A Q&A

Pesticide Residues in Fruits and Vegetables

Amadeo Fernandez-Alba, Ph.D.

Detecting and quantifying pesticide residues in fruits and vegetables is a crucial public health function for analytical laboratories throughout the world. Professor Amadeo Fernàndez-Alba, a full professor in Analytical Chemistry at the University of Almeria, Spain, has headed the European Reference Laboratory for Pesticide Residues in Fruits and Vegetables (EURL-FV) since 2006. Professor Fernàndez-Alba's EURL responsibilities include building and coordinating the network of official laboratories. He also serves on many national and international committees, and has contributed to 24-book chapters and written or co-written 244 scientific publications.

LCGC: Can you please explain the main functions of the EURL-FV and its role in the pesticides community?

Fernàndez-Alba: The main activities of the European Reference Laboratory are to improve the quality of the pesticide residue results from all of the official laboratories here in Europe. And second, to harmonize the results – it means that the results in different laboratories will be very similar if not equal.

The quality improvement is especially focused on two aspects. One is in scope, to cover as many residues as possible in a single analysis, and the second is sensitivity. That means to improve the sensitivity and to try to reach the low detection limits required by the regulations.

LCGC: You edit the European SANTE guidelines. Can you give a brief overview of the purpose and influence of that document?

Fernàndez-Alba: Well, the guidelines started in 1996 and it's a common responsibility of 12 scientific panelists, very expert people that know practically the whole history of these guidelines. And these guidelines are coordinated by the four EURLs for pesticide residues together with the National Reference Laboratory in Sweden.

The main activities are to update the guidelines, to take into account advancements in the instrumentation, the development of new methods and changes in activities. Currently we update these guidelines every two years in order to incorporate all the new activities and new analytical instrument platforms that appear in the market.

LCGC: Those guidelines are used throughout Europe?

Fernàndez-Alba: Yes. Well, the guidelines, they are compulsory or mandatory for all the official laboratories in Europe. But the guidelines are used in other laboratories and also in non-EU countries. For us, it's a very good point for discussion when we visit laboratories in non-EU countries like in Kenya or China etc. We can see clearly how they appreciate the document because it is seen as one set of international guidelines that is very useful for all laboratories all over the world.

LCGC: Turning to specific techniques, LC triple quadrupole has been the gold standard for quantitation and identification of pesticides, and it's accepted by regulatory authorities. Do you think high-resolution mass spectrometry can provide equally valid data and become equally accepted?

Fernàndez-Alba: Yes, absolutely. We can often see a very similar performance in quantitation, especially with the latest high-resolution mass spectrometry platforms that have appeared in the market. That is probably the weak point for this type of instrumentation in the past, but with the new upgraded instruments, the quantitation is now very similar in analytical performance to the triple quads. And then HRMS instruments have some important advantages from the point of view of the identification capabilities, especially in the analysis of complex samples. These developments are very much appreciated by the laboratories.

LCGC: What do you see is the main benefits and limitations of high-resolution mass spectrometry compared to LC triple quadrupoles?

Fernàndez-Alba: Well, as I have commented before, probably for sure it's the higher capability for accurate identification as the high resolution can often distinguish compounds that are isobaric when using nominal mass. And when high resolution is combined with mass accuracy, typically below 5 ppm, then the identification capabilities are very impressive. This type of instrumentation is very important to increase the performance of the residue laboratories.

LCGC: In your view, how important are resolution and mass accuracy?

Fernàndez-Alba: Well, it is similar to what I have commented on with the identification. It's a key aspect especially nowadays when the laboratories start analyzing hundreds of different commodities with different complexity and containing different natural components. In addition, the laboratories are extending the scope of the methods and this is often close to 500-600 different pesticides, and therefore the total number of pesticide-commodity combinations can be challenging.

The difficulties (interferences) that can appear in the identification are really high. So it means that HRMS instrumentation is very, very important especially with respect to avoiding false positive detection, and especially false negative detections.

LCGC: Can you outline some specific scenarios where high-resolution accurate mass spectrometry may provide a really definitive advantage over LC triple quadrupole MS?

Fernàndez-Alba: We can speak about different possibilities. For example, with non-target analysis when we have to detect and identify compounds that we would not expect to be present in the sample. One of the benefits of these platforms is to work in full scan, a non-targeted acquisition. Therefore we can screen for unexpected residues against a library containing spectra and retention times for a large number of pesticides. At the same time we can simultaneously conduct targeted analysis for those pesticides expected to be present. The difference is that when we calibrate the system with standards we are then able to quantify the concentrations and identify the residues by comparison of fragment ion ratios against the standards.

Another advantages of fullscan techniques is the capability to detect transformation products which are formed when pesticides degrade during sample processing, extraction, chromatographic separation and detection. This is because it is possible to detect these compounds, even if standards are not available. It is very difficult to identify compounds without standards, but at least we will get some indication that certain pesticides are possibly being used.

Also, we have the possibility to work in analysis *a posteriori*: after seeing the analysis, we can return to the data (months later) and re-evaluate the raw data for the presence of a residue or contaminant that has since emerged and for which we had no awareness at the time of the analysis. This is possible because we collect all of the ion information all of the time.

And now, this instrumentation is becoming more and more important as the associated software better enables us to treat this big amount of data with special statistical techniques. This can be very useful for a range of different issues, not only pesticide residues, but for example; authenticity, food fraud, metabolomics, etc. The possibility to apply advanced statistical treatments is a very important consideration for implementation of these new technologies.

LCGC: What benefits do you think Orbitrap LC-MS brings to the pesticide community?

Fernàndez-Alba: Well, Orbitrap technology is one of the possibilities for the high-resolution mass spectrometry together with time of flight. But it's a very interesting approach because from my experience with this type of instrumentation, the Orbitrap LC-MS, the resolution that we can achieve is quite high, in some cases it's 100,000 or even higher. So, it can facilitate or improve all these benefits that I have commented on before.

And also, the second benefit is its robustness. The design is a very specific design for high-resolution. The robustness of this instrumentation is important. And working in routine laboratories, robustness is one of the main issues to consider.