CHAPTER 4

CONCLUSIONS AND SUGGESTION

FOR FURTHER WORK

4.1 Conclusion

A gas diffusion unit combined with chemiluminescence (CL) detection in the continuous-flow system was developed for the determination of nitrite and nitrate. The gas diffusion chemiluminescence systems were fabricated from easily available materials and instruments. The proposed method presents good reproducibility, high sensitivity (RSD =1.55%) and sample throughput (60 h^{-1}). The method is simple and inexpensive. This method was successfully applied to the determination of nitrite and nitrate in fermented food products.

This research work consists of two parts. The first was tested for the sensitivity of chemiluminescence reactivity of nitrite by a simple flow injection chemiluminescence method (FI-CL), which is based on CL light induced by luminol in an alkaline medium. The preliminary flow injection configuration used for the determination of nitrite was designed to provide reaction conditions for magnifying and enhancing effect on the CL generated by the reaction of luminol in the presence of hydrogen peroxide (H_2O_2) media as described in Figure 2.1. This method involves the injection of each nitrite standard into sulfuric acid carrier stream,which then is merged at a T-piece with stream of H_2O_2 solution. The reagent stream is consisting of luminol in alkaline medium. In aqueous solutions, the most commonly used chemiluminescent species is luminol (5-amino-2,3-dihydrophthalazine-1,4-dione), which reacts with H_2O_2 in alkaline solution to yield 3-aminophthalate in an excited electronic state which returns to ground state with the production of light. The light intensity can easily be monitored with a photomultiplier tube with no wavelength discrimination, then the intensity of enhanced emission is proportional to the concentration of nitrite ; thus the amount of nitrite can be determined by measuring the increase in CL intensity. This CL reaction seems promising as a basis for development of a gas diffusion flow injection chemiluminescence procedure for nitrite determination. The FI-CL system was modified to a novel GDFL-CL system for determination of nitrite in real samples base on CL induced by luminol and hydrogen peroxide in an acidic medium. Optimum conditions for determining nitrite were investigated. Various factors influencing the sensitivity of the method were optimized using the univariate method. The optimum conditions are summarized in Table 3.1. The linear calibration graph over the ranges of 1×10^{-8} - 7×10^{-7} mol L^{-1} was established (Figure 3.14) with a regression equation: $Y = 1 \times 10^{8} X + 3.8983$ for 1×10^{-8} – 7×10^{-7} mol L^{-1} nitrite with the correlation coefficient of 0.9990. The method was sensitive as little as 1×10^{-8} mol L^{-1} of nitrite could be determined. The reproducibility obtainable for determining 1×10^{-7} mol L^{-1} of nitrite standard by the proposed method (n=12) was 1.55%. The method has been satisfactorily applied to the assay of nitrite in food samples with the sample throughput of 60 h^{-1}. Validation of the proposed method for nitrite determination was also performed by comparison of the results obtained by both the proposed and the standard method (AOAC, Colorimetric) using the same samples. The flow based method is simple, inexpensive, accurate and reproducible which is suitable for the monitoring of nitrite in food samples. The nitrite contents found in food samples were found to be over the range of 4.08×10^{-4} – 6.67×10^{-4} mg kg^{-1}, which value can be accepted with Food and Drug Administration (FDA).

The second part involved the development of the on-line photoreduction of nitrate to nitrite. Regarding to results obtained in the first part, the good sensitivity obtained by the proposed gas diffusion flow injection chemiluminescence system led to further investigation to develop the on-line photoreduction device. The on-line photoreduction gas diffusion flow injection chemiluminescence system was based on photoreduction of nitrate to nitrite using a low pressure UV lamp set at 254 nm wavelength. Its sensitivity was improved by using zinc oxide nanoparticles as catalyst. The optimum conditions are summarized in Table 3.6. The linear calibration graph over the ranges of 4×10^{-3} – 1×10^{-1} mol L^{-1} was established (Figure 3.20) with a regression equation: Y = 206.47X + 3.7807 nitrate (r^2 = 0.9990). The method was sensitive at 4×10^{-3} mol L^{-1} of nitrate could be determined. The reproducibility of nitrate standard by the proposed method (n=12) was 2.56% with the sample throughput of 10 h^{-1}. Validation of the proposed method for nitrate determination was also performed by comparison of the results obtained by both the proposed and the standard method (AOAC, Xylenol method) using the same samples. The method is simple, inexpensive and accurate for the monitoring of nitrate in food samples. It was found that the concentration of nitrate in food samples over the range of 7.04 – 51.46 mg kg^{-1}, which value was acceptable with Food and Drug Administration (FDA) regulation.

4.2 Suggestion for further work

This developed system could be used for routine determination of nitrate and nitrite in many sources of samples such as food samples, cosmetic samples, pharmaceutical formulations and biological materials.

In order to improve the sensitivity, the effect of surfactants should be investigated for determination of nitrate and nitrite.

Moreover, an alternative Zn minicolumn (McKelvie, 1999) reduction could be used for determination of nitrate and nitrite in on-line flow injection system.