A Mixer with Magnetic Stirring for Flow Injection Systems

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Abstract

In this article a mixer with magnetic stirring for flow injection systems, constructed in polyacrylate and PTFE is described. The device was compared with a common **T** shape mixer. An increase of about 11.7 % on the signal was observed. However the main result is the increase of the precision of spectrophotometric measurements from 5.5 % (RSD) to 2.3 % (RSD).

Keywords: mixer; flow injection system; magnetic stirring

1. Introduction

In middle of the 1970's, Ruzicka and Hansen [1] developed an analytical system based on the sample injection in a flow carrier, non-segmented by air bubbles, which was named Flow Injection Analysis by them.

This technique presented high repercussion in the scientific community. Nowadays it is widely applied due to its simplicity and versatility [2], where several kinds of procedures can be done in line: gas diffusion, isothermic distillation, liquid-liquid extraction, etc. [3].

The basic components of a FIA system are: propulsion of the fluids, manifold for fluids transport and for chemical reactions, sample injection and detection.

The most used mode of propulsion is the peristaltic pump, due to its operational simplicity and high torque. The use of syringes is presently finding more extensive use through the MSFIA (Multisyringe flow injection analysis) method. The use of mini pumps is also gaining importance and the propulsion through gas pressure or by the action of the gravity cannot be forgotten.

Historically, various devices have been used to introduce samples in the flow system. For example, rotary valves and proportional injectors.

A variety of methods can be used for the detection, such as spectrophotometry in general, electrochemistry, reflectometry, thermometry, etc.

The transportation of the fluids along the system is done by tubes and connections. Chemical reactions occur in reactors and bobbins after the joining of the reacting solutions. One problem that sometimes occurs in such cases is related with the mixing efficiency of the solutions. Mixing inefficiency can affect the analytical system in terms of determination frequency, accuracy, precision, detection and determination concentrations, etc.

With the objective to enhance the mixing efficiency of the reactive solutions in a flow system we designed and constructed a very simple reactor with magnetic agitation which is described below.

2. Experimental

2.1 Equipments

*Common confluence **T** shape mixer:* piece constructed in polyacrylate: 2 cm wide; 2 cm large; 1 cm high.

Mixer with magnetic stirring: constructed in polyacrylate according to the scheme shown in Fig. 1 and in the picture (Fig. 2). The upper piece was made in PTFE. Into the reaction chamber of the mixer a cylindrical magnetic bar coated with PTFE: *ca.* 4 mm long; diameter *ca.* 2 mm (volume about 13 µL) was introduced.

Magnetic stirrer: IKA model lab disc.

Peristaltic pump: Cole Palmer Masterflex.

Spectrophotometer: Single-beam Femto model 600 (325 - 1100 nm).

Chart Recorder: Cole Parmer Series 8375.

Flow cuvette: 1.00 cm path length quartz cuvette.

Sampling valve: This sampling valve has been described previously in detail [4,5].

Pumping tubes: Ismatec two-stop tubes, blue-green, SC0019, Tygon®, internal diameter 1.75 mm.

Conducting tubes: Polyethylene tubes, i.d. 1 mm, o.d. 1.8 mm.

The manifold of the flow system is depicted in Fig. 3. The conditions of the FI procedure for the determination of vancomycin are summarized in Table 1.

2.2. Reagents and solutions

All the reagents were of analytical grade, excepting vancomycin that was a certified pharmaceutical product gently furnished by the pharmaceutical laboratory ABL Antibióticos do Brasil Ltda.

Copper acetate solution (3.0×10^{-2} mol L^{-1}): 0.5989 g of $Cu(CH_3COO)_2.H_2O$ was dissolved in 100 ml of distilled water.

Vancomycin solution (1.0×10^{-2} mol L^{-1}): 0.7425 g of the product (Molar mass = 1495 g) was dissolved in 50 mL of distilled water.

Table 1 Parameters used in the flow system for copper (II) / vancomycin[a] and for water /ethanol aqueous solution[b] mixing

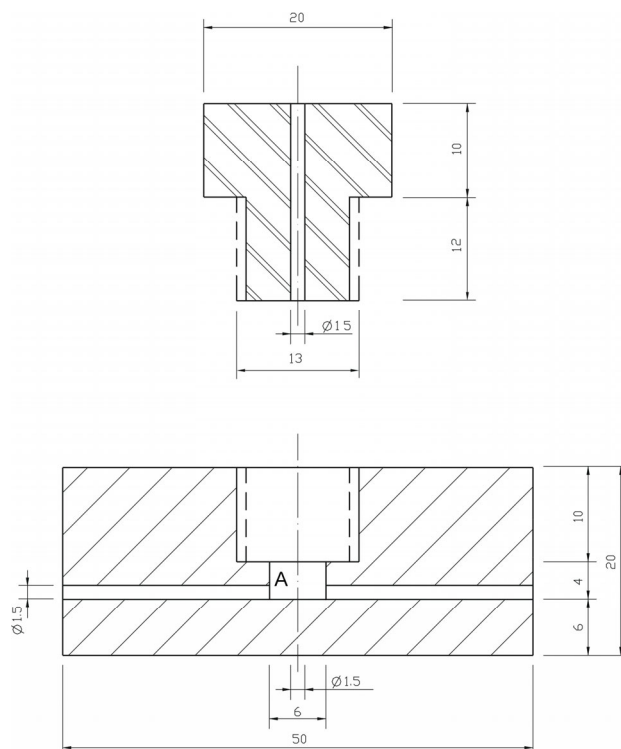|	
Total flow rate[a,b]	2.0 mL min^{-1}
Copper (II) / Vancomycin – molar rate[a]	3:1
Wavelenght[a,b]	525 nm
Sample loop volume[a]	150 µL
Copper (II) solution loop volume[a]	200 µL
Copper (II) concentration[a]	3.0×10^{-2} mol L^{-1}
Vancomycin concentration[a]	1.0×10^{-2} mol L^{-1}
Reaction bobbin length[a,b,c]	50 cm

[c] Polyethylene tube, i.d. 1 mm, o.d. 1.8 mm.

Fig. 1 Mixer with magnetic stirring. Lower part made with transparent polyacrylate. Upper part made in white PTFE. The mixing chamber is indicated by the letter **A**. The magnetic bar coated with PTFE was introduced in this chamber. The total volume of the mixing chamber is about 113 µL; as the volume of the magnetic bar is about 13 µL the effective volume of the mixing chamber is about 100 µL. Measures in the figure are expressed in millimeters.
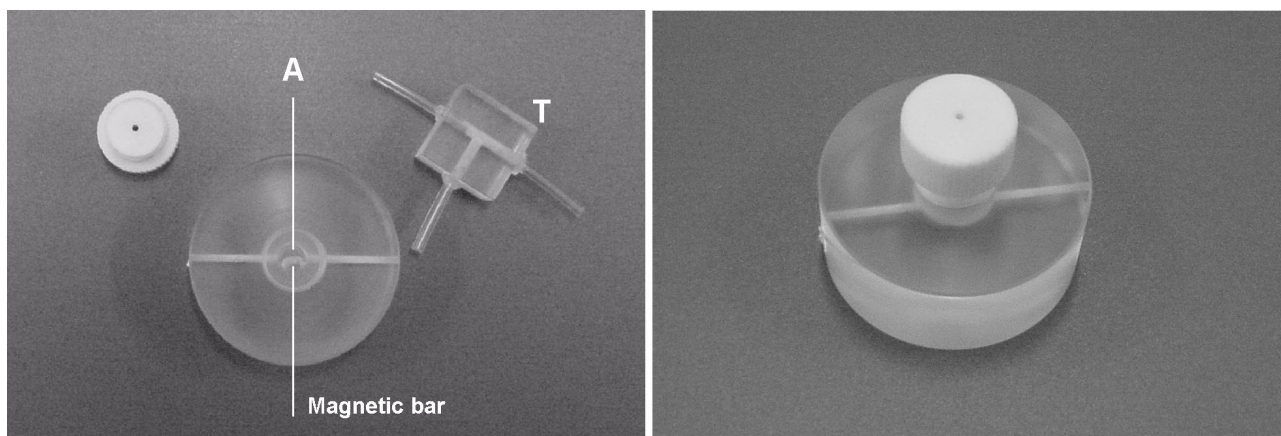

Fig. 2 Picture of the proposed mixer and of a common **T** shape mixer (**T**). The letter **A** indicates the mixing chamber as in Fig. 1.
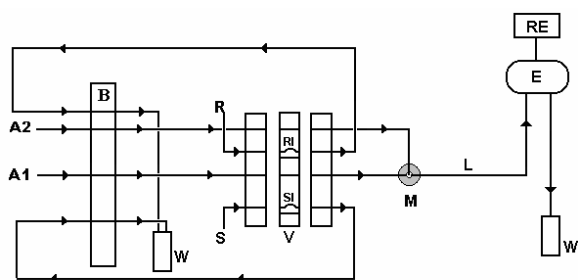
Fig. 3 Scheme of the flow manifold used. **A2** = flow of water; **R** = introduction of the copper (II) solution 3.0×10^{-2} mol L^{-1}; **A1** = flow of water; **S** = introduction of vancomycin solution 1.0×10^{-2} mol L^{-1}; Flow rate **A1** = flow rate **A2**; **B** = peristaltic pump; **W** = waste; **RI** = reagent injection loop; **SI** = sample injection loop; **V** = sampling valve [4,5]; **M** = mixer with stirring; **L** = reaction bobbin (50 cm); **E** = spectrophotometer; **RE** = chart recorder.

3. Results and discussion

Reactors (mixers) are unities inside the transportation system where sample and analytical reagents chemically interact, so that in the moment of its passage through the detection unity the adequate characteristics for the measurement have been achieved [6].

Therefore, in the cases of substances which reactions with the analytical reagent are relatively slow the use of a reactor that offers higher mixing efficiency will present advantages in comparison to the common mixers where the mixture of the solutions occur only by its simple confluence and diffusion.

In the present case, the evaluation of the proposed mixer was done with the system vancomycin chloridrate and ions copper (II) with spectrophotometric monitoring at 555 nm [7]. In this work the wavelength of 525 nm was used in order to decrease the interference of the absorption of the copper (II) acetate solution.

Complementary experiments to verify the efficiency of the mixer, using an ethanol water solution, was performed. The results can be observed in Fig. 4. In the same system depicted in Fig. 3 water was introduced in A_1 and ethanol 25% v/v water solution in A_2. Sampling valve was not operated and the signal was monitored in the spectrophotometer at 525 nm to verify the intensity of the noise provoked by refraction index gradients. The use of **T** shape mixer (**c**) leads to the noisiest profile, followed by the case of the proposed mixer without stirring (**a**). The best situation was observed when stirring was used (**b**).

In Fig. 5 and in Table 2 the results obtained with the proposed mixer in comparison with a common **T** shape mixer are presented. The **a** peaks correspond to the proposed mixer

used without stirring but with the magnetic bar inside in order to maintain the internal volume constant. The **b** peaks were obtained with stirring. An increase of about 11.7 % on the signal was observed but the main result is the increase of the precision from 5.5 % (RSD) to 2.3 % (RSD). The **c** peaks were obtained with a common **T** shape mixer. It can be easily noted that the signal is quite higher with the stirred mixer (about 31.8 %). The RSD (4.9 %) of the **c** peaks is about 21 % smaller than the RSD obtained with the proposed mixer without stirring (6.2 %). This means that the smaller RSD obtained with stirring is not due to the mixer shape but to the additional mixing effect with the rotating magnetic bar.

Therefore the use of the proposed mixer with stirring in the case of the reaction of vancomycin with copper (II) ions is advantageous as it increases the analytical signal and also the precision.

The main body of the mixer (lower part) was constructed in transparent polyacrylate to allow visual monitoring. Its size is relatively big in order to obtain a weight (about 54 g in this case) that stabilizes the mixer on the magnetic stirrer avoiding the use of a clamp. If a clamp is used a much smaller mixer can be utilized.

The connections of the conducting tubes are done directly in the holes which diameters in this mixer are always 1.5 mm. As the external diameter of the conducting tubes is a little higher, when they are introduced in the holes very tight connections are obtained. Alternatively, as showed in the **T** mixer (Fig. 2) pieces of flexible Tygon® tubes can be fixed to the mixer and used for connections with the polyethylene conducting tubes.
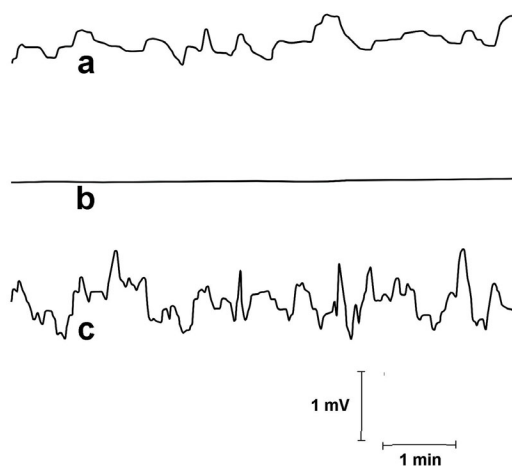

Fig. 4. Observed baselines: Water mixed with ethanol 25 % v/v water solution. The profile **a** corresponds to the proposed mixer without stirring; the **b** to the proposed mixer with stirring. The **c** profile corresponds to the usual T shape form mixer. Spectrophotometric monitoring carried out at 525 nm.
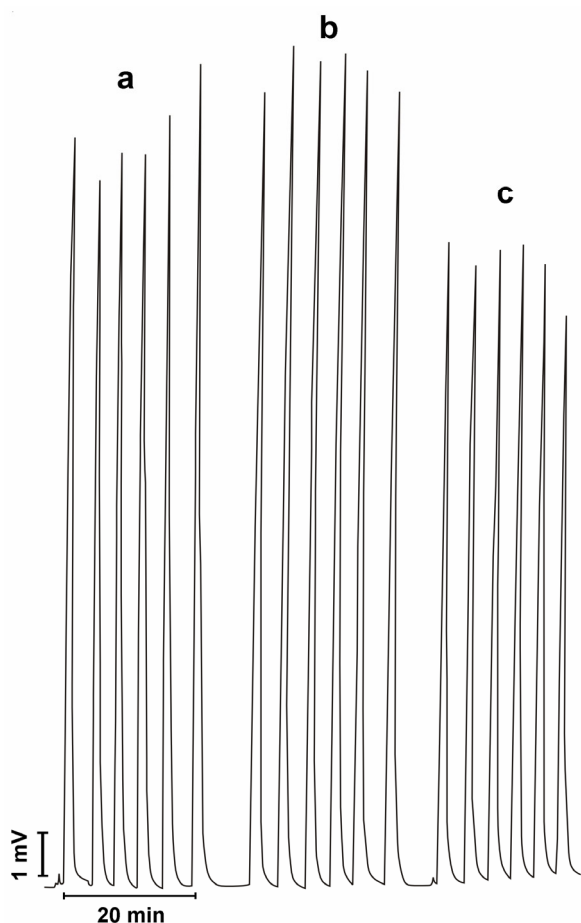
4. Conclusions

The proposed mixer, used in the vancomycin-copper (II) system, furnished a significant increase in the analytical signal in comparison with a common **T** shape mixer. An important decrease in the RSD was also observed.

It was also observed that the proposed mixer with magnetic stirring shows very good efficiency in the case of mixing of water with ethanol-water solution, where the signal noise provoked by refraction index gradients was not observed.

Considering the observed results, this kind of mixer with magnetic stirring can be recommended for use in FIA manifolds where the analytical reactions present relatively slow rates and in all cases where the influence of refractive index gradients must be minimized.

5. References

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Fig. 5. Fiagram obtained with the proposed mixer and with a **T** shape mixer: **a** is the group of peaks corresponding to the signals that result using the mixer without stirring; **b** is the group of peaks obtained with stirring; **c** is the group of peaks that resulted with the use of a common **T** shape mixer.

Table 2 Comparison of the results obtained with the mixers. **a**= results obtained using the proposed mixer without stirring; **b**= results obtained with stirring; **c**= results obtained with a common T shape mixer. In all cases the number of determination was 6 (n=6).

Group of peaks	Height signal (cm)	Relative height	SD (cm)	RSD (%)
a	10.55	1.17	0.65	6.2
b	11.37	1.33	0.30	2.6
c	8.58	1.00	0.42	4.9