 mg intravitreal injections) plus PRP and PRP (monotherapy) in the treatment of patients with high risk proliferative diabetic retinopathy.

Patients who signed informed consent participated in a screening period, lasting 1 to 30 days, to evaluate patient eligibility.

If patient's eligibility was confirmed, patients were randomized at month 0 in a 1:1 ratio to one of the treatment arms, i.e. standard photocoagulation (PRP), combination treatment of pegaptanib (0.3 mg) intravitreal injections plus PRP. Only one eye were selected / treated as the study eye.

Patients:

Number of patients and randomization ratio:

- 30 eyes from 30 diabetic patients were planned but only 22 patients were included
- Randomized 1:1
- Group 1 (PRP group): 15 eyes planned but only 11 eyes were included
- Group 2 (pegaptanib + PRP): 15 eyes planned but only 11 eyes were included

Clinical Site / Trial Site:

- Centre for Clinical Trials – AIBILI, Coimbra, Portugal.

Sample size justification:

Thirty (30) patients were planned to be included in this study (15 per treatment arm).

The sample size was computed for the primary objective, retinal neovascularisation regression at month-12, for a statistical power of 80% considering:

- a 60% improvement rate in the PRP group and a 99% improvement in the pegaptanib+PRP group, for a 1-sided alpha level of 0.05, and
- a drop-out rate of 10% for the 12-month period.

General Statistical Analyses:

Three populations were defined for analyses: the intent-to-treat (ITT) population (all randomized patients who received at least one treatment and for whom data from at least one follow-up visit is available); the per protocol (PP) population (patients from the ITT population who did not deviate from the protocol, i.e. with no major protocol violation), and; the safety population (patients that received at least one treatment and for whom safety data from at least one follow-up visit is available). The primary efficacy analysis was performed for the ITT and PP populations using the Pearson Chi-Square test for the proportion of patients with retinal neovascularization regression at month-12 in the two treatment arms.

Population:

The study population consists of 22 randomized male and female patients ≥18 years of age with either Type I or Type II diabetes mellitus and with high risk proliferative diabetic retinopathy.

Inclusion/Exclusion criteria:

Key Inclusion criteria

1. High-risk proliferative diabetic retinopathy (HR-PDR) eyes.
2. BCVA at baseline > 20/320 (25 letters in the ETDRS Chart) in the study eye.
3. Clear ocular media and adequate pupillary dilatation to permit good quality fundus photography.
4. Intraocular pressure < 21 mmHg.
5. Type I, or Type II diabetic subjects as defined by the WHO criteria of either gender, and aged ≥ 18 years.
6. Women using effective contraception, post-menopausal for at least 12 months prior to trial entry, or surgically sterile.
7. Ability to provide written informed consent.
8. Ability to return for all trial visits.

Key Exclusion criteria

1. Eyes with prior scatter (panretinal).
2. Focal/grid photocoagulation, within the previous 6 months.
3. Fibrovascular proliferation with retinal traction.
4. Other cause of retinal neovascularization (retinal vein occlusion, radiation retinopathy or others).
5. Atrophy/scarring/fibrosis/ hard exudates involving the center of the macula.
6. Subjects who have received YAG laser within the previous 6 months.
7. Peripheral retinal cryoablation, or laser retinopexy (for retinal tears only),

8. Significant media opacities, which might interfere with visual acuity, assessment of toxicity or fundus photography.
9. Subjects should not be entered if there was likelihood that they will require cataract surgery within the following 1 year.
10. Any intraocular surgery within 6 months before trial enrolment.
11. Previous vitrectomy.
12. HbA1C level >11% or recent signs of uncontrolled diabetes.
13. Any of the following underlying systemic diseases:
    - History or evidence of severe cardiac disease, e.g. NYHA Functional Class III or IV, clinical or medical history of unstable angina, acute coronary syndrome, myocardial infarction, or revascularization procedure within 6 months prior to baseline, or ventricular tachyarrhythmia requiring treatment.
    - History or evidence of clinically significant peripheral vascular disease such as intermittent claudication or prior amputation.
    - Clinically significant impaired renal function (serum creatinine >2.5 mg/dL or s/p renal transplant or receiving dialysis).
    - Clinically significant impaired hepatic function.
    - Stroke (within 12 months of trial entry).
    - Any major surgical procedure within one month before trial enrolment.
14. Previous radiation to the head in the region of the study eye.
15. Any prior treatment with an investigational agent for diabetic retinopathy or anti-VEGF therapy (including intravitreal, subconjunctival or subtenons corticosteroids) during the past 90 days for any other condition.
16. Known serious allergies to fluorescein used in angiography, or to components of Macugen® formulation.
17. Systolic BP > 170 (2 different readings) or diastolic BP > 100 (2 different readings).
18. Acute ocular or periocular infection.
19. Previous filtering surgery (e.g., trabeculectomy) or placement of a glaucoma drainage devise (e.g., tube-shunt surgery).
20. Use of other investigational drugs at the time of enrollment.
21. History of hypersensitivity to any of the study drugs or to drugs of similar chemical classes.
22. History of malignancy of any organ system (other than localized basal cell carcinoma of the skin), treated or untreated, within the past 5 years, regardless of whether there is evidence of local recurrence or metastases.

23. Pregnant or nursing (lactating) women, where pregnancy is defined as the state of a female after conception and until the termination of gestation, confirmed by a positive hCG laboratory test (> 5 mIU/mL).
24. Women of child-bearing potential, defined as all women physiologically capable of becoming pregnant UNLESS they were: women whose career, lifestyle, or sexual orientation precludes intercourse with a male partner; women whose partners have been sterilized by vasectomy or other means using a highly effective method of birth control (i.e. one that results in a less than 1% per year failure rate when used consistently and correctly, such as implants, injectables, combined oral contraceptives, and some intrauterine devices - IUDs). Periodic abstinence (e.g. calendar, ovulation, symptothermal, post-ovulation methods) was not acceptable.

Investigational and reference therapy:

Macugen® (pegaptanib), 0.3 mg pegaptanib intravitreal injection plus modified panretinal photocoagulation (PRP).

Standard panretinal photocoagulation (PRP) (according DRS protocol).

Evaluation criteria:

Screening visit:

Ophthalmological examination, BCVA, 7-fields digital fundus photography, fluorescein angiography, Optical Coherence Tomography (OCT) and Visual Fields.

Laboratory tests will be performed at screening visit and at the end of the study (month 12).

Month 3, 6 and 12:

Ophthalmologic examination, BCVA, 7-fields digital fundus photography, fluorescein angiography, Optical Coherence Tomography (OCT) and Visual Fields.

See below table for evaluation assessments performed during the protocol.

Group 1: Subjects receiving scatter photocoagulation, standard PRP according to the DRS protocol, at week 0. For the next period of follow-up subjects may have receive additional treatments.

Group 2: Subjects receiving intravitreal injections at week 0, 6 and 12 plus PRP 2 weeks (± 1 week) after the 1st injection (see above). For the remaining period of follow-up, subjects may have received additional treatments. Injections interval had a minimum of 6 weeks and PRP interval had a minimum of 12 weeks.

Rescue treatment with focal and/or grid laser was allowed at any time during study participation for treatment of DME for all patients included in the study.

PRP when performed in an eye with DME can increase this edema, so patients were allowed to perform focal and/or grid laser to avoid this situation.

Rescue treatment with focal or grid laser was performed according to the ETDRS protocol.

Assessments

Study phase	Screening phase	Open label treatment phase								
Visit Number	1	2	3	4	5	6	7	8	9	10
Study Month		0	1.5	3	4.5	6	7.5	9	10.5	12
Study Week		0	6	12	18	24	30	36	42	48
Procedure										
Informed Consent	X	-	-	-	-	-	-	-	-	-
Medical History and Demographics	X	-	-	-	-	-	-	-	-	-
Inclusion/Exclusion	X	-	-	-	-	-	-	-	-	-
Vital Signs	X	-	-	X	-	-	-	-	-	X
Laboratory Tests	X	-	-		-	-	-	-	-	X
Urine Pregnancy Test**	X	-	-	-	-	-	-	-	-	-
HbA1C	X	-	-	-	-	-	-	-	-	-
Ophthalmic Exam	X	X	X	X	X	X	X	X	X	X
Best-Corrected Visual Acuity	X	-	-	X	-	X	-	-	-	X
Color Fundus Photography	X	-	-	x	-	X	-	-	-	X
Fluorescein Angiography	X	-	-	X	-	X	-	-	-	X
Optical Coherence Tomography	X	-	-	X	-	X	-	-	-	X
Visual Fields	X	-	-	X	-	X	-	-	-	X
Group 1 – PRP[1]	-	X	(X)	(X)	(X)	(X)	(X)	(X)	(X)	-
Group 2 – Macugen[2]	-	X	X	X	(X)	(X)	(X)	(X)	(X)	-
Group 2 – PRP[2]	-	X		(X)	(X)	(X)	(X)	(X)	(X)	-
Concomitant Medications	X	X	X	X	X	X	X	X	X	X
Adverse Events	X	X	X	X	X	X	X	X	X	X
Study Discharge	-	-	-	-	-	-	-	-	-	X

** For patients of childbearing potential.

[1] PRP according DRS

[2] Injections with 6 weeks intervals & PRP

[2] PRP (2 weeks ± 1 week after injection) with at least 3 months intervals

Efficacy assessments:

- Visual acuity assessments using the ETDRS charts at a test distance of 4 meters

- Color Fundus photography
- Fluorescein angiography
- Optical coherence tomography
- Visual Fields
- Use of rescue treatment

Safety assessments:

- Visual acuity assessments using the ETDRS charts at a test distance of 4 meters
- Visual Fields
- Ophthalmic examinations (slit lamp exam with biomicroscopy, fundus exam and intra ocular pressure)
- Adverse events (ocular and non-ocular)

2.2 Study Conclusions

This was an exploratory study with a limited number of patients. The regression of the area of neovascularization in the group treated with Macugen associated with progressive PRP laser was not significantly better than the standard treatment with PRP laser in the treatment of eyes with HR-PDR.

Nevertheless, the eyes that received the combined treatment had less occurrence of central macular thickness increase.

As expected, visual fields remained significantly more preserved in Group 2, and this is a plus for this option of treatment, because of the associated problems in quality of life, night vision and ability to drive, reported with conventional full PRP laser.

Regarding safety, we had two cases needing vitrectomy in Group 1 versus none in Group 2, indicating that serious complications may be more frequent with conventional PRP treatment.

Considering all these factors, this study suggests that VEGF combined treatment may offer a better alternative than PRP alone to treat HR-PDR eyes, but larger prospective studies are needed to confirm this treatment option.

3 List of tables and list of figures

4 List of abbreviations and definition of terms

AE	Adverse Event
BCVA	Best Corrected Visual Acuity
BP	Blood Pressure
CI	Coordinating Investigator
CRF	Case Report/Record Form
CSR	Clinical Study Report
DA	Disc Area
DME	Diabetic Macular Edema
DR	Diabetic Retinopathy
DRS	Diabetic Retinopathy Study
ETDRS	Early Treatment Diabetic Retinopathy Study
FPFV	First Patient First Visit
HbA1C	Haemoglobin A1C
HDL	High Density Lipoprotein
ICH	International Conference on Harmonization (of Technical Requirements for Registration of Pharmaceuticals for Human Use)
IEC	Independent Ethics Committee
INN	International Nonproprietary Name
IQR	Interquartile Range
IRB	Institutional Review Board
LPLV	Last Patient Last Visit
NVD	Neovascularization of the Disc
NVE	Neovascularization Elsewere
NYHA	New York Heart Association
OCT	Optical Coherence Tomography
RA	Regulatory Authorities
RBC	Red Blood Cells
PDR	Proliferative Diabetic Retinopathy
PI	Principal Investigator
PRP	Panretinal Photocoagulation
SAE	Serious Adverse Event
SmPC	Summary Product Characteristics
VEGF	Endothelial Growth Factor
WBC	Whole Blood Cells

5 Ethics

5.1 Independent Ethics Committee (IEC) or Institutional Review Board (IRB)

The study and any amendments were reviewed by the National Independent Ethics Committee CEIC – Comissão de Ética para a Investigação Clínica. The Institutional Review Board of AIBILI (CES – Comissão de Ética para a Saúde) was notified directly by the Independent Ethics Committee (CEIC).
The study was additionally approved by:

1. The National Data Protection Committee, CNPD – Comissão Nacional de Protecção de Dados.
2. The National Competent Authorithy (CA), INFARMED – Autoridade Nacional do Medicamento e Produtos de Saúde, I.P.

Approvals are provided in appendix.

5.2 Ethical conduct of the study

The study was conducted according to the ethical principles of the Declaration of Helsinki.

5.3 Patient information and consent

Informed consent was obtained from each subject in writing before randomization. The study was described by a study coordinator and/or investigator, who answered any questions, and written information was also provided. Samples of the written information was given to each subject
A sample of patients information and consent form is provided in aappendix.

6 Investigators and study administrative structure

The administrative structure is found below (e.g. principal investigator, affiliations and qualifications, other important staff, and Coordinating Center team).

Study Personnel

Coordinating Investigator	José Cunha-Vaz, AIBILI
Study Physicians	José Cunha-Vaz & João Figueira, AIBILI
Coordinating Center Responsible	Cecília Martinho (until 2010) & Sandrina Nunes Coimbra Coordinating Centre for Clinical Research (4C-AIBILI)
Study Statistician	Sandrina Nunes, 4C-AIBILI
Study Data Manager	Miguel Costa, 4C-AIBILI
Study Manager	Ana Pedroso, 4C-AIBILI
Study Monitors	Ana Pedroso & Sónia Simões (trainee), 4C-AIBILI

The protocol and protocol approval page are provided in appendix.

The site personnel is described in the delegation of duties of the clinical center (provided in appendix)

The principal investigator approval page if provided in appendix.

7 Introduction

Panretinal photocoagulation (PRP) can cause regression of retinal neovascularization and reduce the risk of severe vision loss in people with proliferative diabetic retinopathy (PDR). However, this destructive treatment may be associated with side effects (e.g. pain, transient blurring, loss of peripheral and/or night vision, increased risk of macular edema and central vision loss) and it is not always efficient in the regression of the retinal neovascularization.

Vascular endothelial growth factor (VEGF) has been shown to play a role in retinal neovascularization and retinal vascular leakage related with PDR and diabetic macular edema.

Anti-VEGF treatments have been hypothesized as an alternative adjunctive treatment for the management of retinal neovascularization and macular edema related with diabetic retinopathy (DR).

The Macugen Diabetic Retinopathy Study Group reported that during the clinical trial testing pegaptanib in the treatment of diabetic macular edema, most subjects with retinal neovascularization at baseline assigned to pegaptanib showed regression of retinal neovascularization by week 36 [Adamis, 2006]. These findings suggest a direct effect of pegaptanib upon retinal neovascularization in patients with diabetes mellitus.

Gonzalez et al. [Gonzalez, 2007] [Gonzalez, 2009] performed a clinical trial comparing the regression of proliferative diabetic retinopathy (PDR), in 10 eyes treated with pegaptanib, versus 10 eyes treated with panretinal photocoagulation (PRP), and concluded that pegaptanib appears to effectively induce regression of PDR and decrease the anatomic extent of diabetic macular edema. Use of pegaptanib may reduce the need for and/or extent of PRP in PDR thus optimizing therapeutic results while diminishing iatrogenic effects inherent to retinal ablation.

This study was designed to evaluate the safety and determine the efficacy of PRP monotherapy or combination therapy (pegaptanib 0.3 mg plus PRP) in patients with Type I or Type II diabetes mellitus and with high risk proliferative diabetic retinopathy.

Where high risk proliferative diabetic retinopathy is defined as:

- NVD $\geq$ 1/4 DA (disc area) or NVE $\geq$ 1/2 DA or
- NVE < 1/2 DA + vitreous and/or pre-retinal haemorrhage and/or rubeosis and/or traccional retinal detachment.
- NVD <1/4 DA + vitreous and/or pre-retinal haemorrhage and/or rubeosis and/or traccional retinal detachment.

Data from this study was supposed to be used to support Portuguese Guidelines for the treatment of Type I or Type II diabetes mellitus patients at high risk proliferative diabetic retinopathy.

8 Study objectives

The primary objective of this study is to demonstrate superiority of one of the treatment arms: PRP monotherapy or combination therapy (pegaptanib 0.3 mg plus PRP) over a 12-month treatment period in the following: Regression of retinal neovascularization.

The progression of retinal neovascularization (1 year end point) was assessed.
Retinal neovascularization was measured in disc area units, and progression of neovascularization was defined as an increasing of 0.5 disc area associated or not with vitreous haemorrhage, and/or pre-retinal haemorrhage, and/or rubeosis, and/or traccional retinal detachment.

The evaluation of the neovascularization was performed based on color fundus photography (retinography) and, if necessary, on fluorescein angiography. The comparison was performed versus the baseline assessment, to find out if there was regression, that may be total (disappearing of the neovessels) or partial (persistency of neovessels , but with decreases of its size – in disc areas).

Secondary objectives are:

1. Changes from baseline in Best-Corrected Visual Acuity (BCVA)
2. Changes from baseline in macular retinal thickness by Optical Coherent Tomography (OCT)
3. Changes from baseline in Visual Fields
4. Recurrence of retinal neovascularization
5. Number of treatments needed
6. Additional focal or grid laser for DME
7. Drug safety profile
8. Need for vitrectomy due to occurrence of vitreous hemorrhage or retinal detachment relative to the two treatment arms.

9 Investigational plan

9.1 Overall study design and plan-description

This was a prospective, randomized, open label, phase II study to assess efficacy and safety of Macugen® (pegaptanib 0.3 mg intravitreal injections) plus panretinal photocoagulation (PRP) and PRP (monotherapy) in the treatment of patients with high risk proliferative diabetic retinopathy.

Patients who signed informed consent participated in a screening period, lasting 1 to 30 days, to evaluate patient eligibility.

If patient's eligibility is confirmed, patients were randomized at month 0 in a 1:1 ratio to one of the two treatment arms, i.e. standard photocoagulation (PRP) (PRP mandatory at week 0) or combination treatment of pegaptanib (0.3 mg) intravitreal injections plus PRP (intravitreal injections mandatory at week 0, 6 and 12 and PRP mandatory 2 weeks ± 1 week after the 1st injection) (Figure 9-1).

Randomized patients entered a treatment phase during 12 months. Visits to assess safety and efficacy were scheduled at 6 week intervals during the period of the study. The assessment to address the primary objective was performed at the end of month-3 and month-12.

Patients withdrawn from the study prior to completion of the 12-month treatment phase were asked to return at month 12 for a final evaluation.

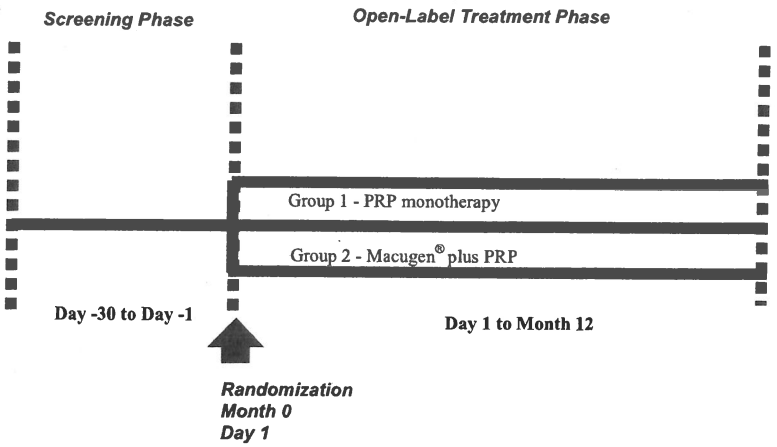

Figure 9-1. Study design.

9.2 Discussion of study design, including the choice of control groups

The control group (Group 1 – PRP) was chosen based on the standard treatment for HR-PDR, i.e., PRP – the only approved treatment for this condition at the time of the beginning of the study.

9.3 Selection of study population

The study population consisted of a representative group of male and female outpatients (≥ 18 years old) who meet all the following inclusion criteria and none of exclusion criteria.

9.3.1 Inclusion criteria

Patients eligible for inclusion in this study had all of the following criteria:

1. High-risk proliferative diabetic retinopathy (HR-PDR) eyes.
2. BCVA at baseline > 20/320 (25 letters in the ETDRS Chart) in the study eye.
3. Clear ocular media and adequate pupillary dilatation to permit good quality fundus photography.
4. Intraocular pressure < 21 mmHg.
5. Type I, or Type II diabetic subjects as defined by the WHO criteria of either gender, and aged ≥ 18 years.
6. Women using effective contraception, post-menopausal for at least 12 months prior to trial entry, or surgically sterile.
7. Ability to provide written informed consent.
8. Ability to return for all trial visits.

9.3.2 Exclusion criteria

1. Eyes with prior scatter (panretinal).
2. Focal/grid photocoagulation, within the previous 6 months.
3. Fibrovascular proliferation with retinal traction.
4. Other cause of retinal neovascularization (retinal vein occlusion, radiation retinopathy or others).
5. Atrophy/scarring/fibrosis/ hard exudates involving the center of the macula.
6. Subjects who have received YAG laser within the previous 6 months.
7. Peripheral retinal cryoablation, or laser retinopexy (for retinal tears only),
8. Significant media opacities, which might interfere with visual acuity, assessment of toxicity or fundus photography.
9. Subjects should not be entered if there was likelihood that they will require cataract surgery within the following 1 year.

10. Any intraocular surgery within 6 months before trial enrolment.
11. Previous vitrectomy.
12. HbA1C level >11% or recent signs of uncontrolled diabetes.
13. Any of the following underlying systemic diseases:
    i. History or evidence of severe cardiac disease, e.g. NYHA Functional Class III or IV, clinical or medical history of unstable angina, acute coronary syndrome, myocardial infarction, or revascularization procedure within 6 months prior to baseline, or ventricular tachyarrhythmia requiring treatment.
    ii. History or evidence of clinically significant peripheral vascular disease such as intermittent claudication or prior amputation.
    iii. Clinically significant impaired renal function (serum creatinine >2.5 mg/dL or s/p renal transplant or receiving dialysis).
    iv. Clinically significant impaired hepatic function.
    v. Stroke (within 12 months of trial entry).
    vi. Any major surgical procedure within one month before trial enrolment.
14. Previous radiation to the head in the region of the study eye.
15. Any prior treatment with an investigational agent for diabetic retinopathy or anti-VEGF therapy (including intravitreal, subconjunctival or subtenons corticosteroids) during the past 90 days for any other condition.
16. Known serious allergies to fluorescein used in angiography, or to components of Macugen® formulation.
17. Systolic BP > 170 (2 different readings) or diastolic BP > 100 (2 different readings).
18. Acute ocular or periocular infection.
19. Previous filtering surgery (e.g., trabeculectomy) or placement of a glaucoma drainage devise (e.g., tube-shunt surgery).
20. Use of other investigational drugs at the time of enrollment.
21. History of hypersensitivity to any of the study drugs or to drugs of similar chemical classes.
22. History of malignancy of any organ system (other than localized basal cell carcinoma of the skin), treated or untreated, within the past 5 years, regardless of whether there is evidence of local recurrence or metastases.
23. Pregnant or nursing (lactating) women, where pregnancy is defined as the state of a female after conception and until the termination of gestation, confirmed by a positive hCG laboratory test (> 5 mIU/mL).

24. Women of child-bearing potential, defined as all women physiologically capable of becoming pregnant UNLESS they were: women whose career, lifestyle, or sexual orientation precludes intercourse with a male partner; women whose partners have been sterilized by vasectomy or other means using a highly effective method of birth control (i.e. one that results in a less than 1% per year failure rate when used consistently and correctly, such as implants, injectables, combined oral contraceptives, and some intrauterine devices - IUDs). Periodic abstinence (e.g. calendar, ovulation, symptothermal, post-ovulation methods) was not acceptable.

Reliable contraception was maintained throughout the study and for 30 days after study drug discontinuation.

No additional exclusions was applied by the investigator, in order to ensure that the study population was representative of all eligible patients.

9.3.3 Removal of patients from therapy or assessment

If a patient prematurely discontinues from the study at any time either at his or her request or at the investigator's discretion, the reason(s) for withdrawal was recorded on the CRF and source document. Patients who withdraw from the study prematurely underwent a termination visit (at month-12), when possible.

If such withdrawal occurs, or if the patient fails to return for visits, the investigator determined the primary reason for a patient's premature withdrawal from the study and recorded this information on the CRF.

Patients could withdraw at any time if the investigator concluded that it was in the patient's best interest for any reason.

Patients could voluntarily withdraw from the study for any reason at any time. They may be considered withdrawn if they stated an intention to withdraw, or fail to return for visits, or became lost to follow up for any other reason. If a patient chooses to stop treatment, the investigator encouraged the patient to return for a last visit at month-12.

Study drug was discontinued and the patient withdrawn from the trial when the investigator determined that continuing it would result in a significant safety risk for that patient. The following circumstances required study drug discontinuation:

- Withdrawal of informed consent.
- Pregnancy.
- Any other protocol deviation that resulted in a significant risk to the patient's safety.

In addition to these requirements for study drug discontinuation, the investigator may discontinue study drug for a given patient if, on balance, he/she though that continuation would be detrimental to the patient's well-being.

9.4 Treatments

9.4.1 Treatments administered

Patients were assigned to one of the following two treatment arms in a ratio of 1:1, i.e.:

- Group 1: The control eyes group received the standard care (PRP), that could be repeated during the 12 months of follow-up according to the Diabetic Retinopathy Study (DRS) protocol.
- Group 2: Trial subjects received a combination treatment of pegaptanib (0.3 mg) intravitreal injections at weeks 0, 6 and 12, plus PRP (2 weeks ± 1 week after the 1st injection). The PRP was performed in the first session in the most peripheral ring of photocoagulations – the DRS third ring. For the next period of follow-up, if neovascularization regression was not completed and/or if there was a recurrence, subjects received additional combination treatments, extending photocoagulations from the ora serrata to the mid-periphery, including DRS second ring, and finally DRS first ring. The intravitreal injections had a minimum interval of 6 weeks, and the PRP had a minimum interval of 12 weeks.

9.4.2 Identity of investigational product(s)

- 0.3 mg pegaptanib (labeled Macugen® 0.3 mg)
- Panretinal laser photocoagulation (PRP)

Pegaptanib was stored according to the label instructions contained on the Summary Product Characteristics (SmPC) and was kept in a secure locked facility.

Once it was used marketed Macugen®, each vial was labeled with the appropriate information stating that the medication was for use in this clinical trial only. Medication labels complied with the legal requirements and were printed in the local language. The storage conditions for study drug were described on the medication label.

Additionally outside the Macugen® box a label was stamped with the statement that the medication was intended to be used only in the designed clinical trial and that one vial was to be used for one patient only.

The laser treatment technique applied followed the DRS protocol (DRS third, second and first rings). For Group 1 the DRS guidelines for PRP treatment was followed (standard PRP, i.e. starting with the first DRS ring) while for Group 2 the treatment started, in the first visit, in the most peripheral ring (DRS third ring).

9.4.3 Method of assigning patients to treatment groups

At Visit 2, month 0, all patients who fulfill all the inclusion/exclusion criteria received the lowest available number on the randomization list. This number assigns them to one of the treatment arms. The investigator entered the randomization number on the CRF.

The randomization numbers were generated using the following procedure to ensure that treatment assignment was unbiased and concealed from patients and investigator staff. A randomization list was produced by AIBILI using a validated system that automates the random assignment of treatment arms to randomization numbers in the specified ratio.

The randomization scheme was reviewed by Biostatistics Personnel and locked by them after approval.

The trial site received initially 2 numbered envelopes corresponding to two treatments/randomization numbers. After confirmed patient eligibility, investigator opens the envelope with the lowest randomization number to assign a treatment to a patient. The opened envelope was signed and dated by the investigator. Patient number and initials were also written on the opened envelope to confirm assignment of this specific treatment to a specific patient. Once an envelope was assigned to a patient it was not be reused.

The randomization number was recorded on the CRF.

9.4.4 Selection of doses in the study

The trial site was supplied by Pfizer with the study drug (Macugen®, 0.3 mg) in commercial packaging labeled and identifying that the specific study medication was for use in this specific study.

Study drug dose adjustments were not permitted.

9.4.5 Selection and timing of dose for each patient

There were two treatment phases for intravitreal injections (Group 2): the loading and the maintenance phase.

Loading Phase (Weeks 0, 6 and 12):

Treatment with intravitreal injections with pegaptanib 0.3 mg was initiated with a fixed loading phase of one injection each 6 weeks for three consecutive visits (Week 0; Week 6 and Week 12). PRP treatment was performed after the 1st injection at Week 0 and repeated if necessary after Week 12 (2 weeks ±1 week).

Maintenance Phase (after Week 12):

If neovascularization regression was not completed and/or if there was a recurrence, subjects received additional combination treatments (pegaptanib 0.3 mg plus PRP – 2 weeks ±1 week after injection). The injections interval had a minimum of 6 weeks, and the PRP interval had a minimum of 12 weeks.

After the suspension of treatment, intravitreal injections were reinitiated if there was a persistent recurrence of retinal neovascularization due to proliferative diabetic retinopathy in the opinion of the investigator (e.g. suggested by fundus observation or angiography and /or fundus photography). In this case the patient was treated at 6 weeks intervals.

All dosages prescribed and dispensed to the patient and all dose changes during the study were recorded on the CRF.

For patients included in Group 1, PRP treatment was performed at Week 0 according to the DRS guidelines. Subsequent laser treatments was performed at intervals no shorter than 3 months from the last treatment if deemed necessary by the investigator.

9.4.6 Blinding

This is a randomized, open label study. Blinding and emergency unblinding procedures are not applicable.

9.4.7 Prior and concomitant therapy

Any concomitant medications including prescription drugs or over-the-counter preparations used by a patient since the date of enrolment (screening visit) until the conclusion of the study participation (except for routine medications given for ocular procedures required by the protocol, i.e. fluorescein, dilating drops, topical antibiotic, topical anesthetic) were recorded on the Concomitant Medications / Non-Drug Therapies CRF page including start and stop dates.

The following treatments were not allowed during the study (for both eyes):

- Treatment with anti-angiogenic drugs (ranibizumab, anecortave acetate, bevacizumab, etc.) or intravitreal corticosteroids in either eye.
- Systemic medications known to be toxic to the lens, retina or optic nerve, including Deferoxamine, Chloroquine/ hydroxychloroquine (Plaquenil), Tamoxifen, Phenothiazines and Ethambutol.
- Treatment with glitazones when newly started during the study.

Concerning the fellow eye, the following treatment is not allowed during the study:

- Anti-VEGF therapy.

The fellow eye received only gold standard (laser photocoagulation) for the treatment of PDR or DME.

The investigator instructed the patient to notify the trial site about any new medications he/she take after the start of the study.

9.4.8 Treatment compliance

Treatment was performed at site. Treatment compliance is not applicable.

9.5 Efficacy and safety variables

9.5.1 Efficacy and safety measurements assessed and flow chart

Table 9-1 lists all of the assessments and indicates with an "X" the visits when they were performed. Patients should be seen for all visits on the designated day or as close to it was possible.

At a minimum, patients were contacted for safety evaluations during the 30 days following the last dose of study drug or last completed visit (whichever is later), including final contact at the 30-day point. Documentation of attempts to contact the patient were recorded in the patient record.

Efficacy assessments

The following assessments were performed for efficacy of pegaptanib on retinal structure, visual function and neovascularization:

- Best-Corrected Visual Acuity (BCVA) with ETDRS chart at 4 meters,
- Optical Coherence Tomography,
- Color fundus photography and fluorescein angiography,
- Visual Fields.

Safety assessments

Safety assessments consisted of monitoring and recording all adverse events (ocular and non-ocular) and serious adverse events (ocular and non-ocular), ophthalmic examinations (slit lamp exam with biomicroscopy, fundus exam and intra ocular pressure), and visual acuity assessments.

Table 9-1. Evaluation and visit schedule.

Study phase	Screening phase	Open label treatment phase								
Visit Number	1	2	3	4	5	6	7	8	9	10
Study Month		0	1.5	3	4.5	6	7.5	9	10.5	12
Study Week		0	6	12	18	24	30	36	42	48
Procedure										
Informed Consent	X	-	-	-	-	-	-	-	-	-
Medical History and Demographics	X	-	-	-	-	-	-	-	-	-
Inclusion/Exclusion	X	-	-	-	-	-	-	-	-	-
Vital Signs	X	-	-	X	-	-	-	-	-	X
Laboratory Tests	X	-	-		-	-	-	-	-	X
Urine Pregnancy Test**	X	-	-	-	-	-	-	-	-	-
HbA1C	X	-	-	-	-	-	-	-	-	-
Ophthalmic Exam	X	X	X	X	X	X	X	X	X	X
Best-Corrected Visual Acuity	X	-	-	X	-	X	-	-	-	X
Color Fundus Photography	X	-	-	x	-	X	-	-	-	X
Fluorescein Angiography	X	-	-	X	-	X	-	-	-	X
Optical Coherence Tomography	X	-	-	X	-	X	-	-	-	X
Visual Fields	X	-	-	X	-	X	-	-	-	X
Group 1 – PRP[1]	-	X	(X)	(X)	(X)	(X)	(X)	(X)	(X)	-
Group 2 – Macugen[2]	-	X	X	X	(X)	(X)	(X)	(X)	(X)	-
Group 2 – PRP[2]	-	X		(X)	(X)	(X)	(X)	(X)	(X)	-
Concomitant Medications	X	X	X	X	X	X	X	X	X	X
Adverse Events	X	X	X	X	X	X	X	X	X	X
Study Discharge	-	-	-	-	-	-	-	-	-	X

** For patients of childbearing potential.

[1] PRP according DRS

[2] Injections with 6 weeks intervals & PRP - [2] PRP (2 weeks ± 1 week after injection) with at least 3 months intervals

9.5.2 Appropriateness of measurements

The efficacy and safety parameters selected are the standard parameters used for this indication and for type of patient population.

9.5.3 Primary efficacy variable(s)

The primary efficacy variable were defined as the regression of retinal neovascularization (assessed on color fundus photography and, if necessary, on fluorescein angiography). Regression, total or partial, was determined by comparison to the baseline evaluation.

9.6 Data quality assurance

AIBILI monitors reviewed the CRFs entered by investigational staff for completeness and accuracy and instruct the site personnel to make any required corrections or additions.

After these actions have been completed and the database has been declared to be complete and accurate, it was locked and data made available for data analysis.

Data from the CRFs were entered into the study database by AIBILI staff using single data entry. Verification was performed manually by a separate member of the AIBILI by comparing the CRF to the data entered into the database.

9.7 Statistical methods planned in the protocol and determination of sample size

9.7.1 Statistical and analytical plans

The ITT and PP sets was used for the primary efficacy analysis according to the following null (H0) and alternative (H1) hypotheses:

- H0: there is no difference at month-12 between the PRP group and the Pegaptanib+PRP group in the proportion of subjects with neovascullariuarion regression.
- H1: there is a difference between these groups.

The hypothesis was tested using the Pearson Chi-Square test and an alpha level of 0.05 was considered.

The ITT analysis was considered the primary analysis. If the ITT and the PP analyses yielded the same results, the PP was used to provide supportative evidence of the magnitude of treatment effect among patients who received the treatment. If the results of the 2 methods differ, exploratory analyses were planned to evaluate the factors that could have a contribution to the differences.

To assess the robustness of the primary analysis, an alternative analysis was planned considering the neovascularization area (as assessed on the color fundus photographs, and, is necessary on the fluorescein angiograms). For this analysis the progression of the neovascularization was compared between the 2 treatments arms for months 0, 3, 6 and 12, using and an ANOVA analysis for repeated measures was planned.

Statistically significant changes from baseline in BCVA, central macular retinal thickness (by OCT) and Visual Fields were tested for the 2 treatment arms using a MANOVA analysis for repeated measures.

For the remaining secondary variables (recurrence of retinal neovascularization; number of treatments needed, additional focal or grid laser for DME; drug safety profile and need for vitrectomy due to occurrence of vitreous hemorrhage or retinal detachment) statistically significant differences between treatment arms were planned to be tested using the Pearson Chi-Square test.

For the safety analysis only treatment-emergent adverse events were considered (e.g., AEs with an onset after the start of the particular treatment, or AEs present prior treatment but with increased severity). The number and percentage of subjects reporting AEs is presented.

No interim analyses were planned for this study. However, interim analyses were planned initially if necessary, due to safety concerns.

9.7.2 Determination of sample size

The sample size calculation was based on the primary efficacy analysis for the proportion of subjects with neovascularization regression at month-12.

Sample size for this study was computed using the SampleSize software (SPSS Inc., Version 2.0) and estimates from the previous studies [ETDRS, 1991], [Adamis, 2006], [Gonzalez, 2007] and [Gonzalez, 2009].

Assuming an 60% improvement rate for the PRP group and a 99% improvement rate for the Pegaptanib+PRP group, with a 1-sided alpha level of 0.05, a statistical power of 80% is achieved with 26 subjects (13 subjects per treatment arm). Accounting for a 10% dropout rate, for the 12-months follow-up period, 30 subjects were planned for in this study (15 subjects per treatment arm).

9.8 Changes in the conduct of the study or planned analyses

The planned statistical planning was reviewed due to the low recruitment rate, only 22 patients were recruited instead of 30 patients as initially planned.

Non-parametric tests were used due to the non-normal distribution of the data.

Data was described using the median and the Interquartile Range (IQR), instead of the mean and standard deviation.

To compare the two treatment groups the Mann-Whitney test was used (for continuous variables) while to compare values between visits the Freidman test (for 3 or more visits) and/or the Wilcoxon test (for 2 visits) were used.

For nominal and/or categorical variables the Person χ^2 was used.

10 Study Patients

10.1 Disposition of patients

Patients disposition by treatment arm are presented in Table 10-1.

Table 10-1. Patient disposition.

	Total	Group 1	Group 2
Patients			
Plannned	30	15	15
Screened	24	---	---
Failed inclusion criteria	2	---	---
Eligible / Randomized	22	11	11
Exposed	22	11	11
Completed	20	9	11
Discontinued	2	2	0
Main cause of discontinuation			
Death	0	---	---
Adverse event(s)	2	2	0
Lack of efficacy	0	---	---
Protocol violation(s)	0	---	---
Administrative reasons	0	---	---
Other	0	---	---

10.2 Protocol deviations

Protocol deviations are listed in Appendix 16.1.10.

The major protocol deviation is related with the recruitment.

The recruitment period was extended from October 2010 (6 months) to October 2011 (1 year and 6 months). Even though, only 22 patients were recruited from the 30 initially planned.

11 Efficacy evaluation

11.1 Data sets analyzed

Subjects were grouped according to the treatment administered (n=22). First group corresponds to standard PRP, which included 11 subjects at baseline and the second group to pegaptanib 0.3mg + modified PRP, which included at baseline 11 subjects Table 11-1.

There were few missing data and therefore, the present analysis includes cases with missing data.

The CONSORT Flow Chart is presented in Figure 11-1.

Table 11-1. Analysis populations.

	Group 1 N = 9	Group 2 N = 11
Analysis population	---	---
Intent-to-treat (ITT)	11	11
Safety (> 1 evaluation)	9	11
Per protocol (PP)	9	11
Evaluable patients	9	11

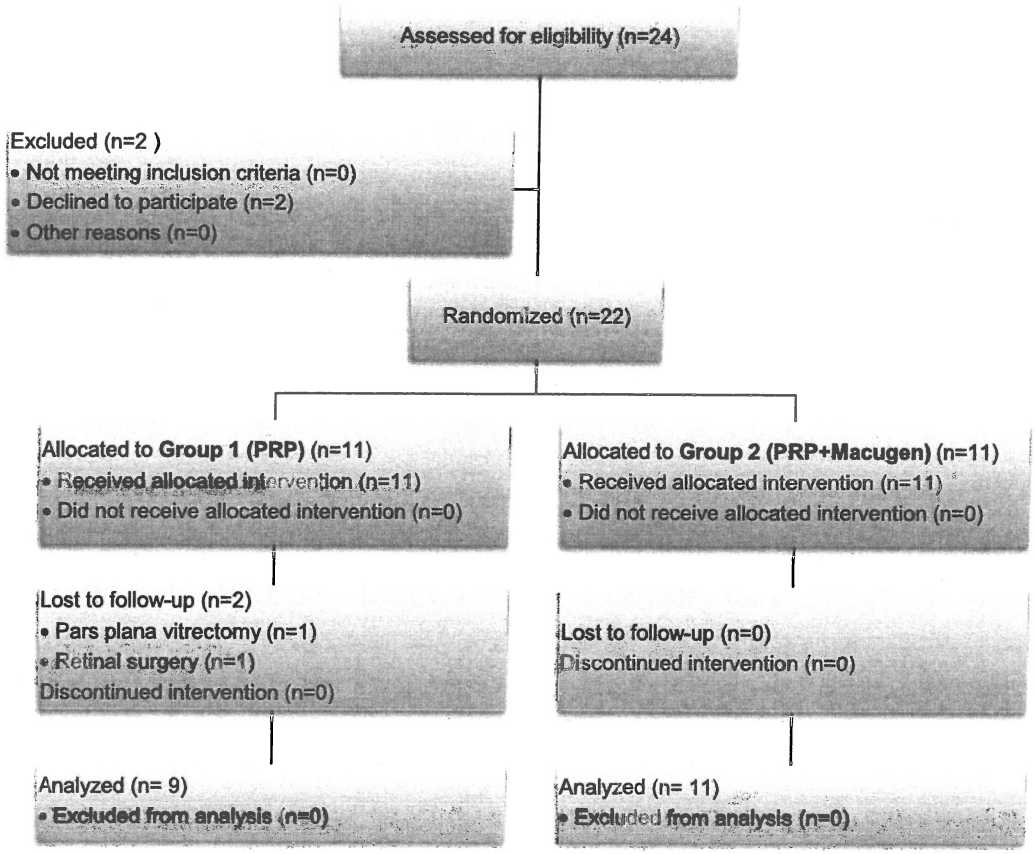

Figure 11-1. CONSORT flow chart.

11.1.1 Demographic and other baseline characteristics

Demographic characteristics are described in the following tables (for the ITT population Table 11-2 and for the PP population Table 11-3).

Regarding the ITT population (Table 11-2), there were 22 subjects included, 11 allocated to standard Group 1 (average age 56 years, range 46-64) and 11 allocated to Group 2 (average age 52 years, range 46-60).

Age distribution was fairly equally distributed across treatment groups, as well as the other parameters, i.e., blood pressure and HbA1C level. Additionally, the same trend was observed for the ophthalmologic examinations, i.e., intraocular pressure, central retinal thickness, visual fields and regression of the NV area.

Table 11-2 Demographic summary by treatment group (baseline population) – ITT population.

		Median (IQR)				Wilcoxon Test
		N	Group 1	N	Group 2	*p-value*
Age (years)		11	56 (46-64)	11	52 (46-60)	0.869
HbA1C level (%)		11	7.5 (7.0-9.1)	11	7.8 (7.0-8.5)	0.948
Heart Rate (bpm)		10	78 (76-80)	10	77 (76-84)	0.802
Blood Pressure	Systolic (mmHg)	10	146 (136-147)	10	139 (135-146)	0.502
	Diastolic (mmHg)	10	80 (77-81)	10	81 (79-82)	0.240
Intraocular pressure (mmHg)		11	17 (16-17)	11	16 (14-19)	0.251
BCVA (letters)		11	78 (73-84)	11	77 (60-78)	0.222
Retinal Thickness (µm)		11	286 (246-343)	9	323 (273-594)	0.425
Visual Fields – Observed Fields (#)		10	45 (34-54)	10	31.5 (17-43)	0.089
Neovascularization area	NVE	11	1.5 (0.11-2.75)	11	2.2 (0.77-3.63)	0.224
	NVD	11	0 (0-0.99)	11	0 (0-1)	0.889
	Total	11	2.53 (0.33-2.95)	11	2.9 (0.88-4.4)	0.341

		N (%)				χ^2 Test
		N	Group 1	N	Group 2	*p-value*
Type of diabetes	1	11	2 (18.2%)	11	3 (27.3%)	1.000
	2		9 (81.8%)		8 (47.1%)	

Regarding the PP population (Table 11-3), there were 20 subjects enrolled in this study, 9 allocated to Group 1 (average age 56 years, range 46-63) and 11 allocated to Group 2 (average age 52 years, range 46-60).

Age distribution was fairly equally distributed across treatment groups, as well as regarding the remaining parameters, i.e., blood pressure and HbA1C level. Additionally, the same trend was observed for the ophthalmologic examinations, i.e., intraocular pressure, central retinal thickness, visual fields and the regression of NV area.

Table 11-3. Demographic summary by treatment group (baseline population) – PP population.

		Median (IQR)				Wilcoxon Test
		N	Group 1	N	Group 2	*p-value*
Age (years)		9	56 (46-63)	11	52 (46-60)	0.91
HbA1C level (%)		9	7.6 (7.0-9.1)	11	7.8 (7.0-8.5)	0.68
Heart Rate (bpm)		9	78 (76-80)	10	77 (76-84)	0.77
Blood Pressure	Systolic (mmHg)	9	139 (136-147)	10	139 (135-146)	0.74
	Diastolic (mmHg)	9	80 (78-81)	10	81 (79-82)	0.36
Intraocular pressure (mmHg)		9	17 (16-17)	11	16 (14-19)	0.28
BCVA (letters)		9	80 (77-84)	11	77 (60-78)	0.14
Retinal Thickness (µm)		9	286 (246-299)	9	323 (273-594)	0.27
Visual Fields – Observed Fields (#)		9	40 (34-51)	10	31.5 (17-43)	0.15
Neovascularization area	NVE	9	1.5 (0.11-2.53)	11	2.2 (0.77-3.63)	0.18
	NVD	9	0 (0-0.33)	11	0 (0-1)	0.97
	Total	9	1.5 (0.33-2.95)	11	2.9 (0.88-4.4)	0.30

		Proportion (%)				χ^2 Test
		N	Group 1	N	Group 2	*p-value*
Type of diabetes	1	9	2 (22.2)	11	3 (27.8)	0.795
	2		7 (77.8)		8 (72.7)	

Since no differences were found between the ITT and the PP populations, efficacy and safety analysis were performed using PP population.

11.2 Efficacy results and tabulations of individual patient data

11.2.1 Analysis of efficacy

The following table and graphics showed the efficacy results regarding treatment for the primary efficacy variable Area of NV (Table 11-4 and Figure 11-2).

No statistically significant differences were found between the combination therapy and the monotherapy groups over the 12-month treatment period.

No statistically significant differences between groups were found at month-12 for the proportion of subjects with neovascularization regression.

Table 11-4. Primary efficacy parameter.

		Median (IQR)		Wilcoxon Test
		Group 1	Group 2	*p-value*
Neovascularization Area (Disc Area)	NV Change (week 48 –screening)	-0.33 (-0.45-0.5)	-0.4 (-1.8-1.1)	0.870
	Screening	1.5 (0.33-2.95)	2.9 (0.88-4.4)	0.305
	W12	1.32 (0.5-2.4)	1.3 (0.35-2.6)	0.761
	W24	1.54 (0.7-2.4)	1.9 (0.15-5.24)	0.970
	W48	1.95 (0.9-2.5)	1.725 (0.15-5.5)	0.806
Between visits	*p*-value	0.972	0.110	---

Table 11-5. Primary efficacy parameter NV regression

		N(%)		χ^2 Test
		Group 1	Group 2	*p-value*
Neovascularization Regression from week 0 to week 48	Yes	5	6	0.964
	No	4	5	

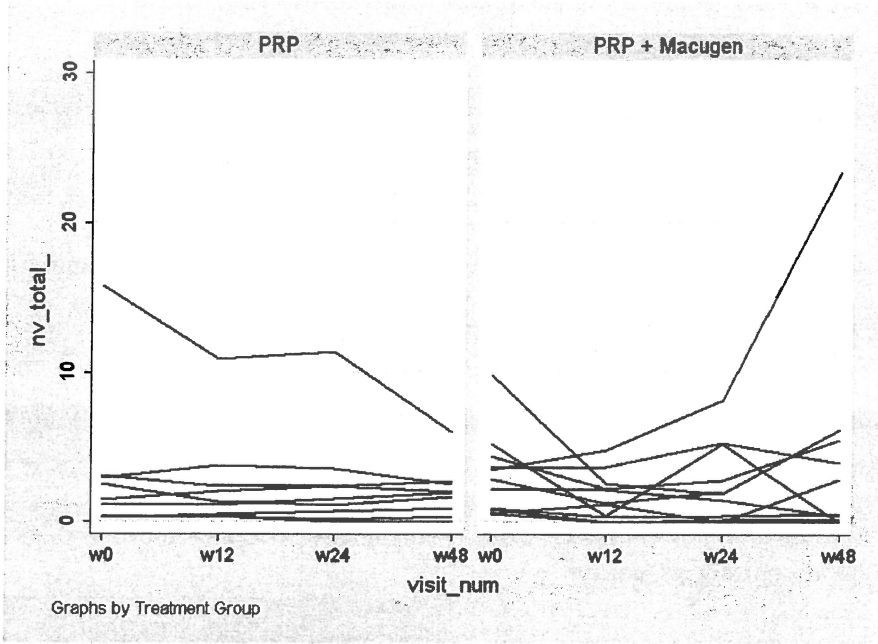

Figure 11-2. Total Neovascularization area (in Disc Area) by patients according to the treatment arms.

Regarding secondary efficacy results (Table 11-6, Figure 11-3Figure 11-4 and Figure 11-5) no significant statistically differences between groups were found between groups for the BCVA, the central macular retinal thickness and for the visual fields. Additionally, 3 subjects from Group 1 (PRP) and 7 subjects from Group 2 (PRP+Macugen) had recurrence of neovascularization at month-12, but with no statistical significant differences.

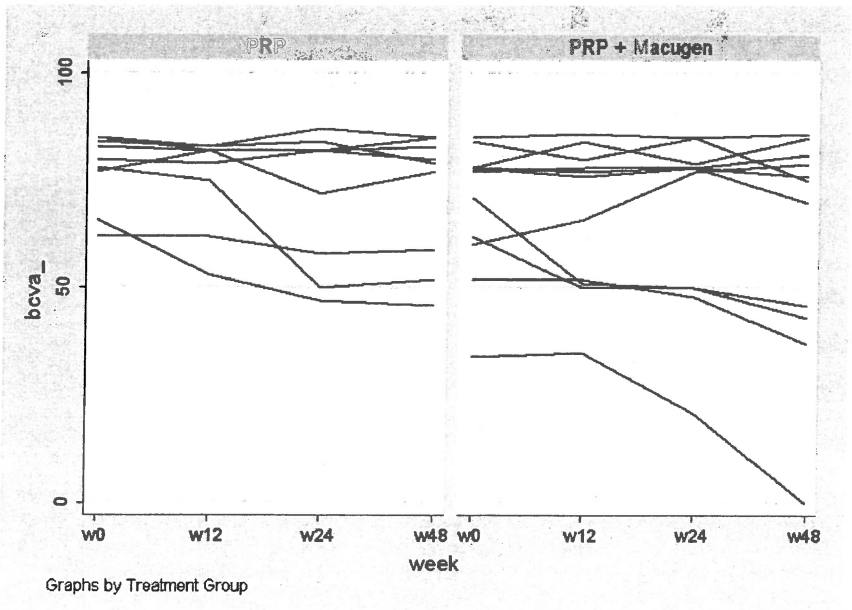

Figure 11-3. BCVA (letters) by patients according to treatment arms.

Table 11-6. Secondary efficacy results.

		Median (IQR)		Wilcoxon Test
		Group 1	Goup 2	*p*-value
BCVA (letters)	Screening	80 (77-84)	77 (60-78)	0.1359
	W12	82 (75-82)	76 (51-80)	0.2091
	W24	82 (58-82)	78 (50-79)	0.4683
	W48	79 (59-83)	75 (43-81)	0.3412
Between visits (Friedman test)	*p*-value	0.221	0.597	---
Retinal Thickness (µm)	Screening	286 (246-299)	323 (273-594)	0.2694
	W12	304 (271-396)	317 (277-569)	0.4250
	W24	361.5 (286-399)	334.5 (299-674)	0.8589
	W48	326 (275.5-417)	322 (256-612)	0.9292
Between visits	*p*-value	0.0455	-----	---
Visual Fields (#)	Screening	40 (34-51)	31.5 (17-43)	0.1529
	W12	37.5 (25.5-46.5)	27 (23-47)	0.5078
	W24	41 (17-47)	31 (5-36)	0.5940
	W48	27 (21-50)	23 (9-48)	0.4869
Between visits	*p*-value	0.0319	0.5465	

		N (%)		χ^2 Test
		Group 1	Group 2	*p-value*
Recurrence of neovascularization	No	6 (60%)	4 (40%)	0.178
	Yes	3 (30%)	7 (70%)	

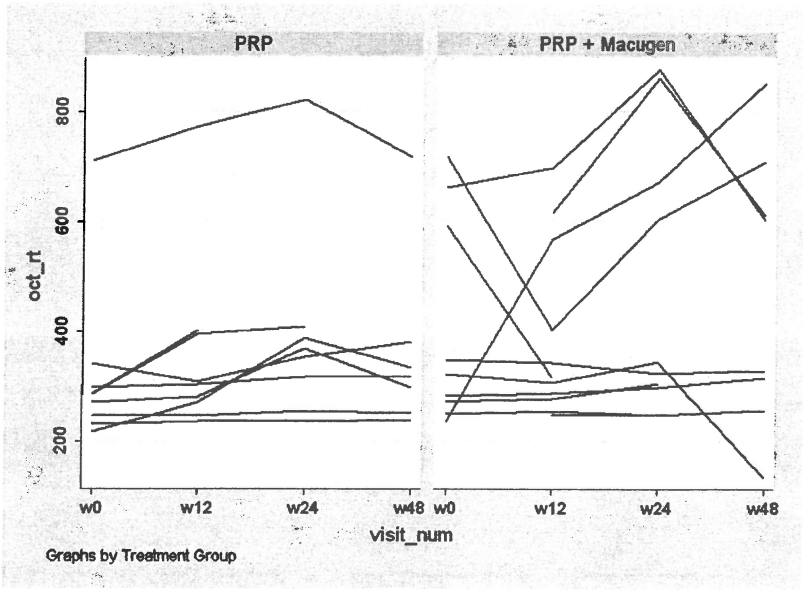

Figure 11-4. Central retinal thickness (µm) by patients according to treatment arms.

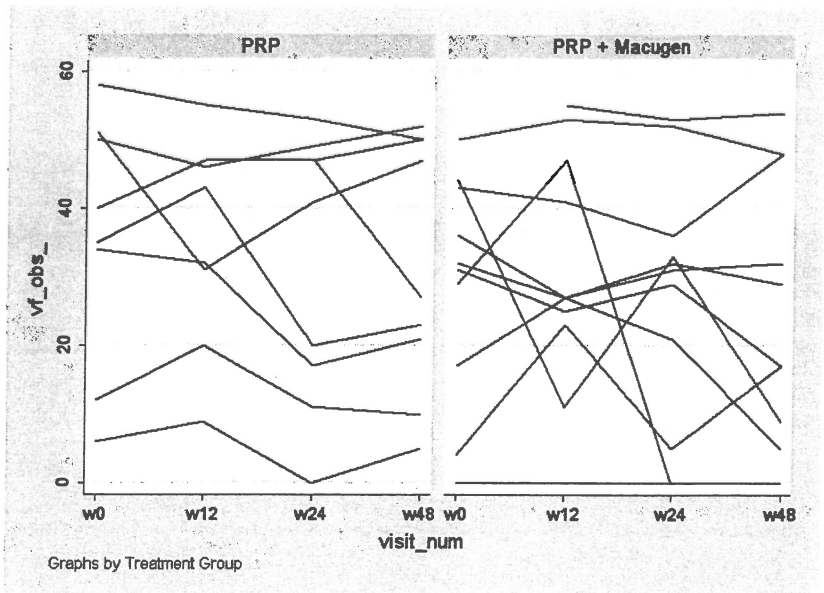

Figure 11-5. Visual Fields (#) by patients according to treatment arms.

Drug safety profile is described in Section **Error! Reference source not found.**.

11.2.2 Statistical / Analytical Issues

Due to the small number of cases (recruitment below 73.3% of the planned and drop-out rate of 9.1%), the distribution of data doesn't follow a normal distribution and therefore non-parametric tests were used (including the Mann-Whitney, Wilcoxon and Friedman tests). The χ^2 test was used for nominal and/or categorical variables. Data was presented using the medians and IQR.

11.2.2.1 Handling of Dropouts or Missing Data

The PP population was used without missing data imputation due to the small number of drop-outs, the small sample size and due to the similar results found between the ITT and the PP populations.

11.2.2.2 Interim Analyses and Data Monitoring

No interim analyses were performed.

Four on-site monitoring visits were performed on: 17 and 18 November 2010, 20 May 2011, 20 February 2012 and 30 November 2012 (close-out visit with 100% source data verification).

11.2.3 Efficacy conclusions

The combination therapy showed no efficacy differences when compared to the standard PRP treatment.

12 Safety evaluation

12.1 Extent of exposure

Table 12-1and Table 12-2.

Regarding the laser treatments:

For Group 1 (PRP) a total of 13 laser treatments were administered from week 0 to week 12, 9 laser treatments from week 12 to week 24 and 9 laser treatments from week 24 to week 48. For Group 2 (PRP+Macugen) a total of 11 laser treatments were administered 2 in the 2nd ring from week 0 to week 12, 8 in the 2nd ring from week 12 to week 24, and 1 in the 2nd ring and 5 in the 3rd ring from week 24 to week 48.

Regarding the study drug (Group 2):

All the patients received 4 or more injections out of 8 Table 12-2.

Fourteen (14) injections of Macugen were administered from week 12 to 24 and 13 additional injections from week 24 to 48 (

Table 12-1).

Table 12-1. Patients treatment exposure.

		No. of Injections	No. of laser treatments (n%)			
	No.	Group 2	Group 1		Group 2 PRP 2nd ring	PRP 3rd ring
Number of treatments week 12	0	0	0 (0.0)	8 (100.0)	2 (18.2)	0 (0.0)
	1	0	5 (62.5)	3 (37.5)		
	2	0	4 (100)	0 (0.0)		
	3	11	4 (100)	0 (0.0)		
Number of treatments from week 12 to week 24	0	1	2 (66.7)	1 (33.3)	8 (72.7)	0 (0.0)
	1	6	7 (41.2)	10 (58.8)		
	2	4	0 (0.0)	0 (0.0)		
	3	0	0 (0.0)	0 (0.0)		
Number of treatments from week 24 to 48	0	3	2 (33.3)	4 (66.7)	1 (9.1)	5 (100.0)
	1	5	6 (46.2)	7 (53.9)		
	2	1	1 (100.0)	0 (0.0)		
	3	2	0 (0.0)	0 (0.0)		

Table 12-2. List of patients study drug exposure (Group 2).

Subject number	Treatment group	Date of the first injection	Number of injections
001	1	-	
002	2	16/08/2010	5/8
003	1	-	
004	1	-	
006	2	10/11/2010	6/8
007	2	15/12/2010	8/8
008	1	-	
009	2	10/01/2011	4/8
010	1	-	
011	2	09/03/2011	8/8
012	1	-	
013	2	21/03/2011	4/8
014	1	-	
015	2	13/04/2011	5/8
016	1	-	
017	2	01/06/2011	5/8
018	1	-	
019	2	07/09/2011	5/8
020	1	-	
021	2	06/10/2011	6/8
022	2	-	
024	2	30/11/2011	4/8

Subjects' concomitant medication was constituted mainly by oral anti-diabetic drugs and insulin analogs, as well as antihypertensive agents (Table 12-3).

Table 12-3. Concomitant medication by patient.

Subject number	Treatment group	Concomitant Medication	INN	Therapeutic class	Visit number
001	1	Insulin NovoMix 30 Penfil	Insulin Aspart	Oral anti-diabetic drugs and insulin analogs	1
001	1	Co-Diovan 160/12.5	Valsartan + Hydrochlorotiazide	Antihypertensives	1
001	1	Nebules	Salbutamol	Drugs for obstructive airway diseases: asthma/COPD	1
001	1	Tromalyt 150 mg	Acetylsalicylic acid	Antithrombotics (thrombolytics. anticoagulants and antiplatelet drugs)	1
001	1	Coveram	Perindopril + Amlodipina	Antihypertensives	1
001	1	Insulin NovoMix 30 Penfil	Insulin Aspart	Oral anti-diabetic drugs and insulin analogs	1
002	2	Risidon 1000 mg	Metformin	Oral anti-diabetic drugs and insulin analogs	1
002	2	Doxi-OM	Calcium dobesilate	Peripheral vasodilators	1
002	2	Zarator	Atorvastatin	Lipid modifying agents	1
002	2	Aspirin	Acetylsalicylic acid	Antithrombotics (thrombolytics.	1

Table 12-3. Concomitant medication by patient.

Subject number	Treatment group	Concomitant Medication	INN	Therapeutic class	Visit number
				anticoagulants and antiplatelet drugs)	
002	2	Algimate	Clonixin	Analgesics and antipyretics	1
002	2	Enalapril	Enalapril	Antihypertensives	1
003	1	Risidon	Metformine	Oral anti-diabetic drugs and insulin analogs	1
003	1	Loftyl	Cloridrato de buflomedil	Antihypertensives	1
003	1	Diovan	Valsartan	Antihypertensives	1
003	1	Insulin (Humulin)	Insulin isophane + Human insulin	Oral anti-diabetic drugs and insulin analogs	1
004	1	Insulin (Humalog)	Insulin lispro	Oral anti-diabetic drugs and insulin analogs	1
004	1	Insulin (Lantus)	Insulin glargine	Oral anti-diabetic drugs and insulin analogs	1
004	1	Cartia	Acetylsalicylic acid	Antithrombotics (thrombolytics. anticoagulants and antiplatelet drugs)	1
004	1	Pravastatin	Pravastatin	Lipid modifying agents	1
004	1	Yag Laser	Yag Laser	Yag Laser	6
006	2	Insulin (Mixtard 30)	Human insulin	Oral anti-diabetic drugs and insulin analogs	1
006	2	Hytacand	Candesartan + Hydrochlorotiazide	Antihypertensives	1
006	2	Magnespasmil	Magnesium lactate	Mineral supplements	1
006	2	Insulin (Mixtard 30)	Human insulin	Oral anti-diabetic drugs and insulin analogs	1
006	2	Hytacand	Candesartan + Hydrochlorotiazide	Antihypertensives	1
006	2	Magnespasmil	Magnesium lactate	Mineral supplements	1
007	2	Diamicron	Glicazide	Oral anti-diabetic drugs and insulin analogs	1
007	2	Janumet	Sitagliptin + Metformin	Oral anti-diabetic drugs and insulin analogs	1
007	2	Floxedol	Ofloxacin	Antibacterials	7
007	2	Oftacilox	Ciprofloxacin	Antibacterials	7
007	2	Minocin	Minocycline	Antibacterials	8
007	2	Predniftalmina	Chloramphenicol + Prednisolone	Antibacterials	6
007	2	Terricil	oxytetracycline	Antibacterials	8
008	1	Risidon	Metformine	Oral anti-diabetic drugs and insulin analogs	
008	1	Pars Plana Vitrectomy	Pars Plana Vitrectomy	Surgery	8
009	2	Daonil	Glibenclamide	Oral anti-diabetic drugs and insulin analogs	1
009	2	Diamicron	Glicazide	Oral anti-diabetic drugs and insulin analogs	1
009	2	Risidon	Metformine	Oral anti-diabetic drugs and insulin analogs	10
010	1	Insulin (Humalog)	Insulin lispro	Oral anti-diabetic drugs and insulin analogs	1
010	1	Insulin (Lantus)	Insulin glargine	Oral anti-diabetic drugs	1

Table 12-3. Concomitant medication by patient.

Subject number	Treatment group	Concomitant Medication	INN	Therapeutic class	Visit number
				and insulin analogs	
011	2	Insulin	Insulin	Oral anti-diabetic drugs and insulin analogs	1
011	2	Ramipril	Rampril	Antihypertensives	1
011	2	Sinvastatin	Sinvastatin	Oral anti-diabetic drugs and insulin analogs	1
011	2	Trentral	Pentoxifillyne	Peripheral vasodilators	1
011	2	Cartia	Acetylsalicylic acid	Antithrombotics (thrombolytics. anticoagulants and antiplatelet drugs)	1
012	1	Velmetia	Sitagliptin + Metformin	Oral anti-diabetic drugs and insulin analogs	1
012	1	Focal Laser	Focal laser	Focal laser	7
012	1	Metformin	Metformin	Oral anti-diabetic drugs and insulin analogs	1
013	2	Coversyl	Perindopril	Antihypertensives	1
013	2	Crestor	Rosuvastatin	Lipid modifying agents	1
013	2	Mixtard	Human insulin	Oral anti-diabetic drugs and insulin analogs	1
014	1	Metformin	Metformin	Oral anti-diabetic drugs and insulin analogs	1
014	1	Losartan	Losartan	Antihypertensives	1
014	1	Tacirel	Trimetazidine	Anti-ischemic agent	1
014	1	Rivotril	Clonzepam	Anticonvulsant	1
014	1	Amlodipina	Amlodipine	Antihypertensives	1
014	1	Ramipril	Ramapril	Antihypertensives	1
014	1	Rytmonorm	Propafenone	Antiarrhythmic agents	1
014	1	Eucreas	Vildagliptin + Metformin	Oral anti-diabetic drugs and insulin analogs	7
014	1	PritorPlus	Telmisartan + Hydrochlorothiazide	Antihypertensives	7
014	1	Novomix Penfill	Insulin aspart	Oral anti-diabetic drugs and insulin analogs	7
014	1	Concor	Bisoprolol	Antihypertensives	7
014	1	Topiramato	Topiramate	Anticonvulsant	7
014	1	Mixtard 30 Penfill	Human insulin	Oral anti-diabetic drugs and insulin analogs	7
015	2	Insulin	Insulin	Oral anti-diabetic drugs and insulin analogs	1
016	1	Insulin	Insulin	Oral anti-diabetic drugs and insulin analogs	1
016	1	Triatec	Ramapril	Antihypertensives	1
016	1	Iron suplements	Iron supplements	Antianemic agents	
016	1	Trentral	Pentoxifillyne	Peripheral vasodilators	5
016	1	Trifiusal	Triflusal	Antithrombotics (thrombolytics. anticoagulants and antiplatelet drugs)	5
016	1	Omeprazol	Omeprazol	Drugs for acid related disorders: drugs for peptic ulcer and GERD/GORD	5
016	1	Centrum	Multivitamins	Vitamins	5
016	1	Pacemaker implementation	Pacemaker implementation	Pacemaker implementation	6

Table 12-3. Concomitant medication by patient.

Subject number	Treatment group	Concomitant Medication	INN	Therapeutic class	Visit number
016	1	Retinal Surgery	Retinal Surgery	Retinal Surgery	6
017	2	Eucreas	Telmisartan + Hydrochlorothiazide	Antihypertensives	1
017	2	Losartan 50 mg	Losartan	Antihypertensives	1
017	2	Sinvastatin	Sinvastatin	Oral anti-diabetic drugs and insulin analogs	1
017	2	Timoptol	Timolol	Antiglaucoma preparation and miotics	4
017	2	Cosopt	Dorzolamida + Timolol	Antiglaucoma preparation and miotics	1
017	2	Losartan 100 mg	Losartan	Antihypertensives	1
017	2	Atorvastatin	Atorvastatin	Lipid modifying agents	1
017	2	Combigan	Brimonidine + Timolol	Antiglaucoma preparation and miotics	1
018	1	Insulin	Insulin	Oral anti-diabetic drugs and insulin analogs	1
019	2	Timolol	Timolol	Antiglaucoma preparation and miotics	1
019	2	Glucovance	Glibenclamide + Metformin	Oral anti-diabetic drugs and insulin analogs	1
019	2	Gliclazida	Gliclazide	Oral anti-diabetic drugs and insulin analogs	1
019	2	Humolog Mix	Insulin lispro	Oral anti-diabetic drugs and insulin analogs	1
019	2	Tens	Lacidipina	Antihypertensives	1
019	2	Co-Diovan	Valsartan + Hydrochlorotiazide	Antihypertensives	1
019	2	Lisinopril + Hidroclorotiazida	Lisinopril + Hydrochlorotiazide	Antihypertensives	1
019	2	Ganfort	Bimatoprost + Timolol	Antiglaucoma preparation and miotics	1
019	2	Lumigan	Bimatropost	Antiglaucoma preparation and miotics	6
019	2	Combigan	Timolol	Antiglaucoma preparation and miotics	6
020	1	Risidon	Metformin	Oral anti-diabetic drugs and insulin analogs	1
020	1	Pravastatin	Pravastatin	Lipid modifying agents	1
020	1	Trental	Pentoxifillyne	Peripheral vasodilators	1
020	1	Brufen	Ibuprofen	Non-steroidal anti-infalmmatory drugs	1
020	1	Herbesser	Diltiazem	Antiarrhytmic agents	1
020	1	Penfil			1
020	1	Aspirin	Acetylsalicylic acid	Antithrombotics (thrombolytics. anticoagulants and antiplatelet drugs)	1
020	1	Clopidogrel	Clopidogrel	Antithrombotics (thrombolytics. anticoagulants and antiplatelet drugs)	7
020	1	Pantoprazol	Pantoprazol	Drugs for acid related disorders: drugs for peptic ulcer and GERD/GORD	7
020	1	Alprazolam	Alprazolam	Anxiolytics	7

Table 12-3. Concomitant medication by patient.

Subject number	Treatment group	Concomitant Medication	INN	Therapeutic class	Visit number
020	1	Cardiac catheterization	Cardiac catheterization	Cardiac catheterization	7
021	2	Janumet	Sitagliptin + Metformin	Oral anti-diabetic drugs and insulin analogs	1
021	2	Supralip	Fenofibrate	Lipid modifying agents	1
021	2	Biloban	Ginkgo Biloba	Peripheral vasodilators	1
021	2	Vastarel	Trimetazidine	Antianginals	1
021	2	Diamicron	Glicazide	Oral anti-diabetic drugs and insulin analogs	1
022	2	Diamicron	Glicazide	Oral anti-diabetic drugs and insulin analogs	1
022	2	Zomarist	Vildagliptin + Metformin	Oral anti-diabetic drugs and insulin analogs	1
022	2	Metformin	Metformin	Oral anti-diabetic drugs and insulin analogs	1
022	2	Ramipril	Rampril	Antihypertensives	1
022	2	Visacor	Rosuvastatin	Lipid modifying agents	1
022	2	Vessel	Sulodexide	Antithrombotics (thrombolytics. anticoagulants and antiplatelet drugs)	1
022	2	Sedoxil	Mexazolam	Anxiolytics	1
024	2	Risidon	Metformin	Oral anti-diabetic drugs and insulin analogs	1
024	2	Diamicron	Glicazide	Oral anti-diabetic drugs and insulin analogs	1
024	2	Glucobay	Arcabose	Oral anti-diabetic drugs and insulin analogs	1
024	2	Amlodipina	Amlodipine	Antihypertensives	1
024	2	Losartan	Losartan	Antihypertensives	1
024	2	Triatec	Ramipril	Antihypertensives	1

12.2 Adverse events (AEs)

12.2.1 Brief summary of adverse events

The adverse events observed in the study were as expected for this population and this class of drug. They were mostly mild and transient AE that did not seem to be dose-related and gave no indication of target organ toxicity. However, one patient from Group 1 (PRP) needed vitrectomy due the occurrence of reubeosis and one subject from Group 1 (PRP) presented one SAE, which was angina pectoris.

12.2.2 Display of adverse events

AEs are summarized and displayed in Table 12-4 and **Error! Reference source not found.**.

Table 12-4. Adverse events overall and most frequent events.

AEs with onset after the start of treatment	Group 1 n (%)	Group 2 n (%)	Total n (%)
Patients with AEs	10 (43.48)	13 (56.52)	23 (100)
Preferred term			
(Bilateral) Blepharitis	0	1 (4.35)	1 (4.35)
(Bilateral) Conjunctivitis	0	1 (4.35)	1 (4.35)
Bradycardia	1 (4.35)	0	1 (4.35)
Cataract conditions	0	1 (4.35)	1 (4.35)
Corneal ulceration	1 (4.35)	0	1 (4.35)
Depression	1 (4.35)	0	1 (4.35)
Diabetic macular oedema	1 (4.35)	0	1 (4.35)
Dyspnea exertional	0	1 (4.35)	1 (4.35)
Vitreous hemorrhage	3 (13.04)	5 (21.74)	8 (34.78)
Vitreomacular traction syndrome	1 (4.35)	0	1 (4.35)
Ocular hypertension	0	3 (13.04)	3 (13.04)
Subhyaloid hemorrhage	1 (4.35)	0	1 (4.35)
Rubeosis iridis	1 (4.35)	1 (4.35)	2 (8.70)

Table 12-5. Adverse events overall and frequently affected system organ classes.

	Group 1 n (%)	Group 2 n (%)	Total n (%)
Patients with AE(s)	10 (43.5)	13 (56.5)	23 (100)
System organ class	---	---	---
Cardiac disorders	1 (4.35)	0 (0)	1 (4.35)
Eye disorders	8 (34.8)	13 (56.5)	21 (91.3)
Psychiatric disorders	1 (4.35)	0 (0)	1 (4.35)

12.2.3 Analysis of adverse events

Results show a total of 23 AEs identified during the study, 10 in Group 1 and 13 in Group 2. Vitreous hemorrhage was reported 8 times (3 in Group 1 and 5 in Group 2). Ocular hypertension was reported 3 times, all of them in Group 2. Rubeosis iridis was reported 2 times (one in each group). Adverse events in higher proportion are included in the eye disorders group (91.3%). Of those AEs, 56.5% are in Group 2. Other non-eye disorders are described, namely 1 cardiac disorder and 1 psychiatric disorder (4.35%), in Group 1.

12.2.4 Listing of adverse events by patient

The listing of adverse events is found in Table 12-6.

Table 12-6. Listing of adverse events by patient.

Subject number	Treatment group	AE	Severity	SAE	Visit number
001	1	None	-	None	-
002	2	Vitreous hemorrhage	1	None	4
002	2	Vitreous hemorrhage	2	None	5
002	2	Vitreous hemorrhage	2	None	6
002	2	Rubeosis Iridis	1	None	6
003	1	None	-	None	-
004	1	Subhyaloid hemorrhage	2	None	6
005	2	None	-	None	-
006	2	None	-	None	-
007	2	Vitreous hemorrhage	2	None	6
007	2	(Bilateral) Conjuntivitis	1	None	7
007	2	(Bilateral) Blepharitis	1	None	8
007	2	Cataract	2	None	8
008	1	Vitreous hemorrhage	2	None	4
008	1	Vitreous hemorrhage	2	None	7
008	1	Rubeosis Iridis	1	None	1
009	2	None	-	None	-
010	1	Corneal ulceration	1	None	6
011	2	None	-	None	-
012	1	Diabetic macular oedema	2	None	4
013	2	None	-	None	-
014	1	None	-	None	-
015	2	None	-	None	-
016	1	Vitreous hemorrhage	1	None	3
016	1	Bradycardia	3	None	6
016	1	Vitreomacular traction syndrome	3	None	6
017	2	Dyspnea exertional	1	None	3
017	2	Ocular hypertension	1	None	4
017	2	Ocular hypertension	1	None	5
017	2	Ocular hypertension	1	None	6
018	1	None	-	None	-
019	2	None	-	None	-
020	1	Depression	2	None	4
020	1	SAE		Angina Pectoris	7

Table 12-6. Listing of adverse events by patient.

Subject number	Treatment group	AE	Severity	SAE	Visit number
021	2	Vitreous hemorrhage	1	None	5
022	2	None	-	None	-
023	2	None	-	None	-
024	2	None	-	None	-

12.3 Deaths. other serious adverse events. and other significant adverse events

SAE and clinically significant AEs are summarized in Table 12-7.

Table 12-7. Deaths, other serious or clinically significant adverse events or related discontinuations.

	Group 1 n (%)	Group 2 n (%)
Patients with AE(s)	10 (43.48)	13 (56.52)
Serious or other significant events		
Death	0	0
SAE(s)	Angina Pectoris	0
Clinically significant AE(s)	10	13
Discontinued due to SAE(s)	0	0
Discontinued due to clin. sign. AE(s)	0	0

12.3.1 Listing of deaths, other serious adverse events, and other significant adverse events

Patients who experienced SAEs and AEs are listed in Table 12-6.

12.3.2 Narratives of deaths, other serious adverse events, and certain other significant adverse events

Only one patient from group 1 (PRP) experienced a SAE (Angina Pectoris) that was not related with the study procedure.

12.3.3 Analysis and discussion of deaths, other serious adverse events, and other significant adverse events

No safety concerns (including SAEs) were identified.

12.4 Clinical laboratory evaluation

12.4.1 Listing of individual laboratory measurements by patient and each abnormal laboratory value

A listing of individual laboratory measurements by patient is found in Table 12-8 and Figure 12-1.

Table 12-8. Individual laboratory measurements by patient – HbA1C.

		HbA1C (%)	
Subject number	**Treatment group**	**Screening**	**Week 48**
001	1	7.0 (Normal)	**8.8 (Abnormal)**
002	2	**8.0 (Abnormal)**	6.8 (Normal)
003	1	7.5 (Normal)	7.2 (Normal)
004	1	**8.8 (Abnormal)**	7.5 (Normal)
005	2	-	-
006	2	**7.6 (Abnormal)**	**8.0 (Abnormal)**
007	2	**8.5 (Abnormal)**	**9.6 (Abnormal)**
008	1	7.1 (Normal)	7.4 (Normal)
009	2	**8.6 (Abnormal)**	7.2 (Normal)
010	1	**9.1 (Abnormal)**	**8.8 (Abnormal)**
011	2	**11.1 (Abnormal)**	**9.2 (Abnormal)**
012	1	**11.0 (Abnormal)**	6.7 (Normal)
013	2	**7.9 (Abnormal)**	6.4 (Normal)
014	1	6.1 (Normal)	6.3 (Normal)
015	2	**7.8 (Abnormal)**	**7.8 (Abnormal)**
016	1	7.4 (Normal)	**7.9 (Abnormal)**
017	2	7.0 (Normal)	**7.6 (Abnormal)**
018	1	6.5 (Normal)	6.7 (Normal)
019	2	7.4 (Normal)	7.7 (Abnormal)
020	1	**7.6 (Abnormal)**	8.4 (Abnormal)
021	2	**5.8 (Abnormal)**	**5.8 (Abnormal)**
022	2	**9.6 (Abnormal)**	**10.2 (Abnormal)**
023	2	-	-
024	2	**5.7 (Abnormal)**	**5.0 (Abnormal)**

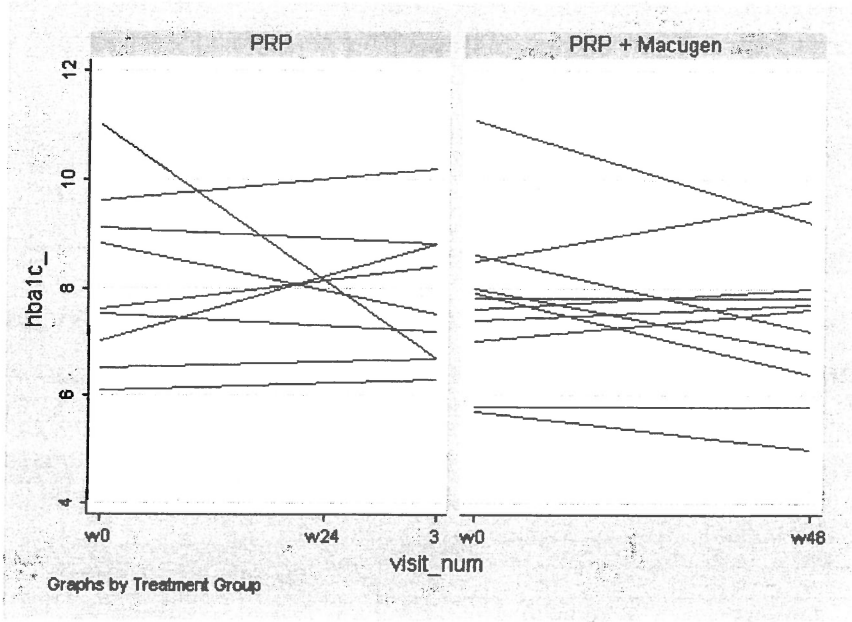

Figure 12-1. Individual laboratory measurements by patient – HbA1C.

12.4.2 Evaluation of Each Laboratory Parameter

HbA1C values by groups are shown in Table 12-9. Increased or decreased HbA1C values are summarized in Table 12-10.

There are more patient with high levels of HbA1C both on screening and week 48. This trend is observed for both groups. A minimal percentage of patients showed low levels of HbA1C.

There were no significant differences regarding HbA1c between screening visit and final visit (Group 1, 7.6% to 7.5% and Group 2, 7.8% to 7.6%).

Table 12-9. Summary of the HbA1C.

		Median (IQR)	
		Group 1	**Group 2**
HbA1C	screening	7.6 (7.0-9.1)	7.8 (7.0-8.5)
	W48	7.5 (6.7-8.8)	7.6 (6.4-8.0)
Global difference between first and final visit	p-value*	0.953	0.264

*Test wilcoxon used

Table 12-10. Laboratory values over time – Abnormal HbA1C.

		Notable abnormality	Median (IQR) Group 1	Median (IQR) Group 2
HbA1C	screening	High (>7.5%)	5 (41.67)	7 (58.33)
		Low (<6.5%)	1 (33.33)	2 (67.67)
	W48	High (>7.5%)	4 (40.0)	6 (60.0)
		Low (<6.5%)	1 (25.0)	3 (75.0)
Global difference between first and final visit	p-value*		0.5367	0.6547

*Test wilcoxon used

12.5 Vital signs, physical findings, and other observations related to safety

Systolic and diastolic blood pressure by groups are summarized in Table 12-11and presented in Figure 12-2 and Figure 12-3.

Table 12-11. Summary of the Systolic and Diastolic blood pressures.

		Group 1	Group 2
Notable abnormality			
BP systolic	screening	139 (136-147)	139 (135-146)
	W12	140 (120-149)	147 (128-154)
	W48	144 (135-156)	148 (126-162)
P-value*		0.317	N/A
BP diastolic	screening	80 (78-81)	81 (79-82)
	W12	77 (68-80)	77 (74-82)
	W48	76 (70-77)	80 (68-85)
P-value*		0.0956	N/A

*Friedman test used

Other safety assessments includes: ophthalmic examinations (slit lamp exam with biomicroscopy. fundus exam and intra ocular pressure) and visual acuity assessments that are presented in Table 12-12, **Error! Reference source not found.** and Table 12-14 (graphical representations are shown in Figure 12-4, Figure 12-5 for Intraocular pressure and for Visual Fields).

Regarding the slit lamp examination, no significant anomalies located at the anterior chamber, cornea and lids were found. Significnat anomalities were found in the iris (rubeosis), the conjunctiva (hyperemia edema) and the lens (cataract + PSC) (**Error! Reference source not found.**).

Ophthalmoscopy (Table 12-14) reveals significant anomalies in the retina, namely, NVE, NVE+DRP, NVE and PDR for both treatment groups. Additionally, Group 1 presented DME and laser scars. On the other hand, it was identified in the macula, CSME, DME, DR and ME for both groups, and laser scars only for Group 1 (PRP).

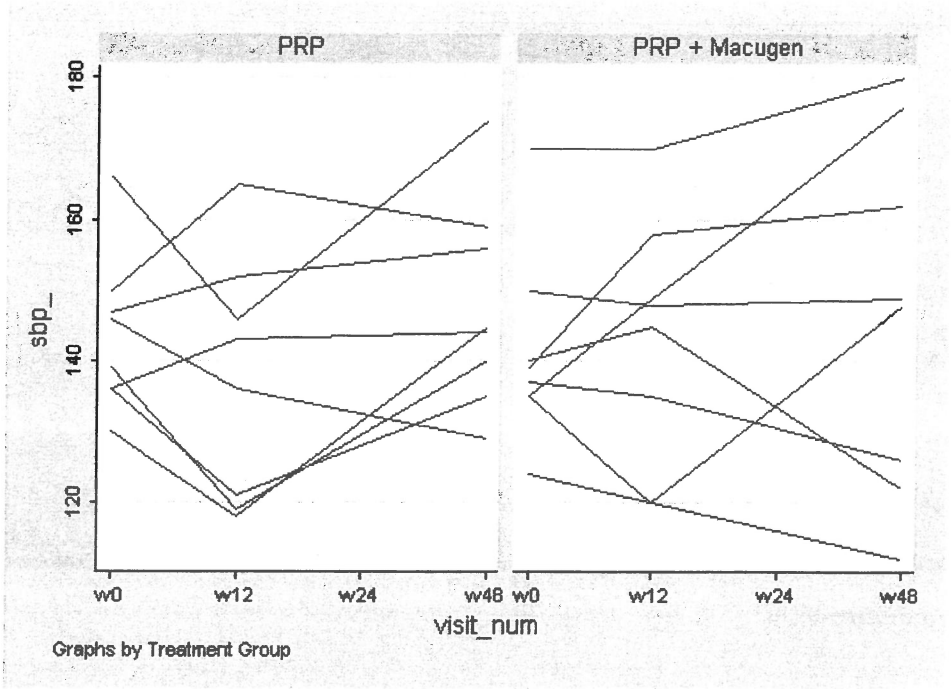

Figure 12-2. Systolic blood pressure according to arm of treatment, by patient.

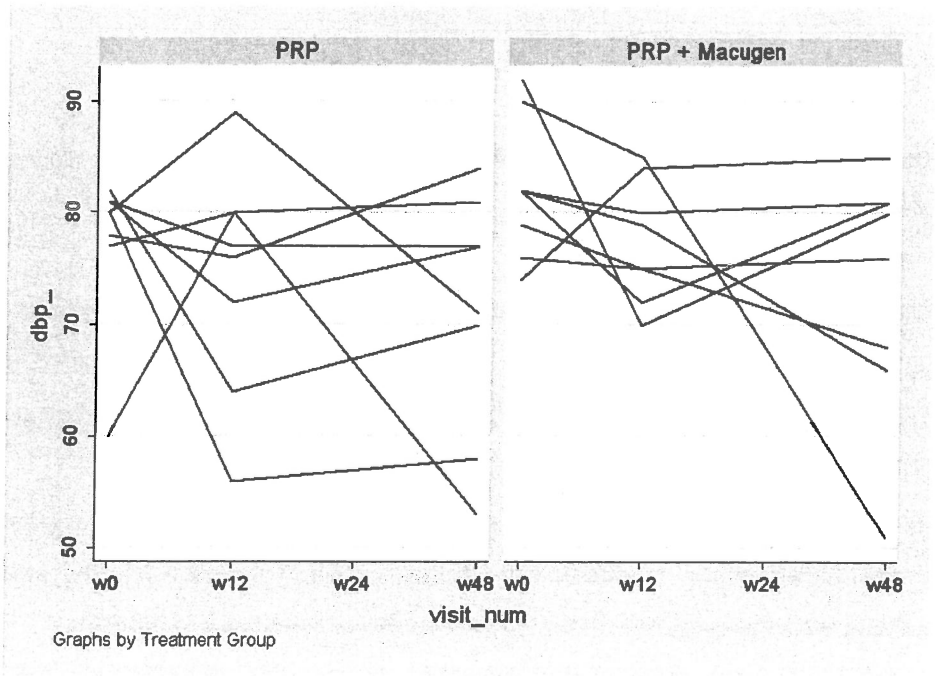

Figure 12-3. Diastolic blood pressure according to arm of treatment, by patient.

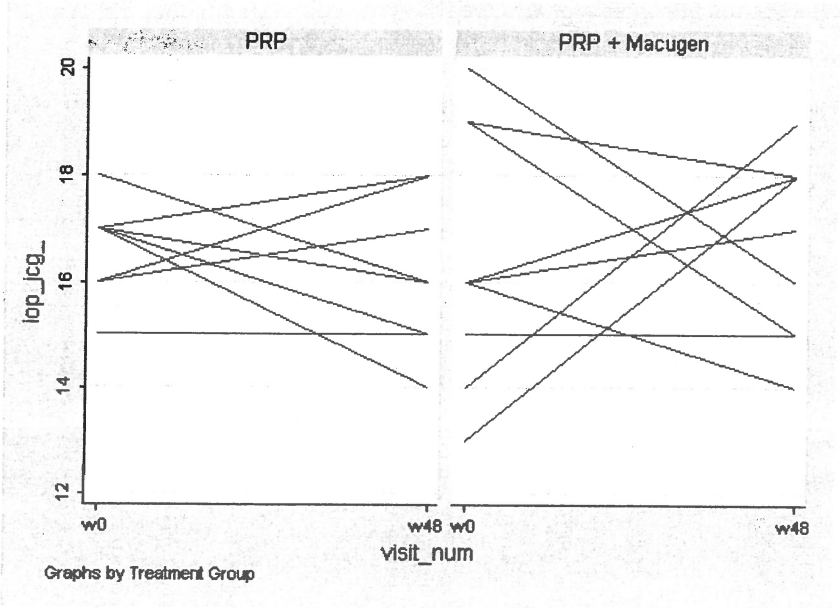

Figure 12-4. Intraocular pressure according to arm of treatment, by patient.

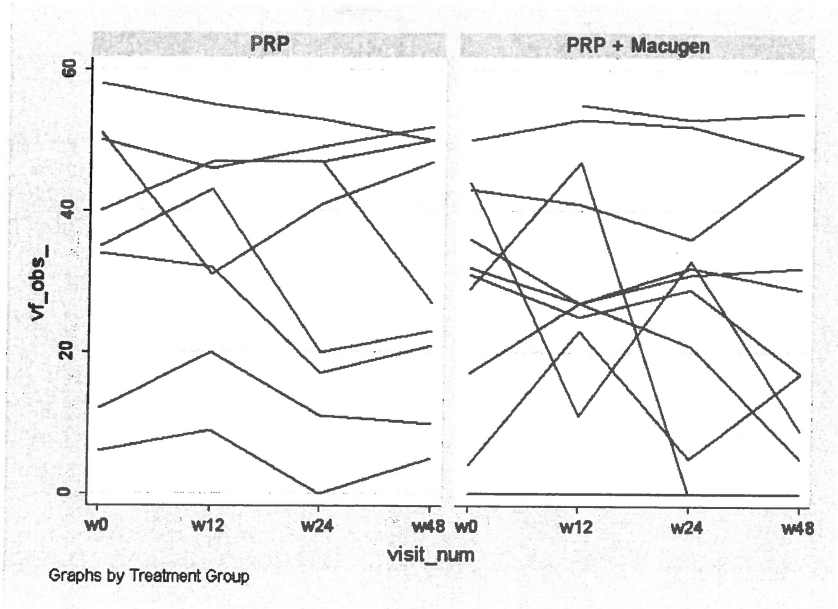

Figure 12-5. Visual fields according to arm of treatment, by patient.

Table 12-12. Summary of the Intraocular pressure and BCVA.

		Median (IQR)	
		Group 1	Group 2
Intraocular pressure	screening	17 (16-17)	16 (14-19)
	W48	16 (15-17)	18 (15-18)
BCVA	screening	80 (77-84)	77 (60-78)
	W12	82 (75-82)	76 (51-80)
	W24	82 (58-82)	78 (50-79)
	W48	79 (59-83)	75 (43-81)

Table 12-13. Slit lamp abnormalities.

		Group 1				Group 2			
		Norm.	Insig. Abn.	Sig. Abn.		Norm.	Insig. Abn.	Sig. Abn.	
Notable abnormality									
Slit lamp exam: Anterior chamber	screening	9	0	0		11	0	0	
	W12	9	0	0		11	0	0	
	W24	9	0	0		11	0	0	
	W48	9	0	0		11	0	0	
Slit lamp exam: cornea	screening	8	1	0		11	0	0	
	W12	9	0	0		11	0	0	
	W24	9	0	0		11	0	0	
	W48	8	1	0		11	0	0	
Slit lamp exam: conjunctiva	screening	9	0	0		11	0	0	
	W12	9	0	0		11	0	0	
	W24	8	0	1	Hyperemia Edema	11	0	0	
	W48	9	0	0		11	0	0	
Slit lamp exam: Iris	screening	9	0	0		11	0	0	
	W12	6	1	2	Rubeosis	11	0	0	
	W24	6	1	2	Rubeosis	11	0	0	
	W48	6	1	2	Rubeosis	11	0	0	
Slit lamp exam: lens	screening	7	2	0		11	0	0	
	W12	7	2	0		11	0	0	
	W24	8	1	0		11	0	0	
	W48	9	0	0		10	0	1	Cataract + PSC*
Slit lamp exam: lids	screening	9	0	0		11	0	0	
	W12	9	0	0		11	0	0	
	W24	9	0	0		11	0	0	
	W48	9	0	0		11	0	0	

*Pseudoexfoliation of lens capsule
Norm. – Normal
Insig.Absn. – Insignificant Anomaly
Sig. Abn. – Significant Anmolay

Table 12-14. Ophthalmoscopy abnormalities.

		Group 1				Group 2			
		Norm.	Insig. Abn.	Sig. Abn.		Norm.	Insig. Abn.	Sig. Abn.	
Notable abnormality									
Ophtalmoscopy: vitreous	screening	9	0	0		11	0	1	HV*[1]
	W12	9	0	0		11	0	0	
	W24	9	0	0		11	0	0	
	W48	9	0	0		11	0	0	
Ophtalmoscopy: retina	screening	1	0	1	DR*[2]	0	0	1	DR
				5	NVE			4	NVE
				1	NVE+RD			1	NVE+NVD +Vascular
				1	PDR*[3]			5	PDR
	W12	1	0	1	DME	3	0	6	NVE
				1	Laser scars			1	NVE+NVD +Vascular
				3	NVE			1	PDR
				1	NVE+DRP				
				1	NVE + Laser Scars				
				1	PDR				
	W24	1	0	1	DME*[4]	1	1	8	NVE
				1	DR+NVD			1	PDR
				1	Laser scars + NVE				
				4	NVE				
				1	PDR				
	W48	1	0	1	DME	2	1	8	NVE
				1	Laser Scars				
				5	NVE				
				1	PDR				
Ophtalmoscopy: macula	screening	4	1	1	DME + Laser scars	3	1	4	CSME*[6]
				2	DR			3	DME
				1	ME*[5]				
	W12	4	0	2	DME	4	0	3	CSME
				1	Laser Scars			4	DME
				1	DR				
				1	ME				
	W24	4	0	1	CSME	3	1	2	CSME
				2	DME			5	DME
				1	Laser Scars + DR				
				1	ME				
	W48	4	0	1	CSME	3	1	1	CSME
				4	DME			4	DME
								1	DR
								1	ME
Ophtalmos	screening	9	0	0		11	0	0	

Table 12-14. Ophthalmoscopy abnormalities.

		Group 1			Group 2		
		Norm.	Insig. Abn.	Sig. Abn.	Norm.	Insig. Abn.	Sig. Abn.
	W12	9	0	0	11	0	0
	W24	9	0	0	11	0	0
	W48	9	0	0	11	0	0
Ophtalmoscopy: optic nerve	screening	9	0	0	11	0	0
	W12	9	0	0	11	0	0
	W24	9	0	0	11	0	0
	W48		0	0	11	0	0
Ophtalmoscopy: optic nerve pallor	screening	9	0	0	11	0	0
	W12	9	0	0	11	0	0
	W24	9	0	0	11	0	0
	W48	9	0	0	11	0	0

*[1] – Hemovitreous; *[2] Diabetic Retinopathy; *[3] Proliferative Diabetic Retinopathy; *[4] Diabetic Macular Edema; *[5] Macular Edema *[6] Clinically significant Macular Edema
Norm. – Normal
Insig.Absn. – Insignificant Anomaly
Sig. Abn. – Significant Anmolay

12.6 Safety conclusions

No safety findings were found in this study.

13 Discussion and overall conclusions

This is a two-arm, parallel, comparative, exploratory, randomized, prospective study to compare the standard treatment for high-risk proliferative diabetic retinopathy (HR-PDR) (Group 1: Pan Retinal Photocoagulation – PRP laser treatment) with a combined therapy of anti-VEGF (Group 2: intravitreal injection of Macugen combined with progressive PRP laser treatment).

Due to the great challenge to enroll patients that met all the study inclusion and exclusion criteria (namely, patients with HR-PDR without previous PRP treatment) only 22 patients were enrolled despite of the efforts made by the research team within the time period allocated for extended enrollment. The target number was 30 patients.

The regression of the neovascularization area from baseline to Week-48 was slightly higher in Group 2 (-0.4DA) when compared to Group 1 (-0.33DA). Nevertheless, the difference found was not statistically significant (p=0.870). Therefore, the primary objective could not be verified, maybe due to the reduced sample size.

The most clinically relevant result of the study was the worsening of the visual fields in the eyes treated with the standard PRP treatment (Group 1) between baseline and Week-48 (40 versus 27 observed dots, p=0.0319). Whereas in the group with the combined treatment the difference between baseline and Week-48 was lower (31.5 versus 23 observed dots, p=0.5465). These results suggest that the visual fields deterioration is related to the retina area that was destroyed by the standard PRP treatment, as in the group of the combined treatment, 6 patients (54.5%) did not need PRP laser in the inner ring, saving the posterior retina that plays such a relevant role for visual function. This combined treatment approach can eventually reduce some side effects related with PRP laser for HR-PDR, like visual field constriction, driving ability and night vision. (Pahor D et. al; 1998) (Buckley SA et. al; 1992) (Aiello LM et. al; 2003).

The central macular thickness (CMT) showed a significant increase between baseline and Week-48 for Group 1 (286 µm versus 326 µm, p=0.0455). Whereas in Group 2, the CMT was constant between baseline and Week-48 (323 µm versus 322 µm, p=0.0455). This protective effect for macular edema can be attributed to the intravitreal injection of Macugen that is an anti-VEGF with recognized action in the treatment of macular edema and/or due to the lower number of spots of the laser treatment in Group 2 (2313 versus 1400 at baseline, p=0.030).

Recurrence of the neovascularization occurred in 30% of the eyes in Group 1, whereas in Group 2 recurrence of neovascularization occurred in 70% of the eyes. Although the difference was not statistically significant (p=0.178). The higher neovascularization recurrence rate in Group 2, may be explained by the study design and/or the transitory effect of the combined treatment with anti-VEGF and PRP laser treatment, but this did not show any effect in the neovascularization area at the final visit.

The number of clinically relevant adverse events for this pathology and treatments (vitreous hemorrhage, vitreomacular traction, subhyaloid hemorrhage and rubeosis iridis) were similar in the 2 groups (6 in each group). Nevertheless, it should be highlighted that vitreous hemorrhage occurred in 3 eyes of Group 1 versus 5 eyes in Group 2. It is noteworthy that, 2 of the 3 patients with these complications of Group 1 needed to be submitted to vitrectomy to

treat the adverse event, whereas none of the patients of Group 2 need this type of intervention. This result suggests that the hemorrhages observed in Group 2 were less dense and also transitory. The anti-VEGF may have influenced positively this result.

There was only one serious adverse event and it was a cardiovascular disease (angina pectoris) that developed during the study in Group 1. The concern in terms of safety that persists in using anti-VEGF treatment in eyes with PDR could not be confirmed in this study.

In summary:
This was an exploratory study with a limited number of patients. The regression of the area of neovascularization in the group treated with Macugen associated with progressive PRP laser was not significantly better than the standard treatment with PRP laser in the treatment of eyes with HR-PDR.

Nevertheless, the eyes that received the combined treatment had less occurrence of central macular thickness increase.
As expected, visual fields remained significantly more preserved in Group 2, and this is a plus for this option of treatment, because of the associated problems in quality of life, night vision and ability to drive, reported with conventional full PRP laser.

Regarding safety, we had two cases needing vitrectomy in Group 1 versus none in Group 2, indicating that serious complications may be more frequent with conventional PRP treatment.
Considering all these factors, this study suggests that VEGF combined treatment may offer a better alternative than PRP alone to treat HR-PDR eyes, but larger prospective studies are needed to confirm this treatment option.

14 Tables, figures and graphs referred to but not included in the text

Tables, figures and graphs are included in the respective sections of this report.

15 Reference list

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16 Appendices

16.1 Study information

16.1.1 Protocol and protocol amendments

16.1.2 List of IECs or IRBs (plus the name of the committee chair if required by the regulatory authority) - representative written information for patient and sample consent forms

16.1.3 Signatures of principal or coordinating investigator(s) or sponsor's responsible medical officer, depending on the regulatory authority's requirement

16.2 Patient data listings