**Supplementary Figure S1.** Effect of S1PR and SphK1 blockers on expression of pro-fibrotic and tissue-remodeling proteins in non-stimulated orbital fibroblasts in GO. Confluent orbital fibroblasts derived from individuals with GO were either untreated or treated with 10
μM W146, 10 μM JTE013, 1 μM FTY720, or 10 μM 5C for 24 h. Expression levels of (A) pro-fibrotic proteins (collagen-Iα, fibronectin, and α-SMA) and (B) tissue-remodeling proteins (MMP-1, MMP-2, MMP-9, and TIMP-1) in cultured cells were assayed by Western blotting. Data in the columns indicate the mean relative density ratios ± SD of three experiments (*P < 0.05 and **P < 0.01 versus untreated control cells).

GO = Graves’ orbitopathy; MMP = matrix metalloproteinase; S1PR = sphingosine-1-phosphate receptor; SphK1 = sphingosine kinase 1; SD = standard deviation; SMA = smooth muscle actin; TIMP = tissue inhibitor of metalloproteinase.
SUPPLEMENTARY FIGURE S2. Effect of siRNA-mediated Sphk1 knockdown on TGF-β-induced expression of pro-fibrotic proteins. Approximately 80% confluent orbital fibroblasts from patients with GO were prepared in 100-mm plates. Transfection of NC or SphK1 siRNA was performed with Lipofectamine 2000 in accordance with the manufacturer’s instructions. After transfection, cells were incubated with or without 5 ng/ml TGF-β for 24 h. Expression levels of collagen-Iα, fibronectin, and α-SMA were then evaluated by Western blotting. Data in the columns indicate the mean relative density ratios ± SD of three experiments (*P < 0.05 versus NC siRNA transfected TGF-β-stimulated cells).

GO = Graves’ orbitopathy; NC = negative control; S1P = sphingosine-1-phosphate; SD = standard deviation; siRNA = short interfering RNA; SMA = smooth muscle actin; SphK1 = sphingosine kinase 1; TGF = transforming growth factor.
SUPPLEMENTARY FIGURE S3. Effect of exogenous S1P on MMP-1 expression in orbital fibroblasts in GO. (A) Confluent orbital fibroblasts derived from individuals with GO were treated with different concentrations of S1P (0–10 μM) for 16 h. Expression levels MMP-1 were then evaluated by Western blotting. Data in the columns indicate the mean relative density ratios ± SD of three experiments (*P < 0.05 versus untreated control cells). (B) Confluent orbital fibroblasts derived from individuals with GO were either untreated or pretreated with 10 μM W146, 10 μM JTE013, 1 μM FTY720, or 10 μM 5C for 1 h prior to treatment with IL-1β (10 ng/ml, 24 h). Expression levels of MMP-1 were then evaluated by Western blotting. Data in the columns indicate the mean relative density ratios ± SD of three experiments (*P < 0.05 and **P < 0.01 versus IL-1β-treated cells without pretreatment).

GO = Graves’ orbitopathy; IL = interleukin; MMP = matrix metalloproteinase; S1P = sphingosine-1-phosphate; SD = standard deviation.