DIN EN 15634-3:2023-05 (E)

Foodstuffs - Detection of food allergens by molecular biological methods - Part 3: Hazelnut (Corylus avellana) - Qualitative detection of a specific DNA sequence in chocolate by real-time PCR

Contents

Page

European foreword ... 3
Introduction ... 4
1 Scope ... 5
2 Normative references ... 5
3 Terms and definitions ... 5
4 Principle ... 5
5 Reagents ... 5
5.1 General ... 5
5.2 Extraction reagents ... 5
5.3 DNA purification by means of solid phase extraction ... 6
5.4 Real-time PCR reagents ... 6
6 Apparatus and equipment ... 7
6.1 General ... 7
6.2 DNA extraction ... 7
6.3 PCR ... 7
7 Procedure ... 8
7.1 General ... 8
7.2 Sample preparation ... 8
7.3 Preparation of extracts ... 8
7.3.1 DNA extraction with CTAB and DNA purification ... 8
7.3.2 Quantification and normalization of DNA concentration ... 9
7.4 Real-time PCR set-up ... 9
7.4.1 Reaction mix for real-time PCR ... 9
7.4.2 Positive control for DNA targets ... 10
7.4.3 Negative control for DNA targets ... 11
7.4.4 PCR inhibition control ... 11
7.4.5 Amplification reagent control ... 11
7.4.6 Extraction blank control ... 11
7.4.7 Positive extraction control ... 11
7.4.8 Temperature/time programme (real-time PCR) ... 11
7.4.9 Accept/Reject criteria ... 12
7.4.10 Identification ... 12
8 Validation ... 12
8.1 General ... 12
8.2 Specificity ... 12
8.3 Ring trial validation study ... 13
8.3.1 Setup of the ring trial study ... 13
8.3.2 Ring trial validation results ... 13
9 Test report ... 15
Bibliography ... 16