Additional File 1

Expression and secretion of a lytic polysaccharide monooxygenase by a fast-growing cyanobacterium

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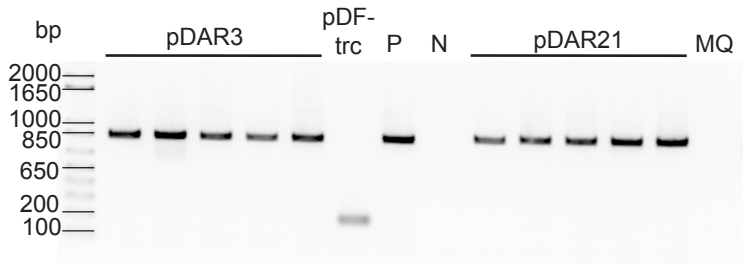

Figure S1. PCR analysis of *S. elongatus* UTEX 2973 strains *Tf*AA10A (pDAR3) and TorA-*Tf*AA10A (pDAR21). Lanes show the colonies from transformation of *S. elongatus* UTEX 2973 with plasmids pDAR3 (*Tf*AA10A), pDAR 21 (TorA-*Tf*AA10A) and an empty vector control (pDF-trc). P denotes a positive control reaction (using pDAR3 plasmid DNA), N denotes a negative control (wild-type *S. elongatus* UTEX 2973) and MQ denotes a H_2O PCR control. The following primers (5' – 3') were used for amplification of the gene of interest: trc-F (attctgaaatgagctgttga) and trc-R (atcaggctgaaaatcttctc).

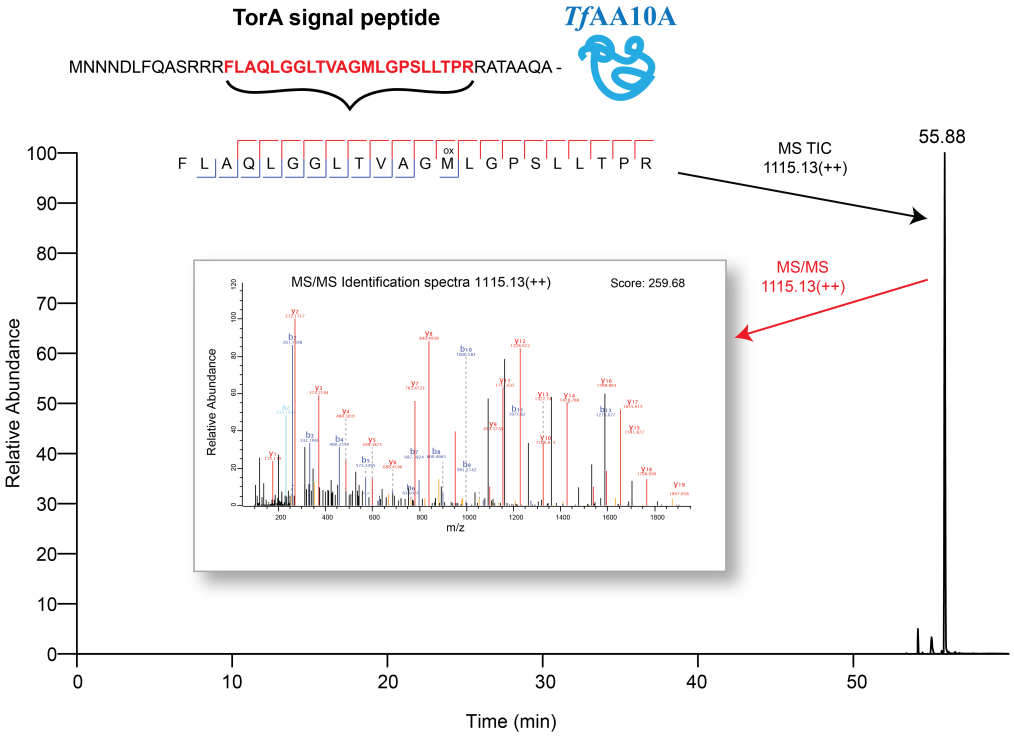

Figure S2. Identification of the TorA signal peptide in the pre-protein TorA-*Tf*AA10A, found in the plasma membrane fraction, by mass spectrometry.

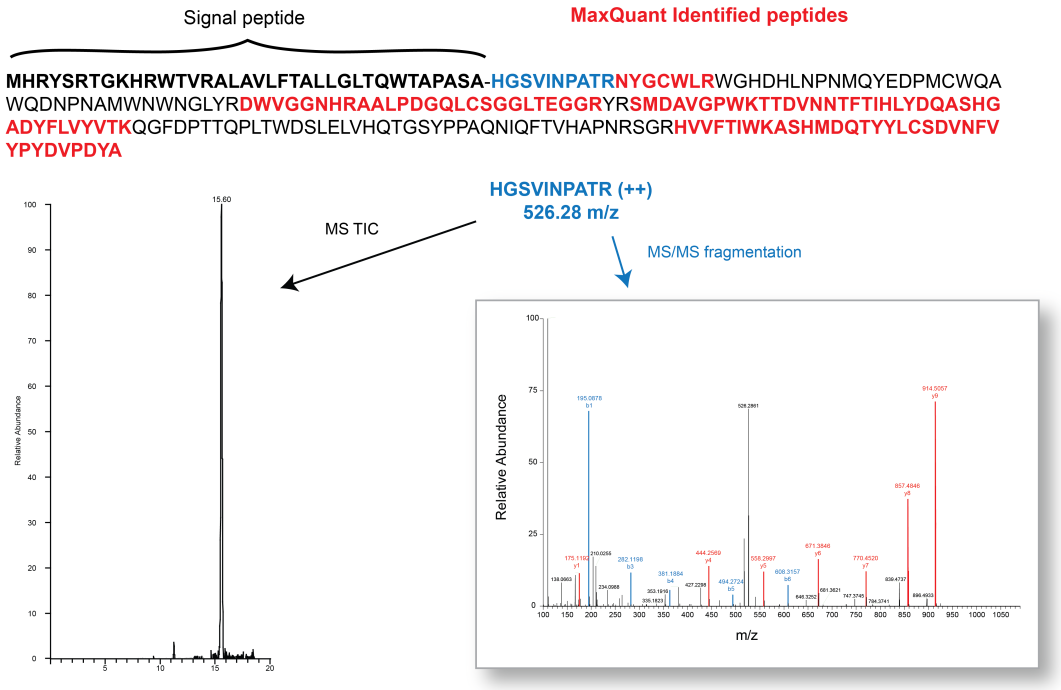

Figure S3. Identification of the secreted *Tf*AA10A by mass spectrometry. The inset highlights the identification of the mature N-terminal tryptic peptide confirming correct processing of the secreted *Tf*AA10A.

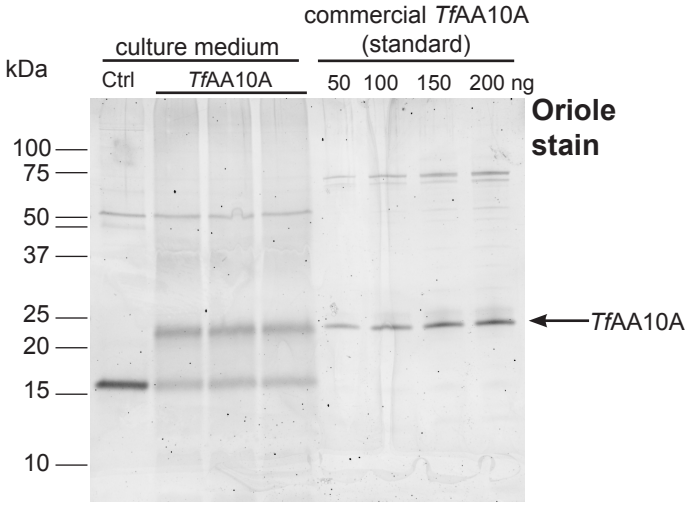

Figure S4. Densitometric analysis of secreted *Tf*AA10A. The first four lanes show culture medium loaded to an equivalent of 200 µL of cell-free medium. The subsequent four lanes show a standard curve of 50 – 200 ng of a commercial *Tf*AA10A. The *Tf*AA10A protein is indicated with an arrow. Ctrl denotes an empty vector control (pDF-trc).

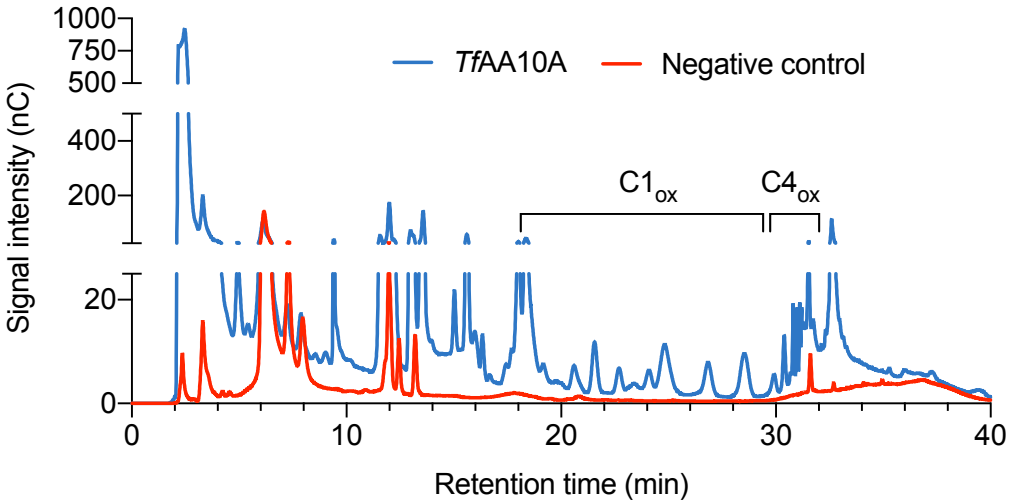

Figure S5. PASC oxidation by *Tf*AA10A. *Tf*AA10A catalyzed oxidation of PASC was performed in 50 mM phosphate buffer (pH 7.5). All reaction mixtures contained 2 mM ascorbate, 0.75% w/v PASC and 2 µg purified and reconstituted *Tf*AA10A. The negative control omitted the enzyme. Reactions were carried out at 50 °C for 24 hours. $C1_{ox}$ – C1 oxidation products. $C4_{ox}$ – C4 oxidation products. Results are representative of n = 3.