TURBIDIMETRIC FLOW ANALYSIS

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ABSTRACT

Historical development, potentialities and limitations of turbidimetric methods in flow analysis are revisited, with emphasis to nucleation rate. Main applications are presented and guidelines for system design are suggested.

HISTORICAL ASPECTS

A very earlier commentary [1] on actual status of turbidity measurements was foretold by Wells in 1927, who claimed that "Every attack on the problem of disperse systems is disappointing, because of the baffling complexity of the phenomena. Diaphanometers, nephelometers, turbidimeters, tyndallmeters, dispersimeters, opacimeters, have been developed and placed on the market, but not one has yet been accepted as a standard instrument for the laboratory... Apparently, turbidity measurements have not proven satisfactory and yet the prospects are more hopeful that they seem. Once the limitations of such optical methods are understood, their real possibilities will be appreciated for what they are worth".

Development of turbidimetric methods of analysis in the last decades has revealed that the Achilles wheel of turbidimetry was more related to processes of solution handling than to quality and performance of measuring instruments. In fact, any variation in the colloidal sol preparation may result in lack of particle size uniformity from one determination to the next, and light scattering varies with the size of the particles as well as their concentration [2]. In this context, the flow system, often considered as a powerful solution manager, is very attractive in view of its unique feature of yielding reproducible colloidal suspensions. So, it is not surprisingly that a turbidimetric sulphate determination was proposed [3] few years after the concept of air-segmented flow analysis was introduced [4].

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At that time, the presence of air bubbles in the analytical path of the existing autoanalyzers was considered essential to reduce carryover, to improve mixing between sample and reagents, and to scrub the inner walls of the tubing and flow cell. Ruzicka and Hansen demonstrated [5] that these tasks could be also achieved without segmentation. The absence of air bubbles lead to a simpler system, termed flow-injection system, and also expanded the potentialities of flow analysis. This explains the increasing number of publications dealing with turbidimetric procedures carried out with the flow-injection analyzer (Tab. 1) after the pioneer work of Krug and collaborators [6].

In flow-injection turbidimetry, laminar flow is characteristic [7] and the solid particles undergo rotation at defined fluid lines [8]. Up to day, however, a quantitative description of dispersion including solid particles seems not to be proposed.

GENERAL

Turbidimetric procedures have been proposed for organic species of pharmaceutical relevance and for some inorganic ions (Tab. 1). Sulphate is by far the most investigated ion, probably because of the low availability of alternative procedures.

Addition of colloid protectors or surfactants is often required (Tab. 1) which, in contrast to batch procedures, is efficiently accomplished in flow-based methodologies [9]. The presence of these agents is an additional guaranty of uniform nucleation, improving measurement reproducibility. Carryover and memory effects can be lessened in view of the better uniformity of the particles, thus reducing washing time and baseline drift. For this task, intermittent addition of a washing solution [10] or a fast washing stream [11] has been additionally exploited.

A noteworthy feature of Tab. 1 is the relatively low sampling rate associated to some listed applications. Although very fast precipitation reactions are concerned, nucleation may be a limiting factor in sample throughput.

NUCLEATION RATE

In a supersaturated solution, the increase in turbidity is observed during the nucleation process which in some cases is remarkably slow. As an extreme example, Nielsen reported [12] that for calcium fluoride solutions, turgidity was observed only after several days.

In flow-injection analysis, slow nucleation was reported by Krug who determined sulphate in natural waters and plant digests [13]. A sulphate standard solution was placed in a situation of "sample infinite volume" [14]: after achievement of a steady state measurement (a, b, c, d - Fig. 1), it was stopped, and further increase in the measurement was followed. Nucleation rate was

dependent mainly on barium chloride (Fig. 1) and sulphate concentrations, acidity, presence of nitric or hydrochloric acid, and surfactant addition. With proper selection of reagent concentrations, amount of added sulphate, and use of intermittent alkaline-EDTA stream, the system handled *ca* 120 samples per hour.

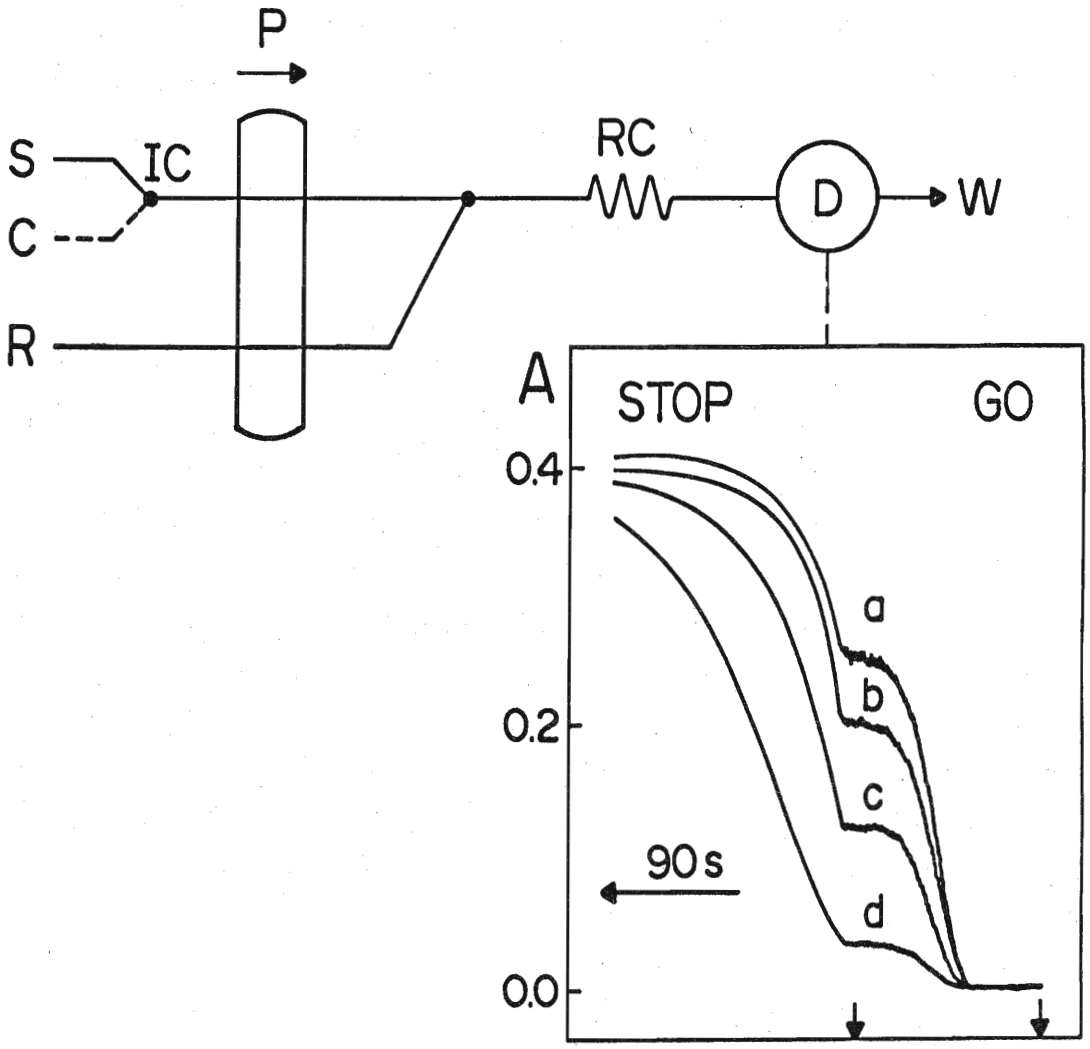

Fig. 1. Flow system for studying nucleation kinetics. S = 60 mg l^{-1} sulphate in 0.02 M HNO_3 (4.0 ml min^{-1}); C = 0.02 M HNO_3 (4.0 ml min^{-1}); R = barium chloride reagent, also 0.05 % w/v in PVA; IC = injector-commutator; P = peristaltic pump; RC = reaction coil (100 cm); D = spectrophotometer (410 nm); W = waste. In the recorder output: a, b, c and d refer to 40.0, 20.0, 10.0 and 5.0 % w/v $BaCl_2.2H_2O$ concentrations in R; vertical arrows indicate instants of sample introduction (right) and peristaltic pump stopping (left). For details, see [13].

The experimental setup of Fig.1 may provide information on trends of the involved chemistry but may be applied to flow-injection turbidimetry with restrictions. Experiments carried out by the authors revealed that for the potassium/tetraphenylborate system, the steady state measurement related to the STOP period was not achieved due to crystal settlement at the detection unit, and for the sucrose/Fehling system, tubing was clogged due to excessive crystal growth.

Nucleation kinetics has been exploited in other situations. In this way, Grases and co-workers determined chemical species able to speed up or inhibit crystal growth [15-20]. In spectrophotometry, absorbance measurements may be carried out after precipitation reactions without the need for crystal separation, as e.g. in the flow-injection determination of chloride by the thiocyanate method [21]. Finally, the feasibility of interference masking by precipitation reactions in flow spectrophotometry seems not to be yet exploited.

ANALYTICAL CHARACTERISTICS

The ever increasing demand for fast and accurate analysis and the favorable characteristics of system robustness and reagent consumption inherent to the flow analyzer explains the growth of applications of turbidimetric methods. With modern system design, drawbacks associated to baseline drift have been circumvented. Simultaneous determinations involving other techniques are possible, too.

Finally, it is interesting to comment the concept of pre-nucleation [22] which is worthwhile in situations where slow nucleation may limit the system design. Primary nuclei are formed outside the analytical path by convergence of the precipitant reagent stream with a flowing solution containing a suitable chemical species. The formed nuclei are then seeded in the main channel by confluence. The interaction between sample zone and precipitant reagent occurs then under more favorable supersaturation conditions. With the approach, nucleation rate is no longer a limiting factor in sampling rate.

Acknowledgments - Partial support from EC (Proj. CI1* CT92-0052), CNPq, CAPES, FINEP and FAPESP is appreciated. The authors express their gratitude to B.F. Reis and L.F.B.P. Gouveia for critical comments.

Tab. 1. Selected procedures. BCP = bromocresol purple; BPB = bromophenol blue; DPHA = lidnone); TPB = tetraphenylborate; * = not reported.

Analyte	sample	reagent	surfactant	range mg l^{-1}
sulphur	plant	$BaCl_2$	gelatine	0-0.66[e]
sulphur	plant	$BaCl_2$	gelatine	10-100[d]
sulphur	plant	$BaCl_2$	TWEEN-20	0-0.66[e]
sulphur	plant	$BaCl_2$	gelatine	0-25[f]
sulphate	plant, water	$BaCl_2$	PVA	10-200[d]
sulphate	water	$BaCl_2$	'PVA	*
total N	plant	Nessler	-	1-5[h]
ammonium	soil extract, water	Nessler	-	0.5-8.0[g]
sulphate	soil, fertilizer, plant	$BaCl_2$	gum arabic	0-15, 0-35[d]
sulphate	water	$BaCl_2$	PVA	20-200[d]
sulphate	soil, plant	$BaCl_2$ (c)	PVP	0-25[f]
total N	plant	Nessler	-	70-350[g]
sulphate	surface, tap waters	$BaCl_2$	gelatine	50-200
sulphate	water	$BaCl_2$	gelatine	50-200
sulphate	water, plant	$BaCl_2$ (c)	PVA	1-30, 5-200[d]
chloride	river water	$AgNO_3$	PVA	up to 14
concanavalin A	serum	yeast mannan	-	0-2000
IgG	serum	anti IgG	-	0-35560
levamisole	pharmaceutical	$[HgI_4]^{2-}$	-	7-32
sulphate	effluent	$BaCl_2$	gelatine	up to 200
calcium	-	oxalate	glycerol	*
chlorhexidine	drug	thymol blue	-	10.5-63
sulphate	soil extract	$BaCl_2$ (c)	gelatine	0-10[f]
phenformin	pharmaceutical	tungstate	-	120-222
DPHA	drug	BPB	-	50-230
amitriptyline	drug	BCP	-	30-200
sulphur	plant	$BaCl_2$ (c)	gum arabic	0-35[d]
sulphate	(b)	$BaCl_2$	-	0-1920
potassium	leave	TPB	glycerol	0-8
promethazine	drug	BPB	-	25-197
sulphate	sea water	$BaCl_2$	-	480-2880
sulphate	water	$Pb(NO_3)_2$ (c)	PVA	5-20
sulphate	river water	$BaCl_2$	PVA	0.10-2.00

(a): S = segmented, U = unsegmented flow; (b): effluents from petroleum industry; (c): adittion of sulphate as seed the injected solution; h: % N, dry basis;

diphenhydramine; IgG = immunoglobulin G; PVA = poly(vinyl alcohol); PVP = poly(vinyl pirro-

flow (a)	sampling rate, h^{-1}	r.s.d.	remarks	year	ref.
S	40	*	three-way valve timer	1965	3
S	40	*	use of washing solution	1972	23
S	30	*	washing soln + intermittent stream	1974	24
S	40	0.047	use of washing solution	1976	10
U	180	0.005-0.02	first flow injection turbidimetry	1977	06
U	250	*	inert carrier stream	1978	25
U	100	0.03	isothermal destillation	1979	26
U	120	*ca* 0.015	measurement of color-turbidity	1979	27
S	30	< 0.006		1980	28
U	*	0.01-0.02	exploitation of pH gradients	1980	29
S	20-30	0.001-0.09	intermittent reagent addition	1980	30
S	80	*ca* 0.01	tartrate to avoid baseline drift	1980	31
U	60	< 0.0095	sample/wash alternating injections	1982	32
U	60	< 0.01	on-line sample filtration	1982	33
U	120	< 0.01	alternating streams	1983	11
U	15	*	interferent removal by ion-exchange	1984	34
U	50	0.02-0.2	stopped-flow/merging zones	1984	35
U	40	0.02-0.06	stopped-flow/merging zones	1985	36-7
U	80	0.009	ion association	1986	38
U	60	< 0.02	rFIA for on-line monitoring	1987	39
U	60	< 0.02	use of stirring chamber	1987	40
U	53	0.015	comparison of reagents	1987	41
S	20	*	ion-exchange for analyte extraction	1988	42
U	67	0.008	ion association	1989	43
U	51	0.003	ion association	1990	44
U	39	0.014	ion association	1990	45
U	120	0.025	alternating streams	1990	09
U	24	*	sulphur speciation	1991	46
U	*	0.005-0.08		1992	47
U	32	0.013	ion association	1992	48
U	50	*		1992	49
U	*	*		1992	50
U	50	< 0.02	analyte concentration by ion-exchange	1993	51

solution; d: S-SO_4 in the injected solution; e: % sulphur, dry basis; f: sulphur; conc. in the injected solution; g: N-NH_4 in

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