



EUROPEAN COMMISSION  
DIRECTORATE GENERAL JRC  
JOINT RESEARCH CENTRE  
INSTITUTE FOR HEALTH AND CONSUMER PROTECTION  
COMMUNITY REFERENCE LABORATORY FOR GM FOOD AND FEED



# **Addendum to the Report on the Verification of an Event-specific Detection Method for Identification of Rice GM-event LLRICE601 Using a Real-time PCR Assay**

## **Method development:**

Bayer CropScience

## **Verification and reporting:**

Joint Research Centre – European Commission  
Biotechnology & GMOs Unit

## EXECUTIVE SUMMARY

Following the Commission Decision of 23/08/2006 (2006/578/EC) "*on emergency measures regarding the non-authorized genetically modified organism LLRICE601 in rice products*", the JRC as Community Reference Laboratory (CRL) for GM Food and Feed, (Regulation EC 1829/2003), has carried out a verification of an event-specific detection method developed by Bayer CropScience and validated by USDA to detect LLRICE601 event. The verification was conducted according to internationally accepted guidelines.

The conclusions of the verification report (<http://gmo-crl.jrc.it/LLRice601update.htm>) stated that additional specificity tests were ongoing at the time of its publication.

The present addendum, while providing specificity data from new experiments, confirms that the LLRice601 event-specific method does not react with the GM events tested under the conditions reported.

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## 1. Specificity

Specificity tests were carried out on ABI7500 on a total of 200 ng gDNA per well, under the conditions described in the Methods. The following Table 4bis is an update of Table 4 presented in the "Report on the Verification of an Event-specific Detection Method for Identification of Rice GM-event LLRICE601 Using a Real-time PCR Assay", published at <http://gmo-crl.jrc.it/LLRice601update.htm>

**Table 4 bis: Results of specificity test**

GM event	% GM	Replicates	Positive GM amplifications / Positive reference amplifications	Ct value for the GM target (PGS0494)	Ct value for the Reference system	$\Delta$ Ct
LL62	100	3	0/3	-	18.49	-
LL601	1	3	3/3	25.07	19.24	5.83
LL25 cotton	100	3	0/3	-	20.81	-
Bt176 maize*	2	3	0/3	-	21.83	-
Starlink maize **	5	3	0/3	-	22.11	-
Ms1 oilseed rape	100	3	0/3	-	20.69	-
Ms8 oilseed rape	100	3	0/3	-	20.17	-
Rf1 oilseed rape	100	3	0/3	-	21.29	-
Rf2 oilseed rape	100	3	0/3	-	21.12	-
Rf3 oilseed rape	100	3	0/3	-	20.29	-
Ms8xRf3	100	3	0/3	-	20.99	-
Ms1xRf1	100	3	0/3	-	20.31	-

GM event	% GM	Replicates	Positive GM amplifications / Positive reference amplifications	Ct value for the GM target (PGS0494)	Ct value for the Reference system	$\Delta Ct$
T45 oilseed rape	100	3	0/3	-	20.21	-
A2704-12 soy	100	3	0/3	-	21.17	-
Wild type non-GM rice seeds	-	3	0/3	-	19.16	-
NTC	-	3	0/0	-	-	-

\* Certified Reference Material (JRC, IRMM, Belgium)

\*\* provided by GIPSA as sample for proficiency test scheme

Table 4bis reports the number of positive amplifications observed with the event-specific method and the average Ct values.

GM-events other than the rice positive control LLRICE601, namely LLRice62, LLCotton25, Bt176 maize, Starlink maize, A2704-12 soy, oilseed rape events T45, Ms1, Ms8, Rf1, Rf2, Rf3, and their stacks Ms8xRf3, Ms1xRf1 did not react with the event-specific method.

## 2. Conclusions

Following the Commission Decision of 23/08/2006 (2006/578/EC) "*on emergency measures regarding the non-authorized genetically modified organism LLRICE601 in rice products*", the JRC as Community Reference Laboratory (CRL) for the GM Food and Feed, (see Regulation EC 1829/2003), has received the samples and the method developed by Bayer CropScience and validated by USDA-GIPSA to identify LLRICE601 event.

The present addendum to the "Report on the Verification of an Event-specific Detection Method for Identification of Rice GM-event LLRICE601 Using a Real-time PCR" confirms that the LLRice601 event-specific method does not react with the GM events tested under the conditions reported.

### **3. Reference**

Report on the Verification of an Event-specific Detection Method for Identification of Rice GM-event LLRICE601 Using a Real-time PCR.

<http://gmo-crl.jrc.it/LLRice601update.htm>