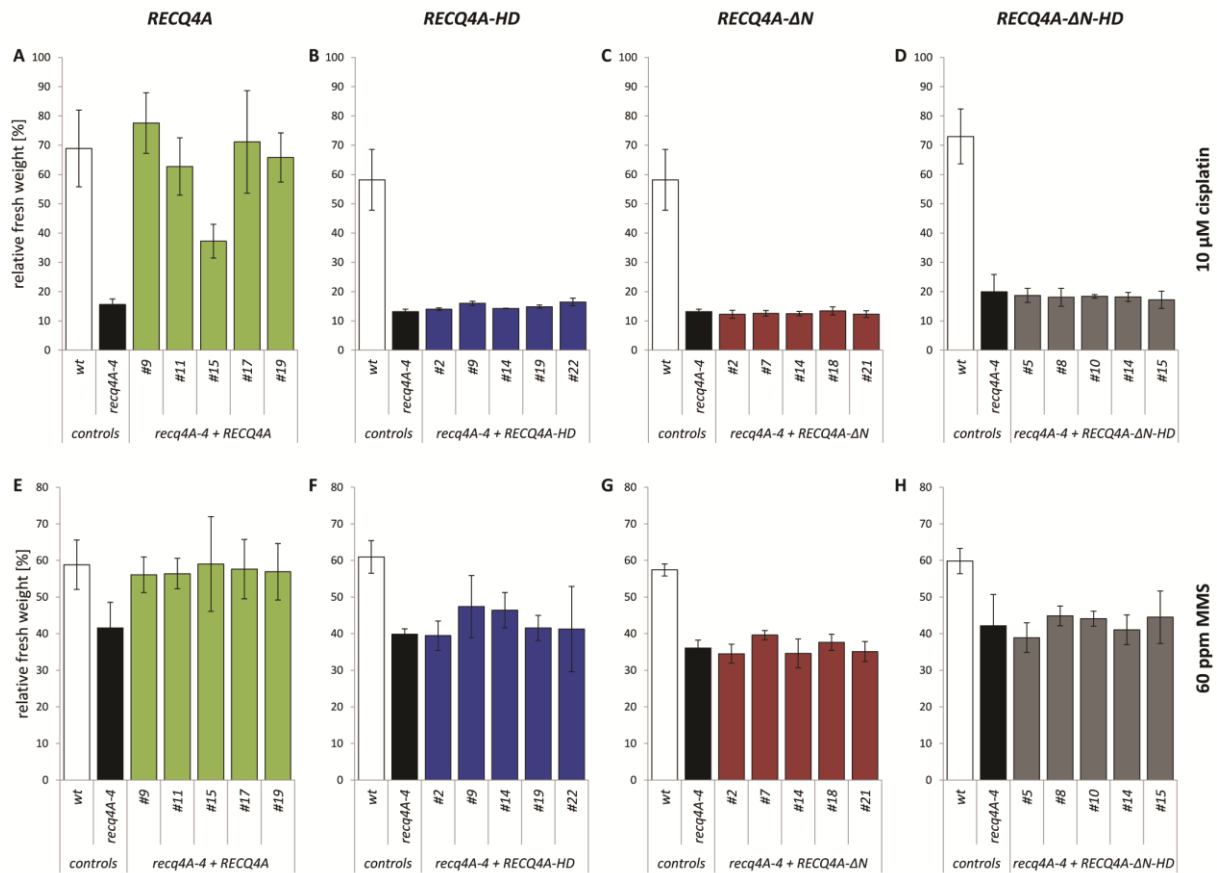
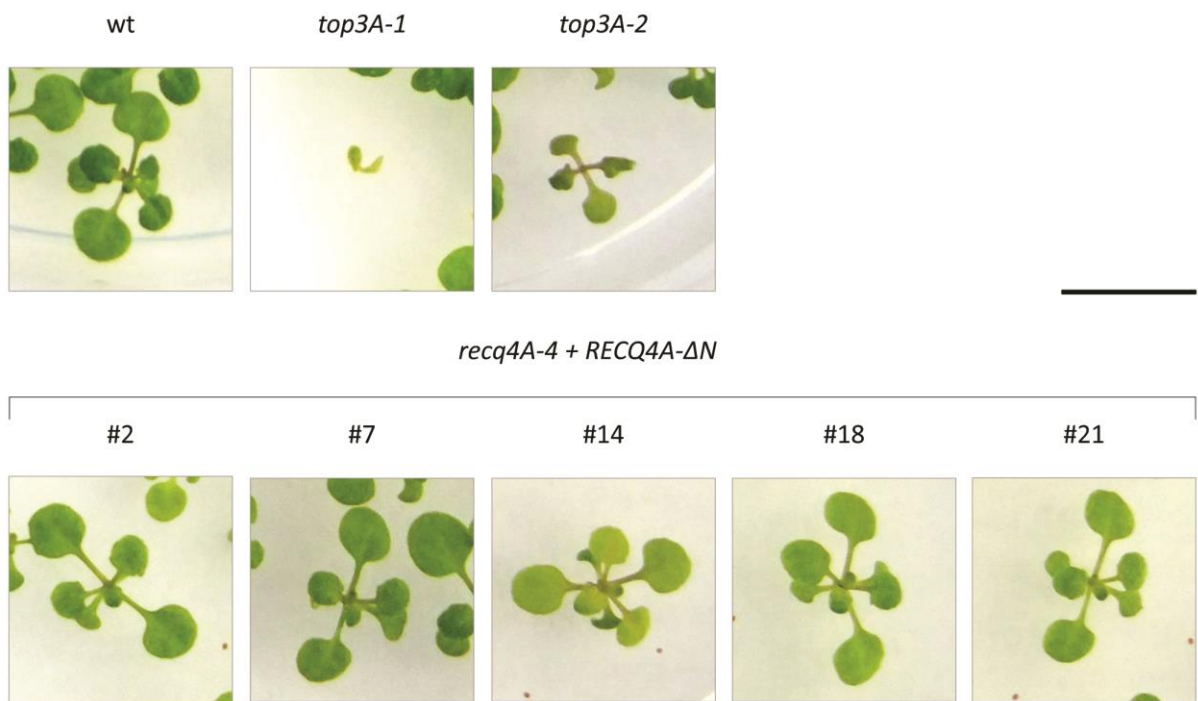


## SUPPLEMENTARY MATERIAL



**Figure S1. Functional analysis of the N-terminus and helicase activity of RECQ4A in the repair of cisplatin (10 μM) and MMS (60 ppm)-induced DNA damage.** The fresh weight of 10 seedlings after 13 days of genotoxin treatment (10 μM cisplatin (A-D), 60 ppm MMS (E-H)) was determined. The relative fresh weight of each line is given as percentage and was calculated from the relation of fresh weight of each line at a respective genotoxin concentration to the fresh weight of the same line without genotoxin treatment. Each assay was performed at least three times as described and the mean values including standard deviations (error bar) are depicted. The expression of the wild type construct *RECQ4A* in *recq4A-4* mutant background (*recq4A-4 + RECQ4A*, green) enables a full complementation of the elevated sensitivity of *recq4A-4* to cisplatin (A) and MMS (E). The constructs *RECQ4A-HD* (blue, B, F), *RECQ4A-ΔN* (red, C, G) and *RECQ4A-ΔN-HD* (grey, D, H) cannot complement the hypersensitivity of *recq4A-4* to cisplatin and MMS.



**Figure S2. Growth phenotypes of *top3A* mutant lines and *recq4A-4* lines transformed with *RECQ4A-ΔN*.** Images of representative 16-days-old plant seedlings of wild type, homozygous *top3A-1* and *top3A-2* mutants as well as different *recq4A-4* mutant lines transformed with *RECQ4A-ΔN* growing on agar plates are shown. After germination and formation of cotyledons, the *top3A-1* mutant stops to grow. *top3A-2* mutant plants are smaller than wild type plants and show malformed, curled leaves. The growth of *recq4A-4* mutant lines expressing *RECQ4A-ΔN* is comparable with wild type and they did not show any growth defects. Scale bar = 1 cm.

**Table S1. Cloning PCRs and primers used to create plasmids for *in vivo* studies.** pPZP221 was used as a basis for the construction of the plant transformation vectors. The different *RECQ4A* constructs consist of a promoter region, a coding region and a terminator region. Amplification of the respective DNA fragment was performed by PCR using the depicted primer combinations and PCR templates. For the creation of homologous regions that are required for assembly of the single DNA fragments by In-Fusion cloning or overlap extension PCR, the DNA fragments were extended by PCR with the appropriate overhang primer. 4A *AtRECQ4A* sequences; 4B *AtRECQ4B* sequences; gDNA genomic DNA; cDNA copy DNA, ORF open reading frame; pro promoter; term terminator.

plasmid	DNA fragment	template	PCR	primer name	primer sequence	
pPZP221-RECQ4A	pro+	gDNA	amplification	RQ4A-Pro-fw RQ4A-PrR	5'-AAGATTCATGATTTGATTTTGG-3' 5'-GACCTCTAGGTTCTGAAC-3'	
	+pro+*	pro+	extending with homologies	RQ4A-Pro-fw-e-pPZP221BamHI RQ4A-PrR	5'-CGGTACCCGGGATCCAAGATTCATGATTTGATTTTGG-3' 5'-GACCTCTAGGTTCTGAAC-3'	
	+ORF+*	cDNA	amplification	RQ4A-ATG(+10)-fw RQL2-stop	5'-GTCACTCCCGAAGTTC-3' 5'-TCACCACGGTAATACTCTTC-3'	
	+term	gDNA	amplification	RQ4A-Stop2-fw RQ4A-Term-rev	5'-GAAGAGTATTACCGTGGTGA-3' 5'-CAAAGCAAACAAGCAGACAA-3'	
	+term+*	+term	extending with homologies	RQ4A-Stop2-fw RQ4A-Term-rev-e-pPZP221BamHI	5'-GAAGAGTATTACCGTGGTGA-3' 5'-CGACTCTAGAGGATCCAAAGCAAACAAGCAGACAA-3'	
pPZP221-RECQ4A-HD	pro+	gDNA	amplification	RQ4A-Pro-fw RQ4A-PrR	5'-AAGATTCATGATTTGATTTTGG-3' 5'-GACCTCTAGGTTCTGAAC-3'	
	+ORF(HD)+	cDNA	site directed mutagenesis, OLE-PCR, amplification	RQ4A-ATG-cDNA RQL2-stop RQ4A-Mut-rev RQ4A-Mut-fw	5'-ATGAGTAGGAGTCACCTCC-3' 5'-TCACCACGGTAATACTCTTC-3' 5'-GTCAAACATCATCCCTCCTC-3' 5'-GAGGAGGGATGAGTTTGAC-3'	
	+term	gDNA	amplification	RQ4A-Stop2-fw RQ4A-Term-rev	5'-GAAGAGTATTACCGTGGTGA-3' 5'-CAAAGCAAACAAGCAGACAA-3'	
	+ORF(HD)-term	+ORF(HD)+ & +term	OLE-PCR	RQ4A-ATG-cDNA RQ4A-Term-rev	5'-ATGAGTAGGAGTCACCTCC-3' 5'-CAAAGCAAACAAGCAGACAA-3'	
	pro-ORF(HD)-term <sup>‡</sup>	pro+ & +ORF(HD)-term	OLE-PCR	RQ4A-Pro-fw RQ4A-Term-rev	5'-AAGATTCATGATTTGATTTTGG-3' 5'-CAAAGCAAACAAGCAGACAA-3'	
pPZP221-RECQ4A-ΔN	pro	gDNA	amplification	RQ4A-Pro-fw RQ4A-Pro-Del-rev	5'-AAGATTCATGATTTGATTTTGG-3' 5'-CATGGGTCTAACATGTTGAAG-3'	
	+pro+*	pro	extending with homologies	RQ4A-Pro-fw-e-pPZP221BamHI ST-rev	5'-CGGTACCCGGGATCCAAGATTCATGATTTGATTTTGG-3' 5'-CAGGAGGAGCAGTGGTCTAACATGTTGAAG-3'	
	ORF(ΔN)	cDNA	amplification	RQ4A-Del-fw RQL2-stop	5'-CTGCCTCTGAATTATGTTC-3' 5'-TCACCACGGTAATACTCTTC-3'	
	+ORF(ΔN)+*	ORF(ΔN)	extending with homologies	ST-fw PJ-rev	5'-TAGACCCATGCTCCTCGAATTATGTTC-3' 5'-GGTCAAACTCACCACGGTAATACTCTTC-3'	
	term	gDNA	amplification	RQ4A-Term-fw RQ4A-Term-rev	5'-AGTTGTGACCACTGAAATTTAG-3' 5'-CAAAGCAAACAAGCAGACAA-3'	
+term+*	term	extending with homologies	PJ-fw RQ4A-Term-rev-e-pPZP221BamHI	5'-ACCGTGGTGAAGTTGACCACTGAAATTTAG-3' 5'-CGACTCTAGAGGATCCAAAGCAAACAAGCAGACAA-3'		
pPZP221-RECQ4A-HD-ΔN	pro	gDNA	amplification	RQ4A-Pro-fw RQ4A-Pro-Del-rev	5'-AAGATTCATGATTTGATTTTGG-3' 5'-CATGGGTCTAACATGTTGAAG-3'	
	+pro+*	pro	extending with homologies	RQ4A-Pro-fw-e-pPZP221BamHI ST-rev	5'-CGGTACCCGGGATCCAAGATTCATGATTTGATTTTGG-3' 5'-CAGGAGGAGCAGTGGTCTAACATGTTGAAG-3'	
	ORF(HD-ΔN)-term	pPZP221-K-RECQ4A-HD	amplification	RQ4A-Del-fw RQ4A-Term-rev	5'-CTGCCTCTGAATTATGTTC-3' 5'-CAAAGCAAACAAGCAGACAA-3'	
	+ORF(HD-ΔN)-term+*	ORF(HD-ΔN)-term	extending with homologies	ST-fw RQ4A-Term-rev-e-pPZP221BamHI	5'-TAGACCCATGCTCCTCGAATTATGTTC-3' 5'-CGACTCTAGAGGATCCAAAGCAAACAAGCAGACAA-3'	
	pPZP221-RECQ-4A(4B)	pro	gDNA	amplification	RQ4A-Pro-fw RQ4A-Pro-rev vor ATG	5'-AAGATTCATGATTTGATTTTGG-3' 5'-GGGTCTAACATGTTGAAGAG-3'
+pro+*		pro	extending with homologies	RQ4A-Pro-fw-e-pPZP221BamHI 4A-P/4B-Nt-rev	5'-CGGTACCCGGGATCCAAGATTCATGATTTGATTTTGG-3' 5'-TCACCACCATGGGTCTAACATGTTGAAGAG-3'	
5'-ORF(4B)		cDNA	amplification	RQL2A(-1) ATG RQ4B-Cr2	5'-ATGGTGGTACAAGAGGAG-3' 5'-CACTTTTTGTCAATTTGAACCT-3'	
+5'-ORF(4B)+*		5'-ORF(4B)	extending with homologies	4A-P/4B-Nt-fw OP-rev	5'-TGTAGACCCATGGTGGTGAAGAGGAG-3' 5'-ACGACTGTCCACTTTTTGTCAATTTGAACCT-3'	
3'-ORF(4A)		cDNA	amplification	RQ4A-C2 RQL2-stop	5'-GAGCAGTCGTGAATTTCCC-3' 5'-TCACCACGGTAATACTCTTC-3'	
+3'-ORF(4A)+*		3'-ORF(4A)	extending with homologies	OP-fw PJ-rev	5'-ACAAAAGTGGAGCAGCTGTAATTTCCC-3' 5'-GGTCAAACTCACCACGGTAATACTCTTC-3'	
term		gDNA	amplification	RQ4A-Term-fw RQ4A-Term-rev	5'-AGTTGTGACCACTGAAATTTAG-3' 5'-CAAAGCAAACAAGCAGACAA-3'	
+term+*		term	extending with homologies	PJ-fw RQ4A-Term-rev-e-pPZP221BamHI	5'-ACCGTGGTGAAGTTGACCACTGAAATTTAG-3' 5'-CGACTCTAGAGGATCCAAAGCAAACAAGCAGACAA-3'	
pPZP221-RECQ-4A(4B)		pro+	gDNA	amplification	RQ4A-Pro-fw RQ4A-Pro-AS	5'-AAGATTCATGATTTGATTTTGG-3' 5'-TTGATTCGAATTAATCATGGGT-3'
		+pro+*	pro+	extending with homologies	RQ4A-Pro-fw-e-pPZP221BamHI RQ4A-Pro-AS	5'-CGGTACCCGGGATCCAAGATTCATGATTTGATTTTGG-3' 5'-TTGATTCGAATTAATCATGGGT-3'
	+5'-ORF(4A)	cDNA	amplification	RQ4A-ATG-fw 4A-Ct-rev	5'-ATGATTAATTCGAATCAAATGAG-3' 5'-AGAAAATAGTATGGTAGCCTT-3'	
	+5'-ORF(4A)+*	+5'-ORF(4A)	extending with homologies	RQ4A-ATG-fw 4A-NI/4B-Ck-rev	5'-ATGATTAATTCGAATCAAATGAG-3' 5'-TCTGGCTCCGAAAAATAGTATGGTAGCCTT-3'	
	3'-ORF(4B)	cDNA	amplification	4B-Ct-fw RQ4B-Stop2-rev	5'-GGAGCCGAGATATAACAAT-3' 5'-TTATTTTTGTCTCACTACC-3'	
	+3'-ORF(4B)+*	3'-ORF(4B)	extending with homologies	4A-NI/4B-Ck-fw IJ-rev	5'-ACTATTTCTGGAGCCAGATATAACAAT-3' 5'-GGTCAAACTTTATTTTTGTCTCACTACC-3'	
	term	gDNA	amplification	RQ4A-Term-fw RQ4A-Term-rev	5'-AGTTGTGACCACTGAAATTTAG-3' 5'-CAAAGCAAACAAGCAGACAA-3'	
	+term+*	term	extending with homologies	IJ-fw RQ4A-Term-rev-e-pPZP221BamHI	5'-CAAAAATAAAGTTGTGACCACTGAAATTTAG-3' 5'-CGACTCTAGAGGATCCAAAGCAAACAAGCAGACAA-3'	

\* indicates existing homologies to the adjacent fragment of the construct or to the linearized plasmid backbone

† indicates starting material for a single In-Fusion reaction with pPZP221 linearized with BamHI to create the respective pPZP221 derivative

‡ indicates starting material for cloning into pCR-Blunt II-TOPO (Invitrogen), followed by a subcloning into pPZP221 using BamHI and PstI restriction sites surrounding the construct