Hydration Analysis and the Pharmaceutical Scanning Electron Microscope

a report by
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Pharmaceutical enterprises require a range of imaging products that provide high-quality information that allows them to optimise their technology, productivity and, ultimately, profitability. With the increasing expectations upon drug delivery systems for efficient and controlled delivery of the active material, there is an equivalent need for analytical tools to provide accurate information on these mechanisms. For 30 years, one of the most effective instruments in this area has been the scanning electron microscope (SEM) owing to its unique ability to provide high-resolution images of specimens under investigation. Complementing the optical microscopes, the SEM continues to be called upon to illuminate the microscopic world of drug discovery and delivery.

Soon after the introduction of the SEM, the analysis of X-rays emitted from the specimen added elemental analysis to the problem-solving abilities of the instrument. Another period of transition has now arrived with the introduction of water vapour to the environment around the specimen. Hydration analysis can now be added to the SEM toolkit. The SEM can now be used in a pressure regime, in which even liquid water can be condensed onto materials of interest. In the context of SEM operations, this pressure regime is known as extended variable pressure. The LEO EVO series of SEMs allow three imaging modes in a single analytical tool. From conventional high vacuum imaging, extended variable pressure imaging and extended pressure imaging, these instruments provide a tremendous range of analytical solutions in fields as diverse as pharmaceutical, life-sciences, semi-conductor and materials.

Instrumentation

In the SEM, a highly focused electron beam is digitally scanned across the specimen and one of several possible responses of the specimen (e.g. the emitted secondary electron current) is measured and presented as a grey scale image. With a very high depth of focus and a large range of magnifications up to and beyond 100,000 times, the modern SEM provides an easy-to-use and productive tool for both elemental analysis and hydration analysis of materials. The output from an elemental analysis is the elemental composition at a point of interest on the specimen. The output from an hydration analysis of a specimen is a record of the interaction of water with the specimen.

Water Vapour and the SEM

If a wet or hydrated specimen is introduced into a previous-generation SEM, the vacuum environment leads to a loss of water from the specimen. This loss will probably lead to changes in microstructure and result in images containing dehydration artefacts. However, if water vapour is deliberately introduced into the chamber of the microscope, a cool, wet specimen can be maintained in a fully hydrated state. The loss of water from the surface is then balanced by the rate of arrival of water at the surface. This steady state, or dynamic balance, maintains the microstructure of the specimen.

At higher water vapour pressures, with the specimen at the same temperature, the loss of water from the surface is inferior to the rate of arrival of water. The dynamic balance is not maintained and liquid water forms on the specimen, leading to the opportunity for hydration analysis.

Figure 1: The LEO EVO 40 SEM

Figure 2a: Fragment of an Aspirin Tablet, Dry

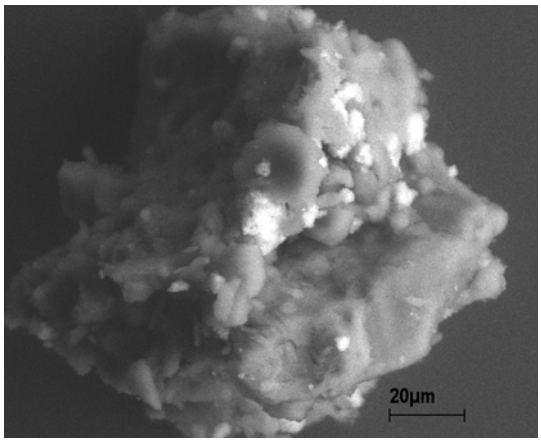
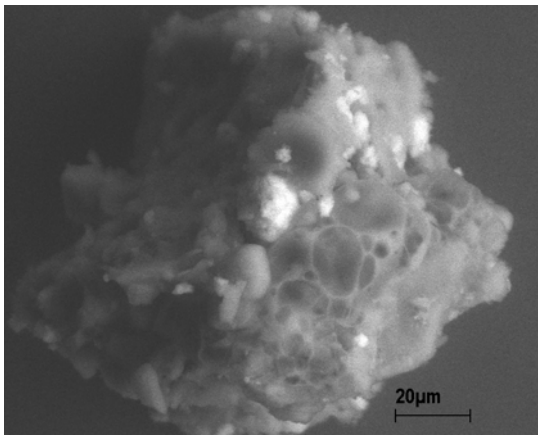


Figure 2b: Fragment as Water Begins to Condense



Hydration and Dehydration of Soluble Aspirin

The interaction of water with soluble aspirin demonstrates the mechanisms by which tablets lose mechanical strength and stability and then release the active material. This process can be observed and recorded in real time in an SEM able to perform hydration analysis. During the wetting phase, the particle absorbs water and fragments. During the drying phase, the reverse processes can be followed if required. In a typical application, the fragments of the tablet are cooled to just above freezing point using a Peltier cooled stage. By cooling the specimen, the water vapour pressure needed to yield liquid water on the specimen is reduced from about 2,400 Pascals at room temperature to 600 Pascals at 0°C. This reduction in pressure helps to achieve clear images.

Images from the hydration analysis of aspirin (see *Figures 2a–2d*) are presented here to illustrate the process. *Figure 2a* shows a fragment of a dry aspirin tablet. *Figure 2b* shows the same sample at the moment when water is condensing onto the aspirin. Some bubble formation is observed. This is interesting in that it shows that a source of gas has been created, which has lifted the surface water film into the spherical shape that we recognise as a bubble. *Figure 2c* shows the tablet in a pool of liquid allowing the active constituents to dissolve. *Figure 2d* shows the same fragment after the water has been removed, causing the specimen to dehydrate.

SEMs continue to form a core analytical tool in the pharmaceutical industry, owing to their capability for high-resolution imaging and the emerging possibilities for hydration analysis with materials. Present and future developments at LEO Electron Microscopy will continue to serve pharmaceutical enterprises with tools that speed up the development cycle by providing mission-critical information on microstructure, composition and morphology. ■

Figure 2c: Fragment in a Pool of Liquid, Allowing the Active Constituents to Dissolve

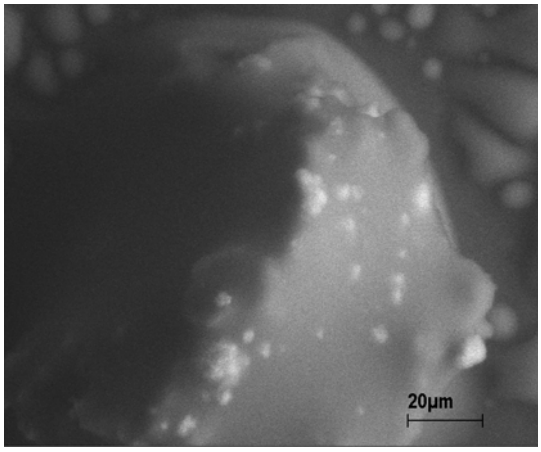
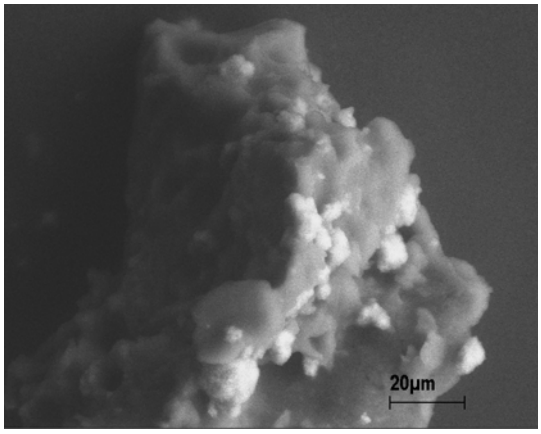


Figure 2d: Fragment after Drying



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