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Deaerating

What is Deaerating? Deaerating is the process of removing dissolved gasses from a material, typically a liquid. Due to the nature of diffusion cells, the formation of bubbles can be a major concern. Most bubbles will move up and can potentially block a portion of the membrane; reducing the area for the drug to diffuse through. There are methods both during preparation and in some cases during the test to deal with bubbles within diffusion cells.

Why Deaerate? Diffusion testing is critically linked to the available surface area of the membrane. This applies to the donor chamber, where the product should be evenly spread on the membrane surface. It also applies to the receptor chamber where any blockage to the membrane would result in a reduction of the diffusion rate. Any liquid typically has a certain amount of air dissolved in it that is invisible to the eye. When heated, or when subject to pressure changes, the molecules of gasses can group together to form bubbles. Removal of the air prior to using the receptor media can prevent this from happening during a test where it may affect results. Bubbles due to deaeration issues are typically small (1-2 mm diameter) and numerous. They will deposit on both the bottom of things and often the sides/walls as well. Bubbles may not always be due to deaeration. If bubbles are larger in diameter (3 mm or larger) there is a chance the cell could be leaking, or could be introduced by poor sampling techniques. Observation during a test is important. The size and number of bubbles can give some indication of what the problem is. In some cases, it may be possible to dislodge and remove the bubbles before there is a significant effect to the data.

Methods of Deaeration: There are a number of ways to deaerate, and choosing the best method may depend largely on the receptor media used for the test.

Vacuum Filtration: This method uses vacuum pump to drawn the receptor media through a filter. Typically the filter is a membrane filter with 0.45 µm porosity. The membrane material would depend on its compatibility with the media being filtered.

- Suitable for aqueous media, or media with a low organic content.
- Heating can further improve deaeration when combined with this method, but is not recommended for media containing volatile organic components.

Sonication: This method makes use of an ultrasonicating bath or probe. Sonication baths/probes with a “degas” or similar mode will operate at a frequency that is ideal for removing gasses. Sonication baths/probes without a degas mode can still be used, but may not be as effective.

- Suitable for most types of media
- Performs better with a dedicated “degas” mode
- Can be further enhanced with heat or vacuum, but that is not recommended for media with a significant organic content.

Heating: Heating alone can remove a significant amount of air from liquids, but is not suitable in all circumstances. Generally speaking, a cold liquid will hold more air than the same liquid at a higher temperature. When using heat alone to deaerate, it is key to ensure the media is held above the test temperature by a few degrees Celsius and then allowed to cool to the test temperature. If the media falls below the testing temperature it may re-absorb air which could then be released during the test.

- Suitable for aqueous media.
- Would typically be heated about 5 °C above the intended test temperature.
- Is more efficient when combined with sonication for vacuum.

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Automated Equipment: There are automated deaeration systems available, but most are focused around dissolution testing in both quantity media, and the method of deaeration. If an automated solution is being considered it is important that customers consider the volumes the systems work with, the method of heating, and the compatibility of wetted materials.

- Can provide a simple automatic method of deaerating media.
- Primarily focused on aqueous media, may not be suitable for any organic content.
- Quantities may be too large to be of practical value for diffusion testing.

Testing Procedures to deal with bubbles:

Prior to testing it's important to make sure that there are not any bubbles on the stir bar or walls of the diffusion cell which could rest against the membrane during the test. Typically running the stirrer for a few minutes prior to applying the membrane will dislodge them. Bubbles that are not removed with this method can be scraped free from walls with an clean pipette.

The most important rule of testing is to make observations. This should be done whether the system is manual or automated. With manual systems observations can be done at every sample point. If there is a significant amount of time between samples (multiple hours) it is good to make inspections in-between sample points as well. With automated systems it is easy to ignore observations and let the instrument handle everything, however if a bubble, or another error occurs, there may be a significant effect on the data and no clear reason why. In most cases this would require retesting, which in many cases can be costly.

If bubbles are seen within the cell the user should determine the following:

1) **How big is the bubble, or bubbles?**
   a. This can be difficult to judge in some cases, but is one of the most important observations.
   b. If inspecting through the side of the cell is not working, an inspection mirror can be useful to look up at the membrane through the donor chamber.

2) **Are there enough of them that they may block a significant portion of the membrane?**
   a. If a single bubble of 1 mm in diameter is observed, the area of the membrane that is blocked is typically quite low. It is unlikely any change from typical results would be observed.
   b. If a single bubble of 4 mm in diameter is observed, the are of the membrane that is blocked is likely to be significant even on 15 mm orifices.
   c. If a large number of bubbles of small diameter are present there is more likely to be a significant effect on the results, but this is not true in all cases.

3) **Can the bubble(s) be removed with minimal impact to the test?**
   a. This depends significantly on how the equipment is setup. Can the cell be moved without spilling the contents of the donor or receptor chamber? If so, can the bubble be moved (tapping usually dislodges them) and guided up the arm? In most cases a momentary loss of stirring has a minimal effect on the results.
   b. If the bubble can't be removed, it will be up to the operator to determine if the data is still worth analyzing. Historical observations with the product being tested can be very beneficial in making this determination. If none exist, then it is usually better to analyze the results with the understanding the presence of a bubble or bubbles may have affected it.

Conclusion: Bubbles covering part or all of a membrane during a diffusion test have the potential render a test useless. By making observations during the test, eliminating bubbles when possible, and deaerating receptor media test results are likely to be more consistent and provide better data.

For more information, or if you have any questions - please email us at: **support@permegear.com**