Application Note

Evaporation from Thermo Scientific Nunc Edge Plate With Solid Reservoir

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Abstract

When using a liquid reservoir in Thermo Scientific Nunc Edge plates in order to counteract evaporation from the wells, there may be a risk of spilling the liquid from the reservoir by abrupt manual or automatic plate moving. Adding a jellying agent such as agarose to the liquid could eliminate this risk. The effect of a 0.5% agarose solution, which was found to be optimal, is here reported.

Materials and methods

Nunc™ Edge plates were filled with 1.75 mL water or 2.5 mL 0.5% agarose (dissolved in water by heating in a microwave oven for 1 minute) per reservoir compartment. After setting of the agarose for half an hour at RT, the Edge™ plates together with standard F96 plates were filled with 100 µL 0.002% aqueous crystal violet solution per well. The plates were incubated on the middle incubator shelf at 37°C, 5% CO_2 and 95% relative humidity for 4 days, during which the incubator door was opened 90° for 15 seconds every hour seven times a day in order to simulate realistic conditions. The total evaporation from the wells and the reservoirs were determined by appropriate plate weighing before and after incubation, and the distribution of the evaporation on the individual wells were determined colorimetrically by the OD values at 590 nm in 50 µL well aliquots according to the following approximation:

$$\%\ \text{well evaporation} = \frac{\text{g total water loss from wells}}{\text{g total water added to wells}} = \frac{OD_{well}}{OD_{average}} \times 100$$

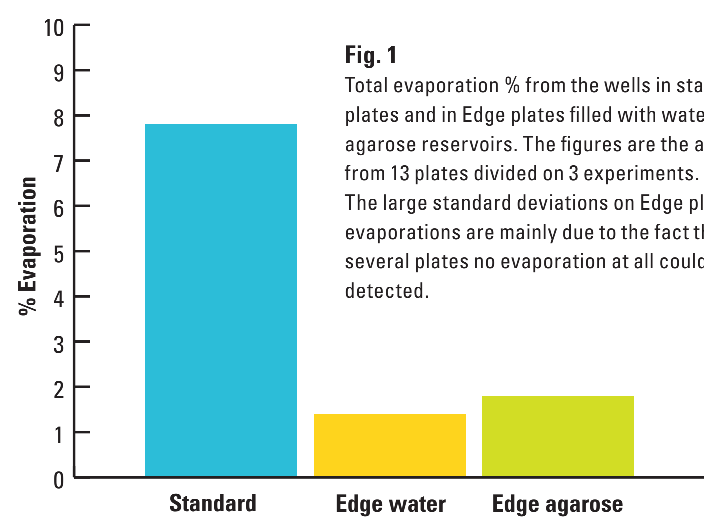

Fig. 1
Total evaporation % from the wells in standard plates and in Edge plates filled with water or agarose reservoirs. The figures are the average from 13 plates divided on 3 experiments.
The large standard deviations on Edge plate evaporations are mainly due to the fact that in several plates no evaporation at all could be detected.

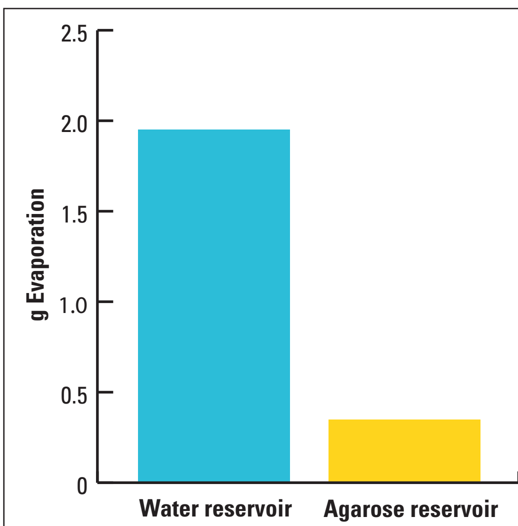

Fig. 2
Average evaporation amounts from the Edge plate reservoirs.

Results

The protection against evaporation by the Edge plate agarose reservoir is substantial, but maybe not quite as effective as by liquid water reservoir (Fig. 1) – although the amounts of evaporation from the respective reservoirs are the same (Fig. 2). Like that of water, the effect of agarose is not only a reduction of the total evaporation from the wells, but also an elimination of the evaporation edge effect (Fig. 3). The incubator humidity, which determines the evaporation velocity, is dependent on incubator door openings, so the 95% relative humidity is decreased to about 90% by frequent door openings and is only restored by the tranquility observed overnight (Fig. 4).

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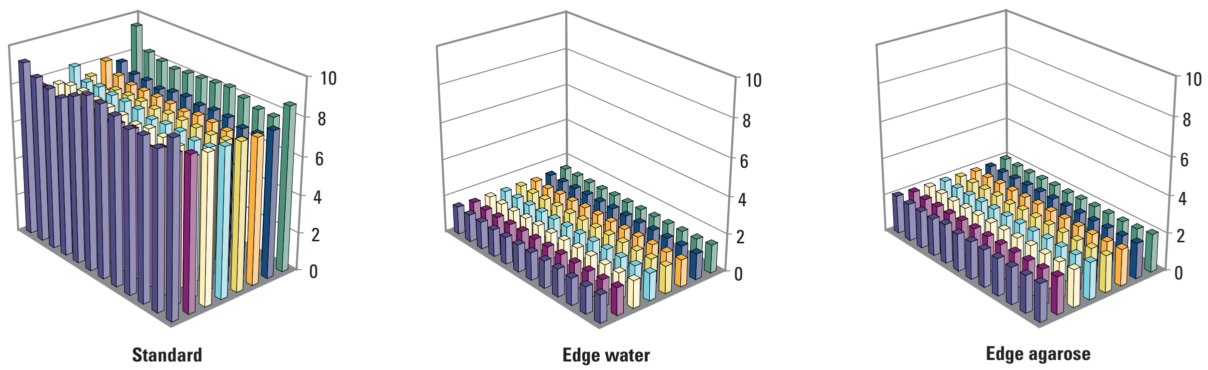

Fig. 3.
Average evaporation percent from the individual wells throughout the 96 well plate matrix for standard plates, and for Edge plates with water and agarose reservoirs respectively.

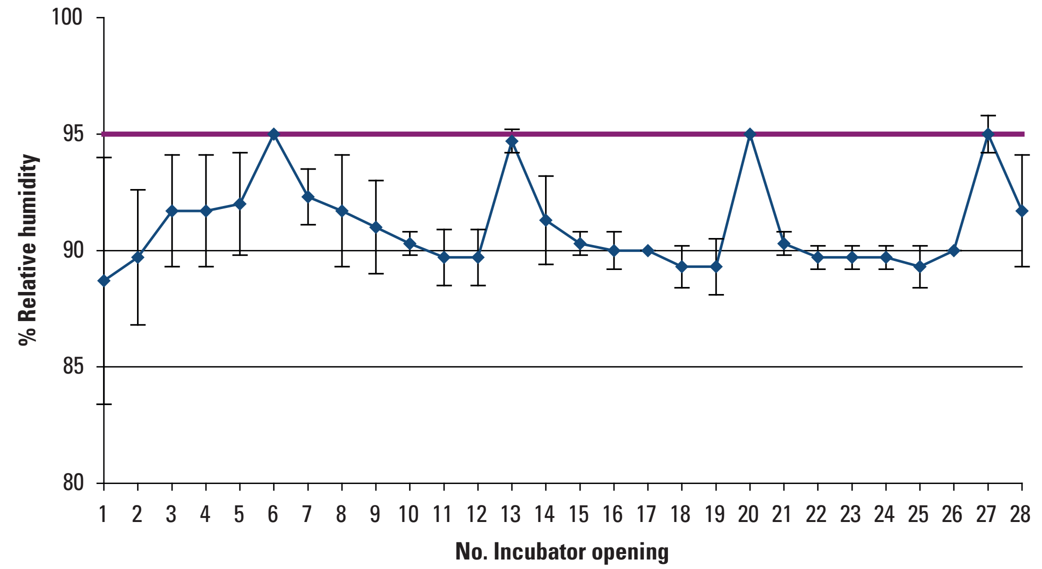

Fig. 4
Average incubator humidity readings from the three experiments just before the incubator door openings. The spike values reaching 95% relative humidity (pink line) appeared as the first readings after night tranquility. The concomitant temperature and CO_2 readings (not shown) were on the other hand practically constant at 37˚C and 5% CO_2 respectively.

Conclusion

The use of a 0.5% agarose gel as Edge plate reservoir is almost as effective as liquid water for protection against evaporation from the plate wells. This may be applied in order to eliminate the risk of spill from the reservoir by plate movement. In particular, one could hereby avoid any plate robot resetting (slow down), which might otherwise be necessary. In addition, one could without risk increase the reservoir volume, thus prolonging the protection period for long-term uses before the reservoir gets exhausted.

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