Supplementary Online Content

Le Q, Chauhan T, Yung M, Tseng C-H, Deng SX. Outcomes of limbal stem cell transplant: a meta-analysis. Published online April 23, 2020. *JAMA Ophthalmol*. doi:10.1001/jamaophthalmol.2020.1120

This supplementary material has been provided by the authors to give readers additional information about their work.

eMethods

Eligibility Criteria

Studies of keratoprosthesis, amniotic membrane transplantation, and cultivated oral mucosal epithelial transplantation were excluded because the purpose of the current study was to evaluate the surgical treatments for limbal stem cell regeneration or replacement and their effect on the reconstruction of ocular surface. Studies that mainly focused on the outcome of optical keratoplasty (penetrating/deep anterior lamellar keratoplasty) after LSCT were excluded unless the outcome of LSCT was presented. To compare the outcomes when different donor sources were used, studies were considered to be eligible only when the exact number of allografts and autografts, and their outcomes were provided separately in the publication. If multiple reports were published from the same authors at the same institutions, these reports were grouped according to the study duration, surgery technique, donor source, and patient information; only the most recent studies with a larger number of patients and a longer follow-up were included to avoid redundant outcomes from an overlapping group of patients.

Quality Assessment

A modified version of the Newcastle-Ottawa Scale (NOS) was used to assess the quality of each cohort study. The NOS was composed of eight items in three categories: selection, comparability and outcome. Each item in the categories of selection and outcome were awarded a maximum of one star, and two stars for the category of comparability. The studies that were assigned at least six stars were considered to be of relatively high quality.

Data Extraction

Demographic and clinical data extracted from each study included study design, sample size, demographic characteristics (gender distribution, mean age, etiology of LSCD, range of LSCD, mean duration between the onset of LSCD and surgery, and prior surgery), surgery type (donor source of the graft, culture system, and substrate for LSCs), and follow-up.

The details of systemic immunosuppressive therapy after allogeneic transplantation were also extracted. We collected the number of medications in immunosuppressive regimen, the dosage, and the treatment duration of each medication used after surgery. Because the immunosuppressive therapy regimens after allogenic LSCT varied among studies, the dosages and durations were categorized. The dosage of medication was categorized as “high,” “regular,” or “low” according to the recommended range in the latest version of clinical guidelines for transplant medications (www.transplant.bc.ca). The duration of immunosuppressive medications used after surgery was classified as “short term (≤3 months),” “mid-term (>3 months but ≤1 year),” and “long-term (>1 year).”

The outcomes extracted from studies were as follows:

1. *Restoration of an intact corneal epithelium*. Some studies included additional outcome measures such as the phenotype of epithelial cells characterized by impression cytology and/or in vivo confocal microscopy at the central cornea and improvement in ocular symptoms and/or vision-related quality of life. "Partial success" was reported in some studies to describe an outcome between "success" and "failure." The total number of cases characterized by "success" and "partial success" was defined as "improvement" in the current study.
2. *Vision improvement*. Two-line improvement in Snellen visual acuity (VA), and pre-surgery and post-surgery LogMar VA were collected. Some studies only provided pre-surgery and post-surgery Snellen VA. They were converted to LogMar VA. "Counting the number of fingers," "hand movement," and "light perception" were converted to LogMar 2.0, 3.0, and 4.0, respectively.
3. *Complications of recipient eyes and donor eyes*. Complications of recipient eyes included recurrent/persistent epithelial erosion, graft rejection, graft failure, infectious keratitis, graft necrosis/loss, corneal melting/perforation, and elevated intraocular pressure(IOP). Complications of donor eyes included hemorrhage and iatrogenic LSCD.

Statistical Analysis

Mean, median, standard deviation (SD), interquartile range, and frequency distribution were used to characterize all studies by surgical technique and donor source. For dichotomized outcomes, which included clinical success, clinical improvement, 2-line Snellen VA improvement, and postoperative complications and adverse events, we used mixed effects logistic models to estimate the overall rate and the rates by subgroups based on surgical technique and donor source, with studies as random effects. Similarly, for the outcomes of clinical success and improvement, we used mixed effects logistic models to evaluate their association with graft source, surgical type, culture system, and immunosuppressive therapy regimen. For the continuous outcome of LogMar VA before and after surgery, robust meta-analysis techniques were used to estimate the change in VA by subgroups based on surgical technique and donor, and the overall estimate of the change was derived. To quantify the outcome heterogeneity among studies, the I^2 statistic was generated to estimate the percentage of variance that is attributable to study heterogeneity based on linearization of logistic models. The likelihood ratio test was used to evaluate the significance of study heterogeneity. Contour-enhanced funnel plots were generated to facilitate the inspection of publication bias. A Modified Macsskill test was performed to formally examine publication bias. All statistical analyses were carried out with R software (www.r-project.org).

eTable 1 Characteristics of eligible studies and quality assessment

	Study Design	Surgical Technique	Sample Size	Mean Age (Y)	Follow-up (M)	Quality Score
El-Hofi AH. 2019	R, NC, CA	CLAL	20	27.4±9.3	29.3±10.5	5
Gupta N. 2018	P, NC, CA	SLET	30	20.7	13.2	5
Basu S. 2018	P, NC, CA	SLET	30	-	25	5
Movahedan A. 2017	R, NC, CO	KLAL/CLAL	165	40.9±15	109.2±35.7	6
Fasolo A. 2017	R, NC	CLET	59	46.3±13.9	72±49.2	7
Cheng J. 2017	R, NC, CA	CLET	80	32.4±13.7	26.4±13.6	6
Arora R. 2017	P, C	SLET/CLAU	20	15.2±10.8 (SLET) 18.1±8.1 (CLAU)	6	6
Basu S. 2016	P, NC, CA	SLET	125	-	18	6
Vazirani J. 2016	R, NC, CA	SLET	68	22	12	5
Scholz SL. 2016	R, NC	CLET	61	48.9±17.5	50.8±32.7	5
Parihar JK. 2016	P, C	CLET/KLAL	50	46±6 (CLET) 48±7 (KLAL)	12	8
Titiyal JS. 2015	P, C, CA	KLAL/CLAL	20	18.1±5.3 (CLAL) 17±6.7 (KLAL)	-	6
Ramirez BE. 2015	P, C, CA	CLET	20	51.6±14.2	36	6
Moreira PB. 2015	R, C, CA	CLAU/CLAL	28	48.8 (CLAU) 31.8 (CLAL)	18.4 (CLAU) 35 (CLAL)	7
Ganger A. 2015	R, C, CA	CLET	62	14.7±10 (autograft) 15.9±10.4 (allograft)	21.4±17.8 (autograft) 24.7±14.6 (allograft)	7
Vazirani J. 2014	R, C, CA	CLET	70	24±12.5	17.5±7	6
Barreiro TP. 2014	R, C, CA	CLAU/CLAL	34	-	19.7±5.6	6
Pellegrini G. 2013	P, NC	CLET	152	46.5±14.6	100.8±30	6
Baradaran-Rafii. 2013	R, NC, CA	KLAL	45	26.7±8.7	26.1±11.8	5

Baradaran-Rafii. 2012	R, NC, CA	CLAU	34	27.3±9.4	17.2±6.3	5
Eberwein P. 2012	R, NC, CO	KLAL	20	44	22.4	5
Basu S. 2012	R, NC, CA	CLET	28	27.9±14.7	57.6±33.6	6
Sangwan VS. 2011	R, NC, CA	CLET	200	24.1±9.9	36±19.2	6
Javadi MA. 2011	R, C, CA	KLAL/CLAL	72	39.6±4.8 (CLAL) 43.7±7.9 (KLAL)	65.6±24.9 (CLAL) 19.6±14.5 (KLAL)	8
Han ES. 2011	R, NC, CA	KLAL	24	39.4±17.4	47.3±22	7
Rama P. 2010	NC	CLET	113	46.5±14.4	34.9±23.2	6
Pauklin M. 2010	P, C	CLET	44	45.4 ± 17.4	28.5±14.9	8
Miri A. 2010	R, C, CO	CLAU/CLAL /KLAL	27	-	47±40.8 (CLAU) 32.6±28.5 (CLAL) 28.1±36.9 (KLAL)	6
Wylegala E. 2008	P, C, CA	CLAU/CLAL /KLAL	90	62.5	31.2	6
Torres J. 2008	R, C, CA	CLAU/CLAL	65	55.8 ± 15.6	19.8±23.5	6
Scocco C. 2008	R, NC, CA	CLAL	39	33.6± 19	48.7±30.6	6
Shimazaki J. 2007	R, C, CA	CLET	27	50.2± 20.7	29.2	7
Maruyama-Hosoi F. 2006	R, NC, CA	KLAL	85	52.5± 19.5	46.6	6
Santos MS. 2005	P, C, CA	CLAU/CLAL	33	35±16	33±12	6
Shimazaki J. 2004	R, C, CA	CLAU/KLAL	32	40.2±14.3 (CLAU) 43.2±19.1 (KLAL)	15.4	7
Ozdemir O. 2004	C	CLAU/CLAL	24	28±11.6 (CLAU) 43±12.4 (KLAL)	13.9±7 (CLAU) 16.2±11.2 (KLAL)	6
Ilari L. 2002	R, NC, CA	KLAL	23	45	60	5
Tsubota K. 1999	NC	KLAL	43	49±23	38.7	6
Schwab IR. 1999	NC	CLET	20	54.8±11.8	9.7±5.9	5

Kenyon KR. 1989	R, NC, CA	CLAU	26	30.8±15	18±11.9	4

C: comparative; CA: case series; CLAL: conjunctival limbal allograft; CLAU: conjunctival limbal autograft; CLET: cultivated limbal epithelial transplantation; CO: case cohort; KLAL: keratolimbal allograft; M: months; NC: non-comparative; P: prospective; R: retrospective; SLET; Y: year

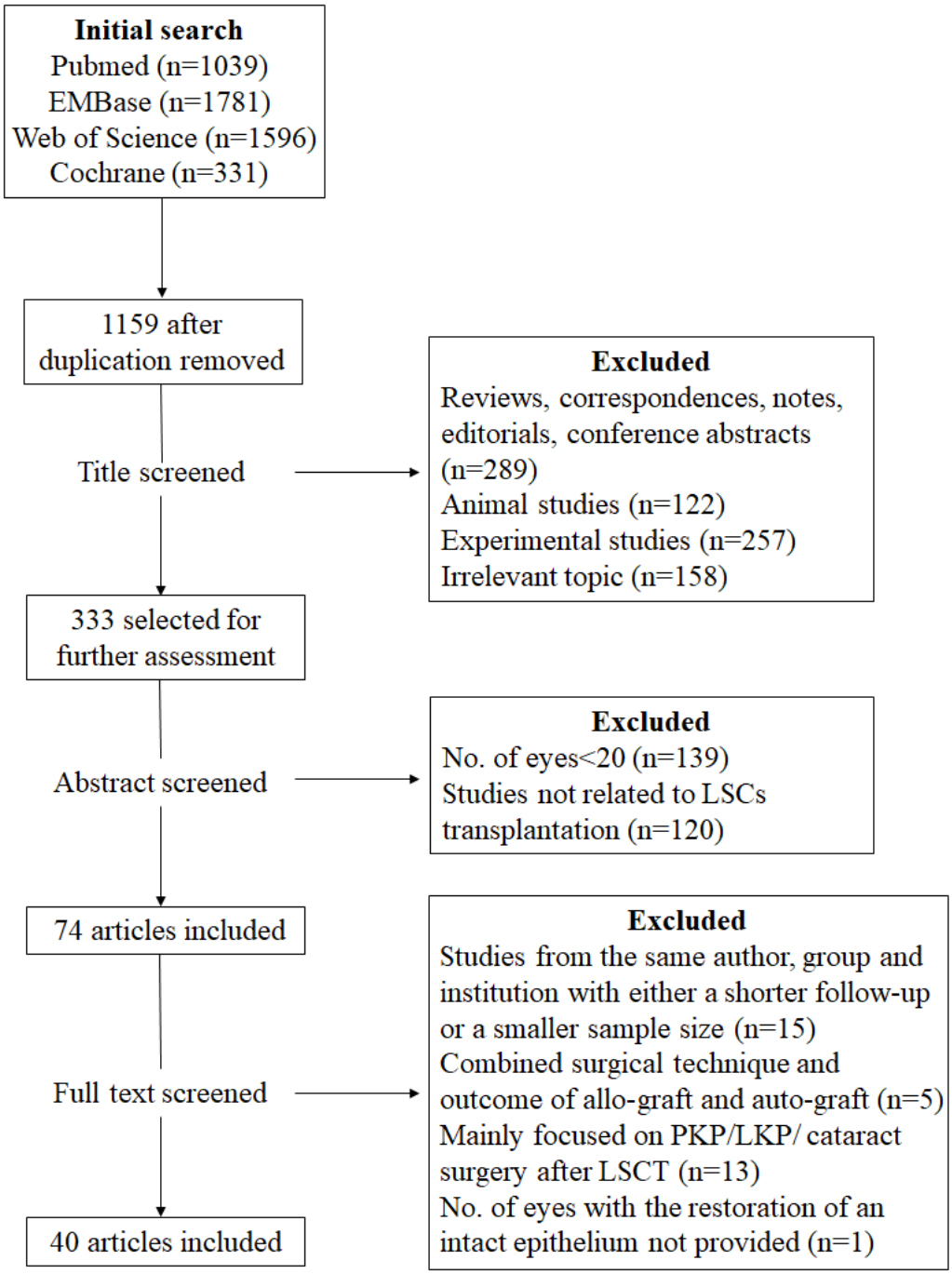
eTable 2 Comparisons on postoperative systemic immunosuppressive therapy and other factors affecting the success rate and the improvement rate

Systemic Immunosuppressive Therapy		No. of studies	Estimated success rate	(95% CI)	OR	(95% CI)	P Value	Estimated improvement rate	(95% CI)	OR	(95% CI)	P Value
No.of Immuno-suppressive Medications	1	10	53%	(39%, 66%)				70%	(57%, 81%)			
	2	10	58%	(42%, 73%)	1.24	(0.611, 2.513)	.55	61%	(42%, 74%)	0.663	(0.300, 1.467)	.31
	3	5	82%	(65%, 91%)	3.844	(1.622, 9.113)	**.002**	86%	(72%, 94%)	2.575	(0.967, 6.858)	.053
Use of Steroid	Long-term	1	74%	(32%, 94%)				80%	(32%, 97%)			
	Mid-term	2	56%	(16%, 89%)	0.443	(0.034, 5.734)	.46	67%	(18%, 95%)	0.486	(0.022, 10.614)	.64
	Short-term	14	56%	(37%, 73%)	0.45	(0.091, 2.217)	.32	68%	(45%, 85%)	0.523	(0.074, 3.698)	.46
Use of Antimetabolites	Long-term	8	75%	(57%, 87%)				82%	(66%, 91%)			
	Mid-term	11	57%	(40%, 72%)	0.436	(0.215, 0.882)	**.02**	64%	(47%, 78%)	0.396	(0.193, 0.814)	**.01**
	Short-term	2	45%	(23%, 69%)	0.274	(0.087, 0.859)	**.02**	46%	(23%, 70%)	0.189	(0.059, 0.602)	**.005**
Dosage of Steroid	High	15	56%	(38%, 73%)				69%	(47%, 85%)			
	Regular	2	54%	(24%, 81%)	0.899	(0.251, 3.221)	.87	64%	(27%, 89%)	0.8	(0.174, 3.676)	.77
Dosage of Antimetabolites	High	2	67%	(33%, 89%)				67%	(31%, 90%)			
	Regular	17	58%	(40%, 73%)	0.66	(0.178, 2.455)	.53	64%	(46%, 79%)	0.888	(0.229, 3.441)	.86
	Low	2	71%	(24%, 95%)	1.192	(0.129, 11.04)	.88	71%	(23%, 95%)	1.188	(0.121, 11.73)	.88
Other Factors												
HLA Matching of Allograft	No	13	51%	(37%, 63%)	1.359	(0.704, 2.622)	.36	54%	(40%, 66%)	1.542	(0.799, 2.977)	.20
	Yes	10	58%	(43%, 71%)				64%	(50%, 76%)			
Use of AM in Direct LSCT	No	11	63%	(45%, 78%)	0.509	(0.208, 1.245)	.14	66%	(48%, 81%)	0.501	(0.200, 1.256)	.14
	Yes	24	46%	(33%, 60%)				50%	(36%, 63%)			

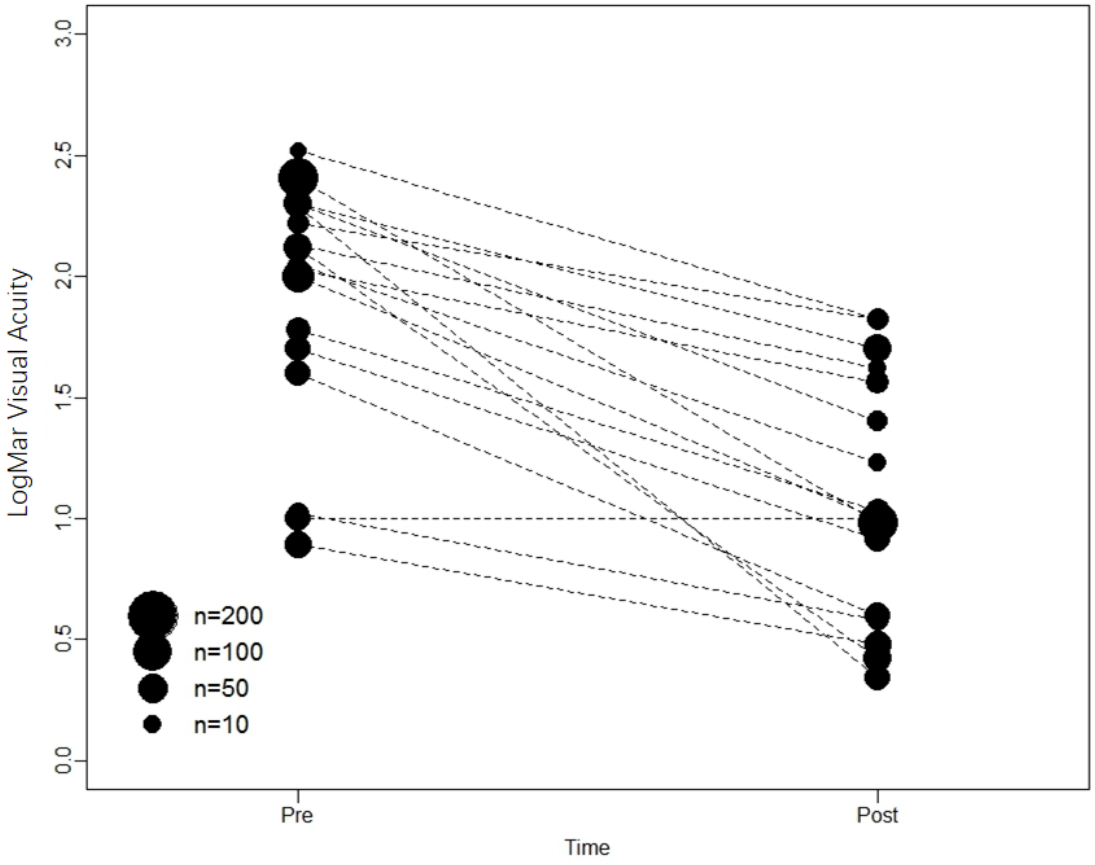
Use of 3T3 Feeder Cells in Cell Culture	No	10	57%	(45%, 68%)	0.808	(0.488, 1.336)	.41	63%	(53%, 72%)	1.931	(1.299, 2.872)	**.001**
	Yes	7	52%	(39%, 64%)				77%	(68%, 83%)			
Serum Used in Cell Culture	Animal	5	57%	(46%, 67%)	0.888	(0.495, 1.593)	.69	75%	(67%, 82%)	0.582	(0.330, 1.027)	.08
	Human	9	54%	(41%, 66%)				64%	(52%, 74%)			
Substrate of Cell Sheet	AM	15	54%	(44%, 64%)	0.751	(0.414, 1.363)	.34	67%	(57%, 76%)	1.677	(0.877, 3.209)	.11
	Fibrin	3	47%	(31%, 64%)				77%	(62%, 88%)			

AM: amniotic membrane; HLA: human lymphocyte antigen; LSCT: limbal stem cell transplantation

eFigure1 Flow chart of study selection



eFigure2 Visual acuity (LogMar) before and after limbal stem cell transplantation.



eFigure3 Funnel plots of publication bias analyses for the success rate (A, B) and improvement rate (C, D). The white, red and orange areas in B and D represent the range of 90%, 95% and 99% confidence interval.