

The measurement of water flux through human skin is important in the cosmetics and pharmaceutical industries. It plays an important role in the evaluation of treatments for skin injuries and diseases. In this note we demonstrate how to measure this quantity using the DVS line of instruments and a Payne type diffusion cell

# Introduction

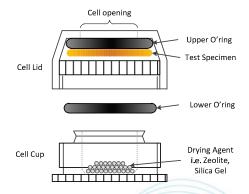
In a previous study, Application Note 52 [1], a Payne type diffusion cell was used to measure moisture flux through various skin simulants and trypsinized human skin under different conditions. Clear differences were observed between the simulants and the trypsinized skin.

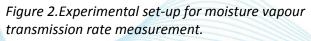
In this study we will use the skin simulant VitroSkin [2] to investigate the ability of skin creams to act as occlusives by measuring their impact on TransEpidermal Water Loss (TEWL) or moisture flux. The manner in which the Payne cell is used will be evaluated by comparing results between dry cell and wet cell measurements.

## Method

As stated in Application Note 52, a Payne type diffusion cell is used for these studies. Figure 1 shows a cross section diagram of the cell.

The base of the cell has a cavity which can be filled with desiccant when performing a dry cell test, or with water if performing a wet cell test. The sample under test is cut into a round 7mm diameter coupon and placed into the cell between two o-rings. A paper punch was found to provide the correct size. Once the test specimen is fitted on the lid, the bottom cell cup and the cell lid are screwed together. The cell is placed on a DVS metal sample pan for measurement in the DVS instrument. The cell has an opening diameter of 4.4 mm on the top, providing an exposed area of 15.20 mm<sup>2</sup> for moisture transport.





The cell with sample and desiccant (or water) will weigh approximately 650 mg. In most cases the absolute weight of the sample is not important as rate of mass loss or gain is the desired value, and is expressed in units of  $g.hr^{-1}.m^{-2}$ .



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To validate the procedure and the seal afforded by the cell a few test runs were performed. These tests also show the maximum and minimum mass uptakes that can be expected during an experiment on real materials.

Dry-cell tests were performed with a 50% RH, 25 °C, and 200 sccm gas flow. Figure 2 shows the result of a test using a cell filled with dry Zeolite. The cell was open to the gas flow with no sample film in place. This test shows the maximum water uptake that may be expected at these conditions.

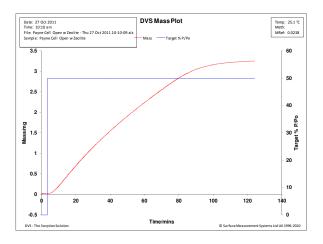


Figure 2.Mass uptake with an open Payne cell filled with dry Zeolite.

The mass of the sample is shown with a red line, while the target humidity is shown with a blue line. Initially the blue line is at 0% RH (right axis), during this time the mass response is nearly flat. The RH is then increased to 50% and the mass response increases almost immediately. The mass uptake is nearly linear with time. At 80 minutes the rate of mass increase begins to decrease and soon approaches a flat response. This indicates that the Zeolite is now saturated with water and cannot uptake any more water vapour. Data from a linear portion of the mass response is plotted in Figure 3, and fitted to a straight line to derive the slope or change in mass versus time. With the known exposed surface area of the cell, it is possible to calculate a steadystate moisture flux of 131 g.hr<sup>-1</sup>.m<sup>-2</sup>.

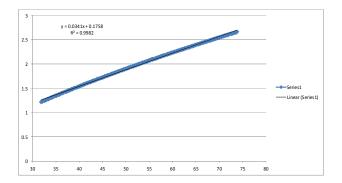


Figure 3.Steady-state mass increase. Slope is used to calculate moisture flux.

Another test was made using thick aluminium foil film as the sample, to demonstrate that a good seal with the o-rings in the cell could be achieved. This test is shown in Figure 4.

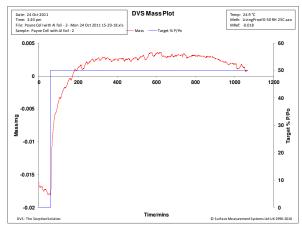


Figure 4.Mass response with an aluminium foil sample.

There is an initial increase in mass, but this is only a 15µg increase and is thought to be due to surface adsorption on the Payne cell components and on the aluminium foil sample. After 200 minutes the mass response becomes flat for an extended period of time. To verify that the Zeolite is still active after the extended test, a small hole was punched into the aluminium foil and the Payne cell was placed back into the instrument at the same RH conditions. As can be seen from Figure 5, the Zeolite was still quite active and the mass increases rapidly at a rate of about 750µg/hr, proving that the flat response

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was due to a good seal and not to the Zeolite having been saturated.

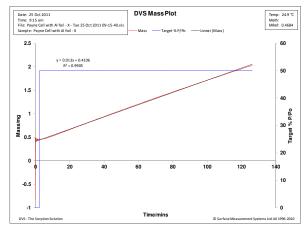


Figure 5.Mass response with punctured aluminium foil sample.

# Results

Samples of VitroSkin were tested using the drycell method with Zeolite as the desiccant. A typical result is shown in Figure 6.

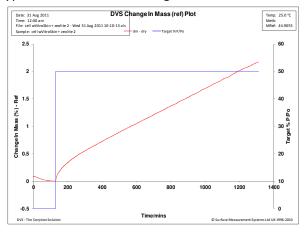


Figure 6.Mass response for a dry VitroSkin sample, 25 °C and 50% RH.

Analysis of the mass response results in a flux of  $2.7 \pm 0.11 \text{ g.hr}^{-1} \text{ .m}^{-2}$  (n=4). Literature values for actual *in-vivo*skin flux rates are in the range of 10 g.hr^{-1} \text{ .m}^{-2}[3,4].

For comparison, data has been taken to determine the diffusion coefficient of water as it diffuses into dry VitroSkin. The method used was the same as Application Note 16. From 0% to 50% RH the D value is  $1.5 \times 10^{-8} \text{ cm}^2/\text{sec.}$ , and from 50% to 90% RH the value is  $2.4 \times 10^{-8} \text{ cm}^2/\text{sec.}$ 

Tests were also done using VitroSkin and a wetcell procedure. The Payne cell was filled with deionized water instead of Zeolite and the rate of mass loss (TEWL) due to water evaporation through the test sample was measured. The cell was positioned so that water was in contact with the test sample. The results are very different than those obtained with a dry-cell test. The flux is over 150 times greater than before. Figure 7 shows the result of one of these wet-cell tests.

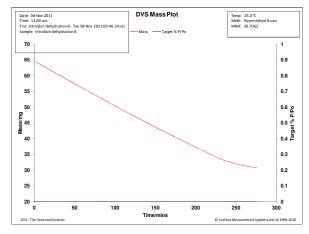


Figure 7.Mass response – VitroSkin wet-cell test.

The steady-state mass loss (i.e. linear portion of the curve) is calculated to be 530 g.hr<sup>-1</sup>.m<sup>-2</sup>. Although this water flux is large compared to the literature value of *in-vivo* skin flux, it should be remembered that the water in this test is free water in direct contact with the membrane and that the membrane is only an approximation to actual skin. This test method is a reasonable model to measure TEWL and the large flux should allow for small differences caused by surface treatments to be observed. The methodology may be improved by replacing the deionized water in the cell with fluid approximating sub-dermal fluid.

To determine the effect of hand creams on the water evaporation rate, or occlusive properties, three hand creams were tested by applying a thin coating to the VitroSkin sample under test. The same procedure was followed for all samples. The VitroSkin sample coupon was placed over the water filled Payne Cell. One o-ring below and one above the VitroSkin assures that water vapour does not escape unless it diffuses through the sample. Once the sample is securely in place, a foam-tipped swab was used to apply a thin coating of the hand cream to be tested. Care was taken to ensure that the coating was uniform. Ideally the Payne cell may be weighed before and after to obtain the mass of the cream being applied.

The mass response from a test using Cream C is shown in Figure 8. Some tests will show a different slope during the first hour of the test. This is thought to be due to water evaporation from the cream under test. The mass response becomes linear after this initial induction phase. This is where all water loss flux values were calculated.

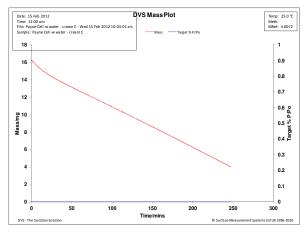


Figure 8.Mass response – VitroSkin with Cream C applied, wet-cell test.

The results shown in Table 1 list the measured TEWL values and any reduction in TEWL compared to the untreated sample. For creams A, B, and C, average values are shown (n=4). Measurements were also made with Glycerol and a petroleum-based lip balm. None of the results exceed the value obtained for the untreated VitroSkin, 530 g.hr<sup>-1</sup>.m<sup>-2</sup>. This implies that all surface treatments reduce the maximum flux or TEWL through the sample.

Treatment	TEWL [g.hr <sup>-1</sup> .m <sup>-2</sup> ]	Reduction in TEWL Versus Untreated
Cream A	397± 29	25%
Cream B	419± 53	21%
Cream C	265± 49	50%
Glycerol	479	10%
Lip Balm	165	69%
Untreated	530	-

Table 1:VitroSkin water flux (wet-cell method) at
25 °C with hand cream applied to top surface.

The data show real differences between the creams tested. The most effective cream in reducing water evaporation through the VitroSkin sample was Cream C with a reduction of 50% compared to the untreated sample. The least effective cream was Cream B with a reduction of 21%. Petrolatum is considered the most effective occlusive agent [5] and this test shows that it does exhibit the best performance of all the compounds in the series. It reduces the TEWL by 69%. Glycerol is a humectant so it was not expected to provide much of a reduction in TEWL. As such it only provides a 10% reduction compared to the untreated skin sample. The values reported for the three creams are the average of four measurements each.





## Conclusions

SYSTEMS

Three hand creams and two reference compounds were tested using DVS and a Payne type diffusion cell as a way to measure TEWL. The results show that it is possible to detect differences in TEWL between hand creams and between reference compounds. A petrolatum based balm exhibited the best performance as expected from literature references regarding this agent. One of the hand creams was clearly better than the other two in TEWL performance. None of the results exhibited a larger flux than was measured from a non-treated sample of VitroSkin, which lends confidence to the technique described.

### Acknowledgement:

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### References

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