The fascinating history of the development of LC-MS; a personal perspective

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Today LC-MS is considered as a commonplace analytical tool that has been around for years. Most users do not give a second thought to the history of the development of this fascinating technique. It is twenty years since the first commercial instrument dedicated to atmospheric pressure ionisation (API) was produced by a small and then unknown company called Sciex, and so now is probably a good time to review some of the significant challenges that were faced to develop the technique. I would like to take you through some of the pioneering work that was carried out in this area, and discuss some of the exotic approaches that were taken by the pioneers of LC-MS prior to the API breakthrough.

I have been working in mass spectrometry for nearly 40 years, and I consider myself fortunate to have grown up during the various stages in the development of LC-MS. What I have attempted to do in this article is give you a personal view of what I consider some of the landmarks on the road to achieving the perfect interface between LC and MS and how that technique plays a major part in the analytical chemists daily life.

Let me take you back to 1968 when the first ever attempt to interface these two apparently incompatible techniques together was published in the Russian Journal of Physical Chemistry by Victor Tal’rose [1]. This was a landmark publication for its time, because it was the first published attempt to connect LC to MS. They managed to spray a very small amount of liquid into a conventional high voltage electron impact mass spectrometer. This was no mean feat, as the ionisation source in the mass spectrometer needed to be at high vacuum (10⁻⁷ torr). Liquid produces a lot of gas as the pressure is reduced, and so this was seen by the mass spectrometry community as an incredible feat, and lots of excitement was generated around this direct liquid introduction (DLI) approach. However, it was soon realised that electron impact, due to its inability to deal with pressures in excess of 10⁻⁶ Torr, was not going to be a practical approach and interest waned. In 1973 Baldwin and McLaффerty recognised that this approach could be viable if the liquid was sprayed into a chemical ionisation source as the amount of liquid entering the mass spectrometer could be increased [2]. They developed a DLI LC-MS interface (Figure 1), which was more robust and was capable of generating a stable ion beam in the mass spectrometer, but the liquid flow rate was still very low.

Even with its limitations, this approach gained enough interest that Hewlett Packard actually developed a commercial interface that was launched at the Pittsburgh Conference in 1979 [3]. Some ground breaking work applying this technique to real problems were published by some eminent chromatographers such as Patrick Arpino and Jack Henion [4,5]. At the same time other research groups were approaching this interfacing problem from different directions, Horning with Dziric and Carrol in 1975 [6], were the first people to develop an atmospheric pressure source, firstly using radioactive Nickel and then subsequently corona discharge interfaced to a mass spectrometer (Figure 2). This interface showed promise, but the spectra were complex due to the presence of cluster ions and so this concept was not really pursued seriously at the time.
In the USA, McFadden et al. [7] were approaching the solvent issue in a completely different way. They wanted to remove the solvent before the sample entered the mass spectrometer, and designed a moving belt interface which comprised of a stainless steel belt, onto which the LC eluent was deposited. The belt then passed under some infra red heaters to evaporate the solvent and then through a complex series of vacuum locks prior to the belt entering a conventional EI/CI mass spectrometer source (Figure 3).

This approach was also developed as a commercial system. The first was developed commercially by Finnigan as an LC-MS interface to their quadrupole instruments, and this was followed by a variation developed by Vacuum Generators who used a polyimide belt instead of the stainless one, so that this approach could be used on high voltage double focussing mass spectrometers [8]. This was later further modified to be used in conjunction with Fast Atom Bombardment ion sources [9]. Around this time some interesting research was being carried out by Thomson and Iribarne [10] who were looking into the fate of charges in evaporating cloud droplets, but, just like Hornings work, this was not viewed with great interest, especially because Vestal had just published his early work on a new and exciting LC-MS interface called Thermospray [11].

This interface took the LC-MS community by storm. For the first time the MS and chromatography communities had an interface that could accommodate reversed phase solvent systems. It did have some significant limitations, in that you could not use phosphate buffer, and for ionisation to occur you needed to have ammonium acetate present, but it was the first real LC-MS approach which addressed many of the desires of the chromatographer. This approach was commercialised by all the main mass spectrometry manufacturers of the time, and the technique was fully embraced by the pharmaceutical industry. Everyone within the mass spectrometry and the chromatography communities were ‘over the moon’ because, for the first time, we had a working LC-MS approach which was capable of dealing with reversed phase solvent systems and which was complimentary with LC-UV detection (although the sensitivity was not as good). Other interfaces also popped up around this time; one was the particle beam interface [12] in 1984, and the other was flowing FAB [13] in 1985, but they only had limited impact on the community because Thermospray had become the technique of choice. The complexities of the interface and the limitations that the technique imposed on the LC solvent composition however, meant that the technique remained predominantly in the hand of the mass spectrometry community.

However, a number of minor events and developments were happening in tandem across the world which, when brought together, would change the LC-MS landscape completely. It is not often in science, that events coincide in this way, but this is an example one of those wonderful periods. So let me try to catalogue the events. Firstly, we find that Jack Henion (of DLI fame circa 1978) who was still working in the veterinary medicine arena at Cornell University, linked up with Bruce Thomson (of ion evaporation fame circa 1978) who was then working for a small mass spectrometry company called Sciex in Toronto who built air monitoring mass spectrometers. They published a seminal paper in Analytical Chemistry titled “Determination of sulfa drugs in biological fluids by LC-MSMS” [14] which used atmospheric pressure chemical ionisation in front of the Sciex mass spectrometer. This approach to LC-MS demonstrated incredible sensitivity at trace levels. This was quickly followed by improvements in the design of the APCI source and further papers by Henion et al. [15,16] soon after. Around the same time John Fenn at Yale University, had developed the first atmospheric pressure electrospray source based on the original work of Dole back in the 1960s [17], and he reported on the multicharging of large bioorganic molecules such as proteins (see Figure 5) [18].

So suddenly and by two independent research groups the world had been exposed to two techniques both using ionisation at atmospheric pressure, both showing incredible sensitivity, and both capable of linking to reverse phase LC. It was also fortunate that through Thomson the triangle was able to be completed, because he had access to an instrument company with a mass spectrometer which sampled at atmospheric pressure, so we had what could be called a marriage made in heaven. The first publications started appearing in the literature under the titles of ‘ion spray’ and ‘APCI’ [19] and all were demonstrating sensitivities at least one to two orders of magnitude greater than other techniques. This was particularly interesting to the drug metabolism industry and by the time Sciex launched their first instrument in 1989, the whole landscape of LC-MS changed irreversibly. This new breed of instruments were simple to use, had no real
Constraints on mobile phase composition or flow rate; the spectra were simple to interpret, as the protonated molecular ion predominated the spectrum (Figure 6), with virtually no fragmentation being detected, even proteins and their digests could be analysed. All of this meant that LC-MS was no longer wholly the domain of the mass spectrometrist and so biochemists, drug metabolism specialists and chromatographers, could all use these systems. In a very small space of time all the other mass spectrometer manufacturers follow suit and produced instruments exclusively dedicated to these API techniques. Without the amazing development of Atmospheric Pressure Ionisation we would not be where we are today. Somewhere in the pharmaceutical industry, mass directed purification was also a result of this development. This incredible sensitivity means that API LC-MS is almost exclusively used in the drug metabolism community to quantify metabolites at low levels. The technique has also revolutionised the detection of trace level contaminants in drug formulations. Outside the pharmaceutical industry, the technique is centre stage in the detection of trace levels of performance enhancing drug within the sports community, and in the horse racing arena.

Without it the life science community would not have progressed in the many ‘omic fields. The protein elucidation field would not have progressed to where it is today, for example the elucidation of large non covalently bound protein complexes is critically dependant on this approach. The list of API application areas is endless. It would not be an exaggeration to say that through the development of atmospheric pressure ionisation techniques, LC-MS has become a central mainstay within the analytical community. In my opinion it is probably the key tool in the analytical chemist’s toolbox, and one that we now take for granted.

Back in 1974, Patrick Arpino drew this famous cartoon (Figure 7), which showed how unlikely it was that two apparently incompatible techniques could end up happily married together, but, within 30 years it was achieved.

I will leave you with one thought before I finish. The next time that you run an LC-MS instrument, or sit down with others to review some LC-MS data, just spend a short time and reflect on all the groundbreaking work and all the heartache that went into the development of this technique by a few pioneers who had the dogged resolve to overcome what was seen at the time to be an insolvable problem.

Analytical Science is a fascinating discipline to be a part of, and this story is one that should be recognised as a real achievement of success against enormous odds.

References

Figure 6. LC-MS analysis of three sulphonamides; MS spectrum shows the predominant MH+ from peak 2 at 4.49 minutes.

Figure 7. LC and MS, will the two ever come together? Highlighting the incompatibility between liquid and gas phase (vacuum based) systems - Patrick Arpino 1974.