

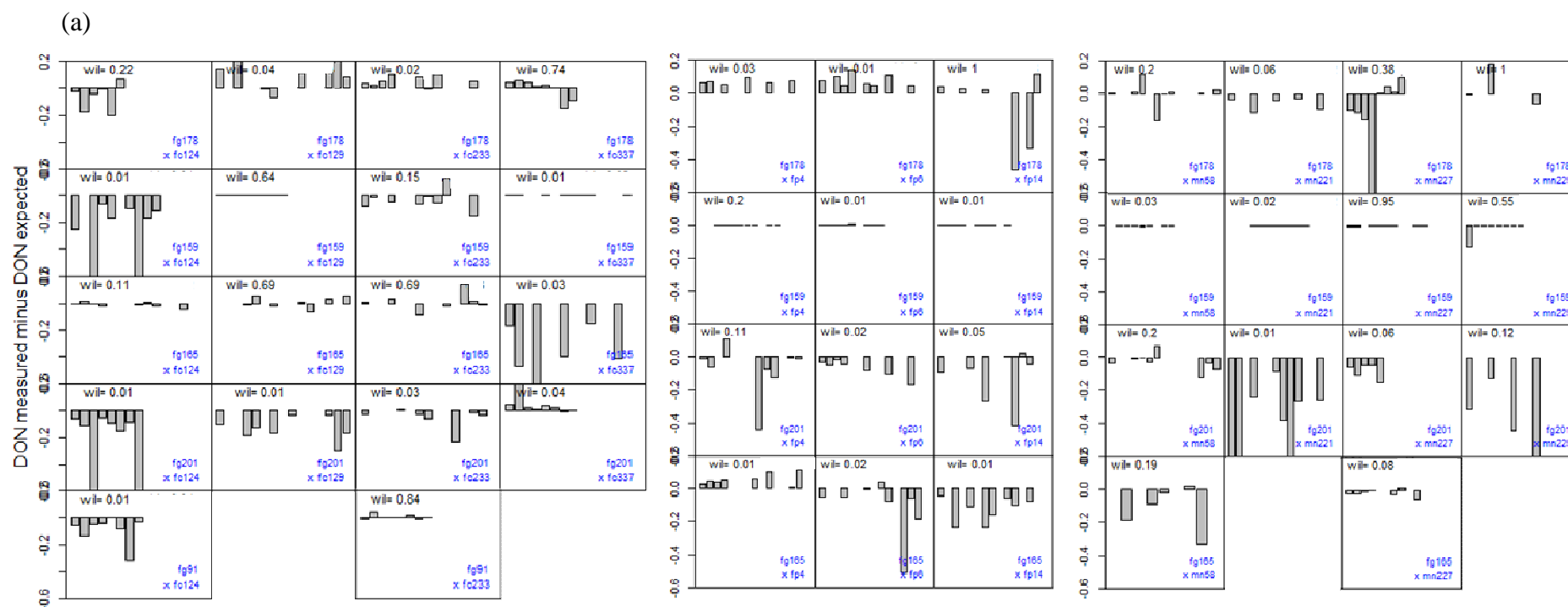
Supporting Information

Extraction and quantification of fungal DNA

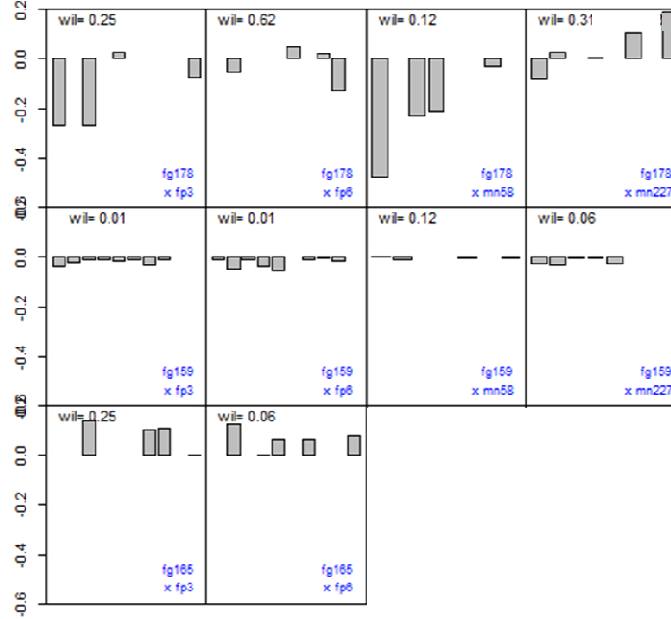
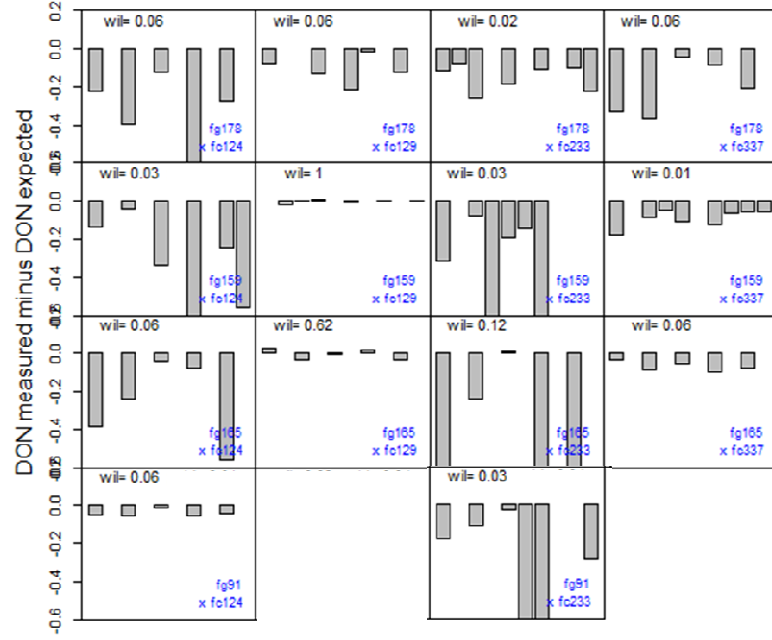
The total DNA was extracted from around 50 mg of grounded material using the DNeasy® Plant Mini Kit according to the manufacturer's instructions (Qiagen Ltd., Courtaboeuf, France). The DNA was quantified with a NanoDrop® ND-1000 spectrophotometer (NanoDrop Technology®, Cambridge, UK) and diluted to a final concentration of 20 ng.µL⁻¹. The amount of both fungal and wheat DNA was estimated by quantitative PCR (qPCR) for each inoculated spike. Species-specific primer pairs and species-specific Taqman® probes were used to enhance the specificity of the test (Table 2). The primers and probes were purchased from Eurogentec (Seraing, Belgium). All the probes used were Taqman® labelled with FAM/TAMRA quencher. Real-time PCR was carried out in a total of 25 µL consisting of 6.25 µL qPCR MasterMix with ROX and Uracil-N-Glycosylase (UNG) at the final concentration of 1X (Eurogentec, Angers, France), species specific primer and probe, and 5 µL of template DNA. The samples were standardized based on the plant DNA quantified with plant EF1α real time PCR primers and SYBR® Green technology. For wheat DNA quantification, we used the MESA GREEN qPCR MasterMix Plus for SYBR® Assay (Eurogentec) at the final concentration of 1X, with the same primer concentration and 5µL of template DNA diluted at 1:10. The PCR reactions were performed in duplicate (and in triplicate on samples used for the standard curve) on ABI PRISM 7900 Sequence Detection System (Applied Biosystems, Foster City, CA, USA) in Applied Biosystem 96 well plates. The amplification conditions included: an initial step of two min at 50°C, 95°C for 10 mn, 40 cycles of 15 s at 95°C and 60 s at 62°C (60°C for *F. poae*). DNA quantifications were done using standard curves of DNA from *F. graminearum*, *F. culmorum*, *F. poae* or wheat extracted from pure cultures and non-contaminated wheat kernels. Each standard curve was generated by ten-fold dilution series

ranging from 1 to $1 \cdot 10^{-3}$ ng. μ L⁻¹ for fungal DNA and 5 to $5 \cdot 10^{-3}$ ng. μ L⁻¹ for wheat DNA. Results were analysed on the AB SDS2.2.2 software (Applied Biosystems). PCR efficiency was in average 98%. The amount of fungal DNA was calculated from cycle threshold (Ct) values using the standard curve, and these values were normalized with the estimated amount of plant DNA based on the plant EF1 α assay.

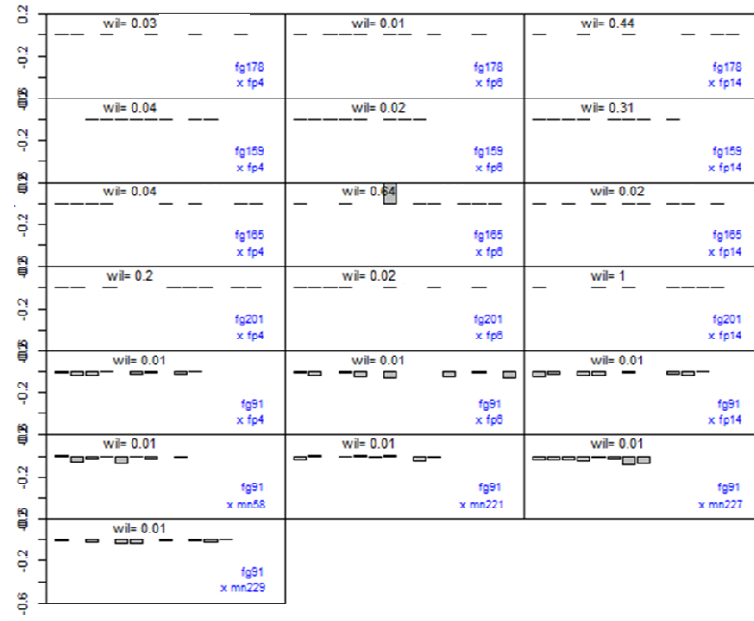
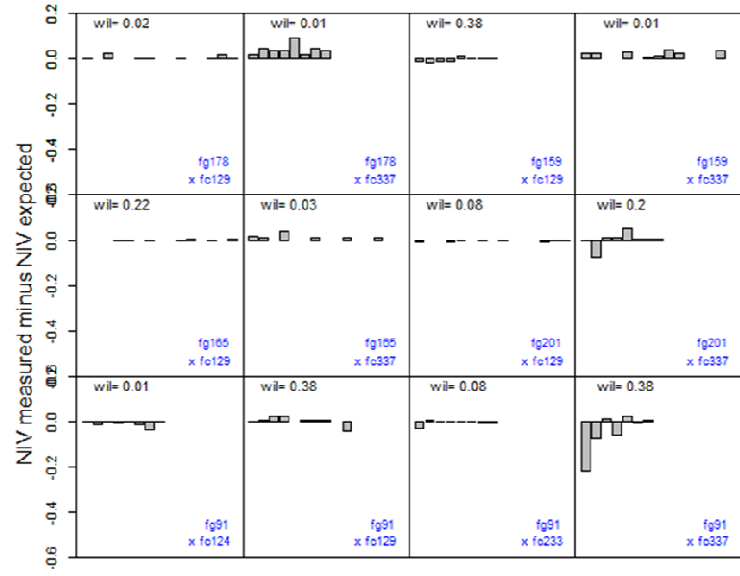
Figure S1 - Differences between measured and expected toxin content (DON and NIV) in individual wheat spikes inoculated with different *Fusarium* and *Microdochium* combinations during the 2010 and 2011 greenhouse experiments (a: DON 2010; b: DON 2011; c: NIV 2010; d: NIV 2011) and 2011 field experiment (g: DON and NIV). The pair of isolates and the p values obtained with the Wilcoxon's test are indicated on each graph; each bar corresponds to a spike from one of the replicates; the gaps correspond to missing data (spikes with a low disease development were not analyzed for toxin content, for cost limitation - see Material & Methods).



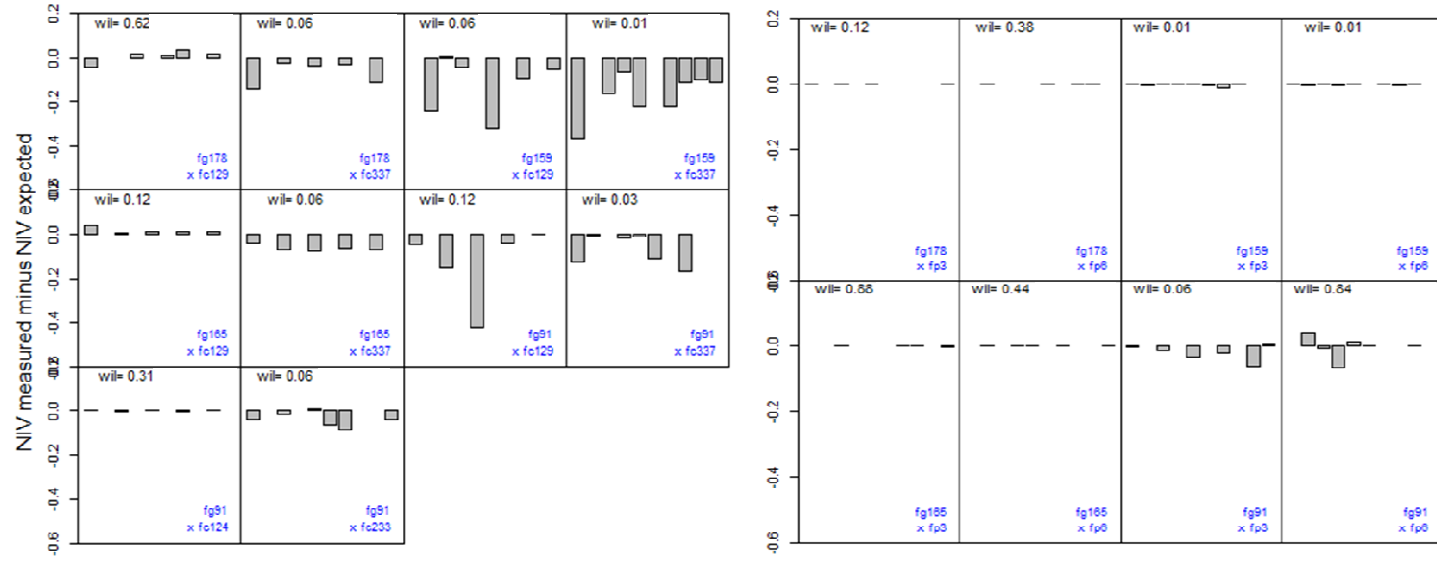
(b)



(c)



(d)



(e)

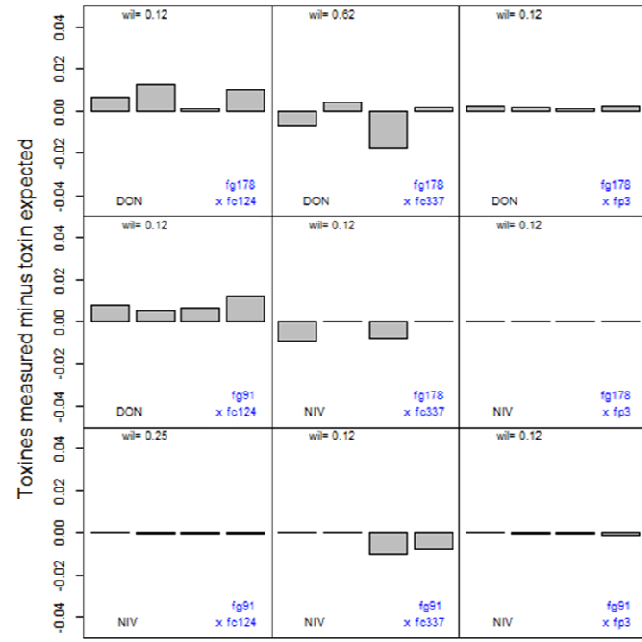


Table S1 - Means values for fungal DNA (ng/ng of plant DNA) quantified in wheat spikes inoculated with different *Fusarium* and *Microdochium* isolates in 2010 (a) and 2011 (b). The quantities of *F. graminearum* DNA obtain in mixed inoculations (first lines, plain text) were compared (***: $p < 0.01$; **: $p < 0.05$; *: $p < 0.1$) to the quantities of *F. graminearum* DNA obtain in single inoculation (SI; first column, in bold); the quantities of *F. culmorum*, *F. poae* or *Microdochium* sp. DNA obtained in mixed inoculations (second lines, in italics) were compared to the quantities of the same species DNA obtained in single inoculation (SI; last row, in bold italics).

(a)	Isolates in competition with <i>F. graminearum</i> (fg)											
	SI	fc124	fc129	fc233	fc337	fp4	fp6	fp14	mm58	mm221	mm229	mn227
fg91	0.027	0.012	0.027	0.011	0.100***	0.038	0.052	0.040	0.039	0.031	0.03	0.065**
		<i>0.088</i>	<i>0.003</i>	<i>0.013*</i>	<i>0.149</i>	<i>0.001</i>	<i>0.001***</i>	<i>0.001*</i>	<i>0.042***</i>	<i>0.049</i>	<i>0.021</i>	<i>0.046**</i>
g159	0.011	0.0001	0.0004	0.0001	0.0003	0.0001	0.002	0.004	0.002	0.002	0.002***	0.004
		<i>0.244**</i>	<i>0.019**</i>	<i>0.080</i>	<i>0.063***</i>	<i>0.004</i>	<i>0.003</i>	<i>0.003</i>	<i>0.065</i>	<i>0.028</i>	<i>0.020</i>	<i>0.051**</i>
fg165	0.034	0.023	0.099	0.034	0.503***	0.108	0.219**	0.204***	0.245**	na	na	0.066
		<i>0.044</i>	<i>0.002**</i>	<i>0.055</i>	<i>0.060***</i>	<i>0.001</i>	<i>0.001***</i>	<i>0.001**</i>	<i>0.058</i>	<i>na</i>	<i>na</i>	<i>0.098</i>
fg178	0.248	0.144	0.113	0.044*	0.144	0.081	0.060	0.301	0.127	0.232	0.198	0.247
		<i>0.084</i>	<i>0.003**</i>	<i>0.010*</i>	<i>0.088**</i>	<i>0.001</i>	<i>0.0002***</i>	<i>0.001</i>	<i>0.046***</i>	<i>0.134***</i>	<i>0.094</i>	<i>0.088</i>
fg201	0.130	0.143	0.234	0.119	0.057	0.148	0.182	0.187	0.093	0.365*	0.249	0.130
		<i>0.087</i>	<i>0.010*</i>	<i>0.011*</i>	<i>0.073***</i>	<i>0.002</i>	<i>0.003***</i>	<i>0.001**</i>	<i>0.050**</i>	<i>0.070</i>	<i>0.141**</i>	<i>0.099</i>
SI		<i>0.108</i>	<i>0.037</i>	<i>0.064</i>	<i>0.205</i>	<i>0.003</i>	<i>0.004</i>	<i>0.002</i>	<i>0.090</i>	<i>0.052</i>	<i>0.043</i>	<i>0.110</i>

(b)	Isolates in competition with <i>F. graminearum</i> (fg)								
	SI	fc124	fc129	fc233	fc337	fp3	fp6	mm58	mn227
fg91	0.212	0.067	0.387	0.380	0.260	0.361	0.323	na	na
		<i>0.063</i>	<i>0.225</i>	<i>0.220</i>	<i>0.113</i>	<i>0.001</i>	<i>0.001***</i>	<i>na</i>	<i>na</i>
fg159	0.060	0.009**	0.013*	0.012**	0.005***	0.030	0.039	0.020	0.029**
		<i>0.464</i>	<i>0.397</i>	<i>0.411</i>	<i>0.329***</i>	<i>0.006</i>	<i>0.006</i>	<i>0.012</i>	<i>0.026</i>
fg165	0.609	0.384	0.284	0.783	0.351	0.322	0.445	na	na
		<i>0.259</i>	<i>0.113</i>	<i>0.236</i>	<i>0.127</i>	<i>0.001</i>	<i>0.001**</i>	<i>na</i>	<i>na</i>
fg178	0.227	0.303	0.204	0.156	0.247	0.398	0.338	0.955	0.404
		<i>0.261</i>	<i>0.120</i>	<i>0.065</i>	<i>0.157</i>	<i>0.002</i>	<i>0.001*</i>	<i>0.100***</i>	<i>0.109*</i>
SI		0.212	0.121	0.158	0.078	0.006	0.004	0.021	0.041