Title: Tocotrienol inhibits TGF-β1-induced human airway smooth muscle cell differentiation

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Body: Background Tocotrienols, members of the vitamin E family, consist of four different isoforms (α, β,γ and δ tocotrienol) that have been shown to exhibit protective effects from brain damage, as well as having anticancer effects in vivo and in vitro. In breast cancer cell lines, γ-tocotrienol inhibited cell growth via a mechanism involving inhibitory differentiation 1 (Id1), which is also a critical mediator in TGF-β1 -induced transdifferentiation of hepatic stellate cells (1, 2). We tested whether γ-tocotrienol modulates TGF-β1 -induced differentiation of human airway smooth muscle (ASM) into a hypercontractile phenotype and synthesis of extracellular matrix proteins. Methods Human ASM cells were stimulated with TGF-β1 (2ng/ml) for 48 hours and the effect of γ-tocotrienol (50µM) on α-smooth muscle actin and fibronectin expression was assessed using Western blotting analysis. We also studied the signaling pathways involved in TGF-β1 stimulation. Results TGF-β1 increased α-smooth muscle actin and fibronectin expression in human ASM cells about 3 to 5 fold. γ-Tocotrienol significantly inhibited this effect. Furthermore, γ-tocotrienol inhibited Id1 expression 3 to 6 hours after TGF-β1 stimulation, and suppressed RhoA activation, but was without effect on Smad2 and Smad 3 phosphorylation. Conclusions These results indicate that γ-tocotrienol could have beneficial effects for airway remodeling in asthma by inhibiting Id1 expression and RhoA activation in ASM cells exposed to elevated TGFβ3 in asthma. Reference 1. Yap, W.N. et al. Cancer Lett. 2010; 291: 187-99. 2. Wiercinska, E. et al. Hepatology 2006; 43: 1032-41.