Optimizing hexane, KOH, and H_2O_2 methods for lipid removal and organic matter digestion in microplastic analysis of human milk

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Abstract

Objective: Microplastics (MPs) contamination is a growing emerging concern with potential health risks, particularly for vulnerable populations such as infants. This study aims to develop and optimize a method for isolating MPs from human breast milk, a complex biological matrix with high lipid and protein content that complicates detection. The goal is to evaluate the effectiveness of a combined chemical extraction approach for isolating MPs and identifying their physical characteristics and polymer types. Methods: Breast milk samples were ethically obtained from the Halimatussaadia Mother's Milk Centre (HMMC). Lipids were removed using hexane followed by potassium hydroxide (KOH) and hydrogen peroxide (H_2O_2) treatments to digest organic matter. The chemical extraction process was optimized with varying concentrations: hexane (0.5:1, 1:1), 30% H_2O_2

(1:2, 1:4), and 10% KOH (1:1, 1:3). The treated samples were analysed using Attenuated Total Reflectance Fourier-Transform Infrared (ATR-FTIR) spectroscopy to identify MPs and determine polymer types. Results: This study presents an optimized method combining hexane, KOH, and H_2O_2 to isolate microplastics (MPs) from human breast milk, identifying 12 MPs polymer types, including polyethylene (PE), polypropylene (PP), and polyethylene terephthalate (PET). This approach improves detection accuracy and provides a foundation for future research into potential health risks associated with MPs exposure in infants. However, due to technological limitations, the most prevalent MPs polymer in the samples could not be confirmed. Despite this, the study demonstrated the reliability of the combined hexane-KOH –H_2O_2 approach for isolating MPs from human milk. Conclusions: This study presents an effective and optimized method for isolating MPs from complex biological fluids such as human breast milk. The findings provide a foundation for future research investigating potential health risks associated with MPs exposure in infants, highlighting the need for larger sample sizes and more advanced analytical techniques.

Author keywords

ATR-FTIR spectroscopy; Human breast milk; Lipid extraction; Microplastics; Polymer identification

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