

Table 8 (*continued*)

Analyte	Reagents	$\lambda$ , nm	Detection limit	Reference
Dansylalanine	Bis(2,4-dinitrophenyl)- oxalate, H <sub>2</sub> O <sub>2</sub>		0.5 nM	405
Fe(II)	Luminol, H <sub>2</sub> O <sub>2</sub>		0.1 pM	389
Fe(II), total Fe	Brillant sulphoflavin, H <sub>2</sub> O <sub>2</sub>		0.45 nM	397
H <sub>2</sub> O <sub>2</sub>	Luminol, Co(II)		10 nM	382
	Luminol, Co(II)	440	5 nM	383
	1,1'-oxalyldiimidazole, 3- aminofluoranthene		10 nM	395
	Bis(2,4,6-trichlorophenyl) oxalate, perylene		6 nM	396
Hydrazine	Hypochlorite, Ni(II)		0.5 nM	406
Morphine	Permanganate, tetraphosphate	430	0.1 nM	407
Nitrate	Luminol		70 nM	389
Nitrate, nitrite	TI(III), iodide, ozone		0.7 ppb (NO <sub>3</sub> <sup>-</sup> ) 0.35 ppb (NO <sub>2</sub> <sup>-</sup> )	393
Nitrite	Luminol, H <sub>2</sub> O <sub>2</sub>	454	1 nM	390
Proteins	Luminol, Cu(II), H <sub>2</sub> O <sub>2</sub>		0.1 ppm	392
Pyrogallol	Periodate, hydroxylamine	455	0.1 $\mu$ M	408
Steroids	Lucigenin			398
	Ce(IV), sulphite		13–19 ppm	409
Sugars	Lucigenin, periodate		1 $\mu$ M (glucose)	398
Sulphide	Fluoresceine, hypochlorite	520	40 ppb	384
Sulphite	Permanganate, riboflavin phosphate	510	90 ppb	410
Surfactants, non-ionic	Hypochlorite, rhodamine B		3 ppm	411

## 6. Other Molecular Spectroscopic Detection Methods

The advantages of flow injection methodology has led to its widespread application in analytical procedures involving less common spectroscopic

detection methods. These adaptations usually result in the favourable mechanisation of determinations, better precision and a higher sampling rate.

The determination of sulphur anions based on their  $S_2$  emission peaks has been achieved using a conventional atomic absorption spectrometer with flame atomisation, operated in the emission mode [412]. The spectrometer is equipped with a specially designed sample holder support device and a circular emission burner. The water-cooled steel cavity is continuously situated within the flame and the sample is introduced into it with a carrier stream of water or hydrogen peroxide. Using a  $3 \mu\text{l}$  sample volume, sulphide, sulphite and sulphate can be determined with detection limits between 20 and 50 ppb of sulphur. The sequential appearance of  $S_2$  emission peaks due to the different thermal stability of the particular sulphur anions allows the resolution of the mixture of anions within 20 s.

Refractive index measurements are very commonly used to characterise chemical species or to evaluate the composition of binary liquid or gaseous mixtures. The concentration gradient of the sample in the solvent, produced during injection, creates a refractive index gradient, which can be used for detection in FIA measurements, similar to the detection method in liquid chromatography, which is based on a refractive index gradient produced by retention processes. Fundamentally, the refractive index of a substance is determined by measuring the change in direction of collimated radiation as it passes from one solution to another. For application as a detection method in flowing streams, a conventional spectrophotometer is used and a light beam deflection (Schlieren optics) is utilised. The signal obtained is dependent on both the magnitude of the concentration change and the distance over which change occurs. When the sample zone passes through a flow cell, the light is focussed into the centre of the detection cell and then dispersed from the centre. The detector registers an absorbance decrease followed by an increase, which results in the appearance of a double peak. The height of both peaks is a function of the analyte concentration in the injected sample. Normal FIA systems with sample injection are usually employed, but it has been demonstrated that the linear working range is narrower compared with the reversed FIA method [413]. This kind of detection has been employed in the determination of glycerol/water mixtures [414], the determination of glycol in deicing/antiicing fluids [413], and the determination of sucrose [415]. In the latter case, a fibre-optics-based detector is employed with an LED as the source of radiation.

Several applications in FIA systems have been reported for optical rotation detection, commonly called polarimetry. Optical rotation measurements

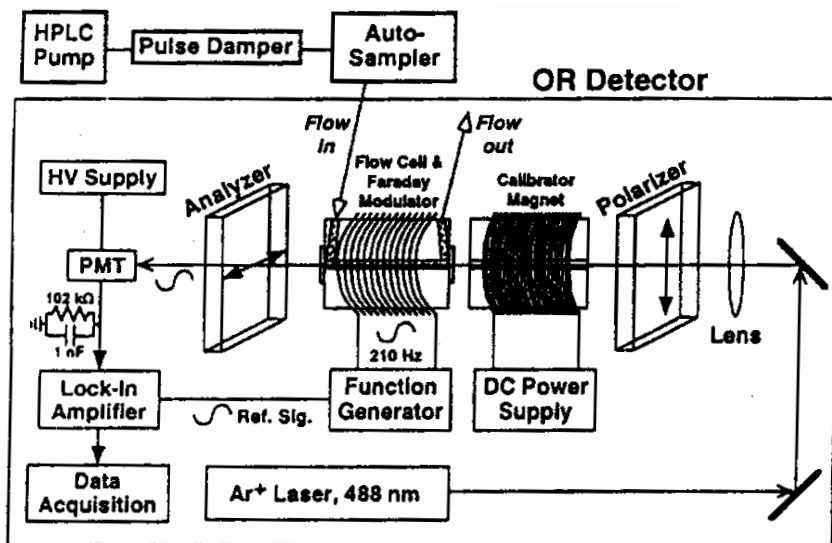


Fig. 11. Schematic diagram of a flow injection system with optical rotation detection [418]. (Reprinted by permission of copyright owner.)

are essential for the characterisation of chiral molecules as this is the only physical property that distinguishes the individual enantiomers. Compared with conventional polarimetry, FIA systems with optical rotation detection significantly reduce sample requirements, lower the detection limits by about two orders of magnitude, and shorten the analysis time to 1-2 min. The first report of a laser-based micropolarimeter for an FIA system describes its use in determining specific rotations from peak height data [416]. A commercially available optical rotation detector, based on a diode laser operating at 820 nm, has been used to determine the amount and enantiomeric purity of a drug in dosage form [417]. The FIA system shown schematically in Fig. 11 gives a 100-fold increase in the sensitivity of optical rotation detection compared with a high-quality conventional polarimeter [418]. It has been employed in the detection of sucrose, for measurements of the specific rotation values of various organic molecules, and for the analysis of the enantiomeric purity of (1*S*, 2*R*)-(+)-ephedrine.

Raman spectroscopy, based on the shifts in wavelength (from that of the incident beam) of the radiation scattered by certain molecules, is considered

to be the complementary technique to infrared spectroscopy. An important advantage is that water does not cause interference, so Raman spectra can be obtained from aqueous solutions and this technique has been applied to the qualitative and quantitative analysis of inorganic, organic and biological systems. The intensity of the Raman signal is greatly enhanced at a rough surface, which is utilised in surface-enhanced Raman spectroscopy (SERS). Generally, this technique is used to study the interactions between molecules and a metal (usually silver) surface, but if the analyte can be reproducibly adsorbed and desorbed from the silver surface, SERS can be used for analytical purposes. There have already been several reports on the use of SERS as a detection method in FIA. The first applications have been developed for the detection of adsorbates such as *p*-aminobenzoic acid [419] and pararosaniline hydrochloride [420] on colloidal silver, although the adhesion of sol particles on the tubing wall is a difficult problem to overcome. When SERS is carried out at a silver electrode, an additional parameter that can be controlled is the potential applied to the electrode. This facilitates the control of adsorption and desorption of the analyte from the surface by the selection of a suitable polarisation potential. This approach has been used for the FIA detection of pyridine using a metallic silver disk electrode, which has to be mechanically polished before the beginning of a set of runs. Additionally, at the beginning of each injection, the surface is regenerated by a short oxidation-reduction cycle [421]. In order to obviate these steps, an *in situ* renewal of a silver substrate has been proposed [422]. Silver ions are introduced into the FIA system and electrodeposited on an inert glassy carbon electrode just before injection of the analyte. When the Raman spectrum of the adsorbate is recorded, the silver film is removed by anodic stripping, preparing the system for the next determination. Such a procedure has been applied in the determination of Fe(II) as its complex with 2,2'-bipyridine with a detection limit of 1 nM of Fe(II). Multicomponent determinations using this technique also seem to be possible.

## 7. References

1. K. Ueno, I. Imamura and K. L. Cheng, *Handbook of Organic Analytical Reagents* (CRC, Boca Raton, 1992).
2. E. B. Sandell and H. Onishi, *Colorimetric Determination of Traces of Metals* (Interscience, New York, 1978).
3. Z. Marczenko, *Separation and Spectrophotometric Determination of Elements* (Horwood, Chichester, 1985).