Application Note

Biology
Life Sciences

XGT

XGT02

Elemental micro-analysis of leaves using EDXRF

X-ray fluorescence (XRF) provides fast characterisation of sample element composition down to parts per million (ppm) levels – however, biologists often ignore this useful technique because it traditionally requires destructive vacuum conditions. Bio-samples containing large amounts of water will quickly dehydrate under these conditions. With the groundbreaking XGT-5000, though, these concerns are removed. Samples are analysed at normal atmospheric pressure, so that the resulting qualitative/quantitative data truly represents a healthy sample. In this application note the unique 10 µm spatial resolution of the XGT-5000 coupled with atmospheric pressure analysis allows leaf structure and behaviour to be explored. By using a combination of single point analysis and mapped imaging it is possible to learn more about the growth of leaves, and their behaviour in the presence of pollutants.

Calcium modules in Mulberry leaves

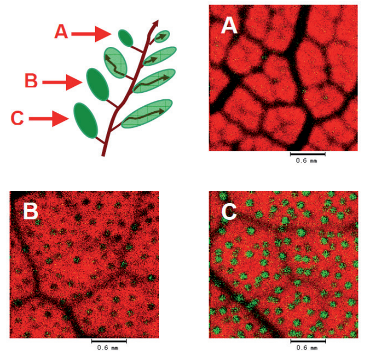


Figure 1: Transmitted x-ray and XRF composite images (Transmission + Ca) acquired from mulberry leaves in different growth stages: (A) young, (B) middle aged, and (C) old.

Mulberry plants are widely distributed across the world, with species native in many countries including America and China. The primary use of mulberry plants is as a food source for silk worms, but their fruit is becoming more popular, and bottled mulberry fruit juice now widespread in Asia. The fruit also has uses within Chinese herbal medicine.

Analysis of leaves taken from a mulberry plant illustrate how the main leaf structure can be imaged using transmitted x-rays, whilst the elemental information of XRF identifies the accumulation of calcium containing nodules (Figure 1). These nodules, typically in the order of 50-150 µm in diameter, are virtually non-existent in new leaf growth, but analysis of older leaves in the plant illustrates a dramatic increase in their concentration.

Plants often evolve strategies to defend themselves against herbivores, and it is possible that mineral incorporation in mulberry leaves could be just such a mechanism against feeding by silkworms. Micro¬analysis of the type described here provides useful information for plant scientists about the nature and structure of calcium accumulation in mulberry plants.

Pollutant uptake in plants

Since the industrial revolution, pollution has become a serious problem in the world, and the known health risks of heavy metals such as lead and cadmium is a cause for alarm.

An emerging method to cope with such pollution is phytoremediation, which uses plants to naturally take up and accumulate the heavy metals within the soil. With the heavy metals fixed within the plant foliage, the plants are then harvested and disposed of in a suitable manner, leaving the soil clean and safe.

Investigation into this forward thinking solution requires fundamental research into the pathways of pollutants within plants, and the XGT-5000 is ideally suited for this. The leaves can be analysed non¬destructively for element concentration and distribution, thus allowing the uptake pathways to be fully characterised.

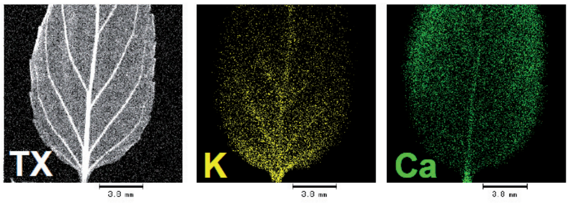


Figure 2: Analysis of leaf prior to introduction of pollutant to root system. Transmitted x-ray (TX), potassium (K) and calcium (Ca) mapped images are shown.

Preliminary results (Figure 2) illustrate how plant leaves can be quickly and easily analysed for element distribution, whilst the additional information provided by simultaneous transmission x-ray analysis picks out the major leaf veins.

A 1% lead solution was introduced to the root system of a living plant - after time for uptake leaves were again analysed with the XGT-5000 (Figure 3). The presence of lead within the veins is unambiguous, and shows the suitability of this technique for following heavy metal pathways in plants.

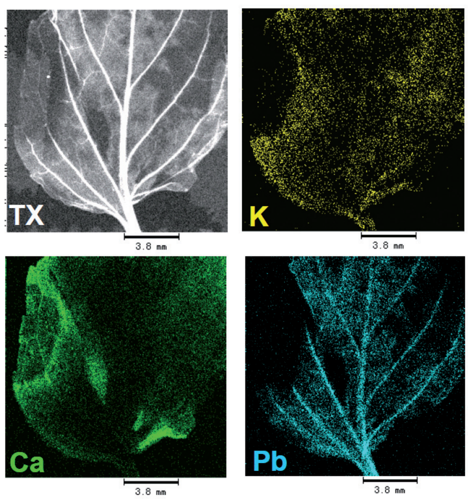


Figure 3: Analysis of leaf after introduction of pollutant to root system. Transmitted x-ray (TX), potassium (K), calcium (Ca) and lead (Pb) mapped images are shown.

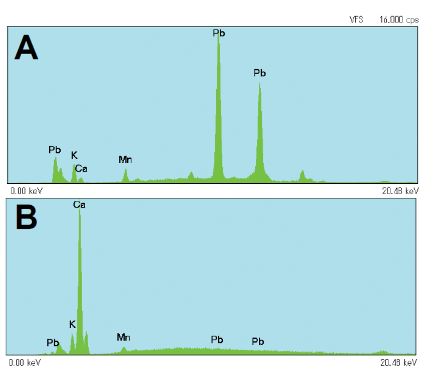
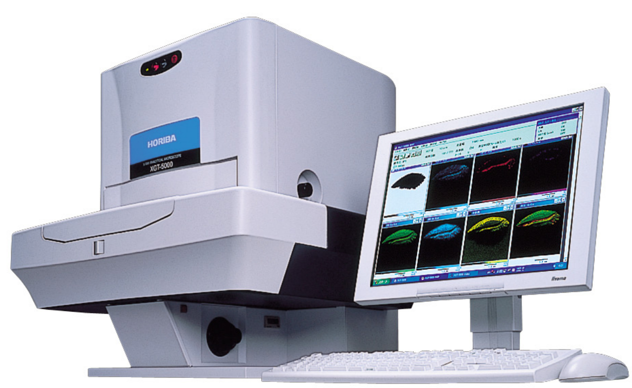


Figure 4: Spectra acquired from (A) high and (B) low lead concentration regions in the leaf shown in Figure 3.

Spectra shown in Figure 4 were acquired from high and low lead concentration regions within the leaf. The lead Lα and Lβ lines are clearly visible in the high concentration spectrum. With limits of detection below 0.1% for the heavy metals (depending upon x-ray beam diameter and sample), it would be possible for even trace amounts of pollutants to be analysed.

Conclusions

The non-destructive nature of micro-XRF analysis with the XGT systems opens this field of elemental analysis to biological applications. The capability for large area, high spatial resolution analysis means that even whole leaves can be analysed – without needing a damaging vacuum.



XGT-5000

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