

Certified Reference Material

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Certificate of Analysis

AOCS 0919-A, non-modified maize

Certified Presence	Certified Value	Measurement Uncertainty	Test Method
MIR162, MZHG0JG and MZIR098 maize DNA absent	0 g/kg	0.4 g/kg	Real-time PCR

Description: This is the first batch of non-modified maize CRM prepared by AOCS for Syngenta Crop Protection, LLC. A total of 7200 seeds (24 aliquots of 300 seeds) were evaluated and all of the aliquots tested negative for MIR162, MZHG0JG and MZIR098 maize. The measurement uncertainty is the expanded uncertainty using the value of the upper bound of impurity. The standard uncertainty can be obtained by dividing the expanded uncertainty by $2\sqrt{3}$ (rectangular distribution). This material is for limited purposes only: see "Intended Use" and "Terms and Conditions."

This certificate is valid through: July 2025

This validity date may be extended when further evidence of stability becomes available. Customers will be notified by AOCS if a stability issue arises at AOCS Headquarters.

Introduced: 18 December 2019

Revised: 10 April 2020, 1 July 2021, 8 July 2022, 10 July 2023, 12 July 2024, 7 January

2025

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AOCS Mission: AOCS advances the science and technology of oils, fats, proteins, surfactants and related materials, enriching the lives of people everywhere.

Characterization

Product Description CRM AOCS 0919-A has been prepared by AOCS from maize seed. CRM AOCS 0919-A is available in 27-mL glass headspace vials containing approximately 10 g of material. Users are informed that this reference material has been produced from seed powder of non-modified maize delivered by Syngenta Crop Protection, LLC.

Homogeneity and Trait Verification The homogeneity of AOCS 0919-A is related to the purity of the seeds. 7200 out of 7200 seeds tested negative for the MIR162, MZHG0JG, and MZIR098 maize events by real-time PCR by Syngenta Crop Protection, LLC. Based on the sample impurity of 0% as determined using SeedCalc8, the batch was expected to be homogenous.

To further confirm homogeneity, ten vials of AOCS 0919-A were evaluated to determine the within-unit and between-unit property value. The property value is defined as the ratio between copies of the event specific target (MIR162, MZHG0JG and MZIR098) and copies of a taxon-specific reference target (*adh1*). Variability of this ratio within CRM vials and between CRM vials is reported as the within-unit relative standard deviation (RSD_w), and the between-unit relative standard deviation (RSD_b), respectively. The results confirm that AOCS 0919-A is homogenous, because all results are below limit of quantification (LOQ). The absence of Events MIR162, MZHG0JG and MZIR098 in this CRM was verified in all ten vials.

The within-unit relative standard deviation (RSD_w), and the between-unit relative standard deviation (RSD_b) for vials of CRM AOCS 0919-A.

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CRM	RSD _w [%]	RSD₀ [%]	
AOCS 0919-A	Below LOQ	Below LOQ	

Stability AOCS Certified Reference Materials (CRMs) are assessed for transport (short-term) and long-term stability and tested for stability on an annual or biennial basis, depending on the type of homogeneity testing that is required, and certificates may be extended based on the outcome of this testing. Customers may request extended certificates, but they are informed that results are based on samples that are obtained from AOCS' inventory. AOCS cannot guarantee the integrity of samples outside of AOCS control.

Analytical Method Used for Certification

The certified value is based on the purity of the non-modified maize material used in the production of this CRM. Eurofins-GeneScan, New Orleans, LA (an ISO 17025 accredited



laboratory) performed quantitative real-time PCR to determine the copy number ratio between the MIR162, MZHG0JG and MZIR098 maize-specific target and the taxon-specific reference target for the assessment of homogeneity. The absence of Events MIR162, MZHG0JG and MZIR098 was verified by converting quantitative results to qualitative data. Purity and stability results were used to determine the expanded measurement uncertainty of the certified value.

Warnings and Precautions

This product is for laboratory use only and is not for consumption. The user of this CRM should follow safety requirements and rules issued by voluntary organizations and government agencies expert in the field of laboratory safety.

Intended Use

This CRM, AOCS 0919-A, is intended for use as quality control material or calibrant in methods for the detection, identification, and/or quantification of biotechnology-derived events.

Instructions for Use

Upon receipt the product should be stored in a sealed container in the dark and at ambient or cooler conditions. The product may have settled during shipment, therefore, thoroughly mix the CRM before use to ensure homogeneity.

If the user of this CRM intends to use it multiple times, proper protocols must be followed to ensure that the sample retains its integrity. Use a clean laboratory spatula to remove the intended sample amount. After the sample has been removed, flush the headsapce of the vial with nitrogen gas, then replace the rubber stopper. Place a new 20 mm tear-off aluminum unlined seal on top of the rubber stopper and crimp it to the vial by using a crimping tool. Store the CRM in the dark and at ambient or cooler conditions and repeat this process for all subsequent uses.

Sample size:

The recommended **sample intake** suitable for DNA extraction and real time PCR is 1 g.

Protocols have been followed to ensure that this CRM is absent of the traits it has been tested against. Please follow all instructions on this certificate to prevent contamination of the CRM.

Note: The CRM AOCS 0919-A certification report is available online and a paper copy could be supplied upon request.



Terms and Conditions (i– ix)

- i. The CRM AOCS 0919-A shall be used solely 1) in assays for (a) detecting the absence of MIR162, MZHG0JG and MZIR098, or (b) quantification of MIR162, MZHG0JG and MZIR098; or 2) for determining whether an assay cross-reacts with CRM AOCS 0919-A. CRM AOCS 0919-A shall be used for no other purpose. Specifically, the CRM may not be used to develop a detection method for non-modified nor trait(s) present therein. No other rights are conveyed by the sale of the CRM 0919-A to any purchaser, including any rights to any pending or granted Syngenta Crop Protection, LLC patents or other Syngenta Crop Protection, LLC intellectual property that may protect the CRM or non-modified or trait(s) present therein or a detection method for non-modified powder.
- ii. Neither the CRM AOCS 0919-A nor any extract or portion thereof shall be resold or redistributed by any purchaser, unless the resale or redistribution is required by national law in force in the purchaser's country.
- iii. Neither the CRM AOCS 0919-A, nor any extract or portion thereof, shall be used for human or animal consumption or human or animal trials.
- iv. Neither the CRM AOCS 0919-A nor non-modified DNA, nor any part of either of these, shall be used for transformation or breeding.
- v. No characterization or derivation of MIR162, MZHG0JG and MZIR098, of the trait or traits present in CRM AOCS 0919-A, or of the CRM AOCS 0919-A shall be performed, except as allowed for in section (i).
- vi. All assay activities undertaken using the CRM AOCS 0919-A shall be conducted in strict compliance with all applicable laws governing such activities, and shall comply with conditions of all permits and authorizations which may be required for such activities; and such activities shall be strictly limited to assays in contained facilities, for example, laboratories.
- vii. Prior to disposal of any used or excess CRM AOCS 0919-A or residues thereof, such material or residue must be treated in a manner that degrades the CRM material, such as by autoclaving.
- viii. CRM AOCS 0919-A shall not be exported nor re-exported in violation of any applicable laws or without securing any necessary export or import clearances or permits.

ix. THE CRM 0919-A IS PROVIDED FOR THE PURPOSE OF IDENTIFYING, DETECTING AND QUANTIFYING MIR162, MZHG0JG AND MZIR098 AND FOR NO OTHER PURPOSE. AOCS HAS TESTED CRM 0919-A WITHIN THE PAST 24 MONTHS AND STATES THAT IT HAS DETERMINED IT TO BE OF SUFFICIENT QUALITY AND FIT FOR THE PURPOSES STATED HEREIN. NO Syngenta Crop Protection, LLC WARRANTY IS PROVIDED WHETHER EXPRESS OR IMPLIED, IN RELATION TO THE CRM AOCS 0919-A AND Syngenta Crop Protection, LLC MATERIALS. Syngenta Crop Protection, LLC MAKES NO REPRESENTATION OR WARRANTY THAT THE USE OF THE CRM AOCS 0919-A, WHETHER BEFORE, OR AFTER THE EFFECTIVE DATE OF THE APPLICABLE CERTIFICATE, WILL NOT INFRINGE ANY PATENT OR OTHER PROPERTY RIGHTS.