Supplementary Figure S1. Western blotting for GFAP expression. Freshly dissected retinas (from 9 month old transgenic and WT mice) were washed and homogenized in PBS buffer. Lysates were prepared by addition of sample buffer (4% (w/v) sodium dodecyl sulfate, 20% (v/v) glycerol, 0.13M Tris, pH 6.8) with Complete™ protease inhibitor cocktail (Roche Applied Science, Cat# 11 697 498 001). Protein levels were measured using a Pierce® BCA protein assay kit (Fisher Thermo Scientific, Cat# PI-23227). After addition of 2% (v/v) 2-mercaptoethanol and 1% (v/v) saturated bromophenol blue protein extracts were boiled for 5 min. Samples (30 µg protein) were resolved by SDS-PAGE on 8-10% acrylamide gels. Proteins were transferred to polyvinylidene fluoride (PVDF) membranes, blocked for one hour with 5% non fat milk diluted in TBS-T (20mM Tris, 137mM NaCl, 0.1% Tween-20, pH 7.6), incubated overnight with mouse anti-GFAP, mouse anti-alpha-tubulin (TU-02, Santa Cruz, CA), or mouse anti-beta-actin (C4; sc-47778, Santa Cruz, CA) antibodies diluted 1:1000 in the blocking solution, washed 3 x 10 min. in TBS-T and reacted for 1h with a sheep anti-mouse IgG, HRP-conjugated ECL antibody (1:5000 in the blocking solution; NA931; GE Healthcare, UK). After washing 3 x 10 min. in TBS-T, protein bands were visualized using ECL reagent (NEL 103, Perkin Elmer) and a Kodak Image Station 440. Note the clear (8-fold) increase in GFAP in ELOVL4/TG1-2 compared to WT retinas.