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*Abstract*

# Effect of Pasteurisation Techniques on Phages in Human Milk †

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**Abstract:** Bacteriophages (phages) are viruses that are the natural predators of bacteria and highly abundant in human milk and the infant gut microbiome. However, the effect of pasteurisation on human milk phages is unknown. This study, therefore, assessed the effect of holder pasteurisation (HP) and UV-C irradiation (UV) on exogenous bacteriophages inoculated into human milk. Ten donor human milk samples inoculated with a thermotolerant *Escherichia coli* phage (T4) and a thermosensitive *Staphylococcus aureus* phage (BYJ20) were subjected to HP and UV treatments. We found that UV effectively inactivated both phages (8/10 samples; 80%), however, HP was ineffective against the thermotolerant T4 phages (0/10; 0% inactivated). This is the first study to assess the impact of UV and HP methods on the viability of human milk phages. This pilot data suggests that HP methods used by milk banks likely destroy thermosensitive, but not thermotolerant, phages, with implications for early-life virome and bacterial microbiome assembly in donor milk fed infants.

**Keywords:** bacteriophage; donor milk; holder pasteurisation; human milk; UV-C

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**Institutional Review Board Statement:** This study was conducted in accordance with the guidelines of the Declaration of Helsinki and approved by the Human Research Ethics Committee at The University of Western Australia (RA/4/1/2369). All participants provided written informed consent.

**Informed Consent Statement:** Informed consent was obtained from all subjects involved in the study.

**Data Availability Statement:** Not applicable.

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