

Paper	Brief Synopsis	Findings	Conclusions
Mariani, C. et al. 2007. Biofilm Ecology of Wooden Shelves Used in Ripening the French Raw Milk Smear Cheese Reblochon de Savoie. <i>J. Dairy Sci.</i> 90:1653-1661.	A study of microbial biofilms on wooden boards at the end of cheese ripening, including identification and enumeration of different groups of microbes and measurements of the physiochemical conditions (aw, pH, salt concentration) at the surface of the boards. Boards from 8 different facilities, and of three different ages (young through old boards) were examined at two points in the year (summer and autumn).	Micrococci-corynebacteria and yeasts and moulds (predominantly <i>Geotrichum</i>) were the dominant bacteria and fungi, respectively.	The microbes that constitute the biofilm on the board surface at the end of ripening are strongly correlated to the cheese surface microorganisms.
		Organisms found at much lower levels across all boards included <i>Leuconostocs</i> , heterofermentative lactobacilli, and enterococci.	Enterococci and staphylococci are considered 'ambivalent' microorganisms, with potential to be advantageous or undesirable. The strain(s) of staphylococcus were not identified in this study, but regardless, they were present at well below the level of concern for potential toxin production.
		Staphylococci and pseudomonads were found on 80 of 100 samples, at low levels similar to the <i>Leuconostocs</i> etc.	<i>Pseudomonas</i> is considered a spoilage microorganism, but was also detected at low levels.
		The shelves had statistically similar microbial counts and physiochemical properties regardless of their age (ranging from 6 months to 14 years) or the facility they came from.	The study only looked at boards after use and before cleaning, not clean shelves ready for use.
		Seasonal variability in the counts of staphylococci and pseudomonads (both lower in Summer than in Autumn) was observed; the levels of the other organisms did not show seasonal variation.	The authors concluded that the strict ripening and cleaning procedures used by the cheesemakers led to the predominance of desirable microflora, while maintaining undesirable microorganisms at a low level.
Mariani, C. et al. 2011. Inhibition of <i>Listeria monocytogenes</i> by resident biofilms present on wooden shelves used for cheese ripening. <i>Food Control.</i> 22:1357-1362.	This study examined the influence of the microbial biofilms on wooden boards used for ripening Reblochon on the growth of two strains of <i>Listeria monocytogenes</i> . Boards were tested directly after use, after cleaning in a brushing machine with cold water only, and after sterilisation by heat treatment. Boards from four different maturers were tested at two different time points (1 and 12 days post-inoculation with <i>Listeria</i>).	Scanning electron microscopy of the boards showed that cleaning with cold water and scrubbing significantly reduced the total population of yeasts and bacteria on the shelves. However, the biofilm persisted even after cleaning and drying.	The inhibition of <i>Listeria</i> by wooden board biofilms could be due to various factors, including production of inhibitory molecules and various forms of acid, and/or to nutrient competition (the 'Jameson effect').
		The boards' pH was not affected by washing or heat-sterilisation. The aw of the boards was reduced by washing-drying and by heat treatment, but this difference did not persist past the inoculation step.	The reproducibility of the anti- <i>Listeria</i> effect across boards from different ripening facilities lends support to the hypothesis that complex biofilms play a significant role in avoiding contamination of cheese.
		The presence of active biofilms (before and after cleaning) significantly reduced the numbers of both strains of <i>Listeria monocytogenes</i> by Day 12. This effect was observed across boards from all farms.	Sterilisation of the wooden boards through ionisation (as opposed to heat treatment) had a similar effect, allowing <i>Listeria</i> to multiply to high levels on the boards.
		In contrast, there was a 'marked multiplication of pathogenic cells' on the autoclaved boards.	For some strains, the cleaned and dried boards had a greater anti- <i>Listeria</i> activity than the boards tested directly after use. This is noteworthy as bringing new cheeses onto the clean shelves of the ripening room is a critical point when contamination might occur.
		The two strains of <i>Listeria monocytogenes</i> showed varying sensitivities to the active biofilms, particularly on Day 1. However, both grew successfully to high levels on the autoclaved boards by Day 12.	Further research is called for to clarify the mechanism(s) of <i>Listeria</i> inhibition and the conditions that establish and promote beneficial biofilms.
Guillier, L. et al. 2008. Modelling the competitive growth between <i>Listeria monocytogenes</i> and biofilm microflora of smear cheese wooden shelves. <i>Int. J. Food Micro.</i> 128: 51-57	This study used a model cheese system to study the competition between biofilm microflora (BM) cultured from wooden shelves used to ripen smear cheeses and two strains of <i>Listeria monocytogenes</i> . The team searched for inhibitory compounds that might be produced by the BM and plotted the growth of the BM and Lm separately and together.	No compounds with an inhibitory effect on <i>Listeria</i> growth were found to be produced by the biofilm microflora at any stage in their growth.	The authors concluded that the inhibition of <i>Listeria</i> growth was due to non-specific competition for nutrients rather than the production of specific inhibitory compounds.
		Biofilm microflora were found to grow more rapidly than <i>Listeria</i> , both separately and together.	The fact that the BM grew more rapidly than the <i>Listeria</i> meant that they were effective at outcompeting it.
		When <i>Listeria</i> and BM were grown together, exponential growth was observed in both populations until the point when the BM reached the stationary phase. At that point, growth of <i>Listeria</i> stopped.	The authors pointed out that under real-world conditions, contamination with <i>Listeria</i> is likely to take place at very low levels compared to the resident BM on the cleaned boards. Therefore 'the potential for growth of <i>L. monocytogenes</i> on smear cheese wooden shelves is rather limited as long as the biofilm microflora is preserved'.
		When <i>Listeria</i> was inoculated into a community where the BM had already reached a stationary phase, no exponential growth was observed. This is despite the fact that models would suggest that <i>Listeria</i> would grow under the physiochemical conditions present.	This result is in accordance with the findings of Mariani et al. (2011) above. It could also help explain why the inhibition occurred consistently regardless of which cheese facility the boards came from; as long as the BM grow faster than the <i>Listeria</i> , the findings suggest the inhibitory results should be the same.
		This finding was reproduced regardless of the various times of relative inoculation of BM and <i>Listeria</i> strains.	The authors also conclude that other undesirable microorganisms (pathogens or spoilage microbes) could be subject to inhibition by the resident BM by the same principle.

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Zangerl, P. et al. 2010. Survival of <i>Listeria monocytogenes</i> after cleaning and sanitation of wooden shelves used for cheese ripening. <i>Eur. J. Wood Preserv.</i> 68:415-419.	An assessment of the effectiveness of three different cleaning methods on spruce cheese boards inoculated with <i>Listeria monocytogenes</i> . Presence of residual Lm was tested for at both the surface of the board (contact plate) and below the surface (in shavings to a depth of 2mm).	Boards were inoculated with a high level of Lm and levels counted after 1h and 24h of incubation. Levels decreased slightly after 1h and rose again after 24h. Therefore the authors concluded the wood had no intrinsic antibacterial properties.	If boards have become contaminated with <i>Listeria</i> , brush cleaning with alkaline detergent is not sufficient completely to sanitise them.
		After 20-24h incubation, contaminated wooden blocks were soaked for 15m in 50C alkaline detergent, then scrubbed for 30s and rinsed with 50C water. Some blocks were then subjected to heat treatment, either at 80C for 5m or at 65C for 15m.	However, proper heat treatment of the wood will remove even high counts of <i>Listeria</i> and render the boards again fit for use. Options in use at different facilities include prolonged immersion of cleaned shelves in boiling water, pressure-cleaning and kilning, etc.
		These blocks were sampled in two ways: direct contact of the surface to a plate and by taking shavings to a depth of 2mm, mixing the shavings with broth, and enumerating the broth using the ISO method.	The authors conclude that wood can and should be used in contact with foods when it provides material advantages (such as humidity regulation) and risks can be reduced by suitable measures.
		When the alkaline detergent was used without subsequent heat treatment, 4 of the 9 surface contact plates grew Lm. All 9 samples of the 2mm deep shavings showed viable Lm detected.	The authors cite work showing that plastic is 'not as hygienic as often stated' and that once plastic boards are scratched or perforated they also become difficult adequately to clean.
		When the alkaline detergent was used in conjunction with heat treatment (both time/temp combinations), no <i>Listeria</i> was recoverable from the surface or the shavings.	Regardless of the material, all food-contact surfaces 'need to be constantly maintained and monitored for cleanliness'.
Frank, J. et al. 1990. Surface-adherent growth of <i>Listeria monocytogenes</i> is associated with increased resistance to surfactant sanitisers and heat. <i>J. Food Prot.</i> 53(7): 550-554.	This study looked at the effect of the mode of growth of <i>Listeria</i> on its susceptibility to various sanitisers and heat treatments. The team looked at cells growing in nutrient broth (planktonic cells), single cells that had adhered to a glass slide after a short incubation, and adherent microcolonies that had grown on a glass slide for an extended period.	The cells growing suspended in broth were most susceptible to chemical and temperature treatments.	The authors concluded that adhesion of bacteria to surfaces has a protective effect against the effects of chemicals and to some degree heat treatment.
		Single cells that had had only a short opportunity (4 hours) to adhere to the glass slides were significantly more resistant to chemical treatments than the planktonic cells. Their initial numbers fell quickly when they were exposed to chemicals, but they were only eliminated with chemical contact times of 12-16 min (quaternary ammonium or anionic acid sanitiser), or 5 min at 70 degrees C.	They note that any <i>Listeria</i> present in food processing plants may be in different states of growth depending on its environment, temperature, and access to nutrients.
		Adherent microcolonies were not entirely destroyed by either chemical or by heating to 70C for five minutes.	They hypothesise that the adherent growth provides more physical protection against the action of chemicals, which work by disrupting the bacterial cell membrane.
		When chemicals were applied to the adherent microcolonies there was a 2-3 log decrease within 30 seconds 'after which a resistant population remained for at least 20 minutes' of continued chemical exposure.	In order for chemicals to sanitise effectively, even on smooth surfaces such as glass, the initial step of cleaning must physically disrupt the adherent cells.
		Increasing the concentration of either chemical treatment did not have a significant impact on the destruction of the adherent cells.	
Gibson, H. et al. 1999. Effectiveness of cleaning techniques used in the food industry in terms of removal of bacterial biofilms. <i>Journal of Applied Microbiology.</i> 87: 41-49.	A more general paper that looks at the relative merits of disinfectant chemicals, a rotating scrubbing brush, and high pressure spray in the removal of bacterial biofilms from stainless steel surfaces. The trial biofilms were composed of <i>Staphylococcus aureus</i> and <i>Pseudomonas aeruginosa</i> rather than rind microflora.	The factors that impact the efficacy of different methods for removal of biofilms (and sanitation in general) are chemical energy (detergents and sanitisers), mechanical energy (scrubbing), temperature, and time.	It is important to remember that stainless steel is not a panacea when it comes to cleaning and disinfection and bacterial biofilms are likely to be present, particularly when equipment (such as racks and shelves) is not able to be cleaned on a constant basis.
		Biofilm formation on stainless steel can also impact the ability of chemicals to disinfect a surface, as the matrix of the biofilm can partially or completely shield the resident bacteria from their effects.	Detergents are more effective at removing food debris than biofilms, which are capable of resisting them to a greater degree. The amount of biofilm produced depends on the species (single or community) of bacteria present in an environment.
		The less-frequently a surface is cleaned, the more probable the development of a biofilm.	The mechanical scrubber was very effective in the removal of biofilms and associated microorganisms.
		Many factory cleaning methods involving application of detergents without vigorous mechanical action are not efficient at removing biofilms, even on stainless steel.	Likewise, the pressure spray wash (another form of mechanical action) was very effective, but carried with it the risk of aerosolisation and spread of contamination through splashing water droplets over a wide area.
		Mechanical scrubbing and high pressure spray proved significantly more effective than gentle application of detergents with low-pressure rinsing.	Practitioners considering the best methods for cleaning their wooden boards (and their stainless steel surfaces) should remember the importance of mechanical action. Use of detergent and sanitiser alone is not necessarily sufficient for effective cleaning. Equipment and procedures should take this into account and environmental testing should be done at an appropriate frequency to verify absence of pathogens.