

# *NtMYB3*, an R2R3-MYB from *Narcissus*, Regulates Flavonoid Biosynthesis

Muhammad Anwar <sup>1,†</sup>, Weijun Yu <sup>1,†</sup>, Hong Yao <sup>1</sup>, Ping Zhou <sup>1</sup>, Andrew C. Allan <sup>2,3</sup>, and Lihui Zeng <sup>1,\*</sup>

<sup>1</sup> College of Horticulture, Fujian Agriculture and Forestry University, Fuzhou 35002, China; anwar\_uar@yahoo.com (M.A.); ywj973618858@hotmail.com (W.Y.); yaohong116@hotmail.com (H.Y.); zhoup1249897684@hotmail.com (P.Z.)

<sup>2</sup> The New Zealand Institute for Plant & Food Research, Mt Albert Research Centre, Private Bag 92169, Auckland, 1025 New Zealand; andrew.allan@plantandfood.co.nz

<sup>3</sup> School of Biological Sciences, University of Auckland, Private Bag 92019, Auckland 1142, New Zealand

\* Correspondence: lhzen@hotmail.com Tel.: +86-591-83789450

† These authors contributed equally to this work.

## Supplementary Material

**Supplemental Table 1.** Analysis of putative cis-acting elements in *NtFLS* promoter.

Motif	Function	Stand	Position <sup>a</sup>	Position(5' - 3')
5' UTR Py-rich stretch	<i>cis</i> -acting element conferring high transcription levels	+	-552	TTTCTCTC TCTCTC
TATA-box	core promoter element around -30 of transcription start	+	-831, -884, -895, -908, 900 -883, -909 -908	TATA ATATAT TATATA
		-	-63 -63 -908	TATA TATAA TATATAA
CAAT-box	common cis-acting element in promoter and enhancer regions	+	-446	CCAAT
		-	-627, -832, -466 -515 -910, -860, -619, -650, -331	CAAT CAAAT CAAT CAAAT
3-AF1 binding site	Light responsive element	-	-421	TAAGAGA GGAA
Box II	part of a light responsive element	+	-213	CCACGTGG C
G-Box	cis-acting regulatory element involved in light responsiveness	-	-215	CACGTG
G-box	cis-acting regulatory element involved in light responsiveness	+	-81	CACGTG
TCT-motif	part of a light responsive element	+	-402	TCTTAC
ACE	cis-acting element involved in light responsiveness	-	-107	ACTACGTT GG
Box I	Light responsive element	+	-199	TTTCAAA
Myb-binding site	MYB combination element	+	-682	CAACAG
MYB	MYB combination element	+	-682	CAACAG

ABRE	cis-acting element involved in the abscisic acid responsiveness	+	-215	ACGTG
		-	-106, -82	ACGTG
ABRE2		-	-214	CCACGTGG
ABRE3a		-	105	TACGTG
ABRE4		+	105	CACGTA
		-	-140	CAACCA
P-box	Gibberellin response element	-	-354	GACCAAA CTCGT
O2-site	cis-acting regulatory element involved in zen metabolism regulation	-	-532	GATGATAT GC
TC-rich repeats	cis-acting element involved in defense and stress responsiveness	+	-497	ATTCTCTA AC
		+	-407	GTTTCTT AC
CAT-box	cis-acting regulatory element related to meristem expression	+	-313	GCCACT
Skn-1_motif	cis-acting regulatory element required for endosperm expression	-	-825	GTCAT

Supplemental Table 2. List of primers used in this studies.

Name	Forward primer (5' to 3')	Reverse primer (5' to 3')	Note
<i>MYB3</i>	AATATGGGTAGGTCTCCTTGTT GTG	GATTACAGGACACGCAAAGTAAATCTA	Cloning of full length of ORF
pSAK- <i>MYB3</i>	TGGATCCAAAGAATTCAATAT GGGTAGGTCTCCTTGTTGTG	TACTCTCGAGAAGCTTGATTACAGGAC ACGCAAAGTAAATCTA	Vector construction
QRT <i>NtCHS</i>		AAATTGCCTCTCTTGAGATCGG ACCTATAATGACCGCGGCTG	
<i>NtCHI</i>	TGGGTAGGTCTCCTTGTT	CAACGTTGACAACATCAGGC	
<i>NtF3H</i>	ACTCCGGATGGCTAAGGACTG TG	CCTTGGTTAAGGCCTCCTTC	
<i>NtFLS</i>	GAAATCCTCCGATCCAGTGA ACAGGGTGAAGTGGTCCAAG	TCCCTGTAGGAGGGAGGATT	
<i>NtLAR</i>	GAACTTGAAGGGAAAAGGGG TCAAGGTCCTTTACGCCATC	ACGAACCTGCTTCTCTTTGG	qRT-PCR analysis
<i>NtANR</i>	CATTTGACTTTCCCAAACGC GGGAATGAAGCTCACTACAGC	ATTGGGCTTTTGAGTTGTGC	
<i>NtDFR</i>	GAGTGCATTGGATGCCTTTT TGCGTTGAAGCTCATACTG	ACTCCGGCCATTTCTCTTGG	
<i>NtUFGT</i>	AATGGAAGTGAATGGTCAAG GC	CCAGCTCCATTAGGTCCTTG	
<i>NtANS</i>		GGAATTAGGCACACACTTGC	
<i>NtACT</i>		TGCCAGATCTTCTCCATGTCATCCCA	