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# What are the effects of raw vs. pasteurized milk consumption on growth rate and fertility in a colony of mice?

Kari Sholing, Lindsay Ganong, and Amy Olson

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## ABSTRACT

*Interest in consuming unprocessed foods has led to assumptions that raw milk has health benefits over pasteurized milk. This study was designed to evaluate the belief that raw milk is nutritionally superior. Eighteen mice were randomly assigned to one of two groups: raw milk or pasteurized milk. Mice were grouped into breeding trios and given a fresh supply of milk every four hours between 7 a.m. and 11 p.m. for 55 days. Milk consumption was tracked at each feeding by measuring the amount of milk that was provided and the amount of milk that was left from the previous feeding. The College of St. Benedict Institutional Animal Care and Use Committee (IACUC) approved this study.*

*On average, the mice in the raw milk group consumed significantly less milk but gained the same amount of weight as the pasteurized milk group. The rate of growth per gram of milk consumed was greater in the raw milk group, but the difference did not reach statistical significance. Lastly, there was no statistical difference between the groups with regard to birth rates or pup mortality rates.*

*Due to the limited number of mice, the positive trend in data did not reach statistical significance. A similar study done on a larger scale could produce significant results. In conclusion, this study does not demonstrate that either milk type is statistically superior in nutritional value.*

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## INTRODUCTION

There is a movement among consumers to seek more local and sustainable food choices and health benefits of unprocessed foods, including raw milk. Current laws often restrict consumers' access to raw milk based on historical risk of contamination. Individuals are driven by their belief that raw milk is higher in nutritional value and more natural in its unaltered state; it is fresh daily, devoid of heat treatment, handled only by the farmer and the consumer, and travels a short distance. Conversely, commercial pasteurized milk has been heat treated, fortified with vitamins A & D, and usually homogenized. Furthermore, milk that is not organic may not be free of hormones, antibiotics, or other questionable and potentially dangerous contaminants. However, there is no current research evidence that supports the belief in the superior nutritional value of raw milk.

No one argues that pasteurization changes milk; what is not clearly understood, however, is how changes in the heat processed milk affect the health of the consumer. Milk is rarely the only component in a person's diet making it difficult to design studies and evaluate outcomes that are exclusive to the consequences of milk consumption.

New evidence of the benefits of raw milk may give the dairy industry greater incentive to find effective and affordable cold processing methods that destroy harmful bacteria while keeping enzymes, proteins, pro- and prebiotics, and other beneficial elements intact and active. Or, it may prompt the development of clean milking production standards that will allow farmers to provide a raw milk source directly to consumers with less risk of contamination.

Raw milk contains probiotics which enhance immunity.<sup>1</sup> In Europe, children given raw milk have stronger immune systems thus resisting infections and allowing their bodies the full opportunity to grow.<sup>2,3</sup> With stronger immune systems, children who consume raw milk have fewer incidences of allergies, asthma, and atopic diseases compared to children consuming pasteurized milk.<sup>4,5,6,7,8</sup> In many cases, the earlier and longer the consumption of raw milk, the greater the protective effects.<sup>8</sup> Infants fed certified raw milk have greater growth rates, fewer gastrointestinal illnesses, and lower mortality rates than infants fed pasteurized milk.<sup>9,10</sup>

Heat processing changes pro-and prebiotics, enzymes, proteins, peptides, and other components in milk. Alternatives to heat processing have been explored to find ways to reduce harmful microbial contents and improve the shelf life of milk. Cold microfiltration (MF) effectively removes vegetative bacteria, spores, and microbial cells while keeping milk proteins intact.<sup>11</sup> Other methods of cold processing that are still in the early technological exploration phase include: ultraviolet, pulsed electric fields (PEF), ultrasound, cold plasma, and high hydrostatic pressure.<sup>12</sup>

The purpose of this study was to determine whether raw milk demonstrates nutritional superiority over pasteurized milk. This study compared the effects of consumption of raw vs. pasteurized milk on growth and fertility rates in 18 mice. Weanling mice were chosen because they had been fed a diet that consisted solely of milk.

## MATERIALS AND METHODS

This experiment was conducted with 18 weanling mice. The mice were 21 days old, and weighed between 15 and 17 grams. The mice were randomly divided into two groups: a raw milk group and a pasteurized group.

During the first 12 days of the experiment, the mice in the two groups were separated by gender. Mice grow rapidly and reach sexual maturation around six weeks for females and eight weeks for males. At the end of the 12 day period, the mice were grouped into breeding trios and put in separate cages.

The mice were weighed individually every five days, and combined weights recorded. Raw milk and commercial milk are visually distinguishable as cream rises to the top in raw milk but not in milk that has been homogenized. In order to keep the weights blind, two people worked together to weigh mice, one person doing the weighing in a separate room and the other removing mice from cages.

As the mice began to reproduce, the number of pups in each litter was recorded. Death rates of pup mice were documented. Any pups that survived until the 21<sup>st</sup> day, the day of weaning, were weighed. Death rates of adult mice were also recorded.

The temperature of the animal room was controlled as much as possible at right around 75\* F. Additionally, the lights were controlled with 12 hours on from 7 pm until 7 am, and 12 hours off from 7 am until 7 pm. Mice are not exclusively nocturnal, but they are more active at night. By reversing their sleep-wake cycle from normal sunlight hours, we were able to provide them with a regular supply of milk during the day when the lights were off and they were most active.

The mice had continual, unlimited access to milk via ceramic dish. The dishes held up to 50 ml of liquid – large enough to contain the appropriate volume of milk for the feeding time period. The dishes, being ceramic, were also heavy enough to prevent spilling as mice climbed over and on the dishes. A fresh milk supply was renewed every four hours from 7 am until 11 pm. The mice were provided with milk between 11 pm and 7 am, but it was not refreshed. The amount of milk going into the containers was measured, and any milk that was not consumed was measured before being dumped.

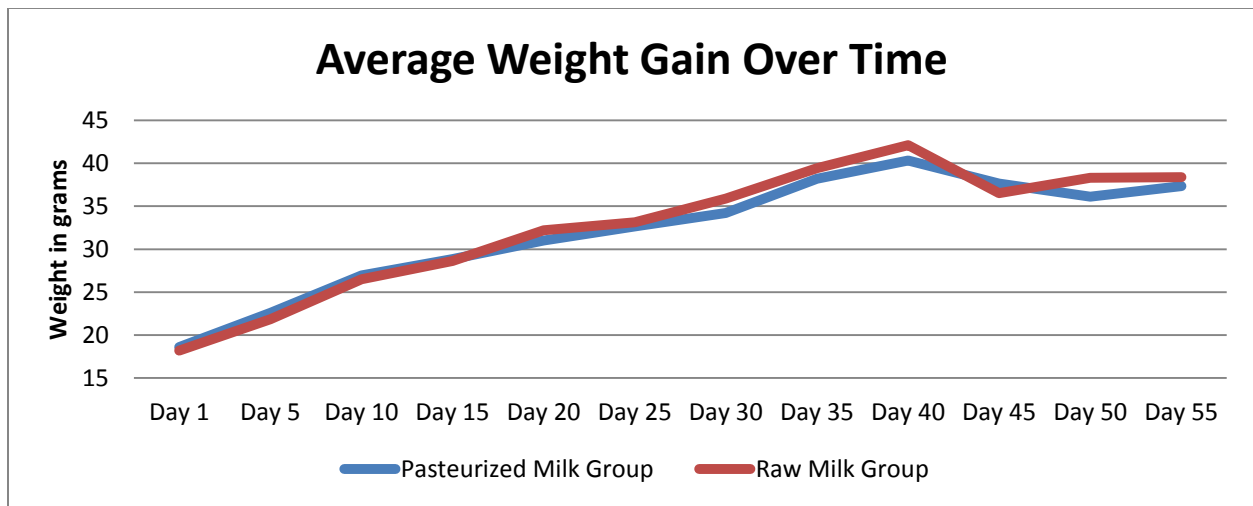
The mice were fed milk as their primary source of food. Milk has no fiber; therefore the mice were supplemented with an unlimited supply of timothy hay as a source of crude fiber. Mice were also provided with unlimited access to water via a drip style container.

The source of the whole raw milk was from cows organically raised with unlimited access to rotationally grazed pasture. The commercial whole milk was also organic; however organic certification regulations stipulate that cows have a minimum access to pasture but not unlimited access. Therefore, there was no way of knowing or guaranteeing that the milk came from grass-fed cows beyond what is required by law for certification. Organic milk was chosen in order to avoid the potential side effects of trace hormones or antibiotics.

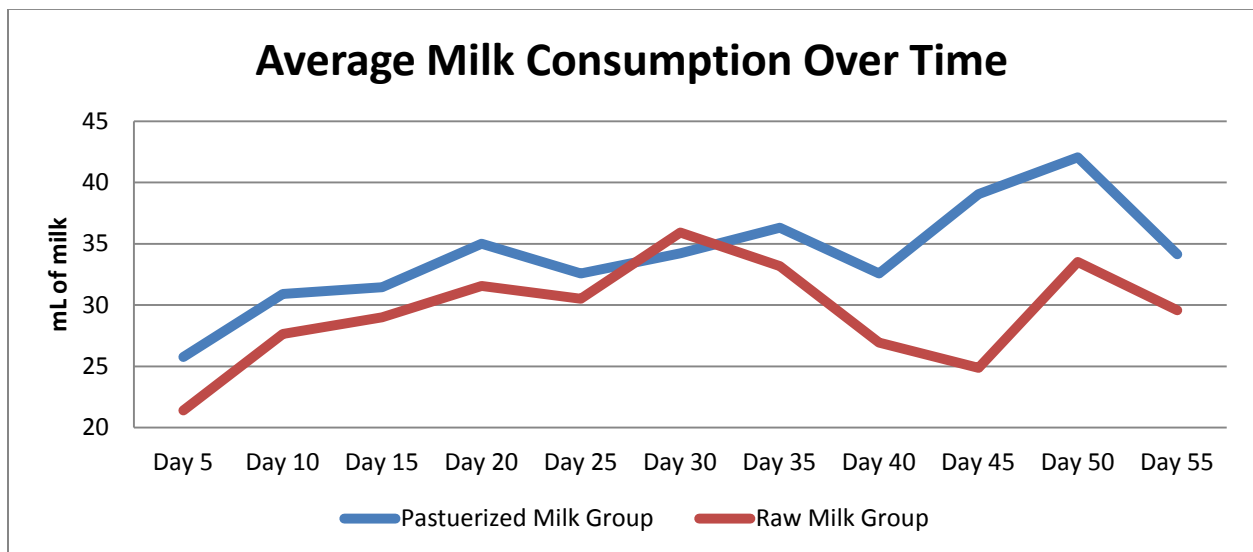
Cages were cleaned regularly but not excessively so as not to disrupt hormone signals for reproduction and to minimize stress as mice become scent comfortable in their environment.

## RESULTS

Data was collected for 55 days.



**Figure 1.** Mice were weighed in a blind fashion every 5 days in each group; weights were averaged.



**Figure 2.** Milk consumption was measured daily by measuring mL of milk provided and milk consumed. Average consumption was calculated and recorded every 5 days.

To test the hypothesis that there would be a significant weight gain between the two groups a Two-sample T-test was conducted with a 95% confidence interval. The P-value score of that test was 0.904. The P-value is greater than 0.05 so there is no significant difference between the groups with regard to weight change.

Another Two-sample T-test was conducted to test for the hypothesis that there would be significant differences in average milk consumption. A 95% confidence interval was used that resulted in a P-value

of 0.021. There is statistical significance between the groups with regard to differences in milk consumption.

A Paired T-test was conducted to determine significance in the difference between groups in weight gain compared to consumption. The P-value score was 0.056 with a 95% confidence interval.

Lastly, a Fisher's exact test was used to compare number of offspring born and the survival rate of the offspring. There were no significant differences between the groups in either category.

## DISCUSSION

The raw milk mice group appeared to grow more efficiently. The difference in growth compared to consumption could have been from the difference in digestibility in the two milk products, or it could have been from the difference in milk fat. The milk fat content in pasteurized milk is regulated at 3.25% for whole milk; raw milk fat content could vary between 3 and 6% depending on the diet of the cows, the breed of cow, and the season. The raw milk fat content was measured periodically at the farm at about 4%. Regular analysis in the lab of the fat content variability on a batch by batch basis would allow for exact caloric measurement compared to growth.

This study was concluded at the 55<sup>th</sup> day of the study instead of the 90<sup>th</sup> day which was much sooner than anticipated. Even though more raw milk offspring survived to day 21, none of the babies were thriving. Their diets were most likely nutritionally inadequate. Cow's milk is really only perfect for cows. A supplement suited for the dietary needs of mice would have been appropriate in addition to milk being the primary food source. This would have ensured greater longevity in the mouse population so that successive generations could have been studied.

This study had a number of limitations. For example, instead of combining mice weights into groups and comparing the averages, tracking weight gains in individual mice would have allowed for greater N values in the data analysis. The only way to track individual mice is to put the mice in breeding pairs instead of trios so the mice are identifiable. The mice groups were regulated by IACUC putting limitations on groupings. Breeding trios were within guidelines but became difficult to work with when the timing of multiple litter pups did not meet regulations. Pup litters born more than a few days apart were required to be separated. With a larger room and more cages, grouping mice in pairs would have been less disturbing for the mice pups, and would have provided the opportunity for easier tracking which in turn would have increased the power for statistical significance. Additionally, there was no control group of mice in order to compare with the two experimental groups.

## CONCLUSION

This study does not demonstrate that either milk is superior in nutritional value for promoting growth and fertility in a colony of mice. However, mice in the raw milk group did appear to grow more efficiently even though the numbers did not reach statistical significance. If this study were conducted with a larger population of mice grouped in pairs instead of trios with individual tracking of weights as well as with an appropriate supplement the results might have been different. Current evidence on probiotics and repeated exposure to environmental bacteria suggest that both strengthen immune

system development. Studies that can obtain measurable benefits and help discover evidence to support consumption of raw milk may make it reasonable to support access to raw milk for consumers in the future.

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