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REVIEW

Milk Consumption, Raw and General, in the Discussion on Health or Hazard

Ton Baars

Research Institute of Organic Agriculture (FiBL), Ackerstrasse 113, 5070 Frick, Switzerland;
Research Institute at Juchowo Farm, Juchowo 54 A, 78 - 446 Silnowo, Poland

In the last decade, consumption of raw milk, milk in general and milk fats suffers from an adverse nutritional and health image and therefore mixed messages are present among consumers. Milk products contain a range of bio-active contents related to health regulation. Heating destroys not only unwanted and beneficial bacteria in milk, but also changes the activity of enzymes and peptides. In this review, attention is paid to the benefits of milk consumption for asthma, allergies and atopy. Raw milk is a single protective factor for asthma and allergies in children. Also milk fat protects against asthma and within the milk fatty acid composition especially the markers rumenic acid and trans-vaccenic acid, are inversely associated with asthma and atopy. From the position of governmental agencies and medical societies, it is not advised to consume any raw milk. It is argued that raw milk can be dangerous due to the potential presence of zoonotic bacteria. In this article special attention is paid to a vero-toxicin producing *E. coli*. This category of virulent bacteria can be present in the intestines, faeces, skin and environment of ruminating animals. In young children, the physical contact with farm animals as well as raw milk intake are therefore included as risk factors. Another potential danger from the consumption of raw foods including raw milk is the spread to humans of antibiotic resistance forms of bacteria. A differentiation of milk qualities is necessary (produced for direct consumption purpose versus meant for pasteurisation), because part of the population wants to drink raw milk to promote their health. Raw milk is produced in several countries as certified Grade-A milk. Hygienic control of this certified milk demonstrates that the risk of zoonotic transfer can be significantly reduced. Since the evidence has increased that raw milk and specific components of milk are protective for human illnesses like asthma and atopy, a re-evaluation of raw milk consumption as well as the intake of beneficial components of milk from grazing animals is needed.

Keywords: Raw Milk, Fatty Acids, Asthma and Allergies, EHEC, Health Discussion.

CONTENTS

1. INTRODUCTION: (RAW) FULL-FAT MILK CONSUMPTION

Human beings have co-evolved with farm animals and during 8,000 years of agriculture man has adapted to the consumption of raw milk and milk products from ruminating animals. As a cultural characteristic large numbers of human beings, especially those with Caucasian background, developed a genetic adaptation to digest lactose after the suckle phase (lactose-tolerance), a feature not found in other mammals.[1]

Nowadays consumption patterns of milk products that were built up over centuries are under attack. The quality of milk is controversially discussed in relation to potential negative health impacts or dangers facing the claims on health promotion of milk intake in general, milk fats[2] and raw milk chiefly.[3,4] In the 1970s and 1980s, medical

REVIEW

societies and government agencies recommend reducing the intake of saturated fat;[5] milk products should be taken as the low-fat or no-fat variant, because of the epidemic of overweight and cardiovascular problems. There is a warning for the consumption of animal fats, especially n–6 fatty acids (FA) and trans-fats. There is an attack on the consumption of all trans-fats, fats from ruminating animals included. A characteristic feature of milk fat from grazing animals, however, is the high level of rumenic acid (C18:2*c*9*t*11) and its precursor vaccenic acid (C18:1*t*11) as characteristic part of the ruminal trans-fats.[6,7] Finally, there is a controversy about how to deal with the epidemiologic findings on raw milk consumption and the inverse incidence of asthma/eczema and milk-allergies in children?[8–10] Since the zoonotic dangers of any raw milk consumption are judged to be unavoidable,[3] official governmental bodies advise that milk should only be consumed after heat treatment. This advice targets pregnant women, young children, older adults or people with weakened immune systems. The listed doubts about milk consumption leads to the question raised:[11] does raw milk consumption lead to health support in terms of tolerance for lactose digestion, prevention of asthma and reduction of milk allergy or should it be seen as a potential health hazard due to the presence of zoonotic bacteria?

The goal of this review paper is to reflect on the relation of asthma and allergies to raw milk and milk fat consumption, to reflect on effects of heat treatment on raw milk properties and to evaluate clinical trials from raw milk consumptions in animal and man; risk of raw milk consumption will be discussed in relation to the potential for transfer of antibiotic resistance as well as zoonotic disease through milk. Information concerning the enterohemolytic (EHEC) and shiga-toxin producing (STEC) subtypes of *E. coli* are taken as an example.

2. EPIDEMIOLOGY OF FARM MILK AND ASTHMA AND ALLERGIES

A model to explain the increase of asthma and allergies within westernised societies is based on the hygiene hypothesis.[12] A mixture of new lifestyle factors is involved in modern living, among others the increased hygiene-standards and disinfectants in households, the repeated use of antibiotics and anthelmintics in early life, the smaller family size and the reduced intake of fermented foods rich in Lactobacilli.[13] Studies on a more traditional lifestyle within Western societies showed that children aged 6–15 with an anthroposophical lifestyle had less atopic disorders than control children. Elements of this lifestyle associated with improved health were the reduced use of antibiotics and antipyretics, longer breastfeeding, less active immunisation in early life and the consumption of organic as well as fermented foods with active lactobacilli.[14,15]

Braun-Fahrländer and Mutius[9] reviewed eleven epidemiological studies in relation to asthma and allergies among farm children. The studies could show that children who grew up on dairy farms and consumed un-boiled, fresh farm milk were strongly protected from asthma, hay fever and allergic sensitization. In the ALEX-study, the protective impact from the farming environment as well as the farm milk consumption by both pregnant mothers and their newborns in their first year of life on later emerging asthma and allergies at primary school age was shown.[16] Later in the PARSIFAL study, the protection of farm milk in both farm and non-farm children was found.[17] Bieli et al.[18] showed the impact of the genetic component and made clear that several genotypes of children were better protected than others. In contrast to the epidemiological studies in Central Europe (ALEX, PASTURE), which were done on smaller sized family farms and where the total farm families were strongly involved in the daily handling of their cattle, also children at larger dairy farms in Shropshire (UK) were investigated.[8] The authors compared three groups: farm children, non-farm children living in villages in the countryside and children whose fathers were farm labourers, but not living on farms, thereby separating the effects from 'living on the farm' and 'raw milk consumption.' Based on IgE skin prick test reaction to allergens in all three groups of children, an inverse relation was found for asthma and atopy with the consumption of raw farm milk, independent of the farm status of the child. Additionally, the authors found as a second pattern a volume of consumption effect, which showed that protection

Ton Baars is a researcher for milk quality in relation to animal and human health aspects. Since 2011 he held a position at The Research Institute of Organic Agriculure (FiBL) in Frick (CH) as well as a research position at the biodynamic farm Juchowo Farm (Silnowo, Pl). In Poland he is building up a research institute for applied organic research. As a trained agronomic ecologist he worked 25 years in The Netherlands at applied research in organic animal husbandry, and later he held a position as Professor for biodynamic Agriculture at Kassel University (2005–2011). At Kassel University research projects have been taken up about raw milk quality and human health. His main focus is on ways to develop raw milk products, which are safe for direct consumption and has a high inner quality to protect and support human health.

was greater if raw milk was more frequently consumed. A differentiation was made according to the symptoms, and unpasteurised milk was inversely associated with current eczema symptoms and seasonal allergic rhinitis, but not with current asthma symptoms. This contrasts with the protective effect of the farming status of the child that protected for asthma and allergic rhinitis, but not eczema. This was confirmed among Greek children[19] as well as among Northern-German children,[20] who showed reduced atopy after raw milk intake. Protective effects from both raw milk and yoghurt intake in New Zealand children were shown.[21] Within the large GABRIELA study (7,682 children) an overall protection of 'living on a farm' was confirmed.[22] Both 'contact with cows' as a farm factor and 'drinking farm milk' were independent factors of protection, suggesting that there are different pathways of protection, the mucosal airways and the gut respectively. The risk reduction in terms of odd ratios for asthma, atopic sensitization and hay fever of farm children after consumption of cow's milk were respectively 0.68, 0.54 and 0.43. For atopic asthmatic children the 'consumption of cow's milk' remained the only significant protective factor. For hay fever the 'contact with cows' and the 'cow's milk consumption' had a significant influence. Finally, atopic sensitization was independently affected by the 'contact with straw' and 'cow's milk consumption.' Overall, the 'contact with straw' was a strong protective factor except for atopic dermatitis. The authors suggest that either arabinogalactan, a plant-derived oligosaccharide present in grass, hay and straw, or the bacteria and fungi in straw might be a protective agent. In the comparison of stool samples of allergic and non-allergic children, differences were found in strains of *Lactococcis lactis* (G121) and also *Acinetobacter iwoffii* (F78), a bacteria present in stable dirt.[23] Within the GABRIELA-study,[24] the authors looked at the diversity of environmental bacterial exposure and showed that there was an inverse relationship between the probability of asthma and the microbial diversity. Effects were independent of living on the farm. However, children living on farms had less asthma because of the exposure to a wider range of microorganisms than non-farm children.

The strongest implication of raw milk as a protective agent came recently from[10] in the GABRIELA-study within the group of farm children. They first confirmed that the so-called 'farm milk' mentioned in earlier studies is drunk by the majority of farm milk consumers (89.3%) as untreated, and un-boiled farm milk. Many had been exposed to un-boiled farm milk during pregnancy, during the first year of life, or both. Compared with the reference group of farm children exclusively drinking shop milk which is heat-treated, the adjusted odd ratio for children who were exclusively taking raw milk was respectively 0.59, 0.74 and 0.51 for asthma, atopic sensitization and hay fever, whereas the consumption of heated farm milk did not lead to any significant difference compared to processed shop milk. In this study the risk reduction was stronger for asthma compared to atopy.

An early epidemiological study on the health status of farm children was done by Price.[25] He used the level of teeth decay plus physical structure of the jaws and cheekbones as a measure for a healthy youth development. He compared farm children in the isolated Swiss Lötschen valley with farm children in other valleys who came into contact with modernised, refined foods. In the Lötschen valley, only salt was imported; for the rest of their food, the villagers had a self-sufficient life style. Although the study might seem anecdotal, the insights into the health situation from the observational studies on a range of close-to-nature people could act as a standard for a 'normal' health situation of children and adults before the effects of the Western lifestyle brought forth all kinds of health problems like asthma, allergies or being overweight. The nutrition of the children consisted mainly of raw milk from goats and cows, sourdough rye bread and raw milk cheese. Meat was consumed once a week and vegetables were rare. Price found hardly any tooth decay among the valley children on the traditional diets (4.6% of all teeth), whereas modernised diets showed much higher frequencies (29.8%). It is plausible that the way of living and health situation of the Swiss farm children is very comparable to traditional Amish children living in Pennsylvania (US). Here,[26] very low incidences of asthma and allergies in these farm children were found. Price[25] did not describe the way farmers were milking and what level of hygiene was present. In those days, wooden buckets and wooden milk transporting systems were used. We can therefore assume that the presence of fermenting bacteria, like lactococcus, was high. In those days, even 'fresh raw milk' described milk with a high microbial content. Even nowadays it could be shown[10] that the farm children exhibiting high protection against asthma consumed raw farm milk with a higher bacterial load of micrococci, staphylococci and lactobacilli.

Earlier, in the 19th century, it was observed[27] that 'summer catarrh' (allergic hay fever) was only present in the middle and upper classes of society. After making an inquiry at the various London pharmacies and elsewhere, he did not find a single case of hay fever amongst the poor. This observation is confirmed by recent data; within the ISAAC study based on information from 20 countries, a highly positive association is present between the country's gross national income as a measure for wealth and the incidence of childhood eczema.[28]

For farm children and the offspring of pregnant farm mothers, the protection is strongest if both consumed raw milk and if the pregnant mother had a daily work routine in the stable.[16] Therefore, it was postulated[29] that the lifestyle on a farm, the way of living of the pregnant mother with her animals, consuming raw milk products, as well as the care of the newborn, who is in near contact with animals and the bacterial environment at the farm, is a beneficial predisposition to develop a healthy functioning immune system. Roughly, the prenatal phase until the early life exposures within the first year of life are the most important periods of a newborn for the training of the immune system. The sensitization of the newborn already starts before birth. The number of animal species present on the farm determined the priming of the immune response of the child.[30] The mother's contact with animals during pregnancy had greater impact than newborn's contact during the first year of life.[31]

Based on the GABRIELA study, it was stated[22] that the hygiene hypothesis might not hold for atopic dermatitis as much as for respiratory allergic diseases. Results from the PARSIFAL and later the GABRIELA studies both showed the protective element within the farm environment. The diversity of the microbial environment protected for asthma, not atopy. Microbes trigger the innate immune system, and due to the wider diversity of microorganisms on farms, farm children are better protected than non-farm children.[24] Lluis and Schaubs[32] summarised the protective elements within the farm environment and stated that the novel studies strengthen the role of microbial exposure, farm milk and grass components, especially early in life. These exposures modulate the immune system, subsequently leading to a long-lasting lower risk of developing atopic diseases.

Apart from milk heating several other lifestyle and nutrition factors are mentioned in relation to protection for asthma and allergies. In the birth cohort study PASTURE, the early introduction of six complementary food items in relation to atopic dermatitis were investigated.[33] Feeding practices in the first year of life affect the risk for atopic dermatitis and each additional introduction of a main food item reduced the risk by 25%. The introduction of yoghurt showed a strong protective effect independently of the overall diversity of introduced foods. There was a risk reduction of 60% (adjusted odd ratio (aOR) 0.41; 95% confidence interval (CI): 0.23–0.73). The authors suggest the protective possibility of probiotic-like bacteria within yoghurt, producing short chain fatty acids (SCFA). SCFA can affect the immune and inflammation response. Supplementation of probiotic cultures in the first two years of life reduced the cumulative prevalence of eczema at 4 years hazard ratio (HR: 0.57; 95% CI: 0.39–0.83) and prevalence of rhinoconjunctivitis at 4 years (HR: 0.38; 95% CI: 0.18–0.83) if children were taking strain HN001, not HN019.[34]

Although there is strong, repeatedly found evidence that the early intake in life of raw milk is an important factor for protection against asthma and atopy later in life,[9,10] it is, however, a different question if raw milk can also cure children dealing with adverse reactions to milk. In the literature,[35,36] parents claim that raw milk can be consumed by children with milk intolerance. In a (double blind placebo controlled trial (DBRCT), raw milk, pasteurized milk and pasteurized and homogenized milk was tested in people who reported intolerance to milk, and could not document any differences in the symptomatic responses.[37] Tendencies were found for less experienced pain and less bloating after consumption of raw milk. Five milk allergic children were tested[35] with both the prick test and an oral double blind placebo controlled milk provocation. As a control, a hypoallergenic formula without any milk components was used. All children showed an allergic reaction when receiving any cow's milk in contrast with the placebo. No reaction was seen when challenged with placebo. There was a tendency toward a shorter reaction time to the skin prick test after being challenged by the raw milk compared to pasteurized and homogenized/pasteurized milk (240 versus 162 and 192 minutes respectively) as well as a lower acceptable amount of milk intake (41 versus 23 and 27 ml respectively), indicating a milder reaction to the raw milk. Cow's milk allergy is usually confined to early childhood, often appearing in the first month of life and disappearing at an age of 2 to 3 years.[36] Based on our own experimental testing of allergic children within a double blind placebo controlled trial, a repeated observation is that children could consume increasing amounts of a selected raw milk from a biodynamic farm, whereas the shop milk, being conventional, pasteurised and homogenised led to a quick allergic response,[38] (results will be published elsewhere). Differences between the studies might be because of differences in raw milk origin.

3. MILK FATS AND PROTECTION AGAINST ASTHMA AND ALLERGIES

The milk's fatty acid composition is not affected by pasteurisation. All farm studies mentioned were carried out at mainly conventional European dairy farms with milk from cows also fed silage, maize and concentrates. This milk does not have a FA profile, which might be beneficial for several health aspects, and therefore the mechanism of protection from raw milk is different from that of pasteurised whole milk or the fatty acid composition.[39,40] It was shown[41] that consumption of whole milk rather than skimmed milk was associated with decreased prevalence of hay fever and asthma. Within the PIAMA-study, consumption of full-fat milk, as well as butter, reduced the incidence of asthma in pre-school children.[42] Within the PARSIFAL-study, an independent reduction of asthma was based on butter compared to margarine consumption.[17] In the prospective KOALA-birth cohort study, children of mothers with a high intake of organic milk ($>$ 90% of total milk product intake) had less eczema (at two years of age) and allergic sensitization (at one year of age).[43] Within the KOALA-study, the independent protective effects from the sum of very long chain n-3-polyunsaturated fatty acids (PUFA) (fish fatty acids: EPA, DPA and DHA) as well as ruminant related PUFA (CLA*c*9*t*11 and its precursor C18:1*t*11) were distinguished.[40] In the German LISA study, the so-called fish FA were investigated,[44] and it could be shown that a high intake of margarine and vegetable oils during the last 4 weeks of pregnancy was positively associated, whereas a high maternal fish intake was inversely associated, with eczema in the offspring's first two years. The quality of the FA in the mother's diet is immediately reflected in the breast milk.[45] FA from industrial origin and hydrogenised plant oils are high in C18:1*t*9 (elaidic acid), whereas diets based on milk products from organic and biodynamic origin are high in C18:1*t*11 (vaccenic acid) and showed a low ratio between the two isomers. Bertschi et al.[46] showed that a dietary supplementation with conjugated linoleic acid (CLA)-rich alpine butter changed human breast milk FA and increased CLA*c*9*t*11, whereas similar results were found for Dutch organic milk intake.[47] In mouse models, diets rich in CLA significantly reduced IgE production and bronchial hyperresponsiveness.[48] The impact of season, region and feeding on a range of FA-markers was evaluated.[7] Since milk is a complex food containing over 200 single FA, it makes sense to judge a milk profile rather than the single FA. The authors showed that season, altitude and grazing intensity are the main factors affecting a milk fat profile. Due to legislation and voluntary restriction of concentrate input in the cow's diet, grass based organic systems during the summer season showed the highest potential to produce such a FA profile, especially in Alpine regions, but also in lowland regions if day and night grazing provides the main fodder for the cows.[49]

Price[25] described the food habits of people with traditional nature-based diets. An interesting finding was that among all traditional people a "sacred food" was present, mainly consumed in relation to fertility and pregnancy. One of the main implicit necessities of food choice and nutrition was *"to bring forward a healthy next generation"* without the help of doctors and the presence of hospitals. Also, the farmers in the Swiss Lötschen valley had a special food in relation to the fertility support, pregnancy and development of the young children. This was the (raw milk) butter, made in June and July *"when the cows were grazing near the glaciers."*[25] Nowadays, the milk fat from grazed mountainous pasture is well known for its high levels of n-3 FA, CLA and vitamins.[49–51] The parallelism between the food habits of all traditional people described[25] is the consumption of raw and raw fermented foods as main elements in their diets. There were no refined sugars or canned foods and the fat quality showed high levels of anti-oxidants, n-3 and n-6 PUFA.

A model to explain the differences between different FA-profiles in relation to health is based on the lipid-hypothesis.[52] The change in the Western diet from n-3 towards a surplus of n-6 as well as increased levels of industrial trans FA originates from increased amounts of meat consumed from monogastric animals fed with concentrates and the use of hydrolysed plant oils from palm and soy used in cooking. This led to a change in ratio of the FA-profile in Western diets, where markers as n-6/n-3 went from 1–2:1 towards 10–25:1 (COLOMBUS-study) and also C18:1*t*9/C18:1*t*10 increased.[6] There is a change from grass-based animals feeding only on roughages of grass products to high yielding cows kept indoors and fed a total mixed ratio of conserved products, like maize silage, grass silage and concentrates. Due to the changes in cows' diets, a change in the overall milk FA profile is found. There is a reduction of branched chain FA, several CLA, like CLA*c*9*t*11 and CLA*t*11*c*13 and its precursor C18:1*t*11 and an increase of trans-fatty acids as C18:1*t*10 and C18:1*t*9.[6] Especially in summer, the differences in milk fat profile are strictly separable.[7] Phytanic acid as a chlorophyll derivate is part of the branched chain fatty acids (BCFA) and as a marker for the green roughage in the cow's diet. An inverse correlation between the phytanic acid concentration in milk fat and the percentage of maize silage plus concentrate in the cow's diet was found.[53]

4. CHANGES OF MILK AFTER HEAT TREATMENT

Pasteurisation of milk can be in different combinations of temperature and time span. The goal of heating milk is to kill unwanted bacteria, which could lead to disease or death.

After Holder pasteurisation (62, 5 °C for 30 minutes) 93% of the human donor breast samples did not show any growth on routine bacterial cultures.[54] Before pasteurisation, some milk samples were contaminated with pathogenic organisms. In a large Swiss study, it was shown that the average number of bacteria in raw cow milk was 20,180 bacteria ml^{-1} (average over 12 monthly samplings). After pasteurisation at 72 °C (15 sec) and 92 °C (20 sec) this was reduced to 1,381 and 4 bacteria ml^{-1} respectively, whereas ultra-high-temperature (UHT) treatment led to bacteriologically sterile milk samples.[55]

4.1. Effects of Heat Treatment on Milk Contents

Although the FA composition of milk is not changed after pasteurisation,[56,57] the enzyme activity is completely (lipase) or partly destroyed (amylase).[57] In mother's milk, the Holder pasteurization was compared with the high pasteurization (75 °C for 15 seconds). The anti-oxidant properties of heated milk were reduced and there was a considerable loss of the activity of gluthatione peroxidase, which is one of the major anti-oxidants. Losses were higher at higher heating temperatures.[58] The available lysine in milk, which is seen as a parameter for the protein quality in human milk, was also lost after pasteurisation; however, losses were now higher at lower temperatures.[59] In a review paper on the effect of pasteurisation of human milk, Holder pasteurisation completely

REVIEW

inactivates all cellular components of milk, including T-cells, B-cells, macrophages, and neutrophils.[60] Holder pasteurisation did not affect the concentrations of vitamins A, D, E, B2 and B12. In contrast, there were significant losses of vitamins B6 (15%), C (36%), and folic acid (31%).[61] Relative to freshly expressed human milk, the concentrations of lysozyme, lactoferrin, lactoperoxidase and secretory immunoglobulin A were reduced 50% to 82% in pasteurised donor milk and the activities of lysozyme and lactoperoxidase were 74% to 88% lower.[62, 63] The major milk protein lactoferrin (LF) can destroy microbes and reduce inflammatory responses.[64] Therefore, the proliferation of bacterial pathogens in pasteurised donor milk was enhanced 1.8- to 4.6-fold compared to fresh or frozen human milk.[62] The loss of immunological components were investigated,[65] and the growth of *Escherichia coli* (*E. coli*) and *Staphylococcus aureus* (*S. aureus*) in fresh raw and pasteurised human milk was experimentally tested. The bacterial growth inhibition was significantly reduced after pasteurisation, showing negative effects on the antibacterial properties of raw human milk. Raw milk, Holder-pasteurized milk, and high-pasteurized milk yielded a reduction in *E. coli* growth of 70%, 52%, and 36%, respectively.[66] The reduction of bactericidal properties of fresh raw cow milk was calculated and the lag-phase of *E. coli* at 38 °C was reduced from 160 minutes in raw milk to 40 minutes in all heat treatments (pasteurised as well as UHT milk).[55] The antimicrobial capacity of normal raw human milk with filtered raw milk being sterile from any bacteria and with a removal of macrophages and neutrophils was compared[67] and after inoculation with *E. coli* and *S. aureus*, raw milk showed a reduced growth of these harmful bacteria. Since no differences were found between the filtered and unfiltered raw milk, the focus should be on the importance of the non-cellular constituents of raw milk.

4.2. Changes in Allergenicity Due to Heating of Milk

Cow's milk allergy affects about 2–3% of children.[68] In farm children differences in levels of asthma, atopy and hay fever were correlated with changes in whey proteins due to heating of the milk consumed.[10] Allergenicity of food depends on the predisposition of the patient and on the food itself, including its processing. Beta-lactoglobulin (LG) as one of the main whey proteins is considered to be the principle milk allergen, because of its absence in human milk. Heat treatment changes the native tertiary structure of the protein and will result in the formation of aggregates between casein micelles and whey proteins via the formation of disulfide bonds between kappa- and/or alpha-s2-casein and whey proteins.[69] Heat sensitive proteins undergo denaturation due to the unfolding of their compact globular conformations and aggregation. The concentration of residual native protein depends on a time versus temperature curve and the sensitivity to heat damage is different for each protein. The order of increasing thermal stability of the proteins taking into account irreversible changes was alkaline phosphatase < lactoferrin (LF) < immuloglobulin-G < bovine serum albumin < lactoglobulin (LG) < lactoalbumin (LA).[70] Whey proteins, such as LG, LA and LF that account for 20% of the total milk proteins, are mostly globular proteins and several IgE-binding epitopes have been identified on these proteins in the past. However, caseins, which represent about 80% of the milk proteins, have also been shown to be major allergens. Antibody production was used to measure immunomodulation of whey proteins after heat treatment. Although native LG is strongly resistant to stomach digestion, a sharp LG immunoreactivity was measured when milk was heated over 70–80 °C, converting native LG into denaturated LG with an aggregated structure.[71] Due to the thermolability of LG, values were already high in fresh cheese pasteurised at 66 °C and even higher after sterilisation at higher temperatures, whereas no LG was found in fresh cheese made from raw, non-heated milk.[72] Similar results were found in cow's milk allergic children consuming fully matured Parmesan cheese, where high temperature treatment was lacking (maximum temperature at processing 55 °C). 58% of the cow's milk allergic people could tolerate an oral provocation with 36-month-old Parmesan cheese.[73] Mature Parmesan (> 20 month of ripening) had a high level of casein-degradation, especially for alphaS1-casein, and therefore the immunological *in-vitro* IgE reaction on caseins decreased. The whey proteins were not degraded during the ripening process and were still present in the mature cheese.

In infants the extent of the protein digestion is affected by factors such as the maturity of the gut and its healthy functioning. Furthermore cows' milk whey peptides need to have a molecular weight greater than 3,000 Da (around 25 residues) in order to stimulate an immune response and an allergen must contain at least two IgE epitopes, each of which a minimum of 15 amino acid residues long, in order to make the antibody binding possible. Fragments combining multiple epitopes at least 1 nm in size are required to elicit histamine release.[74] Song et al.[71] could define the immunoreactive site of the newly built epitopes after thermal denaturation of LG and showed that in parallel the binding capacity for retinol, palmitic acid and Vit D3 was reduced after heating above 70 °C.[71, 75] Therefore the transport as well as the uptake function of LG after denaturation might be destroyed. Based on infant *in-vitro* digestion models, it was suggested that IgE-mediated allergy would be connected to an incomplete digestion of dietary proteins causing an inappropriate immune response in the gut and connected with the resistant fragments of beta-caseins found, which corresponded with detected epitopes.[74] In *in-vitro* digestion models, it was observed that raw milk was digested significantly faster with human proteolytic enzymes than the pasteurized and high-heated milk.[76] Milk processing led to differences in peptide patterns and heat treatment of milk tended to increase the number of peptides found in digested samples.[69] Antigens such as aggregated LA and LG, induced after pasteurization, could not be taken up by intestinal epithelial cells, but instead were directed to the mast cells of Peyer's Patches.[77]

During anaphylaxis, there is an acute oxidative stress in the respiratory tract. In mice models, sensitization followed by the crossing of the epithelial barrier of protein aggregates and the anaphylactic reaction has been tested. Subcutaneous reactions and oral provocation of sensitized mice gave different reactions to untreated raw milk and homogenized milk. When given orally, no antibody production was found compared to subcutaneous reaction (respectively 8 and 71% of the animals).[78] After immunisation of mice with different milk types: raw, skimmed, pasteurized and pasteurized plus homogenized, the antibody production of all specificities within the group immunized with pasteurized/homogenized milk was higher than the group immunized with raw or skimmed milk.[79] After immunisation of rats with pure LG, native or heat-denaturated, native LG caused a more intense immune reaction in terms of IgE production compared to denaturated LG, but heat-denaturated LG caused a greater increase in mucosa inflammatory cells in the gastro-intestinal tract, resembling a delayed type hypersensitivity reaction instead of a Type I allergic reaction as measured by total IgE levels.[80] The immunity response in sensitised mice depends on the heating temperature. Sterilised milk proteins loose their allergenicity compared to raw milk and pasteurised milk.[81]

Neerven et al.[82] discussed the question, which underlying elements of raw cow's milk consumption contribute to the protection against allergies. They argued that a chain of heat-induced changes leads to an unbalanced immune response. Denaturation changes the functionality of several milk proteins, the microbiota-composition is changed and therefore the gut barrier functions are changed. Serum from food allergic people was used[83] to determine the antigen-production *in-vitro* in reaction on raw/crude- versus processed/cooked food ingredients. Additionally to the immediate IgE reaction, measurements were made for a delayed immune reaction based on IgA, IgM and IgG antibody production against modified food antigens. Allergy sensitive people showed severe immune reactions on the cooked food via non IgE production.

5. STUDIES ON HEALTH, GROWTH AND REPRODUCTION

5.1. Animal Studies

Several animal studies have been done with raw and heat-treated milk. In the studies, attention was paid to the generation intervals and the reproductive effects. In studies, where milk was the main food of the animals without any compensational effects of vitamins, there are negative effects found associated with heating the milk. In rats, the five-week development of pups was compared, starting at two weeks of age.[84] The diet consisted of bread and milk from different treatments: raw, sterilised at 96 °C and sterilised full cream milk. The last diet was associated with the highest weight gain, whereas the slope of weight gain was better in the raw milk group compared to the sterilised milk group. In all groups, all furs were fine and all females became pregnant at the end of the trial. Although no differences were found in growth between raw and heated milk, the reproduction of the pasteurized milk group was reduced to 51% compared to the raw milk group.[85] Animals receiving sterilized milk could not produce any offspring after the 3rd generation, whereas raw milk animals could reproduce even after seven generations. In a critique,[86] it was mentioned that the heated milk could not be compared with a normal pasteurized milk, because the milk was sterilized twice for 30 minutes each time. In a follow-up study only some of these findings could be repeated.[87] Growth of rats on sterilized milk was about 10% less after one year. The reproduction in terms of number of pregnant females plus number of born pups was higher after raw milk, although in both groups, none of the pups survived after the fourth day of life. In guinea pigs, full recovery of scurvy was found in the group consuming heat-treated, dried milk powder after changing to raw milk.[88] In a comparison of three generations, cats were fed with a diet of milk, meat and cod liver oil. One group received raw milk, another pasteurised milk and the third one condensed milk. It was reported that cats on a raw milk diet showed normal growth and reproduction as well as a normal bone structure, whereas cats with a diet of pasteurized milk could not successfully reproduce, the animals showed an aggressive behaviour and their bone structure was changed.[89] In a trial with 9 consecutive generations of rats, diets were composed based on freeze-dried milk powder from different origins, but all from the same batch of milk: raw, low pasteurisation (72 °C, 15 sec), high pasteurisation (92 °C, 20 sec) and UHT milk.[90] Milk was the main part of the diet (66%) and all diets were balanced with a vitamin-mixture. In the average values for protein and fat in the diets, no differences were present. There were several highly significant differences between raw and pasteurised milk in growth, fertility, litter yield and internal organs. The animal weight was always higher after raw milk, values up to 10% difference were found. The fertility over 9 generations was very high in all treatments. Especially in the later generations (8th and 9th), there were more pups born and more of them survived by the end of the suckling phase after raw milk intake. From the 4th generation onwards, the total weight of all pups born was highest after raw milk. The internal organs were also different. Spleen and adrenals were systematically heavier in raw milk fed animals, although not in relative relation to their total (heavier) body weight. In the blood serum of males from the 6th generation, significant differences were found for chloride, urea, glutamate pyruvate transaminase (GPT) and triglycerides, which were all lower after raw milk.

In guinea pigs fed with heated skim milk, animals developed so-called wrist and muscular stiffness (known as the Wulzen syndrome) in contrast to those fed raw milk.[91] Calcium depletion had taken place in the wrong places, causing calcification of soft tissues but also an increase in blood pressure due to calcification of the arteries. However, stiffness could be cured with raw cream among others. Chemical analysis showed that heat labile sterols (stigmasterol) were responsible for this deficiency. Also, fresh green feed (e.g., kale) could prevent stiffness. Stigmasterol interacts with hormone functioning (progesteron) and is a precursor in the building of Vitamin D3.[92] The mechanism of the 'anti-stiffness-factor' is through fat-soluble vitamins regulating the calcium and phosphorus metabolism.[93]

The interesting element of the rat study[90] is the long run over 9 generations and the compensation of the feeding ratio with vitamins, etc. The rats received a balanced diet especially in terms of important vitamins. Nevertheless, differences still remain due to the heating of the milk. In earlier studies, like for instance the cat study[89] or earlier animal studies in guinea pigs,[91] no balanced or compensational feeding ratio was offered, and the heat-treated diets were deficient in some essential nutrients. Such studies offer another view on milk heating and are important to show some baseline changes through diets. Modern human diets are much more in balance and we are eating other things than only milk and milk products. Therefore, the effects of heating on health in modern diets may not become visible immediately.

5.2. Human Studies

Following the heating of mother's milk, there is a loss of the protective effect of the milk. The clinical impact of loss of immunomodular properties through milk heating was shown in a randomised trial where endangered human neonates showed higher infection rates after pasteurisation of human milk.[94] Pasteurisation of mother's milk in preterm infants resulted in reduced fat absorption and reduced bone growth[95] or a lower gain.[96] Lower weight gain was also measured in preterm children fed pasteurised pooled human milk as well as lower serum alkaline phosphatase, but higher phosphorus blood values. Serum calcium and 25-hydroxyvitamin D (25-OHD) concentrations were similar in the two groups.[97] Fat absorption was reduced, probably due to the inactivation of milk lipase after heating and the still inadequate production of the child's own lipase.[98]

Apart from the allergenic effect in relation to milk heating and further milk processing, there are also differences found in growth rates of children as part of a nutritional evaluation of milk, processed and unprocessed.[99] Results based on these early trials should not be taken as being too significant. The trials were not randomized and could be biased to the selection of, for instance, schoolteachers.[86] In the statistical recalculation of the trials among school children in the 1920s,[99] it was shown that the increase in length and weight was smaller after consumption of pasteurized milk compared to raw milk.[100] Effects were stronger among the boys than for the girls. Reduction of weight increase due to the consumption of pasteurized milk was 66.0 and 91.1% respectively, whereas reduction of length increase was even more striking (49.8 and 70.1% respectively). Therefore, the authors conclude that consumption of pasteurized milk had only half the value of raw milk in the case of the boys and 70% in the case of the girls. Raw milk appeared to have a greater effect, raising the possibility that at least part of the 'milk effect' could be caused by substances other than fat, protein or sugar (which do not differ between the two milk types).[101] A larger weight gain was found in babies on raw milk without additional supplementation of cod liver oil and orange juice than babies on pasteurised milk alone or pasteurised milk supplemented with cod liver oil and orange juice.[102] Calcium from raw milk is better available and the retention of Calcium was more efficient in children and adults after drinking of raw milk compared to pasteurised or dried milk.[103]

6. RAW MILK RISKS

There are several risks associated with the consumption of raw milk. First, there is the risk of the transfer of antibiotic resistant (ABR) genes through all kinds of innocent microbes in raw milk. Secondly, and already well described and warned about, is the risk of contamination by pathogenic bacteria.

6.1. Raw Milk and the Distribution of AB Resistance Genes

A rapid increase in multi-drug resistant pathogens has occurred in the last decades and some of them (f.i., *S. aureus, Enterococci*) are showing resistance against most known antimicrobial agents. This implies a serious risk to public health[104] and leads to increased problems for bacterial infected humans in hospitals. The driving factor behind the emerging drug resistance is the extensive use of antibiotics in animal husbandry as growth promoters in pigs, poultry and veal. The problem is that any use of antibiotics leads to a higher risk of resistance selection among pathogens. In practice, a sub-therapeutic level of antibiotics administered may especially select for further resistance.[105] Measurements in the stool of infants showed the presence of ABR genes and, already in infants younger than two weeks, high numbers of such genes were detected.[104]

Raw milk is not the only vehicle for antibiotic resistant bacteria. Fermented raw foods can often harbour high concentrations of resistant Enterococci.[106] In so-called 'ready-to-eat' products like raw milk cheese and quark, genes resistant to antibiotics can be increased in number and transferred. Killing of bacteria though pasteurisation reduced the prevalence of ABR genes. Raw milk cheeses had a higher prevalence than pasteurised cheeses (75% versus 37%).[107] Similar results were found for 'erythromycin'-resistance (raw vs. pasteurised 55% versus 23%), although the overall levels of contamination were lower than for tetracyclin. Also, in starter cultures used for fermented milk products or in probiotic cultures, antibiotic resistance has been detected.[107] The genus Enterococcus, as a typical part of the intestinal flora, has an exceptional ability to acquire and transmit ABR genes and is considered to be a major player in the dissemination of ABR genes worldwide.[106] Gene transfer can take place vertically, within the bacterial species, but also horizontally, between different bacterial species, like *S. aureus*, *Lactococcus spec.* and *Listeria spec.*[108] Horizontal gene transfer of ABR determinants, mainly via mobile genetic elements such as plasmids and transposons, contributes to a large extent to the increasing prevalence of bacteria resistant to a single or to multiple antibiotics.[104] Typical bacterial species that cause mastitis in dairy cows have few, if any, mechanisms for transfer of antibiotic resistance to other bacteria, as occurs with intestinal bacteria.[105] Horizontal ABR gene transfer from *Enterococcus faecalis* to *Listeria monocytogenes* and to commensal bacteria can occur in the presence of competing faecal microbiota in a colonic fermentation.[104]

After testing 12 main antibiotics available for udder pathogens, no differences could be found in resistance frequency between organic and conventional Swiss cows, although there should be a strong restriction in the use of antibiotics at organic farms. There was a tendency for organic cows to show higher susceptibility for drug resistance than cows from integrated production systems, especially in isolates from *Streptococcus uberis*.[105] In Swiss cheeses, resistance from Enterococci to tetracyline were as high in organic as in conventional samples (60–61%).[107] No differences were found in penicillin resistance between organic and conventional cows.[109] Also a larger proportion of isolates within Staphylococcus were mentioned[110] from organic rather than conventional farms that were susceptible to erythromycin, pirlimycin and tetracycline. In contrast, in a review article, evaluating 17 different studies, a lower prevalence of antibiotic resistance on organic farms was found,[111] although only in U.S. studies, not in European studies. One of the suggestions for this difference might be the geographic differences in the use of antimicrobial drugs, which is not allowed at US organic farms.

6.2. Raw Milk, Safety and Zoonotic Bacteria

With raw milk consumption a list of potential zoonotic dangers exist. The most commonly reported risks in connection with raw milk are Listeria, Salmonella, Campylobacter and EHEC/STEC. Outbreaks between 1990 and 2006 due to milk consumption in the U.S. according to the official CDC (Centers for Disease Control and Prevention) data were evaluated.[112] The majority of outbreaks attributed to the consumption of unpasteurized milk were caused by Campylobacter and occurred in private homes and resulted in relatively few illnesses. Although listeriosis is a serious foodborne illness, there are no raw milk outbreaks attributed to *Listeria monocytogenes* in the numerous listing in the US.

Here we will especially discuss the virulent sub-strains of *E. coli*. Enterohemorraghic *Escherichia coli* (EHEC) includes a phenotypically diverse population of Shiga toxin producing *E. coli* that causes foodborne and waterborne diseases.[113] EHEC is a subgroup of STEC or VTEC: Shiga toxin encoding *E. coli* or Verotoxin building *E. coli* bacteria. To become virulent, different genetic characteristics must be present in the infecting strain, one of them being the production of Shiga toxin (coding for Stx1 or Stx2 with variants), it is also necessary that bacteria attach to the intestine cells of the host (encoding for intimin eae)[114, 115] and enter these cells. EHEC is one of the most problematic zoonotic diseases, because of its low infective dose. The incubation of the EHEC in humans is between 2–5 days. Sero-groups O26, O103, O111, O145 and O157 belong to the major EHEC strains.[116] The recent outbreak in the Hamburg area (Niedersachsen State, Germany) in June 2011 showed how risky new strains of EHEC are. Fifty-three people died and over 4,300 people had diarrhoea. In this outbreak it had nothing to do with raw milk, but with uncooked vegetables and sprouts for salads. New high-risk pathovars with other aggregation mechanisms[115] can develop rapidly (in this case, STEC O104:H4), and it seemed that human beings, rather than animals, were the reservoirs of this new, antibiotic-resistant strain that acquired the Stx2-gene. Outbreaks of *E. coli* O157 vary dramatically in the severity of illness and the frequency of the most serious complication, haemolytic uremic syndrome (HUS).[113] Looking at the disease pattern in humans, there is a difference in virulence among different outbreaks. Sometimes large numbers of people only experience abdominal pain and diarrhoea while in other outbreaks only a limited number of people are involved, but with severe complaints like bloody diarrhoea, kidney failure, hospitalisation and even death. Although proven raw milk cases are present, raw milk dangers seem to be less in comparison to infectious dangers from human to human contact, animal contact, dirty toilets, undercooked ground beef and unhygienic environments (such as sandboxes for children). A cross-contamination through toilets from bearers is a possible vector for the spread of EHEC.

REVIEW

Currently, we have been dealing in most cases with O157 strains in relation to raw milk outbreaks, although most of the clinical isolates from people with reported illnesses in Europe belong to the non-O157 groups.[116, 117] Reported illnesses in children are mainly associated with the O157 group.[118] *E. coli* O157:H7 does not cause illness in cattle, but colonization in bovines and sheep are the major nonhuman reservoirs for this organism.[119] Risk factors were age specific. In children under 3 years of age, having touched a ruminant had the highest odds of disease, and raw milk was the only food identified as a risk factor. The prevalence of STEC or EHEC positive raw milk samples is different between countries. In 2010 17, 6 and 2, 3% of German raw milk and raw milk cheese were STEC positive respectively.[120] In French raw milk samples, 21.0% and 5.7% were positive for STEC and EHEC respectively, whereas Swiss data showed only 2.5% STEC positive samples. Not only are the prevalences different in different countries, the predominant virulent subtypes of *E. coli* differ. Dangers are not only present in fresh milk; STEC can still survive in fresh cheeses up to two months of age.[121] This was also found for lactic cheeses.[122] Even if the bacteria are killed, some toxins produced by the bacteria still might be active in milk.

Due to genomic research, it is now possible for further specification of STEC O157, showing genomic variations in Shiga toxin production, Stx1 and Stx2 being the main variants. Different virulence of the Stx-gene alleles (f.i., Stx1, Stx2, Stx2c) and their combination might be the reason for this.[113, 123] In The Netherlands, it was suggested that only a fraction of STEC O157 strains in the bovine reservoir are associated with causing disease in humans.[123] The human isolates showed a larger variation in Stx-genotypes than the bovine samples. All human isolates were characterised by the presence of the *tir* gene, responsible for the adherence of the bacteria in the intestine. In southern Sweden mainly one type of STEC O157 (presence of Stx2 and Stx2c) was involved that was highly pathogenic in human.[124] This strain was correlated with two larger foodborne outbreaks. Further genetic differentiation, based on genotyping of STEC O157 strains, is necessary. The presence of the Stx2, eae, and katP genes, together with a combination of several Stx2 variants, was clearly associated with human-virulent strains. In contrast, dairy food STEC strains were characterized by a predominance of Stx1, with a minority of isolates harbouring eae, espP, and/or katP.[125] The current molecular assays used in public health laboratories might be adequate for identification of STEC O157:H7. However, these laboratories lack the further discriminating power needed to resolve its genetically homogenous population structure. So, before dairy food is made suspect for STEC O157 a further determination of all genetic aspects is necessary (which pathogenic Stx strain and whether additional adhesion and colonisation features (eae) present?).[126]

Transmission routes of *E. coli* vary depending on hygiene measures on the farm in the production chain and killing through heating. Studies are inconsistent, however, the types of feed and feeding patterns affect the prevalence of STEC O157:H7 in cows. High levels of concentrate rather than a diet of roughage in beef animals showed increased numbers of the bacteria.[127] The same was found for dried distiller grains.[128] Even in newborn calves STEC can be found within the first 24 hours of their life and, in the phase of young stock, bacteria are spread through the herd.[129] Infected calves kept on pasture were less exposed or even had cleaned themselves compared to calves kept indoors.[130] Calves fed milk replacers showed lower incidences of colonization than those fed with colostrum and real milk.

Shedding from cows shows a complex dynamic. So-called super-shedders can be responsible for increased transmission and persistent colonization rates in the herd.[131, 132] There are seasonal shedding patterns. In a review, consistent seasonal patterns across regional boundaries were shown. Higher temperatures during summer could increase the growth rate of the bacteria. For the Nordic hemisphere, STEC findings had a summer peak from July to September. Mechanisms behind this pattern could be the population occurrence of the bacteria as well as the behaviour of the infected human beings who are outside more and have more contact with animals during summer or eating faeces-contaminated food (salad).[133] Besides the primary infection, there is also a risk of secondary and tertiary transmission from person-to-person,[134] because of the very high concentration of the virulent bacteria in the stools of infected persons.

Risk reduction of STEC concentration in raw milk and its products can be achieved by aggressive hygienic practices and periodic monitoring of the milk. The on-farm management and hygiene measures that reduce the number of environmental bacteria were investigated[135] and five management practices were mentioned to reduce the bacterial pressure: Dry and clean bedding, stable rearing groups, regular emptying and cleaning of water troughs, avoiding contact between different animal herds and maintaining a closed herd policy. Persistent STEC shedders should be identified and removed from the herd. Growth of STEC can be reduced if milk is sufficiently and quickly been cooled, after rapid acidification of the curd, or during long ripening of hard cheese or when during the cheese process, the curd has been heated at 54 °C during 30 minutes.[136] In the bacteriological evaluation of commercial cheeses, it was concluded that the application and maintenance of good hygiene practices throughout the food chain is the key to prevent contamination and bacterial growth.[137] This was confirmed in 41 commercial US raw milk cheeses after 60-day of ripening and where no zoonotic bacteria could be detected.[138] A periodic monitoring of coliform levels in raw milk could be a tool to control the hygiene praxis on the farm. The outbreak described was preceded by three repeated milk samples with elevated levels of coliform bacteria (>50 cfu ml^{-1}). Slow rises of coliform-values over time and single spikes should be taken as a signal to examine the hygiene during milking as well as the equipment cleaning technique.[134]

The answer to EHEC contamination by official bodies is to increase hygienic practices (e.g., washing hands), the heating of food (milk included), the avoidance of eating salads, sprouts, etc., and avoidance of contact with farm and zoo animals. Also, water can be a source of infection as people and water can bear EHEC; therefore, risk sources are being re-evaluated. Additionally, alternative non-thermal processing treatments will be investigated (high pressure, pulsed-electric field, ionizing radiation, UV radiation, and ultrasound) to inactivate STEC with minimal alteration to sensory and nutrient characteristics. However, it has yet to be determined if and to what level the native character of food will be changed with these treatments.

7. DEALING WITH RISKS

The raw milk issue has been controversially and emotionally discussed, and microbiologists and health officials repeatedly warn of the danger of raw milk consumption. Researchers dealing with the epidemiology of raw milk research often insert a risk warning at the end of their scientific paper (see, e.g., the review paper of Ref. [9]). After his conclusion that farm children who drank raw rather than cooked milk were better protected from asthma and atopy, Loss et al.[10] for instance, said: *"however, on the basis of current knowledge, raw milk consumption cannot be recommended because it might contain pathogens. Once the mechanisms underlying the protective farm milk effect are better understood, ways of processing and preserving a safe and preventive milk can be developed."* Coenen,[139] investigating the hygienic quality and presence of zoonotic bacteria in certified German raw milk (Vorzugsmilch) and showing the much better quality of this milk type, said: *"A health risk after consumption of Vorzugsmilch is basically possible, because the presence of zoonotic germs in low doses as EHEC and Campylobacter jejuni never can be excluded."* There is a tendency to look at technical rather than ecological solutions to control risks and to find the single key factor within the protection. In his presentation at the first raw milk symposium, Kneifel[140] said: *"The challenge is to maintain the native status of the product and to get rid of its shady side. Therefore, we need high tech solutions."*

Several issues seem to be relevant in the discussion 'health or hazard': Freedom of food and nutrition choice[141] against general protection of the population; finding acceptable ways to reduce the risks in the production chain rather than a plea for (non-existing) zero-risks and a well-balanced discussion about the pros and cons of raw milk consumption. This means finding ways to differentiate milk qualities in terms of hygienic attention at milking and processing rather than speaking generally about the dangers of *any* raw milk (*a* milk vs. *the* milk). Additionally, Vuitton[142] remarks that *"we all strive to zero-risk, however an immediate fight to zero risk may mean immediate threat to sustainable agriculture and the emergence of other types of more insidious risks both for health and for the environment."* In the discussion about the risks of raw, expressed breast milk fed to preterm infants, raw milk is clearly preferred above pasteurised mother's milk. One hundred percent safe raw human milk does not exist, but clear correlations fail between bacterial intake and infant disease, and microbiological testing of expressed breast milk identifies bacterial growth in >75% of samples, with 7%–36% of all samples containing pathogens.[143] The screening of raw expressed mother's milk delivered for premature infants showed that these infants with an undeveloped immune system were frequently exposed to large numbers of milk-associated coagulase-negative Staphylococci, whereas the exposure to *S. aureus* and gram-negative aerobic bacteria was less frequent.[144] Nevertheless, the authors expressed their surprise that they were unable to document *any* adverse events that could be directly related to ingestion of bacteria in raw breast-milk. Since the benefits of using a mother's own milk for preterm infants out-weigh any potential disadvantages they started an education program to improve the bacteriological quality of the delivered milk instead of reducing the pathogenic risks by pasteurisation. To improve the safety of expressed milk, a HACCP system (control system of hazard-reducing control points) was developed in which the contamination and multiplication of hazardous microorganisms could be reduced.[143]

Besides a technological approach to find solutions, it is necessary to look for an ecological approach in which the environmental prerequisites and modes of transmission of contamination become clear. What are the ways and measures to improve the immune system of young children so that they can handle the load of infectious bacteria that will always be present in the environment? At the farm level, it is necessary to understand why some animals can become resistant to strains of EHEC without becoming EHEC shedders. Rather than focussing on the presence of EHEC in individual animals, it makes sense to look at the absence as well and on environmental factors that inhibit their growth. In expressed human milk, not EHEC, but *S. aureus*, presents one of the biggest health issues. In a study on the role of commensals in breast milk, several bacterial strains were found that were very effective in the suppression of the growth of *S. aureus*. It was stated, that *"the published studies of breast milk microbiota are focused on pathogenic bacteria as possible sources of infection. In contrast, the species diversity and the importance of the normal bacterial flora have received little attention so* *far,*" a statement, which seems also to be true for cow's raw milk. The highest inhibition came from *Lactococcus lactis*, a strain producing a bacteriocin, nisin.[145]

Another relevant issue is to evaluate successful farms, those that are able to deliver raw milk over a long period of time without any evidence of zoonotic illness among its customers. To learn from these farms can be an important tool for best practices in relation to hygienic management. Last but not least, rather than focusing only on health risks there should be a balance between benefits and risks. It therefore makes sense not only to investigate case studies from patients with zoonotic diseases but also to look at individual cases described by farmers of patients who say that they can only consume raw milk. This is also stated by Vuitton,[142] who calls for a more balanced judgement of raw milk. He gives an example from Norway, where raw milk can be sold legally if packaging indicates that *"the product is associated with a marked increased risk of transmission of severe diseases."* The author[142] suggests this warning be completed with *"but the product is associated with a significant protection against other types of severe diseases"* and concludes *"that this would be scientifically correct!"* A differentiation of milk qualities, absent in the discussion about milk dangers, is necessary.[146] Raw milk intended for consumption fresh or for raw milk processing in cheese, is a distinctly different product than collected bulk milk meant for pasteurisation. Our modern opinion of milk should be connected with the reductionist views on food quality:[146] *"we do not have the multiple terms that the Inuit have for snow, because, until the last fifty years or so, milk was surprisingly undifferentiated, relative at least to our modern retail cornucopia."* We should add to this that an ecological-environmental focus as well as a focus on case-orientation could open the ways of understanding why raw milk supports human health. Another research focus is needed, one that is more connected with words like health support and health promotion.

In the scientific discussion, single cases, are always discounted as anecdotal. But for some reason, individual cases of diseases are taken more seriously than individual cases of healing, which is often seen as a coincidence. There are accepted scientific methods to evaluate the cause and effect within single cases[147] and methods and prerequisites are analysed dealing with complex situations of healing. A better scientific evaluation of single cases, both negative and positive, would be helpful to balance the present polemic between advocates on both sides of the borderline. A significant number of people benefit from milk only after they find that although they are unable to drink commercial milk for a number of reasons, they are able to enjoy drinking milk that is raw.

7.1. Reducing Risks by Adopted Regulations

The possibility of consuming raw milk is very different in Western countries. The control of raw milk delivery varies between countries even in the 1900s. In the UK, grades of milk were introduced in 1917 as a possible answer to the high prevalence of tuberculosis in UK herds; however, the adoption of the system was too slow and therefore pasteurisation was introduced gradually by dairy companies in the inter-war period.[148] In Germany since 1931, there is the possibility of selling a legally controlled raw shop milk, called Vorzugsmilch. This milk is made available for direct consumption as raw milk, and not produced from the background of any further standardisation or heating process within a creamery. In the monthly veterinarian control of bulk tank milk samples a differentiation is made between process indicators looking after hygiene and bacteria in connection with zoonotic diseases. There is a zero-tolerance for the presence of pathogens like Campylobacter, Salmonella, EHEC, Brucellosis, Tuberculosis, whereas process indicators are used to control and improve the milking and cleaning practice. In Italy and Slovenia, raw milk vending machines are upcoming ways to sell raw milk and in most European countries raw milk can be bought in small amounts directly from a farm if a warning label is applied to say that the milk should be heat treated (>70 °C) before consumption, storing temperatures should be below 5 °C and consumption should be within three days. Testing milk from vending machines in Italy, not a single pathogen in any of the samples was found.[149] Thirty-three farms served 60 machines and in total 3,800 litres of milk per day were sold. In previous studies, however, in their screening, one *E. coli* O157:H7, one *C. jejuni* and one *S. typhimurium* were detected from three out of 99 raw milk samples collected from vending machines in the same area. Several studies have shown that the so-called 'Vorzugsmilch' or 'certified grade-A raw milk' in Germany has a much higher hygienic and zoonotic standard than 'normal' raw milk sent to creameries and to be pasteurised. These studies showed that, due to the attention to hygienic handling and processing of raw milk, in combination with a strict control system of raw milk for sale, there is already a huge reduction of potential health risk. Total bacterial counts and zoonotic bacteria are low or even absent in 'Vorzugsmilch' in comparison to 'normal' raw milk. Recently, in Vorzugsmilch, the maximum levels for coliform bacteria have been lowered as an answer to the risk of EHEC. Risks of Campylobacter, Listeria and Salmonella from 'Vorzugsmilch' can be negligible.[120, 150–152]

In an editorial comment Jay-Russell[153] plies for additional raw milk regulations such as pathogen testing, sanitation standards, and warning labels. On the other hand she signalise also a need for more research into best management practices for raw dairy production at farms with a long history of producing safe milk without any contamination. The five main testing parameters suggested[154] include somatic cell count (SCC), plate counts, preliminary incubation, coliform counts, and specific pathogen testing. One of the most important recommendations for the sale of raw milk at farms is the strict control on cow health (udder), zoonosis and process hygiene at the time of milking. The certified German farmers who produce Vorzugsmilch are under a strict HACCP system and strict microbiological standards. In Certified Grade A milk as Vorzugsmilch lower overall bacterial counts, somatic cell counts and the absence of several zoonotic bacteria were found in comparison to general raw milk (Table I). The German Federal Institute for Risk Assessment controls the zoonotic pressure in food.[150–152] In the German zoonotic data, raw milk and raw Vorzugsmilch is presented separately and the data from 2008 to 2010 show the same findings as other studies (Table II). In general, hardly any positive samples for zoonotic bacteria were found and the indicators for a hygienic process quality of Vorzugsmilch is in most cases much better than general raw milk samples.

The routine control of the German Association of producers of Vorzugsmilch (BDVM) reported that in the last 15 years milk was sampled for EHEC: in the examination of about 100 milk samples per year from the Vorzugsmilch deliverers up to the present time only one single test for EHEC was positive. It shows that there is hardly any potential risk within this type of milk.[155]

These reports suggest that the presence of pre- and post-harvest control measures to effectively reduce contamination might be related to the reduction in the pathogens in raw milk, suggested as one of the measures to reduce the zoonotic danger,[156] although a 100% risk reduction is illusive. If we make the comparison with the development of a HACCP for expressed mother's milk in relation to the risk assessment for preterm children, Cossey et al.[143] should be quoted, when they said: *"Those who believe that the number or type of bacterial contaminants ingested with milk can be assumed to affect the incidence and severity of illness, especially in preterm infants, should not attempt to define safe limits, but rather should consider **not** feeding raw expressed milk to high-risk infants."* This might also be true for raw cow's milk.

Table I. Microbiology and somatic cell counts of general raw milk compared to certified Grade-A milk (Vorzugsmilch) in Germany.[139]

	Raw milk	Vorzugsmilch
Farms sampled (N)	115	35
Milk samples (N)	149	74
Somatic cell count (× 1,000) ml^{-1}	190	160
Total bacterial counts (× 1,000) ml^{-1}	49.0	8.7
Escherichia coli ml^{-1}	1.1	0.08
Coliform bacteria ml^{-1}	110	10
STEC ml^{-1}	0.7	0.0
Salmonella ml^{-1}	0.0	0.0
Campylobacter ml^{-1}	0.0	0.0
Listeria ml^{-1}	10.1	16.2[a]
Bacillus cereus ml^{-1}	8.1	0.0
Staphylococcus aureus ml^{-1}	0.02	0.08

STEC = shiga-toxin producing subtypes of *E. coli*.
[a]All positive samples were found at one single farm where a range of samples was taken.

Table II. The percentage of positive samples for several zoonotic bacteria of general raw milk (raw) and Vorzugsmilch (VZ) samples in Germany (2008–2010); (derived from Refs. [150–152]).

	Year					
	2008		2009		2010	
Raw milk type	Raw	VZ	Raw	VZ	Raw	VZ
Samples (N)	ca.100	ca.150	ca.300	ca.175	ca.300	30
STEC ml^{-1}	4.9	1.8	1.5	0.0	1.4	0.0
MRSA ml^{-1}	nd	nd	4.1	nd	4.7	10.0
Salmonella ml^{-1}	0.0	0.0	0.0	0.0	0.0	0.0
Campylobacter ml^{-1}	0.8	1.3	0.8	0.0	1.9	0.0
Listeria ml^{-1}	0.0	0.5	1.2	1.8	4.6	0.0
Yiersinia ml^{-1}	4.8	1.6	9.1	1.4	9.3	3.5

STEC = shiga-toxin producing subtypes of *E. coli*; MRSA = methicillin-resistant *Staphylococcus aureus*; nd = not determined.

8. CONCLUSION: RAW MILK, HEALTH OR HAZARD

—Pasteurisation of milk negatively affects the digestibility and the protection against asthma and atopy. Immunological properties present in raw milk are lost or changed through milk heating. A range of epidemiological studies pinpointed raw farm milk as the protective agent, both in farm and in non-farm children. The protective elements within raw milk are only beginning to be understood and may be complex.

—Medical doctors and official governmental bodies do not promote the consumption of general raw milk because of its reputation as potential vehicles of zoonotic illness, especially for those groups who might profit most from raw milk intake: Young children and pregnant women. With regard to food safety it is necessary to discriminate between different types of milk. Raw milk is far more than just milk that has not been pasteurized. To improve the safety of raw milk, which can be delivered for direct consumption, increased hygienic standards during the processing and delivery of raw milk is helpful, like in the German Vorzugsmilch.

—It is impossible to reach 100% zoonotic safety in raw milk delivery. The pressure to reach 100% safety, on the other hand, can frustrate the development of a raw milk market. It makes sense to evaluate and understand examples of farms, where long-term relationships are present between consumers and raw milk suppliers and there have not been any incidence of disease. The immune system

REVIEW

and health status of raw milk consumers should be investigated as well.
—Rather than working to repress zoonotic bacteria we should be investigating their ecology to understand the circumstances under which they become a danger to human beings.
—A case-to-case approach is a good scientific tool to evaluate positive and negative effects from raw milk intake within a small or limited number of consumers or patients. Case control studies on raw milk consumption are necessary to evaluate the impact on health.
—It is becoming necessary to use molecular techniques to discriminate between STEC strains. Not every STEC means an endangered milk sample. Even though we know less about the other gastrointestinal infectious agents it is likely that the same is true for them.

Conflict of Interest

There is no conflict of interest.

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