

Title: Effects of topically administered neuroprotective drugs in early stages of diabetic retinopathy. Results of the EUROCONDOR clinical trial

Short running title: Neuroprotection for diabetic retinopathy

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ABSTRACT

The primary objective of this study was to assess whether the topical administration of two neuroprotective drugs (brimonidine and somatostatin) could prevent or arrest retinal neurodysfunction in diabetic patients with type 2 diabetes. For this purpose, adults aged between 45 and 75 years with a diabetes duration ≥ 5 years, and an ETDRS level ≤ 35 were randomly assigned to one of 3 arms: placebo, somatostatin and brimonidine. The primary outcome was the change in Implicit Time (IT) assessed by mfERG between baseline and at the end of follow-up (96 weeks). A total of 449 eligible patients were allocated to brimonidine (n=152), somatostatin (n=145) and placebo (n=152). When the primary end-point was evaluated in the whole population we did not find any neuroprotective effect of brimonidine or somatostatin. However, in the subset of patients with pre-existing retinal neurodysfunction (34.7%), IT worsened in the placebo group ($p < 0.001$), but remained unchanged in the brimonidine and somatostatin groups. In conclusion, the topical administration of the selected neuroprotective agents appears useful in preventing the worsening of pre-existing retinal neurodysfunction. This finding points to screening retinal neurodysfunction as a critical issue to identify a subset of patients in whom neuroprotective treatment might be of benefit.

INTRODUCTION

Diabetic retinopathy (DR) is classically considered a microvascular disease. However, growing evidence suggests that abnormalities in retinal function can be detected in patients without any evidence of microvascular abnormalities (1,2). In addition, it has been suggested that diabetes-induced retinal dysfunction might contribute to the development of microvascular abnormalities (2). Therefore, it is reasonable to hypothesize that therapeutic strategies aimed at neuroprotection may also be effective in preventing the development and progression of microvascular disease. In fact, there is experimental evidence to support this concept (3,4).

In the early stages of diabetes a high proportion of patients present deficiencies such as decreased hue discrimination and contrast sensitivity, delayed dark adaptation, abnormal visual fields, and impairment of vision related quality of life (5-7). Therefore, neuroprotection itself can be considered a therapeutic target, independently of its potential to prevent the development or progression of microangiopathy (8).

A number of therapeutic strategies based on the main pathogenic mechanisms involved in neurodegeneration have been proposed (9). Systemic administration of drugs blocking these pathways is very unlikely to reach the retina at pharmacological concentrations and, in addition, could have serious adverse effects. On the other hand, if the early stages of DR are the therapeutic target, aggressive treatments such as intravitreal injections would be unacceptable. Topical

treatment with neuroprotective agents in the form of eye drops has been neglected as a possible option because of a general assumption that the posterior chamber of the eye cannot be reached by this route. However, there is emerging evidence that several peptides administered by eye drops are able to reach the retina in pharmacological concentrations, at least in animal models (3, 10-13). Topical administration has the advantage of concentrating drug action to the eye while potentially minimizing systemic effects.

On this basis we conducted the first clinical trial aimed at evaluating the effects of topically administered neuroprotective agents in diabetic patients with no or mild DR. The selected drugs were brimonidine and somatostatin, which have already shown their neuroprotective action in preclinical studies (11, 14). The primary objective was to assess whether these drugs administered topically were able to prevent or arrest neurodegeneration as assessed by mfERG. The main secondary objectives were to evaluate their safety, and to examine their potential impact on the development or progression of DR in terms of microvascular disease.

METHODS

Study design and participants

In this randomized, placebo-controlled, phase II-III trial of parallel groups a total of 450 patients with type 2 diabetes were enrolled at 11 European centers belonging to the EUROCONDOR consortium. The trial (NCT01726075) was funded by the European Commission 7th Framework Programme. The protocol (Supplementary Material) was approved by the local research ethics committee at each site. All participants provided their written informed consent.

Eligibility criteria were age between 45 and 75 years, a diagnosis of type 2 diabetes with a known duration ≥ 5 years, and an Early Treatment for Diabetic Retinopathy Study (ETDRS) ≤ 35 . Exclusion criteria were previously detailed (15).

Randomization and masking

Eligible patients (n= 449) (Supplementary Fig. S1) were randomly allocated in a 1:1:1 ratio to placebo, somatostatin 0.1%, and brimonidine tartrate 0.2% (1 drop BID in each eye in all cases).

Randomization was based on a minimization algorithm that balanced the three groups, stratified by ETDRS level (<20 vs. 20-35).

Procedures

Each patient underwent a comprehensive ophthalmic examination as previously reported (15). Only one eye from each patient was included in the study. If both eyes met the inclusion criteria, one of them was chosen randomly.

Multifocal ERG Recording and Analysis

The mfERGs were recorded in the study eye using the RETI-port/scan 21 (Roland Consult, Berlin, Germany) visual electrophysiology system. Detailed information regarding the methodology used has been previously reported (15).

OCT Imaging and Analysis

SD-OCT images were acquired according to standardized protocols by CIRRUS HD-OCT (Zeiss Meditec), or by Topcon 3D-OCT 2000 (Topcon Corporation), henceforth designated as Topcon, depending on the equipment available at each site. A total of 284 patients underwent CIRRUS HD-OCT imaging, while 165 patients underwent Topcon 3D-OCT 2000 imaging. Further details on the methodology have been previously reported (15).

Follow-up, outcome measurements and adverse events and compliance

Patients were followed and treated during a 96-week period. Study visits were scheduled as detailed in the protocol (Suppl Material) every 24 weeks.

The presence of neurodysfunction was defined as an eye with ≥ 6 altered hexagons for Implicit Time (IT) (16). An altered hexagon was defined as a hexagon with a z-score 2 or higher for IT in comparison with a normative database that was previously created (17).

The primary outcome was the change in the IT assessed by mfERG (between baseline and the end of follow-up).

Secondary outcomes were: other neurodegenerative variables (thickness of the retinal nerve fibre layer [RNFL] and ganglion cell layer [GCL] assessed by SD-OCT), microvascular variables (microaneurysm turnover assessed by colour fundus photography, central retinal thickness assessed by SD-OCT, DR severity assessed by the ETDRS scale, best corrected visual acuity (BCVA) assessed by the ETDRS scale and visual field defects assessed by the Visual Fields Test).

Safety evaluation included assessment of: intra-ocular pressure (IOP), BCVA, conjunctival redness, biomicroscopy, visual fields, blood pressure, heart rate and laboratory safety variables (i.e. blood count and blood biochemistry). In addition, reported adverse events such as overall drop discomfort were recorded.

A compliance of 60% was considered appropriate for this study. If compliance was below 60% or patients interrupted the study medication for more than 1 month, they were not evaluable for efficacy

Sample size calculation

Assuming that, at the end of the study, the placebo group would present 50% of abnormal mfERG IT versus 30% in the patients receiving the active drugs, the number of patients required in each group to demonstrate neuroprotection would be 93. However, as the progression of microvascular changes was also going to be analyzed, the progression rate for patients with very early ETDRS stages at study entry needed to be taken into account. Therefore, assuming that 30% of these patients would present some degree of worsening during the follow-up and that active therapy would reduce this figure to 15%, the number of patients required in each arm was 120. These estimates were performed to assess the efficacy of neuroprotective drugs (somatostatin or brimonidine) vs. placebo with a 2-side risk level of 0.05 and a statistical power of 80%. Taking into account a dropout rate of 20%, the final number of patients to be included in each arm would be 150.

Statistical analyses

For primary analyses (efficacy), we did the analyses per protocol (restricted to participants who completed the study) and by intention-to-treat. Pre-specified analyses for both the prevention and progression of neurodysfunction were performed. No imputations were done for missing data. The safety analysis population included the subjects who received at least one dose of study treatment.

Statistical analysis was performed by TechnoSTAT (www.technostat.co.il) in close collaboration with the statistical team from AIBILI and VHIR. We used the two-tailed paired, or independent samples Student's t-test, and ANOVA for continuous variables. Mixed-effect linear regression models adjusted by HbA1c were performed to evaluate IT progression during follow-up. To examine the association between categorical variables the Chi-Square was used. Data are expressed as mean (\pm standard deviation) for continuous data, and as percentages for categorical data. All analyses were done with SAS® Version 9.4 under Windows® 2008 Terminal.

RESULTS

Baseline characteristics and drop-outs

Between February 5, 2013 and 6 November, 2013, eligible patients (n=449) with type 2 diabetes were randomized. The characteristics of the treatment groups at randomization were well balanced in terms of age, HbA1c and cardiovascular risk factors as previously reported (15).

According to the pre-defined criteria of this study, we found only 156 (34.7%) patients with mfERG abnormalities at baseline (patients with neurodysfunction).

During the 2 years of follow-up a 24% (n=109) drop-out rate was observed, occurring mainly in the first year (Supplementary Fig. S2). The characteristics of patients included in the analysis of efficacy per-protocol according to treatment are shown in Table 1.

Safety

Detailed information of serious adverse events (SAEs) is shown in supplemental material (Tables S1 and S2). Only 1 SAE (ocular hyperaemia) was considered related to the investigational drugs (brimonidine). The most frequent ocular adverse events are detailed in Supplementary Table S3. Brimonidine had more frequent adjudicated ocular adverse events.

Effectiveness

We did not find any significant effect of brimonidine or somatostatin in comparison with placebo on the number of abnormal hexagons during follow-up in the whole population when the analysis was performed per-protocol (Table 2) or by intention-to-treat (p=0.75 and p=0.24, respectively).

Pre-specified subanalyses were performed separately to examine the effects of the neuroprotective drugs in preventing neurodysfunction or arresting its progression. When patients without

neurodysfunction at baseline were analysed, we did not find significant differences in the incidence of neurodysfunction at the end of the study. In contrast, both somatostatin and brimonidine were able to arrest the progression of pre-existing neurodysfunction. Therefore, those patients in whom some degree of neurodegeneration was already present (≥ 6 abnormal hexagons at the baseline), somatostatin and brimonidine were effective in preventing the increase of the mean IT that was observed in the placebo group (Figure 1A). However, when patients were analyzed per intention-to-treat a clear tendency to increasing the IT in the placebo group was also observed but did not reach statistical significance ($p=0.06$) (Figure 1B).

It is worth mentioning that there were no differences between the groups regarding the mean HbA1c during the trial. Therefore, our findings are not influenced by differences in the glycaemic control (Supplementary Table S4 and Table S5).

Regarding the main pre-specified secondary objectives, we did not find any effect of brimonidine or somatostatin in preventing or arresting microvascular disease.

Analyses of SD-OCT data revealed no difference in retinal thickness between the placebo, brimonidine or somatostatin arms at study entry or during follow-up (Table 3).

DISCUSSION

The concept of DR as microvascular disease has evolved into that of a more complex diabetic complication in which neurodegeneration plays a significant role (8). In fact, the American Diabetes Association has recently defined DR as a highly specific neurovascular complication (18). This is the first clinical trial using neuroprotective drugs for treating DR. We have found that eye drops containing neuroprotective agents (brimonidine or somatostatin) did not exert any apparent

effect in terms of primary prevention of neurodysfunction or in modulating the appearance and progression of microvascular disease, at least over 2 years of follow-up and using the methodology previously described. In the subgroup of patients with pre-existing retinal dysfunction, the agents tested were able to arrest the progression of IT only in patients who completed the study without any major protocol violations (per-protocol). Overall, our results suggest that topical administration of somatostatin or brimonidine failed in achieving the primary end point of this clinical trial. However, the subset of patients with neurodysfunction could be envisaged as a promising target population in future clinical trials designed to elucidate this issue. The lack of effectiveness in preventing neurodysfunction could be attributed to the short follow-up (2 years) and to the excellent metabolic control of the patients included in the study.

The mechanisms by which brimonidine and somatostatin arrest the progression of neurodysfunction remain to be fully elucidated. Brimonidine has been shown to effectively promote the survival and function of retinal ganglion cells in a variety of animal models unrelated to diabetes (14). This is the first study showing that topical administration of brimonidine arrests the progression of retinal neurodysfunction in patients with type 2 diabetes.

Somatostatin is abundantly produced by the human retina, the main source being the retinal pigment epithelium (19). Somatostatin exerts relevant functions in the retina (20). In the diabetic retina, a significant downregulation of somatostatin production has been reported (21-23). Therefore, a replacement treatment by the topical route can be envisaged as a reasonable approach for treating DR. The main reasons by which systemic administration of somatostatin analogues failed in arrest progression of DR have been recently reviewed (20), but one of the most important is their inability to cross the blood-retinal barrier.

Our results point to screening for retinal neurodysfunction as a critical issue to identify those patients in whom neuroprotective treatment might be of benefit. In this regard, mfERG is probably not a good option because it is a cumbersome and time-consuming method and, therefore, should be reserved for clinical trials. In addition, mfERG reflects only macular cone photoreceptor function and does not assess broader retinal integrity. Apart from mfERG other methods addressed to measure retinal function have been proposed (9). Among these methods, fundus driven microperimetry is a very sensitive, reliable and rapid method and, consequently, can be a useful tool to screen for neurodysfunction in clinical practice (24, 25).

Our study has several limitations. First, we found a lower prevalence of neurodysfunction than expected. Second, a low progression rate of microvascular disease was found. The inclusion by design of 43% of patients without any microvascular abnormalities at study entry, the short follow-up (2 years) and the excellent HbA1c and blood pressure levels throughout were the main factors accounting for this low rate of DR progression. Third, the drop-out rate was higher than anticipated (24% vs. 20%), but the number of patients who completed the study was higher than required to achieve the primary end point. Finally, there was no way to determine quantitatively whether or not somatostatin and bimonidine reached the human neurosensory retinas in meaningful concentrations.

In conclusion, we did not find any significant effect of topical administration of brimonidine or somatostatin in preventing or arresting both neurodysfunction and microvascular disease in the whole population included in this study. However, these neuroprotective agents could play a role in reducing the progression of pre-existing neurodysfunction. Further studies using new

technologies and with longer follow-up addressed to confirm the neuroprotective effects in this subset of type 2 diabetic population, and whether they result in reduction of microvascular disease are needed.

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Declaration of interests

Berta Ponsati is an employee of BCN Peptides which holds the intellectual property related to the use of ocular somatostatin to treat diabetic retinopathy. No other potential conflicts of interest relevant to this article were reported.

Contributors

CH and MAC analyzed the data, wrote the first draft and edited the manuscript. MP, FB, JG, SPH, SJA, CE, UF, JG-A, JG, GEL, RL, PM, EM, BP, LR and PS researched data, contributed to the discussion and reviewed the manuscript. J.C-V., and R.S. contributed to the study design, discussion, reviewed the manuscript and are the guarantors of this work and, as such, had full access to all the data in the study and take responsibility for the integrity of the data and the accuracy of the data analysis.

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Table 1. Baseline characteristics of patients with type 2 diabetes included in the analysis per-protocol.

	Placebo N=123	Brimonidine N=97	Somatostatin N=120
Age (years)	62.4±7.1	63.7±6.0	62.6±6.6
Gender (% males)	66.1	66.0	65.0
BMI (Kg/m ²)	30.8±5.6	30.8±5.3	31.1±5.4
Diabetes duration (years)	11.6±5.8	11.1±5.5	11.4±5.5
Diabetes treatment (%)			
Diet	4.8	2.1	4.2
Oral agents	65.3	76.3	73.3
Oral agents + Insulin	24.2	21.6	20.8
Insulin	5.6	0.0	1.7
HbA1C (%)	7.21±0.97	7.22±1.09	7.11±0.92
Hypertension (%)	71.0	73.2	71.7
Dyslipidemia (%)	69.4	67.0	67.5
Micro/macroalbuminuria (%)	19.3	22.7	19.1
Cardiovascular disease (%)	19.4	14.4	21.7
ETDRS <20/20-35(%)	42.7/57.3	38.1/61.9	43.3/56.7
BCVA letter score	85.9±5.2	86.1±5.2	85.7±4.6

Table 2. Effect of the investigational drugs on the number of abnormal hexagons assessed by mfERG.

	Patients in whom abnormal hexagons did not increase compared to baseline N (%)	Patients in whom abnormal hexagons increased compared to baseline N (%)
Placebo (n=123)	69 (56.1)	54 (43.9)
BRIM (n=97)	48 (49.5)	49 (50.5)
SST (n=120)	55 (45.8)	65 (54.2)

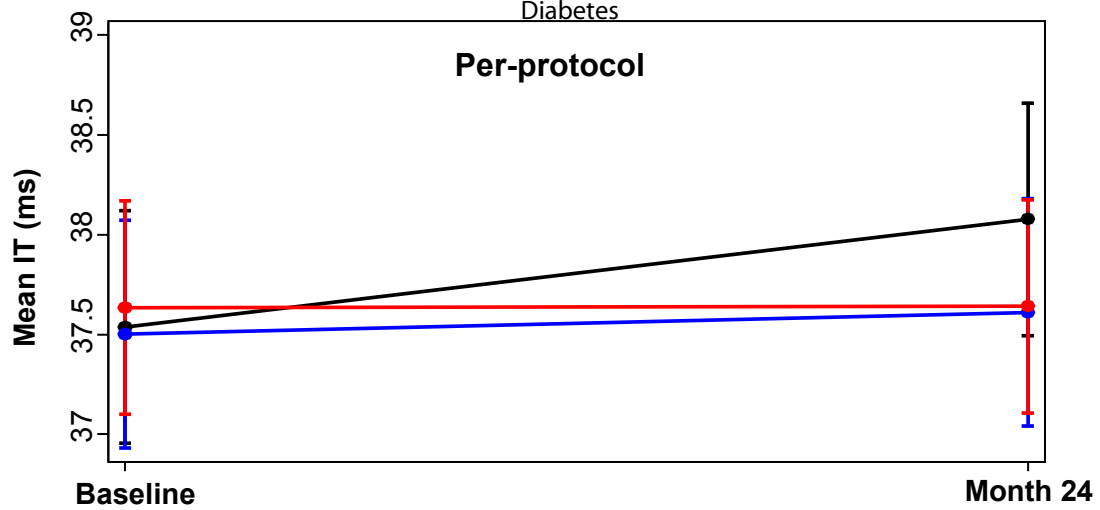
BRIM: brimonidine; SST: somatostatin. BRIM vs. placebo: p=0.29; SST vs. placebo: p=0.11

Table 3. Changes in retinal thickness (RT) measured by SD-OCT. Data are mean \pm standard deviation.

	Baseline (μm)	24 months (μm)	p
Placebo	255.14 \pm 25.93	253.63 \pm 25.16	0.06
BRIM	255.94 \pm 27.03	255.30 \pm 29.13	0.64
SST	256.20 \pm 28.23	257.14 \pm 29.73	0.31

FIGURE LEGEND

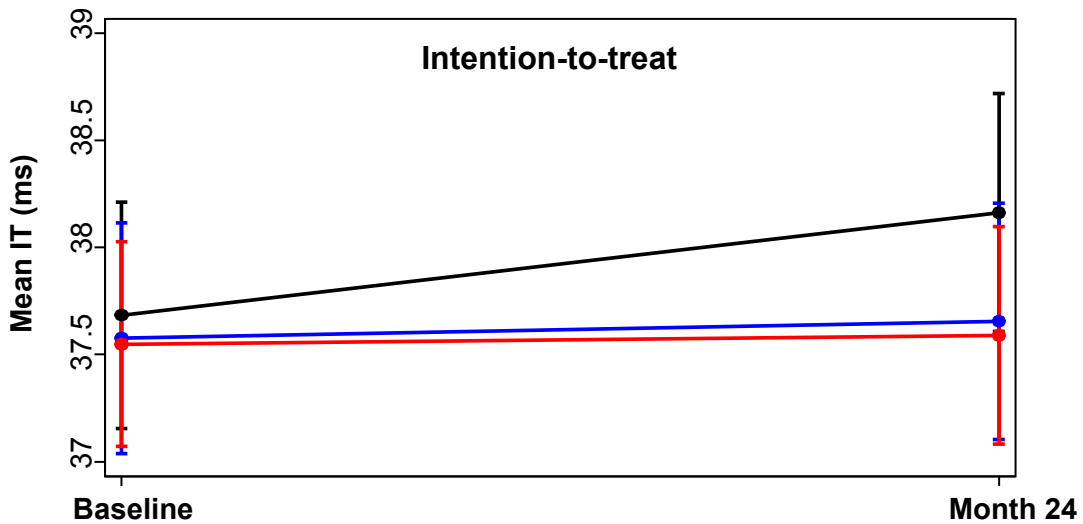
Figure 1. Progression of IT (ms) during follow-up in patients with pre-existing neurodysfunction: A) Analysis per-protocol and B) Analysis by intention-to-treat. Black: placebo group; Red: brimonidine group; Blue: somatostatin group. Difference (change in IT) from baseline to month 24 is expressed as mean (standard deviation).



	Difference	95% Confidence Interval	p-value
Placebo (n= 39)	0.539 (0.264)	0.021 ; 1.057	0.04
BRIM (n= 39)	0.006 (0.242)	-0.468 ; 0.481	0.97
SST (n= 41)	0.108 (0.258)	-0.398 ; 0.614	0.67

SD of Random effect =1.46

B)



	Difference	95% Confidence Interval	p-value
Placebo (n= 49)	0.479 (0.259)	-0.028;0.987	0.06
BRIM (n= 60)	0.041 (0.236)	-.0422;0.50	0.86
SST (n= 47)	0.078 (0.254)	-.421;0.58	0.76

SD of Random effect =1.49

SUPPLEMENTAL MATERIAL**Clinical Protocol**

Link to the protocol published in the clinical trials register:

<https://www.clinicaltrialsregister.eu/ctr-search/trial/2012-001200-38/DE>

Table S1. Serious adverse events by body system, preferred term and treatment group.
Brimonidine vs. Placebo Group

Body System / Preferred Term		Treatment Group		
		Brimonidine	Placebo	Any
Any	Any	26; 21 (14%)	34; 23 (15%)	60; 44 (14%)
Blood and lymphatic system disorders	Any	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)
	Anaemia	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)
Cardiac disorders	Any	5; 5 (3%)	7; 5 (3%)	12; 10 (3%)
	Angina unstable	0; 0 (0%)	2; 2 (1%)	2; 2 (1%)
	Atrial fibrillation	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)
	Atrial flutter	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)
	Cardiac arrest	1; 1 (1%)	0; 0 (0%)	1; 1 (0%)
	Cardiac failure	1; 1 (1%)	1; 1 (1%)	2; 2 (1%)
	Coronary artery disease	2; 2 (1%)	1; 1 (1%)	3; 3 (1%)
	Myocardial infarction	1; 1 (1%)	1; 1 (1%)	2; 2 (1%)
Eye disorders	Any	2; 2 (1%)	0; 0 (0%)	2; 2 (1%)
	Glaucoma	1; 1 (1%)	0; 0 (0%)	1; 1 (0%)
	Ocular hyperaemia	1; 1 (1%)	0; 0 (0%)	1; 1 (0%)
Gastrointestinal disorders	Any	2; 2 (1%)	1; 1 (1%)	3; 3 (1%)
	Colitis ischaemic	1; 1 (1%)	0; 0 (0%)	1; 1 (0%)
	Enterocolitis	1; 1 (1%)	0; 0 (0%)	1; 1 (0%)
	Lower gastrointestinal haemorrhage	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)
General disorders and administration site conditions	Any	1; 1 (1%)	0; 0 (0%)	1; 1 (0%)
	Condition aggravated	1; 1 (1%)	0; 0 (0%)	1; 1 (0%)

Body System / Preferred Term		Treatment Group		
		Brimonidine	Placebo	Any
Hepatobiliary disorders	Any	1; 1 (1%)	0; 0 (0%)	1; 1 (0%)
	Jaundice cholestatic	1; 1 (1%)	0; 0 (0%)	1; 1 (0%)
Infections and infestations	Any	1; 1 (1%)	2; 2 (1%)	3; 3 (1%)
	Gastroenteritis norovirus	1; 1 (1%)	0; 0 (0%)	1; 1 (0%)
	Influenza	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)
	Urinary tract infection	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)
Injury, poisoning and procedural complications	Any	1; 1 (1%)	0; 0 (0%)	1; 1 (0%)
	Tibia fracture	1; 1 (1%)	0; 0 (0%)	1; 1 (0%)
Metabolism and nutrition disorders	Any	1; 1 (1%)	0; 0 (0%)	1; 1 (0%)
	Dehydration	1; 1 (1%)	0; 0 (0%)	1; 1 (0%)
Musculoskeletal and connective tissue disorders	Any	1; 1 (1%)	1; 1 (1%)	2; 2 (1%)
	Bursitis	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)
	Osteoarthritis	1; 1 (1%)	0; 0 (0%)	1; 1 (0%)
Neoplasms benign, malignant and unspecified (incl cysts and polyps)	Any	5; 5 (3%)	9; 8 (5%)	14; 13 (4%)
	Breast cancer	2; 2 (1%)	1; 1 (1%)	3; 3 (1%)
	Chronic myeloid leukaemia	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)
	Gastric cancer	1; 1 (1%)	0; 0 (0%)	1; 1 (0%)
	Hepatic cancer	1; 1 (1%)	0; 0 (0%)	1; 1 (0%)
	Malignant melanoma	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)
	Malignant melanoma stage i	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)
	Metastases to bone	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)
	Rectal adenocarcinoma	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)
	Rectal cancer	1; 1 (1%)	0; 0 (0%)	1; 1 (0%)
	Renal cell carcinoma	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)
	Tongue neoplasm malignant stage unspecified	0; 0 (0%)	2; 2 (1%)	2; 2 (1%)
Nervous system disorders	Any	1; 1 (1%)	5; 4 (3%)	6; 5 (2%)
	Basilar migraine	1; 1 (1%)	0; 0 (0%)	1; 1 (0%)
	Coma	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)
	Ischaemic stroke	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)
	Radicular pain	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)
	Radiculitis	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)

Body System / Preferred Term		Treatment Group		
		Brimonidine	Placebo	Any
	Transient ischaemic attack	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)
Renal and urinary disorders	Any	1; 1 (1%)	1; 1 (1%)	2; 2 (1%)
	Acute kidney injury	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)
	Renal failure	1; 1 (1%)	0; 0 (0%)	1; 1 (0%)
Reproductive system and breast disorders	Any	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)
	Benign prostatic hyperplasia	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)
Respiratory, thoracic and mediastinal disorders	Any	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)
	Obliterative bronchiolitis	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)
Skin and subcutaneous tissue disorders	Any	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)
	Skin ulcer	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)
Surgical and medical procedures	Any	3; 3 (2%)	3; 3 (2%)	6; 6 (2%)
	Breast operation	1; 1 (1%)	0; 0 (0%)	1; 1 (0%)
	Hip arthroplasty	1; 1 (1%)	0; 0 (0%)	1; 1 (0%)
	Skin neoplasm excision	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)
	Surgery	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)
	Transurethral prostatectomy	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)
	Umbilical hernia repair	1; 1 (1%)	0; 0 (0%)	1; 1 (0%)
Vascular disorders	Any	1; 1 (1%)	1; 1 (1%)	2; 2 (1%)
	Aortic aneurysm	1; 1 (1%)	0; 0 (0%)	1; 1 (0%)
	Peripheral artery stenosis	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)

Table S2. Serious adverse events by body system, preferred term and treatment group.
Somatostatin vs. Placebo Group

Body System / Preferred Term		Treatment Group		
		Somatostatin	Placebo	Any
Any	Any	19; 12 (8%)	34; 23 (15%)	53; 35 (12%)
Blood and lymphatic system disorders	Any	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)
	Anaemia	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)
Cardiac disorders	Any	5; 4 (3%)	7; 5 (3%)	12; 9 (3%)
	Angina pectoris	1; 1 (1%)	0; 0 (0%)	1; 1 (0%)
	Angina unstable	0; 0 (0%)	2; 2 (1%)	2; 2 (1%)
	Atrial fibrillation	1; 1 (1%)	1; 1 (1%)	2; 2 (1%)
	Atrial flutter	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)
	Atrioventricular block complete	1; 1 (1%)	0; 0 (0%)	1; 1 (0%)
	Cardiac failure	2; 2 (1%)	1; 1 (1%)	3; 3 (1%)
	Coronary artery disease	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)
	Myocardial infarction	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)
Eye disorders	Any	1; 1 (1%)	0; 0 (0%)	1; 1 (0%)
	Visual impairment	1; 1 (1%)	0; 0 (0%)	1; 1 (0%)
Gastrointestinal disorders	Any	1; 1 (1%)	1; 1 (1%)	2; 2 (1%)
	Duodenal ulcer haemorrhage	1; 1 (1%)	0; 0 (0%)	1; 1 (0%)
	Lower gastrointestinal haemorrhage	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)
General disorders and administration site conditions	Any	1; 1 (1%)	0; 0 (0%)	1; 1 (0%)
	Chest pain	1; 1 (1%)	0; 0 (0%)	1; 1 (0%)
Hepatobiliary disorders	Any	1; 1 (1%)	0; 0 (0%)	1; 1 (0%)
	Cholelithiasis	1; 1 (1%)	0; 0 (0%)	1; 1 (0%)
Infections and infestations	Any	3; 3 (2%)	2; 2 (1%)	5; 5 (2%)
	Influenza	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)
	Lower respiratory tract infection	1; 1 (1%)	0; 0 (0%)	1; 1 (0%)
	Pneumonia	1; 1 (1%)	0; 0 (0%)	1; 1 (0%)
	Respiratory tract infection	1; 1 (1%)	0; 0 (0%)	1; 1 (0%)
	Urinary tract infection	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)
Injury, poisoning and procedural complications	Any	1; 1 (1%)	0; 0 (0%)	1; 1 (0%)

Body System / Preferred Term		Treatment Group		
		Somatostatin	Placebo	Any
	Hip fracture	1; 1 (1%)	0; 0 (0%)	1; 1 (0%)
Musculoskeletal and connective tissue disorders	Any	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)
	Bursitis	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)
Neoplasms benign, malignant and unspecified (incl cysts and polyps)	Any	0; 0 (0%)	9; 8 (5%)	9; 8 (3%)
	Breast cancer	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)
	Chronic myeloid leukaemia	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)
	Malignant melanoma	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)
	Malignant melanoma stage i	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)
	Metastases to bone	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)
	Rectal adenocarcinoma	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)
	Renal cell carcinoma	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)
	Tongue neoplasm malignant stage unspecified	0; 0 (0%)	2; 2 (1%)	2; 2 (1%)
Nervous system disorders	Any	0; 0 (0%)	5; 4 (3%)	5; 4 (1%)
	Coma	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)
	Ischaemic stroke	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)
	Radicular pain	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)
	Radiculitis	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)
	Transient ischaemic attack	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)
Renal and urinary disorders	Any	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)
	Acute kidney injury	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)
Reproductive system and breast disorders	Any	2; 2 (1%)	1; 1 (1%)	3; 3 (1%)
	Benign prostatic hyperplasia	1; 1 (1%)	1; 1 (1%)	2; 2 (1%)
	Priapism	1; 1 (1%)	0; 0 (0%)	1; 1 (0%)
Respiratory, thoracic and mediastinal disorders	Any	3; 2 (1%)	1; 1 (1%)	4; 3 (1%)
	Acute respiratory failure	1; 1 (1%)	0; 0 (0%)	1; 1 (0%)
	Diaphragmatic paralysis	1; 1 (1%)	0; 0 (0%)	1; 1 (0%)
	Obliterative bronchiolitis	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)
	Pleural effusion	1; 1 (1%)	0; 0 (0%)	1; 1 (0%)
Skin and subcutaneous tissue disorders	Any	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)
	Skin ulcer	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)
Surgical and medical procedures	Any	1; 1 (1%)	3; 3 (2%)	4; 4 (1%)

Body System / Preferred Term		Treatment Group		
		Somatostatin	Placebo	Any
	Cardiac pacemaker insertion	1; 1 (1%)	0; 0 (0%)	1; 1 (0%)
	Skin neoplasm excision	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)
	Surgery	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)
	Transurethral prostatectomy	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)
Vascular disorders	Any	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)
	Peripheral artery stenosis	0; 0 (0%)	1; 1 (1%)	1; 1 (0%)

Table S3. Ocular adverse events by treatment group.

	Placebo	BRIM	SST
Eye pain	21	41	18
Ocular hyperaemia	4	49	14
Eye pruritus	14	14	12
Anterior chamber disorder	14	8	10
Foreign body sensation	7	17	6
Dry eye	9	9	10
Lacrimation increased	7	13	4
Eye discharge	8	3	12
Vision blurred	2	7	14
Conjunctival follicles	0	16	1
Conjunctivitis	5	7	5
Blepharitis	5	6	2
Conjunctivitis allergic	1	10	2
Eyelid oedema	1	12	0
Conjunctival hyperaemia	0	9	1
Punctate keratitis	4	4	1

BRIM: brimonidine; SST: somatostatin

Table S4. HbA1c measurements at baseline and during follow-up

	Placebo N= 123	BRIM N= 97	SST N= 120	p
HbA1C at baseline (%)	7.21±0.97	7.22±1.09	7.11±0.92	n.s
HbA1C at month 6 (%)	7.23±1.08	7.19±1.14	7.14±1.07	n.s
HbA1C at month 12 (%)	7.28±1.26	7.19±1.20	7.13±0.94	n.s
HbA1C at month 18 (%)	7.22±1.27	7.01±1.13	7.25±1.17	n.s
HbA1C at month 24 (%)	7.35±1.33	7.02±1.27	7.35±1.08	n.s
Mean HbA1C (%)	7.26±1.06	7.13±1.03	7.21±0.90	n.s

Data are mean±SD

BRIM: brimonidine; SST: somatostatin

Table S5. HbA1c measurements at baseline and during follow-up in patients with neurodysfunction at baseline

	Placebo N= 39	BRIM N= 39	SST N= 41	p
HbA1C at baseline (%)	7.19±0.98	7.25±1.12	7.12±0.91	n.s
HbA1C at month 6 (%)	7.03±1.06	7.32±1.22	7.13±1.03	n.s
HbA1C at month 12 (%)	7.09±1.04	7.30±1.33	7.17±0.98	n.s
HbA1C at month 18 (%)	7.03±0.99	6.95±0.99	7.17±1.10	n.s
HbA1C at month 24 (%)	7.16±1.10	7.05±1.05	7.37±1.19	n.s
Mean HbA1C (%)	7.09±0.94	7.15±1.05	7.21±0.94	n.s

Data are mean±SD

BRIM: brimonidine; SST: somatostatin

Figure S1. Trial profile

Diabetes

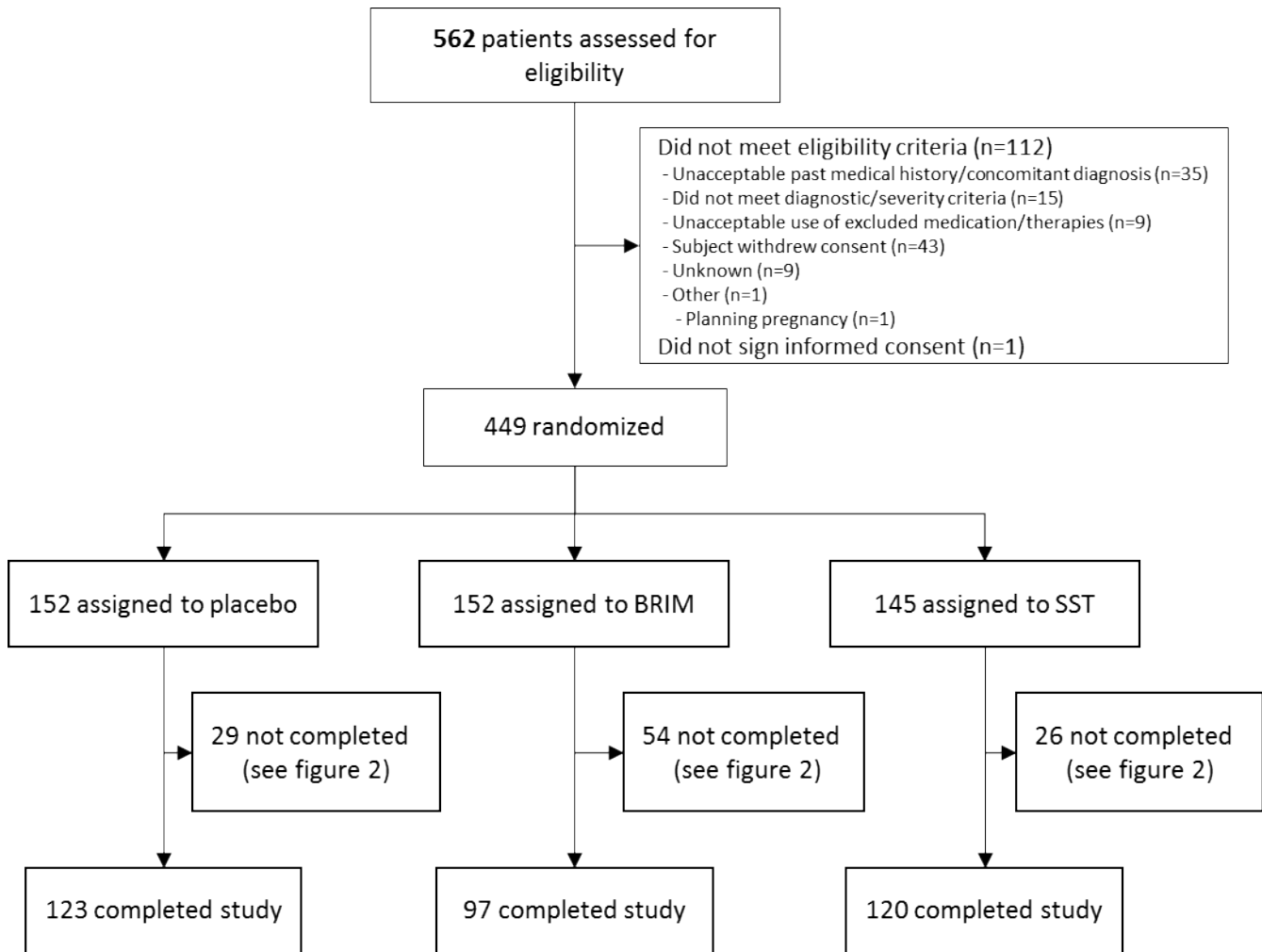
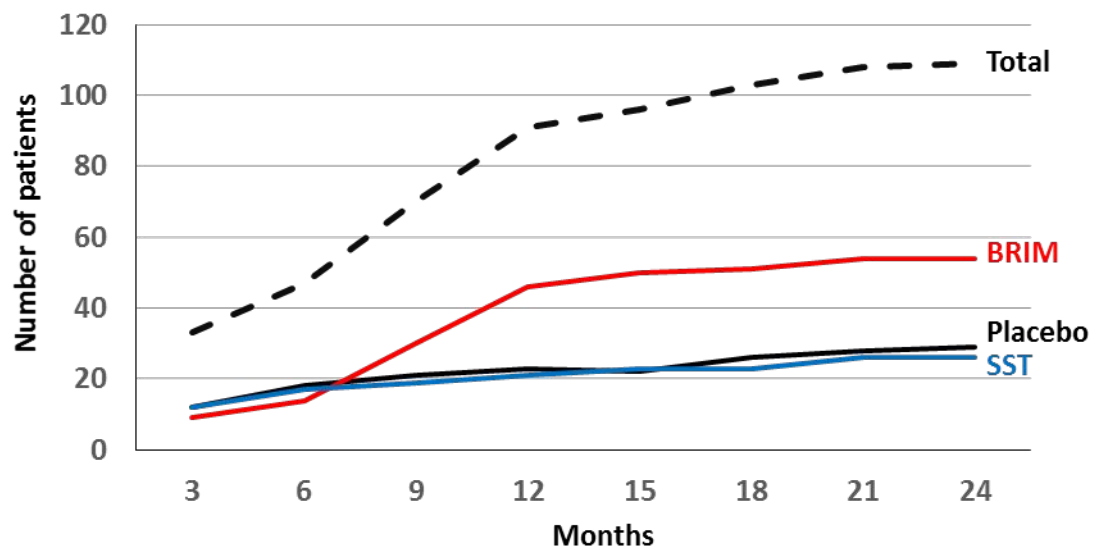


Figure S2. Evolution (upper panel) and causes (lower panel) of dropouts.

	Placebo	BRIM	SST	Total
Ocular adverse effects	5	35	3	43
Withdrawal of Informed Consent	12	8	13	33
Other (i.e. intercurrent illness)	12	11	10	33
Total patients, n (%)	29 (19.3%)	54 (36%)	26 (17.3%)	109 (24.2%)

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