# MEASUREMENT OF PATHOGEN TRANSFER IN AEROSOLS FOLLOWING LAND APPLICATION OF MANURE

***P. Hobbs[1], D. Davies[2], J. Williams[1], H. Bakewell[2], K. Smith[3]***
*[1]Institute of Grassland and Environmental Research, North Wyke Research Station, Okehampton, Devon EX20 2SB. phil.hobbs@bbsrc.ac.uk*
*[2]Institute of Grassland and Environmental Research, Plas Gogerddan, Aberystwyth, SY23 3EB, UK*
*[3]ADAS, Woodthorne, Wergs Road, Wolverhampton, WV6 8TQ*

## ABSTRACT

The study of the transfer of pathogens through the air from manure spreading that may cause disease has been brought to the fore by the concern over increasing numbers of illnesses caused by microbial infections relating to food. Sampling procedures and methods of detection and collection affect understanding concerning transmission. The recent environmental history of the pathogen is important, for example an organism that has previously been exposed to drying and rewetting will be more likely to survive aerosol travel and often exhibit increasing pathogenic capacity. Enteric and K12 *Eschericha (E) coli* marker organisms demonstrated that pathogens can be transported over 200 m at a wind speed of 2.3 $m.sec^{-1}$ and over 400m at 4.2 $m.sec^{-1}$. Models of the data show that pathogens can be transported over 1500 m. Experimentation indicates that we may be able to model transportation as an aerosol, but viability of a pathogen makes assessment of disease transmission a risk assessment rather than numerical evaluation. Some early experimental studies that are relevant to the transfer of pathogens (including marker organisms) as aerosols will be used to highlight monitoring difficulties.

## INTRODUCTION

Bioaerosols containing pathogens can travel distances up to 10 km and remain viable (Dowd et al., 2000), however, there is uncertainty as to which pathogens remain viable or pathogenic during aerosol transportation. Mechanisms by which pathogens survive and their capacity to endure climatic variations vary not only between species but within species (personal communication, Prof. T. Humphery). Desiccation is probably the most difficult problem for most microbial species to combat. Aerosols can acquire or lose mass as water depending on the relative humidity of the air as a particle travels on the wind. Larger particles can travel further as they evaporate and bacteria internal to the aerosol are likely to survive longer because they are less exposed to dehydration (Lighthart, 1989). At 'take-off' as either a dry or wet microbe, changes in water content will inevitably follow. Oxygen (and ozone), solar radiation and the effects of pollutants such as $SO_2$ and $NO_2$ can all reduce viability of pathogens travelling as, or within, an aerosol.

Particulates from manure and dirty water are transported on winds and may drift onto edible crops, watercourses, or be inhaled by the local human population and animals (Nicholson et al*.,* 2000). Particles below 10 microns will stay suspended in low wind speeds (Nicholson, 1995) and can enter the mammalian lung (Brunekreef, et al*.,* 1995).

Animal viruses, coliphages and bacteria have been identified in aerosols from wastewater irrigation on farms *(*Brenner et al*.,* 1988). Concern over pathogen bioaerosol transport was highlighted at Walkerton, Ontario where *E. coli* 0157:H7 and other pathogens from biosolids contaminated the local drinking water supply. Liquid manure resulted in viable microbes 228 m downwind using Anderson samplers and 274 m downwind using static impaction plates

(Evenden, 1972). Generally with wind speeds below 2.2 m.sec$^{-1}$, only 10% of the bacteria were recovered 200m from the source. Further wind speeds of between 1.3 to 6.7 m.sec$^{-1}$ were positively associated with distance at which bacteria were collected. Aerobacter (90%) and *E. coli* (10%) were the principal micro-organisms recovered by these techniques.

During the use of a vacuum tanker application with fresh slurry, faecal and fungal flora were abundant but declined to background levels after 200m (wind speed 2.2 m.sec$^{-1}$). Boutin et al., (1988) also showed that the particle profile, 50%> 10-micron, remained stable over 100m for samples of pig slurry from a spraying irrigation gun, with transfer occurring up to 200m. The use of an irrigation or rain gun system for spreading slurry or dirty water will increase the possibilities of pathogen transfer compared with spreading by a trailing shoe or injector system. However, if the slurry is not subject to UV radiation the pathogens may survive longer. Solid manure or FYM spread by a flail spreader has the lowest probability of pathogen transfer by air, however, some biosolids have been shown to emit organisms at high rates after land application and when stored uncovered in the field (Boutin et al., 1988).

In this study we determined the transmission of pathogens via aerosol dispersion downwind of a land spreading source with a range of effluents using specialised sampling equipment. Aerosol particles were collected directly on agar plates (with an appropriate medium according to the target organism) via portable air sampling units.

## MATERIALS AND METHODS

Slurry spreading was arranged upwind of an array of air sampling units in which aerosols impact on an agar plate with appropriate target-selective agar medium using a Burkard air sampler (Burkard Scientific Ltd., PO Box 55, Uxbridge, Middlesex, UB8 2RT, UK.). Cattle slurry was seeded with a culture of the non-pathogenic *E. coli* K12 marker strain which is resistant to an antibiotic, nalidixic acid to provide a conclusive link to the source and the sampling position. Air samplers sited at five distances downwind from the source up to 200m, with 2-5 samplers at each distance (depending on the distance from source) at a height of 1.5 m.

## RESULTS AND DISCUSSION

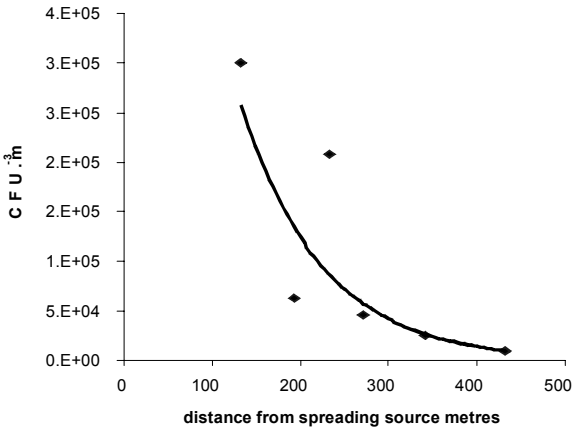


***Figure 1***. *Concentrations of enteric micro-organisms in the air from a splash plate spreader during the application of cattle slurry in wind speeds of 4.2 m. sec$^{-1}$*

Aerosols were not visible after distances of 20 metres from the spreader. However, as results in Table 1 demonstrate, pathogens can be transported distances greater than 200m. K12 *E. coli* was added to the slurry to confirm the source as being from the cattle slurry. According to the projections from the experiment, K12 *E. coli* can be transported between 500 and 1500m in wind conditions from 2.8 to 4.2 m.sec$^{-1}$. There were insufficient data to determine any decay rate of survivability or transmission because there was no sta-

tistical difference between the 50m and the 200m colony forming units either collectively or individually. During runs 1 and 2, enteric bacteria were counted at 9.3 x $10^4$ CFU.$ml^{-1}$ in the dairy washings. During trials 3 and 4 K12 *E. coli* was at concentrations of 2.5 x $10^5$ and 6.2 x $10^5$ CFUs $ml^{-1}$ in the cattle slurry. A further experiment showed that at higher wind speeds of 4.2 m $sec^{-1}$ (Fig.1) enteric organisms can be transported over 400m. If we examine the microbial counts with distance transported from the source and then determine what size particles can be transported at these wind speeds then we concluded that the particles that are associated with pathogen transfer are of the size 5 to 30 microns.

***Table 1***. *The experimental data from four trials spreading dairy washings and cattle slurry*

| **Run** | **1** | **2** | **3** | **4** |
|---|---|---|---|---|
| **Tanker passes** | 1 | 2 | 6 | 6 |
| **sampling time mins.** | 9 | 9 | 9 | 9 |
| **Sampled air volume (litres)** | 180 | 180 | 180 | 180 |
| **Slurry origin** | Dairy washings | Dairy washings | cattle slurry | cattle slurry |
| **pH** | 7.44 | 7.44 | 6.77 | 6.77 |
| **Dry matter % w/w** | 0.7 | 0.7 | 2.9 | 2.9 |
| **Wind speed hourly average m $sec^{-1}$** | 1.31 | 1.31 | 2.29 | 2.29 |
| **Beaufort scale** | 3-4 | 3-4 | 4-5 | 4-5 |
| **Direction degrees** | 276 | 276 | 261 | 261 |
| **Std dev of direction** | 15.6 | 15.6 | 7.6 | 7.6 |
| **Organism counted** | **Enterobacteria** | **Enterobacteria** | **K12 *E.coli*** | **K12 *E.coli*** |
| **Distance from spreader metres** | Averaged cfu/litre air/pass | Averaged cfu/litre air/pass | Averaged cfu/litre air/pass | Averaged cfu/litre air/pass |
| **200** | 19.3 | 11.1 | 0.30 | 0.61 |
| **150** | 29.2 | 8.6 | 0.58 | 0.60 |
| **100** | 12.7 | 6.4 | 0.48 | 0.56 |
| **50** | 17.7 | 13.0 | 0.57 | 0.68 |
| **-50** | 13.9 | 12.0 | 0.51 | 0.64 |

***Table 2***. *Experimental data from trials with dairy cattle slurry spread at high wind speeds*

| **Run** | 5 |
|---|---|
| **Tanker passes** | 6 |
| **sampling time mins.** | 9 |
| **Sampled air volume** | 180 |
| **Slurry origin** | Fresh cattle slurry |
| **pH** | 7.42 |
| **Dry matter %w/w** | 3.5 |
| **Wind speed hourly average m $sec^{-1}$** | 4.2 |
| **Beaufort scale** | 5-6 |
| **Direction degrees** | 270 |
| **Std dev of direction** | 15.6 |
| **Organism counted** | **Enterobacteria** |
| **Distance from spreader metres** | Averaged cfu/litre air/pass |
| 133 | 300000 |
| 193 | 62963 |
| 232 | 207778 |
| 271 | 46296 |
| 342 | 25926 |

In the second experiment, wind speed increased to an hourly average of 4.2 $m.sec^{-1}$ and enteric organisms were collected using Burkard samplers.

## CONCLUSIONS

The transfer of pathogens via aerosols from land spreading of cattle slurry using a splash plate spreader was detected. This was confirmed at over 200m at wind speeds of 2 $m.sec^{-1}$ and over 400m at wind speeds of 4.2 $m.sec^{-1}$ by the presence of enteric bacteria and the marker organism K12 *E.coli*. Projected estimations from these data indicate that *E.coli* K12 can be transported at least up to 1500m. Enteric bacteria were at a lower concentration in the spreader but were present in greater numbers on the Burkard plates than the K12 *E.coli.* This may be explained by several possibilities which include; a greater ability to survive; that enteric organisms were located inside the aerosol particles whereas the K12 *E.coli* may have been outside or detached from the particles and therefore more exposed to desiccation. Higher wind speeds increase pathogen concentration and transmission distance in the air.

## REFERENCES

Boutin, P., Torre, M., Serceau, R., Rideau, P. J. 1988. Atmospheric Bacterial-Contamination From Landspreading Of Animal Wastes - Evaluation Of The Respiratory Risk For People Nearby. J. Agric. Eng. Res., 39: 149-160.

Brunekreef, B., Dockery, D. W., Krzyzanowski, M. 1995. Epidemiologic studies on short-term effects of low-levels of major ambient air-pollution components. Environ. Health Persp., 103: 3-13.

Brenner, K. P., Scarpino, P. V., Clark, C. S. 1988. Animal viruses, coliphages, and bacteria in aerosols and waste-water at a spray irrigation site. Appl. Environ. Microbiol., 54: 409-415.

Dowd, S. E., Gerba, C. P., Pepper, I. L., Pillai, S. D. 2000. Bioaerosol transport modeling and risk assessment in relation to biosolid placement. J. Environ. Qual., 29: 343-348.

Evenden J J. *Bacterial aerosols generated by spray disposal of cattle manure*. 1972. Thesis/Dissertation

Lighthart, B. Kim, J. W. 1989. Simulation of airborne microbial droplet transport. App. Environ. Microbiol., 55: 2349-2355.

Nicholson F.A., Hutchinson M.L., Smith K.A., Keevil C.W., Chambers B.J., Moore A. 2000. A study on farm manure applications to agricultural land and an assessment of the risks of pathogen transfer into the food chain. MAFF Report London.

Nicholson K.W. 1995. Physical aspects of bioaerosol sampling and deposition. In Bioaerosols Handbook C.S Cox and C. M. Wathes, (eds.), CRC Press, New York. pp. 27-53.