Optic nerves were exposed surgically in an anesthetized adult Sprague Dawley rats through using a supraorbital approach and crushed using an aneurysm clip (YASARGIL). Aneurysm clip, which was placed 2 mm behind the posterior eye pole, as described in previous reports. Cultured neural stem cells (NSCs) were transplanted into the subretinal space immediately after the optic nerve crush using a transscleral approach. A blunt needle attached to a 10-μL syringe (Hamilton, Reno, NV) was introduced tangentially through the sclerotomy site into the subretinal region and caused retinal detachment. The retinal detachment was confirmed microscopically. The same procedure was then repeated to slowly inject a suspension of pigment epithelium-derived factor (PEDF)-modified NSCs (2 μL of 2.0 × 10^5 cells). In this study, 72 rats with ongoing optic nerve injury were randomly assigned to three groups: a group that received injections of phosphate-buffered saline (PBS) (n = 24), a group that received weekly injections of PEDF (n = 24), and a group that received PEDF-modified NSCs (n = 24). Subsequently, 0.67 nM of PEDF dissolved in 5 μL of sterile PBS was injected immediately after the optic nerve crush (day 0) and at 1 and 2 weeks thereafter. The rats (from each group) were examined at each of the time points post-injection (2 or 4 weeks). At each time point, samples were harvested and placed into a protein extraction buffer. Equal amounts of protein were denatured for 5 minutes at 95 °C in a sample buffer and separated by sodium dodecyl sulfate polyacrylamide gel electrophoresis (SDS-PAGE).