EVALUATION OF METHODS FOR RECOVERY OF GI NOROVIRUS ON READY-TO-EAT FOODS

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Norovirus (NoV) is a highly infectious foodborne viral pathogen of the Caliciviridae family that is a common cause of gastroenteritis in adults and children. The Norovirus genus is subdivided into 7 genogroups, with GI, GII, and GIV causing human disease. Ready-to-eat (RTE) foods such as deli sandwiches, prepared meals, buffet foods and restaurant lunches have been considered as the causative food vehicles in described foodborne NoV outbreaks. Since then, numerous methods have been developed for recovering noroviruses from various foodstuffs. The aim of this study was to evaluate polyethylene glycol (PEG) precipitation and the TRIzol® method on the recovery of GI NoV from ham and turkey meat samples and to detect GI NoV from these foods by RT-qPCR using murine norovirus (MNV-1) as process control. The food samples were divided in 25g aliquots artificially contaminated with 250 μL of faecal samples containing GI NoV. MNV-1 was propagated in RAW 264.7 cells and 10⁶ genome copies were used on the food samples. The viral particles were washed from the foods surfaces using elution buffer (Tris-Glycine pH 9.5) including 3% meat extract followed by PEG precipitation. The other recovery method was based on TRIzol® reagent as previously described and purification of the RNA was performed by the use of the manual QIAMP Mini kit. After reverse transcription, cDNA was submitted to RT-qPCR, and the data analysis was performed using the ABI 7500 RT-qPCR instrument. Quantitative data showed that PEG precipitation resulted in a recovery of GI NoV with efficiencies of 1.83% to 19.40% from ham samples, and 5.32% to 19.56% from turkey meat samples. MNV-1 was recovered with efficiencies of 0.74% to 6.00% from ham samples, and 0.40% to 0.97% from turkey meat samples. After the method using TRIzol® reagent to recovery GI NoV from the foods, quantitative data showed efficiencies of 14.88% to 75.61% from ham samples, and 2.59% to 26.08% from turkey meat samples. MNV-1 was recovered with efficiencies of 9.05% to 32.99% from ham samples, and 5.88% to 26.27% from turkey meat samples. The assays could react different to the presence of residual components on foods, although better recovery by method using TRIzol® reagent has been observed to GI NoV from ham and turkey meat samples. The evaluated detection method was capable of detecting NoV on the foods, but recoveries can be influenced by the inoculum level and by the food type. This research is within the scope of the activities of FIOCRUZ as a collaborating center of PAHO/WHO of Public and Environmental Health.

Keywords: Norovirus; viral recovery methods; ready-to-eat foods.