New Phytologist

***New Phytologist* Supporting Information**

Article title: **Mutational studies of the Aux/IAA proteins in Physcomitrella reveal novel insights into their function**
Authors: S. Tao and M. Estelle
Article acceptance date: 09 January 2018

The following Supporting Information is available for this article:

Fig. S1 qPCR of the *Aux/IAA* gene expression level in the wild-type moss protonemata.

Fig. S2 Yeast 2-hybrid analysis of the effects of IAA1a mutations.

Table S1 Primers used in this study

Fig. S1 qPCR of the *Aux/IAA* gene expression level in the wild-type moss protonemata treated with mock (-) and 10 μM IAA (+). All the three *Aux/IAA* genes encoding the auxin signaling repressors are up-regulated upon auxin treatment, indicating a negative feedback of the signal transduction. Error bars represent SEM. "a" difference between mock and IAA-treated samples is significant at $p<0.05$ (Student's t-test), n=4.

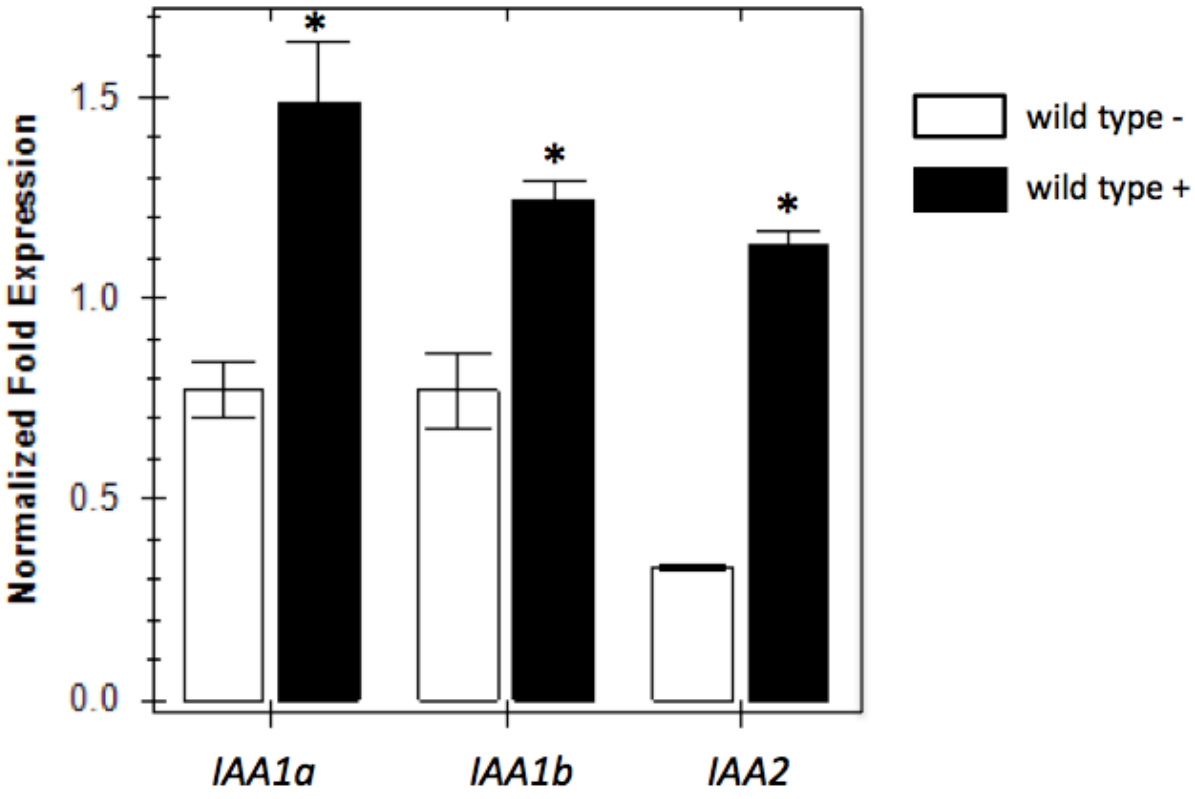

Fig. S2 Yeast 2-hybrid analysis of the effects of IAA1a mutations on protein interaction. (a) IAA1a$^{\Delta ear}$ did not show interaction with the moss TPL1 or TPL2 proteins. (b) K and OPCA mutations disrupted the IAA1a self-interaction and its interaction with moss ARFa8. (c) IAA1a^{K362A} did not show self-interaction but still interacted with proteins that had intact PBI domain like ARFa8 and IAA1a. EV: empty vector expressing only the Y2H DNA-binding domain or activation domain.

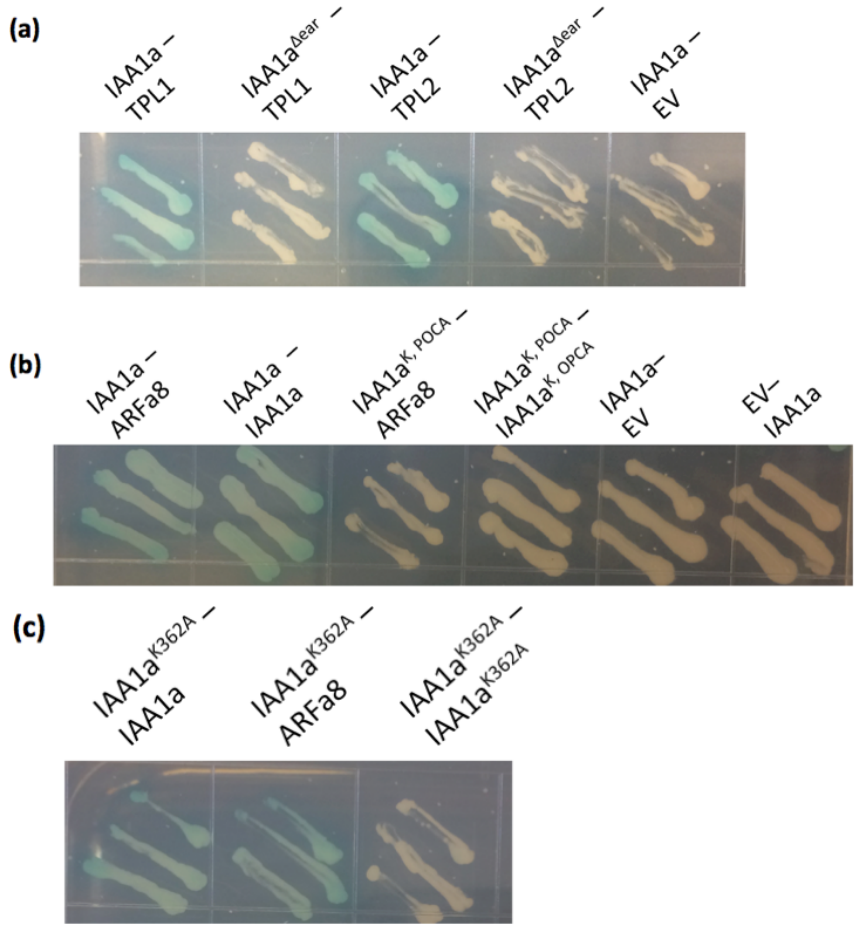

Table S1 Primers used in this study

Primer name	Sequence	Usage
PstI-Luc-F	CGCGACCTGCAGATGGAAGACGCCAAAAACAT	Cloning of *Luciferase*
Luc-XbaI-R	CGTCTGTCTAGATTACACGGCGATCTTTCCG	
PstI-IAA1a-F	CGCGACCTGCAGATGAACGTCAGCGAAGGTTG	Cloning of *IAA1a*
IAA1a-PstI-R	ATGCCTGCAGCCCCACCGCCACTTGC	
HindIII-IAA1a5'g-F	ATGCAAGCTTGATTGAAGAGTGGCAGTGGGTG	Cloning of *IAA1a* 5' genomic DNA
IAA1a5'g-HindIII-R	CGTCTGAAGCTTTGAATAATGTCCAACTTCACTGCTAAAAG	
SpeI-IAA1a3'g-F	CACCACTAGTGGAGATTGCAATGGCGGGATGTGCG	Cloning of *IAA1a* 3' genomic DNA
IAA1a3'g-SpeI-R	CCACTAGTCGTAAAATACTTACAATCATGAAATCCTTCCC	
K325R-F	GTGGGTTGGCCACCCGTGAGGAACTTCAACAAGATGAAC	IAA1aK325R mutagenesis
K325R-R	GTTCATCTTGTTGAAGTTCCTCACGGGTGGCCAACCCAC	
VK-F	GTGGGTTGGCCACCCATCGGGAACTTCAACAAGATGAAC	V324IK325G mutagenesis
VK-R	GTTCATCTTGTTGAAGTTCCCGATGGGTGGCCAACCCAC	
K326A-F	CGAGCGGGAACCTTGTGGCGATCTACATGGATGGTGTG	IAA1aK362A mutagenesis
K362A-R	CACACCATCCATGTAGATCGCCACAAGGTTCCCGCTCG	
D435A-F	GTGTTGATATACGAGGCTCACGAGGGAGACTCG	IAA1aD435A mutagenesis
D435A-R	CGAGTCTCCCTCGTGAGCCTCGTATATCAACAC	
D439A-F	GGCTCACGAGGGAGCCTCGATGCTGGTCGG	IAA1aD439A mutagenesis
D439A-R	CCGACCAGCATCGAGGCTCCCTCGTGAGCC	
ear-F1	GAACGAAAGTGCCTCGGCCCTCCTGCGGC	IAA1aΔear mutagenesis
ear-R1	GCCGCAGGAGGGCCGAGGCACTTTCGTTC	
ear-F2	GAATGGAACAGAGCAGCTGGCACTTGCCGAAAGGC	IAA1aΔear mutagenesis
ear-R2	GCCTTTCGGCAAGTGC CAGCTGCTCTGTTCCATTC	
EF1α-F	ACGCGTTGTTGGCTTTCACTTTG	qPCR
EF1α-R	GTGGTTGCGTCCATCTTGTTGC	qPCR
IAA1a-F	ATCCGGGAGTCCGAGCTTC	qPCR
IAA1a-R	GGTTCTGCGCAGGAGGTG	qPCR
IAA1b-F	CGGTGGTCAGAATGGGTCA	qPCR
IAA1b-R	CCCACAGTCTGGTTCTGCG	qPCR
IAA2-F	TGCCTTGGGACTGGTTCATC	qPCR
IAA2-R	CACAGCACCTTGGGCTTTCA	qPCR
CBS-F	TTGGAAAGACCGCCCAGCTATC	qPCR
CBS-R	GCTCCGTTAAACTCTCAGAACCAC	qPCR
Dox-F	AGCGATCCCACCAAATTCAGCTC	qPCR
Dox-R	TGAACAACGTGGGCTCCAATCC	qPCR
RSL4-F	TCAAACGGCCGAAACATTCTACG	qPCR
RSL4-R	CAGCTCCGCTCCTTTCAGAATATG	qPCR