A Novel, non-invasive method of measuring in-pack oxygen concentration and its application in the study of staling of a fruit-flavoured alcoholic beverage

R.I. White, C.A. Boulton, A. Mundy

Coors Brewers Ltd., Technical Centre, P.O. Box 12, Cross Street, Burton-on-Trent, Staffs. DE141XH, United Kingdom. E-mail -: rod.white@coorsbrewers.com

SUMMARY

Current techniques for the measurement of oxygen concentration in small-pack products are destructive. Here is described a non-invasive optical method of oxygen measurement suitable for glass and PET packages. The results are compared with those obtained using an invasive electrochemical procedure. The optical technique has two key advantages. Firstly it can be used to measure the oxygen concentration in both the liquid and gas headspace. Secondly, the oxygen concentration in individual containers can be measured repeatedly over time. This technique has been used to monitor the oxygen concentration in a bottled non-carbonated fruit flavoured alcoholic beverage.

INTRODUCTION

Dissolved oxygen concentration is an important determinant of flavour stability of alcoholic beverages. Oxidative reactions are associated with the formation of compounds that contribute to staling. These reactions occur slowly throughout the shelf life of the product. In order to minimise these effects it is usual to take steps to ensure that products are packaged with a very low oxygen concentration. In the case of carbonated products packaged into glass bottles this is achieved by a high-pressure water jet, which causes foaming in the headspace of the bottle and consequently air to be expelled. However, in the case of non-carbonated products high pressure jetting is not possible and alternative strategies must be employed. For example, filling bottles under an atmosphere of an inert gas such as nitrogen.

The in-package oxygen concentration is usually determined via an electro-chemical technique using a flow-through chamber containing an oxygen electrode [1]. A disadvantage of this procedure is that the package must be pierced in order to take a measurement.

In this study, a novel non-invasive fluorimetric method is described that is able to measure simultaneously oxygen concentrations in both the liquid and gas phases of a bottled non-carbonated fruit flavoured alcoholic beverage. The results are compared with those obtained using an electrochemical technique.

Principle of operation

The equipment used in this study is termed the OxySense 101 (OxySense, Inc., Las Vegas, Nevada, USA). It relies on the properties of a ruthenium based metal organic fluorescent dye. To ensure durability and for ease of handling the dye is immobilised in a gas permeable hydrophobic polymer mixture coated onto a small glass disc,

termed an *Oxydot*. Discs can be placed anywhere on the inner surface of the bottle allowing for oxygen measurements to be made at any discrete point within the bottle, including the headspace (gas phase) or the liquid phase. The dye absorbs light between 400 - 500 nm and has a strong emission peak at 600nm. The width of the emitted peak provides a measure of the duration of fluorescence. It is inversely related to oxygen concentration since the latter exerts a quantifiable quenching effect on the duration of fluorescence.

The fluorimeter takes the form of a "pen" which, in order to make measurements, is placed on the surface of the bottle adjacent to the disc (Figure 1). The pen is linked via optical fibre to an associated electronics package that regulates the high voltage supply to the photomultiplier detector.

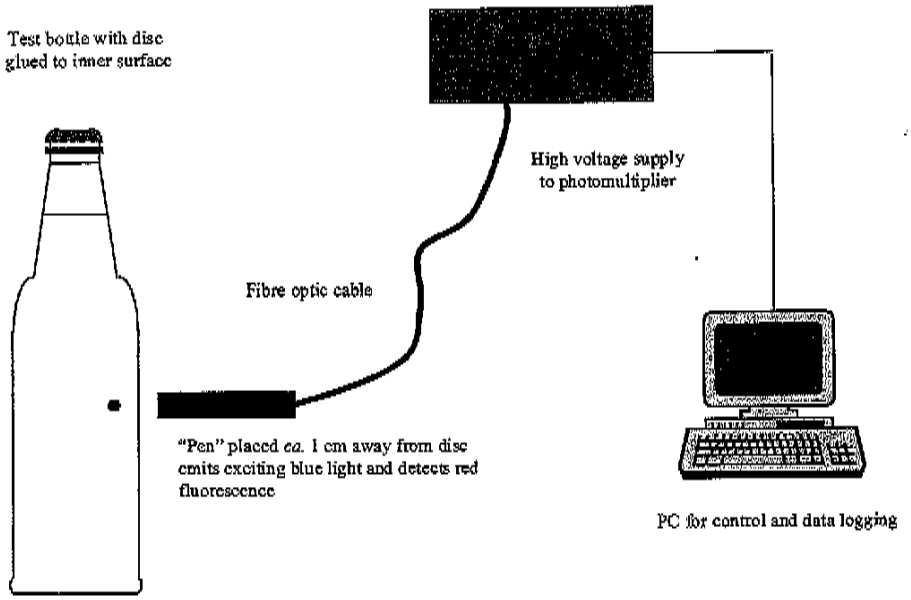

Figure 1. Principal components of the fluorimetric device for measuring in-pack oxygen concentration

The signal from the fluorimeter is transferred to a computer. The fluorescence signal is displayed in real time on a graphical display (Figure 2). When a stable signal is observed the data (average of 1000 pulses) is captured using a computer mouse or keyboard command. The magnitude of fluorescence is temperature-dependent and before making measurements an appropriate value must be entered into the software loaded onto the computer. In addition, specific calibration factors, supplied with individual batches of discs, must also be entered.

The colour of the glass is important. The technique can be used most successfully on clear glass. In the case of green glass the signal is weaker and the gain of an amplifier must be increased in order to make measurements. This increases both the

sensitivity and background noise of the instrument. In this situation, in order to make accurate measurements the averaged results from several replicate bottles should be

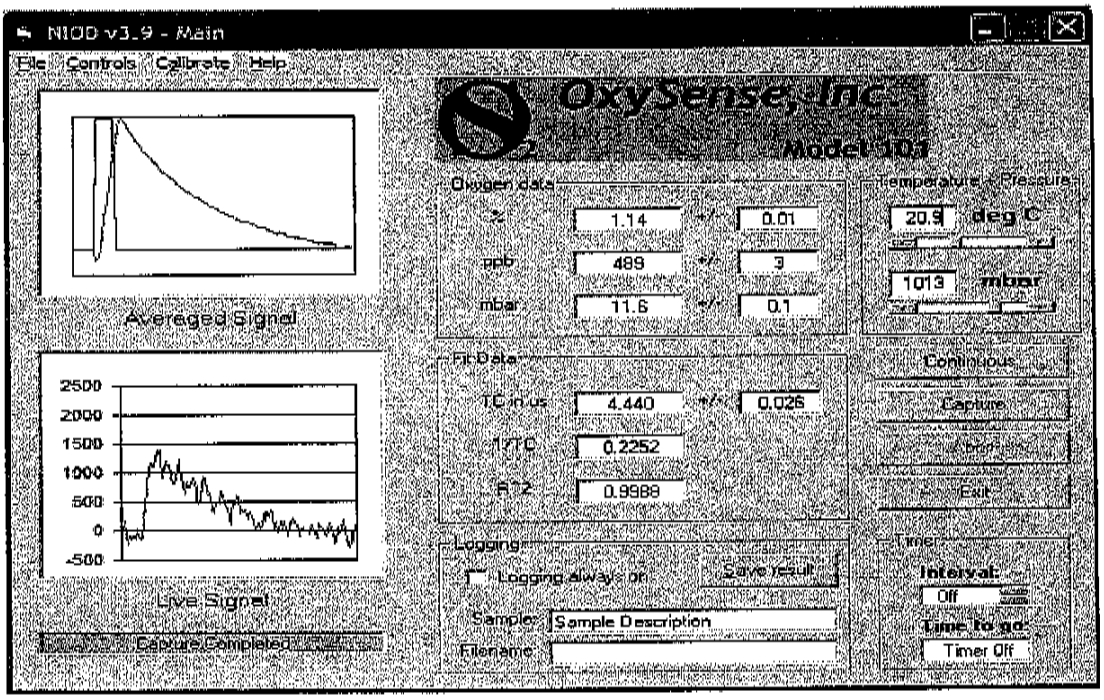

Figure 2. Computer screen showing the graphical display of fluorescence (bottom left) and readout of oxygen concentration (ppb)

obtained. In the case of brown glass the signal is completely absorbed rendering this technique unusable.

METHODS

Electrochemical dissolved oxygen measurements were made using an invasive technique [1] in which the liquid contained within bottles removed from the pacakging line was forced via the application of CO_2 top-pressure into a measuring chamber containing a polarographic oxygen electrode.

For the fluorimetric procedure Oxydot disc(s) were glued onto the insides of clean empty bottles using a silicon-based oxygen-permeable adhesive and a special applicator provided for this purpose. Bottles, with attached discs, were placed on the filler in-feed conveyor, filled and capped. Test bottles were labelled so that they could be easily recognised and recovered from the packaging line prior to analysis. The filler was run at normal speed and test bottles were thereby filled, together with normal bottles, under standard conditions.

RESULTS AND DISCUSSION

The fluorescent technique has been applied to a study of the measurement of oxygen concentration in the headspace and liquid phases of a bottled non-carbonated fruit flavoured alcoholic beverage. The results were compared with those obtained using the electrochemical technique. In previous investigations using the electrochemical procedure it was observed that the results were not reproducible. For these analyses bottles were agitated for 5 min using a mechanical shaker operating at 200rpm. The electrochemical method produced satisfactory and reproducible results with carbonated products. This suggested that the control of dissolved oxygen during packaging of the non-carbonated product was inadequate. Alternatively, the procedure used to measure dissolved oxygen in this product could have been the source of the inconsistency.

The effect of agitation time on dissolved oxygen measurements of the non-carbonated product was investigated. It was shown that using the electrochemical procedure, bottles required to be agitated for at least 15 min in order to obtain a stable and maximum dissolved oxygen concentration (Figure 3). The results of fluorimetric analyses made within the liquid phase were essentially identical to those obtained with the electrochemical method. Conversely, the oxygen concentrations in the gas headspaces of bottles declined with increased agitation time.

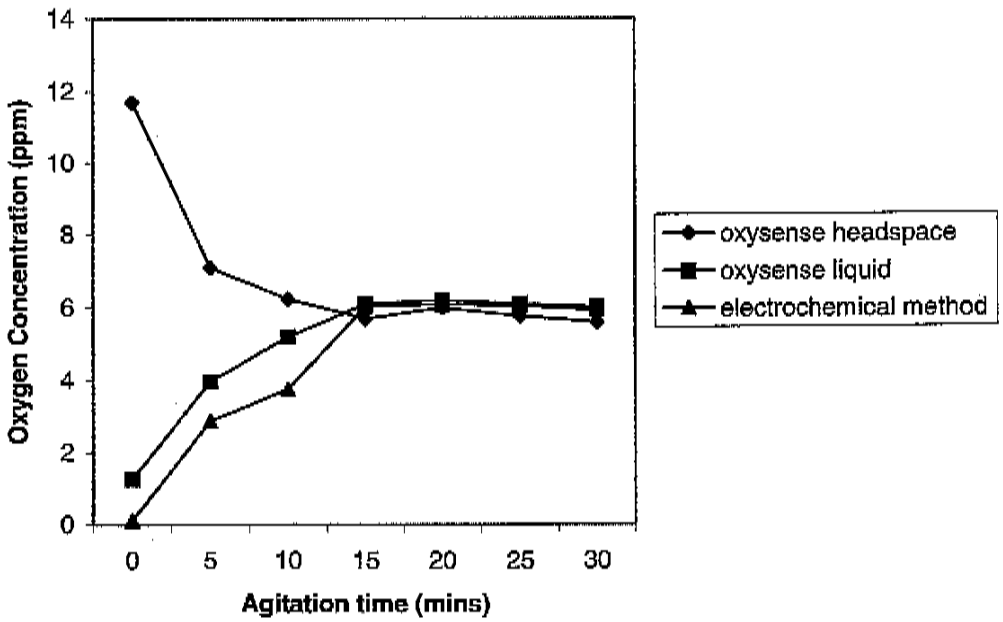

Figure 3. Effect of agitation time on dissolved oxygen concentration of a non-carbonated fruit flavoured alcoholic beverage measured by electrochemical and fluorimetric methods. Fluorimetric analyses are the means of determinations made with 5 individual bottles. Each bottle was fitted with one disc in the headspace and another half way down the bottle in the liquid phase. Electrochemical analyses are the means of determinations made with 5 individual bottles at each time point.

The results shown in Figure 3 indicated that 5 minutes agitation was insufficient to achieve equilibrium in dissolved oxygen concentration between the gas and liquid phases in the bottles. Furthermore, the lack of equilibrium was due to the oxygen concentration within the bottle headspace being higher than might be expected. The differences in oxygen concentrations in the gas and liquid phases of bottles immediately after packaging were immediately detectable using the fluorimetric technique. Measurements of oxygen concentration in the liquid phase after 5 minutes agitation were under estimates of the true in pack oxygen concentration.
The heterogeneity of oxygen concentration within bottles soon after filling and with no agitation was investigated by making measurements using discs located at different heights within individual bottles. The results confirmed that there was a gradient of decreasing oxygen concentration from the headspace throughout the liquid (Figure 4).

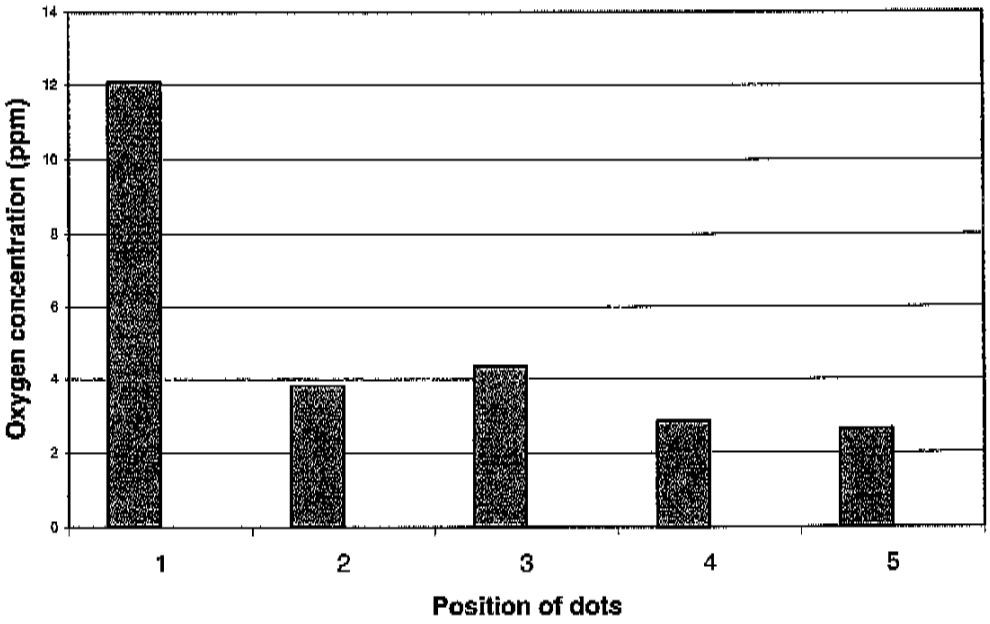

Figure 4. Oxygen determinations made at different locations within bottles (1 = headspace, 2 = bottle neck, 3 = bottle shoulder, 4 = bottle body upper, 5 = bottle body lower) via the fluorescence technique. Analyses (means of 5 individual bottles) were performed on non-agitated bottles 1h after filling.

An advantage of the fluorescence technique is the ability to measure oxygen concentration within the same sample over a period of time. The change in oxygen concentration in bottles of two different fruit flavours of a non-carbonated alcoholic beverage during 40 days is shown in Figure 5. In both liquids oxygen concentration declined throughout the storage period. The patterns of decline in oxygen concentration were different for each liquid although in both cases oxygen disappeared most rapidly in the first 10 days after packaging.

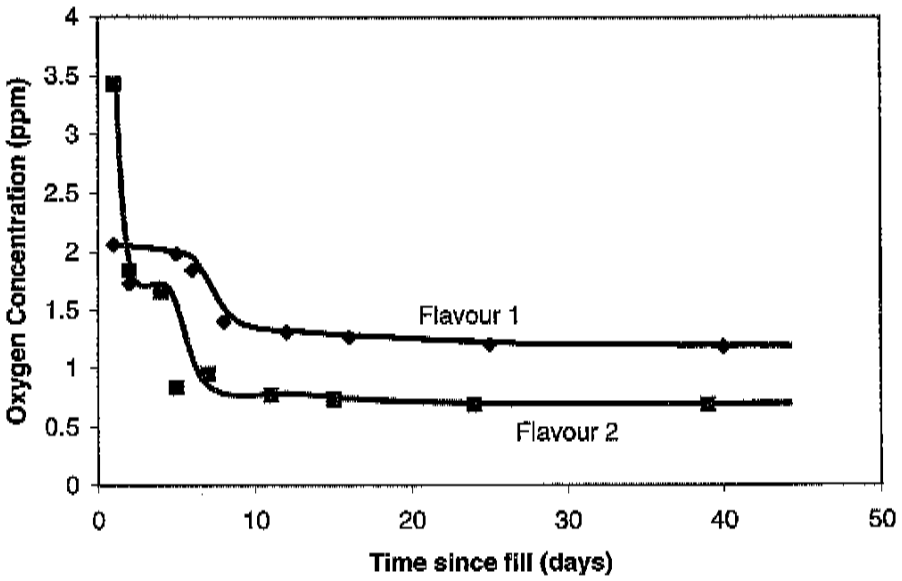

Figure 5. Change in dissolved oxygen concentration with time in individual bottles of two fruit flavours of a non-carbonated alcoholic beverage. Bottles were filled on a production packaging line. Fluorescence measurements (mean of 5 bottles) were made after mechanical agitation for 15 min at 200 rpm to ensure equilibration of oxygen concentration between the liquid and gas phases.

Sensory analysis of the flavoured alcoholic beverage used in these tests has indicated that staling occurs over a period of time. It has been assumed that these undesirable changes are partially the result of oxidation reactions.

CONCLUSIONS

The technique described allows the accurate determination of oxygen concentration in bottled beverages. The results are comparable with those obtained by electrochemical methods. The technique offers several advantages. Firstly it allows simultaneous measurement of oxygen concentration in both the gas and liquid phases. Secondly the same package can be repeatedly measured, enabling oxygen concentrations to be monitored throughout key processes such as pasteurisation and storage. Thirdly the technique could be especially useful for monitoring new package types such as barrier-protected PET.

In future studies it is intended to correlate changes in oxygen concentration with the formation of chemical markers of staling.

The technique has been applied to a study of in-pack oxygen concentration in a bottled non-carbonated Product with a relatively high oxygen concentration. In future

work we will examine the accuracy of the technique at the low oxygen concentrations found in beer.

REFERENCES

[1] European Brewery Convention (1998) *Analytica-EBC*, Method 11.5, *Total oxygen in packaged beer*, Fachverlay Hans Carl, Nurnberg, Germany.

ACKNOWLEDGEMENTS

The authors thank the directors of Coors Brewers for permission to publish this paper.