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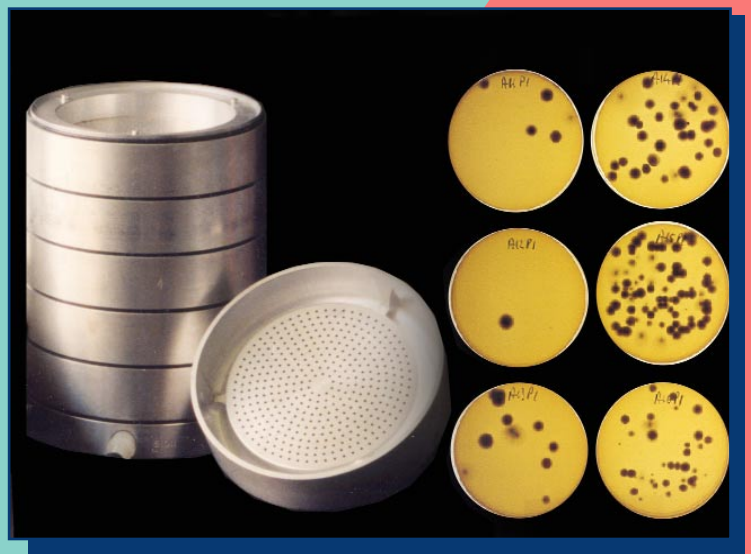
Improved Food Safety through Sterility of Air in Food Processing and Packaging

Dr. D.J. O'Callaghan

The control of particulate and microbial levels in a process environment should be undertaken as a co-ordinated exercise. The large fluctuations in microbial levels found in these studies underline the necessity of an effective and well maintained air filtration system, good work practices and regular monitoring of air quality using appropriate air sampling equipment.



Teagasc
The Science of Farming and Food

Improved Food Safety through Sterility of Air in Food Processing and Packaging

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Project Team:
D. O'Callaghan (Leader)
P. Murphy
D. Lynch
S. Coleman

The Dairy Products Research Centre
Moorepark, Fermoy, Co. Cork.

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Teagasc 19 Sandymount Avenue
Ballsbridge Dublin 4

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Summary and Conclusions

The guarantee of food quality, and particularly food safety, places an onerous responsibility on all food processors. EU Regulations (EN 29000 - 29004) define this responsibility not only in terms of the quality of the end produce, but also the precautions which must be taken in the preparation and processing of foods to minimise the risk of contamination. For all food products, and especially for those which are heat sensitive (e.g. milk or egg-based) and subject to minimal heat treatment, all aspects of the process must be rigorously controlled to prevent microbial contamination. This includes, not only the processing equipment, but also the processing environment including the air, which may reduce product shelf life, and in certain circumstances, pose a serious hazard to food safety.

While air filtration is now standard practice in food processing environments, significant increases in aerial microbial counts may occur intermittently due to failure to adhere to Good Manufacturing Practices and/or failure of the air filtration system due to design, inadequate maintenance, malfunction, etc.

Frequent and effective monitoring of the air sterility, in the process environment is therefore essential to alert processors of the potential risks, and the possible need for corrective action. However, the lack of reliable quantitative air sampling techniques, coupled to the uncertainty about the behaviour or control of micro-organisms within air filtration systems, present serious obstacles to effective control of processing environments.

The aims of this research were to develop reliable methods for evaluating the level of airborne micro-organisms in food processing facilities and to study the viability and behaviour of micro-organisms in air filtration systems.

The main conclusions were as follows:

- *Assessment of air quality involves monitoring both particulate and microbial levels as there is no simple relationship between these phenomena in a food process environment.*
- *The large fluctuations in microbial levels found in air in these studies underline the necessity of frequent and regular air sampling in processing facilities.*
- *It was established that micro-organisms can grow inside an air filter under certain environmental conditions and give rise to intermittent microbial germ contamination of the "cleaned" air.*
- *It was demonstrated that flowing air affects the survival of micro-organisms and the survival rate is dependent on filter class. Hence more emphasis on filter design aimed at effective microbial control is advised.*
- *A combined system for filtering and sterilisation by ozone was demonstrated to be an efficient technique for extending the microbial separation efficiency of air filters.*

Survey of sampling systems

A wide range of techniques for sampling micro-organisms in air were investigated and some of these were applied to monitor background and in-process levels of airborne micro-organisms in manufacturing processes. A number of these sampling systems, including impaction types (Biotest, RCS, Biotest RCS plus, SAS), slit/impaction type (Casella), sieve/impaction (Anderson), membrane type (Sartorius MD8), custom designed liquid impinger types (cyclone and all glass impinger) were evaluated in a challenging commercial dairy plant environment for sensitivity in monitoring microbial levels as well as ease of operation.

Impaction type samplers were operator friendly but sensitivity was dependent on the area of agar surface exposed to the sampled air. The multi-stage sieve/impaction Anderson sampler, while less convenient for field use, has been well established for research use and discriminates according to micro-organism *(Fig. 1)*. Slit samplers were found to be the most sensitive, due to the relatively large agar surface exposed to the sampled air. These samplers further combined the advantages of ease of operation with time-based profile of microbial deposition. A prototype with a longer running time per sample (i.e. capable of monitoring over an entire work shift) was developed for use in the course of the project.

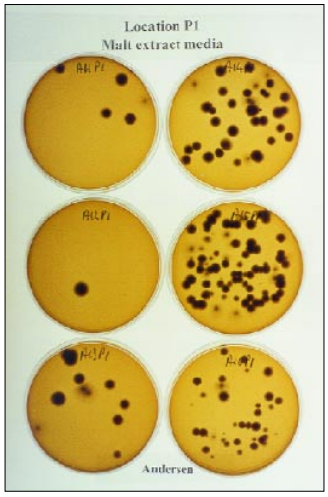


Fig. 1. Colony forming units shown from successive stages of an Anderson air sampler.

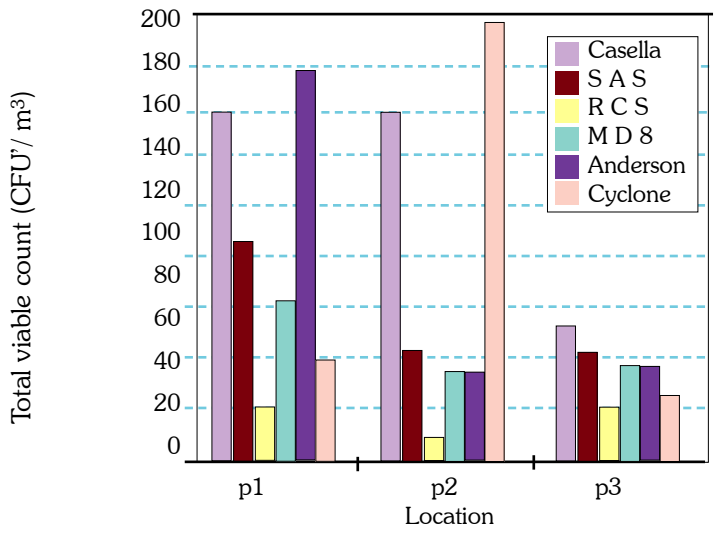
Membrane type samplers were found to be potentially quite sensitive provided the agar films were processed after exposure with due care by trained laboratory personnel.

The impinger type samplers were not considered successful due to poor microbial recovery from the sampled air and were relatively difficult to set up and subsequently required expert management.

In general the microbiological sampling of air in any location is subject to high statistical variations dependent on sampler type, volume of air sampled, collection efficiency, surface area and suitability of bacterial growth medium, etc. (Fig. 2). Hence, accurate quantification depends on appropriate sampler selection and frequent monitoring to establish trends and patterns as a basis for a reliable quality control system.

Anderson 6-stage sieve air sampler.

Fig. 2. Comparison of microbial recovery (CFU/m^3) of six air samplers used at three locations (p1, p2 and p3.)



On-site evaluation of filters in food process environments

Milk Powder Plant

Air filtration is now standard practice in commercial dairy and food plants. However the bacterial status of the filters used is not well understood. Hence a glass fibre filter (EU7), in use for several months in a typical commercial drying plant, was removed for investigation.

Stereo-micrographs of this glass fibre filter showed that milk powder particles dominated the contamination found on the inlet face of the filter, with both large and small milk powder particles seen to adhere to the filter fibres, mainly in clusters but also individually. Smaller bacterial rods, some appearing in clumps, could also be seen adhering to the filter fibres. In another study, air samples were taken from an F5 deep bed filter unit which was integrated in an air conditioning system for air supply to the laboratory in the plant. A variety of Gram-positive and Gram-negative micro-organisms, typical of soil, were found on the intake surface of the filter which had been in use for about 6 months. However, at the exhaust side of the filter, the Gram-positive bacteria represented the major proportion of the micro-flora, suggesting that either the micro-organisms were able to grow in the filter and leave at the exhaust side or were able to pass selectively through the filter. The predominant micro-organisms found in the

laboratory air space were Gram-positive cocci (especially Staphylococcus spp.) which are known to be related to human activity. *It was concluded that the filter was effective in removing bacterial contamination from the external air supply.*

When evaluating filter systems in a milk powder plant it was found that most of the filters were performing well and in accordance with the manufacturer's specifications. The HEPA (High Efficiency Particulate Arrestance) filters which were in place for a long period of time (two production seasons) were all working close to specification and would not need to be replaced from a particle efficiency point of view (*Table 1*). However one filter (C) was failing to arrest the smaller particles (<1 μm) possibly due to a very high air velocity through this filter which was well in excess of recommended limits.

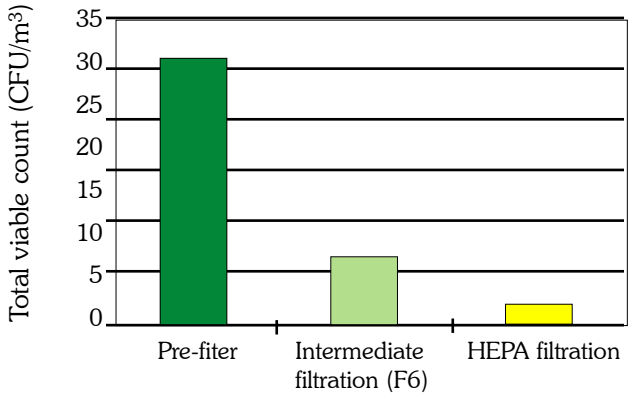
Filter	A		B		C		D		E	
Type	*Bag filter (A)*		*EU3 (B)*		*EU8 (C)*		*HEPA (D)*		*Prefilter+HEPA (E)*	
Reduction (%)	*Ex*	*Me*	*Ex*	*Me*	*Ex*	*Me*	*Ex*	*Me*	*Ex*	*Me*
Sizeband (µm)										
>10	100	100	90	100	97	100	100	100	100	100
5 - 10	100	100	75	0*	97	96	100	100	100	100
1 - 5	60	78	40	86	97	64	100	100	100	99.87
0.7 - 1.0	30	71	20	77	83	53	100	100	100	99.87
0.5 - 0.7	20	51	15	52	55	22	100	99.996	100	99.87
0.3 - 0.5	15	25	10	23	55	13	99.995	99.994	99.995	99.698

** count was 1 before and 1 after*

Table 1. Expected (Ex) and measured (Me) reduction (%) in particle counts by sizeband for various filters.

While no filters tested produced sterile air in the process environment, air filters were found to reduce bacterial counts depending on specification, number of stages, level of maintenance, working age, etc. (Fig.3).

Fig. 3. Typical reduction in bacterial count in a two-stage air filtration system



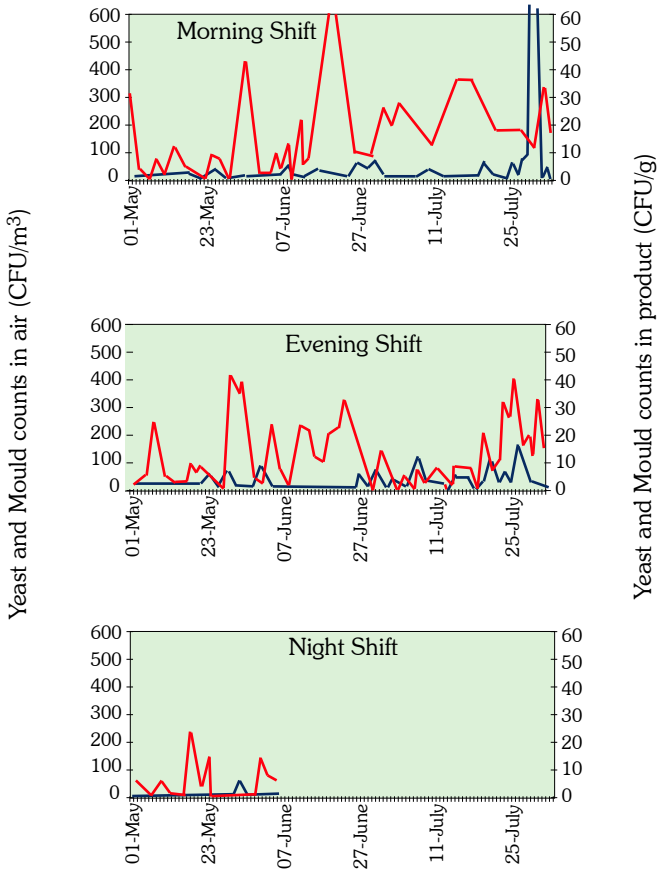
Processed Cheese Plant

Microbial Counts in the ambient air and in the product

The microbial status of air in a large commercial cheese plant, in the course of a typical process runs, was evaluated over a 3 month period.

The critical process involved the extrusion of hot (80°C) molten cheese onto a cold stainless steel roller from which it was delivered in a flat strip on a conveyor to a slicer and packaging unit. During this period the product was exposed to the ambient air for a period of 1 minute approximately. The ambient air in the room was filtered and kept under positive pressure.

Fig 4. Comparison of Yeast and Mould counts in process air ( ) and in product ( ) taken over a 3-month period.



During the 3 month evaluation period the air and the product were sampled on a daily basis, for Total Microbial and Yeast and Mould counts.

Higher than expected microbiological counts, with large day to day variations, were recorded (Fig. 4). In most instances the total microbial content of the process air was higher than that in the product. However, no obvious correlation was observed between the Total Microbial or Yeast and Mould counts in the process ambient air and those in the product.

Evaluation of a fogging system for air cleaning found that a 3-shift schedule would not allow sterilisation by fogging on a daily basis and hence fogging could only take place at weekends as it can only be performed when plant is down and unoccupied.

The results indicate that in the majority of cases the process air during the third shift was cleanest during the night shift, having the lowest micro-biological count. This was contrary to what was expected, i.e. that the cleaner air would be in the first shift, but might be explained by the fact that during the night shift there was less movement, fewer personnel and, better compliance with entry and exit procedures. However, it should be noted that high sporadic microbiological counts occurred in all shifts.

In some cases the first shift showed higher counts than the second, which could be attributed to the plant having been idle overnight, when airborne dust particles would settle in ducts and on equipment. When the plant was started all of this contamination could easily be disturbed and become airborne again.
Microbial counts obtained with the Settle Plates did not correlate to any great extent with those obtained using the impaction sampler, highlighting the inaccuracy of techniques which do not sample known volumes of air in the course of a working shift (e.g. start of shift, end of shift, shut down of production line, etc.). Settle Plate counts for Yeasts and Moulds taken over a further 41-day period, made up of roughly equal parts of two-shift and three-shift operations, showed that high counts were more frequent with three-shift operations. However, when Mould counts obtained by Settle Plate were plotted against those obtained with the Biotest sampler, there was no correlation, indicating that the Settle Plate technique is not quantitatively reliable because it is not related to a defined airflow.

Particulate counts in the ambient air
During a two day working period the particulate counts were monitored continuously at four locations (air inlet, cheese slicer, cheese packing and air exhaust) in the process room *(Fig. 5)*.

Consistently, the lowest counts were found close to the air inlet, typically 700 to 7000 /m^3. The counts measured in the three other locations were relatively similar, but higher by a factor of 10 to 100 approximately than those at the inlet. Hence, the counts at the centre of the room are influenced by factors other than the quality of the inlet air, such as the generation and movement of dust particles by operators and machines, door opening, etc.

It can be concluded that incoming air is only one source of aerial contamination and that filtration of incoming air will only be effective if accompanied by other dust control measures such as personal clothing regimes and washdown cycles.

No relationship was identified between temperature, relative humidity or absolute humidity and the particle/microbial counts obtained.

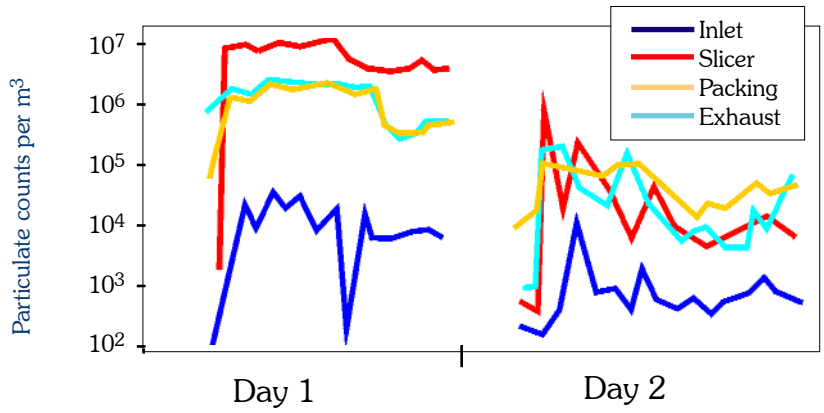


Fig. 5. Continuous measurement of particulate counts at four locations in a process cheese plant.

Comparison between microbial counts and particulate levels

Particulate measurement based on optical techniques is immediate and reliable. On the other hand the quantification of microbes in air is constrained by the requirement to incubate samples for up to 3 days. Hence in an attempt to eliminate the microbial detection interval, studies were undertaken to determine the feasibility of predicting microbial counts from particulate counts.

Total Bacterial Counts (TBC – 3 days at 30°C) and Mould Counts were taken in processing areas on a daily basis using Anderson, Mattson-Garvin samplers and Settle Plates and compared with total particle counts by size band.

The best correlation with dust particle count was for Mould Counts with the Settle Plate technique with respect to the two micron size bands (0.7 - 1.0 and 1.0 - 5.0 micron, significant at $P < 0.01$). This correlation suggests that much of the mould found on Settle Plates was deposited by mould laden dust particles. However, no significant overall correlation was obtained when using any of the sampling instruments (impaction plate or slit sampler) for either Mould or TBC with any size band in the particle counter.

Evaluation of Filter Performance on Test Rig

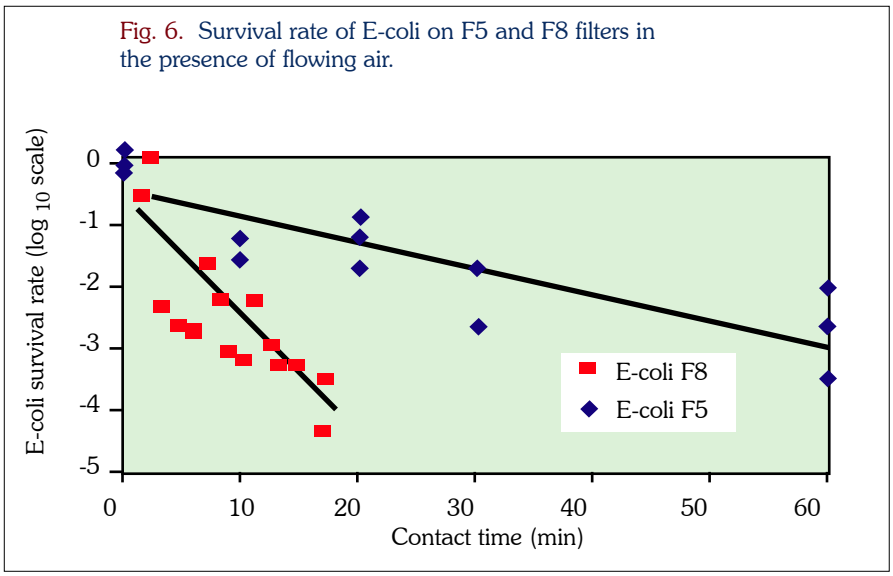
The aim of these experiments was to evaluate the performance of filter media in removing particles and bacteria from air.

Particle separation

Firstly a typical two-stage air filtration system was evaluated for in-process particulate separation efficiency, using a particle counter at the inlet and outlet of each stage. The two stages consisted of an F5 prefilter and an F8 final filter.

The F5 filter was found to be highly effective in removing particles greater than 1μm in size while the F8 filter proved highly effective in removing the smaller particles. Hence both filters played a complementary and effective role in air filtration.

Fig. 6. Survival rate of E-coli on F5 and F8 filters in the presence of flowing air.



Microbial separation

Trials were then undertaken on the laboratory test rig to determine the effect of air-flow on bacteria in filter media. Deep-bed non-woven fibre filter media of F5 and F8 grades, were each impregnated with an aerosol containing marker bacteria (*E. coli)* resulting in counts in the filter material of between 10^6 and 10^3 Colony Forming Units (CFUs)/g. Following impregnation, non-contaminated air was passed through the filters for up to one hour and the bacterial levels in the filter material monitored at 10 minute intervals. Using F8 filters the *E. coli* counts in the filter material dropped from 10^6 to 10^3 CFU/g approximately in 10 minutes *(Fig. 6)*. The velocity of air flow through the filter was 0.5 m/s. The count of micro-organisms shown depended on the amount of air handled. However, to achieve a similar count reduction using the F5 filter, 60 minutes of purging with air was required. Repeating this experiment with *Micrococcis luteus*, it was found that, although the reductions in counts were slower than for E. coli, the filter pore size effect was similar.

These results suggest that air flow through the filter material has a bactericidal effect (possibly due to dehydration) and may vary for different microbial species.

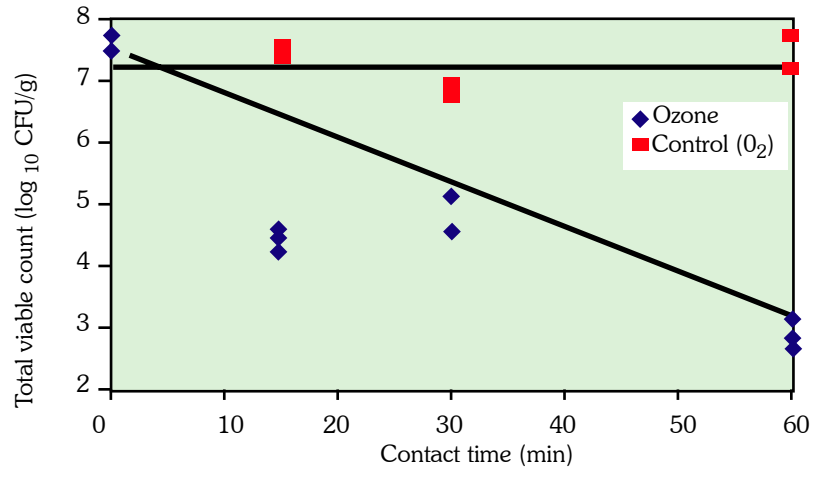
Evaluation of a sterilisable filter system

The accumulation of organic matter, especially of food origin, in a filter provides a medium for microbial proliferation. Hence the efficiency of the filter in blocking microbes may quickly be offset by the dispersal of micro-organisms which grow in the filter material. To extend the performance of the filter in providing microbial free air, the use of ozone as a sterilising agent for the filter was evaluated, since ozone combines effective bactericidal action with minimum environmental impact. A filter system was adapted, allowing for periodic ozone saturation of the filter.

A suspension of *M. luteus* was sprayed on an F8 filter material for 30 minutes giving a concentration of about 10^7 CFU/g on the filter material samples before applying sterilisation. These tests showed that the survival of micro-organisms on the filter materials was dependent on the exposure time to ozone. After fifteen minutes exposure to ozone a one thousand fold decrease in the concentration of micro-organisms was found (*Fig.7*). Exposure to oxygen (as a control) gave no reduction on the viability of the micro-organisms.

These results showed that a combined system for filtering and air sterilisation using ozone at predetermined intervals, with ca. 15-30 minutes exposure is a practical technique to extend the microbial separation efficiency of the filter. For commercial application, existing filtering systems may be adapted by retro-fitting the sterilisation unit ensuring that the ozone treated air is by-passed to the atmosphere.

Fig. 7. Effect of ozone exposure time on microbial survival on filter material



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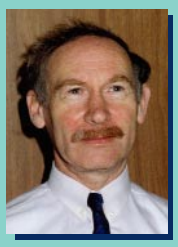

Dr. P. Murphy

DAIRY PRODUCTS RESEARCH CENTRE
Moorepark, Fermoy, Co. Cork, Ireland
Tel: +353 (0)25 42222 - Fax: +353 (0)25 42340
e-mail: reception@moorepark.teagasc.ie