



**Figure 2-figure supplement 4. Complementation of *brush* by overexpressing *ATCNGC19* or *ATCNGC20*.** (A) Hairy root complementation assay in *brush* by overexpression of *ATCNGC19* and *ATCNGC20* relative to empty vector control. (B) Box plot showing the number of *DsRED*-positive nodules per transformed plant (n=10 for all constructs). (C) Yeast two-hybrid interaction assay with the BRUSH NT fused to the GAL4 binding domain (BD) and either *ATCNGC19* NT or *ATCNGC20* NT fused to the GAL4 activation domain (AD). Shown are control (-LW: leucine, tryptophan), low-stringency (-LWH: leucine, tryptophan, histidine), or high stringency (-LWAH: leucine, tryptophan, adenine, histidine) plates for assaying interaction. Yeast optical densities (600 nm) were adjusted to 0.5 and diluted 10-fold. The lower panel in (A) contains images of the *DsRED* channel to visualize *Mesorhizobium loti*. Roots were observed six weeks after the addition of rhizobia. Scale bars in (A) represent 2 mm. Different letters in (B) indicate different statistical groups (ANOVA followed by Tukey's HSD test,  $F_{(2, 26)} = 18.99$ , p-value < 0.05).