Comparative Investigations on the Blocking Efficiency of Filter Tips

The blocking efficiency of Filter Tips is investigated using airborne particles. Filter tips mounted in air-displacement pipettes are used to prevent potential contamination of the pipette cones by e.g. DNA fragments. The more effective the filter, the less the risk of contamination. This is of utmost importance for preventing false-positive results, especially in highly sensitive PCR experiments. In this investigation Starlab TipOne 200 μl filter tips have been compared with those of nine other manufacturers.

Today a many research organizations are using PCR routinely for the detection of DNA sequences. The increase of this technique leads to an increasing risk of contamination leading to false positive results. Since e.g. double-stranded DNA, the main product of PCR, is very longlived it seems obvious that in the PCR procedure pipetting of DNA or PCR product is one of the most insecure steps. Cross-contamination occurs if incorrect pipetting causes splashes or drips. Yet even if pipettes are handled properly aerosols can still be generated, containing DNA molecules which can thus contaminate the pipette and then subsequent different batches. This demands increasing efforts to reduce penetration of contaminants by means of filter tips.

Measurement of Blocking Efficiency of Filter Tips: Particle Sizes Used for the Test

Since contamination is believed to occur mainly by airborne particles or macro molecules it is appropriate to use airborne particles for testing as well [1]. Pipette tip filters were tested with regard to their blocking efficiency using airborne particles in a size range corresponding to maximum penetration probability [2] (compare Fig. 1) of the filter tips. This makes the tests as rigorous as possible.

Experimental Setup and Determination of Blocking Efficiency

Sodium chloride particles where generated by dispersing an aqueous sodium chloride solution and subsequently drying. A suitable concentration of the solution was chosen to generate particles with a mean diameter of approx. 100 nm. These particles where passed through a filter tip and the particles penetrating were counted (Fig. 2). Comparing measurements with and without filters mounted in the tips gives the total retention of particles in the filters. Following experiments described in the literature the flow rate through the tips was set to 1 ml/sec.

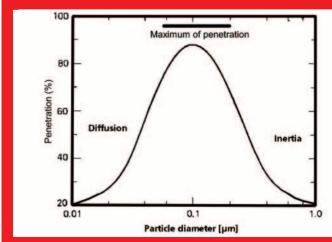


Fig. 1: shape of penetration curve of a filter

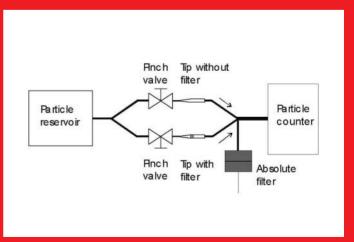
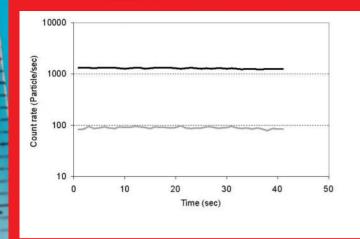


Fig. 2: Setup for the measurement of the particle blocking efficiency



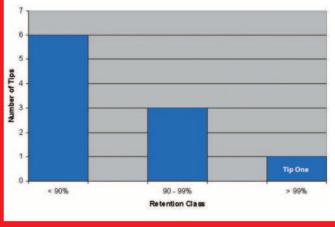


Fig. 3: Typical shape of count rate curves measured resulting in a blocking of R=93% aerosols. The upper curve represents the tip without filter, lower curve tip with filter.

Fig. 4: Summary of blocking measurements

For the determination of the blocking efficiency of the filter tips the total number of particles penetrated through a tip with a filter mounted during a defined period of time is compared to the corresponding total number penetrated through a tip without a filter. If $N_F,_{\Lambda T}$ is the number of particles penetrated through the tip in time period $_{\Lambda T}$ and $N_W,_{\Lambda T}$ is the number of particles penetrated through the tip without a filter in the same time period the particle blocking is defined as:

$$R = 1 - \frac{N_{F,\Delta T}}{N_{W,\Delta T}}$$

Since the particle concentration in the reservoir is not constant during the measurements the two types of tips are alternately measured several times. This procedure is repeated three times for every type of tip.

Results

Fig. 3 shows typical count rate curves measured for tips without (upper, black) and with (lower, grey) filter. The retention (R) measured for the different filter tips covers a wide range. Therefore filter tips were classified into three different retention classes depending on their blocking efficiency: a lower retention class containing all filter tips with retention values smaller than 90 %, a medium class for all tips with retention values be-

tween 90% and 99% and a class summarizing all tips with values better than 99%.

Fig. 4 summarizes the results of the measurements. $60\,\%$ of the filter tips investigated belong to the class below $90\,\%$ retention. $30\,\%$ show retention values

Туре	Penetration	
Company 1, 200 µl		6,8 %
Company 2, 180 µl		29 %
Company 3, 100 µl		20 %
Company 4,	200 μΙ	20 %
Company 5, 2	200 μl	26 %
Company 6,	100 μΙ	14 %
Company 7, 2	200 μl	18 %
Company 8,	200 μΙ	6 %
Company 9, 200 µl		8,3 %
Starlab, TipOne 200 μ l graduated Filter Tips $<$ 1 $\%$		

Fig. 5: Result of single retention measurements

between 90 % and 99 % and only one filter tip, the Starlab TipOne $200\,\mu l$ tip shows values better than 99 %. The individual results are listed in Fig. 5.

Summary

In this article the results of comparative measurements of ten different filter tips available on the market are reported. The parameter investigated is the blocking efficiency of the filter mounted in the tips. Since it can be assumed that a contamination risk is strongly accompanied with the penetration of molecules through the filter the possibility of false

positive results in e.g. DNA detection can be reduced by orders of magnitude by the use of filter tips with high blocking efficiency. In the cases investigated three different retention classes could be defined. Therefore in the cases investigated the risk of contamination can vary by more than two orders of magnitude depending on the filter tip used.

References

[1] Hinds, W. C.: Aerosol Technology, 2nd edition, John Wiley & Sons, New York, (1999)

[2] Brown, R. C.: Air Filtration, Pergamon, Oxford (1993)

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