

Table S1: *Specc1*^{ΔC510} primer sequences

Primer	Sequence
ZFN Targeting Forward	GTCTGAGCAGAAGGGCAAAG
ZFN Targeting Reverse	AAGAACGTGTTTACGTGGGC
Δ154 Genomic Forward	AGTGAGAACGAAAGGCTGGG
Δ154 Genomic Reverse	GAAGAGGAGCGGCTGACAAT
Δ154 RT-PCR Forward	AGAGCAGCGCTACATGGATT
Δ154 RT-PCR Reverse	CTTCTTGGCCTCTTCCTGGA

Table S2: Specc1^{cGT} primer sequences

Primer #	Name	Sequence
965	Ad-SA-XhoI_R	TGAAGCCGCTCGAGACTGGAAAGACCGCGAAGA
1112	mPrdm16in2cGT-3'TargetPCR_F	GGGAGGATTGGGAAGACAAT
1115	mPrdm16in2cGT-5'TargetPCR_R	ACTGGAAAGACCGCGAAGAG
1125	bGeo_RT-R1	CGCCATGTCACAGATCATCAAG
1686	mSpecc1l-5'HomA-attB4f	GGGACAACCTTTGTATAGAAAAGTTGTTGTCCTC TGACACGCACTT
1687	mSpecc1l-5'HomA-attB1r	GGGACTGCTTTTTTGTACAACTTGACTGTTGC CATCTGCAACTT
1688	mSpecc1l-3'HomB-attB2f	GGGACAGCTTTCTTGTACAAAGTGGAAGTACTC TCATCTGCCGACTGT
1689	mSpecc1l-3'HomB-attB3r	GGGACAACCTTTGTATAATAAAGTTGCACATTGA CTACGGGAAGCA
1712	Specc1l-HomA-5R	GGATGTCTTGATCCCTGGAA
1713	Specc1l-HomA-3F	AACTCTCAAGCCCTCCACAG
1717	Specc1l-HomB-3F	CACACAGCAGGCATCCATAA
1752	Specc1l-5'TargTest_F (w/ oBB1115)	TTCATGTCAAATGCCCAAGG
1753	Specc1l-3'TargTest_R (w/ oBB1112)	GGCAATGAGCACAGAACTGA
1754	Specc1lx2_F (w/ oBB1125)	ACCAAGAGGCAGCCCAGAAT
	pUC_F	CTAGTCCACTGGCACAGCCTAC
	Hsvtk_R	AACTCCAGAGAGGGCAGAG

Figure S1:

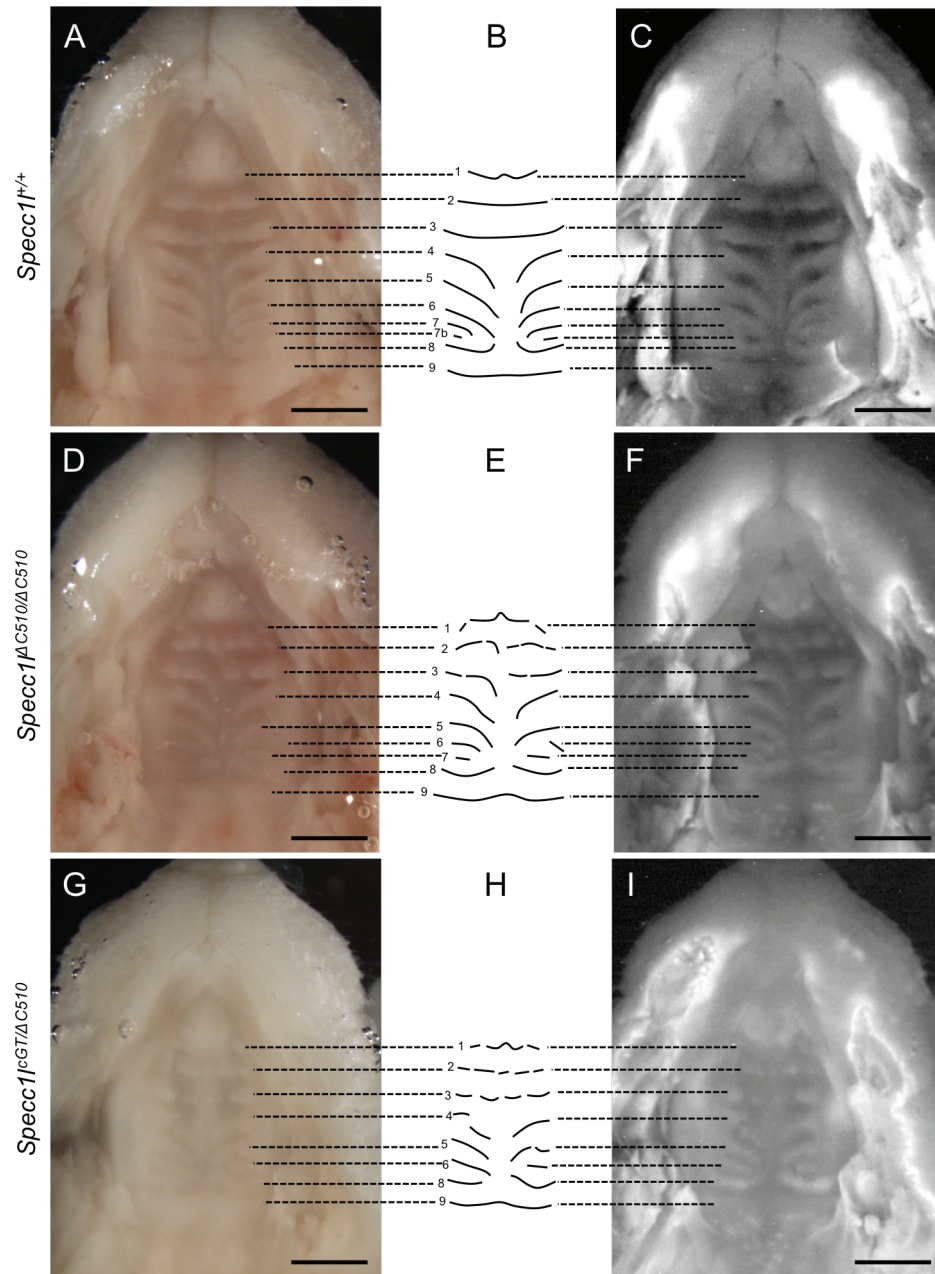


Figure S1: At E18.5 rugae patterning is disrupted moderately in *Specc1*^{ΔC510/ΔC510} embryos and severely in *Specc1*^{cGT/ΔC510} embryos. *Specc1*^{+/+} embryos (top row) with the rugae numbering schematic (B) used in this study. *Specc1*^{ΔC510/ΔC510} mutant embryos (middle row) exhibit moderate defects in rugae development outlined in schematic (E) with several discontinuous rugae with the absence of 7b. *Specc1*^{cGT/ΔC510} mutant embryos (bottom row) display the greatest degree of rugae developmental defects with several discontinuous rugae and the complete absence of 7 and 7b outlined in schematic (H). Scale bars are 1mm.

Figure S2:

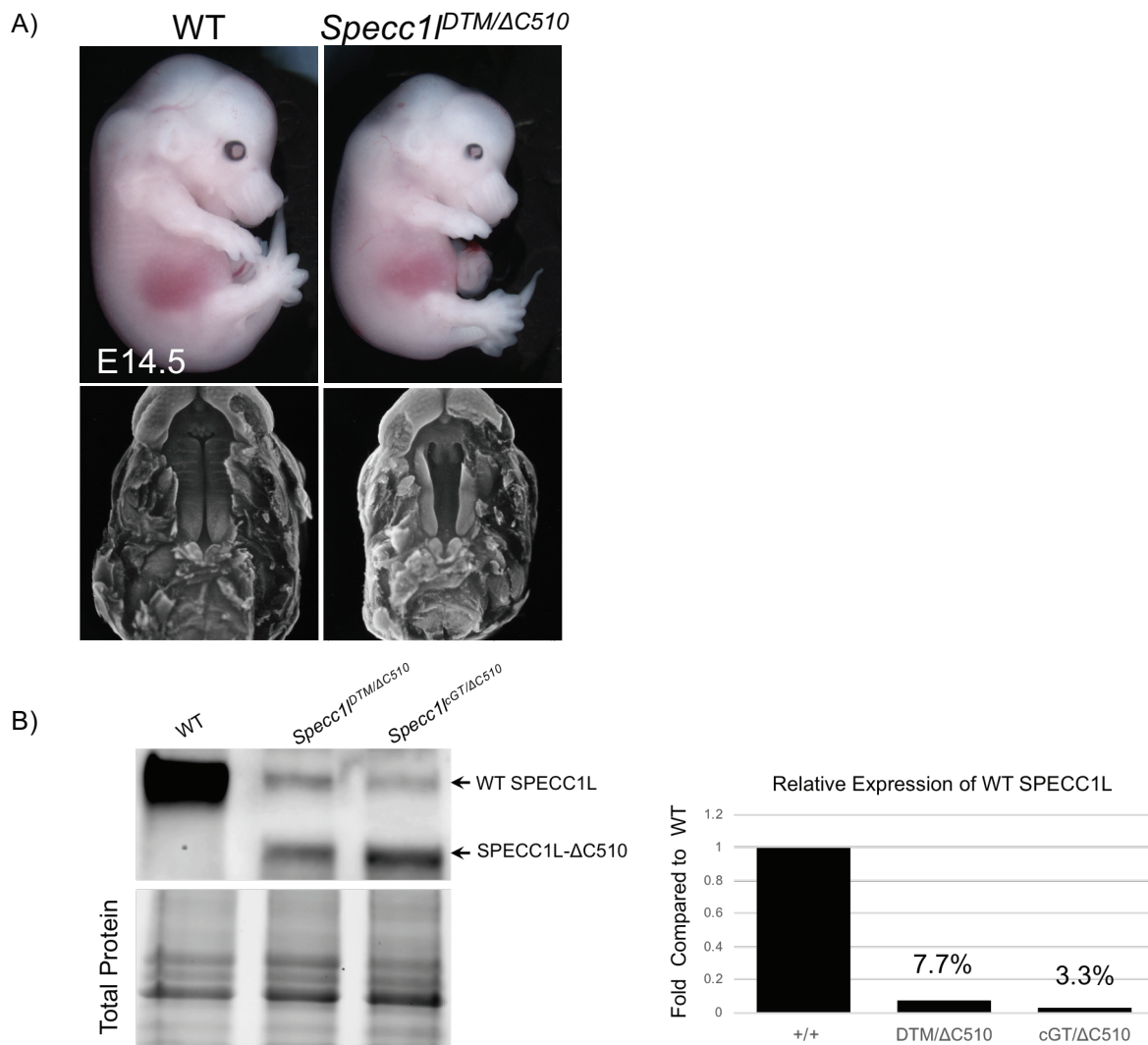


Figure S2: *Specc1*^{DTM/ΔC510} E14.5 embryos also show palate elevation delay. **A)** Representative images of E14.5 wildtype (left) and *Specc1*^{DTM/ΔC510} mutant (right) whole embryos and palatal shelves stained with DAPI. **B)** Lysates from WT, *Specc1*^{DTM/ΔC510} mutant, and *Specc1*^{cGT/ΔC510} mutant were immunoblotted with α -SPECC1L. Wildtype (WT) SPECC1L levels are drastically reduced in the compound heterozygotes, with cGT gene-trap allele showing lower levels than DTM gene-trap allele, as expected. The truncated SPECC1L-ΔC510 band can be observed migrating faster, as shown in Figure 1.

Figure S3:

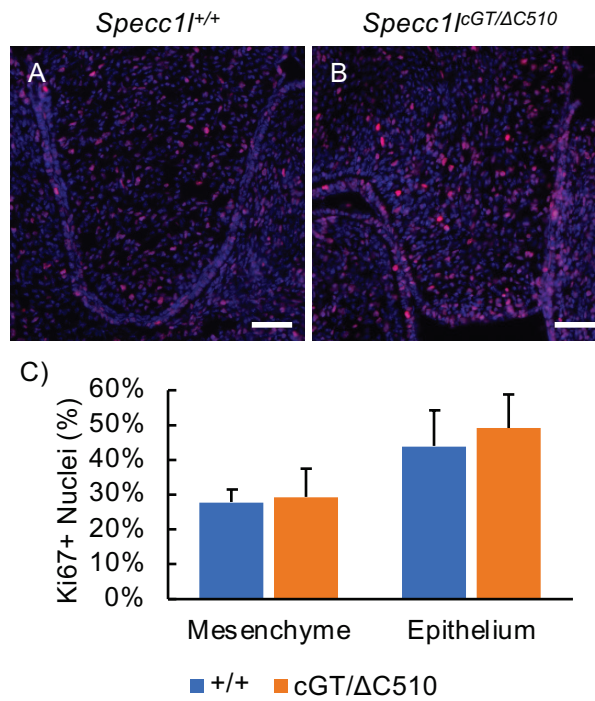


Figure S3: Palates of *Specc1*^{cGT/ΔC510} E13.5 embryos show no changes in cell proliferation. A- B) Representative images of palatal shelves from E13.5 wildtype and *Specc1*^{cGT/ΔC510} mutant coronal sections immunostained for Ki67 (red) and counterstained with DAPI (blue). Scale bars are 50μm. **C)** The proliferation index was determined by measuring the proportion of Ki67+ nuclei in the mesenchyme and epithelium of the palatal shelves of three embryo pairs. The index is not significantly different between wildtype and mutant samples. Error bars show SEM.

Figure S4:

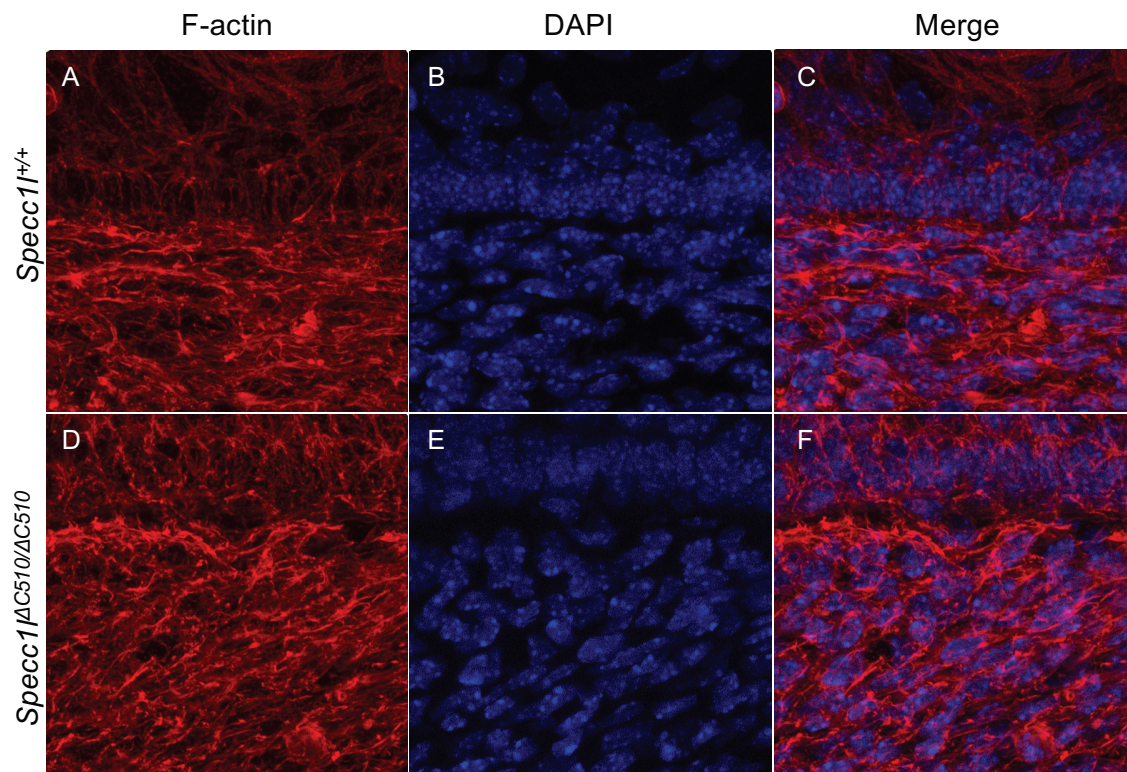


Figure S4: *Specc1*^{ΔC510/ΔC510} mutant embryos do not show differences in F-actin staining.

Actin staining of sagittal embryonic head sections at E18.5. Images were taken of the basal oral epithelium and underlying mesenchyme of the jaw. 100x magnification.

Figure S5:

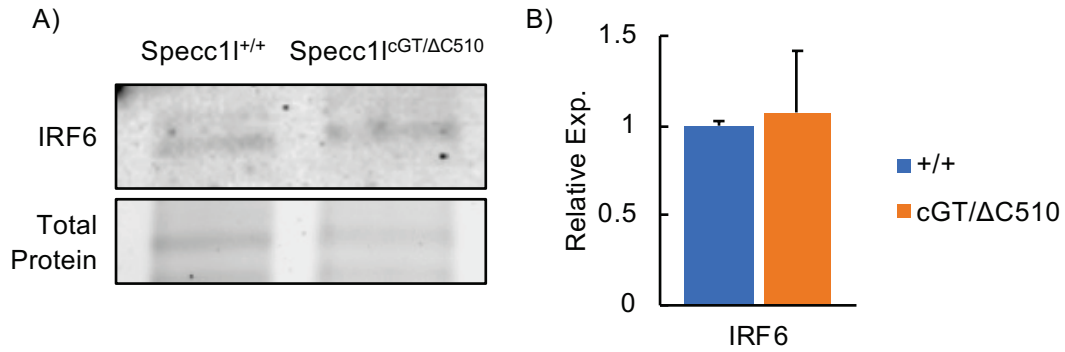


Figure S5: IRF6 protein levels are unchanged in *Specc1*^{cGT/ΔC510} mutants. **A)** Representative images from IRF6 western blots of wildtype and *Specc1*^{cGT/ΔC510} mutant craniofacial lysate. **B)** Bar graph showing relative expression normalized by total protein. There is no significant difference in IRF6 protein levels between the wildtype and mutant samples. n=3. Error bars show SEM.

Figure S6:

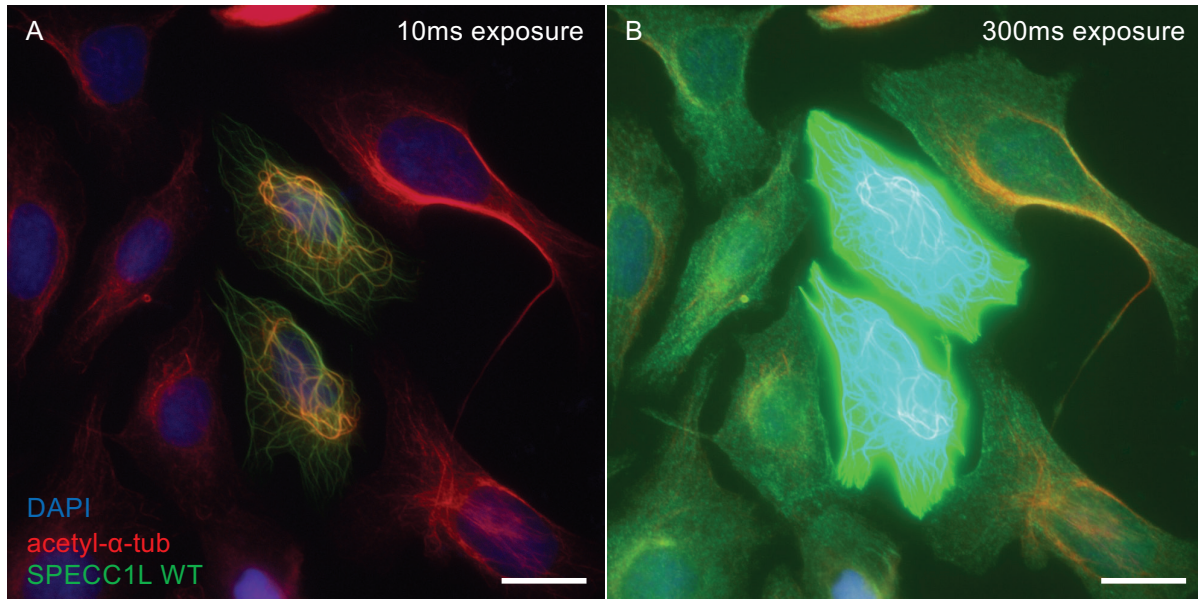


Figure S6: Verification of SPECC1L staining in transfected cells. A-B) Representative images of transfected cells taken at 10ms (A) and 300ms (B) exposure to demonstrate the difference in the level of overexpressed SPECC1L compared to endogenous (J). Scale bars are 20 μ m.