Influence of Conditions of GC- Flame Ionization Detector Analysis on Parameters of Peaks in Highly Concentrated Compounds

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Abstract

The aim of this research was to investigate an influence of flow velocity on the main parameters (retention time, tailing factor, relative area and number of theoretical plates) of peaks belonging to volatile compounds in the GC- Flame ionization detector analysis. Our results showed that as the flow increases, the retention time declines significantly ($p<0.01$). The other above-mentioned parameters of peaks also change depending on the characteristics of the solvents. The obtained results provide a platform for increasing accuracy of impurity tests for many volatile compounds and development of novel methods of analysis.

Keywords: GC-FID; Flow velocity; Tailing factor; Theoretical plate

Abbreviation: FID: Flame Ionization Detector

Introduction

GC-FID analysis is actively used for determination of purity of volatile compounds in environmental, clinical, pharmaceutical, biochemical, forensic, food science and petrochemical laboratories. The present study allows to optimize a set of conditions [1] for the development of new analytical methods for assessment of concentrations in chromatography. A direct injection of analyzed substances is one of the most widespread and the simplest methods applied [2,3]. The purity of compound is determined in accordance with the relative area of peak of the dominant compound in the mixture. In spite of the fact that various may give a certain inaccuracy, this method is simple in its realization and calculations, and is sufficiently widely used. One of the keys for the successful GC application is the geometrical clarity of the eluted peak boundaries [4]. The tailing factor is defined by the United States Pharmacopeia as the distance from the front edge of the peak to the back edge, divided by the distance from the front edge to the centerline, with all distances measured at 5% of the maximum peak height. In addition, the tailing factor must be close enough to 1 because it is an important prerequisite of correct functioning of majority kinds of software. When an injection of highly concentrated compound is performed, often the ideal form of the peak is not achieved. Dependence of number of theoretical plates (N) on the physical properties of the analyzed compounds was investigated many years ago, in the 60th years of previous century [5,6].

Despite the progress in the development of the chromatographic dynamic models, the studies on the conditions of the GC-FID analysis with regard to the parameters of eluted peaks are rare. Yet, the traditional research has tended to focus on the actual outcomes of the column estimation, rather than on the on the main parameters of peaks. For example, a general rate model permitted reasonably accurate predictions for plug flow through open columns, but it was inaccurate for pressure driven flow cases [7]; the volume averaging model was accurate for some columns [8]. To the best of our knowledge, the question of how conditions of analysis influence the number of theoretical plates was not investigated, as the number of theoretical plates was considered mainly for control of the efficiency of a column [9]. Influence of evolution factor on chromatography was also discussed [10]. Theoretically, a whole complex of physical properties of compounds has an impact on such parameters including the tailing factor and number of theoretical plates. These properties include boiling temperature, enthalpy and evaporation, polarity, dipole moment, viscosity, and specific interactions of the compound with the gas phase [1,2].

A main physical principal of GC chromatography is dynamic equilibrium of sorption/de sorption in the gas flow, the flow velocity is important ruling factor. Connection between retention time and boiling point of compounds is more or less clear but in what way parameters of the peak of the compound are related with properties of this compound is less understood. To the best of our knowledge, an influence of flow velocity on the main parameters of peaks was never considered in such way. The purpose of this study is to investigate the influence of flow velocity on the main parameters of peaks of volatile compounds (solvents) in GC-FID analysis.

Materials and Methods

Compounds

The analyzed compounds (1,4-Dioxane, N,N Dimethylformamide, N,N Dimethylacetamide, Cyclohexanone, Methyl-Ethyl Ketone, N-Propanol, Dichloromethane, Ethyl acetate) were purchased from Merck, Kenilworth, N.J., U.S.A.

GC-FID analysis

Varian GC-800 analytical system was applied with FID detector. The split/splitless liner was int $d_2$mm. A carrier gas was helium UHP. A column (Cyanomethyl- Phenylsilane), 75m×0.53mmi.d was used.

Statistical analysis

Independent experiments were repeated three times. All data were analyzed using Statistica for Windows software (Stat Soft, Inc., Tulsa, OK), and $p<0.05$ was chosen as the minimal acceptable level of significance. Simple regression models were subsequently used to eliminate non-significant effects. Values are presented as means ±SD.

Result and Discussion

Physical properties of the of the analyzed compounds were collected (Table 1). The above-mentioned compounds were selected for GC- Flame Ionization Detector Analysis because of the following reasons: (a) these compounds represent an interesting model for analysis because they have different boiling points and relative polarities (Table 1); (b) these compounds are the most frequently used for impurity test. The novelty of the proposed study lies at estimating the influence of flow velocity on the main parameters of peaks of volatile compounds. (Table 2) demonstrates that once the flow increases, the retention time decreases significantly ($p<0.01$). The approximately linear relation is observed for N-Propanol, Methyl-Ethyl Ketone and Ethyl acetate, but it is highly different from 1 for 1,4-Dioxane, N, N Dimethylformamide, N, N Dimethylacetamide, Cyclohexanone and Dichloromethane. Abnormally high value of the retention time is observed for Dichloromethane at boiling temperature that may be explained by the interaction of its molecules with column phase. The movement of the compounds in the column is determined by three main factors. The first factor is an interaction of the compounds with the active phase of the column mainly because of sorption and as a result of Van der Waals interaction. The second factor is the desorption by means of the column temperature. A balance between the sorption and the desorption depends directly on the column temperature, and indirectly on flow velocity by means of forming the concentration gradient between the sorption and the desorption phases. The third factor is the front migration of the material at the maximal concentration which is proportional to the flow velocity. The retention time is described as (upon condition of constant temperature)

Table 1: Boiling point, relative polarity and topological tolar surface area of the of the analyzed compounds.

Analyte	Boiling Point °C	Relative Polarity	Topological Polar Surface Area, Å^2
1,4-Dioxane	101	0.164	18.5
N,N Dimethylformamide	153	0.386	20.3
N,N Dimethylacetamide	165	6.3	20.3
Cyclohexanone	155.6	0.281	17.1
Methyl-Ethyl Ketone	79.64	0.327	17.1
N-Propanol	97	0.785	20.23
Dichloromethane	39.6	0.309	0
Ethyl acetate	77.1	0.228	26.3

Table 2: Flow, retention time and area of the analyzed compounds.

Analyte	Flow, ml/min		Retention Time, min		Area	
	Result	D (% from Based)	Result	D (% from Based)	Result	D (% from Based)
1,4-Dioxane	4.5	-25	12.22	9.547288212	9624313	-2.400087913
	5	-16.66666667	11.46	2.73419991	9019730.9	-8.531139533
	5.5	-8.333333333	11.445	2.599731062	9819390.1	-0.421816029
	6	0	11.155	0	9860985.317	0
	6.5	8.333333333	10.89	-2.375616316	9501405.85	-3.646486179
N,N Dimethylformamide	9	-10	10.4	2.851491676	1469824.9	-1.583667263
	10	0	10.11166667	0	1493476.6	0
	11	10	9.85	-2.587769903	1514476	1.406074926
N,N Dimethylacetamide	4.7	-6	13.57	1.635251529	1661728	1.735700039
	5	0	13.35166667	0	1633377.467	0
	5.3	6	13.15	-1.510423168	1674978.1	2.546908733
Cyclohexanone	4.9	-9.259259259	15.52166667	-2.563297761	3121584.767	6.452422845
	5.4	0	15.93	0	2932375.5	0
	5.9	9.259259259	15.14	-4.959196485	3007229.95	2.55268979
Methyl-Ethyl Ketone	4.6	-8	10.73	7.694881231	1305803.35	-0.706090325
	5	0	9.963333333	0	1315089.067	0
	5.4	8	9.36	-6.055536969	1348872.8	2.568931199
N-Propanol	5.2	-10.34482759	4.75	11.98428291	2205567.45	-1.177026053
	5.8	0	4.241666667	0	2231836.75	0
	6.4	10.34482759	3.825	-9.823182711	2280297.4	2.171334888
Dichloromethane	4.5	-10	10.04	6.205923836	16896853.65	-6.117738826
	5	0	9.453333333	0	17997919.35	0
	5.5	10	8.965	-5.165726375	17329167.4	-3.715718117
Ethyl acetate	4.3	-4.444444444	14.655	3.973040085	885325.25	-4.066310795
	4.5	0	14.095	0	922851.25	0
	4.7	4.444444444	13.445	-4.611564385	936665.85	1.496947639

RT~1/Flow+⟨o⟩

where RT- Retention Time, o- Irrelevant Factor or as

RT~1/Flow+⟨O⟩

Where O-relevant factor.

If it is $\frac{dRT}{dFlow} < 0$, Then $\frac{dRT}{dFlow} \rightarrow const$, but if it is $\frac{dRT}{dFlow} \neq const$ the strong interaction of the compounds with the column material is observed. In this case the compound cannot be retained in the gas phase. The tailing factor depends on the flow in different ways. For example, there is a significant decrease in the length of the tailing factor ($p<0.01$) in cases of 1,4-dioxane and dichloromethane which was not observed for other tested compounds (Table 3). Of note, 1,4-dioxane and dichloromethane are molecules that have two highly electronegative centers, and they are symmetrical. There is no significant dependence between the flow rate and the relative area of the main peak (Table 3).

Table 3: Tailing factor, number of theoretical plates and relative area of the analyzed compounds.

Analyte	Tailing Factor		Number of Theoretical Plates		Relative Area (%)	
	Result	D (% from Based)	Result	D (% from Based)	Result %	D (% from Based)
1,4-Dioxane	21.74	975.350371	82042	87.90157715	99.93	0.01
	3.87	91.42621599	53408	22.32085313	99.92	0.01
	3.015	49.13437758	48839.69	11.8579976	99.9085	-0.013176989
	2.021666667	0	43662.22	0	99.92166667	0
	1.535	-24.0725474	39569.69	-9.373160595	99.9335	0.01184261

N,N Dimethylformamide	0.53	-2.09476E-14	26754.17	8.49710297	99.9395	-0.000833831
	0.53	0	24658.88	0	99.94033333	0
	0.53	-2.09476E-14	23636.3	-4.146904512	99.94	-0.000333532
N,N Dimethylacetamide	0.54	0.309597523	69917.63	1.659210549	99.944	0.000833807
	0.538333333	0	68776.48	0	99.94316667	0
	0.54	0.309597523	66322.28	-3.568375721	99.945	0.001834376
Cyclohexanone	0.541666667	0.308641975	57294.79	-12.33821576	99.93733333	-0.00066708
	0.54	0	65358.91	0	99.938	0
	0.54	0	52699.13	-19.36963912	99.939	0.00100062
Methyl-Ethyl Ketone	1.595	3.908794788	39884.13	3.479676117	99.9895	0.026176265
	1.535	0	38542.96	0	99.96333333	0
	1.56	1.628664495	37265.49	-3.314405966	99.986	0.022674981
N-Propanol	0.58	-0.571428571	2438.09	-10.34482759	99.998	0.043519802
	0.583333333	0	2413.702	0	99.9545	0
	0.58	-0.571428571	2417.23	10.34482759	99.998	0.043519802
Dichloromethane	46.91	5.676954269	4701.755	-1.168682026	99.966	-0.010502468
	44.39	0	4757.353	0	99.9765	0
	40.5	-8.763234963	5310.94	11.63644211	99.964	-0.012502938
Ethyl acetate	0.715	0.486486486	36549.2	4.785935996	99.9805	0.010002951
	0.711538462	0	34879.87	0	99.9705	0
	0.72	1.189189189	33806.72	-3.076721947	99.982	0.011503394

The number of theoretical plates depends on the flow that is connected with polarity and properties of the compounds. For N-Propanol and Methyl-Ethyl Ketone these dependencies are the same, while in case of N, N Dimethylformamide and N, N Dimethylacetamide (the compounds with similar structures) the number of theoretical plates depends on the flow qualitatively, but not quantitatively (Table 3).

Concluding Remarks

Our results show that as a result of flow increase, the retention time declines. The remaining parameters of peaks vary according to the properties of the solvents. The obtained results may allow increasing the accuracy of impurity test for many volatile compounds, and may facilitate development of novel methods of analysis for impurity test by understanding of ruling parameters. The further investigations are important in order to elucidate the influence of the column phase and the temperature gradient on the main parameters of peaks of volatile compounds in the GC-FID analysis.

References

1. Pollock GS, Eldridge RB (2000) Neural network modeling of structured packing height equivalent to a theoretical plate. Ind Eng Chem Res 39: 1520-1525.
2. Babuška I, Li L (1992) The problem of plate modeling: Theoretical and computational results. Computer Methods in Applied Mechanics and Engineering 100(2): 249-273.
3. Velayudhan A, Ladisch MR (1993) In: Tsao GT (Ed.), Springer Berlin Heidelberg, Berlin, Germany, pp. 123-145.
4. Shou M, Qiu H (2020) Development of a rapid GC-FID method to simultaneously determine triethylamine, diisopropylamine, and 1,1,3,3-tetramethylguanidine residues in an active pharmaceutical ingredient. Journal of Pharmaceutical Analysis 11(2): 251-256.
5. Wicke E (1967) J. C. Giddings: Dynamics of Chromatography. Part. I: Principles and Theory. Marcel Dekker, New York, 1965. XII und 323 Seiten. 39 Abb. Preis:$ 11.50. Reports of the Bunsen Society for Physical Chemistry 71(2): 236-236.
6. Kucera E (1965) Contribution to the theory of chromatography: Linear non-equilibrium elution chromatography. J Chromatogr 19(2): 237-248.
7. Lieres E, Andersson J (2010) A fast and accurate solver for the general rate model of column liquid chromatography. Computers & Chemical Engineering 34(8): 1180-1191.
8. Yan X, Wang Q (2013) Comparative analysis of chromatography dynamic models in predicting the plate height contributed by interphase mass transfer. Chemical Engineering Science 104: 760-766.
9. Stauffer E, Dolan JA, Newman R (2008) Fire Debris Analysis. (1st edn.), Academic Press, USA, pp. 1-672.
10. Kaiser RE (1977) The correct measurement and interpretation of evaluation factors in chromatography: The real plate number, the separation number, the dosage quality. Part 1: Gas chromatography. Chromatographia 10(6): 323-338.