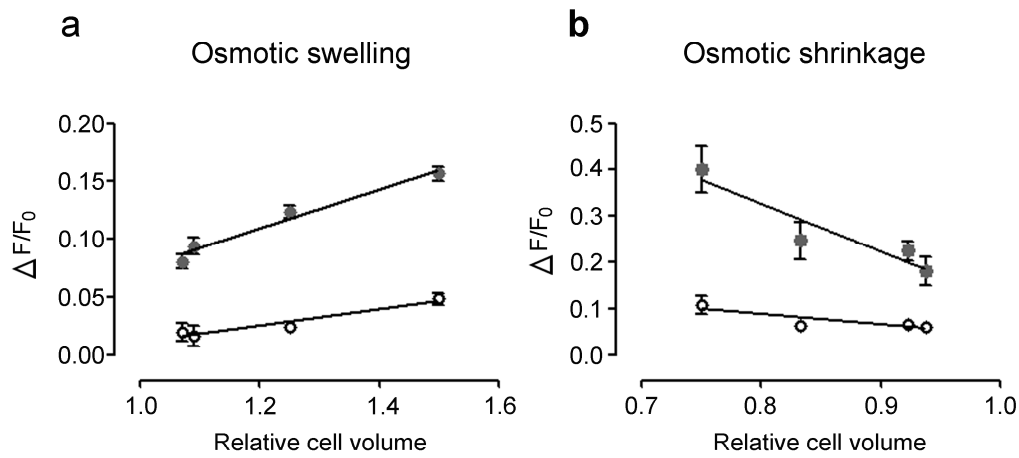


# **Supplementary Material**

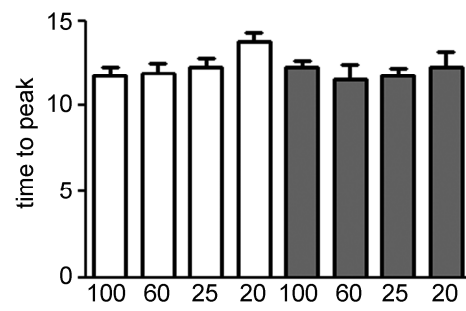
## **Cell Volume Regulation Mechanisms in Differentiated Astrocytes**

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**Fig. S1.** Characterization of the relationship between the osmotic change in cell volume and the intracellular fluorescent intensity in calcein-loaded astrocytes grown on HTlc compared to those grown on PDL. Cells were exposed to a series of hypertonic and hypotonic gradient sizes (20, 25, 60, 100 mOsm/L), and the average calcein signal was determined as the difference between the fluorescence intensity in 12 replicate wells before and after application of the osmotic challenges. The  $\Delta F/F_0$  ratio of the fluorescence variation at time  $t$  to initial fluorescence  $F_0$  was plotted with the relative change in cell volume computed from the ratio of solution osmolarities. The linear regression line is indicated for both PDL cells (white circles) and HTlc cells (grey circles). (means  $\pm$  SE,  $n=3$  independent experimental data).



**Fig. S2.** Histogram showing the mean $\pm$ SE values of the time to peak of calcium responses recorded from PDL (white bars) and HTlc (grey bars) astrocytes exposed to a range of hypotonic extracellular osmolarities as reported in Fig. 3A. (n=3 independent experimental data).