What is your background and role within this research project?

I come from a medical engineering background and was previously developing vacuum therapy devices and cardiac implants, including delivery systems. In 2008, I joined the Hartmann Human Lactation Research Group at the University of Western Australia and learned how to analyse human milk components. Later, I started a PhD in which I investigated the effect of novel pasteurisation methods on certain human milk components.

When is an infant considered preterm, and why is human milk important for these babies?

A preterm infant is born earlier than 37 weeks gestational age. The earlier an infant is born, the more difficult it is for the mother to initiate lactation, as her baby is unable to suckle at the breast. The mother, therefore, does not often have sufficient stimulus to produce milk, and milk is not being removed to ensure increased production. Therefore, the mother

AFTER GIVING BIRTH, a mother’s body will automatically produce milk that has a unique composition of fats, carbohydrates, proteins and vitamins to provide targeted nourishment for her baby. The living cells that make up breast milk help protect babies from infections; in fact, feeding a baby breast milk is one of the most effective ways to ensure child health and survival, according to the World Health Organization (WHO). It describes breastfeeding as ‘an unequalled way of providing ideal food for the healthy growth and development of infants’. It recommends that for the first six months of an infant’s life, the baby should be exclusively breastfed to achieve optimal growth, development and health. This recommendation is especially important to follow when a baby is born prematurely. While there are artificial formulas and supplements specifically designed for premature babies, they lack the antibodies and other protective agents found in breast milk. Therefore, when a mother cannot provide her child with breast milk, donor human milk is the best alternative.

PASTEURISING DONOR HUMAN MILK

Before a donor’s breast milk can be fed to newborns, it must first be pasteurised to prevent the transmission of bacteria and other pathogens. The process of pasteurisation is regulated at both a national and local level, adhering to milk banking guidelines. The most common method of pasteurisation used at such facilities is known as Holder pasteurisation, which is capable of eliminating common bacteria and pathogens transmitted through human

Novel pasteurisation methods for donor human milk

The standard treatment method for donor human milk deactivates bioactive components essential for preterm infants’ health. Research from the University of Western Australia has discovered alternative pasteurisation methods...
must pump her milk for her baby. However, due to the stresses of having a premature baby and reliance on the pump, many women are unable to reach full milk production.

In the case of the preterm infant, all of the infant’s systems are immature. Human milk is highly beneficial in this situation, mainly because of its bioactive components, including immunological and developmental proteins, digestive enzymes and cellular components.

Research has shown the use of the mothers’ own milk leads to lower mortality rates, shorter hospital stays with lower incidence rates of various infections, better neurodevelopment and better health in adulthood. However, until the preterm mother produces enough milk, a substitute has to be used to ensure adequate nutrition and growth in the preterm infant, as poor growth is associated with detrimental health outcomes. In these cases, donor human milk is considered superior to preterm formula.

Milk banks pasteurise donor human milk using the Holder pasteurisation method. Why does the milk need to be pasteurised, and what are the advantages and disadvantages of the current method?

There are no global guidelines mandating that donor human milk has to be pasteurised. However, many regional and local best practice guidelines require pasteurisation to prevent transmission of diseases from a donor to a preterm infant. The most common method used is the Holder method in which human milk is heated to 62.5 °C, kept at that temperature for 30 minutes and immediately cooled. The advantage of this method is that it is a relatively simple process that has been used successfully for almost a century.

This process is capable of destroying a wide range of pathogens, from vegetative bacteria such as Staphylococcus aureus and Escherichia coli to viruses such as HIV and cytomegalovirus (CMV). However, the disadvantage is that the process inactivates many major bioactive human milk components.

Can you talk about these bioactive components and explain their importance for preterm infants?

Not all human milk components are affected by Holder pasteurisation. For example, lactose, epidermal growth factors and vitamins A, D and E are preserved. However, as an example, the activity of bile salt stimulated lipase (BSSL) is eradicated completely. This enzyme is used by the infant to break down the lipids in human milk, thus aiding in digestion. The effects of the absence of BSSL have been shown in research, where the fat absorption was reduced in preterm infants fed pasteurised compared to unpasteurised human milk. Furthermore, some of the major immunological proteins including lactoferrin, lysozyme and secretory IgA are reduced by 80, 60 and 30 per cent, respectively. These proteins are known for their anti-inflammatory and anti-infectious activity, and they offer protection to the preterm infant.

You investigated two alternative pasteurisation methods: ultrasonication and ultraviolet-C irradiation. Did either one preserve the bioactive components in donor human milk?

Ultraviolet-C irradiation showed superior results when using lipase as a biomarker of treatment. The waste heat during the ultrasonication significantly denatures lipase, whereas ultraviolet-C does not reduce lipase activity at the dosages required to reduce vegetative bacteria. Because ultraviolet-C does not produce heat, we focused on this method for the greater part of the project.

We subsequently showed that lipase and alkaline phosphatase activity were not changed during ultraviolet-C pasteurisation compared to a complete loss during Holder pasteurisation. The fatty acid composition of human milk was also not changed during the ultraviolet-C irradiation. Furthermore, the loss of lactoferrin, lysozyme and secretory IgA was significantly less than that with the Holder method.

Lukas Christen, a PhD candidate at the University, has led the study with Professor Peter Hartmann and the Human Lactation Research Group. Christen has taken a multidisciplinary approach to his research, which now focuses on the optimisation of the treatment of human milk, and his experiment is the first of its kind to incorporate non-thermal methods of pasteurisation.

Though common in the food industry, where non-thermal methods help optimise the taste and flavour of food as well as manufacturing costs, no such research has been conducted on human milk. Before embarking on the study, Christen hypothesised that: “A novel treatment method can eliminate pathogens as effectively as the current thermal method with a higher retention of bioactive components in donor human milk and therefore increase its overall quality”.

ULTRASONICATION AND ULTRAVIOLET-C IRRADIATION

In order to test the ultrasonication and ultraviolet-C irradiation pasteurisation methods, the Hartmann Human Lactation Research Group gathered milk from a variety of mothers whose milk supply was plentiful. The team introduced bacterium into the milk until it reached the ‘maximum’ level, as defined by Australian national milk banking recommendations. They then used both alternative methods to test the samples of milk several times, to show beyond any doubt the applied pasteurisation processes were responsible for causing the changes between the treated and untreated samples.
INTELLIGENCE

ULTRAVIOLET-C IRRADIATION: A NOVEL PASTEURISATION METHOD FOR DONOR HUMAN MILK

OBJECTIVES

Holder pasteurisation (milk held at 62.5 °C for 30 minutes) is the standard treatment method for donor human milk. Although this method is able to deactivate most bacteria, it also deactivates important bioactive components. Therefore, the objective of this study was to investigate ultraviolet irradiation as an alternative treatment method for donor human milk.

KEY COLLABORATORS

Research Assistant Professor Ching Tat Lai; Professor Peter Hartmann; Associate Professor Donna T Geddes, School of Chemistry and Biochemistry, Faculty of Science, University of Western Australia

Dr Ben Hartmann, Perron Rotary Express Milk Bank, King Edward Memorial Hospital, Australia, and the Centre for Neonatal Research and Education, University of Western Australia

PARTNERS

Medela AG and Carag AG, Martin Elbel, Head Corporate Communications, Medela AG, Lättichstrasse 4b, 6341 Baar, Switzerland

FUNDING

Medela AG, Switzerland • Carag AG, Switzerland

CONTACT

Lukas Christen
PhD Candidate
Carag AG
Bahnhofstrasse 9
6340 Baar
Switzerland
T +41 41 766 02 02
E lukas.christen@carag.com

www.plosone.org/article/info%3Adoi%2F10.1371%2Fjournal.pone.0068120
www.plosone.org/article/info%3Adoi%2F10.1371%2Fjournal.pone.0085867

LUKAS CHRISTEN completed his PhD in Biochemistry in 2013 at University of Western Australia with Professor Peter Hartmann and the Human Lactation Research Group. The thesis is currently under examination. Lukas’ background includes a degree in medical engineering and several years working in a medical device engineering company in Switzerland. With a multidisciplinary approach, Lukas’ research focuses on the optimisation of the treatment of human milk. In particular, the improvement of the pasteurisation process of donor human milk with an aim of reducing the loss of bioactive components.

Studying these two models of pasteurisation has been particularly fascinating for the team, with each method varying widely in its ability to preserve bioactivity. With ultrasonication, the researchers sent shockwaves through the milk, which caused certain cells to die. Ultraviolet-C irradiation, however, directly affected the DNA, causing microorganisms to become inactive.

A shortcoming of the ultrasonication method was its inability to eliminate microorganisms that lack a particular type of membrane, such as non-enveloped viruses. “Some bacteria, such as Staphylococcus aureus, have a high resistance to the shockwaves and require higher ultrasound energies. Another disadvantage of this technique is that high ultrasound energies create waste heat, which is known to inactivate bioactive components. However, ultraviolet-C irradiation has the disadvantage that its ability to penetrate opaque liquids is very limited,” explains Christen. The research team realised that in order to overcome this, it would have to present the microorganisms to the photon, instead of the photon to the microorganisms. They achieved this by carefully stirring human milk around an ultraviolet source, with the method proving viable for reducing the number of microorganisms.

RESULTS AND THE FUTURE

Using lipase as a biomarker, ultraviolet-C irradiation proved a better method for preserving the bioactive components in donor human milk. This is because the method did not lessen lipase activity, though ultrasonication did. Further research discovered that, unlike Holder pasteurisation in which alkaline phosphatase and lipase activities diminished, both enzyme activities remained essentially unchanged during ultraviolet-C pasteurisation. Also, the fat content of milk did not change, and overall the process proved successful for protecting several essential proteins, including lactoferrin, lysozyme and secretory IgA.

The study has been a success, with results favouring ultraviolet-C irradiation as an alternative method of pasteurisation to improve the quality of donor breast milk. The next step will be to test the effectiveness of treated milk on preterm infants. Though yet to be studied, the team hypothesises that by conserving lipase, the speed at which premature babies gain weight will increase. Moreover, by conserving a higher number of immunological proteins, they suspect inflammation and infection rates will fall.

The team plans to carry out more research on ultraviolet-C irradiation, and it will continue to test its viability as an alternative method for pasteurising human milk. “Further experiments are being planned to determine if there are any negative effects to other human milk components due to ultraviolet-C pasteurisation. Further to this, we are investigating the effect of this treatment on viruses in human milk,” elucidates Christen.

Indeed, for the safety of premature infants, it is vital to preserve the bioactive components of the pasteurisation process. The Hartmann Human Lactation Research Group is very close to showing that ultraviolet-C irradiation treatment of human milk could be the best possible pasteurisation method to address the preservation of these components.

PRESENTING TO THE PUBLIC

Lukas Christen will present his research results at:

• The 9th International Breastfeeding and Lactation Symposium; Madrid, Spain; 4-5 April 2014

Hartmann Human Lactation Research Group members will present the team’s research at:

• Experimental Biology 2014; San Diego, California, USA; 26-30 April 2014

• The meeting for the International Society for Research in Human Milk and Lactation; Kiawah Island, South Carolina, USA; 23-27 October 2014

VERY IMPORTANT PATRONS

Medela AG – producer of technologically advanced breast pumps and medical vacuum technology
Carag AG – a medical technology engineering company