Cheese May Be Good for Blood Circulation

- Dairy foods protect against cardiovascular disease.
- Salt is a risk factor for cardiovascular disease.
- Natural cheese consumed with a high-salt diet may minimize the adverse effects of dietary salt on microvascular function, despite cheese containing salt.
- Increased natural cheese consumption could benefit healthy elderly people even on a high-salt diet.

Cheese is much more than just food. It is a part of the compelling story of ancient and modern human civilization. The huge range of cheeses today reflects the diversity of human taste and history. Cheese types also became a metaphor for public opinion. As Charles de Gaulle frustratingly said, "How can you govern a country (France), which has 246 varieties of cheese." Adding to this impressive résumé of achievements, investigators recently demonstrated that natural cheese may also be good for blood circulation in older adults [1].

A Brief History of Cheese

People at the dawn of civilization realized that milk from livestock quickly spoiled and became unsuitable for human consumption. All that nutrition and energy in milk could be wasted at a time when the next meal was often uncertain. Cheese was the solution to this problem as it provided a convenient and long-term means of concentrated storage of the nutrients in milk. Cheese could be eaten when times were tough or traded when plentiful.

About 20–30% of cheese is protein, which, when digested into its constituent amino acids, is the basic building material for human growth, development, and metabolism in the young, and body maintenance in adults. Cheese also has fats that provide energy. Why is cheese storable for long periods? Initially in the process, cheese makers add special bacterial species to milk, which live on the sugar (lactose) in milk and ultimately give each cheese type its characteristic taste. Depletion of the lactose in the milk by these added bacteria makes it hard for other spoilage bacteria to live. Later in the process, the cheese maker adds salt and removes moisture, which also prevents spoilage. Salt is an excellent food preservative, but today it is known that too much dietary salt is bad for a person’s health [2].

Health Benefits of Dairy Products

Multiple groups of investigators who examined results from many scientific experiments and clinical trials conclude that the currently recommended level of dairy food consumption is linked with decreased risk of cardiovascular disease (CVD) [3-5]. CVD is a broad class of diseases that affect the heart or blood vessels and includes coronary heart disease, stroke, heart failure, and high blood pressure [6]. The American Heart Association recently reported that CVD in the USA affected nearly half of the adult population in 2016 and it is the leading cause of death in the world [6]. Sobering statistics! The greatest risk factor for...
CVD is age, but diet and lifestyle are strong and modifiable risk factors [6]. There is not much that can be done about ageing, but modifiable risk factors point toward practical ways of reducing the incidence of CVD. Most people eat far less than the recommended level of dairy products in their diets, especially the elderly [2], and hence are at greater risk of CVD. Scientists have also demonstrated that eating the recommended amount of dairy food each day improves cardiovascular health by lowering blood pressure and reducing arterial stiffness [7, 8].

Health authorities may be able to reduce the incidence of CVD by better promoting their recommendation that people eat three dairy servings each day, with a focus on low-fat dairy products [2]. But it’s not that simple. Some dairy products like cheese contain salt, which when present in non-dairy foods is a risk factor for CVD [9, 10]. Unexpectedly, a recent clinical trial demonstrated there is something special about cheese consumption. It preserves normal blood flow in very small blood vessels in healthy elderly people with normal blood pressure, despite their high-salt diet [1]. Turophiles (cheese lovers) should celebrate!

**The Salt of It**

Scientists in many investigations conclude that too much dietary salt, or sodium chloride, reduces the kidneys’ ability to remove water, which results in extra fluid in the blood, increased blood pressure, and a chronic strain on major blood vessels [1, 11]. An excess of salt in a diet also independently affects the functions of very small blood vessels by reducing blood capillary density in tissues, changing the capillary internal structure, and altering the responsiveness of capillaries to normal chemical regulatory signals [11]. Scientists suggest that these latter effects in small blood vessels are not well understood and could be a warning of future CVD risk for people who have normal blood pressure [1, 11]. The United States Department of Health and Human Services and the United States Department of Agriculture indicate that most people in the USA far exceed their recommended daily intake of salt [2]. Although dairy products decrease the risk of CVD [3-5], scientists suggest that it is important to independently measure the effect of cheese consumption by elderly people on both low- and high-salt diets.

**Cheese is Good for Blood Circulation**

Billie Alba and five colleagues from Pennsylvania State University and the Ohio State University recently published results from a clinical trial assessing the impact of cheese on blood microcirculation in an elderly but healthy population averaging 64 years of age [1]. Although the number of people assessed in the clinical trial was small, the results were very interesting, and potentially very useful.

The clinical trial was a turophile’s dream come true. The participants in the trial who were lucky enough to be allocated to the right group on day one received a choice of excellent natural cheeses, four times a day and were also given a controlled diet containing either high or low quantities of salt. Others were initially not so lucky. They received only the high- or low-salt diet but no cheese, to rub salt into the wound. At the end of seven days on the diets, the investigators assessed the participants for their levels of a range of factors including blood pressure, urinary sodium, and several blood components. The investigators also measured dilation (expansion) of very small blood vessels in the arm in response to a range of concentrations of a normal signalling chemical, acetylcholine. They explained that the latter measurement assessed a normal function of small blood vessels, i.e., localized microvascular response to acetylcholine. The inability of very small blood vessels to dilate, possibly due to stiffness, may be an early indicator of increased risk of CVD [1, 12, 13]. After a week’s rest, the groups were swapped to a different diet by the investigators and again assessed for their responses after seven days. In this way, each participant received each of the four diets (low-salt, low-salt with cheese, high-salt, and high-salt with cheese). Thankfully, all participants eventually sampled the variety of cheeses on offer.

The investigators ensured that the four diets contained equal quantities of total fat, carbohydrate, and protein to control for the additional nutrients in cheese. Importantly, the clinical trial was “blinded” to the investigators, i.e., the core investigators did not know who was receiving a specific diet at any time in the trial. This design feature prevents inherent investigator bias creeping into the experimental data.

The first result that captured the investigator’s attention was the amount of urinary sodium excreted by
the trial participants on both high-salt diets. It was about three times greater than that for participants on the two low-salt diets. That’s a big difference. The kidneys of the elderly participants were certainly working well! Moreover, as the investigators reported that the blood pressure of the participants was largely unaffected by the high-salt diets, all the elderly participants except one were classed as “salt-insensitive.” This result independently confirmed an important selection criterion used for the original enlistment of participants in the clinical trial, i.e., the participant blood pressure had to be normal and unaffected by salt. The investigators removed the person who was salt-sensitive from the trial and one other person for noncompliance with the diets.

The investigators then demonstrated that microvascular dilation in response to localized administration of acetylcholine in the arm was diminished on the high-salt diet compared with either the low-salt diet or the low-salt diet with cheese [1]. This result demonstrated that the high-salt diet (without cheese) compromised normal microvascular function even though the participants had normal blood pressure. Perhaps this was an early warning sign. The exciting result was that microvascular function for the participant group receiving the high-salt diet and cheese was the same as for the groups on the low-salt diet or the low-salt diet with cheese. Thus, consuming cheese with the high-salt diet somehow neutralized the adverse effects of the high-salt diet on microvascular function, despite the cheese containing salt. What’s so special in cheese?

Alba and colleagues also investigated the mechanism underlying the beneficial effect of cheese in healthy people on a high-salt diet [1]. Their experiments and results from other scientists led Alba and colleagues to conclude that dairy proteins in cheese prevented salt-induced oxidative stress in small blood vessels [1, 14–17]. Oxidative stress in the body results from an imbalance between free radicals and anti-oxidant chemicals. Free radicals are highly reactive small molecules that indiscriminately alter nearby molecules carrying out important biological functions. Cheese proteins tipped the oxidative balance against free radicals leading to less molecular damage and maintenance of normal biological function.

Implications

It’s hard to change a lifetime of dietary habits. Some are bad, like a high-salt diet. But there is hope. The incorporation of the recommended amount of natural cheese into a high-salt diet "may be an effective strategy to reduce cardiovascular disease in salt-insensitive, older adults" with normal blood pressure and no other health problems [1]. The cheese résumé just became more impressive. Pass the Gouda and Edam, please!

Human Milk Reduces Gut Inflammation after Bone Marrow Transplant

- Complications associated with bone marrow transplants include damage to the gut and disruption of normal gut microbiota.
- A small pilot study found that enteral feeding of human milk in children undergoing bone marrow transplants was associated with a reduced incidence of complications, such as graft-versus-host disease, a reduction in markers of inflammation, and a reduction in markers of intestinal injury.
- These promising results require validation in a larger, randomized, and blinded study.

The human newborn’s gastrointestinal (GI) tract is immature and heavily reliant on components from human milk to successfully adapt to the novel challenges of life outside of the uterus. Recent research has highlighted the important role of milk’s bioactive components in establishing a healthy gut microbiome [1–4]. Starting life off with the right mix of bacteria in the GI tract is essential not only for the development of the gut but also for mucosal immunity [1, 4]. It is so essential, in fact, the gut microbiome has been referred to as an ancillary immune organ [1].

Colonizing commensal bacterial populations influence the production of proteins that help gut barrier cells stick together, train immune cells to recognize disease-causing bacteria, and prevent gut inflammation [1–3]. Dr. Stella Davies, Director of the Division of Bone Marrow Transplantation and Immune Deficiency at Cincinnati Children’s Hospital and a Professor of Pediatrics at University of Cincinnati, was listening to colleagues at Cincinnati Children’s Hospital Center for Breastfeeding Medicine discuss these very attributes of human milk in the neonate’s gut when it occurred to her that milk’s ingredients could be beneficial to bone marrow transplant patients as well.

With any type of transplant, the non-functioning or diseased body part must be removed before being replaced. Organs can be removed surgically, but removal of damaged bone marrow requires intentionally giving a patient a lethal dose of chemotherapy before transfusion with healthy bone marrow stem cells [5, 6]. Such a high dose of chemotherapy, unfortunately, has the potential to damage other healthy tissues in the process, particularly the gut. Gut mucosal cells respond to injury by producing inflammatory chemical messengers, which in turn alter the composition of the gut’s bacterial communities. The disruptions in the microbiome lead to dysbiosis—less diversity in bacterial species, reduction in beneficial species, and greater numbers of potentially pathogenic species—and have been linked to the development of graft-versus-host disease (GVHD) and bloodstream infections (BSI), both of which are significant complications of bone marrow transplants [5, 6].

Dr. Davies hypothesized that providing human milk to children after bone marrow transplants could prevent dysbiosis and reduce the incidence of GVHD [5, 6]. There have been concerns over the safety and efficacy of probiotics, such as *Lactobacillus*, to immune-compromised patients to help combat dysbiosis; namely, the risk of these bacteria moving from the intestines into the bloodstream (translocation) [7]. But
Dr. Davies believed that human milk would not have these same issues because it contains numerous immune components that could potentially protect gut tissue by preventing an inflammatory reaction [6].

Davies did have one major concern, however, about testing her hypothesis. “I was unsure of whether parents would be accepting of giving their children human milk,” she explains. But as it turns out, this was the only thing she was wrong about.

After establishing that human milk was well tolerated by children (ages 6–40 months) undergoing bone marrow transplants [5], Davies and colleagues performed a randomized trial to compare transplant outcomes in children (age 0–5 years) receiving human milk or a standard formula via feeding tubes [6]. Initially, their study design called for randomly assigning 42 patients to the human milk versus control arm of the study in a 2 to 1 ratio, or approximately 28 patients receiving human milk and 14 controls [6]. However, nine patients that were breastfeeding at the time of enrollment were automatically assigned to the human milk arm (and were given mother’s own milk orally rather than donor milk). These nine were added to the 24 randomly assigned patients for a total of 33 children receiving human milk.

The study design was further complicated by the fact that parents did not want their children to be controls. From the nine randomly assigned control participants, three left the study when they found out they were not getting human milk and two secretly obtained human milk from other sources and informed the study staff a year after completion, leaving only four valid controls [6]. It was the opposite of what Davies had anticipated. “Mothers understood how important breast milk is and desperately wanted to have it for their babies,” she recalls.

Although it would be ideal to provide human milk for all of the children, controls are necessary to demonstrate efficacy of treatment. Luckily, the team was able to include 10 controls from a previous study that met the study eligibility requirements and had all of the necessary samples stored, bringing the control arm to 14 patients [6].

In addition to looking at incidence of GVHD and BSI, the team collected stool and plasma samples at enrollment and on day 14 after treatment, and performed analyses on all samples that would tell them about the levels of inflammatory and anti-inflammatory chemical messengers in the blood, the types of bacteria present in the stool (a proxy for the gut microbiome), and even individual metabolites, such as fatty acids and amino acids [6].

As predicted, GVHD incidence was lower in the human milk arm, particularly in incidence of grades 2–4 (or the more severe forms) of GVHD [5, 6]. This result is likely due to a confluence of factors demonstrating reduced intestinal inflammation in patients receiving human milk: bacteria associated with GVHD and BSI had lower detection in stool from the human milk arm compared with the control arm; plasma REG3α, a marker of intestinal injury, was lower in the human milk group, suggesting a protective effect of human milk on gut mucosal cells and possibly gut barrier function; and T-cell activation, a marker of an inflammatory response, was lower in children receiving human milk.

These findings are quite remarkable, but they must be considered provisional due to the small size of the study, the lack of blinding (patients knew what treatment they were getting), and the fact that not all patients were randomized.

Davies acknowledges these limitations. “We absolutely need to blind the study and randomize it,” says Davies about the next steps. Additional studies may also include only some milk components, such as human milk oligosaccharides, or oligosaccharides and immune factors, in order to pinpoint which specific ingredients are responsible for the differences in gut inflammation. But Davies admits that it is highly likely it is a synergistic effect.

“Ideally, what we would like to do is identify the components that were valuable and synthesize these to put into a pill,” explains Davies. "But it could very well be that it is a bit of a magic mix and it is several
components working together.”


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Chew on This: Softer Diets of Preindustrial Dairy Farmers Influenced the Shape of Their Skull

- A 40-year old hypothesis proposes that as human populations transitioned from a hunter-gatherer subsistence to agriculture, skulls became less robust because of reduced demands on chewing.
- A 2017 study provided the most thorough test of this hypothesis, using over 500 crania and 500 mandibles from foraging and farming populations across the globe.
- The study identified a modest effect of diets of grains and dairy on skull shape, size, and form, with the most substantial changes found in foraging populations.
- Softer foods, like milk and cheese, did have an effect on size and shape of the skull but were minor relative to the influence of temperature and neutral evolutionary forces such as genetic drift and gene flow.

The human family tree has an extinct genus that is remarkable for their massive jawbones, molars, and cranial crests (picture a bony mohawk). All of these anatomical features are proposed adaptations to the tough, fibrous diet of genus Paranthropus; hard and chewy diets require large chewing muscles, which in turn require larger jaw and cranial bones (and crests!) for points of attachment.

Although Paranthropus were an evolutionary side-branch and not directly related to modern humans, these same form-function principles should also apply to the skeleton of Homo sapiens. Over 40 years ago, the masticatory-functional hypothesis was proposed as a way of explaining variation in the shape and form of craniums and mandibles (which together are known as the skull) between early human farming populations and the foraging populations from which they descended. The hypothesis argues that as populations adopted agricultural practices and began to consume softer foods, such as grains and cheese, demands on mastication were reduced and skulls became less robust. Could softer diets bring about changes in the human skull just as tougher diets did in Paranthropus?

It seems like a straightforward hypothesis to test—take measurements on crania and jaws associated with agricultural and foraging populations and then compare the findings. But, as is usually the case with studying humans, it is much more complicated in practice. Diet is only one factor that influences skull size and shape; differences among human groups could also be the result of temperature or neutral evolutionary forces such as gene flow (migration) and genetic drift (small population size, isolation). To tease out these other factors requires a large sampling of skeletal material from across the globe; to date,
studies have been small and mainly sampled from geographically local populations.

But a 2017 study [1] hoped to finally put the hypothesis to a rigorous test by assessing skull shape, form, and size on a global scale. Katz and colleagues selected 37 cranial and 23 mandibular landmarks and digitally recorded three-dimensional data from 559 crania and 534 mandibles, representing 25 groups of pre-industrial foragers and farmers from across the globe [1].

Here’s where things get a bit tricky—these landmark data represent the size and shape of an individual’s skull, which are phenotypes, or observable traits. Some phenotypes, like blood type, can be tied solely to genes (e.g., if you have O blood, you have two copies of the O gene). But many phenotypes, like those measured in this study, are the result of the interaction of genetic and environmental traits. To understand the degree of influence from diet, a non-genetic factor, Katz and colleagues applied a sophisticated quantitative genetics model to their 3D landmark data [1].

Each cranium and mandible was fit to one of three models, each with a different diet predictor: Milk (dairy), Mush (cereals such as rice or wheat), and Soft (all Milk and Mush populations). Dairy and cereals were selected because they represent the softest agricultural diets; if reduced mastication has an effect on the size, shape, or form of the cranium or jaw, it should be easiest to detect in populations with these diets. For each model, the team indicated whether the diet predictor was present or absent. Skeletal elements from foragers would have all three predictors listed as absent, whereas those of dairy farmers would have Milk and Soft listed as present. In addition to diet, each model also took into account the sex of the skeletal element and temperature; both factors have been demonstrated to influence human cranial and mandibular diversity [2].

Katz and colleagues found several specific differences in size, shape, and form between foragers and farmers, with the most noticeable changes in the Milk group: the ramus (which is the part of the mandible that projects upward, along the side of the face) was narrower, which suggests smaller masseter muscles; the point of attachment for the temporalis muscle (which covers the sides of the head by the temples) was lower down on the cranium, suggesting a smaller temporalis muscle; and mandibles were generally smaller in size and less robust. All signs pointed to a reduction in bite force, especially in dairy farmers, exactly as predicted by the masticatory-functional hypothesis [1].

When the results of this study were published in 2017, a string of popular science articles followed with titles implying that eating cheese and other dairy foods directly changed the shape of the human skull. The reality is that the study found a significant but small effect of diet, which is exactly what Katz and colleagues had predicted [1].

Because of migration (and the movement of genes), most human genetic variation is found within groups rather than between them. Variation in human skull shape and form is known to conform to this same pattern; this means that a cranium from a forager and a cranium from a farmer have the potential to be more similar to one another than crania from two farmers from the same population.

Add to that an evolutionary perspective on the transition to agriculture; even before the archaeological evidence for agriculture, human forager populations were cooking, cutting, grinding, and processing foods [1]. As such, the difference in chewing demands between foragers and farmers was probably not terribly drastic. Eating cheese and milk on its own didn’t change the shape of the human skull, but rather it is one category of human cultural practices that influenced the size, shape, and form of the human skull. That might not make for as catchy of a title, but it certainly makes for a more interesting story.


Future Plastic: Biofilms Derived from Colostral Milk Proteins

- Milk protein-based edible films can protect food from spoilage and provide an environmentally friendlier alternative to plastic.
- Cow milk immunoglobulins (Igs) have been added to milk protein-based edible films to increase antimicrobial properties.
- Igs increase the solubility, tensile strength, adhesiveness, and appearance of milk protein-based edible films.

We all know that plastics are bad for the environment, and there is ongoing research indicating they are harmful to humans as well. When microplastics—less than 5 mm in length—get into oceans and tributaries, they end up in the fish and plants that we may consume. But plastic is an integral part of our lives. Computers, cars, and many household appliances are, or include components made of, plastic. Medical equipment like syringes, gloves, and the little plastic filters that go over thermometers for each new patient are one-time use items that help ensure good hygiene. And, of course, much of the food we buy is wrapped in plastic for both convenience as well as protection from contamination. In fact, it’s hard to imagine giving up the assurances that plastic can provide us when it comes to keeping our food safe. But advances in the development of milk protein-based edible films may soon make those wrappers not only less wasteful but even beneficial to our health, thus letting us have our cake and safely eating it, too.

Edible films are not new, as researchers have been experimenting with different ways of making them for more than two decades [1]. In a previous article in SPLASH!, we reported on a study conducted by Jean-Luc Audic and his colleagues from the Université de Rennes 1 in Rennes, France of a then-new film derived from milk casein [2]. Audic et al. demonstrated certain functional properties of milk proteins and whey in the production of protein-based films and biomaterials [2]. Among the benefits of that particular film were protective properties from light, oxygen, and humidity, making it a viable option for packaging foods like string cheese. It was also easily dissolvable, which could make it well-suited for packaged noodles that could be dropped into boiling water without needing to unwrap. Recently, researchers have looked at new ways to improve milk-based films’ antimicrobial properties that could both protect foods from spoilage and also possibly boost our health. These types of films are often dubbed “active films.”

Previously, milk-based edible films were sometimes given antimicrobial properties via non–milk additives. Mixing in organic sorbic, propionic, and benzoic acids, fatty acid esters (glyceryl monolaurate), polypeptides (lysozyme, peroxidase, and nisin), and even plant essential oils (cinnamon, oregano, and lemongrass) have all produced some level of antimicrobial benefit, primarily measured by keeping food from going bad [3]. But in a recent study, researchers looked for a more targeted way to not only prevent contamination but to improve the antimicrobial potential of edible films with the addition of another milk component, immunoglobulin.

In March 2019, research scientists from the Natural Resources Institute Finland improved the quality of active films by successfully incorporating active immunoglobulins (Igs) and further doping them with antibacterial proteins that enhance the films’ antimicrobial properties [1]. The amount of Igs incorporated also affected the mechanical properties of the films such as adhesion and tensile strength. Pirjo Rantamäki and her colleagues believed that their team was the first to produce data of the novel Ig-incorporated biofilms.

In their study, Rantamäki and colleagues targeted particular microbes, believing that “If milk Igs which are specific against spoilage microbes or pathogens could be incorporated and released from film materials
in biologically active form, then such materials could be used in products for passive immunization against harmful microbes” [1]. In particular, the team looked at how edible films with added Igs harvested from bovine colostrum could minimize and even prevent dental cavities caused by *Streptococcus mutans*, a common cause of tooth decay.

Rantamäki and her team collected four milkings from 84 pregnant Friesian cows that were immunized against *Streptococcus mutans* and *Streptococcus sobrinus*, both of which are responsible for tooth decay. Milkings from nonimmunized cows were also collected for control purposes. Three types of protein sources were used for the study: β-lactoglobulin-enriched whey protein and two commercial milk proteins. The commercial milk protein used in the study consisted of WPC-75 and DSE1908, and sodium caseinate was also used as a film-forming material.

The films made from each protein source were tested for a number of mechanical properties. Solubility is an important test factor because researchers hope films that dissolve easily in water or saliva could be an effective means of pharmaceutical delivery. When testing solubility, the Rantamäki’s team used water, simulated saliva, and real human saliva and found that the Ig-blended edible films were, in fact, more easily dissolved. “It seems that in films containing high concentration of Igs, the dominant bonds were more easily loosened and more rapidly dissolved,” the researchers observed. “This result can be important regarding the release of various biologically active compounds from edible films.” In particular, Rantamäki’s team was interested in how Igs might be broken down by saliva and help prevent cavities. They found that “When the IP [immune preparation] used in the present edible film study was applied as a mouth rinse by adult volunteers for 3 days, it resulted in a higher resting pH in dental plaque and decreased the relative number of mutans streptococci significantly in the test group when compared with the control group” [1]. It would seem that the Igs could, in fact, bring an added benefit to milk protein-based edible films.

Increasing Ig fractions not only increased the film’s solubility, but also its tensile strength, adhesiveness, stickiness, and appearance. The team found that with increasing Ig concentrations, the film would require more force before tearing. Similarly, the adhesiveness of films increased when increased Ig concentrations were used. WPC-75 films made with increased Ig fractions were also smoother and more transparent than films made without Ig. All of these mechanical properties, of course, increase the appeal of using such films in the food and pharmaceutical industries.

The benefits of using Igs in edible films are plentiful. As Rantamäki’s team notes, “The emergence of antibiotic-resistant pathogen strains will in near future put emphasis on the need to develop alternative ways to prevent and treat oral and gastrointestinal infections” [1]. In the near future, we may see edible films that can prevent *Escherichia coli* or *Salmonella*, and perhaps the wrappers or coatings for candies, lozenges, and even chewing gum could help keep us healthy while minimizing plastic waste. Milk protein-based edible films are biodegradable and reduce the waste that ends up in landfills and our oceans. With the ongoing development of these active films, one thing is undeniably clear. The future is no longer plastic.


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