Graefe's Arch Clin Exp Ophthalmol
DOI 10.1007/s00417-006-0318-2

CLINICAL INVESTIGATION

Effect of calcium dobesilate on progression of early diabetic retinopathy: a randomised double-blind study

Maria L. Ribeiro
Andras I. Seres
Angela M. Carneiro
Michael Stur
Alain Zourdani
Patricia Caillon
José G. Cunha-Vaz
on behalf of the DX-Retinopathy Study Group

Received: 12 September 2005
Revised: 21 February 2006
Accepted: 23 February 2006

The study was supported by a grant from OM PHARMA, Meyrin/Geneva, Switzerland. There are no financial interest and no conflict of interest for M.L. Ribeiro, the authors and the members of the study group. P. Caillon is the Clinical Project Manager of the study at OM PHARMA.
The authors have full control of all primary data and agree to allow *Graefe's Archive for Clinical and Experimental Ophthalmology* to review their data if requested.

M. L. Ribeiro (✉) · J. G. Cunha-Vaz
AIBILI, Clinical Trial Centre,
Azinhaga de Santa Comba,
3000-548 Coimbra, Portugal
e-mail: lr@aibili.pt
Tel.: +351-239-480124
Fax: +351-239-483593

A. I. Seres
Department of Ophthalmology,
Semmelweis University,
1083 Budapest, Hungary

A. M. Carneiro
Department of Ophthalmology,
Hospital de São João,
4200 Porto, Portugal

M. Stur
Department of Ophthalmology,
Medical University of Vienna,
1090 Vienna, Austria

A. Zourdani
Department of Ophthalmology,
University-Paris XII-Creteil,
94010 Creteil, France

P. Caillon
OM PHARMA,
Rue du Bois-du-Lan 22,
1217 Meyrin/Geneva, Switzerland

Abstract *Background:* The study was carried out to confirm the effect of calcium dobesilate (CaD) compared to placebo (PLA) on the blood-retinal barrier (BRB) permeability in early diabetic retinopathy (DR). *Methods:* Adults with type II diabetes and early diabetic retinopathy (below level 47 of ETDRS grading and PVPR between 20 and 50×10^{-6}/min, plasma-free fluorescein) were included in this double-blind placebo-controlled study. Treatment was 2 g daily for 24 months. The primary parameter, posterior vitreous penetration ratio (PVPR), was measured every 6 months by fluorophotometry. Secondary parameters were fundus photography, fluorescein angiography and safety assessments. Metabolic control was performed every 3 months. *Results:* A total of 194 patients started the treatment (98 CaD, 96 PLA) and 137 completed the 24-month study (69 CaD, 68 PLA). Both treatment groups were comparable at baseline, with ETDRS level 10 in about 59% of patients. Mean PVPR change from baseline after 24 months was significantly (P=0.002) lower in the CaD group [−3.87 (SD 12.03)] than in the PLA group [+2.03 (SD 12.86)], corresponding to a 13.2% decrease in the CaD group and a 7.3% increase in the PLA group. PVPR evolution was also analysed by HbA1c classes (<7%, between 7 and 9%, ≥9%) and results confirmed the superiority of CaD independently of the diabetes control level. A highly significant difference [CaD: −3.38 (SD 13.44) versus PLA: +3.50 (SD 13.70)] was also obtained in a subgroup of patients without anti-hypertensive and/or lipid-lowering agents (P=0.002 at 24 months). A further analysis of the secondary parameters showed significant changes in favour of CaD in the evolution from baseline to the last visit of haemorrhages (P=0.029), DR level (P=0.0006) and microaneurysms (P=0.013). Regarding safety, only 2.5% (n=5 patients/events) of all adverse events reported were assessed as possibly or probably related to the test drug, while all serious adverse events were reported as unlikely. There was no statistical difference between groups. *Conclusion:* Calcium dobesilate 2 g daily for 2 years shows a significantly better activity than placebo on prevention of BRB disruption, independently of diabetes control. Tolerance was very good.

Keywords Diabetic retinopathy · Blood-retinal barrier · Posterior vitreous penetration ratio · Calcium dobesilate

Introduction

Type II diabetes mellitus is one of the most prevalent and costly chronic diseases in Western society, with a rising prevalence [34, 47, 49]. One complication is diabetic retinopathy (DR), in itself a leading cause of visual impairment and blindness in working age adults [7]. Progression of DR can be delayed by tight glycemic control [37], but this can be difficult in practice [46].

Retinal laser photocoagulation can prevent and sometimes improve visual loss [27], but the quality of life of the patient may be altered, e.g. some patients may be unable to drive after surgery due to loss of night and peripheral vision [36, 41]. Moreover, patients undergoing laser photocoagulation may experience immediate worsening after the intervention [1].

Clearly, pharmacological treatment in the early stages of DR to slow or stop its progression would be desirable [14, 24]. Some treatments have been suggested, including aspirin [2, 15], ACE inhibitors [9, 38], angiotensin I and II receptor antagonists [10, 23, 29, 45] and inhibition of the beta isoform of protein kinase C [19]. However, the therapeutic benefit of such agents has yet to be demonstrated in large controlled clinical trials.

Calcium dobesilate (CaD) has been widely prescribed for many years to prevent progression of DR [3]. This synthetic compound is known to be effective in microcirculatory disorders [20, 25, 30, 44].

Several double-blind placebo-controlled clinical studies performed more than 20 years ago have demonstrated the effectiveness of CaD at slowing progression of diabetic retinopathy [4, 20, 42], with efficacy endpoints such as leakage, fluorescein angiography and fundus photography parameters in relatively small cohorts. Except for one study with a treatment duration of 2 years [42], all lasted less than 1 year. Fluorescein leakage, also known as posterior vitreous penetration ratio (PVPR), was the primary endpoint of a pilot study at the high dose of 2 g/day for 12 months and in which the permeability of the blood-retinal barrier (BRB) was stabilised as reflected by PVPR [30].

The objective of the present study was to confirm the efficacy of calcium dobesilate to restore the BRB in a larger patient population at an oral dose of 2 g/day over 2 years.

Materials and methods

Patients

Inclusion criteria were subjects of both sexes, aged 40–75 years, with stable adult onset type 2 diabetes, minimal evidence of retinopathy (score less than 47 on the Wisconsin grading scale [16]) and evidence of mild fluorescein leakage in vitreous fluorometric examination (PVPR between 20 and 50×10^{-6}/min, plasma-free fluorescein, normal value=$15.9\pm4.7\times10^{-6}$/min [12]).

Exclusion criteria were signs of other eye disorders such as macular oedema, retinal vascular diseases or vitreous syneresis and previous laser therapy.

This randomised double-blind placebo-controlled study was conducted in eight European centres. The trial was approved by the competent ethics committees and national authorities, and all patients provided written informed consent prior to enrollment.

Treatment

After final selection, the patient was randomly assigned to active treatment (two capsules of calcium dobesilate 500 mg, Doxium 500, OM PHARMA, Meyrin/Geneva, Switzerland) or a matching placebo (PLA), administered once in the morning and once in the evening after meals, for the duration of the study. Treatment compliance was assessed at each monthly visit by counting the capsules in returned medication bottles. The study lasted 24 months and was monitored regularly to ensure compliance with Good Clinical Practice.

Clinical evaluation

The primary efficacy endpoint was the change in PVPR (measurement of fluorescein leakage) over the entire study, at 24 months and at the last available visit. Leakage was measured every 6 months with a Fluorotron Master (OcuMetrics, Mountain View, Calif., USA). Scans were taken before intravenous administration of fluorescein (14 mg/kg) and 60 min after injection. Blood samples were collected after 10, 15 and 50 min to measure plasma fluorescein concentration. The details of the procedure have been described elsewhere [11, 30].

Retinographic assessments by fundus photography according to Wisconsin Grading [16, 28] and angiographic assessments according to Early Treatment Diabetic Retinopathy Study (ETDRS) Grading [17], determined every 6 months, were used as secondary efficacy endpoints. Visual acuity and intra-ocular pressure were also monitored.

Adverse events were assessed and laboratory analyses were performed every 3 months to evaluate the safety of the drug. These laboratory assessments included measurement of fasting blood glucose and glycosylated haemoglobin (HbA1c) to assess the degree of metabolic control.

Statistical methods

All patients who received at least one dose of study medication were included in the safety analysis. Efficacy was analysed for the intent-to-treat (ITT) population (all

patients entering the treatment phase with at least one efficacy measure after baseline). The worst eye of each patient, as assessed by PVPR and fundus photography, was used for the primary efficacy analysis.

A further analysis of the secondary parameters was conducted to investigate if the choice of the worst eye based on PVPR levels introduced a bias which denied the unveiling of a difference between CaD and PLA in the results of the stereoscopic retinography. This analysis was based on the "worst" fundus eye instead of the "worst" PVPR eye of the previous analysis. The "worst" fundus eye for each patient was defined as the one with the higher value of DR level at baseline. The following fundus parameters were investigated: haemorrhages, microaneurysms and DR level. Exudates were not analysed since too few patients showed these signs during the trial.

Baseline comparisons between groups were performed to verify the homogeneity of groups for the demographic variables using two-sided tests (Table 1).

For the efficacy analysis, ANCOVA by treatment group was performed with all the slopes of the individual PVPR values by time and with the intercept as covariate. The slopes were weighted according to the duration of treatment. The 24-month point and the last time point available for change from baseline of PVPR were also compared between treatment groups by unpaired Student *t*-test or Mann-Whitney test and within groups by paired Student *t*-test or Wilcoxon signed rank test, according to the normality of the distributions.

A subgroup analysis was included in the statistical plan while the data were still blinded depending on the average levels of HbA1c: HbA1c<7.0%, 7.0%≤HbA1c<9.0%, HbA1c≥9.0%.

Secondary efficacy parameters expressed as raw values and changes from baseline were compared for the two groups with the Mann-Whitney test.

Robust multiple regression analyses were also performed for each study group to investigate the influence of other factors on the permeability of the BRB.

The safety variables (adverse events and laboratory values) were displayed descriptively, and the two treatment groups compared using the Fisher exact test or the Mann-Whitney test.

For the sample size calculation, the difference in penetration ratio between calcium dobesilate and placebo was assumed to be 3.4×10^{-6}/min (adjusted from 0.6 reported in the pilot study based on total plasma fluorescein) [30]. Sixty-four patients were required in each treatment group to obtain an alpha of 0.05 and a beta of 0.8, but because of the expected high rate of drop-outs in a 24-month study with out-patients, 240 patients were planned to provide approximately 130 analysable cases.

For all comparisons, *P*-values of 0.05 or less were considered statistically significant. Statistical calculations

Table 1 Demographic and baseline characteristics of ITT population

Characteristics	Calcium dobesilate (*n*=87)			Placebo (*n*=82)			Comparisons
Parametric	Mean	SD	Median	Mean	SD	Median	2-sided test
Age (years)	54.7	7.2	54.5	54.8	7.4	55.6	0.8392
Onset of diabetes (years)	8.52	5.12	7.00	8.07	4.20	7.00	0.7263
Duration of diabetes treatment (years)	6.68	5.19	5.00	6.94	4.31	5.00	0.4979
BMI (kg/m^2)	27.97	3.86	27.38	28.76	4.40	28.09	0.2247
HbA1c (%)	8.20	1.78	7.9	8.01	1.58	7.8	0.5643
Pulse rate (beat/min)	73.4	11.4	72.0	73.4	9.7	72.0	0.8850
BP systolic (mmHg)	134.3	16.2	130	137.8	16.5	140	0.1672
BP diastolic (mmHg)	79.7	9.7	80.0	80.7	8.3	80	0.8285
IOP (mmHg)	15.34	2.40	16.0	15.48	2.28	16.0	0.6936
PVPR (10^{-6}/min)	29.30	10.21	28.04	27.81	8.26	26.06	0.3709
Non-parametric	Description		Missing	Description		Missing	2-sided test
Sex (M/F)	58/29		–	43/39		–	0.0624
Race (W/B/A/other)	85/0/1/1		–	82/0/0/0		–	0.3853
Smoking (Y/N)	6/80		1	7/74		1	0.7769
Dietary restrictions (Y/N)	85/2		–	80/2		–	1.0000
Conc. disease (Y/N)	34/53		–	32/50		–	1.0000
Conc. medication (Y/N)	33/54		–	33/49		–	0.6793
Eye considered (RE/LE)	42/45		–	37/45		–	0.7581
DR level (10/15/20/35)	51/3/18/14		1	48/2/17/15		–	0.9674

BMI body mass index; *HbA1c* glycosylated haemoglobin; *BP* blood pressure; *IOP* intraocular pressure; *PVPR* posterior vitreous penetration ratio; *W* white; *B* black; *A* Asian; *RE* right eye; *LE* left eye; *DR level* diabetic retinopathy level according to ETDRS

were conducted using NCSS version 2004 (NCSS, Kaysville, USA), Systat v.9.0 (SPSS Inc, Chicago, Ill., USA) and Testimate v.5.02 (IDV, Gauting, Germany).

Results

Patients

Of the 299 patients screened for the study, 102 were not randomised because they did not comply with the inclusion/exclusion criteria (Fig. 1). The main reason for exclusion was PVPR outside inclusion limits (80 patients). After the first selection visit, 197 patients were in fact eligible for randomisation. Three of them did not take any medication for different reasons (withdrawal of consent before medication dispensation, lost to follow up after first medication dispensation, and withdrawal of consent at the second visit but returning his entire medication to the investigator) and were therefore excluded from all analyses. Treatment was actually started by 194 patients (98 CaD, 96 PLA), who formed the safety population. The efficacy population (ITT population) comprised the 169 patients (87 CaD, 82 PLA) who attended the first efficacy visit at month 6. The study was completed by 137 patients (69 CaD, 68 PLA). The main reason for discontinuation was the occurrence of an adverse event (nine treated with calcium dobesilate, eight with placebo) or its consequences (e.g. introduction of unauthorised medications).

The demographic and baseline characteristics of the two treatment groups were similar except for sex, which showed a trend to dissimilarity with a higher proportion of men in the active treatment group. DR level at baseline was similar in both groups, with level 10 in 59.3% of CaD group and 58.5% of PLA group (Table 1). Mean treatment duration was 676.5 (SD 136.5) days for the CaD group and 679.4 (SD 131.8) days for the PLA group. Compliance, measured as a percentage of theoretical consumption of 4 capsules a day was 95% for both groups.

Efficacy

The change in posterior vitreous penetration ratio (PVPR) was significantly greater in the active treatment group from baseline to the last available visit (P=0.006) and to the 24-month value (P=0.002) with respect to placebo (Fig. 2). This was confirmed by the ANCOVA comparison of the slopes for the individual regression lines.

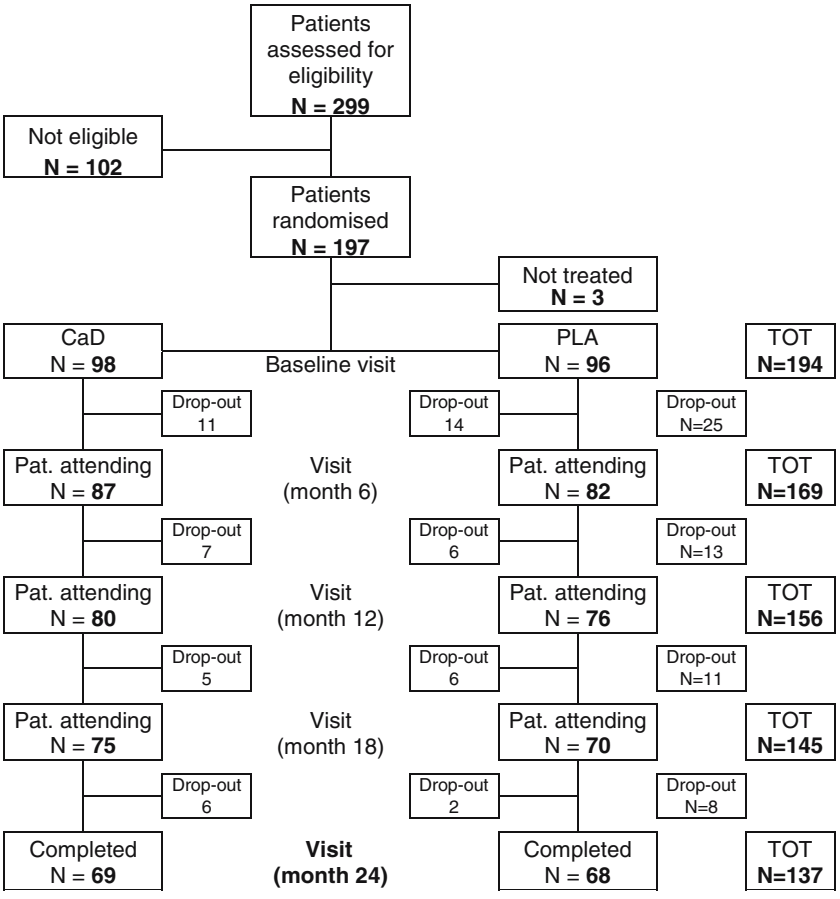

Fig. 1 Patient disposition

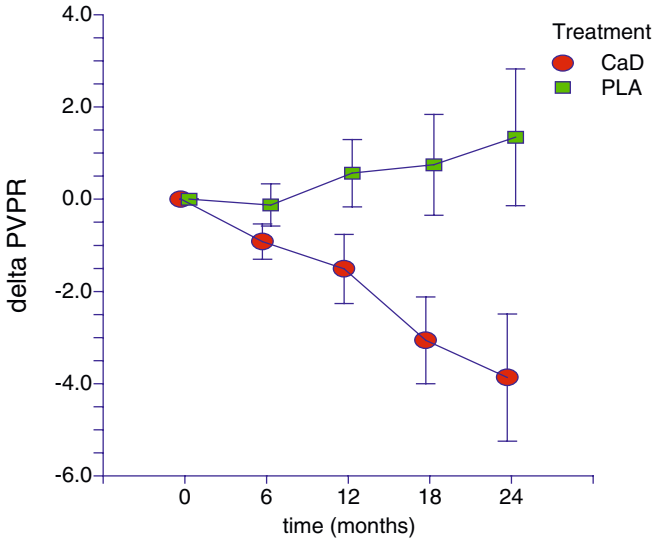

Fig. 2 Progression of PVPR (mean±SEM) during the study, P=0.002 at M24; data obtained by robust regression. *CaD* calcium dobesilate; *PLA* placebo

PVPR decreased progressively from baseline until 24 months in the active treatment group [−3.87 (SD 12.03) 10^{-6}/min corresponding to a percentage decrease of −13.2% versus. baseline (P<0.001)]. In the placebo group, PVPR increased by 7.3% at 24 months with respect to baseline, but this variation was not significant. The results were similar for change from baseline to last available visit, with a significant decrease of PVPR in the active treatment group of −12.4% and a non-significant increase in the placebo group by 4.5%. Figure 3 illustrates the individual evolutions of PVPR from baseline to 24 months. PVPR decreased from baseline in significantly more patients in the active treatment group (61 patients improved, 25 did not) compared with the placebo group (43 patients improved, 39 did not) at the last visit (P=0.018 in the Fisher Exact test). These differences were independent of sex and of centre.

As shown in Table 2, the only variables that significantly correlated with change in PVPR from baseline in the multiple regression analysis were PVPR at baseline (the higher the level, the bigger the difference in favour of active treatment; P<0.001) and treatment (treatment favoured slower progression; P=0.002).

In the subgroup analysis with respect to diabetes control, PVPR in calcium dobesilate-treated group was significantly improved at month 24 for the patients with HbA1c<7% (P=0.043) and between 7 and 9% (P=0.05), and over the whole study period [P=0.044 by ANCOVA of individual slopes [21]] for patients with HbA1c≥9% (Fig. 4).

Regarding concomitant medications, 33 patients in both the active treatment group and placebo group received chronic concomitant medications other than treatment for diabetes. These medications comprised mainly antihypertensive and/or lipid-lowering agents. Figure 5 shows the effect of treatment on PVPR progression according to whether patients took these agents or not. Such treatment had a favourable influence on PVPR in the placebo group, without affecting the efficacy of calcium

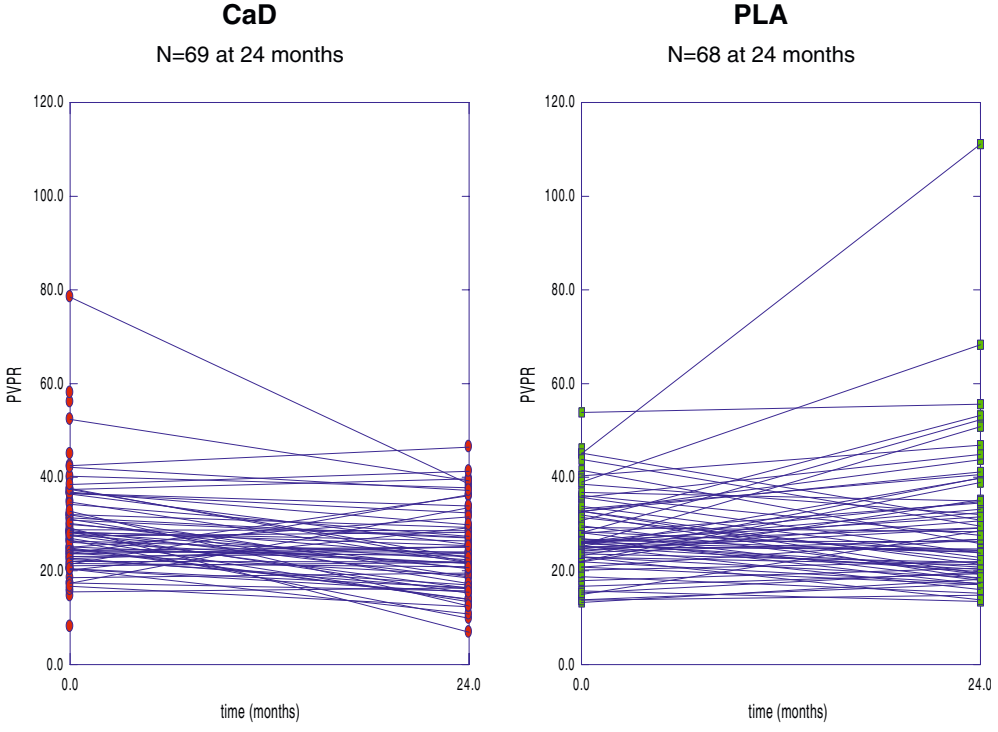

Fig. 3 Individual evolutions of PVPR from baseline to 24 months. *CaD* calcium dobesilate; *PLA* placebo; *n* number of patients

Table 2 Factors affecting posterior vitreous penetration ratio (*PVPR*) progression: a robust multiple regression analysis

PVPR at 24 months (vs baseline) Factor full model R^2=0.2951	Regression coefficient	Standard error	*t*-value	*P*-value
Intercept	−1.0016	13.4956	−0.074	0.9410
Age (years)	0.0815	0.1279	0.637	0.5253
Diastolic blood pressure (mmHg)	0.0590	0.1021	0.578	0.5643
Systolic blood pressure (mmHg)	0.0012	0.0625	0.019	0.9850
Previous duration of diabetes (years)	0.1941	0.1857	1.045	0.2981
Glycosylated haemoglobin (%)	0.1039	0.5128	0.203	0.8398
Body mass index (kg/m^2)	−0.1915	0.1887	−1.015	0.3123
Sex (M=1/F=2)	−0.2939	1.7779	−0.165	0.8690
Intra-ocular pressure (mmHg)	−0.2378	0.4134	−0.575	0.5663
Other concomitant medications (Y=1/N=0)	−3.6403	1.8493	−1.969	0.0513
PVPR at baseline	−0.3445	0.0979	−3.520	0.0006
Treatment (CaD=1/PLA=2)	5.0778	1.6030	3.168	0.0020

dobesilate on this parameter. The evolution of PVPR from baseline to 24 months in the active group was confirmed to be significant in the presence of these drugs (*P*=0.011), which was not the case in PLA group (*P*=0.223). In patients without these medications, PVPR was significantly improved at 24 months (*P*=0.002) in CaD group as compared with placebo [CaD: −3.38 (SD 13.44) versus PLA: +3.50 (SD 13.70)].

There were no consistent or significant differences between groups in the evolution of best corrected visual acuity, intra-ocular pressure, glycaemia or HbA1c.

The fundus or angiography parameters were also similar for both groups at baseline and at study end, with the worst eye based on PVPR. The further analysis of the fundus parameters, with worst eye based on DR level revealed, however, the following results for haemorrhages, microaneurysms and DR level.

Haemorrhages evolution from baseline until the last visit was significantly in favour of CaD compared to PLA (*P*=0.029) as shown in Fig. 6. While the CaD group decreased non-significantly, a significant increase was observed in the placebo group (*P*=0.044). The number of haemorrhages decreased in significantly more patients in the active treatment group compared to the placebo group at the last visit (*P*=0.036).

The evolution of microaneurysms count from baseline until the last visit showed a trend between groups in favour of CaD (*P*=0.066). Moreover, the number of microaneurysms was improved in significantly more patients with the active treatment compared to placebo at the last visit (*P*=0.038). All these results are confirmed by the highly significant worsening for PLA (*P*<0.001) from baseline to the last visit, compared to no significant variations for CaD. By ranking microaneurysms by classes [18], the difference of progression between groups appears even more evident (Fig. 6) and also gives a significant result in favour of CaD (*P*=0.013).

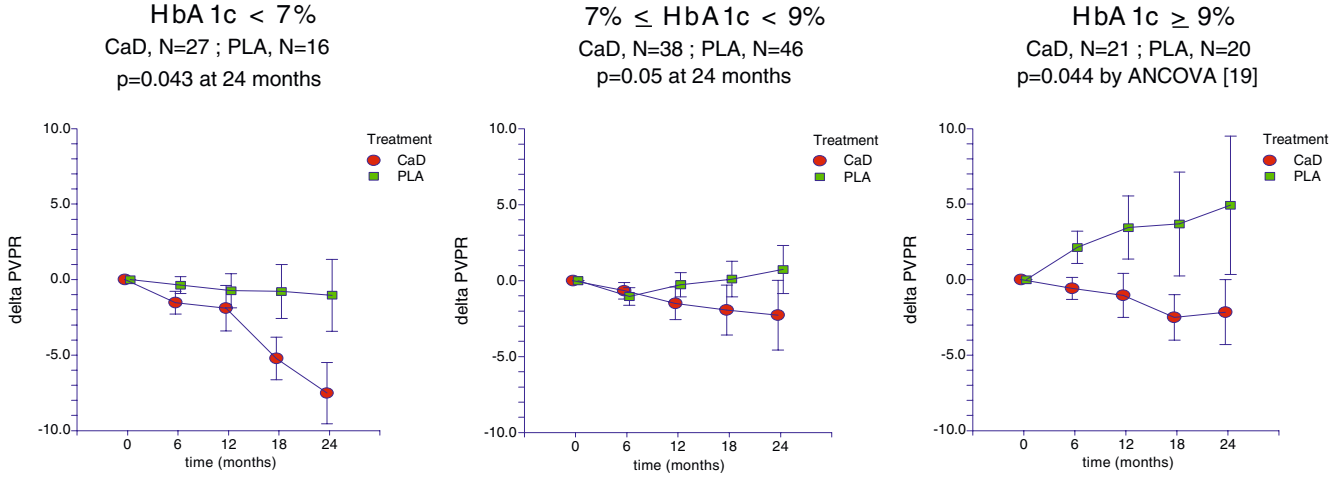

Fig. 4 Evolution of PVPR (mean±SEM) according to HbA1c level (data obtained by robust regression). *CaD* calcium dobesilate; *PLA* placebo; *n* number of patients at baseline

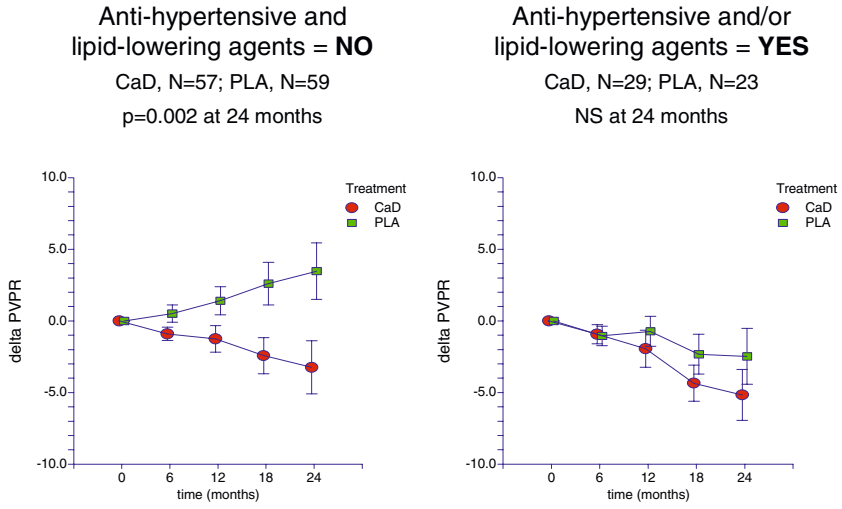

Fig. 5 Progression of PVPR (mean±SEM) according to administration of concomitant medications. *CaD* calcium dobesilate; *PLA* placebo; *n* number of patients at baseline; *NS* not significant

Finally, DR level evolution from baseline until the last visit showed a highly significant difference between groups in favour of CaD ($P<0.001$) as shown by Fig. 6. Again, DR level improved in significantly more patients in the active treatment group than in the placebo group ($P<0.001$).

This analysis of fundus parameters, considering the worst eye based on DR level, confirmed the significant difference in favour of CaD for haemorrhages, microaneurysms and DR level.

Safety

Of the 194 patients (98 CaD, 96 PLA) forming the safety population, 93 (49 CaD, 44 PLA) complained of 202 adverse events (115 CaD, 87 PLA), but only five (three CaD, two PLA) were assessed by the investigators as probably or possibly related to the test drug (Table 3). Two patients died during the study because of myocardial infarction (one in each group), neither of which was considered related to the study drug, and a third patient (PLA) was killed in a car accident. The causal relationship of all serious adverse events to the study drug was reported as "unlikely" (21 CaD, 18 PLA). None of these comparisons was significant between treatment groups.

Discussion

Vascular leakage is a very early process in the pathophysiology of diabetic retinopathy [31, 32], and can be a prelude to a cascade of events that may lead to eventual blindness. Fluorescein leakage into the vitreous is a surrogate endpoint that may potentially predict outcome of clinically relevant endpoints such as visual loss from macular oedema or the need for photocoagulation. Indeed, previous studies have found high rates of leakage to be associated with an increased risk of macular oedema and need for photocoagulation [8, 13, 50].

Calcium dobesilate was recently found to act on basic biochemical mechanisms involved in diabetic retinopathy. In vitro and in vivo results suggest that CaD has antioxidant

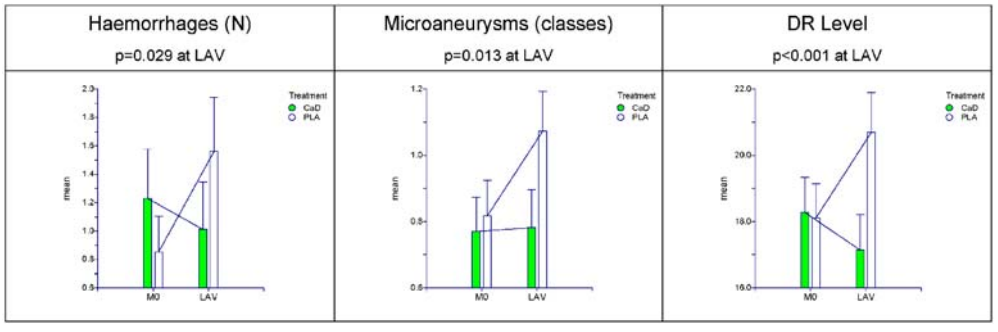

Fig. 6 Evolution of fundus parameters from baseline to the last visit. *CaD* calcium dobesilate; *PLA* placebo; *LAV* last available visit

Table 3 Adverse events with probable or possible relationship with the study drug

Group	Description	Relationship	Severity	Action taken	Outcome	Sequelae
CaD	Constipation	Possible	Severe	Treatment stopped	Resolved	None
CaD	Collapse	Probable	Moderate	None	Resolved	None
CaD	Tachycardia	Possible	Mild	None	No change	None
PLA	Constipation	Possible	Mild	None	Resolved	None
PLA	Rash	Probable	Moderate	Treatment stopped	Unknown	None

properties via a direct scavenging effect of free radicals [5, 6, 50]. Moreover, CaD strongly reduces retinal levels of AGEs and VEGF and reversed retinal hyper-permeability in diabetic rats with pre-proliferative retinopathy [40]. The most plausible explanation of these results is that calcium dobesilate first reduced carboxymethyl-lysine-advanced glycation end product (CML-AGE) formation via its antioxidant properties and that this in turn decreased retinal VEGF over-expression and secondary albumin leakage. The above pharmacological evidence suggests that CaD can stabilise the blood-retinal-barrier in patients with diabetic retinopathy by an antioxidant mechanism [22].

In the last 20 years, several double-blind placebo-controlled clinical studies demonstrated the efficacy of calcium dobesilate at slowing DR progression. Most of them were performed with small cohorts, low dosage and short study duration and their evaluation parameters were only based on fluorescein angiography or fundus photography, but none of them indeed evaluated PVPR.

In the 1990s, a controlled study against no therapy [26], with a dosage of 1.5 g CaD /day in the treated group and a follow-up of 2 years, did not show any reduction in DR progression based on the same parameters. According to the authors, the intake of study drug was poorly controlled, which could explain the lack of efficacy.

On the other hand, a pilot double-blind placebo-controlled study with 47 patients [30] reported that PVPR remained stable in patients with early diabetic retinopathy on calcium dobesilate 2 g/day for 12 months. As in the pilot study, we studied the effect of calcium dobesilate on PVPR in patients with diabetic retinopathy in its early phases, but over 2 years and in a much larger patient collective.

In the present study, the demographics and baseline characteristics of the two treatment groups were comparable, except that there were more men than women in the CaD group, which did not influence the outcome of the analyses.

This study showed a progressive significant improvement in PVPR with respect to baseline in the group treated with calcium dobesilate during the 24 months of the study ($P<0.0001$). The placebo group, on the other hand, showed no significant change with respect to baseline. Direct comparison of calcium dobesilate treatment with placebo showed a significant effect of active treatment ($P=0.006$ for change from baseline to last available value, $P=0.002$ for change from baseline to 24-month value). These results were further confirmed by ANCOVA comparison of the corrected slopes for the individual regression lines.

As expected in a population with minimal retinopathy, treatment with calcium dobesilate did not influence visual acuity or intra-ocular pressure.

Fundus parameters, however, showed significant differences in favour of CaD in the evolution of haemorrhages, microaneurysms and DR level, when considering the worst eye based on DR level at entry.

Higher levels of glycosylated haemoglobin correlate with a faster rate of progression of diabetic retinopathy [33, 43]. Given the importance of levels of HbA1c on progression of diabetic retinopathy, we analysed PVPR evolution by degree of metabolic control. In this analysis, control of BRB leakage was significantly better in the group treated with calcium dobesilate compared to placebo, regardless of degree of metabolic control. The fundus parameters evolution by subgroup of HbA1c also gave significant differences in favour of CaD at the last visit, mainly for patients with HbA1c>9%.

Hypertension correlates with poor prognosis of diabetic retinopathy [39]. According to one study [35], high systemic blood pressure contributes to abnormal blood-retinal barrier permeability but anti-hypertensive therapy reverses this abnormal permeability. Hypercholesterolemia has been described as an additional independent risk factor in diabetic retinopathy, thus lipid-lowering agents may improve prognosis of diabetic retinopathy. The possible influence of anti-hypertensive and/or lipid-lowering agents on the main outcome of the current study was therefore investigated. Almost 40% of our ITT population were taking at least one of these agents. Use of such agents appeared to reduce the PVPR ratio in both the calcium dobesilate group and the placebo group. Such treatment had a favourable influence on PVPR only in placebo-treated patients, without affecting the efficacy of CaD on this parameter. However, a significant reduction with respect to baseline was seen only in the active treatment group.

We conclude that calcium dobesilate significantly lowered the permeability of the BRB as measured by the PVPR. Its effect was manifest regardless of the degree of metabolic control and the use of anti-hypertensive and/or lipid-lowering agents. Moreover, CaD had a significant beneficial effect in controlling the haemorrhages and the

global evolution of DR. Our findings suggest that calcium dobesilate can slow progression of leakage in diabetic retinopathy.

The experience gained in this study, together with previous ones, could be used to design further studies on the efficacy of CaD on visual acuity outcome and the need for photocoagulation in macular oedema.

Acknowledgements The authors would like to thank the statistician, Giancesare Gamba, PhD (Biometrix SA, CH-1196 Gland) and the other members of the DX-Retinopathy Study group: Austria – N. Maar, M. Tittl (Vienna); France – G. Coscas, G. Soubrane, L. Perlemuter, Z. Kahal, H. Oubraham (Créteil); Germany – C. Ohrloff (Frankfurt); Hungary – I. Süveges, A. Borbandy (Budapest); Portugal – E. Leite, C. Lobo (Coimbra) and JF. Castro-Correia, F. Falcão-dos-Reis (Porto); Switzerland – P. Leuenberger, A. Dosso, A. Golay, F. Ustun, L. Sekkat, M. Bagnoud (Geneva) and E. Messmer (Zürich).

References

1. Bailey CC, Sparrow JM (2001) Visual symptomatology in patients with sight-threatening diabetic retinopathy. Diabet Med 18:883–888
2. Bergerhoff K, Clar C, Richter B (2002) Aspirin in diabetic retinopathy. A systematic review. Endocrinol Metab Clin N Am 31:779–793
3. Berthet P, Farine JC, Barras JP (1999) Calcium dobesilate: pharmacological profile related to its use in diabetic retinopathy. Int J Clin Pract 53:631–636
4. Binkhorst PG, Van Bijsterveld OP (1976) Calcium Dobesilate versus placebo in the treatment of diabetic retinopathy: a double-blind cross-over study. Curr Ther Res Clin Exp 20:283–288
5. Brunet J, Farine JC, Garay RP et al (1998) In vitro antioxydant properties of calcium dobesilate. Fundam Clin Pharmacol 12:205–212
6. Brunet J, Farine JC, Garay RP et al (1998) Angioprotective action of calcium dobesilate against reactive oxygen species-induced capillary permeability in the rat. Eur J Pharmacol 358:213–220
7. Buch H, Vinding T, La Cour M et al (2004) Prevalence and causes of visual impairment and blindness among 9980 Scandinavian adults: The Copenhagen City Eye Study. Ophthalmology 111:53–61
8. Bursell SE, Delori Fac, Yoshida A et al (1984) Vitreous fluorophotometric evaluation of diabetics. Invest Ophthalmol Vis Sci 25:703–710
9. Chaturvedi N, Sjolie AK, Stephenson JM et al (1998) Effect of lisinopril on progression of retinopathy in normotensive people with type 1 diabetes. The EUCLID Study Group. EURODIAB Controlled Trial of Lisinopril in Insulin–Dependent Diabetes Mellitus. Lancet 351:28–31
10. Chaturvedi N, Sjoelie AK, Svensson A (2002) The Diabetic Retinopathy Candesartan Trials (DIRECT) Programme, rationale and study design. J Renin Angiotensin Aldosterone Syst 3:255–261
11. Cunha-Vaz JG, Gray JR, Zeimer RC, Mota MC, Ishimoto BH, Leite EB (1985) Characterization of the early stages of diabetic retinopathy by vitreous fluorophotometry. Diabetes 34:53–59
12. Cunha-Vaz JG, Leite E, Castro Sousa JP, Faria de Abreu JR (1993) Blood-retinal barrier permeability and its relation to progression of diabetic retinopathy. A four year follow-up study. Graefe's Arch Clin Exp Ophthalmol 231:141–145
13. Cunha-Vaz J, Lobo C, Sousa JC et al (1998) Progression of retinopathy and alteration of the blood-retinal barrier in patients with type 2 diabetes: a 7-year prospective follow-up study. Graefe's Arch Clin Exp Ophthalmol 236:264–268
14. Cunha-Vaz JG (2000) Diabetic retinopathy: surrogate outcomes for drug development for diabetic retinopathy. Ophthalmologica 214:377–380
15. DAMAD Study Group (1989) Effect of aspirin alone and aspirin plus dipyridamole in early diabetic retinopathy. A multicenter randomized controlled clinical trial. The DAMAD Study Group. Diabetes 38:491–498
16. Davis MD, Hubbard LD, Trautman J, Klein R (1985) Studies of retinopathy. Methodology for assessment and classification with fundus photographs. Diabetes 34 (suppl. 3):42–49
17. ETDRS Research Group (1991) Classification of diabetic retinopathy from fluorescein angiograms. ETDRS report number 11. Early Treatment Diabetic Retinopathy Study Research Group. Ophthalmology 98 (5 Suppl):807–822
18. Feman SS, Leonard-Martin TC, Andrews JS, Armbruster CC, Burdge TL, Debelak JD, Lanier A, Fischer AG (1995) A quantative system to evaluate diabetic retinopathy from fundus photographs. IOVS 36:174–181
19. Frank RN (2002) Potential new medical therapies for diabetic retinopathy: protein kinase C inhibitors. Am J Ophthalmol 133:693–698
20. Freyler H (1974) Microvascular protection with calcium dobesilate (Doxium) in diabetic retinopathy. Ophthalmologica 168:400–416
21. Frison LJ, Pocock SJ (1997) Linearly divergent treatment effects in clinical trials with repeated measures: efficient analysis using summary statistics. Stat Med 16:2855–2872
22. Garay P, Hannaert P, Chiavaroli C (2005) Calcium dobesilate in the treatment of diabetic retinopathy. Treat Endocrinol 4:221–232
23. Gilbert RE, Krum H, Wilkinson-Berka J, Kelly DJ (2003) The renin-angiotensin system and the long-term complications of diabetes: pathophysiological and therapeutic considerations. Diabet Med 20:607–621
24. Giusti C (2002) Is medical treatment for diabetic retinopathy still an unreal dream? Med Hypoth 59:706–709
25. Gloviczki P, Fowl RJ, Hollier LH et al (1985) Prevention of platelet deposition by ibuprofen and calcium dobesilate in expanded polytetrafluoroethylene vascular grafts. Am J Surg 150:589–592
26. Haas A (1995) Einfluss von Kalziumdobesilat auf die Progression der diabetischen Retinopathie. Klin Monatsbl Augenheilkd 206:17–21
27. Harper CA (1999) Treatment of diabetic retinopathy. Clin Exp Optom 82:98–101
28. Klein BEK, Davis MD, Segal P et al (1984) Diabetic retinopathy. Assessment of severity and progression. Ophthalmology 91:10–17
29. Knudsen ST, Bek T, Poulsen PL et al (2003) Effects of losartan on diabetic maculopathy in type 2 diabetic patients: a randomized, double-masked study. J Intern Med 254:147–158

30. Leite EB, Mota MC, Faria de Abreu JR, Cunha-Vaz JG (1990) Effect of calcium dobesilate on the blood-retinal barrier in early diabetic retinopathy. Int Ophthalmol 14:81–88
31. Lobo CL, Bernardes RC, de Abreu JR, Cunha-Vaz JG (2001) One-year follow-up of blood-retinal barrier and retinal thickness alterations in patients with type 2 diabetes and mild nonproliferative retinopathy. Arch Ophthalmol 119:1469–1474
32. Lobo CL, Bernardes RC, Figueira JP et al (2004) Three-year follow-up of blood-retinal barrier and retinal thickness alterations in patients with type 2 diabetes mellitus and mild nonproliferative diabetic retinopathy. Arch Ophthalmol 122:211–217
33. Nakagami T, Kawahara R, Hori S, Omori Y (1997) Glycemic control and prevention of retinopathy in Japanese NIDDM patients. A 10-year follow-up study. Diabetes Care 20:621–622
34. Narayan KM, Boyle JP, Thompson TJ et al (2003) Lifetime risk for diabetes mellitus in the United States. JAMA 290:1884–1890
35. Parving HH, Larsen M, Hommel E, Lund-Andersen H (1989) Effect of antihypertensive treatment on blood-retinal barrier permeability to fluorescein in hypertensive type 1 (insulin-dependent) diabetic patients with background retinopathy. Diabetologia 32:440–444
36. Pearson AR, Keightley SJ, Casswell AG (1998) How good are we at assessing driving visual fields in diabetics? Eye 12:938–942
37. Polak BC, Crijns H, Casparie AF, Niessen LW (2003) Cost-effectiveness of glycemic control and ophthalmological care in diabetic retinopathy. Health Policy 64:89–97
38. Pradhan R, Fong D, March C, Jack R et al (2002) Angiotensin-converting enzyme inhibition for the treatment of moderate to severe diabetic retinopathy in normotensive type 2 diabetic patients. A pilot study. J Diabetes Complications 16:377–381
39. Ratzmann KP, Raskovic M, Thoelke H (1989) Significance of proteinuria and hypertension in the prognosis of type 1 diabetes. Results of a 10-year follow-up study on micro- and macrovascular disease mortality[Article in German]. Dtsch Med Wochenschr 114:1311–1315
40. Rota I, Chiavaroli C, Garay RP et al (2004) Reduction of retinal albumin leakage by the antioxidant calcium dobesilate in streptozotocin-diabetic rats. Eur J Pharmacol 495:217–224
41. Russell PW, Sekuler R, Fetkenhour C (1985) Visual function after pan-retinal photocoagulation: a survey. Diabetes Care 8:57–63
42. Salama-Benarroch I, Nano H, Perez H et al (1977) Assessment of calcium dobesilate in diabetic retinopathy. A double-blind clinical investigation. Ophthalmologica 174:47–51
43. Saum SL, Thomas E, Lewis AM, Croft PR (2002) The effect of diabetic control on the incidence of, and changes in, retinopathy in type 2 non-insulin dependent diabetic patients. Br J Gen Pract 52:214–216
44. Schmidt M, Michal M (1989) Inhibition of sorbitol formation in human erythrocytes by calcium dobesilate. Arzneimittleforschung/Drug Res 39:493–495
45. Sjolie AK, Chaturvedi N (2002) The retinal renin-angiotensin system: implications for therapy in diabetic retinopathy. J Hum Hypertens 16 (Suppl 3):S42–S46
46. Snoek FJ (2000) Barriers to good glycaemic control: the patient's perspective. Int J Obes Relat Metab Disord 24(Suppl 3):S12–S20
47. Stovring H, Andersen M, Beck-Nielsen H et al (2003) Rising prevalence of diabetes: evidence from a Danish pharmaco-epidemiological database. Lancet 362:537–538
48. Szabo ME, Haines D, Garay E et al (2001) Antioxydant properties of calcium dobesilate in ischemic/reperfused diabetic rat retina. Eur J Pharmacol 428:277–286
49. Ubink-Veltmaat LJ, Bilo HJ, Groenier KH et al (2003) Prevalence, incidence and mortality of type 2 diabetes mellitus revisited: a prospective population-based study in the Netherlands (ZODIAC-1). Eur J Epidemiol 18:793–800
50. Van Schaik HJ, Benitez del Castillo JM, Caubergh MJ et al (1998–99) Evaluation of diabetic retinopathy by fluorophotometry. European concerted action on ocular fluorometry. Int Ophthalmol 22:97–104