

## TECHNICAL REPORT

### PASTEURIZATION OF HUMAN MILK TO PREVENT TRANSMISSION OF CHAGAS DISEASE

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#### SUMMARY

Although admittedly transmission of *Trypanosoma cruzi* infection through breastfeeding is a rare event, it involves serious risks. To test the effectiveness of pasteurization in preventing this mode of infection, three sets of samples of human milk were tested: *a* – contaminated with *T. cruzi* and pasteurized; *b* – contaminated with *T. cruzi* and non-pasteurized; *c* – non-contaminated and pasteurized. Samples from all sets were orally and intraperitoneally administered to 90 BALB/c mice. The animals inoculated with contaminated, non-pasteurized samples, got the infection. Controls and the animals inoculated with contaminated and pasteurized milk were not infected. The hypothesis was accepted that pasteurization inactivates *T. cruzi* trypomastigotes.

**KEYWORDS:** *Trypanosoma cruzi*; Pasteurization of human milk; Milk banks.

Control of populations of *Trypanosoma cruzi* vectors, to a large extent achieved in Brazil, was followed by a marked reduction in the frequency of new cases of Chagas disease. Alternative modes of transmission, breastfeeding among them, were thus put to the foreground. MAZZA *et al.*<sup>6</sup> reported in 1936 the first finding of *T. cruzi* in human milk, namely from a patient in the acute phase of Chagas disease. DISKO & KRAMPITZ<sup>5</sup> (1971) observed numerous *T. cruzi* trypomastigotes in the milk of experimentally infected mice and pointed out that this was not due to contamination of milk with blood from ruptured vessels. However, they were unable to demonstrate the infection of suckling mice via contaminated milk. Likewise, CAMPOS *et al.*<sup>4</sup> found that experimentally infected female mice failed to infect their litters through suckling. MILES<sup>9</sup> (1972) detected *T. cruzi* trypomastigotes and anti-*T. cruzi* antibodies in the milk of mice in the acute phase of the infection. MEDINA-LOPES<sup>7,8</sup> reported in 1983 and 1988, respectively, two cases of infants, born of infected mothers, who developed acute Chagas disease while being breastfed, which was attributed to the ingestion of blood from papillary fissures. BITTENCOURT *et al.*<sup>3</sup> (1988) and AMATO NETO *et al.*<sup>2</sup> (1992) could not demonstrate the presence of *T. cruzi* in the milk of infected mothers. AMATO NETO *et al.*<sup>1</sup> (1999) evaluated experimentally the penetration, through the digestive tract, of *T. cruzi* trypomastigotes suspended in liquid foods. Although admittedly it is a rare event, human to human transmission of *T. cruzi* through breastfeeding must be prevented whenever possible. Accordingly, infected women reaching fertile age should be regarded as a potential health hazard to their offspring. One should not underestimate the risk of transmission from infected nursing mothers who, possibly unaware of their condition, are also milk bank donors. This risk obviously increases when the mothers

involved are in the acute phase of the infection.

Pasteurization inactivates the causative agents of many diseases (including HIV)<sup>10</sup>, in which case it will not be necessary to deprive infants of their mother's milk. To evaluate the effectiveness of pasteurization in inactivating *T. cruzi* trypomastigotes suspended in human milk, an experiment was set up. A volume of 50 mL of human milk from a healthy donor, kindly supplied by the Milk Bank of Hospital Estadual Infantil "Darcy Vargas", kept at the temperature of -18 °C until used, was divided into 5 samples. To samples 1, 2, 3 and 4 respectively, we added 2,500, 5,000, 10,000 and 2,500 *T. cruzi* trypomastigotes (Y strain) from laboratory-infected BALB/c mice. No contaminated material was added to sample 5. Samples 1, 2, 3 and 5 were pasteurized by immersion in a water bath at 62.5 °C during 30 minutes, and then in melting ice for 10 minutes. We used ten batches of mice (A, B, C, D, E, F, G, H, I, and J), supplied by the Laboratory Animal Facility Center, Faculty of Medicine of the University of São Paulo. Batches A to H were composed of ten mice, I and J of 5 mice each. The milk samples were administered orally and intraperitoneally to the animals, as described in Table 1. Starting from the 5<sup>th</sup> day of administration, fresh preparations of peripheral blood from each animal were examined (400× magnification) at 3-day intervals during 60 days. As shown in Table 1, positive results were observed only in the animals to which non-pasteurized, contaminated milk was given, thus confirming the hypothesis of the effectiveness of human milk pasteurization in inactivating suspended *T. cruzi* trypomastigotes. As the nutritional value of the milk is mostly preserved after pasteurization, and the process is quite simple, its use is recommended whenever the mother is presumed infected.

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**Table 1**

Results of search for *Trypanosoma cruzi* trypomastigotes in the peripheral blood of BALB/c mice inoculated according with the experimental conditions described

Batch	Number of animals	Milk sample	Parasites per mL	Volume (mL)	Administration	Result
A	10	1	250	0.1	Oral	Negative
B	10	1	250	0.5	Intraperitoneal	Negative
C	10	2	500	0.1	Oral	Negative
D	10	2	500	0.5	Intraperitoneal	Negative
E	10	3	1,000	0.1	Oral	Negative
F	10	3	1,000	0.5	Intraperitoneal	Negative
G	10	4 *	250	0.1	Oral	Positive
H	10	4 *	250	0.5	Intraperitoneal	Positive
I	5	5	0	0.1	Oral	Negative
J	5	5	0	0.5	Intraperitoneal	Negative

\* Non-pasteurized

## RESUMO

### Pasteurização de leite humano para evitar a transmissão da doença de Chagas

A amamentação é modo alternativo de transmitir-se a doença de Chagas. Embora admitida como evento raro, a infecção por esta via é preocupante. Para evitá-la é sugerida a pasteurização. Separaram-se para o ensaio três conjuntos de amostras de leite humano: *a* – contaminadas por *Trypanosoma cruzi* e pasteurizadas; *b* – contaminadas por *T. cruzi* e não pasteurizadas; *c* – não contaminadas e pasteurizadas. Frações dos três conjuntos foram inoculadas por vias oral e intraperitoneal em 90 camundongos BALB/c. Os animais inoculados com leite contaminado e não pasteurizado infectaram-se. Os controles e os inoculados com leite contaminado e pasteurizado não se infectaram. Aceitou-se a hipótese de a pasteurização ter inativado as formas tripomastigotas de *T. cruzi* em suspensão no leite.

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