

Faecal Contamination of Lettuce Heads after Manure Application

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In recent years, an increasing number of disease outbreaks have been associated with consumption of contaminated vegetables. Thus, it has been speculated to what extent such contamination is associated with application of animal manure as fertilizer, which is particularly practiced in organic vegetable production where conventional fertilizers are prohibited. A field survey was therefore performed aiming to assess the survival and transfer of *E. coli* from animal manure to lettuces, with *E. coli* serving as an indicator of bacterial enteric pathogens.

Animal manure was applied to 3 Danish fields prior to planting of lettuce seedlings, then 5-8 weeks later at the normal time of harvest, inner and outer leaves of 10 lettuce heads were pooled into one sample unit with a total of 50 pools per field. Additionally, in one field, 15 soil samples were collected weekly until the harvest time. *E. coli* was enumerated by plating 1 mL of 10-fold serial dilutions of 5 g of homogenized sample material, i.e. manure, soil and lettuce onto Petrifilm™ Select *E. coli* count plates (3M), which were then incubated 24 h at 44°C.

The manure applied to the fields contained 3.0-4.5 Log₁₀ *E. coli* CFU/g and *E. coli* was found in 36-54% of the pooled lettuce samples with a detection limit of 10 CFU/g. Numbers of *E. coli* in 14-20% of pooled lettuce samples exceeded a satisfactory microbiological hygiene criteria level of 100 CFU/g.

The highest percentage of faecally contaminated lettuce heads (54%) coincided with the shortest growth period studied indicating that the time gap between application of manure and harvest and the survival of *E. coli* (and pathogens) influences the contamination of lettuce via manure amended soil. However, at the time of harvest, the numbers of *E. coli* in 5 of 15 soil samples were reduced below the detection limit and no samples exceeded 100 CFU/g. This is in contrast to the lettuce samples, where 20% of faecally contaminated samples had >100 *E. coli*/g, which may indicate that faeces contamination of crops could originate from alternative sources, such as contaminated water and wildlife. Comparisons of the genotype of isolated *E. coli* strains could help to elucidate this.