recruited. fMRI data was gathered with a 3T Siemens TIM Trio scanner. Participants were presented with a standard GoNoGo task. Accumulated points were displayed every 20 frames with a target goal of 1000 to win a prize. NoGo error rate was maintained at 50±10% by adjusting stimulus duration to control difficulty. Unbeknownst to subjects, trials were structured as “win/lose/recovery” paradigms to induce frustration and index emotional regulation. “Win” blocks allowed point increases, “lose” blocks led to net negative points, and “recovery” blocks allowed re-accumulation to ultimately win the prize. BioImage Suite was used to analyze whole-brain intrinsic connectivity between tasks with cluster-corrected group-level T-maps. P<0.05 was significant.

RESULTS: Seven right unilateral-coronal (ULC; average age 12.2, 3 females), six metopic (MCS; average age 11.5 years, 2 females), and respective matched controls were included. During “win”, ULC displayed significantly increased activity in the right inferiortemporal gyrus(p<0.05), calcaneus(p<0.03), and cerebellar vermis(p<0.01), and reductions in the right parietal operculum of the supramarginal gyrus(SMG; p<0.03). “Lose” induced decreased left superior/middle frontal(p<0.05), left precuneus(p<0.01) and left pre/post central gyri activity(p<0.03). In “recovery”, ULC cohort experienced increases in the left pre/post central gyri (p<0.05), left inferior temporal gyri(p<0.02), left paracentral lobule(p<0.01), bilateral precuneus/calcaneus (p<0.01), right anterior subinsula(p<0.05), and cerebellar vermis(p<0.01). MCS participants during “win” demonstrated significantly decreased activity in the left supramarginal/angular gyrus(p<0.03) and bilateral posterior cingulate(p<0.01). “Lose” induced decreased activity in similar regions with additional increases at the anterior paracentral lobule/supplementary motor area(p<0.03). “Recovery” prompted similar suppression in the posterior cingulate gyrus(p<0.05) and activation in the anterior paracentral lobule/supplementary motor area(p<0.05). Sagittal synostosis studies are ongoing.

CONCLUSION: NSC adolescents respond variably to reward, frustration, and emotional regulation. During “win” and “recovery” R-ULC patients evidenced increased activity in regions associated with visual recognition/processing and motor coordination. “Lose” prompted broad suppression ipsilateral to the calvarial defect, particularly in areas responsible for inhibition, suggesting poor self-control when frustrated. ULC patients experienced a surge of activity during “recovery” in reward regulation and visual processing, suggesting possible difficulties in down-regulating emotional based sensory input. Importantly, while ULC morphological defects are isolated to the anterior cranium, posterior brain connectivity were influenced. MSO adolescents had comparable activity suppression across all conditions, located in regions producing emotional regulation and recognition. This suggests global deficits but less pronounced dynamic processing differences. This study provides insight into higher level neural networks in patients with NSC, providing evidence for targeted neuropsychiatric therapy depending on the suture phenotype.

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84

nAG (a Salamander-Derived Protein) as an Inhibitor of TGF-β Signaling and Fibrotic Responses

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PURPOSE: Salamanders have the amazing ability to regenerate their limbs within 30 days when amputated. The key protein responsible for this regeneration is the nAG protein, which stands for newt anterior gradient. In previous studies, a nAG gene was designed that was suitable for human use that demonstrated local injection of recombinant nAG protein reduces hypertrophic scarring in a rabbit ear model. Fibrotic disorders of the skin such as scleroderma, hypertrophic scarring and keloids are characterized by excessive pro-fibrotic responses was determined by measuring the Extracellular Matrix (collagen III, Fibronectin), connective

METHODS: Fibroblasts from lesional scleroderma patient skin were treated with nAG protein in doses of 100pM,1nM and 10nM for 24 hrs and were then left untreated or treated with 20 pM of TGF-β. The inhibition of TGF-β-mediated pro-fibrotic responses was determined by measuring the
tissue growth factor (CTGF) and myofibroblast alpha smooth muscle actin (α-SMA) protein production by Western blot as well as immunofluorescence and validated at the mRNA level by Quantitative PCR. Activation of the TGF-β pathway was determined by measuring the TGF-β receptor 1 (ALK5) and phosphorylated Smad2/3 levels by using Western blot and immunofluorescence. Luciferase assay was used to measure nAG protein’s capacity to inhibit the three TGF-β isomers. Cell migration was assessed using a scratch assay and finally colocalization of nAG protein with TGF-β receptor 1 was evaluated using confocal microscopy.

RESULTS: Both the Western Blot and the immunofluorescence results revealed that the application of the nAG protein to human scleroderma fibroblasts in the presence of TGF-β successfully inhibited the fibrotic response shown by a decrease in the fibrotic factors such as collagen III, alpha smooth muscle actin (α-SMA), connective tissue growth factor (CTGF) and fibronectin. In addition, immunofluorescence and western blot after 1 hour of treatment with nAG revealed a significant decrease in Phosphorylated Smad2/3, and a decrease in the TGF-β receptor 1 (ALK5) inhibiting the TGF-β pathway. Luciferase assay revealed nAG’s inhibition of the canonical TGF-β pathway to be most specific with TGF-β1 isomer reducing activity by 83%. Cell migration was significantly inhibited with nAG protein treatment and confocal microscopy revealed the colocalization of nAG protein with ALK5 receptors.

CONCLUSION: Fibrosis in scleroderma fibroblasts was effectively inhibited when treated with nAG protein, demonstrated by the decrease in ECM using Western Blot and immunofluorescence. Although much about the mechanism of the nAG protein is still unknown, the decrease in pSmad2/3 and ALK5 receptors most potently with TGF-β1 after treatment, inhibition of cell migration and binding of nAG protein with ALK5 receptors suggest that nAG blocks the canonical TGF-β pathway. This research is anticipated to lead to the development of an injectable antifibrotic agent for the treatment of fibrotic disorders such as scleroderma, hypertrophic scars and keloids.

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