



Trade Science Inc.

November-December 2005

Volume 1 Issue 1-2

# Analytical CHEMISTRY

An Indian Journal

Full Paper

ACAIJ, 1(1-2), 2005 [27-31]

## Study Of Factors Effects On Static Headspace Gas Chromatography



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Received: 24<sup>th</sup> August, 2005

Accepted: 10<sup>th</sup> September, 2005

Web Publication Date : 13<sup>th</sup> October, 2005



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### ABSTRACT

Depending on the determination of ethanol and benzene in water, we studied the theory of static headspace gas chromatography, and discussed the effect of vapor-liquid phase ratio, distribution constant, equilibrium temperature, equilibrium time and inorganic salts on headspace concentration. The results showed that the effect of these factors on headspace concentration varied significantly in various volatile residual solvents in pharmaceuticals.

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### KEYWORDS

Static headspace gas  
chromatography;  
Vapor-liquid phase ratio;  
Distribution constant;  
Equilibrium temperature;  
Equilibrium time.

### INTRODUCTION

Residual solvents in pharmaceuticals are defined as organic volatile chemicals that are used or produced in the manufacture of drug substances or excipients, or in the preparation of drug products. The solvents are not completely removed by practical manufacturing techniques. Appropriate selection of the solvent for the synthesis of drug substance may enhance the yield, or determine characteristics such as crystal form purity, and solubility. Therefore, the solvents may sometimes be a critical parameter in the synthetic process. However, there is no therapeutic benefit from residual solvents; all residual

solvents should be removed to the extent possible to meet product specifications, good manufacturing practices or other quality-based requirements. Drug products should contain no higher levels of residual solvents than can be supported by safety date. The content of solvents in pharmaceuticals should be evaluated and justified.

The common method to determination of residual solvents in pharmaceuticals is gas-chromatography (GC). According to the ways of injection, they are divided into the direct injection with solution and the headspace injection. There are many disadvantages of direct injection with solution, such as the solvent interference, column contamination and

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high, detection limit etc. The advantages are simple and convenient. Recently, solid-phase micro-extraction<sup>[1-6]</sup> has been used to determine the residual solvents in pharmaceuticals, however, it costs too much and the repeatability needs to be improved. The headspace technique has two ways. One is dynamic<sup>[7-11]</sup> (purge and trap) with lower detection limit, but the analytical period is too long and needs special facilities. The other is static<sup>[11-19]</sup> (vapor-phase extraction) which has been applied widely. Compared with the dynamic the static has some particularly advantages when they are used to analysis the residual solvents in pharmaceuticals. Not only has the analytical procedure been simplified, but also with short analytical time. The key problem is the headspace concentration is too low. This paper mainly discussed the factors effect on static headspace gas chromatography.

### Principle

In isothermal and hermetical vapor-liquid phase system in which the volatile chemicals reached to equilibrium, the relationship among headspace concentration, distribution constant and vapor-liquid phase ratio according to the conservation of mass is below:

$$C_g = C_0 / (\beta + K) \quad (1)$$

Where  $\beta$  is the phase ratio, the ratio between the volume of the gas phase ( $V_g$ ) and that of the liquid phase ( $V_w$ ) ( $\beta = V_g/V_w$ )  $K$  is the distribution constant, the ratio of the equilibrium concentration of a solute in the liquid phase ( $C_w$ ) to that in the vapor phase ( $C_g$ ) in chromatography ( $K=C_w/C_g$ ). Therefore, the inverse of  $K$  is equal to the Henry's constant. It can be seen from equation (1) that as long as determining  $C_g$ , we can obtain the original concentration of volatile organic compounds in water ( $C_0$ ).

## EXPERIMENTAL

### Apparatus and reagents

The headspace apparatus included GC-4009 Gas Chromatograph with a flame ionisation detector (FID) (Beijing East & West Analytical Instruments, China), N2010 processing station (Zhejiang Univer-

sity Zhida Information Engineering Co., Ltd, Hangzhou, China), AG-1605 air pump (Beijing Keep-Science Analysis Sci & Tech Co.Ltd. Beijing, China), 1102GC column oven (Shanghai Analytical Instruments, Shanghai, China), 1.0 ml injectors (Shanghai-mishawa medical Co.Ltd. Shanghai, China), headspace vials (12.6 ml, self-prepared), PTFE coated butyl rubber septa (Agilent, USA), Pincher which was used to press cap stopper (Agilent, USA). Ethanol (Chromatogram-grade; Purity=99.8, Kangkede, Tianjin, China), Benzene (chromatographic standard, Beijing chemical plant, Beijing, China), Sterile water for injection (Pinghu Shapu Aisi, Zhejiang, China), Sodium chloride (A.R. Purity=99.5%, Tianjin University. Tianjin, China).

### Headspace gas-chromatography parameters

Gas chromatography was achieved with a capillary column DB-WAX (J&W Scientific, 30 m×0.25 mm I.D×0.25  $\mu$ m). Nitrogen was carrier gas with a flow rate of 3.0 ml min<sup>-1</sup>. Column temperature was at 50°C. Both the injector and the detector temperature were both at 160°C. The split ratio was 10:1. The injection volume of the headspace vapor was 1.0 ml.

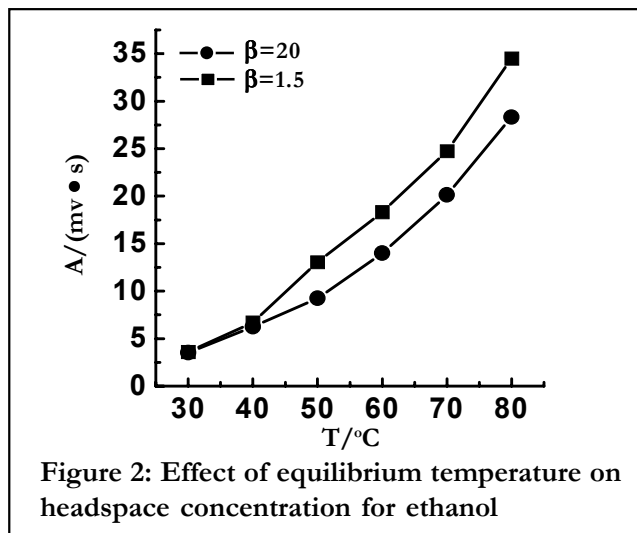
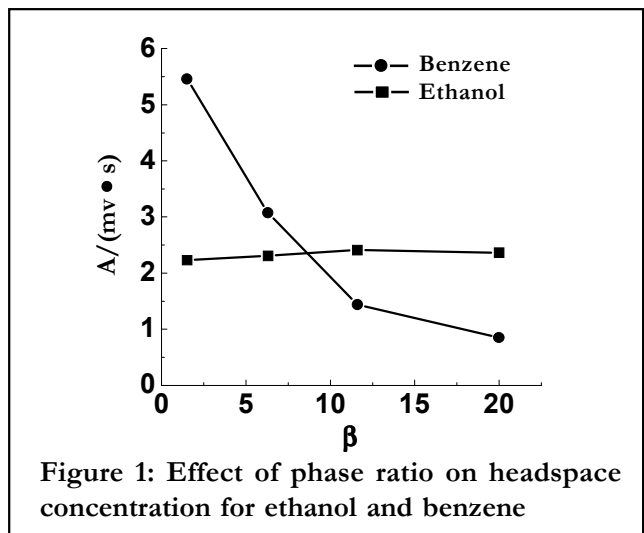
### Procedure

Add certain volume of ethanol-water solution (80.7  $\mu$ g ml<sup>-1</sup>) into headspace vials and close the vials at once and then put them into the oven respectively. When the vials were achieved equilibrium, GC analyzed 1.0 ml headspace vapor. The benzene-water solution was discussed the same as the ethanol-water solution above.

## RESULTS AND DISCUSSION

### Influence of vapor-liquid phase ratio

After lay aside an hour in 25°C the samples were analyzed by GC. The headspace chromatographic peak area of ethanol and benzene in different vapor-liquid phase ratio was shown in figure 1. We could see that there was little influence on the peak area of ethanol with the different vapor-liquid phase ratio. But the peak area of benzene changed sharply from 20 to 1.5, which increased six times. The re-



sults were indicated that there was obvious influence on headspace concentration through the change of vapor-liquid phase ratio to the component with small distribution constant or big Henry's constant. This conclusion was the same as the result of calculation according to eq. (1). Eq. (1) showed that in a certain vapor-liquid equilibrium system,  $K$  and  $C_0$  was constant,  $\beta$  was direct proportion to headspace vapor concentration, in other words, when the liquid phase ( $V_w$ ) increased,  $\beta$  decreased and  $C_g$  increased. So if the injection volume of different samples was the same, the peak area must increase. But the peak area for factual samples was also related with  $K$ . If  $K \gg \beta$ ; the eq. (1) was considered as:  $C_g \approx C_0/K$ , there was little influence on the peak area with the change of sample volume, or the influence was serious.

#### Influence of distribution constant<sup>[20]</sup>

We could observe the influence of distribution constant from format (1). When vapor-liquid phase ratio was 1.5 and distribution constant was more than 10, the headspace concentration decreased sharply; meanwhile, when distribution constant was 20, there was little influence.

#### Influence of equilibrium temperature

The equilibrium of vapor-liquid was controlled by temperature, in headspace GC, there was great influence on distribution constant  $K$  and little on  $\beta$  with the change of temperature. We must consider both of them. In a certain sample system,  $\beta$  was con-

stant, the headspace concentration was inverse proportion to distribution constant  $K$ . From eq. (1) we knew, when  $K \gg \beta$ , the influence of temperature was obvious; when  $K \ll \beta$ ,  $K$  increased with the increasing of temperature, but  $K + \beta$  changed a little, so the change of the headspace concentration was tiny. From the results (Figure 2 and Figure 3) of the ethanol-water solution and the benzene-water solution in different temperature after lay aside 30 minutes we concluded that there was great influence on the ethanol-water solution with big  $K$ , there was little influence on the benzene-water solution with small  $K$ , in other words, when  $K < 5$ , there was meaningless to enhance headspace concentration depending on increase of equilibrium temperature.

#### Influence of equilibrium time

The analysis cycle of headspace GC usually lived up to equilibrium time, so to decrease equilibrium time was the key to improve the speed of headspace GC. Equilibrium time was rested with the diffusion speed of determined component molecule from sample solution to vapor phase. The faster the diffusion speed was, the higher the molecular-diffusivity and the shorter the equilibrium time was. In addition, the molecular-diffusivity depended on the size of molecules, the viscosity of medium and temperature. The higher the temperature was, the lower the viscosity of medium was and the higher the molecular-diffusivity was. So to increase the temperature could save equilibrium time. When equilibrium tem-

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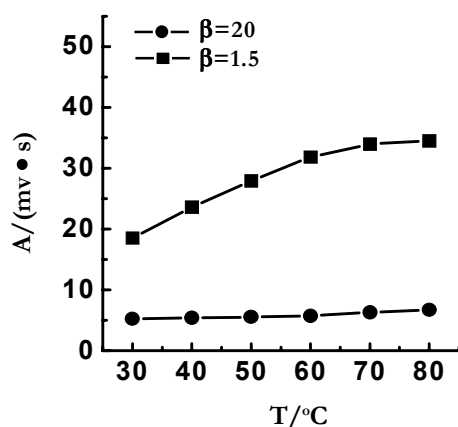


Figure 3: Effect of equilibrium temperature on headspace concentration for benzene

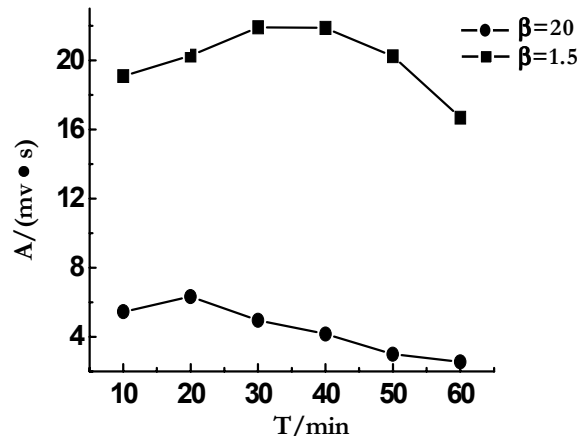


Figure 4: Effect of equilibrium time on headspace concentration for benzene

perature was at 60°C and the phase ratios were 1.5 and 20, the influence on ethanol and benzene were shown in figure 4 and figure 5. The results were indicated that the higher the phase ratio was, the shorter equilibrium time was. For component with small  $K$ , to decrease phase ratio could enhance  $C_g$  greatly and prolonged equilibrium time; the component with big  $K$ , to decrease phase ratio had little effect on increasing headspace concentration. So in the system with small  $K$ , we could save equilibrium time depending on enhancing phase ratio.

### Influence of inorganic salt

Activity coefficient ( $\gamma$ ) could describe the interaction between solute and solvent molecule. The distribution constant was inverse proportion to the vapor pressure multiplied with activity coefficient:

$K=1/(\rho \cdot \gamma)$ , so we could increase the headspace concentration by increasing activity coefficient. The common way was to add inorganic electrolyte into sample water solution that is so called salting-out effect. This way could also effectively reduce the matrix effect in factual sample. To add 0.0, 0.2, 1.0, and 1.5 g NaCl to headspace vials, respectively, and then add 5.0 ml of ethanol solution ( $80.7 \mu\text{g ml}^{-1}$ ) and 5.0 ml of benzene solution ( $0.879 \mu\text{g ml}^{-1}$ ), lay aside 30 minutes in 40°C. The relationship between headspace peak area of ethanol and benzene and the concentration of NaCl, was shown in figure 6. We found that salt with low concentration had no influence. So we chose high concentration or saturated salt solution. When the phase ratio was 1.5, NaCl could make headspace chromatographic peak area of ethanol enhance three times, but little on

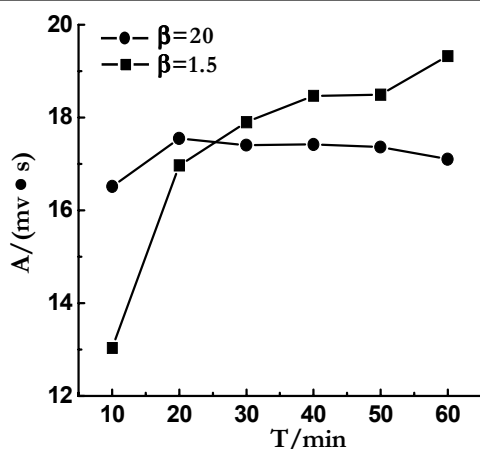


Figure 5: Effect of equilibrium time on headspace concentration for ethanol

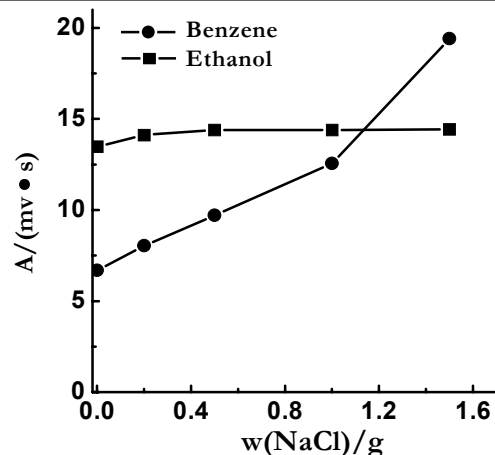


Figure 6: Effect of inorganic salt on headspace concentration

benzene, it was shown that the salting-out effect is selective, generally the influence on the polar components was greater than on the non-polar components.

### CONCLUSION

In this paper, we introduced the mechanism of static headspace gas chromatography. Compared with the dynamic the static has some particularly advantages when they are used to analysis the residual solvents in pharmaceuticals. Not only has the analytical procedure been simplified, but also with short analytical time. This method has been applied to the determination of various volatile residual solvents in pharmaceuticals.

### ACKNOWLEDGMENTS

The Natural Science Foundations of China and Hebei Province, and a Visiting Scholar Foundation of the key Laboratory at Hebei University supported this work.

### REFERENCES

- [1] J.Pawliszyn; 'Solid Phase Microextraction-Theory and Practice', Wiley-VCH, New York, (1997).
- [2] M.Chiarotti, R.Marsili, A.M.Pineiro; J.Chromato.B, **772**, 249-256 (2002).
- [3] S.A.Coran, V.Gainnellini, S.Furlanetto et al.; J. Chromato. A, **915**, 209-216 (2001).
- [4] S.Scypinski, A.M.Smith; Wercinski S.A.S., Dekker M., 'Solid Phase Microextraction A Practical Guide', New York, USA, 111 (1999).
- [5] H.Lord, J.Pawliszyn; J.Chromato.A, **885**, 153 (2000).
- [6] A.R.Raghani; J.Pharm. and Bio., **290**, 507-518 (2002).
- [7] T.P.Wampler, W.A.Bowe, E.J.Levy; J.Chromato.Sci., **23**, 64-67 (1985).
- [8] V.N.Thomas, T.A.Ramstad; Acta Pharm.Nord., **4**, 97-104 (1992).
- [9] T.D.Cyr, R.C.Lawrence, E.G.Lovering; Pharm.Res., **9**, 1224-1226 (1992).
- [10] F.Ruchatz, P.Kleinebudde, B.W.Müller; J.Pharm., **142**, 67-73 (1996).
- [11] Claudia Witschi, Eric Doelker; J.Pharm. and Bio., **43**, 215-242 (1997).
- [12] C.Bicchi, A.Bertolino; Farmaco.Ed.Prat., **37**, 88-97 (1981).
- [13] J.P.Guimbard, M.Person, J.P.Vargnaud; J.Chromato., **403**, 109-121 (1987).
- [14] D.Herlitz, G.Herold, H.Nufer; Pharm.Ind., **49**, 200-203 (1987).
- [15] D.Herlitz, G.Herold, W.Neckel, H.Nufer; Pharm.Ind. **50**, 251-256 (1988).
- [16] H.Hachenberg; 'Die Headspace Gaschromatographie als Analysen-und Meâmethode', DANI Verlag, Frankfurt, (1988).
- [17] Z.Penton; J.High Resolution Chromato., **15**, 329-331 (1992).
- [18] J.M.Kevin, W.B.Thomas, F.C.David et al., J.Chromato. B, **686**, 85-95 (1996).
- [19] K.Theresa, Natishan, W.Yan; J.Chromato.A, **800**, 275-281 (1998).
- [20] Y.H.Wang; Journal of Peking University, **39**, 96-900 (2003).