Talanta 102 (2012) 53–58

Contents lists available at SciVerse ScienceDirect

Talanta

journal homepage: www.elsevier.com/locate/talanta

A flow system with zone merging and zone trapping in the main reactor applied to spectrophotometric catalytic determination of cobalt in grasses

Taciana F. Gomes, Ana C.F. Vida, Milton K. Sasaki, Claudineia R. Silva, Valdemir A.F. Barros, Elias A.G. Zagatto*

Centre for Nuclear Energy in Agriculture, University of S. Paulo Box 96, Piracicaba 13400-970, Brazil

ARTICLE INFO

Available online 20 July 2012

Keywords:
Flow analysis
Spectrophotometry
Zone stopping
Zone trapping
Cobalt determination
Pasture

ABSTRACT

A flow system with zone merging and zone trapping in the main reactor was proposed. The sample and reagent inserted aliquots merge together and the resulting zone is directed towards a displaceable reactor inside which its most concentrated portion is trapped. After the pre-set TRAP period, the handled sample is released towards detection. A comparison with an analogous flow system exploiting zone stopping revealed the superior characteristics of sampling rate and system operation; moreover, the sample inserted volume plays little influence on sampling rate. The system was applied to the spectrophotometric determination of cobalt in pastures, and enhanced figures of merit (sampling rate $=18$ h^{-1}; peak height r.s.d. $=0.7\%$, detection limit $=0.046$ μg L^{-1} Co; reagent consumption $=330$ μg of Tiron per measurement; $98\% <$ recovery $< 114\%$) were estimated.

1. Introduction

Flow-based analytical procedures involving fast chemical reactions are generally characterized by enhanced figures of merit. Implementation of methods relying on relatively slow reactions can however be limited, as long sample residence time and/or reduced sample dispersion are usually needed.

Reducing flow rates permits an increase in the mean sample residence time without pronounced sample dispersion, but plays a deleterious effect in sampling rate [1]. Improving the characteristics of the flow system, e.g. designing more efficient reactors and/or exploiting segmented, mono-segmented or pulsed flows [2], is also relevant in the context. Another possibility is to halt the sample zone in the analytical path: as convection ceases during the incubation period, sample dispersion is hindered, allowing longer reaction times to be efficiently attained. To this end, zone stopping and zone trapping are worthwhile.

Zone stopping was originally implemented in a flow injection system with zone merging [3] and has been worldwide accepted. Zone trapping was proposed in relation to a flow injection system involving continuous reagent pumping [4]; its acceptance has been however not so expressive probably because of the high reagent consumption involved. The feasibility of designing a flow system with zone trapping and zone merging was recently demonstrated and applied to the spectrophotometric determination of nitrite in natural waters [5]. The performances of similar flow systems exploiting zone stopping or zone trapping were compared and the advantages of the flow system with zone trapping in terms of sampling rate, inserted sample volume and ease of operation became evident. It should be stressed that the handled sample was halted in the flow through cuvette, either by stopping the pump or by exploiting detector relocation.

There are however situations where the sample should be halted inside the main reactor as, e.g. when parallel reactors are needed for increasing sampling rate [6] or when the reactor is immersed in a thermostatic water-bath for heating purposes [7].

The aim of this work was therefore critically to compare zone stopping and zone trapping in the main reactor of a flow system with zone merging. As an application, the catalytic determination of cobalt in pastures using Tiron was selected. The method involves its oxidation by hydrogen peroxide in alkaline medium catalysed by Co(II) and spectrophotometric monitoring at 426 nm [8]. The method was implemented in an ordinary flow injection system [9], but sensitivity was limited because the mean sample residence time could not be increased at will. The drawback can be circumvented by taking advantage of zone halting.

Cobalt determination in pastures is needed for an efficient farm management. In spite of being required by some enzymatic systems and important for the synthesis of vitamin B_{12}, cobalt is not essential to plants. Thus, its deficiency in pastures does not affect the forage growth but may limit the milk production as it is essential to cattle [10]. The cobalt content in the plants is then an indicative of the need for adding cobalt to the fertilizers and/or to the livestock animal supplements.

---

* Corresponding author.
E-mail address: ezagatto@cena.usp.br (E.A.G. Zagatto).

0039-9140/$ - see front matter © 2012 Elsevier B.V. All rights reserved.
http://dx.doi.org/10.1016/j.talanta.2012.07.032

2. Experimental

2.1. Solutions

All solutions were prepared with chemicals of analytical-grade quality and deionised water (specific conductance $<0.1\ \mu S\ cm^{-1}$). The substances used for assessing selectivity towards iron and manganese ($>99.998\%$ w/w purity) were purchased from Alfa Aesar (Ward Hill, MA, USA).

The pasture samples were digested following a procedure similar to that used earlier [11] but involving 0.5 g of dried and grinded sample, 5.0 mL HNO_3, 1.3 mL $HClO_4$ and a final digest volume of 50.0 mL. The mean digest acidity was ca. 0.25 mol L^{-1} $HClO_4$.

The 10.00 mg L^{-1} Co stock solution was prepared by weighing 23.85 mg $CoSO_47H_2O$, dissolving it in about 450 mL of a 0.01 mol L^{-1} HNO_3 solution and filling the volume up to 500 mL with this acidic solution. Working standard solutions covering the 0.00–1.50 $\mu g\ L^{-1}$ Co(II) range were prepared in 0.25 mol L^{-1} $HClO_4$.

The sample and reagent carrier streams (C_S and C_{R1}, Fig. 1) were a 0.25 mol L^{-1} $HClO_4$ solution and water, respectively. The R_1 reagent was a 0.005 mol L^{-1} Tiron (disodium1,2-dihydroxybenzenedisulfonate) water solution; R_2 was a 0.2 mol L^{-1} pyrophosphate (based on $Na_4P_2O_710H_2O$) plus 5.0 mg L^{-1} Mn^{2+} (based on MnO_2) solution; R_3 was freshly prepared a 0.005 mol $L^{-1}H_2O_2$ solution and R_4 was a 2.0 mol L^{-1} NaOH solution.

2.2. Apparatus

A model IPC-4-V2.00 Ismatec peristaltic pump, a homemade injector–commutator similar to that used in earlier work [5] and a USB 2000 UV–vis Ocean Optics spectrophotometer with an Ultem Z-shaped flow cell (inner volume = 18 μL, optical path = 10 mm) were used. Wavelength and integration time were set as 426 nm and 80 ms. Sampling loops, coiled reactors and transmission lines were made of polyethylene tubing (i.d. = 0.8 mm) of the non-collapsible wall type, and PEEK connectors were used. The in- and out-let ends of the R_{C2} reactor were attached to the movable central portion of the injector–commutator (Fig. 1). For data acquisition and treatment, the control software provided by the spectrophotometer manufacturer was used.

2.3. Flow diagrams

The flow systems in Fig. 1 were designed as similar to each other as possible, and the main difference between them was the strategy for sample halting. Fig. 1, left, refers to the flow system exploiting zone trapping. When the injector–commutator remains in the specified position, the sample and the Tiron solutions are filling their corresponding sampling loops and the R_{C2} reactor is kept outside the analytical path. Switching the injector–commutator inserts the selected sample and reagent aliquots into their carrier streams and displaces the main reactor towards the analytical path. Immediately after sample insertion, the R_2 reagent is added. The sample and reagent zones merge together at the x confluence, and the other previously mixed reagents are further added (y confluence). Tiron oxidation by hydrogen peroxide in alkaline medium proceeds inside the following R_{C2} coiled reactor, yielding the Tiron semi-quinone coloured radical [8]. When the most concentrated portion of the handled sample is passing through the main reactor, the injector–commutator is switched back to position specified in Fig. 1. The sample portion inside the reactor is then trapped and next sample and reagent aliquots are selected. After the pre-selected TRAP period, the injector–commutator is switched again, inserting the next sample and Tiron solutions into their carrier streams and re-displacing the R_{C2} reactor to the analytical path. The sample trapped portion is then released and pushed through a 15 cm transmission line towards detection.

The flow system exploiting zone stopping (Fig. 1, right) is similarly operated, but involves switching pump off when the handled sample zone is passing through the R_{C2} reactor. The entire sample zone is then halted during the pre-set STOP period.

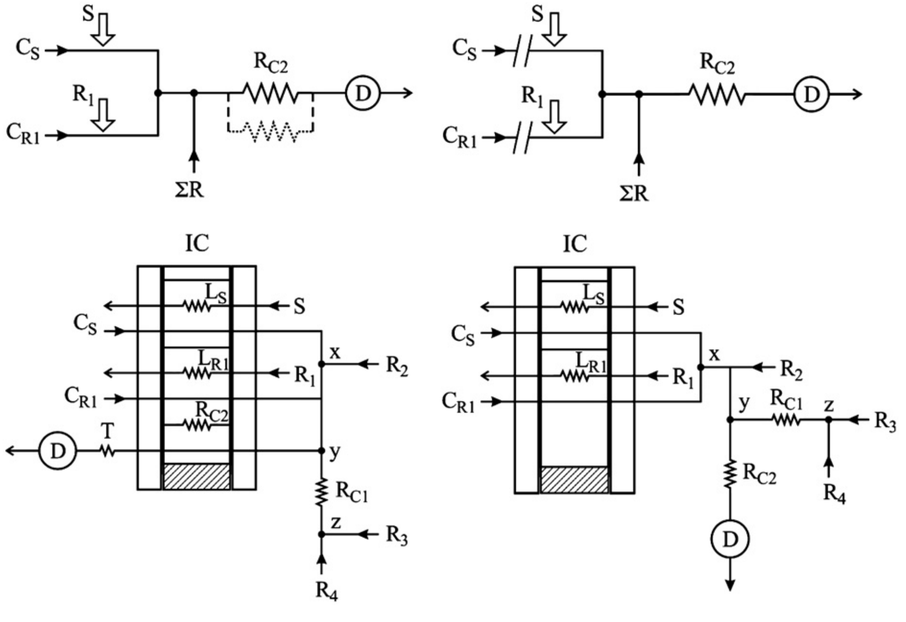

Fig. 1. Didactic representations (upper) and flow diagrams (lower) of the flow systems with zone trapping (left) and zone stopping (right). Empty arrows = sample/reagent injections; // = intermittent streams; dotted lines = alternative position of the R_{C2} reactor; black arrows = sites where pumping is applied; S and R_1 = sample and Tiron reagent solutions; C_S and C_{R1} = corresponding carrier streams (3.0 and 0.6 mL min^{-1}); L_S and L_{R1} = sampling loops (70 and 40 cm); IC = injector–commutator; dashed area = alternative IC position; R_2 = 0.2 mol L^{-1} $P_20_7^{4-}$ plus 5.0 mg L^{-1} Mn^{2+} at 0.6 mL min^{-1}; R_3 = 0.005 mol $L^{-1}H_20_2$ at 0.6 mL min^{-1}; R_4 = 2.0 mol L^{-1} NaOH at 0.6 mL min^{-1}; x, y and z = confluent sites; R_{C1} and R_{C2} = 100 cm and 50 cm coiled reactors; T = 10 cm transmission line; D = spectrophotometric flow cell (426 nm). For details, see text.

Thereafter, the pump is restarted thus directing the handled sample towards detection.

In both flow systems, the recorded peak height is considered as the measurement basis. The signal related to the blank solution reflects the development of uncatalyzed oxidation, whereas those related to the standard or sample solutions refer to Tiron catalysed plus uncatalyzed oxidation.

2.4. Procedure

A dye approach strategy was exploited in the experiments carried out for optimizing the overlap of the merging zones and for selecting the instants of sample trapping or stopping. To this end, 10–100 mg L^{-1} bromocresol green solutions, also 0.01 mol L^{-1} $Na_2B_4O_7$, were used to mimic the sample and the R_1 reagent (Fig. 1), and the establishment of undesirable concentration gradients along the flowing zones was avoided by using a 0.01 mol L^{-1} $Na_2B_4O_7$ solution as the carrier streams. In these experiments, flow rates were maintained as in the earlier work [9] and wavelength was set as 612 nm.

Length of the R_{C2} reactor was selected as short as possible to minimise broadening of the sample zone, yet long enough to permit suitable mixing conditions. With a 50 cm reactor length, good mixing conditions were provided, as confirmed by the baseline stability (thickness <0.001 absorbance) and measurement repeatability (peak height r.s.d. typically <0.01). Influence of the sample and reagent inserted volumes were investigated between 250–900 μL ($50<L_S<180$ cm) and 50–350 μL ($10<L_{R1}<70$ cm), respectively. Timing of the injector–commutator was defined in order to select the most concentrated sample portion and to define the pre-set incubation period, aiming at peak height optimization.

In the experiments carried out for investigating the influence of the main parameters involved in the in-line Tiron oxidation, 0.00–1.00 μg L^{-1} Co solutions, also 0.1% (v/v) HNO_3, were used. Accordingly, the sample carrier stream was a 0.1% v/v HNO_3 solution. Influence of reagent concentrations and alkalinity of the reaction medium were investigated by varying the concentrations of Tiron, H_2O_2 and NaOH between 0.001–0.01, 0.0005–0.01 and 0.1–4.0 mol L^{-1}, respectively. The halting period was varied between 0.0 and 6.0 min. Influence of the temperature of the reaction medium was investigated by immersing the R_{C2} reactor into a thermostatic bath and varying its temperature between 20 and 60 °C.

In the experiments carried out for selectivity improvement, the NaOH, Tiron and H_2O_2 concentrations were 0.5, 0.005 and 0.005 mol L^{-1}, respectively and 1.00 μg L^{-1} Co solutions without or with the presence of a potential interfering chemical species were prepared in 0.1% (v/v) HNO_3. The chemical species Fe^{3+}, Mn^{2+}, Cu^{2+}, Zn^{2+}, Mg^{2+}, Ca^{2+}, Ni^{2+}, V^{3+} and Cr^{6+} (based on Fe_2O_3, MnO_2, $CuSO_4 \cdot 5H_2O$, $ZnSO_4$, MgO_2, $CaCO_3$, $NiSO_4 \cdot 7H_2O$, V_2O_5 and $K_2Cr_2O_7$, respectively) were selected as potential interferents, and their concentrations were successively raised (1.0×10^{-6}, 1.0×10^{-5}, 1.0×10^{-4}, 1.0×10^{-3} g L^{-1}…): for each interferent, the highest tested concentration was that manifesting $>10\%$ variation in recorded peak height. Aiming at selectivity improvement, different solutions (0.2 mol L^{-1} sodium pyrophosphate, 0.2 mol L^{-1} sodium pyrophosphate plus 5 mg L^{-1} Mn^{2+}, 0.01 mol L^{-1} triethanolamine, 0.01 mol L^{-1} ammonium citrate, 0.01 mol L^{-1} potassium sodium tartrate, 0.2 mol L^{-1} potassium oxalate and 0.1 mol L^{-1} ascorbic acid) were successively added via the *x* confluence (Fig. 1).

After system dimensioning, the mean sample residence time (time interval between sample insertion and peak maximum achievement [1]), washing time (time interval between peak maximum and baseline restoration [1]), analytical sensitivity, measurement repeatability ($n=10$) and sampling frequency [5] were estimated. The flow system in Fig. 1, left, was then applied to the pasture analysis and the main analytical figures of merit were evaluated. For obtaining the recovery data, 1.00 mL of a 3.00 μg L^{-1} Co solution was added to a 5.00 mL sample digest and the volume was filled up to 10 mL with a 0.25 mol L^{-1} $HClO_4$ solution.

3. Results and discussion

During development of this work, good repeatability of the measurements (r.s.d. usually $<5\%$) was always noted, demonstrating the ruggedness of the different tested system prototypes and the reproducibility of the in-line Tiron oxidation.

3.1. Experiments involving the dye approach

Increasing the length of the L_S loop increased the recorded peak height, the effect being more pronounced for shorter loops. Beyond 70 cm, the improvement in sensitivity was too low, because ca. 95% of the maximum attainable peak height was already reached. A 70 cm L_S loop was then chosen, meaning a sample inserted volume of about 350 μL.

Inserting the dye solution via L_S or L_R allowed one to confirm that overlap between the involved zones was successfully attained with a L_R loop length of 40 cm (ca. 200 μL) and with 15 cm and 5 cm transmission lines between the injection port and confluence *x* (Fig. 1). The sample zone arrived then slightly later than the reagent one, and the reagent zone was broader relatively to the sample one (Fig. 2). A tendency towards a steady reagent concentration was noted at the most concentrated portion of the handled sample, and this aspect is a guarantee for system ruggedness. It should be emphasized that a recorder tracing identical to that in Fig. 2 was obtained by using the flow system in Fig. 1, right, without stopping the peristaltic pump.

In the flow system with zone trapping, the injector–commutator was switched 11 s after sample/reagent insertions for attaining best sensitivity. This time setting is not critical in relation to the flow system with zone stopping and, for didactical purposes, an 11 s GO period was also set.

The dye approach was also useful for comparing the management of the sample and reagent zones in relation to the zone stopping and zone trapping strategies. The following points can be inferred from Fig. 3:

- Wash time (t_w): This parameter is shorter in the system with zone trapping because the front and trailing portions of the sample zone are discarded during the TRAP period. On the

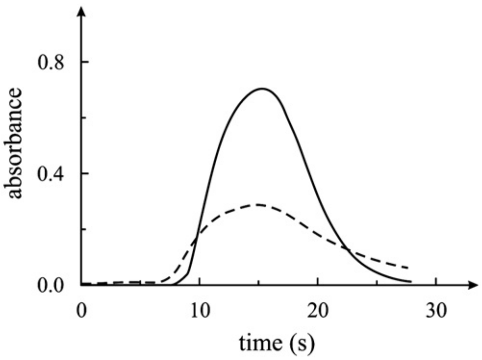

Fig. 2. Recorder tracing for assessment of sample/reagent overlap. Figure refers to the flow system in Fig. 1, left into which a 50.0 mg L^{-1} bromocresol green buffered solution is inserted via L_S (full line) or L_{R1} (traced line) sampling loops. Switching the injector–commutator not performed after sample (or reagent) insertion.

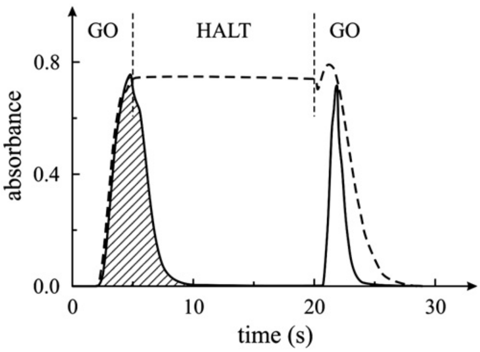

Fig. 3. Recorder tracing associated with the flow systems with zone trapping (full line) and zone stopping (traced line). GO and HALT = handled sample in movement and halted; dashed area = non-trapped portion of the flowing sample that passes through the detector without being halted; experimental conditions as in Fig. 1.

other hand, the entire sample zone is halted in the system with zone stopping; sample discarding becomes then less efficient. Although the difference in wash times related to the flow systems in Fig. 1 is remarkable, sampling rate (F) is not so different, as this parameter depends mainly on the halting period according to the relationship [5]: $F = 3600/(t_{TRAP} + t_w)$, and this period is much higher than t_w. An efficient expedient to take advantage of the short t_w value inherent to the flow system with zone trapping is to use several parallel trapping reactors [12], and this aspect is presently under investigation.

- Carryover: The flow system with zone trapping is characterized by a lower carryover coefficient [13] relatively to that with zone stopping. This aspect can be explained by recalling that, in the flow system with zone trapping, the continuously flowing carrier stream removes the remaining portion of the previous sample still inside the manifold portion between insertion port and R_{C2} reactor.
- Sensitivity: For the same HALT period, peak heights associated with the flow system with zone stopping are slightly higher, ca. 5% in the present situation. This aspect refers to the sample axial dispersion taking place during transport of the handled sample from the R_{C2} reactor to the flow-through detector. Regarding the flow system with zone stopping, the diluent flowing stream contains the front and trailing edges of the sample zone; as the coloured product is formed also in these portions, lowering of the analytical signal due to dispersion becomes less pronounced. On the other hand, a blank solution acts as a diluent in the flow system with zone trapping thus lowering the analytical signal.
- Recorded peak shape: With the dye approach, instant measurements ideally indicate the instant concentrations of the sample (or reagent) inserted solution. Analysis of peak shape is then useful for evaluating temporal distribution of the volumetric fractions relatively to the halted and non-halted sample portions. Regarding zone trapping, the dashed area in Fig. 3 corresponds to the non-trapped sample portions that are monitored earlier, as they are sent directly to the flow-through detector. The corresponding recorded signal resembles a typical signal of an ordinary flow injection analyzer with a distortion near peak maximum which refers to the sample portion removed by the R_{C2} trapping reactor and further released. This portion is larger than the trapped portion (peak on the right), and this aspect is relevant for preventing an excessive sample dispersion. Regarding zone stopping, the large horizontal tracing in Fig. 3 refers to the steady measurement of the sample portion halted inside the detector. Again, presence of this sample portion inside the detector during the STOP period reflects the large sample amount needed to prevent dispersion. It should be emphasized that, in analytical procedures involving highly coloured samples where sensitivity is not critical, a low sample volume can be selected: the dashed area related to the zone trapping system would be very small and horizontal tracing related to the zone stopping system would be very low.
- Peak distortion: A distortion in recorded signal due to the ON/OFF pump switching was observed for the flow system with zone stopping. This distortion limits the signal-to-noise ratio, and was probably caused by the plasticity of the manifold tubing, often referred to as an inertial effect. The drawback was not noted for the zone trapping system as a continuous pump operation was involved.
- Kinetic aspects: Shape of the recorded peaks related to the flow systems in Fig. 1 depends also on kinetic aspects, as the involved Tiron oxidation is relatively slow. For pasture analysis, the dashed area associated with zone trapping is very low (about 0.076 absorbance of for 1.00 μg L^{-1} Co) whereas the horizontal tracing associated with sample stopping is not observed, an increasing absorbance during the STOP period being noted instead.

3.2. Tiron oxidation

Alkalinity is an important parameter in the system design. As in earlier work [9], increasing the NaOH concentration in the R_4 reagent up to 0.5 mol L^{-1} NaOH improved sensitivity; beyond this value, however, a sensitivity drop was noted probably because of the higher decomposition rate of the coloured formed product under more alkaline conditions [9]. Moreover, the measurement repeatability deteriorated for higher NaOH concentrations. The drawback was minimised by placing a large ($>$ 90 cm) R_{C1} (Fig. 1) reactor for improving the mixing of the NaOH and H_2O_2 solutions. For a 3 min halt period, best sensitivity was noted for 0.5 mol L^{-1}, the selected NaOH concentration. Sensitivity was improved also by increasing the Tiron and H_2O_2 concentrations. Increasing these concentrations up to 0.005 mol L^{-1} increased the recorded peak height in asymptotical manner, the effect being more pronounced in relation to Tiron. Beyond this concentration, any increase in reagent concentration did not improve the Tiron oxidation rate, thus sensitivity. The above mentioned concentration was then chosen for the R_1 and R_3 reagents, these values being different relatively to earlier work in view of the different mean sample resident time involved.

Halting period is an important parameter in the system design, too. The increase in absorbance during the halting period followed an asymptotical function, approaching the maximum value (represented by a plateau) after 6 min. As about 90% of the maximum value was attained with 3 min, this value was preliminarily selected.

Temperature of the reaction medium proved to be an important parameter, as demonstrated in parallel experiments carried out without sample halting. Under room temperature, 4% of the maximum signal was observed and this value reached 20% by increasing the water-bath temperature to 60 °C. It was decided however to operate the flow system under room temperature considering that about 90% of maximum signal was attained by exploiting sample halting, that a simple manifold architecture was aimed, and that the possibility of release of air bubbles that might affect sample dispersion and measurement was minimized under room temperature.

Regarding selectivity, Ca^{2+}, Mn^{2+} and Fe^{3+} were the main interferences, considering their expected concentrations in the digests [14] and their influence on Tiron oxidation at concentrations higher than 100, 1 and 10 mg L^{-1} respectively (Table 1). Addition of citrate, tartrate or triethanolamine was not effective

Table 1
Influence of potential chemical species. Data refer to peak heights, in absorbance. $S = 1.00$ µg Co L^{-1} (1Co)+N mg L^{-1} of a potential interfering species. Different masking solutions added as R_2 (Fig. 1); a = water, b = 0.01 mol L^{-1} ammonium citrate, c = 0.01 mol L^{-1} potassium sodium tartrate, d = 0.01 mol L^{-1} triethanolamine, e = 0.2 mol L^{-1} sodium pyrophosphate, f = 0.2 mol L^{-1} sodium pyrophosphate + 5.0 mg L^{-1} Mn^{2+}, g = 0.2 mol L^{-1} potassium oxalate. Column at right (∗) refers to the addition of potential interfering species in absence of cobalt and any masking agent. For details, see text.

S	a	b	c	d	e	f	g	∗
1Co	0.90	0.07	0.47	0.04	0.72	0.93	1.28	–
1Co+10Zn	1.15	0.09	0.69	0.04	0.82	0.97	0.98	0.18
1Co+10Mg	1.00	0.09	0.42	0.03	0.80	0.98	1.43	0.14
1Co+10Al	0.88	0.08	0.60	0.03	0.74	0.90	1.48	0.12
1Co+10V	0.87	0.09	0.49	0.05	0.66	0.98	0.56	0.11
1Co+1Cr	0.83	0.11	0.47	0.06	0.60	0.98	0.48	0.11
1Co+1Ni	0.78	0.11	0.68	0.05	0.79	0.97	0.52	0.11
1Co+1Mn	1.07	0.66	1.00	0.70	1.05	0.93	1.35	0.79
1Co+10Cu	0.88	0.10	0.50	0.04	0.72	0.95	0.80	0.14
1Co+100Ca	0.03	0.03	0.03	0.02	0.17	0.93	0.65	0.02
1Co+10Fe	1.73	0.45	1.49	0.36	1.57	0.95	1.84	1.39

Table 2
Recovery data. Data refer to concentrations in the digests, before and after spiking. Expected increase in concentration: 0.300 µg L^{-1}.

Sample	Co concentrations (µgL^{-1})		Recovery (%)
	Original	Found	
A	1.03 ± 0.02	1.30 ± 0.05	98
B	0.88 ± 0.02	1.35 ± 0.03	114
C	1.09 ± 0.00	1.47 ± 0.03	105
D	1.30 ± 0.05	1.65 ± 0.03	103
E	1.25 ± 0.06	1.75 ± 0.06	112

in circumventing the interference caused by these ions; moreover, this addition led to a drop in sensitivity. Addition of oxalate was the least effective, as it did not reduce the interference effects caused to any of the investigated potential interfering species. Addition of ascorbic acid cannot be recommended; although Fe^{3+} interference was suppressed, the analytical signal underwent a pronounced reduction.

Pyrophosphate was effective for reducing most of the interferences (Table 1) but a slight interference from Mn^{2+} still persisted. It was then decided to add this chemical species together with pyrophosphate, as this strategy relying on use of a compensating solution was exploited in earlier work [9]. Considering the solubility of sodium pyrophosphate in alkaline medium [15] and the expected manganese concentration in the digest, the R_2 reagent (Fig. 1) was a 0.2 mol L^{-1} $Na_4P_2O_7$ plus 5.0 mg L^{-1} Mn^{2+} solution. In this situation, the proposed procedure can be applied to pasture analyses.

3.3. Application

The flow system with zone merging and zone trapping proved to be very rugged, and baseline shift was not noted during 4 h operation periods. As a consequence of the involved zone merging configuration, only 330 µg Tiron are consumed per determination, which corresponds to about 10% of that related to the original procedure [9] involving continuous reagent addition. Precise results (r.s.d. = 0.7%, $n = 10$, for a typical sample with 0.60 µg L^{-1} Co) are obtained at a rate of 18 h^{-1} (see also Table 2) Lambert–Beer's law is followed up to 2 µg L^{-1} Co. A typical analytical curve is described by

$$h = 0.4625 + 0.4492\,[\mathrm{Co}] \quad (r = 0.9981,\ n = 7)$$

where h = recorded peak height, in absorbance; [Co] = analyte concentration in the plant digests, in µg L^{-1}.

It should be recalled that the high linear coefficient of the regression equation reflects the development of the uncatalized indicator reaction.

Recovery data ranging from 98% to 114% (Table 2) were obtained with the flow system in Fig. 1, left. Detection limit relying on the 3σ criterion [16] was estimated as 0.046 µg L^{-1} Co. These figures of merit emphasises the potentiality of applying the proposed flow system to pasture analysis.

4. Conclusions

A noteworthy advantage of the flow systems with zone merging and zone halting in the reactor is that the sample volume may undergo a pronounced increase without impairing sampling rate, and this aspect matches the one already reported for flow systems with zone halting in the detector [5]. Under all the investigated conditions, the wash time inherent to the flow system with zone trapping was shorter in relation to zone stopping. An additional potentiality is that several zones can be halted in parallel reactors, thus expanding the potentialities of the zone trapping strategy. As large volumes and long halting times can be set, sensitivity of flow-based procedures relying on relatively slow chemical reactions is improved. The peristaltic pump is operated continuously, thus ON/OFF turning and eventual heating are not limiting aspects leading to a longer pump life time.

In the present application, sample/reagent insertions were done simultaneously with the movement of the R_{C2} reactor (Fig. 1) as a single three section injector–commutator was used. Rotary valves can be used for building up the flow system with zone trapping. The related flow diagram is analogous to that proposed in relation to the stopped-in-loop flow system [17]. A more versatile analyser could be designed by using two independent commutators, and studies focusing on this matter are presently in progress.

Acknowledgements

Partial support from CNPq and fellowships from FAPESP to CRS, ACFV and MKS are greatly appreciated.

References

[1] S.D. Kolev, I.D. McKelvie (Eds.), Wilson and Wilson's Comprehensive Analytical Chemistry, vol. 54, Elsevier, Amsterdam, 2008.
[2] E.A.G. Zagatto, C.C. Oliveira, A. Townshend, P.J. Worsfold, Flow Analysis with Spectrophotometric and Luminometric Detection, Elsevier, Amsterdam, 2012.
[3] J. Ruzicka, E.H. Hansen, Anal. Chim. Acta 106 (1979) 207–224.

[4] F.J. Krug, B.F. Reis, M.F. Gine, J.R. Ferreira, A.O. Jacintho, E.A.G. Zagatto, Anal. Chim. Acta 151 (1983) 39–48.

[5] A.C.F. Vida, M.K. Sasaki, T.F. Gomes, C.R. Silva, M.A. Feres, E.A.G. Zagatto, Talanta 85 (2011) 259–263.

[6] R.A.S. Lapa, J.L.F.C. Lima, J.L.M. Santos, Anal. Chim. Acta 407 (2000) 225–231.

[7] N. Teshima, M. Kuno, M. Ueda, H. Ueda, S. Ohno, T. Sakai, Talanta 79 (2009) 517–522.

[8] M. Otto, J. Rentsch, G. Werner, Anal. Chim. Acta 147 (1983) 267–275.

[9] M.A.Z. Arruda, E.A.G. Zagatto, A.O. Jacintho, S.M.B. Brienza, J. Braz., Chem. Soc. 2 (1991) 47–50.

[10] L.R. McDowell, Anim. Feed Sci. Technol. 60 (1996) 247–271.

[11] F.J. Krug, H. Bergamin-Filho, E.A.G. Zagatto, S.S. Joergensen, Analyst 102 (1977) 503–508.

[12] B.F. Rocks, R.A. Sherwood, M.M. Hosseinmard, C. Riley, Anal. Chim. Acta 179 (1986) 225–231.

[13] M.A. Evenson, G.P. Hicks, R.E. Thiers, Clin. Chem. 16 (1970) 606–611.

[14] P. Martin-Prevel, J. Gagnard, P. Gautier, Plant Analysis as a Guide to the Nutrient Requirements of Temperate and Tropical Crops, Lavoisier Pub, Paris, 1987.

[15] R.C. Weast (Ed.), 45th ed.,The Chemical Rubber Co., Cleveland, OH, 1964.

[16] L.A. Currie, Pure Appl. Chem. 67 (1995) 1699–1723.

[17] N. Teshima, S. Ohno, T. Sakai, Anal. Sci. 23 (2007) 1–2.