J. Plant Biol. (2010) 53:233–239
DOI 10.1007/s12374-010-9110-2

ORIGINAL RESEARCH

Overexpression and Inactivation of *UGT73B2* Modulate Tolerance to Oxidative Stress in *Arabidopsis*

In A Kim · Jung-Ok Heo · Kwang Suk Chang · Shin Ae Lee · Mi-Hyun Lee · Chae Eun Lim · Jun Lim

Received: 14 March 2010 / Revised: 19 April 2010 / Accepted: 4 May 2010 / Published online: 19 May 2010
© The Botanical Society of Korea 2010

Abstract Glycosyltransferases (GTs) play an important role in modulating solubility, stability, bioavailability, and bioactivity of secondary metabolites, such as flavonoids. In *Arabidopsis thaliana*, at least 120 family 1 uridine diphosphate (UDP)-glycosyltransferases (UGTs) have been predicted. However, little is known about their substrates or their physiological roles *in planta*. To define the role of UGT73B2 *in planta*, we first characterized its expression pattern using transgenic *Arabidopsis* plants carrying the *cis*-elements of *UGT73B2* fused to the GUS reporter. During vegetative phase, its expression was high in embryonic and postembryonic roots, where it may play a physiological role in the glycosylation of flavonoids. Loss of function of *UGT73B2* alone or in conjunction with its closest homologs, *UGT73B1* and *UGT73B3*, confers greater tolerance to oxidative stress, whereas overexpression of *UGT73B2* increases sensitivity to oxidative stress. In addition, growth phenotypes of mutant and transgenic seedlings correlate well with ROS levels *in planta*. Our results suggest that the glycosylation of flavonoids by *UGT73B2*—and/or its closest homologs—modulate the response of plants to oxidative stress.

Keywords *Arabidopsis* · Flavonoids · Glycosylation · Methyl viologen · Oxidative stress · UGT73B2

In A Kim and Jung-Ok Heo contributed equally to this work.

I. A. Kim · J.-O. Heo · K. S. Chang · S. A. Lee · M.-H. Lee · J. Lim (✉)
Department of Bioscience and Biotechnology, Konkuk University, 1 Hwayang-dong, Gwangjin-gu, Seoul 143-701, South Korea
e-mail: jlim@konkuk.ac.kr

K. S. Chang · J. Lim
Bio/Molecular Informatics Center, Konkuk University, 1 Hwayang-dong, Gwangjin-gu, Seoul 143-701, South Korea

C. E. Lim
Vascular Plants Research Division, Biological Resources Research Department, National Institute of Biological Sciences (NIBR), Environmental Research Complex, Gyeongseo-dong, Seo-gu, Incheon 404-708, South Korea

Introduction

Due to their sessile nature, plants need to cope with a variety of environmental stresses: abiotic and biotic. To allow plants greater flexibility and plasticity under various stress conditions, it is essential that a variety of small molecules, either intrinsic or extrinsic, including hormones, metabolites, and xenobiotics, be dynamically modified (Dixon and Paiva 1995; Hemm et al. 2004). In particular, glycosylation catalyzed by a superfamily of glycosyltransferases (GTs), plays an important role in modulation of the solubility, stability, bioavailability, and bioactivity of various small molecules (Heller and Forkmann 1994; Jones and Vogt 2001; Ross et al. 2001). In plants, secondary metabolites such as flavonoids perform multiple functions, including roles in oxidative stress, UV protection, and disease resistance (Chong et al. 2002; Gachon et al. 2005; Langlois-Meurinne et al. 2005; Li et al. 1993).

In the *Arabidopsis* genome, at least 120 family 1 uridine diphosphate (UDP)-glycosyltransferases (UGTs), which transfer UDP-activated sugar moieties to specific acceptor molecules, have been predicted (Li et al. 2001; Ross et al. 2001). Phylogenetic analyses revealed that the family 1 UGTs were classified into 14 groups: A to N (Bowles 2002; Li et al. 2001; Ross et al. 2001). To date, no definite conclusion on the function of these UGTs has been made from their primary sequence information (Langlois-Meurinne

et al. 2005). It is generally believed that plants synthesize a vast array of natural products to accommodate abiotic and biotic stresses and that, in many cases, a glycosylation reaction by a UGT is the last step in the biosynthesis of natural products (Paquette et al. 2003). Thus, the evolution and explosion of UGTs in the *Arabidopsis* genome to cope with constant changes in environment is reasonable.

One of the 14 UGT groups, group D is composed of 13 UGT members, which generally exhibit high substrate specificity for small compounds, such as flavonoids, with regioselectivity (Heller and Forkmann 1994; Kim et al. 2006; Lim et al. 2002; Vogt and Jones 2000). It has been reported that flavonoids serve as antioxidants to reduce production of reactive oxygen species (ROS) (Asada and Takahashi 1987; Bowler et al. 1992). In the group D UGTs, UGT73B2 was previously reported to preferentially transfer a glucose group to the 3-hydroxyl group of flavonoids *in vitro* (Kim et al. 2006), and loss-of-function mutations in *UGT73B1*, *UGT73B2*, or *UGT73B3* (located in tandem on the same chromosome) enhanced resistance to oxidative stress (Lim et al. 2006). In addition, Langlois-Meurinne et al. (2005) demonstrated the potential role of UGT73B3, along with another group D member UGT73B5, in defense responses to *Pseudomonas*.

The aim of this study was to determine the physiological role of UGT73B2 in a subfamily of group D because recent work demonstrated that UGT73B2 used flavonoids as substrates to transfer a glucose moiety from UDP-glucose to the 3-hydroxyl group of flavonoids *in vitro* (Kim et al. 2006). We thus characterized mutant and transgenic *Arabidopsis* plants that exhibited the modulated expression levels of *UGT73B2* and its closest homologs (*UGT73B1* and *UGT73B3*). Under oxidative stress, loss-of-function of either *UGT73B2* or all three genes (*UGT73B1*, *UGT73B2*, and *UGT73B3*) confers greater tolerance to the stress, whereas overexpression of *UGT73B2* causes slight, but significant, sensitivity to oxidative stress. In addition, growth phenotypes of mutant and transgenic seedlings correlate well with ROS levels *in planta*. Our results suggest that the glycosylation of flavonoids by UGT73B2, probably also by its closest homologs (*UGT73B1* and *UGT73B3*), modulates plants' response to oxidative stress.

Materials and Methods

Plant Materials, Growth Conditions, Oxidative Stress Treatments, and Quantitative RT-PCR (qRT-PCR)

In this study, *Arabidopsis thaliana* ecotype Columbia (Col-0) and its derivatives were used. A loss-of function mutation in *UGT73B2* (At4g34135) (SALK_043888) was previously described (Lim et al. 2006). Seeds were surface-sterilized in 5% sodium hypochlorite and 0.15% Tween 20 for 3 min, rinsed thoroughly in distilled water, and planted on MS agar plates (1X Murashige-Skoog salts, pH 5.7; 1% sucrose; and 0.8% agar). They were stratificated for 3 days at 4°C and grown vertically under continuous light conditions at 22°C, as previously described (Lim et al. 2006; Lim et al. 2008).

To induce oxidative stress, plants were grown on 1× MS agar plates containing 0, 0.5, or 1 μM of methyl viologen (MV; paraquat; 1, 1'-dimethyl-4, 4' bipyridinium dichloride, Aldrich, USA). The root lengths and fresh weights of the seedlings ($n \geq 50$) were measured using the Image J program (NIH Image, USA). To score the survival rates, plants were first grown in MS agar plates for 3 days (as mentioned above) and were transferred to new MS agar plates with 3 μM MV. At 7 days after transfer (DAT), only surviving plants were counted ($n > 260$).

For qRT-PCR, total RNA was isolated from seedlings grown in MS agar plates (with or without MV), using an RNeasy Plant Mini kit (Qiagen, USA) according to the manufacturer's instruction. Using the Mx3000P® QPCR System (Stratagene, USA), qRT-PCR experiments were carried out as described previously (Lee et al. 2008; Lim et al. 2008). Sequence information for primers used in PCR is listed in Table 1.

Transgenic Plants, Transformation, and GUS Assay

To generate overexpression lines, the full-length (1,452 bp) open reading frame (ORF) of the *UGT73B2* gene was amplified by PCR using gene-specific primers, as listed in Table 1. Subsequently, the amplified product was subcloned into pENTR Directional TOPO™, a Gateway entry vector (Invitrogen, USA). Only an error-free insert was transferred to the pEarley100 binary vector (Earley et al. 2006) containing the cauliflower mosaic virus (CaMV) 35S promoter using LR recombinase, according to the manufacturer's instructions (Invitrogen, USA).

To produce *pUGT73B2::GUS* transgenic plants, an approximately 1.2-kb fragment upstream from the ATG start codon of *UGT73B2* was amplified by PCR and subcloned into pBI101 as described in Lim et al (2008) with modifications. The PCR product was cut with *Pst*I and *Bam*HI, and the digested *Pst*I-*Bam*HI fragment was inserted into the multicloning sites (MCS) of pBI101 cut with *Pst*I and *Bam*HI. Transgenic lines containing the *pUGT73B2::GUS* construct were selected and analyzed for GUS activity, as previously described (Lee et al. 2008).

For RNA interference (RNAi), we selected the most highly similar region among the three closely related genes: *UGT73B1* (At4g34138), *UGT73B2* (At4g34135), and *UGT73B3* (At4g34131). The 0.9-kb PCR-amplified partial fragment of the GUS gene was subcloned into the

Table 1 Primer sequences used in this study

Primer name	5′-3′	Primer name	5′-3′	Use
UGT73B2-F	CACCATGGGTAGTGATCATCATCGA	UGT73B2-R	AACGTCAATTTCAATACCTTTGGT	*35S::UGT73B2*
pUGT73B2-F	AACTGCAGAACCAATGCATTGGATCAGGACGCAAACCCAACA	pUGT73B2-R	CGGGATCCCGGATTGGCTGGTTGAAAAGAAAAC	*pUGT73B2::GUS*
NotI-73B-F	GCGGCCGCCTTCCCTTTCATGGCTTATGGTC	BamHI-73B-R	GGATCCTGTCGGCGATAAGACAGTCTGGTC	RNAi
KpnI-73B-F	GGTACCCTTCCCTTTCATGGCTTATGGTC	EcoRI-73B-R	GAATTCTGTCGGCGATAAGACAGTCTGGTC	RNAi
ACT2-F	TCGCTGACCGTATGAGCAAAGAA	ACT2-R	TGGAATGTGCTGAGGGAAGCA	qRT-PCR
UGT73B2qRT-F	AGAGGCGGAGACGGGCAAA	UGT73B2qRT-R	TGAACTAAACTCTTCCATGAAGC	qRT-PCR

pBluescript SK vector, using *Bam*HI and *Eco*RI sites (pBSK-dGUS). Based on the sequence information of the most conserved region between *UGT73B2* and *UGT73B3*, a 359-bp fragment of *UGT73B2* was amplified with *Not*I-*Bam*HI (forward) and *Kpn*I-*Eco*RI (reverse) linker primers (Table 1). The fragment was cut and subcloned into the pBSK-dGUS vector. Subsequently, the cassette of *UGT73B2* and dGUS was cut with *Xba*I and transferred into the pMBP1 binary vector (Suh et al. 1998) for RNAi *in planta*.

Plant transformation was performed by the floral-dip method (Clough and Bent 1998) with Col-0 wild-type plants for overexpressor and transcriptional fusion, and *ugt73b1* for RNAi, respectively. Transgenic plants in the T_3 generation were used for analysis throughout our experiments.

Analysis of ROS Production by NBT Assays

To qualitatively estimate ROS generation, we performed NBT (nitroblue tetrazolium) staining in mutant and transgenic seedling roots as described by Blanvillain et al. (2009) with minor modifications. At 10 days after germination (DAG), seedlings grown under oxidative stress (0.5 μM MV) were stained for 3 h with 0.5 mg ml^{-1} NBT in 1× phosphate-buffered saline (PBS). Plants were fixed with 70% methanol prior to observation, and pictures were obtained using an Axio Imager. A1 microscope equipped with an AxioCam MRc5 digital camera (Carl Zeiss, Germany).

Results and Discussion

Tissue-Specific Expression Patterns of *UGT73B2*

To define the role of *UGT73B2 in planta*, we analyzed its expression pattern with transgenic *Arabidopsis* plants harboring the *cis*-elements of *UGT73B2* fused to the GUS reporter gene (*pUGT73B2::GUS*). Expression of *pUGT73B2::GUS* was primarily observed in the seedling root, specifically in the stem cell niche where the quiescent center (QC) and the initial cells are located, and in the mature root cap cells (Fig. 1a, b). No expression was observed in the shoot meristem or cotyledons (Fig. 1a). In the mature embryo, blue staining was detected in the radicle (Fig. 1c). We also found GUS expression in the stem cell niche and from the endodermis onward in the lateral root (Fig. 1d). In aerial parts, we found GUS expression in the anthers and sepals of the flower (Fig. 1e–g). Our GUS analysis revealed that *UGT73B2* is expressed highly in embryonic and postembryonic roots as well as in flowers with tissue-specific patterns. Based on the tissue-specific expression patterns of *UGT73B2,* we speculate that its major physiological role takes place in roots during the vegetative phase and in anthers and sepals during the reproductive phase.

Characterization of *ugt73b2* and Transgenic Plants Under Oxidative Stress

To better understand the physiological role of UGT73B2 in *Arabidopsis*, we generated and characterized overexpressors of *UGT73B2* driven by the cauliflower mosaic virus (CaMV) 35S promoter along with loss-of-function *ugt73b2* mutants that were previously described (Lim et al. 2006). The expression level of *UGT73B2* in the overexpressors (OX) was substantially high by approximately sevenfold as compared with that of the wild-type (data not shown). It was previously reported that two closest homologs (*UGT73B1* and *UGT73B3*) are located in tandem with *UGT73B2* on the same chromosome (Lim et al. 2006). In addition, expression of the *UGT* genes was induced in different mutant backgrounds. For example, *UGT73B1* expression substantially increased in *ugt73b2* mutants, suggesting possible redundancy in their function. Thus, we attempted to produce plants with a loss of function of all three genes by genetically crossing each other; but, our attempts were unsuccessful because the genes are very tightly linked on the same chromosome. Alternatively, we have generated transgenic plants with substantially decreased expression of all three genes by RNA interference (RNAi) (see Materials and Methods).

Previously, it was shown that *ugt73b2* displayed enhanced resistance to oxidative stress caused by methyl viologen (MV), which is known to promote ROS generation (Lim et al. 2006; Mylona et al. 2007). Under standard growth conditions, *ugt73b2*, overexpressor (OX), and RNAi plants showed no obvious morphological defects, but *ugt73b2* and RNAi appeared to grow better than the wild-type (Fig. 2a–c). We further investigated growth phenotypes of OX and RNAi seedlings in comparison with *ugt73b2* and wild-type under oxidative stress. As MV concentrations increased, the growth of wild-type, *ugt73b2*, OX, and RNAi seedlings was inhibited to varying degree compared with those grown under control conditions (Fig. 2a). We measured the root lengths and fresh weights of wild-type, *ugt73b2*, OX, and RNAi seedlings. We found that the root length of RNAi plants increased, as was that of *ugt73b2* single mutants, which have been shown to be tolerant to MV (Fig. 2a, b). In addition, fresh weights of both *ugt73b2* and RNAi were more than those of wild-type and OX seedlings (Fig. 2c). Collectively, *ugt73b2* and RNAi grew better than wild-type and OX seedlings under MV-induced oxidative stress (Fig. 2a–c). These results indicate that reduced function of either *UGT73B2* alone—or in conjunction with the two homologs—confers relatively resistant growth phenotypes to oxidative stress. Taken together, these results suggest that *UGT73B2* plays a physiological role in a glycosylation pathway in response to oxidative stress.

Characterization of Stress Tolerance Under Extreme Oxidative Stress

Since both *ugt73b2* and RNAi seedlings grew better than wild-type and OX under the MV-induced oxidative stress, we next investigated the adaptive fitness of the plants under extreme conditions. We grew wild-type, *ugt73b2*, OX, and

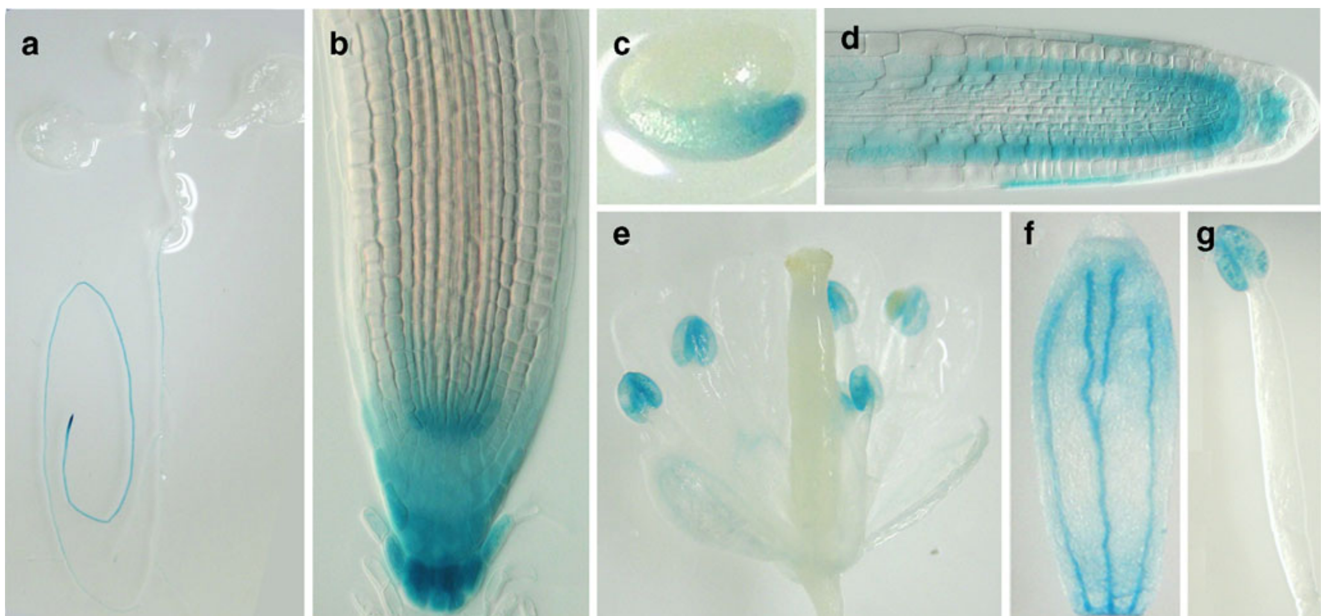

Fig. 1 Expression patterns of *UGT73B2* by *pUGT73B2::GUS* transcriptional fusion. **a** 10-day-old light-grown seedling. **b** Primary root. **c** Mature embryo. **d** Lateral root. **e** Flower. **f** Sepal. **g** Stamen

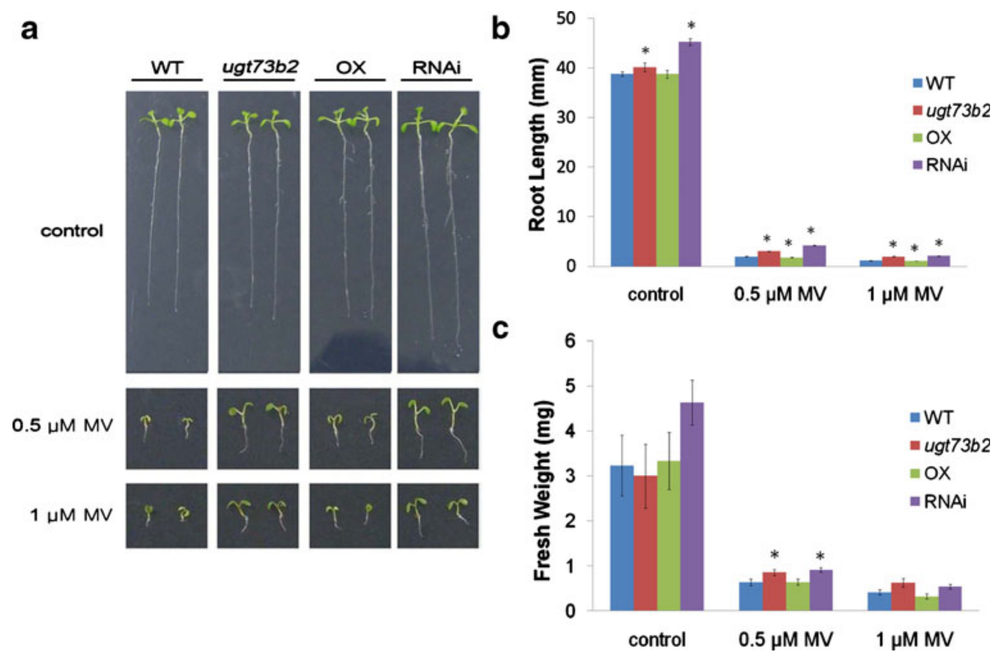

Fig. 2 Effects of MV-induced oxidative stress on wild-type, *ugt73b2*, overexpressor, and RNAi seedlings. **a** Phenotypes of 10-day-old light-grown seedlings at different concentrations. **b** Root lengths. **c** Fresh weights

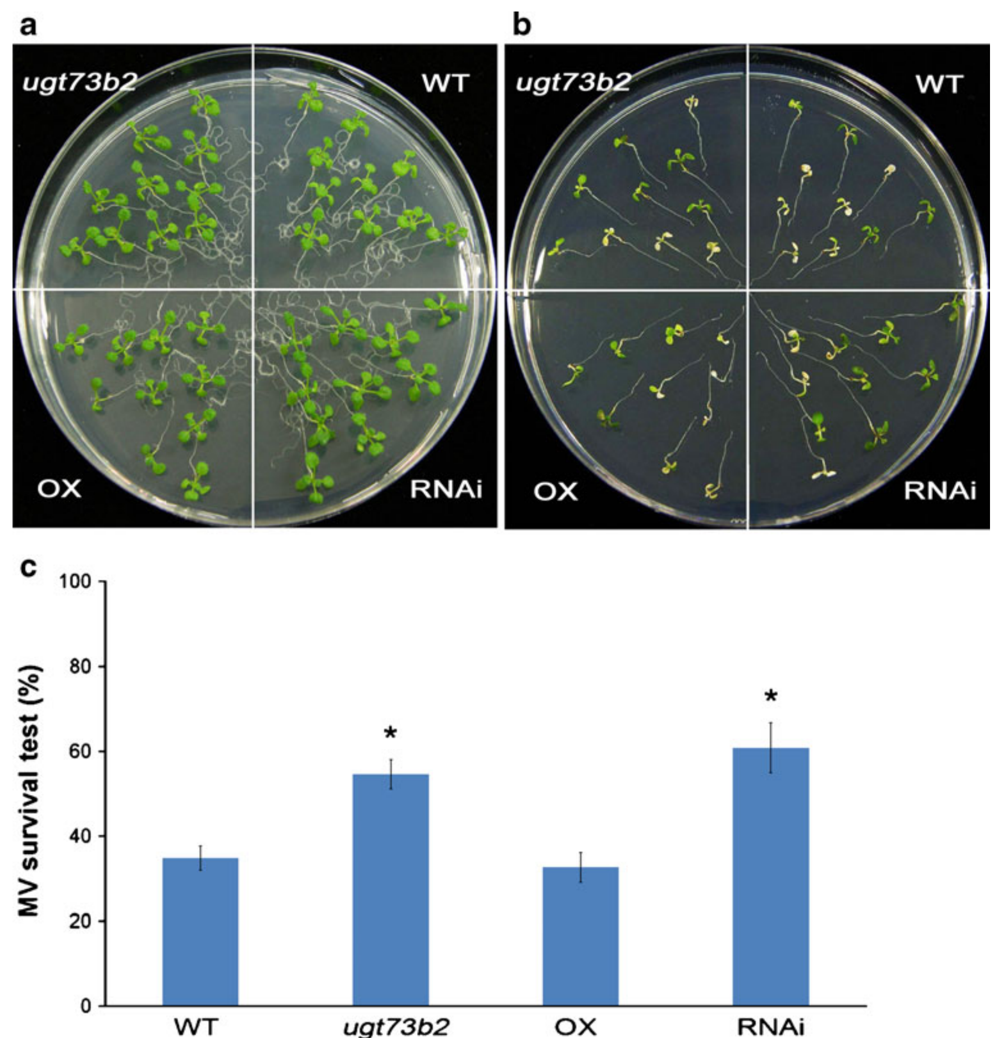

Fig. 3 Survival phenotypes under extremely adverse oxidative stress. **a** MS agar plate without MV. **b** MS agar plate with 3 μM MV. **c** Survival rates. Values that are statistically significant ($p<0.05$) are indicated by an *asterisk*

RNAi for 3 days in normal conditions and transferred the seedlings to extreme conditions (MV 3 µM; see Materials and Methods). At seven DAT, we scored the survival rates of wild-type, *ugt73b2*, OX, and RNAi plants. Under extremely adverse conditions, approximately 35% (119 out of 342) wild-type survived, whereas survival rates of both *ugt73b2* and RNAi significantly increased to approximately 55% (192 out of 352) and 61% (161 out of 265), respectively (Fig. 3a–c). It was previously suggested that smaller-sized individuals appear to have better adaptive fitness under extremely adverse conditions (Achard et al. 2006). Interestingly, our analysis showed that both *ugt73b2* and RNAi have better fitness under adverse conditions, even though these seedlings are larger. Loss of function of *UGT73B2* and its closest homologs also produce plants with greater fitness in extremely adverse conditions.

Analysis of ROS Production in Mutant and Transgenic Seedling Roots

To better understand the correlation between the reduced function of *UGT73B2* and ROS production, we monitored ROS production qualitatively by the standard NBT (nitroblue tetrazolium) staining assay. Under normal conditions, *ugt73b2* and RNAi showed similar staining patterns to those of wild-type and OX roots (Fig. 4a–d). However, we found that NBT staining substantially decreased in both *ugt73b2* and RNAi roots compared with roots of wild-type and OX under oxidative stress (Fig. 4e–h). These data indicate that ROS generation in both *ugt73b2* and RNAi is indeed reduced, which well correlates with tolerant growth phenotypes under oxidative stress.

It is well established that flavonoids, which are known to be substrates of UGT73B2 *in vitro*, act as antioxidants by reducing ROS production (Cotelle et al. 1992; Hanasaki et al. 1994; Jovanovic et al. 1994; Kim et al. 2006; Rice-Evans et al. 1996; Yuting et al. 1990; Zhou and Zheng 1991). In particular, it is thought that flavonoid aglycones are potentially more effective antioxidants than flavonoid glycosides (Rice-Evans et al. 1996). Thus, it is likely that loss-of-function of UGT73B2 alone and/or with the homologs is unable to efficiently convert aglycones to glycosides in a given pool of flavonoids. Relative accumulation of aglycones, more effective antioxidants, in the pool of flavonoids results in improved tolerance to oxidative stress caused by MV treatments. It is also probable that each of the three UGTs may act only on the subset of specific substrates in a given pool of flavonoids in a spatiotemporal-specific manner. Nevertheless, we can speculate that the glycosylation of flavonoids by *UGT73B2* (and/or its closest homologs) modulate plants' response to oxidative stress.

Conclusion

During the vegetative phase, the *UGT73B2* gene is expressed highly in embryonic and postembryonic roots, where it possibly plays a physiological role in the glycosyl-

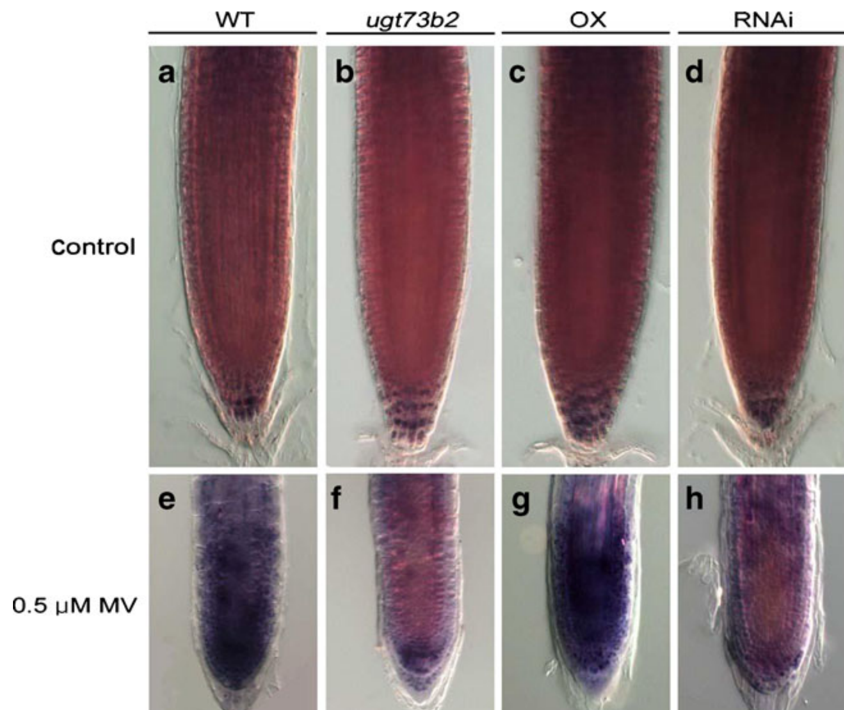

Fig. 4 NBT staining patterns of wild-type, *ugt73b2*, overexpressor, and RNAi roots. Seedlings grown in normal conditions (**a**–**d**). Seedlings grown in MS agar plates with 0.5 µM MV (**e**–**h**). **a**, **e** Col-0 wild-type. **b**, **f** *ugt73b2*. **c**, **g** overexpressor. **d**, **h** RNAi

ation of secondary metabolites, such as flavonoids. Loss of function of *UGT73B2* alone and/or with its two closest homologs (*UGT73B1* and *UGT73B3*) confers greater tolerance to oxidative stress, whereas overexpression of *UGT73B2* raises sensitivity to oxidative stress slightly, but significantly. Our results suggest that the glycosylation of flavonoids by *UGT73B2* (and/or its closest homologs) modulates the response of plants to oxidative stress.

Acknowledgments We thank the members of the Lim laboratory for helpful suggestions and discussions. This research was supported by grants from the National Research Foundation (NRF2009-0093824 and NRF2010-0015691).

References

Achard P, Cheng H, Grauwe LD, Decat J, Schoutteten H, Moritz T, Straeten DVD, Peng J, Harberd N (2006) Integration of plant responses to environmentally activated phytohormonal signals. Science 311:91–94

Asada K, Takahashi M (1987) Production and scanvenging of active oxygen in Photosynthesis. In: Kyle DJ, Osmond CB, Arentzen CJ (eds), Photoinhibition, Elsevier, pp. 227–287

Bowler C, Montagu MV, Inze D (1992) Superoxide dismutases and stress tolerance. Annu Rev Plant Physiol Mol Biol 43:83–116

Bowles D (2002) A multigene family of glycosyltransferases in a model plant, *Arabidopsis thaliana*. Biochem Soc Trans 30:301–306

Blanvillain R, Kim JK, Wu S, Lima A, Ow DW (2009) Oxidative stress 3 is chromatin-associated factor involved in tolerance to heavy metals and oxidative stress. Plant J 57:654–665

Chong J, Baltz R, Schmitt C, Beffa R, Fritig B, Saindrenan P (2002) Downregulation of a pathogen-responsive tobacco UDP-Glc: phenylpropanoid glucosyltransferase reduces scopoletin glucoside accumulation, enhances oxidative stress, and weakens virus resistance. Plant Cell 14:1093–1107

Clough S, Bent A (1998) Floral dip: a simplified method for *Agrobacterium*-mediated transformation of *Arabidopsis thaliana*. Plant J 16:735–743

Cotelle N, Bernier JL, Henichart JP, Catteau JP, Gaydou E, Wallet JC (1992) Scavenger and antioxidant properties of ten synthetic flavones. Free Radic Biol Med 13:211–219

Dixon RA, Paiva NL (1995) Stress-induced phenylpropanoid metabolism. Plant Cell 7:1085–1097

Earley KW, Haag JR, Pontes O, Opper K, Juehne T, Song K, Pikaard CS (2006) Gateway-compatible vectors for plant functional genomics and proteomics. Plant J 45:616–629

Gachon CMM, Langlois-Meurinne M, Saindrenan P (2005) Plant secondary metabolism glycosyltransferases: the emerging functional analysis. Trends Plant Sci 10:542–549

Hanasaki Y, Ogawa S, Fukui S (1994) The correlation between active oxygens scavenging and antioxidative effects of flavonoids. Free Radic Biol Med 16:845–850

Heller W, Forkmann G (1994) Biosynthesis of flavonoids. In: Harborne JB (ed) The flavonoids: advances in research since 1986. Chapman & Hall, London, pp 499–536

Hemm MR, Rider SD, Ogas J, Murry DJ, Chapple C (2004) Light induces phenylpropanoid metabolism in *Arabidopsis* roots. Plant J 38:765–778

Jones P, Vogt T (2001) Glycosyltransferases in secondary plant metabolism: tranquilizers and stimulant controllers. Planta 213:164–174

Jovanovic SV, Steenken S, Tosic M, Marjanovic B, Simic MG (1994) Flavonoids as antioxidants. J Am Chem Soc 116:4846–4851

Kim JH, Kim BG, Ko JH, Lee Y, Hur H-G, Lim Y, Ahn J-H (2006) Molecular cloning, expression, and characterization of a flavonoid glycosyltransferase from *Arabidopsis thaliana*. Plant Sci 170:897–903

Langlois-Meurinne M, Gachon CMM, Saindrenan P (2005) Pathogen-responsive expression of glycosyltransferase genes *UGT73B3* and *UGT73B5* is necessary for resistance to *Pseudomonas syringae* pv *tomato* in *Arabidopsis*. Plant Physiol 139:1890–1901

Lee MH, Kim B, Song SK, Heo JO, Yu NI, Lee SA, Kim M, Kim DG, Sohn SO, Lim CE, Chang KS, Lee MM, Lim J (2008) Large-scale analysis of the GRAS gene family in *Arabidopsis thaliana*. Plant Mol Biol 67:659–670

Li Y, Baldauf S, Lim E-K, Bolwes DJ (2001) Phylogenetic analysis of the UDP-glycosyltransferase multigene family of *Arabidopsis thaliana*. J Biol Chem 276:4338–4343

Li J, Ou-Lee TM, Raba R, Amundson RG, Last RL (1993) *Arabidopsis* flavonoid mutants are hypersensitive to UV-B irradiation. Plant Cell 5:171–179

Lim CE, Ahn JH, Lim J (2006) Molecular genetic analysis of tandemly located glycosyltransferase genes, *UGT73B1*, *UGT73B2*, and *UGT73B3*, in *Arabidopsis thaliana*. J Plant Biol 49:309–314

Lim CE, Choi JN, Kim IA, Lee SA, Hwang Y-S, Lee CH, Lim J (2008) Improved resistance to oxidative stress by a loss-of-function mutation in the *Arabidopsis UGT71C1* gene. Mol Cells 25:368–375

Lim E-K, Doucet CJ, Elias L, Worral D, Li Y, Ross J, Bowles DJ (2002) Activity of the group 1 glycosyltransferase of *Arabidopsis* towards salicylic acid, para-hydroxybenzoic acid and other benzoates. J Biol Chem 277:586–592

Mylona PV, Polidoros AN, Scandalios JG (2007) Antioxidant gene responses to ROS-generating xenobiotics in developing and germinated scutella of maize. J Exp Bot 58:1301–1312

Paquette S, Moller BL, Bak S (2003) On the origin of family 1 plant glycosyltransferases. Phytochemistry 62:399–413

Rice-Evans CA, Miller NJ, Paganga G (1996) Structure-antioxidant activity relationships of flavonoids and phenolic acids. Free Redic Biol Med 20:933–956

Ross J, Li Y, Lim E-K, Bowles DJ (2001) Higher plant glycosyltransferases. Genome Biol 2:3004.1–3004.6

Suh MC, Choi D, Liu JR (1998) Cadmium resistance in transgenic tobacco plants expressing the *Nicotiana glutinosa* L. metallothionein-like gene. Mol Cells 8:678–684

Vogt T, Jones P (2000) Glycosyltransferase in plant natural product synthesis: characterization of a supergene family. Trends Plant Sci 5:380–386

Yuting C, Rongliang Z, Zhongjian J, Yong J (1990) Flavonoids as superoxide scavengers and antioxidants. Free Radic Biol Med 9:19–21

Zhou YC, Zheng RL (1991) Phenolic compounds and an analogue as superoxide anion scavengers and antioxidants. Biochem Pharmacol 42:1177–1179