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ORIGINAL RESEARCH

Real-World Outcomes of Allogeneic Immunosafe Plasma Rich in Growth Factors Eye Drops for Refractory Ocular Surface Diseases: A Prospective Observational Study

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ABSTRACT

Introduction: Immunosafe plasma rich in growth factors eye drops (is-ePRGF) have shown anti-inflammatory and regenerative effects on ocular surface diseases (OSD). However, medical or technical issues may preclude some patients from undergoing autologous blood extraction. We aimed to evaluate the safety and clinical outcomes of allogeneic is-ePRGF therapy for refractory OSD in real-world practice.

Methods: A single-center cohort was conducted involving consecutive patients with severe OSD nonresponsive to conventional therapy. All participants received allogeneic is-ePRGF derived from healthy family donors as compassionate treatment (one drop four times daily for 6 weeks per cycle), with a minimum follow-up of 3 months. Primary outcomes included symptom changes assessed by Ocular Surface Disease Index (OSDI) and Symptom Assessment iN Dry Eye (SANDE) questionnaires, resolution of persistent epithelial defects (PED), and adverse events (AE). Secondary outcomes included best-corrected visual acuity (BCVA), intraocular pressure (IOP), corneal staining, Schirmer I test, tear break-up time (BUT), conjunctival bulbar redness, and meibomian gland dysfunction (MGD). Multilevel mixed-effects models were used to account for intra-patient and inter-eye correlations.

Results: A total of 30 patients (58 eyes; mean age 53.4 ± 22.6 years; 50% female) were included. The most common diagnoses were severe dry eye disease and neurotrophic keratopathy, frequently associated with autoimmune/inflammatory conditions. Overall, 12 patients (40%) had previously received autologous therapies. Donors were primarily first-degree relatives. Median follow-up was 9 months (range 3–30 months), with

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a mean of 6.2±3 cycles. OSDI and SANDE scores significantly improved ($p<0.001$). Among 12 cases with PED, 8 (67%) fully resolved. No AE occurred. BCVA ($p=0.010$), Schirmer test, BUT, conjunctival bulbar redness, corneal staining, and MGD severity improved significantly ($p<0.001$); IOP remained stable ($p=0.132$).

Conclusions: Allogeneic is-ePRGF was a safe and effective alternative for refractory OSD when autologous sources were not available or suitable. Standardization of regulatory frameworks for allogeneic blood-based therapies is needed to support broader clinical adoption.

Keywords: Allogeneic blood-based eye drops; Dry eye disease; Hemoderivatives; Ocular surface disease; Plasma rich in growth factors; PRGF

Key Summary Points

Why carry out this study?

Blood-based therapy has emerged as a promising option for refractory ocular surface diseases (OSD); however, certain patients are medically or technically unsuitable for autologous blood donation.

Allogeneic immunosafe plasma rich in growth factors eye drops (is-ePRGF) may offer a viable alternative to address this unmet need.

What was learned from the study?

Allogeneic is-ePRGF derived from healthy donors significantly improved both symptoms and clinical signs, showing a safety profile in patients with refractory OSD who were ineligible for autologous blood donation.

The use of allogeneic sources in blood-based therapy may enhance biological consistency, facilitate standardized quality control, streamline processing, and expand access to regenerative treatments for complex cases.

However, harmonization of regulatory framework for blood-derived therapies is essential to support clinical implementation.

INTRODUCTION

Ocular surface disease (OSD) is an umbrella term that encompasses a wide range of pathologies affecting any structure of the ocular surface, along with related disorders and their corresponding responses. Refractory OSD refers to a subset of conditions that does not adequately respond to conventional treatments, leading to vision-threatening complications such as corneal nerve damage, melting, and perforation. It is estimated that approximately 10–15‰ patients with moderate-to-severe dry eye disease (DED) may develop a refractory form of the disease, although prevalence can vary depending on diagnostic criteria and patient population [1]. The exact underlying mechanism is multifactorial and may involve chronic and dysregulated inflammation, altered ocular surface immunity, and neurotrophic abnormalities that impair tissue healing [2, 3].

Current management of these refractory cases includes the use of preservative-free artificial tears, topical corticosteroids, immunomodulators (e.g., cyclosporine A, lifitegrast), autologous serum (AS) eye drops, and, in more severe cases, systemic immunosuppressants. However, these treatments may be associated with suboptimal efficacy or adverse effects. For example, long-term use of corticosteroids can lead to increased intraocular pressure and cataract formation, while immunomodulators may be poorly tolerated and slow-acting. In addition, recent evidence suggests that the presence of circulating proinflammatory cytokines or systemic medications in AS may affect its therapeutic consistency and safety in some patients [4, 5].

Plasma-based eye drops have gained popularity in treating these complex cases owing to their favorable safety and efficacy profiles [5, 6]. Over the last two decades, immunosafe plasma rich in growth factors (is-PRGF), an autologous platelet-rich plasma (PRP) characterized by the absence of leukocytes, a controlled platelet-fibrinogen activation step, long-term biostability, and a versatile formulation (e.g., for topical application as eye drops, is-ePRGF; injections; or surgical use as membrane or clot, termed is-mPRGF/is-cPRGF), has been introduced in the armamentarium for

refractory OSD cases as an anti-inflammatory and regenerative agent [7–12], showing higher biological potential than AS or insulin [13, 14].

Unfortunately, not all patients who could benefit from the biological properties of is-ePRGF are suitable for blood extraction. Certain patients may have poor venous access, low hemoglobin levels, hematological disorders, anticoagulant medication, or age-related issues [15]. Moreover, the blood of patients with inflammatory or autoimmune diseases may have high levels of proinflammatory mediators [16, 17] or cytotoxic drugs that could negatively impact the ocular surface [18, 19]. Immunosuppression may also reduce the epitheliotropic properties of hemoderivatives, requiring higher concentrations for effective outcomes [20]. An allogeneic source from adult peripheral blood or umbilical cord blood (UCB) is an alternative that could address these limitations [21–24].

However, conducting randomized controlled trials to evaluate the safety and effectiveness of a therapy for severe OSD is challenging owing to the nature of the disease, the wide range of local and systemic comorbidities, and differences in the types and dosages of prior local and systemic medications. This situation limits the translatability of the results into real-world clinical settings [25]. Currently, there is some indirect evidence of the comparative efficacy and effectiveness of blood-based eye drops [26]; however, no direct evidence on allogeneic is-ePRGF has been published [6]. Therefore, we aimed to report the feasibility and clinical outcomes of allogeneic is-ePRGF from healthy donors in refractory OSD cases within real-world practice.

METHODS

Study Design

This was an observational, single-center study performed at the Instituto Oftalmológico Fernández-Vega, Oviedo, Spain. The protocol was approved by the Ethics Committees for Investigation with Medicinal Products of the Principality of Asturias (CEImPA 2023.281), and the procedures were conducted in adherence with the tenets of the Declaration of Helsinki. Written informed consents were obtained from every patient and donor before the procedure as routine clinical practice, including permission to use their medical records for scientific purposes. Authorization was granted by the Spanish Agency for Medicines and Health Products (AEMPS) to initiate allogeneic is-ePRGF as compassionate treatment for every enrolled patient.

Study Participants

Medical records from consecutive patients diagnosed with severe OSD who showed no improvement after at least 3 months of conventional treatment (e.g., non-preservative artificial tears, topical/oral antibiotics, topical/oral corticosteroids, therapeutic contact lenses, eye patches, punctal occlusion, autologous hemoderivatives, or cyclosporine) were reviewed. For each case, a request was submitted to AEMPS, and once authorization was obtained, patients received is-ePRGF under a compassionate use protocol. We included patients who had a minimum follow-up of 3 months. Patients with active ocular surface infections were excluded.

Donor Selection

Allogeneic is-ePRGF was procured from healthy family members (related or not) with no history of blood-borne diseases or blood transfusion and no systemic active inflammation. To be eligible for blood donation, the relative had to be a minimum of 18 years of age and weigh at least 50 kg. An ad hoc blood donor questionnaire was completed by the donor, and serological tests for *Treponema pallidum*, human immunodeficiency virus, hepatitis B virus, and hepatitis C virus were conducted prior to venipuncture. ABO blood type matching was not considered.

Allogeneic is-ePRGF Preparation

Allogeneic is-ePRGF follows the same standardized protocol as the autologous one, by which platelets are around 2.84-fold concentrated,

and red and white cells are discarded [11, 27]. The streamlined procedure used the single-use Endoret® Kit (BTI Biotechnology Institute, S.L., Vitoria, Spain) and has been published elsewhere [8, 9, 12]. Briefly, blood was collected into 9-mL tubes containing 3.8% sodium citrate and centrifuged at 580 *g* for 8 min for blood component separation. Then, under highly sterile conditions inside a biohazard laminar flow hood, the supernatant was

◂**Fig. 1** Allogeneic immunosafe plasma rich in growth factors eye drops (is-ePRGF) protocol. (**a**) Blood was collected into 9-mL tubes containing anticoagulant. (**b**) Samples were centrifuged at 580 *g* for 8 min at room temperature. (**c**) Then, the supernatant was collected using a closed system under a laminar flow hood. (**d**) It was activated with 10% calcium chloride (**e**) at 50 µL per 1 mL of plasma. (**f**) Next, the released plasma was incubated at 37 °C for 1 h, and (**g**) heat-treated at 56 °C for 60 min to reduce the immunologic components. (**h**) Finally, the treated plasma was filtered and (**i**) aliquoted in a single-use kit. (**j**) A sample of is-ePRGF was obtained for microbiological analysis control in each preparation. (**k**) A close-up view of an empty disposable kit, each containing 32 dispensers of 0.5 mL. This should be stored at −20 °C until use. (**l**) The dispenser in use can be kept in an individual container and stored at 4 °C for up to 72 h

collected by a closed technique, avoiding the buffy coat that contains leukocytes. Next, it was activated with 10% CaCl2 (50 µL per 1 mL of plasma) and incubated at 37 °C for 1 h. The released plasma was heated at 56 °C for 60 min to reduce the immunologic components [8, 28]. After that, the supernatants were filtered, aliquoted, and stored in a sealed and disposable kit at –20 °C until use. A sample was taken for microbiological control. (Fig. 1).

Treatment Regimen

Once the microbiology control tested negative, the patients were instructed to apply one eye drop four times daily for 6 weeks ("one cycle") on the affected eye. The number of cycles could be extended as needed, and other concomitant local or systemic medications could be continued except for autologous hemoderivatives, which the allogeneic is-ePRGF should replace.

To ensure sterile conditions, storing the is-ePRGF dispensers at –20 °C for up to 3 months and keeping the bottle in use for up to 72 h at 4 °C was recommended.

Data Extraction

Data collection included demographic and clinical details such as age at presentation, gender, ocular and systemic comorbidities, prior medication, and baseline and last follow-up clinical subjective and objective outcomes. In addition, the donor–recipient relationship was recorded.

Outcome Measures

Assessments were performed during routine clinic hours (9 a.m.–5 p.m.); timing relative to drop instillation or waking was not standardized, reflecting real-world practice. The primary outcomes included symptom changes, the rate of healing for epithelial defects/corneal ulcers, and the frequency of adverse events (AE). Changes in symptoms from baseline conditions were measured using the Ocular Surface Disease Index (OSDI) and the Symptom Assessment iN Dry Eye (SANDE) questionnaires. The OSDI is a 12-item questionnaire that assesses certain DED symptoms in the past week of a patient's life; its final score ranges from 0 to 100, with higher scores representing more significant disability. The SANDE questionnaire assesses the frequency and severity of dryness and/or irritation symptoms using a visual analog scale from 0 to 100 [29].

For patients presenting persistent epithelial defects (PED) or corneal ulcers, the response was categorized as complete resolution (null to minimal corneal staining), partial improvement (at least 50% of healing), stable (no change in the corneal defect), or worsening (enlargement or deepening of the corneal defect). We recorded AE, including but not limited to a burning sensation, redness, a sticky sensation, and an allergic reaction.

Secondary outcomes included corneal fluorescein staining, Schirmer I test, tear break-up time (BUT), conjunctival bulbar redness, best-corrected visual acuity (BCVA) at decimal scale, and intraocular pressure (IOP).

The area and density of fluorescein corneal staining were graded using the following scale: stained area (0 = no dotted staining, 1 = staining less than 1/3 of the cornea, 2 = staining between 1/3 and 2/3 of the cornea, and 3 = more than 2/3 of the cornea) and density (0 = no dotted staining, 1 = sparse density, 2 = moderate density, and 3 = high density with overlapping corneal

lesions) [30]. The degree of conjunctival bulbar redness was assessed by the Oculus Index (Keratograph 5 M®, Oculus, Wetzlar, Germany). This system automatically generates a bulbar redness score ranging from 0 to 4, determined by the percentage ratio of the vessel area to the total analyzed area [31].

The severity of meibomian gland dysfunction (MGD) was graded on a scale from level 0 to level 5 according to the International Workshop on Meibomian Gland Dysfunction: level 0 = no dysfunction; 1 = subclinical, nonobvious MGD; altered quality, only on expression; no gland loss; 2 = minimally altered quality of expressed meibum from scattered glands; none to minor loss; 3 = mildly altered meibum quality; occasional lid margin signs; mild gland loss; 4 = moderately increased opacity and viscosity of meibum; plugging; increased marginal vascularity; loss of orifice definition; moderate gland loss; 5 = marked, diffuse MGD; cicatricial or noncicatricial; multiple lid margin signs; lid deformity and marked lid margin hyperemia; severe gland loss [32].

Statistical Analysis

The Shapiro–Wilk test was used to assess normal distribution, and the F-test was used to assess variance equality. Descriptive statistical data were presented as mean ± standard deviation (SD) or median and range for continuous data and percentages for categorical variables. For summarizing inter-eye continuous correlated data, linear mixed models were used to show the variability accounting for both within- and between-patient differences.

Continuous variables before and after treatment with allogeneic is-ePRGF at the patient-level (e.g., symptoms) were compared using a paired *t*-test or Welch's *t*-test, and at eye-level (e.g., BCVA), multilevel mixed-effects linear regression models were used. Size effects were calculated using Cohen's *d*. For ordinal categorical data (e.g., MGD severity), mixed-effects ordinal logistic regression models were used to adjust for nonindependence of eyes within patients, clustering at the patient level and comparing repeated measures within eye (e.g., before/after allogeneic is-ePRGF). All statistical analyses were completed in Stata® version 15.1 (StataCorp. 2015, Stata Statistical Software: Release 15. College Station, TX, USA). A *p*-value less than 0.05 was considered to be statistically significant.

RESULTS

Demographic Data and Clinical Characteristics

We included 30 patients (58 eyes), with a male-to-female ratio of 1:1. The mean age was 55.4 ± 22.6 years (range: 5–90 years). The patients' offspring were the most common blood donors (11, 36.7%) in this cohort, followed by siblings (6, 20.0%), mothers and spouses (5, 16.7% each), and family-in-law (3, 10.0%). Most of the donors were females (18, 60%).

The systemic and ocular pathologies are detailed in Table 1. Most patients had autoimmune and inflammatory diseases under immunosuppressive treatment. Five cases had low platelet count, three reported blood clotting disorders, and two had chronic anemia. Poor venous access limited autologous phlebotomy in three patients and severe encephalopathy in one pediatric case. Severe DED and neurotrophic keratopathy (NK) were the most common OSD. A total of 46 out of 58 eyes (79%) had punctate epithelial erosion (PEE), while the remaining 12 (21%) exhibited PED. In addition, 12 patients (40%) had already been treated with autologous therapy without response. After initiating allogeneic is-ePRGF, the median follow-up time was 9 months (range: 3–30 months).

Primary Outcomes

A total of 29 patients completed the questionnaires after at least 3 months of treatment, with a mean number of cycles of 6.2 ± 3 (range: 2–14 cycles) during the follow-up. Before treatment, the mean OSDI score was 42.9 ± 9 (range: 25–67); 93.1% (27 cases) reported severe symptoms, while only 2 patients experienced moderate symptoms. After treatment, the OSDI

Table 1 Baseline demographic and clinical characteristics

ID	Age (years)/ gender	Systemic pathologies	Ocular clinical diagnosis	Time from symptom onset (months)	Prior ocular treatment	Healthy donor	FU (months)
1	34/F	Juvenile rheumatoid arthritis under immunosuppressive treatment, DM	Severe DED with necrotizing scleritis	45	AT, DEX, CSA, auto-PRGF	Mother	30
2	90/F	Rosacea, severe sleep apnea syndrome	Severe DED, drug-induced CC	45	AT, COG, HCT, SCL, insulin	Daughter	30
3	85/F	Sjögren's syndrome, chronic anemia	Severe DED, terminal glaucoma	36	AT, FML, AHT, CSA	Son	30
4	53/M	GvHD post-BMT for leukemia, Factor V Leiden-positive, hypothyroidism	Severe DED with PED, oGvHD	12	AT, COG, DEX, VAO, CSA, AS	Daughter	24
5	64/M	Amyloidosis, thrombocytopenia	NK with PED	24	AT, COG, HCT, CSA, AS	Granddaughter	24
6	54/M	Post-transfusion hepatitis C infection	NK with PED, post-PK (× 3)	36	AT, DP, HCT, SCL, AS, AMT	Brother	24
7	22/M	SJS, autoimmune thrombocytopenia	CC and PEE	40	AT, COG, DEX, CSA	Mother	24
8	11/F	Linear IgA dermatosis, celiac disease	CC and PEE	23	AT, COG, DEX, CSA, insulin	Mother	18
9	79/F	Hemophilia C, MMP	Ocular cicatricial pemphigoid, severe DED	40	AT, COG, DEX, CSA	Daughter	18
10	82/M	Rosacea, lichen planus	Severe DED and MGD	65	AT, HCT, AZT, CSA, auto-PRGF	Son	15
11	11/F	GvHD post-BMT for leukemia	Severe DED, moderate MGD, oGvHD	62	AT, HC, CSA	Mother	12
12	43/M	GvHD post-BMT for leukemia, hepatitis C infection	Severe DED, monocular vision, oGvHD	48	AT, COG, DEX, CSA	Brother	12

Table 1 continued

ID	Age (years)/ gender	Systemic pathologies	Ocular clinical diagnosis	Time from symptom onset (months)	Prior ocular treatment	Healthy donor	FU (months)
13	70/M	DM, colon cancer under chemotherapy	NK with PED	40	AT, COG, FML, insulin	Daughter	12
14	61/M	Rosacea, thrombocytopenia, post-kidney transplant	Severe DED, moderate MGD	12	AT, FML, CSA	Wife	9
15	47/F	Darier–White disease, Gilbert syndrome, thalassemia minor	Severe DED, allergic conjunctivitis	12	AT, DEX, CSA	Husband	9
16	81/F	Rheumatoid arthritis under immunosuppressive treatment	Severe DED	12	AT, DEX, CSA	Daughter-in-law	9
17	58/M	GvHD post-BMT for leukemia, under chemotherapy	Severe MGD and DED, oGvHD	48	AT, FML, insulin, AS, CSA	Sister	9
18	50/M	Klinefelter syndrome, nephropathy, and autoimmune thrombocytopenia	Recurrent erosion syndrome	3	AT, COG, HCT, SCL, CSA	Daughter	6
19	48/F	Post-BMT for mycosis fungoides	Cicatricial MGD with PEE	36	AT, HC, CSA, auto-PRGF	Sister	6
20	58/F	Leukemia under chemotherapy	Severe DED, benign essential blepharospasm	36	AT, COG, DP, DEX, BT, AS	Son	6
21	45/M	Amyotrophic lateral sclerosis	Exposure keratopathy with PED	36	AT, COG, EP, insulin	Brother	6
22	65/F	GvHD post-BMT for leukemia, under chemotherapy	Severe DED, CC, oGvHD	6	AT, FML, AS	Husband	6
23	50/F	Breast cancer recurrence under chemotherapy and radiotherapy	Severe DED, moderate MGD, blepharospasm	12	AT, DEX, CSA, IPL	Husband	6
24	85/F	Colon cancer under chemotherapy	NK with PED, severe MGD, glaucoma, monocular vision, CC	6	AT, COG, AHT, FML, insulin, AS	Daughter	6

Table 1 continued

ID	Age (years)/ gender	Systemic pathologies	Ocular clinical diagnosis	Time from symptom onset (months)	Prior ocular treatment	Healthy donor	FU (months)
25	51/M	Rosacea, sarcoidosis with systemic activity	Severe DED, leukoma post-corneal perforation	8	AT, COG, DP, FML, CSA	Son-in-law	6
26	83/M	MMP under immunosuppressive treatment, rosacea	CC, severe MGD	12	AT, DEX, CSA, auto-PRGF	Son	6
27	5/M	Leigh syndrome, infantile epileptic spasms, severe encephalopathy, developmental delay	Recurrent erosion syndrome, allergic conjunctivitis	12	AT, COG, DP, FML, CSA	Mother	3
28	57/F	Hypothyroidism, epilepsy, end-stage chronic renal disease under hemodialysis, and anticoagulant medication	Severe MGD and DED, post-LASIK	10	AT, COG, DEX, CSA	Husband	3
29	70/F	Scleroderma, thrombocytopenia, auricular fibrillation, post-thyroidectomy, serum cholinesterase deficiency	Severe DED, allergic conjunctivitis	12	AT, COG, FML, CSA, insulin	Sister	3
30	49/M	Uncontrolled DM, polyneuropathy, IgA deficiency, psoriasis	NK with PED, moderate MGD	6	AT, COG, DP, AS	Sister-in-law	3

AMT amniotic membrane transplantation, *AT* artificial tears, *AS* autologous serum, *AHT* antihypertensives, *Auto-PRGF* autologous plasma-rich in growth factors, *BMT* bone marrow transplantation, *BT* botulinum toxin, *CC* cicatrizing conjunctivitis, *COG* carbomer ophthalmic gel, *CSA* cyclosporine, *DED* dry eye disease, *DEX* dexamethasone, *DM* diabetes mellitus, *DP* dexpanthenol gel, *EP* eye patch, *F* female, *FML* fluorometholone, *FU* follow-up, *GvHD* Graft-versus-host disease, *HCT* hydrocortisone, *IPL* intense pulsed light, *M* male, *MGD* meibomian gland dysfunction, *MMP* mucous membrane pemphigoid, *NK* neurotrophic keratopathy, *oGvHD* ocular GvHD, *PEE* punctate epithelial erosions, *PED* persistent epithelial defects, *PK* penetrating keratoplasty, *SJS* Stevens–Johnson syndrome, *VAO* vitamin A ointment

score reduced significantly to 32.8 ± 6 (range: 15–45)($p < 0.001$, Cohen's d: 1.32, large effect size), and the number of patients with severe symptoms reduced to 17 cases (58.6%). When patients were asked about the frequency and severity of the symptoms of dryness and/or irritation compared with the pretreatment state using the SANDE questionnaire, a mean reduction of 31.2 ± 16.4 in frequency ($p < 0.001$, Cohen's d: 2.07, large effect size) and 32.1 ± 14.5 in severity ($p < 0.001$, Cohen's d: 2.00, large effect size) was reported (Table 2). Out of 12 cases with PED, 8 (67%) showed complete resolution, while 4 exhibited partial improvement. No worsening of PED was observed; those with partial healing were moved to amniotic membrane combined with is-mPRGF transplantation. No AE were reported. Neither of these patients had infections or microbial keratitis after initiating is-ePRGF treatment.

Secondary Outcomes

At the last follow-up, the multilevel mixed-effects linear regression demonstrated a statistically significant improvement in BCVA from baseline values ($p = 0.010$, Cohen's d: –0.27, moderate effect size), while IOP remained stable ($p = 0.132$) (Fig. 2). During the follow-up, one patient reported vision loss in her left eye after

Table 2 Clinical parameters before and after allogeneic is-ePRGF treatment

	Before is-ePRGF (mean ± SD)	After is-ePRGF (mean ± SD)	P^*	Cohen's $d^\dagger$
Symptoms				
OSDI (score)	42.9 ± 9 $n = 29$	32.8 ± 6 $n = 29$	**< 0.001**	**1.32**
SANDE—frequency (score)	79 ± 13 $n = 29$	47.7 ± 17 $n = 29$	**< 0.001**	**2.07**
SANDE—severity (score)	76.9 ± 15 $n = 29$	44.8 ± 17 $n = 29$	**< 0.001**	**2.00**
Signs				
BCVA (decimal scale)	0.52 ± 0.33 $n = 56$	0.61 ± 0.33 $n = 56$	**0.010**	−0.27
IOP (mmHg)	12.5 ± 2.5 $n = 58$	12 ± 2.5 $n = 58$	0.132	0.2
BUT (s)	2.8 ± 1.9 $n = 50$	4.7 ± 1.7 $n = 50$	**< 0.001**	**−1.05**
Schirmer test (mm)	4.2 ± 2 $n = 52$	5.6 ± 2.5 $n = 52$	**< 0.001**	−0.62

BCVA best-corrected visual acuity, *BUT* break-up-time, *IOP* intraocular pressure, *is-ePRGF* immunosafe plasma rich in growth factors eye drops, *OSDI* Ocular Surface Index, *SANDE* Symptom Assessment iN Dry Eye, *SD* standard deviation

[*] Paired t-test or Welch's t-test for patient-level comparisons and multilevel mixed-effect linear regression for eye-level data. Bold values denote statistical significance

[†] Size effect; bold values denote large effect

her third chemotherapy session; she developed macular edema associated with branch retinal vein occlusion. In addition, Schirmer test ($p<0.001$, Cohen's d: –0.62, moderate effect size) and BUT ($p<0.001$, Cohen's d: –1.05, large effect size) values significantly increased after allogeneic is-ePRGF treatment (Table 2).

Furthermore, the mixed-effects ordinal logistic model showed a significant improvement after treatment across conjunctival bulbar redness, fluorescein-stained area and density, and MGD, after adjusting for eye-level and patient-level correlation (Fig. 3). Before treatment, most cases (96%) exhibited grade 2–3 conjunctival ocular redness, which decreased to 7% (four cases) at the last follow-up. Regarding PEE assessed by fluorescein staining, the proportion of cases with severe staining (grade 3) fell from 48% to 0% in area and from 34% to 0% in density. At baseline, 52% and 21% of the cases showed moderate and severe MGD, respectively; after treatment, these figures reduced to 21% and 17%, respectively (Fig. 4).

DISCUSSION

Allogeneic platelet concentrates have long been used in transfusion medicine to prevent and correct bleeding disorders; as such, they are recognized by the World Health Organization as essential medicines for adults and children [33]. Over recent years, interest has sharply grown in evaluating their properties in the field of ophthalmology [34]. Herein, we report that

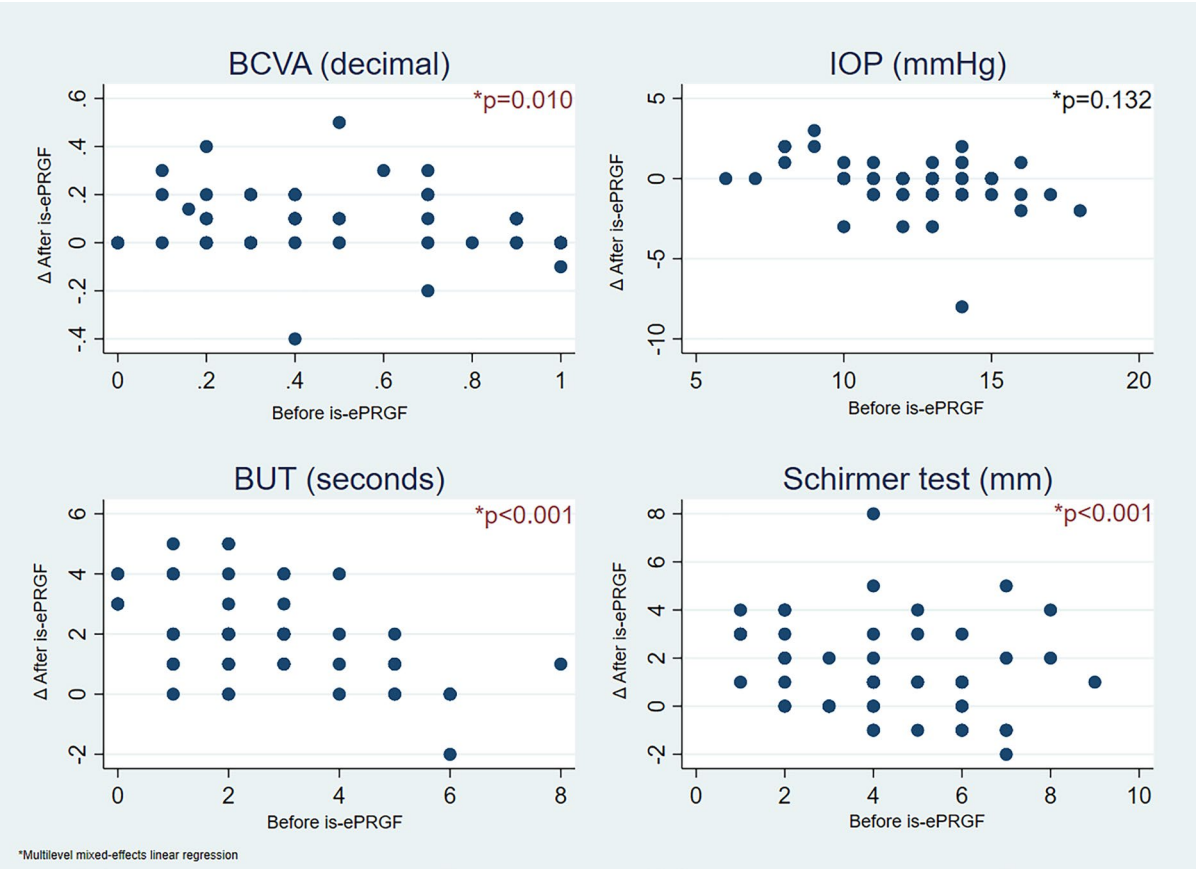

Fig. 2 Paired comparisons between baselines values and changes (Δ) in BCVA, IOP, BUT, and Schirmer test following allogeneic is-ePRGF treatment. *BCVA* best-corrected visual acuity, *IOP* intraocular pressure, *BUT* break-up time, *is-ePRGF* immunosafe plasma rich in growth factors eye drops

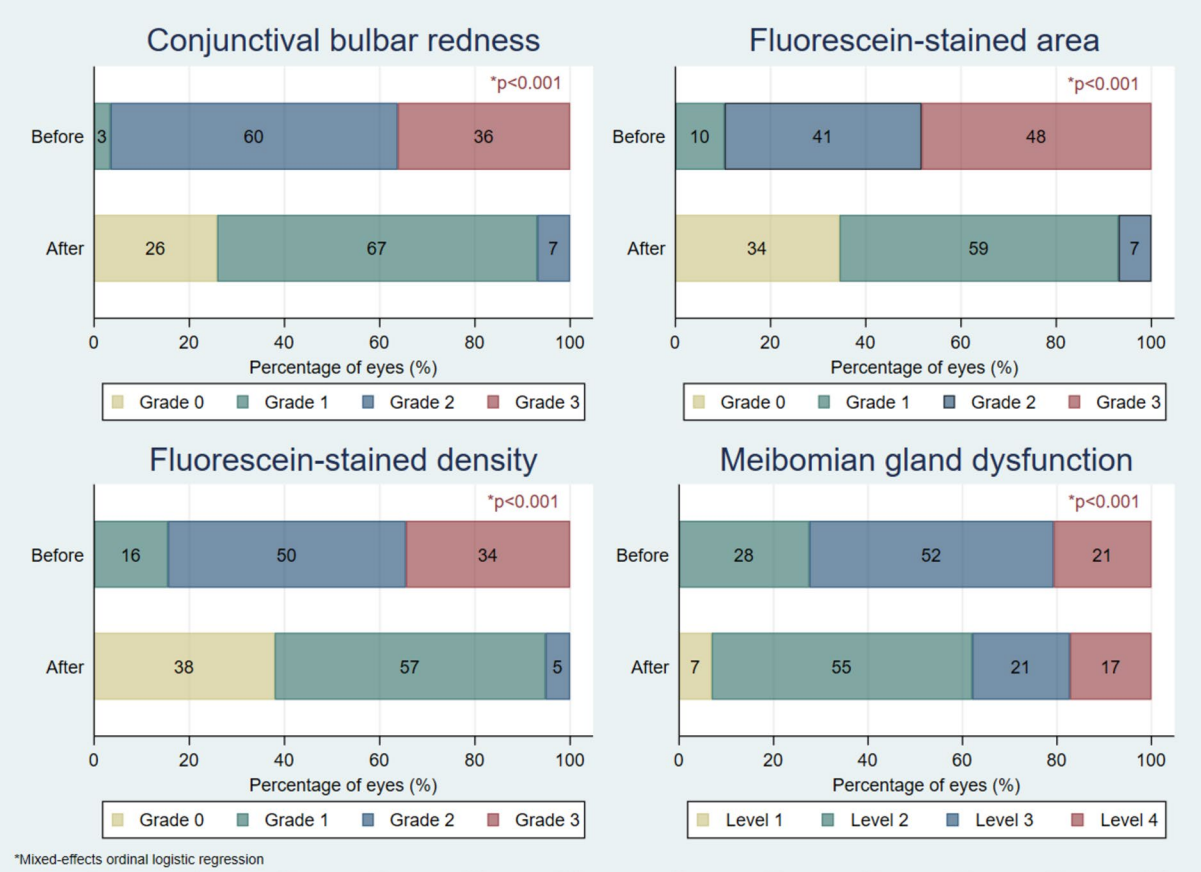

Fig. 3 Changes in clinical grading scores following allogeneic immunosafe plasma rich in growth factors eye drops treatment

allogeneic is-ePRGF was well tolerated and effective in reducing the frequency and severity of symptoms as well as in improving BCVA, conjunctival redness, corneal staining, MGD, BUT, and Shirmer test values in refractory OSD cases.

Platelets are best known for their vital role in maintaining blood hemostasis; however, they also perform essential functions in innate and adaptive immunity as well as in wound healing and tissue repair mechanisms that could meet needs in cell therapy, regenerative medicine, and targeted drug delivery [35]. Autologous platelet concentrates, such as is-ePRGF, have shown successful outcomes as regenerative therapy for various ocular disorders [11] in both preclinical and clinical studies [27].

However, certain circumstances, such as frail patients with severe, debilitating systemic diseases or hematological disorders, may prevent the patients' own blood extraction [21]. In our cohort, some patients reported blood clotting disorders, anemia, and low platelet counts or being on anticoagulant medication; these conditions, along with poor venous access, severe encephalopathy, and the need for frequent extractions due to chronic conditions, limited the autologous treatment available to our patients. Moreover, a subset of OSD characterized by a persistent inflammatory reaction and poor response to conventional treatment has been recognized as being associated with a systemic inflammatory influx [4]. These patients may have higher concentrations of proinflammatory cytokines or cytotoxic drugs in their bloodstream, which may affect the efficacy and safety of the autologous preparation [16–21]. In our series, most patients had autoimmune and inflammatory diseases on immunosuppressive treatment; in addition, 40% of our patients had already been treated with autologous therapy without response.

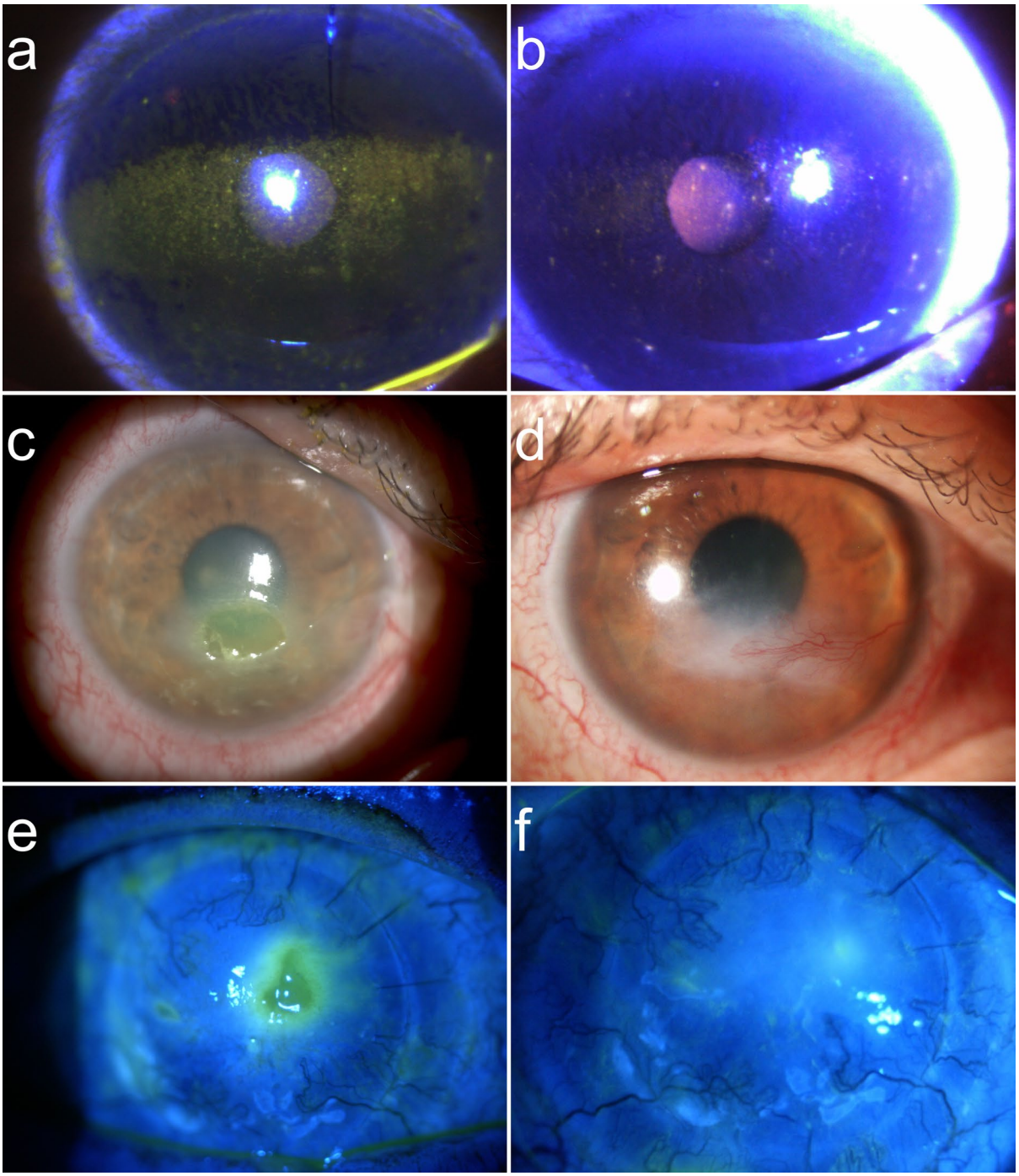

Fig. 4 Representative slit-lamp clinical photographs of patients before (**a**, **c**, **e**) and after (**b**, **d**, **f**) treatment with allogeneic immunosafe plasma rich in growth factors eye drops. The first case (**a**, **b**) shows a marked reduction in corneal fluorescein staining, while the second (**c**, **d**) and third (**e**, **f**) cases demonstrated resolution of persisted corneal epithelial defects

The lack of response to previous treatments may be attributed to the underlying systemic inflammatory state. Prior studies have demonstrated that elevated levels of IL-1β, IL-6, IL-10, and VEGF, along with reduced IL-2, are associated with persistent inflammation in patients with inflammatory OSD [17]. Furthermore, Hwang et al. [16] reported significantly higher serum concentrations of proinflammatory cytokines—such as TNF-α, IL-1β, IL-6, and IL-8—in patients with secondary Sjögren's syndrome. The authors hypothesized that these altered serum cytokine profiles may adversely affect the ocular surface in patients using AS. Similarly, Chmielewska et al. [17] found that patients treated with serum eye drops containing high cytokine levels experienced adverse effects, including a foreign body sensation, decreased visual acuity, and reduced ocular lubrication. It is also noteworthy that previous studies have detected orally administered immunosuppressants in AS at clinically relevant concentrations, which may significantly influence both the efficacy and tolerability of these topical therapies [18, 19]; in this context, Janus et al. [23] reported that, in patients with Sjögren's syndrome, treatment outcomes were more favorable with allogeneic AS compared with autologous AS.

Interestingly, nearly one quarter of our "nonresponsive" patients (7, 23%) were using insulin eye drops. Although several studies have reported topical insulin as an effective adjuvant therapy for promoting epithelial wound healing—particularly in refractory cases—the exact mechanism of action remains unclear. Our research group previously evaluated the biological potential of PRGF eye drops in comparison with AS and insulin. The results showed that PRGF induced a significantly higher proliferation rate in human conjunctival fibroblasts and human keratocytes compared with AS or insulin. Moreover, PRGF treatment led to a significantly greater reduction in wound area and markedly decreased the number of myofibroblast-differentiated cells at both concentrations tested [13].

The beneficial effects of PRGF are well documented (Table 3). In our cases, allogeneic is-ePRGF treatment has shown successful clinical outcomes similar to those previously reported with autologous source. Clinical studies on autologous therapy have assessed a wide range of OSD, including DED, different stages of NK, cicatrizing conjunctivitis (Stevens–Johnson syndrome, ocular cicatricial pemphigoid), post-refractive surgery epithelial defects, graft-versus-host disease, Sjögren's syndrome, and keratoneuralgia, reporting significant improvement in symptoms and ocular surface parameters [9, 10, 12, 18, 30, 31, 36–59]. A recent systematic review [60] analyzed 1168 eyes under autologous PRGF treatment and found eight minor AE (0.68%), including self-resolving eye irritation, eye redness, burning/itching, and eyelid inflammation; in contrast, we did not find major or minor AE during the follow-up in our cases.

As far as we know, eight clinical studies [61–68] have been published evaluating allogeneic platelet concentrates in OSD cases (Table 4). Five of these studies addressed umbilical cord blood-based eye drops, such as platelet lysate [64–66, 68] and PRP [67], which are prepared from umbilical cord units donated for hematopoietic stem cell transplantation that are not suitable for this use. In addition, three studies reported the use of PRP as eye drops [61, 63] or a clot [62], which were prepared by blood banks from whole blood bags of periodic blood donations [62] or individual donors with the same blood type as the patient [61]. At our institution, the is-ePRGF treatment was prepared and picked up at the institutional pharmacy under a compassionate use protocol. In our cases, blood was collected from the patients' family members and tested for transmissible diseases. Our encouraging results with allogeneic is-ePRGF are supported by those previous studies testing different allogeneic platelet concentrate eye drops.

However, some concerns arise from using allogeneic sources, mainly focused on the risks of alloimmunization, immunogenicity, and transmission of infectious diseases. Since the ocular surface bears ABO antigens and some human leukocyte antigens (HLA) [69], plasma contains antibodies and complement, and platelets have a relatively higher number of HLA-I antigens on their surface compared with erythrocytes and granulocytes [70], there is a theoretical risk of type II hypersensitivity reaction. However,

Table 3 Clinical studies addressing ocular surface diseases using plasma rich in growth factors therapy

Author, year	Country	Design	*n* eyes	*n* patients	Diagnosis	Intervention regimen	Comparator	Results
Medical treatment								
Lozano-Sanroma, 2024[36]	Spain	PT	23	12	DED, congenital aniridia	is-ePRGF QID × 6 m	None	Improvements in symptoms, ocular redness, and ocular damage. No adverse effects (AE)
Ghalibafan, 2023[37]	USA	RT	26	15	NK stage I	ePRGF 4–6 ×/d × 2 m	None	Improvement in CS, corneal staining, BCVA, BUT, and Schirmer test, and reduction in MMP-9 levels. No AE
Lozano-Sanroma, 2023[31]	Spain	RT	102	52	DED	ePRGF QID × 3 m	CT	Improvement in symptoms, ocular redness, and BUT. No AE
Barros, 2023[38]	Spain	RT	83	83	DED	ePRGF QID × 3 m	CT	The length, number of branches, nerve density, and BUT values significantly increased
Wang, 2022[39]	USA	RT	16	16	Keratoneuralgia	ePRGF QID × 3 m	None	Improvement in symptoms. No AE

Table 3 continued

Author, year	Country	Design	*n* eyes	*n* patients	Diagnosis	Intervention regimen	Comparator	Results
Soifer, 2022[12]	USA	RT	153	153	OSD	ePRGF 4–6 ×/d × 3 m	None	Improvement in symptoms and corneal staining. Ocular burning sensation (1 case), progressive corneal thinning (2 cases), elective tarsorrhaphies (2 cases)
Wang, 2022[40]	USA	RT	42	26	OSD	ePRGF QID × 3 m + SL	None	Decreases in symptoms and the number of concurrent treatments. No AE
Sánchez-González, 2021[41]	Spain	RT	48	24	Post-TPRK surgery	ePRGF TID × 6 w + CL until complete healing	None	Reduction in pain score on day 3 and day 7. Mean time to healing of 2.5 ± 1.2 d. No AE
Sánchez-Ávila, 2021[42]	Spain	RT	117	69	Post-PRK surgery	ePRGF QID × 6 w + CL until complete healing	MMC	No differences in efficacy, safety, and predictability indexes between groups. Hyperemia, eye pain, superficial keratitis with MMC. No AE with ePRGF
Sánchez-Ávila, 2021[43]	Spain	RT	79	74	Noninfectious corneal ulcer	ePRGF QID × 6 w + Cacicol®QOD until healing	None	Complete healing in 96%. Improvement in BCVA and symptoms, while IOP remained unchanged

Table 3 continued

Author, year	Country	Design	*n* eyes	*n* patients	Diagnosis	Intervention regimen	Comparator	Results
Gea-Navarrete, 2021[44]	Spain	RT	24	24	DED	ePRGF regimen not specified	None	Improvement in 75% of cases, satisfaction in 100%. Improvement after switching in 86%
de la Sen-Corcuera, 2020[45]	Spain	RT	10	6	CC	Phase I: is-ePRGF QID × 1 m. Phase II: + SCI ePRGF/m, Phase III: + surgery	None	Phases I (20%), II (70%), and III (20%). Reduction in clinical inflammation and IOP
Sánchez-Ávila, 2018[46]	Spain	RT	77	42	Post-LASIK surgery	is-ePRGF QID × 6 w	CT	Improvement in symptoms, IOP value, BUT, and Schirmer test with is-ePRGF. Improvement in BUT with CT
Sánchez-Ávila, 2018[47]	Spain	RT	8	6	OSD in patients with glaucoma	is-ePRGF QID × 6 w	None	Complete healing in 14.5 ± 5.5 w. Improvement in symptoms, IOP, and BCVA. Itching (1 case)
Sánchez-Ávila, 2018[48]	Spain	RT	38	31	NK stage II and III	ePRGF QID × 6 w	None	Improvement in symptoms and BCVA. Complete healing in 97% in 11.4 ± 13.7 w. Itching (1 case)

Table 3 continued

Author, year	Country	Design	*n* eyes	*n* patients	Diagnosis	Intervention regimen	Comparator	Results
Sánchez-Ávila, 2018[30]	Spain	RT	23	12	GvHD	is-ePRGF QID × 6 w	None	Improvement in area and density of corneal staining, BCVA, BUT, and Schirmer test. No AE
Sánchez-Ávila, 2017[18]	Spain	RT	52	26	SS	is-ePRGF QID × 6 w	None	Improvement in symptoms, BCVA. Mild eye irritation (2 cases)
Guarnieri, 2017[49]	Spain	CR	1	1	Corneal melting after erlotinib	ePRGF QID × 12 m	None	Clinical improvement, corneal integrity was maintained. ePRGF prevented an imminent corneal perforation
Sánchez-Ávila, 2016[50]	Spain	CR	1	1	NK stage III	ePRGF QID × 3 m + Cacicol® q/m × 5 m	None	Complete healing, improvement in symptoms, VA
Merayo-Lloves, 2016[51]	Spain	RT	156	83	Evaporative DED	is-ePRGF QID × 6 w	None	Improvement in symptoms, Schirmer test. Mild itching (2 cases), dizziness (2 cases); all resolved
Merayo-Lloves, 2015[9]	Spain	RT	80	41	OSD	is-ePRGF QID × 6 w	None	Improvement in symptoms and BCVA. Eye redness (1 case), eyelid inflammation (1 case)

Table 3 continued

Author, year	Country	Design	*n* eyes	*n* patients	Diagnosis	Intervention regimen	Comparator	Results
Vendreil, 2015[52]	Spain	RT	14	7	Severe DED	ePRGF 5 ×/d × 3 m	None	Improvement in symptoms and ocular surface staining
Valdez-Payan, 2014[53]	Mexico	PT	40	20	DED	NR	AS + AT or AT alone	All groups showed clinical improvements and were better with ePRGF
López-Plandolit, 2011[54]	Spain	PT	16	16	DED	20% ePRGF QID × 3 m	None	Reduction in symptoms
López-Plandolit, 2010[55]	Spain	PT	20	18	PED	50% ePRGF q2h × 3 d and then prn	None	Complete healing (85%) in 10.9 w. Additional medical or surgical medication was required in 80%
Rocha, 2007[56]	Spain	CR	1	1	Post-LASIK Flap necrosis	ePRGF q2h, tapering × 6 w	None	Progressive healing, no change in BCVA
Surgical treatment								
Rahhal-Ortuño, 2021[57]	Spain	CR	1	1	Corneal dellen	mPRGF + ePRGF QID × 2 m	None	Complete dellen resolution at 1 m
Sabater, 2021[10]	USA	RT	15	14	OSD	mPRGF w/wo AMT + ePRGF QID × 2 m	None	The mPRGF dissolution in 21 ± 3 d. Improvement/maintenance of VA (93%), corneal healing (83%). Severe LSCD and graft failure (1 case). No AE

Table 3 continued

Author, year	Country	Design	*n* eyes	*n* patients	Diagnosis	Intervention regimen	Comparator	Results
Idoipe, 2021[58]	Spain	PT	49	NR	Primary pterygium	mPRGF + ePRGF QID x 1 m	CAG or AMT	Lower recurrence with CAG. No differences in BCVA and IOP. Improvement in symptoms in all groups. No AE
Sánchez-Ávila, 2018[59]	Spain	RT	15	15	OSD	mPRGF w/wo AMT + is-ePRGF QID × 6 w	None	Inflammation control in 2.5 ± 1.1 m. Complete healing in 87% in 2.9 ± 1.2 m. Improvement in BCVA. No AE

AMT amniotic membrane transplantation, *AS* autologous serum, *AT* artificial tears, *BCVA* best-corrected visual acuity, *BUT* break-up time, *CAG* conjunctival autograft, *CC* cicatrizing conjunctivitis, *CL* contact lens, *CT* conventional treatment, *CR* case report, *CS* corneal sensitivity, *d* day(s), *DED* dry eye disease, *ePRGF* PRGF eye drops, *F* female, *GF* growth factors, *GvHD* graft-versus-host disease, *IOP* intraocular pressure, *is* immunosafe, *LASIK* laser-assisted keratomileusis, *LSCD* limbal stem cell deficiency, *M* male, *m* month(s), *MMC* mitomycin C, *mPRGF* fibrin membrane PRGF, *NK* neurotrophic keratitis, *NR* not reported, *OSD* ocular surface disease, *PED* persistent epithelial defect, *PRGF* plasma rich in growth factors, *prn* "pro re nata" = as needed, *PRK* Photorefractive Keratectomy, *PT* prospective, *q* every, *QID* four times a day, *QOD* every other day, *RT* retrospective, *RCT* randomized clinical trial, *SL* scleral contact lenses, *SD* standard deviation, *SS* Sjögren's syndrome, *SCI* subconjunctival injection, *TID* three times a day, *TPRK* Transepithelial Photorefractive Keratectomy, *VA* visual acuity, *w* week(s), *w/wo* with/without, *y* years

Table 4 Clinical studies addressing ocular surface diseases using allogeneic platelet concentrates therapy

Author, year	Country/setting	Design	*n* eyes/*n* patients	Diagnosis	Donor/matching	Allogeneic intervention	FU	Results
Peripheral blood								
Mancini, 2023 [61]	Italy/blood bank	RT	6/3	DED (SS)	Not specified/ABO matching	ePRP, 6 ×/d × 3 m	6 m	Improvement in symptoms, visual acuity, corneal sensitivity, and corneal staining
Romano, 2022 [62]	Italy/blood bank	RT	4/4	NK and DED	Unrelated/not specified	cPRP + fibrin glue + SCL	4 m	Complete healing (100%). Improvement in symptoms, conjunctival hyperemia, and corneal staining. No variations in visual acuity, corneal sensitivity, and Schirmer test
Passilongo, 2019*[63]	Italy/blood bank	RT	62/31	DED	Not specified/not specified	ePRP versus 25% serum	2 m	Higher improvement in symptoms and corneal staining with allogeneic PRP than with allogeneic serum

Table 4 continued

Author, year	Country/setting	Design	*n* eyes/*n* patients	Diagnosis	Donor/matching	Allogeneic intervention	FU	Results
Umbilical cord blood								
Foti, 2024 [64]	Italy/CB bank	PT	98/49	SS, systemic sclerosis, oGvHD, NK, SJS	Unrelated/no matching	ePL 6 ×/d	70 d	Improvement in symptoms, Schirmer test, BUT, BCVA, corneal staining, and meibography. Pain and inflammation were markedly reduced. No AE
Gagliano, 2024 [65]	Italy/CB bank	PT	44/22	oGvHD	Unrelated/no matching	ePL 6 ×/d	3 m	Improvement in symptoms, Schirmer test, BUT, BCVA, corneal staining, and meibography. Pain and inflammation were markedly reduced. No AE
Kamel Farag, 2023 [66]	Egypt/CB bank	PT	40/40	Persistent corneal ulcers (DED, post-keratoplasty, CCB, post-infection)	Unrelated/no matching	ePL 4–6 ×/d	9.5–14.5 d	Full (50–71%) and partial ulcer recovery (12–50%). Improvement in symptoms. No AE

Table 4 continued

Author, year	Country/setting	Design	*n* eyes/*n* patients	Diagnosis	Donor/matching	Allogeneic intervention	FU	Results
Wong, 2023 [67]	Singapore/CB bank	PT	40/40	Recalcitrant DED	Unrelated/no matching	30% ePPP 1–3 ×/d	5.5 ± 1.6 m	Improvement in symptoms, corneal staining, and BUT. No change in visual acuity or Schirmer test. No AE
Samarkanova, 2021 [68]	Spain/CB bank	RT	46/33	NK, corneal ulcers, CCB, oGvHD, DED	Unrelated/no matching	ePL 4–6 ×/d	19–96 d	Full (78%) and partial ulcer recovery (19%). Improvement in symptoms and corneal clarity. No AE

AE adverse events, *BCVA* best-corrected visual acuity, *BUT* break-up time, *CB* cord blood, *CCB* corneal chemical burn, *cPRP* PRP clot, *d* day(s), *DED* dry eye disease, *ePRP* PRP eye drops, *ePL* PL eye drops, *FU* follow-up, *m* month(s), *oGvHD* ocular GvHD, *RT* retrospective, *PL* platelet lysate, *PPP* platelet-poor plasma *PRP* platelet-rich plasma, *PT* prospective, *SCL* soft contact lens, *SS* Sjögren's syndrome, *SJS* Stevens–Johnson syndrome. *This reference corresponds to a 2019 ARVO Annual Meeting Abstract and a XVIII SICSSO presentation, no publication-related full-text was found

some authors have reported successful and safe treatment with ABO-unselect allogeneic serum from peripheral blood [71–73] or from UCB [74], and even with finger-prick allogenic blood [75]. Moreover, ABO-matching is unnecessary for corneal transplants [76] or UCB transplantation [77]. In our patients, ABO-matching was not considered, and no AE was observed. In this context, to avoid putative side effects, our standardized production of is-ePRGF includes a heat treatment that has been shown to reduce the complement system activity and IgE concentrations without reducing the biological properties [8, 28].

To minimize the risk of disease transmission, testing donors and the final blood-based product is strongly recommended [78]. In this study, we included healthy donors with no history of blood transfusion. They were asked to complete a blood donor health questionnaire and undergo testing for blood-borne diseases. Moreover, in our standardized protocol, we followed a closed-system technique, and all procedures were performed under a biohazard laminar flow hood; during the final steps, a sample for microbiological control was taken. Therefore, we implemented a control system to ensure that the product was only used when the results of the microbiological and serological tests were clear. In this study, we chose to prepare the product from relatives rather than volunteer donors for two main reasons. First, feasibility—as demonstrated in previously published studies evaluating allogeneic serum tears [71–73, 75], and second, the preparation was conducted within our institution under standardized and approved protocols, rather than at a transfusion center or blood bank, where blood collection programs are typically designed for volunteer donors.

Blood transfusion has gained acceptance as a safe and effective treatment, making the use of topical allogeneic blood-based eye drops both ethical and legal [71, 72]. Nevertheless, implementation depends on the regulatory guidelines of each country and varies among multiple centers globally. For instance, in Europe, three countries classify blood-based eye drops as Advanced Therapy Medicinal Products (ATMPs), two as non-ATMPs, seven as blood products, and eight with no specific regulation for them [78]. Moreover, the coronavirus disease 2019 (COVID-19) outbreak has adversely affected blood bank supplies and autologous preparations, leading many centers to transition to allogeneic therapy, which offers advantages such as improved standardization for clinical consistency, more efficient production, and enhanced safety control [74, 78]. Furthermore, the use of allogeneic is-ePRGF may expand access to this regenerative therapy for not only complex clinical cases but also for a broader patient population, which can significantly contribute to the achievement of many Sustainable Development Goals (SDG) adopted by the United Nations [79]. This development paves the way for creating tailored products for specific conditions, ensuring robust quality control [78].

While the results are encouraging, we must recognize the limitations of this study. The non-comparative design, the unobserved confounders, and the relatively small sample size limit the generalizability of the findings. As an observational study, we cannot entirely exclude the influence of ongoing concomitant treatments. Although is-ePRGF was the only new intervention introduced, the improvements observed should be interpreted as real-world outcomes and not definitive proof of causality. In addition, other imaging software tools—such as ImageJ—could enhance the precision of measurements in cases involving PED and may be considered for future projects. Nevertheless, unlike clinical trials, which typically have strict participant inclusion and exclusion criteria and may not reflect the general population, we included a variety of underlying etiologies refractory to conventional treatments that may be more representative of everyday clinical practice. This is the first study to report on the use of allogeneic is-ePRGF in refractory OSD and further contributes to a body of knowledge.

CONCLUSIONS

In this cohort, allogeneic is-ePRGF was a safe and effective option for refractory OSD when autologous sources were not available or suitable. The use of allogeneic source from selected

donors could have better biological properties and may broaden access to is-ePRGF therapy; however, harmonization in blood-based therapy regulation is warranted.

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Data Availability. The datasets used and/or analyzed during the current study are available from the corresponding author upon reasonable request.

Declarations

Conflicts of Interest. The authors declare that Eduardo Anitua is the Scientific Director, and Ronald M. Sánchez-Ávila is a scientist at BTI Biotechnology Institute, a biomedical company that investigates the fields of regenerative medicine and PRGF-Endoret Technology. The other authors declare no conflicts of interest in developing this study.

Ethical Approval. The protocol was approved by the Ethics Committees for Investigation with Medicinal Products of the Principality of Asturias (CEImPA 2023.281), and the procedures were conducted in adherence with the tenets of the Declaration of Helsinki. Written informed consents were obtained from every patient and donor before the procedure as routine clinical practice, including permission to use their medical records for scientific purposes. Authorization was granted by the Spanish Agency for Medicines and Health Products (AEMPS) to initiate allogeneic is-ePRGF as compassionate treatment for every enrolled patient.

REFERENCES

1. Stapleton F, Argüeso P, Asbell P, et al. TFOS DEWS III digest report. Am J Ophthalmol. 2025;S0002–9394(25):00276–84.
2. Bron AJ, de Paiva CS, Chauhan SK, et al. TFOS DEWS II pathophysiology report. Ocul Surf. 2017;15(3):438–510. https://doi.org/10.1016/j.jtos.2017.05.011.
3. Soifer M, Azar NS, Mousa HM, Perez VL. Ocular surface inflammatory disorders (OSID): a collective of systemic etiologies which cause or amplify dry eye syndrome. Front Med (Lausanne). 2022;6(9): 949202.
4. Jones L, Craig JP, Markoulli M, et al. TFOS DEWS III management and therapy report. Am J Ophthalmol. 2025;S0002–9394(25):00274.
5. Giannaccare G, Versura P, Buzzi M, Primavera L, Pellegrini M, Campos EC. Blood derived eye drops for the treatment of cornea and ocular surface diseases. Transfus Apher Sci. 2017;56(4):595–604.
6. Zhang Y, Li N, Ge Z, Li F. Blood component therapy for dry eye disease: a systematic review and network meta-analysis. Front Med (Lausanne). 2024;16(11):1500160.
7. Anitua E, Sanchez M, Merayo-Lloves J, De la Fuente M, Muruzabal F, Orive G. Plasma rich in growth factors (PRGF-Endoret) stimulates proliferation and migration of primary keratocytes and conjunctival fibroblasts and inhibits and reverts TGF-beta1-Induced myodifferentiation. Invest Ophthalmol Vis Sci. 2011;52(9):6066–73.
8. Anitua E, Muruzabal F, De la Fuente M, Merayo-Lloves J, Orive G. Effects of heat-treatment on plasma rich in growth factors-derived autologous eye drop. Exp Eye Res. 2014;119:27–34.
9. Merayo-Lloves J, Sanchez RM, Riestra AC, et al. Autologous plasma rich in growth factors eyedrops in refractory cases of ocular surface disorders. Ophthalmic Res. 2015;55(2):53–61.
10. Sabater AL, Mousa HM, Quinones X, et al. Use of autologous plasma rich in growth factors fibrin membrane in the surgical management of ocular surface diseases. Int Ophthalmol. 2021;41(7):2347–58.
11. Anitua E, de la Sen-Corcuera B, Orive G, et al. Progress in the use of plasma rich in growth factors in ophthalmology: from ocular surface to ocular fundus. Expert Opin Biol Ther. 2022;22(1):31–45.
12. Soifer M, Tovar A, Wang M, et al. A multicenter report of the use of plasma rich in growth factors (PRGF) for the treatment of patients with ocular surface diseases in North America. Ocul Surf. 2022;25:40–8.
13. Anitua E, de la Fuente M, Sánchez-Ávila RM, de la Sen-Corcuera B, Merayo-Lloves J, Muruzábal F. Beneficial effects of plasma rich in growth factors (PRGF) versus autologous serum and topical insulin in ocular surface cells. Curr Eye Res. 2023;48(5):456–64.
14. Anitua E, Muruzabal F, Pino A, Prado R, Azkargorta M, Elortza F, Merayo-Lloves J. Proteomic characterization of plasma rich in growth factors and undiluted autologous serum. Int J Mol Sci. 2021;22(22):12176.
15. Harritshøj LH, Nielsen C, Ullum H, Hansen MB, Julian HO. Ready-made allogeneic ABO-specific serum eye drops: production from regular male blood donors, clinical routine, safety and efficacy. Acta Ophthalmol. 2014;92(8):783–6.
16. Hwang J, Chung SH, Jeon S, Kwok SK, Park SH, Kim MS. Comparison of clinical efficacies of autologous serum eye drops in patients with primary and secondary Sjögren syndrome. Cornea. 2014;33(7):663–7.
17. Chmielewska K, Janus J, Mikołowska A, Wrzodak K, Stącel M, Antoniewicz-Papis J. Correlation between serum cytokine levels and the effect of allogeneic serum-based eye drops. Transfus Apher Sci. 2024;63(3): 103912.
18. Steele WH, Stuart JF, Whiting B, et al. Serum, tear and salivary concentrations of methotrexate in man. Br J Clin Pharmacol. 1979;7(2):207–11.
19. Tahmaz V, Wiesen MHJ, Gehlsen U, et al. Detection of systemic immunosuppressants in autologous serum eye drops (ASED) in patients with severe chronic ocular graft versus host disease. Graefes Arch Clin Exp Ophthalmol. 2021;259(1):121–8.

20. Harloff S, Hartwig D, Kasper K, Wedel T, Müller M, Geerling G. Epitheliotrophe Kapazität von Serum-Augentropfen gesunder versus immunsupprimierter Patienten mit rheumatoider Arthritis [Epitheliotrophic capacity of serum eye drops from healthy donors versus serum from immunosuppressed patients with rheumatoid arthritis]. Klin Monbl Augenheilkd. 2008;225(3):200–6.

21. Badami KG, McKellar M. Allogeneic serum eye drops: time these became the norm? Br J Ophthalmol. 2012;96(8):1151–2.

22. Stenwall PA, Bergström M, Seiron P, Sellberg F, Olsson T, Knutson F, Berglund D. Improving the anti-inflammatory effect of serum eye drops using allogeneic serum permissive for regulatory T cell induction. Acta Ophthalmol. 2015;93(7):654–7.

23. Janus J, Chmielewska K, Antoniewicz-Papis J. Allogeneic serum-based eye drops may give better results than autologous drops in Sjögren's syndrome dry eye. Transfus Apher Sci. 2024;63(5): 103991.

24. Dormann F, Hähnel V, Müller V, Burkhardt R, Offner R. Allogenic and autologous nondiluted serum eye drops-validation strategy compliant with good manufacturing practice. Transfus Med. 2024;34(4):319–25.

25. Foulks GN. Challenges and pitfalls in clinical trials of treatment for dry eye. Ocul Surf. 2003;1(1):20–30.

26. Alvarado R, Alfonso-Sanchez J, Merayo-Lloves J. Comparative efficacy and safety of allogeneic blood-based eye drops for ocular surface diseases: a systematic review of randomized clinical trials. Invest Ophthalmol Vis Sci. 2024;65(7):3612.

27. Anitua E, Muruzabal F, Tayebba A, et al. Autologous serum and plasma rich in growth factors in ophthalmology: preclinical and clinical studies. Acta Ophthalmol. 2015;93(8):e605–14.

28. Sanchez-Avila RM, Merayo-Lloves J, Riestra AC, et al. The effect of immunologically safe plasma rich in growth factor eye drops in patients with Sjögren syndrome. J Ocul Pharmacol Ther. 2017;33(5):391–9.

29. Wolffsohn JS, Arita R, Chalmers R, et al. TFOS DEWS II diagnostic methodology report. Ocul Surf. 2017;15(3):539–74.

30. Sanchez-Avila RM, Merayo-Lloves J, Muruzabal F, Orive G, Anitua E. Plasma rich in growth factors for the treatment of dry eye from patients with graft versus host diseases. Eur J Ophthalmol. 2020;30(1):94–103.

31. Lozano-Sanroma J, Barros A, Alcalde I, et al. Impact of plasma rich in growth factors (PRGF) eye drops on ocular redness and symptomatology in patients with dry eye disease. Medicina (Kaunas). 2023;59(5):928.

32. Tomlinson A, Bron AJ, Korb DR, et al. The International Workshop on Meibomian Gland Dysfunction: report of the Diagnosis Subcommittee. Invest Ophthalmol Vis Sci. 2011;52(4):2006–49.

33. WHO model lists of essential medicines 2023. https://www.who.int/publications/i/item/WHO-MHP-HPS-EML-2023.02. 2023

34. You J, Hodge C, Hoque M, Petsoglou C, Sutton G. Human platelets and derived products in treating ocular surface diseases—A systematic review. Clin Ophthalmol. 2020;12(14):3195–210.

35. Burnouf T, Chou ML, Lundy DJ, Chuang EY, Tseng CL, Goubran H. Expanding applications of allogeneic platelets, platelet lysates, and platelet extracellular vesicles in cell therapy, regenerative medicine, and targeted drug delivery. J Biomed Sci. 2023;30(1):79.

36. Lozano-Sanroma J, Barros A, Alcalde I, et al. Efficacy and safety of plasma rich in growth factor in patients with congenital aniridia and dry eye disease. Diseases. 2024;12(4):76.

37. Ghalibafan S, Osei KA, Amescua G, Sabater AL. Efficacy of plasma rich in growth factors (PRGF) in stage 1 neurotrophic keratitis. Eye. 2024;38:1390–1.

38. Barros A, Lozano-Sanroma J, Queiruga-Piñeiro J, et al. Recovery of corneal innervation after treatment in dry eye disease: a confocal microscopy study. J Clin Med. 2023;12(5):1841.

39. Wang M, Yennam S, Pflugfelder S. Initial experiences using plasma rich in growth factors to treat keratoneuralgia. Front Med (Lausanne). 2022;24(9): 946828.

40. Wang M, Yennam S, McMillin J, et al. Combined therapy of ocular surface disease with plasma rich in growth factors and scleral contact lenses. Ocul Surf. 2022;23:162–8.

41. Sánchez-González JM, Alonso-Aliste F, Borroni D, et al. Plasma rich in growth factors (PRGF) in transepithelial photorefractive keratectomy (TPRK). J Clin Med. 2021;10(9):1939.

42. Sanchez-Avila RM, Uribe-Badillo EE, Sanz JF, et al. Plasma rich in growth factors versus mitomycin C in photorefractive keratectomy. Medicine (Baltimore). 2021;100(3): e24139.

43. Sánchez-Ávila RM, Uribe-Badillo E, Fernández-Vega González C, et al. Use of plasma rich in growth factors and ReGeneraTing agent matrix for the treatment of corneal diseases. Vision (Basel). 2021;5(3):34.

44. Gea-Navarrete S, Montero-Hernández M, Gallego-Lago I, Santiuste Román A, Montesinos-Orti S, Soler-Company E. Resultados en salud y aspectos fármaco-económicos del tratamiento con PRGF-Endoret en ojo seco. Rev. OFIL.ILAPHAR. 2021; 31 (4): 357–360.

45. de la Sen-Corcuera B, Montero-Iruzubieta J, Sánchez-Ávila RM, et al. Plasma rich in growth factors for the treatment of cicatrizing conjunctivitis. Clin Ophthalmol. 2020;17(14):1619–27.

46. Sanchez-Avila RM, Merayo-Lloves J, Fernandez ML, et al. Plasma rich in growth factors for the treatment of dry eye after LASIK surgery. Ophthalm Res. 2018;60(2):80–6.

47. Sánchez-Avila RM, Merayo-Lloves J, Fernández ML, et al. Plasma rich in growth factors eye drops to treat secondary ocular surface disorders in patients with glaucoma. Int Med Case Rep J. 2018;1(11):97–103.

48. Sanchez-Avila RM, Merayo-Lloves J, Riestra AC, et al. Treatment of patients with neurotrophic keratitis stages 2 and 3 with plasma rich in growth factors (PRGF-Endoret) eye-drops. Int Ophthalmol. 2018;38(3):1193–204.

49. Guarnieri A, Alfonso-Bartolozzi B, Ciufo G, Moreno-Montañés J, Gil-Bazo I. Plasma rich in growth factors for the treatment of rapidly progressing refractory corneal melting due to erlotinib in nonsmall cell lung cancer. Medicine (Baltimore). 2017;96(22): e7000.

50. Sánchez-Ávila RM, Merayo-Lloves JM. Neurotrophic ulcer after cataract surgery: new treatment. EC Opthalmology. 2016;3(5):359–65.

51. Merayo-Lloves J, Sanchez-Avila RM, Riestra AC, et al. Safety and efficacy of autologous plasma rich in growth factors eye drops for the treatment of evaporative dry eye. Ophthalmic Res. 2016;56(2):68–73.

52. Vendreli C, Piludu S, Mantaras J, et al. PRGF-ENDORET en la queratopatía por síndrome de ojo seco severo. 46 Congreso de la Sociedad Catalana de Oftalmología. Noviembre 2015. Barcelona. Available from: https://www.scoft.cat/oftalmologia2015/docs/comunicaciones/20_comunicados.pdf

53. Valdez Payan EL, Fernandez O, Velasco R, et al. Plasma rich in growth factors (PRGF) compared with autologous serum for dry eye treatment. Invest Ophthalmol Vis Sci. 2014;55(13):3671.

54. López-Plandolit S, Morales MC, Freire V, Grau AE, Durán JA. Efficacy of plasma rich in growth factors for the treatment of dry eye. Cornea. 2011;30(12):1312–7.

55. López-Plandolit S, Morales MC, Freire V, Etxebarría J, Durán JA. Plasma rich in growth factors as a therapeutic agent for persistent corneal epithelial defects. Cornea. 2010;29(8):843–8.

56. Rocha GA, Acera A, Durán JA. Laser in situ keratomileusis flap necrosis after trigeminal nerve palsy. Arch Ophthalmol. 2007;125(10):1423–5.

57. Rahhal-Ortuño M, Fernández-Santodomingo AS, Martínez-Rubio C, Barranco-González H, Almor-Palacios I, Rodrigo-Hernández A. Use of plasma rich in growth factors (PRGF-Endoret®) fibrin membrane to cover corneal dellen. Rom J Ophthalmol. 2021;65(3):293–5.

58. Idoipe M, de la Sen-Corcuera B, Sánchez-Ávila RM, et al. Membrane of plasma rich in growth factors in primary pterygium surgery compared to amniotic membrane transplantation and conjunctival autograft. J Clin Med. 2021;10(23):5711.

59. Sanchez-Avila RM, Merayo-Lloves J, Riestra AC, et al. Plasma rich in growth factors membrane as coadjuvant treatment in the surgery of ocular surface disorders. Medicine (Baltimore). 2018;97(17): e0242.

60. Gemae MR, Patil NS, Yu CW, Agarwal M, Slomovic AR, Chan CC. Use of plasma rich in growth factors for ocular surface disorders: a systematic review. Cornea. 2024

61. Mancini M, Postorino EI, Gargiulo L, Aragona P. Use of allogeneic platelet-rich plasma for the treatment of autoimmune ocular surface disorders: case series. Front Ophthalmol (Lausanne). 2023;5(3):1215848.

62. Romano V, Bignotti S, Forbice E, Bianchetti A, Almici C, Semeraro F. Allogeneic solid platelet-rich plasma for persistent epithelial neurotrophic defects: a protocol and pilot study. Cornea. 2023;42(4):498–506.

63. Passilongo M, Bergamo L, Romanelli F, Lovisi A, Colosio M. Comparison between homologous serum eye drops and platelet-rich plasma eye drops as treatment for dry eye disease. Invest Ophthalmol Vis Sci. 2019;60(9):6742.

64. Foti R, Zeppieri M, Foti R, et al. Umbilical cord blood-derived products in autoimmune systemic

syndromes with severe dryness: a pilot study. Medicina (Kaunas). 2024;60(11):1764.

65. Gagliano C, Foti R, Zeppieri M, et al. Umbilical cord blood platelet lysate eyedrops for the treatment of severe ocular surface disorders in graft vs host disease patients: clinical study. Life (Basel). 2024;14(10):1268.

66. Kamel Farag R, Dawood M, Elesawi M. Safety and efficacy of eye drops from umbilical cord blood platelet lysate to treat resistant corneal ulcer. Med Hypothesis Discov Innov Ophthalmol. 2023;11(4):189–202.

67. Wong J, Govindasamy G, Prasath A, et al. Allogeneic umbilical cord plasma eyedrops for the treatment of recalcitrant dry eye disease patients. J Clin Med. 2023;12(21):6750.

68. Samarkanova D, Martin S, Bisbe L, et al. Barcelona CBED Study Group (Appendix I) Clinical evaluation of allogeneic eye drops from cord blood platelet lysate. Blood Transfus. 2021;19(4):347–56.

69. Pleyer U, Schlickeiser S. The taming of the shrew? The immunology of corneal transplantation. Acta Ophthalmol. 2009;87:488–97.

70. Cantisani R, Del Re V, Toraldo F, et al. HLA class I expression on human platelets is highly variable and correlates with distinct allele group frequencies. Blood Transfus. 2024;22(5):440–9.

71. Chiang CC, Lin JM, Chen WL, Tsai YY. Allogeneic serum eye drops for the treatment of severe dry eye in patients with chronic graft-versus-host disease. Cornea. 2007;26(7):861–3.

72. Chiang CC, Chen WL, Lin JM, Tsai YY. Allogeneic serum eye drops for the treatment of persistent corneal epithelial defect. Eye (Lond). 2009;23(2):290–3.

73. Na KS, Kim MS. Allogeneic serum eye drops for the treatment of dry eye patients with chronic graft-versus-host disease. J Ocul Pharmacol Ther. 2012;28(5):479–83.

74. Giannaccare G, Carnevali A, Senni C, Logozzo L, Scorcia V. Umbilical cord blood and serum for the treatment of ocular diseases: a comprehensive review. Ophthalmol Ther. 2020;9(2):235–48.

75. Pujari R, Deshmukh R, Sheth C, Rajan MS. Maternal finger-prick allogenic blood for persistent corneal epithelial defects. BMJ Case Rep. 2021;14(4):e241138.

76. Dunn SP, Stark WJ, Stulting RD, et al.; Cornea Donor Study Investigator Group. The effect of ABO blood incompatibility on corneal transplant failure in conditions with low-risk of graft rejection. Am J Ophthalmol. 2009;147(3):432–438

77. Solves P, Carpio N, Carretero C, et al. ABO incompatibility does not influence transfusion requirements in patients undergoing single-unit umbilical cord blood transplantation. Bone Marrow Transplant. 2017;52(3):394–9.

78. Gabriel C, Marks DC, Henschler R, Schallmoser K, Burnouf T, Koh MBC. Eye drops of human origin–Current status and future needs: report on the workshop organized by the ISBT Working Party for Cellular Therapies. Vox Sang. 2023;118(4):301–9.

79. Burton MJ, Ramke J, Marques AP, et al. The Lancet Global Health Commission on Global Eye Health: vision beyond 2020. Lancet Glob Health. 2021;9(4):e489–e551.