

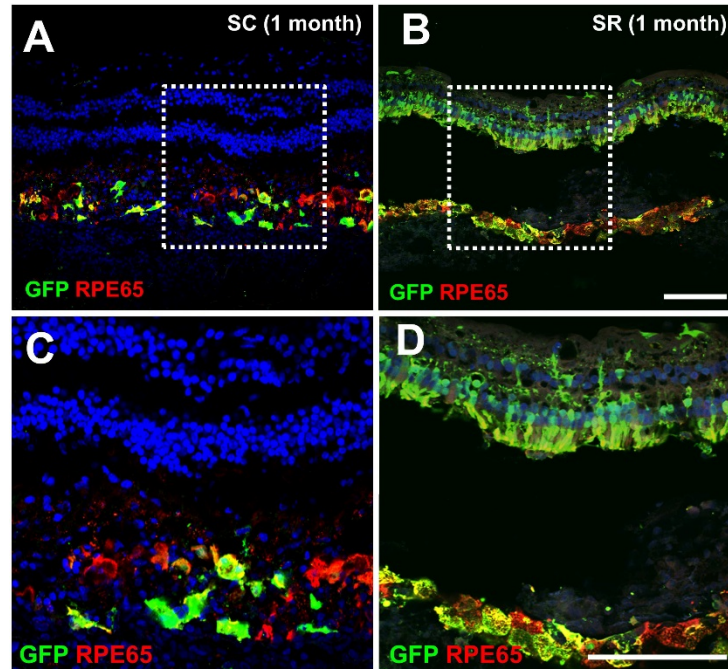
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**Supplemental Information**

**Suprachoroidal and Subretinal Injections  
of AAV Using Transscleral Microneedles for  
Retinal Gene Delivery in Nonhuman Primates**

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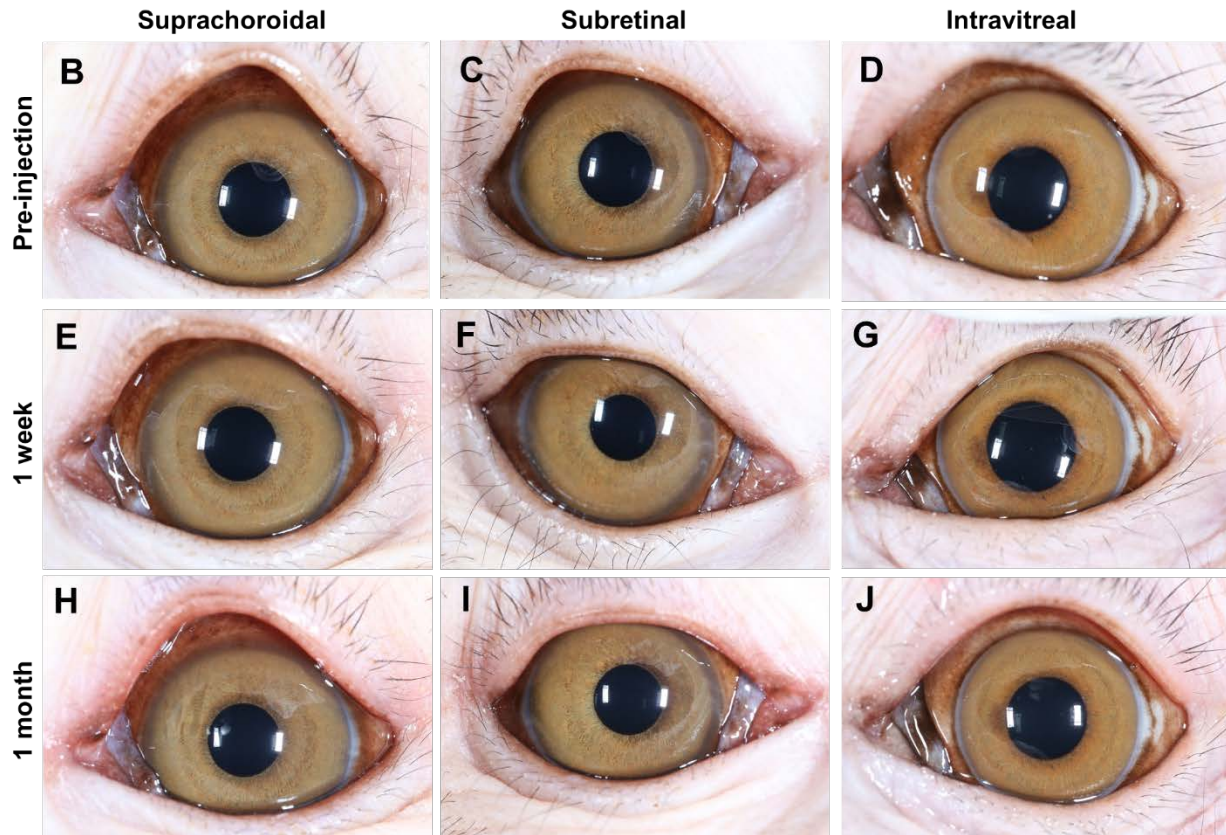
## Supplementary Materials



**Fig S1.** Retinal anatomy and GFP expression near injection sites after suprachoroidal and subretinal injection of AAV8 in rhesus macaques. **(A-B)** Confocal fluorescence images with **(C-D)** magnified views of the white-dashed boxes in **(A-B)** showing GFP expression (green) co-immunostained with anti-RPE65 (red) to label retinal pigment epithelium (RPE) in areas of retinal structure disruption near injection sites after suprachoroidal (**A, C**) or subretinal (**B, D**) injections of AAV8. Scale bars: 100 μm.

**A**

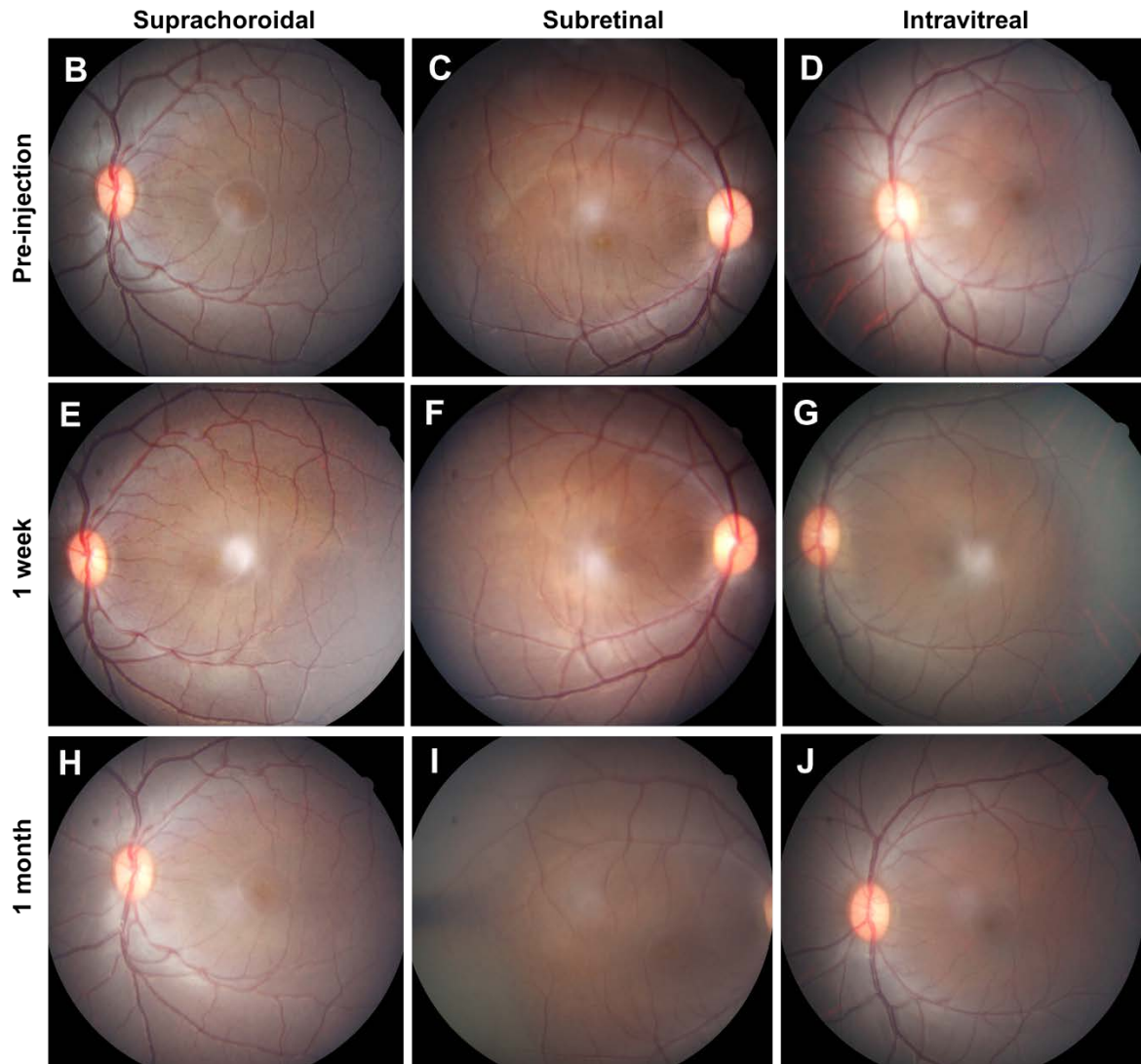
ID	Eye	Dose (vg/eye)	Injectant Location	Pre-injection AC Cell	1 Week AC Cell	2 Week AC Cell	1 Month AC Cell	2 Months AC Cell	3 Months AC Cell
Rhesus 01	OD	7 x 10 <sup>11</sup>	SC	0	0.5	0	0		0
	OS	7 x 10 <sup>11</sup>	SC	0	0	0	0		0
Rhesus 02	OD	7 x 10 <sup>12</sup>	SC	0	0.5	2*	0		0
	OS	7 x 10 <sup>12</sup>	SC	0	0.5	2*	0		0
Rhesus 03	OD	7 x 10 <sup>12</sup>	SR	0 <sup>#</sup>	0	0	0	0	
	OS	7 x 10 <sup>12</sup>	SC	0 <sup>#</sup>	0	0	0	0	
Rhesus 04	OD	7 x 10 <sup>12</sup>	SR	0 <sup>#</sup>	0	0	0		
	OS	7 x 10 <sup>12</sup>	SC	0 <sup>#</sup>	0	0	0		
Rhesus 05	OD	7 x 10 <sup>12</sup>	IVT	0 <sup>#</sup>	0	0	0		
	OS	7 x 10 <sup>12</sup>	IVT	0 <sup>#</sup>	0	0	0		



**Fig S2.** Anterior chamber inflammation following suprachoroidal, subretinal, and intravitreal injections of AAV8. (A) Table outlining study animals and eyes, AAV8 dose, injectant location, and degree of anterior chamber (AC) cell before and after intraocular injections. AC cells were graded based on Standardization of Uveitis Nomenclature (SUN) criteria. \*Rhesus 02 exhibited AC inflammation at 2 weeks and was treated oral prednisone (1mg/kg), while <sup>#</sup>subsequent animals received periorbital subtenon triamcinolone acetate (40mg) at time of the AAV8 injection. (B-J) Representative external photographs show minimal anterior uveitis before (B-D), and at 1 week (E-G) or 1 month (H-J) after suprachoroidal (B, E, H), subretinal (C, F, I) or intravitreal (D, G, J) injections of AAV8.

**A**

ID	Eye	Dose (vg/eye)	Injectant Location	Pre-injection Vitreous Haze	1 Week Vitreous Haze	2 Week Vitreous Haze	1 Month Vitreous Haze	2 Months Vitreous Haze	3 Months Vitreous Haze
Rhesus 01	OD	7 x 10 <sup>11</sup>	SC	0	0	0	0		0
	OS	7 x 10 <sup>11</sup>	SC	0	0	0	0		0
Rhesus 02	OD	7 x 10 <sup>12</sup>	SC	0	0	0	0		0
	OS	7 x 10 <sup>12</sup>	SC	0	0	0	0		0
Rhesus 03	OD	7 x 10 <sup>12</sup>	SR	0	0	0	0	0	
	OS	7 x 10 <sup>12</sup>	SC	0	0	0	0	0	
Rhesus 04	OD	7 x 10 <sup>12</sup>	SR	0	0	0	0	0	
	OS	7 x 10 <sup>12</sup>	SC	0	0	0	0		
Rhesus 05	OD	7 x 10 <sup>12</sup>	IVT	0	0	0	0		
	OS	7 x 10 <sup>12</sup>	IVT	0	0	0	0		



**Fig S3.** Posterior inflammation following suprachoroidal, subretinal, and intravitreal injections of AAV8. **(A)** Table outlining study animals and eyes, AAV8 dose, injectant location, and vitreous haze before and after intraocular injections. Vitreous inflammation was graded based on National Eye Institute (NEI) scale for vitreous haze. **(B-J)** Representative fundus photographs show minimal vitritis before (B-D), and at 1 week (E-G) or 1 month (H-J) after suprachoroidal (B, E, H), subretinal (C, F, I) or intravitreal (D, G, J) injections of AAV8.

**Table S1.** Intraocular pressure after suprachoroidal, subretinal, or intravitreal AAV8 injections

<b>ID</b>	<b>Eye</b>	<b>Dose</b> (vg/eye)	<b>Injectant</b> <b>Location</b>	<b>Pre-</b> <b>injection</b> <b>IOP</b>	<b>1</b> <b>Week</b> <b>IOP</b>	<b>1</b> <b>Month</b> <b>IOP</b>	<b>2</b> <b>Months</b> <b>IOP</b>	<b>3</b> <b>Months</b> <b>IOP</b>
<b>Rhesus 01</b>	OD	$7 \times 10^{11}$	SC	14	7	7		14
	OS	$7 \times 10^{11}$	SC	19	10	8		20
<b>Rhesus 02</b>	OD	$7 \times 10^{12}$	SC	17	13	23		13
	OS	$7 \times 10^{12}$	SC	18	18	23		15
<b>Rhesus 03</b>	OD	$7 \times 10^{12}$	SR	19	16	16	21	
	OS	$7 \times 10^{12}$	SC	24	19	19	19	
<b>Rhesus 04</b>	OD	$7 \times 10^{12}$	SR	14	13	18		
	OS	$7 \times 10^{12}$	SC	17	15	16		
<b>Rhesus 05</b>	OD	$7 \times 10^{12}$	IVT	21	15	15		
	OS	$7 \times 10^{12}$	IVT	19	14	14		