1987

A Simple Lecture Demonstration of Flow Injection Analysis

Julian Tyson
University of Massachusetts Amherst

A simple lecture demonstration of flow injection analysis

Julian Tyson

Department of Chemistry, University of Technology, Loughborough, Leicestershire, LE11 3TU, UK

Einfache Vorlesungsdemonstration der Fließinjektionsanalyse

Summary. The difficulties of explaining the basic concepts of dispersion in flow injection analysis are discussed with particular reference to comparisons with segmented flow analysis and high performance liquid chromatography. The problems with diagrammatic illustration of the laminar flow processes and subsequent dispersion of an injected sample zone are explained. A simple demonstration based on a flow cell for a slide projector consisting of a peristaltic pump tube sandwiched between two slightly modified slide mounts is described.

Introduction

Although the basic concept of flow injection analysis (FIA) is simple to explain, it is difficult to convey an accurate impression of the basic dispersion processes to an audience meeting the concepts for the first time in a lecture theatre. Such audiences will consist of students at various stages of undergraduate or post-graduate training or will consist of practising analytical chemists who may already have experience of segmented flow analysis (SFA) and/or high performance liquid chromatography (HPLC). This latter audience may be the most difficult to convey an accurate description of flow injection dispersion to, as there is a tendency amongst chemists meeting FIA for the first time to get the impression that FIA is either "SFA without the bubbles" or "HPLC without the column" or "HPLC with post-column derivatisation without the column". Indeed, such descriptions of FIA may be proposed by lecturers themselves.

The SFA analogy

Such descriptions, however, are quite inappropriate for conveying the basic ideas of dispersion mechanisms in FIA, although there might be some merit in the descriptions as far as the equipment for FIA is concerned. The concept of dispersion as a parameter exploitable for analytical purposes does not really arise in SFA methods. With these techniques [1] sufficient sample is introduced into the flow line to produce a flat topped "peak" (i.e., a steady state response is obtained). The reagent is added at a confluence point and each sample is segmented into about 15 sub-sample zones by the addition of air prior to the reagent addition. Mixing between sample and reagent is obtained by the circulation induced in each liquid segment from the walls to the centre (so-called "bolus flow"). This circulation is considerably aided by wetting of the tube walls by the liquid. In order to promote this wetting, glass tubes are used (with diameters up to 2.5 mm) and a surfactant is often added to sample solutions (which also improves the bubble pattern as a result of the reduced surface tension of the water). Complete mixing in each segment is also promoted by repeated inversion of the segments as the tubing consists of coils in the vertical plane. Typically the 15 sample segments will each occupy about 14 mm of tubing. If the typical length of a bubble is 5 mm, the entire sample will occupy a length of approximately 280 mm when travelling through the manifold.

The HPLC analogy

In contrast, the concept of dispersion is very much a key parameter in HPLC. However, as a rule, an HPLC separation is designed to achieve separation between the components of the sample mixture and thus dispersion is considered only in terms of its effects on peak width. The factors which control the extent of this dispersion [2] are mainly those which arise from processes within the column such as (a) tortuous flow between particles of stationary phase (b) axial diffusion and (c) slow equilibration between mobile and stationary zones. The system will be designed to minimise extra-column broadening effects by use of the minimum lengths of tubing of diameters $\leqslant 0.2$ mm. Sample volumes may be small (compared with the typical flow injection procedure) and it may thus be difficult for a chromatographer to visualise what would be the effect of removing the column from such a system. The inclusion of post-column derivatisation is possibly a more useful analogy as the system will have been designed to produce mixing without band broadening (by the use of low volume mixing chambers or by flow effects due to tightly coiled or other physically contorted tubing patterns). Neither of these two mechanisms is appropriate for the basis of an explanation of the basic dispersion mechanisms in FIA. Chromatographers do make use of a dispersion model (the axially dispersed plug flow model [3]) for calculating the extra broadening effects due to connecting tubing, which does have more relevance to flow injection dispersion mechanisms. The model is always used in a form which allows the calculations of an additional contribution to the width of a Gaussian shaped peak in volume units, and thus it is doubtful whether analytical chemists familiar with this expression

could readily visualise its application to a typical flow injection experiment. This is even more unlikely when dispersion in FIA is formulated in terms of the extent of dilution at the peak maximum rather than the width of the peak. Furthermore, the model is only formulated for inputs to the flow system of zero initial width (the so-called "delta" function). Clearly the chromatography model is not readily applied to FIA [4].

Visual aids

What is needed, therefore, for an introductory lecture on FIA is an appropriate visual aid. Although diagrams of the longitudinal cross-section of the tube would appear to be the most suitable approach, such diagrams suffer from a number of limitations. The first of these concerns representation of the differences in concentration at different locations in the tube. Whereas the initial conditions are easy to represent when the undispersed sample slug has just been intercalated into the flowing stream (time, $t = 0$), the situation at $t > 0$ requires skillful shading if tone or depth of colour is to be used to represent concentrations. The second of these limitations concerns the space available for the display of such a diagram. A typical slide might show a suitably enlarged version of Fig. 1, and indeed pictures such as this are often used in text books, papers and manufacturers' literature. However, a few simple calculations show that such pictures are seriously misleading and convey a false impression of the shape of the dispersed sample zone. Considering a single line manifold with tubing of internal diameter 0.5 mm, volume injected 50 µl and flow rate of 1 ml min^{-1} as typical of a basic FIA system, it is readily calculated that the length of tubing occupied by sample at $t = 0$ is 254 mm. Thus, whatever scale is chosen to represent the tubing, the length of the initial sample zone should be about 500× the "height" (representing the internal diameter). If the original 'art work' for such as slide were to represent the initial sample zone by a length of 25 cm, then the tube diameter would be represented by a distance of 0.5 mm (i.e., the dimensions of the real situation). Clearly this is not possible. Alternatively it would be considered that the volume represented in Fig. 1 is 1 µl; hardly typical of flow injection procedures. A further limitation becomes apparent when the linear flow velocity is calculated. The average linear flow rate is 85 mm s^{-1} and an explanation of dispersion mechanisms which started with the formation of concentration gradients due to laminar flow, leads to the central stream-line flowing at twice the average linear velocity, i.e., 170 mm s^{-1}. Thus the position shown in Fig. 1 b shows the distortion of the initial sharp boundary that would occur after the first 0.015 s. Again, such diagrams are of limited use when describing the processes which occur during several seconds residence time in the system.

One possible approach is to use different scales for the width and length of the tubing. Some very elegant diagrams have recently been produced by Vanderslice et al. [5] which are different tones of grey to represent different concentrations. Distances are scaled in reduced units of length and thus to the uninitiated eye, the bolus shapes so produced might well reinforce the impression that the sample zone really has a compact "hollow-bullet" shape.

To give a more accurate impression of the processes occurring during passage from an injector to detector and

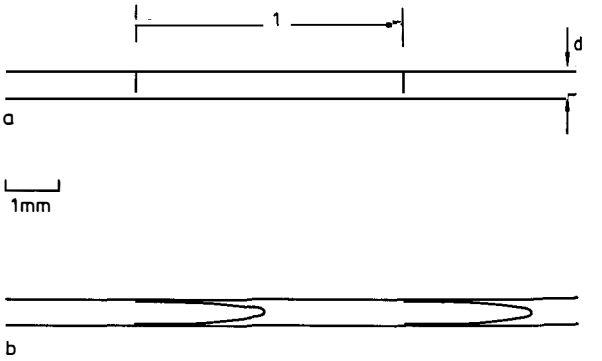

Fig. 1 a, b. a Typical representation of the start of a FIA experiment. The tube internal diameter, *d*, is 0.5 mm, thus the length, *l*, of sample solution of 5 mm corresponds to an injection volume of 1 µl. **b** Typical representation of the formation of a flow injection bolus shape. At a flow rate of 1 ml min^{-1} the picture shown corresponds to 0.015 s of pure laminar flow

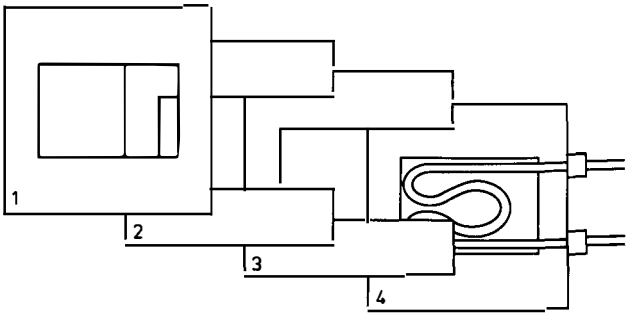

Fig. 2. Construction of demonstration cell. *1* Unmodified half of slide mount, *2* modified (part of side removed) half of slide mount glued to, *3* similarly modified half of second slide mount. *4* unmodified half of second slide mount showing positions of peristaltic pump tube when all four halves mounted together. Halves *1* and *4* still have their glass windows, whereas these have been removed for halves *2* and *3*

to show the shape of the peaks produced a simple demonstration cell for a slide projector has been devised. This may be used in conjunction with any suitable flow injection manifold and chemistry (no chemistry is, of course, needed to illustrate physical dispersion). For the single line manifold the carrier stream may be delivered from a disposable plastic syringe. This is particularly convenient as it avoids the need to make arrangements for an electrically driven pumping system. It also demonstrates to the operator just how little back pressure a typical flow injection system produces. Flow rates can be readily judged by observing the rate of drops produced at the end of the waste tubing (as a rough guide, the number of drops in 15 s divided by 10 gives the numerical value of the flow rate in ml min^{-1}).

The cell is constructed (see Fig. 2) from two slide mounts of the type which consist of two halves each with a thin glass window. In normal operation the two halves are kept together by a simple "male-female" friction fastening. The two mounts are glued together after removing the "middle" two windows and cutting an access slot in the side. A transparent (standard PVC) peristaltic pump tube is "snaked"

between the windows to provide the transparent optical path. Suitable tubing is that colour coded with orange, white or red shoulders (internal diameters 0.89, 1.02 and 1.14 mm respectively) with which simple push fit connections may be made with the flow lines from the manifold and to waste. As the "path length" of the cell is effectively the internal diameter of tubing, the concentrations of absorbing species need to be correspondingly increased. There is no difficulty in seeing the development of a colour with the standard chloride reagent [mercury(II)thiocyanate, acid and Fe^{3+}] and a sample containing several hundred ppm chloride. Acid-base indicators at elevated concentrations provide particularly striking effects. Bromothymol blue at a concentration of about 10^{-3} M (saturated) is very suitable. Reagents containing large amounts of organic solvents should be avoided as these degas rather readily.

Although, as discussed above, the linear flow velocity is high the parabolic concentration profiles can be clearly seen and it is possible to stop the flow and examine the pattern produced in detail. The relative slowness of diffusion as a dispersion mechanism compared with convection is readily "observed".

Conclusion

A demonstration, for lecture theatre presentations, of the basic dispersion effects in FIA has been devised. Even experienced flow injection practitioners may be surprised by what can be seen with such a simple piece of apparatus. As Karlberg has remarked [6] "Flow injection analysis should not be explained. It ought to be demonstrated."

References

1. Coakley WA (1981) Handbook of automated analysis continuous flow techniques. Marcel Dekker, New York
2. Knox JH (ed) (1980) High-performance liquid chromatography. Edinburgh University Press, Edinburgh
3. Levenspiel O (1972) Chemical reaction engineering, chap 9, 2nd edn. Wiley New, York
4. Stone DC, Tyson JF (1987) Analyst 112:515–521
5. Vanderslice JT, Rosenfeld AG, Beecher GR (1986) Anal Chim Acta 179:119–129
6. Karlberg B (1981) In: Ruzicka J, Hansen EH (eds) Flow injection analysis. Wiley, New York

Received June 20, 1987