J. Titze, H. Walter, F. Jacob, A. Friess and H. Parlar

Evaluation of a new Optical Sensor for Measuring dissolved Oxygen by Comparison with Standard Analytical Methods

The determination of the concentration of dissolved oxygen (O_2) plays an important role in different stages of beer production. Only precise, stable and fast measurement procedures for determining the oxygen content in beer are therefore suitable for the use in breweries. A research project was carried out in order to evaluate the newly developed oxygen measurement system built in the CO_2/O_2 Gehaltemeter type c-DGM of Haffmans BV by comparison with other standard analytical references such as the Orbisphere 3650 from Hach Ultra Analytics GmbH and the DIGOX 6 from Dr. Thiedig & Co. The purpose of this study was to determine the precision and accuracy as two main criteria for the evaluation of a suitable oxygen measurement in the brewing industries, since the precision describes the quality of a measurement procedure and the accuracy the level of agreement between the indicated and real value. The measurement system of the c-DGM was compared with the certified reference systems during a series of tests. The measurement methods of the reference systems are specified in Brautechnische Analysenmethoden, volume II of the Mitteleuropäische Brautechnische Analysenkommission (MEBAK) and are considered to be the recognized procedure for oxygen regulation in the brewing industry. It was observed that in comparison to the reference systems, the oxygen measurement system built in the c-DGM exhibited an effective work performance regarding precision and accuracy, and moreover signalized a fast response and low calibration efforts.

Descriptors: oxygen measurement, dissolved oxygen (O_2), oxygen content in beer

1 Introduction

The determination of dissolved oxygen concentration plays a major role in different steps of beer production. The oxygen intake, for example, during mash and wort boiling should be inhibited due to induced processes such as oxidation of unsaturated fatty acids, decrease of phenolic compounds, influences on the degradation of starch, lautering and finally the extract recovery [1, 2]. In contrast to that, oxygen serves the reproduction of yeast during the starting phase. The excess of oxygen, however, can lead to the formation of unwanted fermentation by-products. Furthermore, beer ingredients can be oxidized after the cease of the main fermentation process [3]. The flavour stability of bottled beers is also influenced by the oxygen content.

Breweries are therefore highly interested in having available fast and reliable methods for determining the oxygen content, especially during filtration, carbonization, blending, bottling and filling. Besides of dissolved oxygen which is uptaken during production, the oxygen remaining in the empty space (head) of bottles, kegs and cans should also be taken under consideration, as there is a possibility that gas exchanges into beer. By using especially conceptualized filters and pressurized plants constructed with tanks pre-filled with carbon dioxide, it is possible to keep oxygen concentrations below 0.1 mg/l at the filler demand, and below 0.3 mg/l in filled bottles. The total oxygen content in industrial scale is currently below 0.1 mg/l. To date, manufacturers guarantee that the oxygen content for fillers reaches a maximum of 0.03 mg/l and for filters can even stay below 0.01 mg/l. This, of course, presents a special challenge for the measurement system in terms of accuracy and precision, as due to those guarantee statements the system needs to have the capacity to measure precisely concentrations of below 10 ppb.

The investigation was launched to examine the newly developed oxygen measurement system built in the CO_2/O_2 Gehaltemeter type c-DGM of Haffmans BV, in comparison to the standard analysis systems, the Orbisphere 3650 from Hach Ultra Analytics GmbH and DIGOX 6 from Dr. Thiedig & Co. All systems were compared to each other in terms of their service capacity in breweries. The evaluation emphasized in particular the precision and accuracy as criteria for the feasibility of the systems during the daily use in brewery industry, because the precision describes the quality of the system and the accuracy is considered a degree of equality between displayed and real value [4, 5].

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Tables and figures see Appendix

2 Determination of dissolved oxygen

2.1 Electrometric methods

In the brewing industry dissolved oxygen is commonly measured by electrometric methods using Clark or similar sensor methods.

These measurement methods are in detail described in Brautechnische Analysenmethoden – volume II of the Mitteleuropäische Brautechnische Analysenkommission (MEBAK) [3] and in EBC Analysis Committee – Section 9 of the European Brewery Convention [6]. The measurement principles are extracted therefrom and briefly described in the following.

2.1.1 Oxygen determination via Clark-Electrode [3, 6]

By using the so-called Clark-Electrode, the dissolved oxygen is determined with the help of two electrodes. The measurement cell consists of the cathode (mostly gold or platinum) and the anode (counter electrode, made of silver), which both are connected by a liquid electrolyte (KCl/KOH solution). As the reference potential serves the potential of the anode, of which the metal, together with the anion contained in the electrolyte solution, forms a reversible electrode the second kind of (double electrode system). The electrode pair is separated from the measurement solution by a gas permeable membrane. By applying an adequate polarisation voltage, the oxygen diffuses through the membrane into the measurement cell and there is reduced on the cathode surface whereupon hydroxyl ions are formed. The potential between anode and cathode should lie above the potential at which the oxygen is reduced to hydroxyl ions, but still in magnitude to keep the electrolytes safe.

Cathode: $O_2 + 4\ e^- + 2\ H_2O \rightarrow 4\ OH^-$

At the anode, the electrode reaction occurs reversibly:

Anode: $4\ Ag + 4\ Cl^- \rightarrow 4\ AgCl + 4\ e^-$

Both reactions can be combined by applying KCl as electrolyte:

$O_2 + 2\ H_2O + 4\ Ag + 4\ KCl \rightarrow 4\ AgCl + 4\ KOH$

According to the amount of oxygen molecules reduced at the cathode, an electrode current is initiated that can be measured with the help of an ampere meter. The current is proportional to the oxygen partial pressure. According to the Henry law, the oxygen concentration in the medium can be determined by taking the oxygen solubility coefficient into account.

2.1.2 Oxygen determination according to Tödt and Teske [3, 6]

Another method for determining the oxygen content works according to the potentiostatic principle, applying a 3-electrode measurement system that was developed by Tödt and Teske. A membrane is not used thereby. The measurement cathode consists of massive silver and the anode as counter electrode of stainless steel. The reference electrode is coated with silver chloride. After applying a defined polarisation voltage, an electrochemical reaction is initiated on the measurement cathode, whereupon oxygen is reduced.

Measurement electrode (silver):

$O_2 + 2\ H_2O + 4\ e^- \rightarrow 4\ OH^-$ (on the cathode)

Counter electrode (stainless steel):

$4\ OH^- \rightarrow O_2 + 2\ H_2O + 4\ e^-$ (on the anode)

The current initiated by this reaction is directly proportional to the amount of dissolved oxygen, if the polarisation voltage matches exactly the level of the diffusion threshold current. In this case, the above mentioned relationship can be described by the equation:

$I = K \times c(O_2)$

where $K = n \times F \times A \times 1/d$, I the measurement current, $c(O_2)$ the oxygen concentration, F the Faraday constant, n the amount of electrons converted per molecule, A the cathode surface, and d the "unstirred boundary layer" thickness. The thickness of the "unstirred boundary layer" depends on the hydrodynamic conditions occurring at the measurement electrode, and on the transfer of oxygen molecules through the layer by temperature dependent diffusion processes. In order to definably adjust the polarisation voltage between both electrodes, a third electrode for comparison is introduced in this measurement instrument. This electrode for comparison stays in electrical contact with the surface of the measurement electrode through a diaphragma, so that no oxygen exchange can occur.

2.2 Optical methods [7, 8]

The oxygen concentration determined by optical methods measures the luminescence of oxygen sensitive layers. The fluorescence changes in dependence of the oxygen partial pressure. With the help of the measured oxygen partial pressure and temperature, the amount of gas in liquid can be calculated. This technology is already being applied in industries such as in wastewater treatment, or for measuring the oxygen ingress into PET bottles [9]. The oxygen sensor determines the oxygen content in the liquid by optical measurement according to luminescence method (dynamic luminescence quenching), which in principle follows the process that an oxygen sensitive layer containing the oxygen is irradiated with blue light as shown in Figure 1 (1). Thereby, molecules present in the oxygen sensitive layer are excited to reach a higher energy level. The electron gets into an excited state (2) and luminescence occurs in form of red light once the electron falls back to normal energy level (3), which then leads to the emission of red light (4).

If oxygen is present, it collides with the molecules in its excited state also present in the oxygen sensitive layer. The oxygen as the quencher results in radiation-less deactivation (dynamic quenching). After collision, an energy transfer between the excited molecule of the oxygen sensitive layer and the oxygen which consequently is transferred from its ground state (triplet state) to its excited singlet state. These molecules then cannot illuminate anymore due to the presence of oxygen whereupon the luminescence signal decreases.

Through this process, a relationship can be established between the oxygen concentration in the sample and the luminescence intensity, as well as the degradation rate of light intensity. The degradation rate as the lifetime of the luminescence is described

in the Stern-Volmer-equation, which depicts the luminescence decay times in absence and presence of oxygen in relation to the oxygen concentration. By measuring the luminescence lifetime and the product temperature, the oxygen concentration can be determined. The optical measurement of dissolved oxygen is independent from pressure, colour, flow rate of liquid, as well as from the age of the LED light source.

3 Experimental

Comparative measurements were performed using the sensor oxygen measurement system and the standard analytical references. The experimental setup involved several preliminary trials, two long-term measurements allowing for the determination of precision, one gas measurement for determining the accuracy, and two concluding tests conceptualized as long-term measurements also for determining the accuracy. The reference instruments were calibrated for each experimental period separately. The c-DGM remained in its as-delivered condition and was not calibrated during the entire investigation. The sensor of the Orbisphere was calibrated in air saturated with water. The calibration in air was chosen and not liquid because the exact atmospheric oxygen concentration in air worldwide is known and only the air pressure and temperature needs to be taken into account. Liquids with known precise oxygen concentrations as required are currently not available.

The clue of the Orbisphere calibration is based on a mathematical 2-point calibration. Thereby, the zero-point is set (electrode current is zero), while the second point is obtained by the atmospheric oxygen concentration. As the value of the oxygen in air is far away from the zero-point, the effect of possible measurement errors by determining the atmospheric oxygen concentration is very low on the accuracy of the calibration curve near the zero-point, which is the defined area of application. For all measurements the membrane type 2956A was used, which was not covered with any kind of lattices. It was renounced to use lattices especially for gas measurements because there is possibility that parts of liquids can accumulate in the space between the membrane and the lattice. Dissolved oxygen contents in the liquids can influence negatively the measurements.

The DIGOX uses an incorporated feature to actively calibrate the system. The calibration cell produces a specific amount of oxygen by electrolyses, which is additionally dissolved in the medium and is detected at the measurement electrode. The disadvantage of this manner of calibration is that the user has to rely on the functionality of the calibration unit. There exist no way to check the functionality by means of certified test gases.

3.1 Preliminary experiments

By using a tap preparation, it was at first the purpose to determine the oxygen content of different beer products sampled directly from the bottle. In order to ensure that the entire band of oxygen contents is covered, one part of the beer samples were separated and aged artificially by applying conventional Forcier-tests according to Brautechnische Analysenmethoden [3]. This preliminary trial served only for a first evaluation as to the feasibility of determining the precision and accuracy. For this, the Orbisphere was connected in-line with the c-DGM, so that, after starting from the tap preparation, the beer sample passed at first through the c-DGM and then through the Orbisphere, which measures the oxygen content by following the Clark-Electrode principle.

3.2 Comparative measurements – c-DGM and DIGOX performance in long-term tests

For determining the precision, a long-term measurement was performed based on insights obtained from this preliminary trial. At first, two 30 l kegs of equal filling were prepared and connected to the c-DGM through a three-way tap. The c-DGM was then connected in-line to the comparing instrument (Figure 2). As the reference here served the DIGOX, which is also accredited for the brewing industry. Its measurement principle according to Tödt and Teske is also well acknowledged and established in the brewing industry (see Brautechnische Analysenmethoden, volume II, chapter 2.32.1.2.2 [3]).

The beers of both kegs differed in terms of their oxygen concentration. The beer flow could be chosen by a three-way tap. At this set-up, keg A contained beer with an oxygen concentration of ca. 0.3 mg/l, whereas the beer sample in keg B exhibited a high oxygen concentration of ca. 4.5 mg/l, both in average respectively. The measurement range of the c-DGM was up to 2 mg/l. The volume flow was adjusted to 10.0 l/h. The oxygen concentrations measured over time were documented using data loggers to evaluate the precision of the c-DGM and DIGOX systems by decline characteristic (from higher to lower oxygen content). The real measurement values were logged every 15 sec.

3.3 Comparative measurements – c-DGM and Orbisphere in long-term tests

The second long-term test served for the precision of the c-DGM oxygen measurement system in combination with the Orbisphere as the reference. The Orbisphere was calibrated as described previously and received a new membrane. In the third test series, the oxygen content in beer was measured on location in a brewery. The measurement systems were connected in-line and, as described previously, the beer sample rich in oxygen could be diverted by a three-way tap (Fig. 3). The flow rate was measured using impulse measurement and adjusted again to 10 l/h. In addition to that, the real measurement value was logged every 15 sec.

3.4 Gas measurement for determining the accuracy

The accuracy determination of the c-DGM and reference instrument was performed with the help of certified test gases (Scott Specialty Gases, The Netherlands). Currently also systems exist that measure accuracy and precision by means test fluids. Such systems however can reach the precision for the oxygen content in the test fluid of ± 10 ppb within the measurement range of 0 and 300 ppb. The specification of the c-DGM in this respect is ± 1 ppb plus 2 % of the measured value, which is more precise than the precision of the test fluids. In order to test accuracy and precision of a test system it is imperative that the reference system is more precise. The test gases used herein possess a certified precision of

± 1 % of the measured value and, thus, are feasible for the purpose of verifying the c-DGM system's accuracy (e. g., the test gas with an oxygen content of 1 %, which means approximately 400 ppb of dissolved oxygen, possesses the precision of ± 4 ppb).

As the DIGOX instrument, which determines the oxygen according to Tödt and Teske, is incapable of measuring gases, only the Orbisphere in combination with c-DGM was evaluated by connecting these two instruments in-line. The accuracy was verified using three test gases that each differed in terms of their oxygen concentrations. The flow rate was adjusted to ca. 25 l/h to avoid built-up pressure that increases the partial oxygen air pressure of the test gases. The c-DGM as well as the Orbisphere and the test gases were acclimatised for one night at room temperature to ensure that the measuring systems and the gases were having the same temperature.

3.5 Beer measurements for determining the accuracy

In order to verify the results obtained from the test gases and to evaluate correctly the real accuracy of the c-DGM and Orbisphere, beer was chosen as the sample matrix. For this purpose, the experimental set-up involved both a connection in-line and in parallel on location in a brewery.

3.5.1 In-line connection

For the set-up, both measurement instruments were operated in-line and connected again directly to the filter outlet of a sheet filter in a filter cellar of a brewery as shown in Figure 4. The flow rate in both devices was kept constant at 10 l/h using impulse measurement and all measurement data were logged.

3.5.2 Parallel connection

Based on the findings gained from the in-line experiments (discussed in 4.5.1), both instruments were operated in parallel and connected to a keg that was freshly filled with beer (Fig. 5). The flow rate, adjusted to 10 l/h, and data logging occurred in the same manner as described previously.

4 Results and Discussion

In the following, the results of the validation series are described chronologically along with the changes made in the experimental set-up.

4.1 Preliminary experiments

Representatively for at least 80 different measurements, Figure 6 shows schematically a typical measurement course, determined with the c-DGM and Orbisphere instruments during oxygen determination in beer sampled from a 0.5 l bottle. It can be seen that the oxygen content measured with the Orbisphere starts from an increased stage and levels to the value measured in beer. The opposite measurement behaviour can be recognized for the c-DGM instrument. An explanation for this can be given by the short response time of the c-DGM, which is due to the fact that nearly oxygen free carbon dioxide is contained in the instrument before and after tapping the beer sample. Also for that reason, the measurement curve of the c-DGM approximates at first the lower value and then sinks again after carbon dioxide is found only in the measurement cell. The time period in which both instruments levelled to the real oxygen content is very short and represents the area with the lowest measurement difference (red zone in Figure 6).

As the sample volume was insufficient (0.5 l), a correct evaluation as to the time point when the measurement of an identical sample in both instruments is guaranteed could not be achieved. Furthermore, the response time and decline characteristics of both systems could not be ascertained. These two cognitions that the response time of both systems seemed to be too low compared to the run duration of the sample, and additionally that a regulation of oxygen content from high to low was not possible, has led to a change of the sample volume, which needed to be increased significantly in the following experiment. Thereby, a continuous measurement of oxygen in beer from rich to low could be guaranteed.

4.2 Comparative results – c-DGM and DIGOX in long-term tests

Figure 7 shows the oxygen curves obtained with experiments run by both instruments. As can be seen, the curves possess a similar appearance and, thus, the velocity and the decline characteristic of the oxygen determination with both instruments are identical. From Figure 7 it can be additionally deduced that a fourfold change between the interconnected kegs was performed within the measurement period (ca. 1 h and 40 min). Both measurement curves document explicitly an increase of the oxygen concentration in beer.

It is noticeable that the c-DGM registers lower measurement values at lower oxygen concentrations (< 0.5 mg/l; keg A) than the DIGOX instrument (with a mean difference of 0.06 mg/l). In order to compare both systems by statistics, involving all results below the oxygen concentration of 0.5 mg/l, then $n = 262$ measurement should be used. As the amount of n exceeds 30, it is allowed to apply the Grubb's Test for outliers according to DIN 53 804 [10, 11]. Figure 8 shows on the left the outliers (in total 5) before and on the right side after elimination. The diagram further reveals a linear correlation between the measured values of both systems and furthermore an offset related to the value of DIGOX. After the outliers were eliminated, the correlation coefficient R^2 is 0.9954 and the offset (the ordinate part of the linear equation) is 0.06 (Fig. 8).

The approach of data correction performed for the measurement values below 0.5 mg/l is needed because a systematic deviation from 0.06 mg/l occurred. The evaluation for normal distribution according to David, Hartley and Pearson [12] is considered positive as to the differences from the measurements of c-DGM (corrected) and of DIGOX. Figure 9 shows the density distribution of the measurement differences after outlier elimination and the confidence interval ($P = 95\%$) of the mean value x (= 0.0001 mg/l). The true mean value μ is between $-0.0009 \text{ mg/l} \leq \mu \leq 0.0012 \text{ mg/l}$.

Each measurement pair was afterwards examined statistically with the help of the t-test according to Gosset [13, 14]. The test with 5 % significance level proved that the results of both instruments differed not significantly. At oxygen concentrations above 0.5 mg/l (keg B) the c-DGM measures slightly higher concentrations than the DIGOX, with a mean difference of 0.28 mg/l. Compared to lower concentrations, the difference in the measurement between both instruments is notably higher at a concentration of 4.5 mg/l. This, however, can be explained due to the measurement range of the c-DGM, which is from 0 to 2 000 ppb. It was therefore refrained from performing a statistical evaluation. The upper measurement limit of the c-DGM instrument is depicted as a line in Figure 7.

With the help of these tests, it could be demonstrated that the measurement speed and the decline characteristic of both instruments is equal. At measurements below 0.5 mg/l, both systems exhibit only a systematic deviation of 0.06 mg/l. After this deviation is corrected, it can be statistically proven that both instruments are capable of equally measuring the oxygen concentration in beer. After offset correction and in the frame of measurement accuracy as stipulated by Brautechnische Analysenmethoden [3], the measurement behaviour and the results obtained by c-DGM and DIGOX can be considered identical.

4.3 Comparative results – c-DGM and Orbisphere in long-term tests

A long-term test was also performed similarly to determine the precision of the c-DGM system using the reference system Orbisphere. The experimental setup, compared to the previous test using the DIGOX, changed only in the manner that the beer sample poor of oxygen was not supplied from a keg but from the filter outlet of a sheet filter in a brewery. The oxygen concentration at the filter outlet was measured with 0.05 mg/l in mean. The mean oxygen concentration in beer from the keg was 0.4 mg/l.

Figure 10 shows the measurement curves of both instruments. As can be compared to the previously performed long-term test, the courses of concentration are nearly identical. The first two slopes of the curves (at 45 and 1275 sec) document the switchover to the beer rich in oxygen, which was fed from the keg. The final maximum was registered towards the end of the filtration, when the filter was flushed with water. Thereby, beer is diluted with water so that the oxygen concentration is increased. Therefore, only beer is measured that comes from the keg afterwards. Without lead time and set-up time, the experiment was performed within 41 min, whereupon in total 165 measurement values were obtained and logged.

Despite similar courses, it is noticeable that the c-DGM measured lower oxygen concentrations than the Orbisphere throughout the entire concentration area (0.014 to 3.52 mg/l). The deviations become increasingly higher the higher the measured oxygen concentration is. This means that there is no linearity in the measurement deviation to recognize over the entire measurement period.

The measurement differences between the Orbisphere and the c-DGM in dependence of the oxygen concentration are plotted in Figure 11. The deviation between both instruments increases non-linearly along with the oxygen concentration. Upon parting the measurement area, a linear relationship for the increase in concentration below 0.5 mg/l can be assumed. It seems, however, that above 0.5 mg/l the differences approximate asymptotically to the limit between 0.6 and 0.7 mg/l. In order to evaluate the response and decline characteristics during oxygen determination within the relevant measurement area (< 0.5 mg/l), the slopes of the measurement curves need to be compared with each other due to differences in the measurements. For this, measurement areas were selected in which it was switched over twice to the beer sample rich in oxygen. The calculation is demonstrated exemplarily for the first switch over point.

In Figure 12, a magnified part extracted from Figure 10 is shown and additionally the maximum slopes (m_1 and m_2) and mean maximum values (M_1 and M_2) of both curves, resulting from the first switchover to the beer rich in oxygen. In order to determine the maximum slope, the maximum of the first derivative of each curve is calculated for the measurement period between 0 and 600 sec. The mean maximum values are obtained when the slope reaches a plateau to become nearly zero. The data of the maximum slope, as well as the mean maximum values are shown in Table 1. From the ratio M/m, it can be ascertained on the hand of the obtained curves that both instruments possess the same responses. The following contemplates the deviations in the area of low oxygen concentrations (< 0.06 mg/l) during the experimental time period between 690 and 1260 sec, as during this timeframe the oxygen concentration remained nearly constant.

Figure 13 shows the measurement deviations between the Orbisphere and c-DGM with clearly an increase of the measurement differences in dependence of increased oxygen concentrations. The differences in the measurements of both instruments within the considered measurement area are too high in magnitude. In current praxis, a measurement of 0.01 mg/l should safely be achievable. The results of the second long-term test revealed that the measurement speed and the decline characteristic of the c-DGM and the Orbisphere as the reference are identical for measuring oxygen concentrations below 0.5 mg/l. However, noticeable is the difference in measurement between both instruments, which magnifies depending on the measured oxygen concentration. Therefore, the currently required maximum precision could not be achieved due to increased differences in the measurement, especially for the concentrations area below 0.05 mg/l, which is currently practice-relevant.

4.4 Results of gas measurement for determining the accuracy

The final tests concerned the accuracy of the c-DGM and Orbisphere systems by using three different test gases, which were composed as follows:

1. Nitrogen (O_2-concentration 0 %)
2. Nitrogen/Oxygen mixture (O_2-concentration: 0.05 %)
3. Nitrogen/Oxygen mixture (O_2-concentration: 1.01 %)

After the Orbisphere and the c-DGM were subjected to sterile air for dryness over several hours, both instruments were connected in-line and admitted to test gases. The results shown in Table 2 are additionally expressed in ppb as to dissolved oxygen, although gas was measured. When dealing with dissolved oxygen, the temperature plays a decisive role in the calculation of ppb-values, because according to Henry law, the dissolved oxygen content is higher at lower temperatures, but otherwise equal oxygen partial pressure.

The measurement series started with using the 2nd test gas. It was noticeable that the Orbisphere needed more time to reach down to the endpoint of ca. 1 %. To explain the longer response, it should be mentioned that the Orbisphere was still equipped with the same membrane type used in previous trials. This membrane was manufactured for using fluids and not gases, for which thinner membranes exist.

The results demonstrate that both systems are capable of determining sufficiently the oxygen concentration in gases. The difference in measurements can be neglected taking deviations into account. The results obtained with all three test gases can be considered identical. The only notice was that the Orbisphere exhibited a longer response time, especially at the beginning of the experiment where the adjustment towards a constant value lasted for more than 45 min. This observation, however, could be ascribed to the applied membrane, which firstly was designed for the use with fluids and, secondly, was oxygen saturated after the treatment with sterile air. The results concluded further that both systems possess the same accuracy. In viewpoint of the previous performed experiments, the question arises now whether this accuracy pertains only to the measurement with gases, or can also be related to measure oxygen concentrations in liquids like beer.

4.5 Results of beer measurement for determining the accuracy

4.5.1 Connection in-line

For the final clarification whether the accuracy of the c-DGM and the Orbisphere is also viable for beer as sample matrix, both systems where connected in-line to the filter outlet of the sheet filter in a brewery (cf. Fig. 4 for the experimental set-up). Figure 14 shows clearly that the Orbisphere registers higher oxygen concentrations than the c-DGM. At this set-up, the Orbisphere was integrated after the c-DGM. These results confirmed those obtained in the previous experiment (4.3). However, they are also in contrast to the measurement results acquired under 4.4, which leads to the assumption that oxygen is entering the system somewhere.

A continuous intake to the system can be excluded according to the results obtained under 4.3. The oxygen curve of the Orbisphere registered as shown in Figure 14 permits rather the conclusion that it draws nearer to the curve of the c-DGM over a longer time period. This, however, would mean that the Orbisphere, as demonstrated by the gas experiments, possesses a noticeably longer response time than the c-DGM. This assumption, however, could not be verified by this experiment because the filtration was finished.

An explanation as to the slower response of the Orbisphere can be given as follows: Through the tube, oxygen air can be taken up from environmental air until the tube is saturated. This diffusion process depends on the tube's material so that there is a dependency to recognize between the material and the diffusion rate, as well as to the oxygen uptake capacity until the oxygen concentration in the tube and liquid (here the beer matrix) reaches an equilibrium. The fact that oxygen can enter the system is not new, but often remains uncovered in practice, as it is often not mentioned in technical manuals.

4.5.2 Results from the parallel connection

From the assumptions of chapter 4.5.1 the final test was modified as follows:

1. To ensure equal lines in length (from the connection point until the instrument), both systems were switched in parallel using a Y-manifold (cf. Fig. 5)

2. All connecting tubes were composed of the same material

3. The tubes were placed in beer for 24 h prior to each experiments in order to reach an oxygen equilibrium

4. The Orbisphere has received a new membrane, which in addition was subjected to the beer matrix

5. Prior to the measurement the Orbisphere was filled with a blank beer sample (aged beer, oxygen free)

Figure 15 shows the two oxygen curves obtained with both systems after the modified set-up. The curves confirm that both instruments deliver stable results with a mean deviation of 0.5 ppb, and thus possess equal accuracy. In this respect, however, it should be noted that any kind of influence on the oxygen content (e.g., by diffusion through tubes) needs to be excluded. It is further recognisable that the Orbisphere exhibits considerable variations at the beginning of the measurements.

5 Conclusion

For its practical application in breweries, a newly developed oxygen measurement system, the c-DGM, based on optical sensors has been validated. The validation involved a comparison with accredited reference systems, for which the Orbisphere and the DIGOX came to operation. The measurement methods of these systems are acknowledged in the brewing industry and thus serve well as a lead for comparison with other methods. The aim of validation was to determine the precision and accuracy as the two main criteria used for evaluating best practices in substance specific measurements. The results obtained from the long-term tests confirmed that the measurement rate and decline characteristic of the c-DGM in comparison to both references were equal. The tests comparing the measurements revealed moreover that the c-DGM possesses an excellent precision, complying very well with the demands defaulted according to Brautechnische Analysenmethoden [3].

With the help of certified test gases, is could be ascertained that the c-DGM and Orbisphere systems both have the capacity to measure sufficiently very low oxygen concentration in gases, with differences of only 2 ppb. The differences in the results lied in the frame of measurement errors and could be neglected. The results acquired with test gases can be considered identical, revealing further that the c-DGM and the Orbisphere have the capacity to measure gases with exactly the same accuracy. Finally, it was examined on the hand of beer as sample matrix whether the obtained accuracy of the gas measurement is valid. For this, information received from preliminary trials were used to optimize the experimental set-up in terms of equalizing the conditions (identical tube material, preconditioning with oxygen depleted beer, and integration of the systems through parallel switch). Thereby, the oxygen that continuously entered the system through diffusion as assumed could be minimized, so that the systems c-DGM and Orbisphere, confirmatively for beer, possessed the same accuracy.

Conclusively, the c-DGM can be described as an effectively working, precise and accurate system, and therefore is well suited to determine the dissolved oxygen content in beverages. Compared to other electrochemical measurement systems currently in use, the system signalizes a fast response and low calibration efforts, which makes it feasible for routine application in the brewing and beverage industry. Due to the highly stability of its optical sensor, future experiments should in involve further long term experiments in order to ascertain whether the sensor still remains in high-quality performance as to precision and accuracy over several months.

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Appendix

Table 1 **Data of the maximum slope and mean maximum value, obtained for the measurement period between 0-600 sec**

	Orbisphere	c-DGM
Slope [mg/(l × s)]	$m_1 = 0.0114$	$m_2 = 0.0076$
Maximum value [mg/l]	$M_1 = 0.442$	$M_2 = 0.294$
Ratio (maximum slope value) [s]	$M_1 : m1 = 0.442/0.0114 = 38.8$	$M_2 : m_2 = 0.294/0.0076 = 38.7$

Table 2 **Accuracy results for both systems determined using three different test gases**

Test gas	Gas specification	Orbisphere	c-DGM
1	$c_{O2} = 0.0$ %	1.7 ppb (22.3 °C)	1.4-1.5 ppb (23.0 °C)
2	$c_{O2} = 0.05$ %	0.049 %; 19.8 ppb (22.5 °C)	0.051 %; 20 ppb (20 °C)
3	$c_{O2} = 1.01$ %	0.944 %; 380 ppb (22.7 °C)	0.935 %; 367 ppb (23.9 °C)

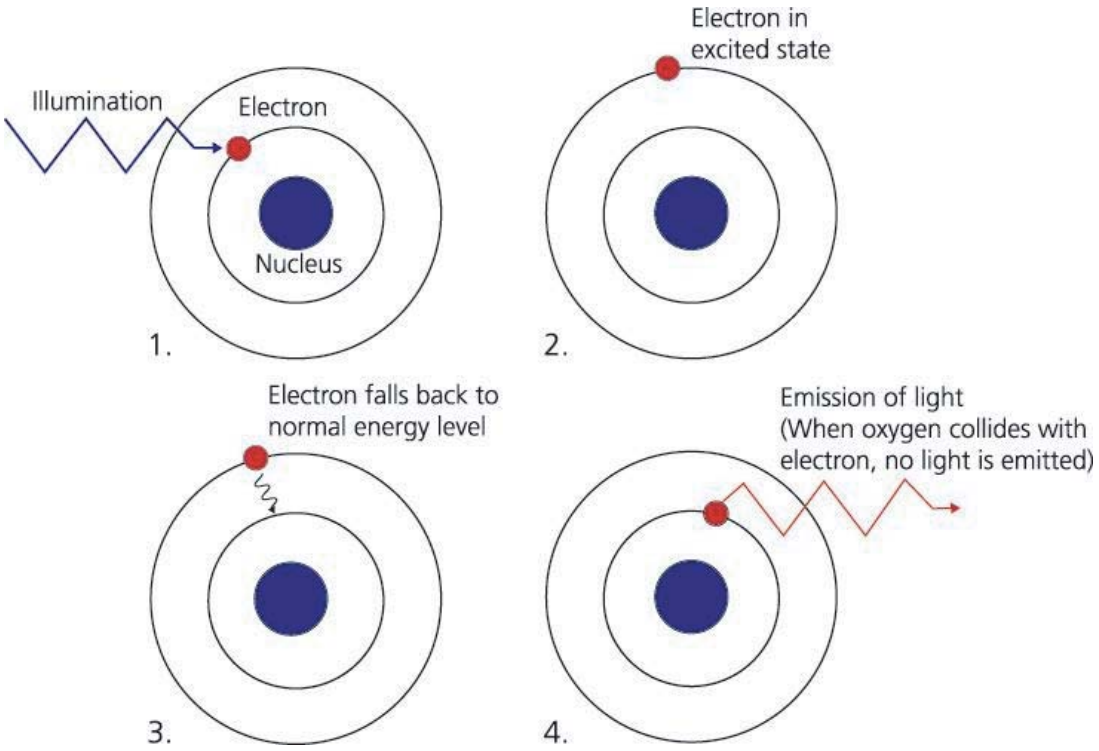
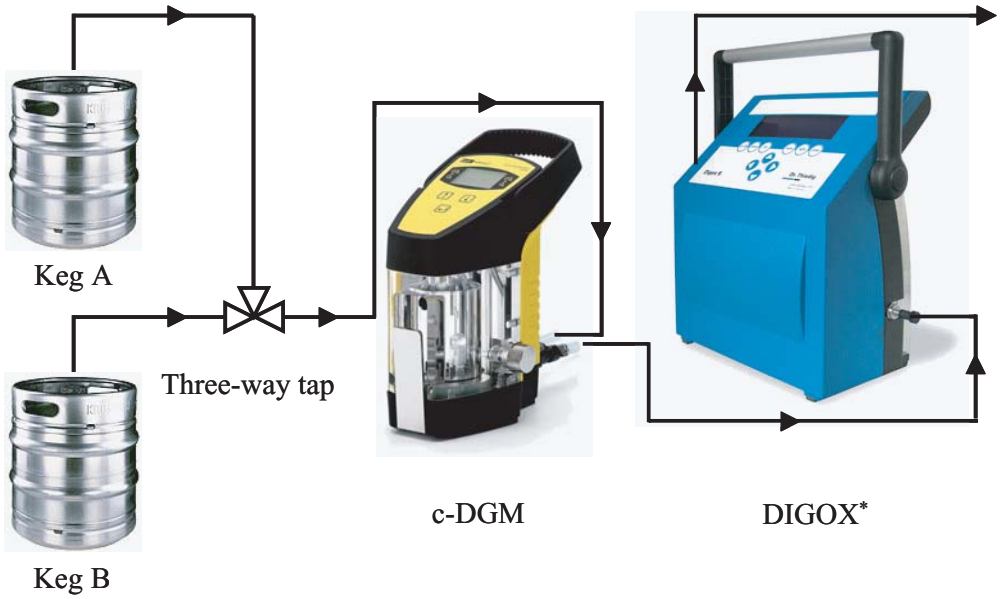
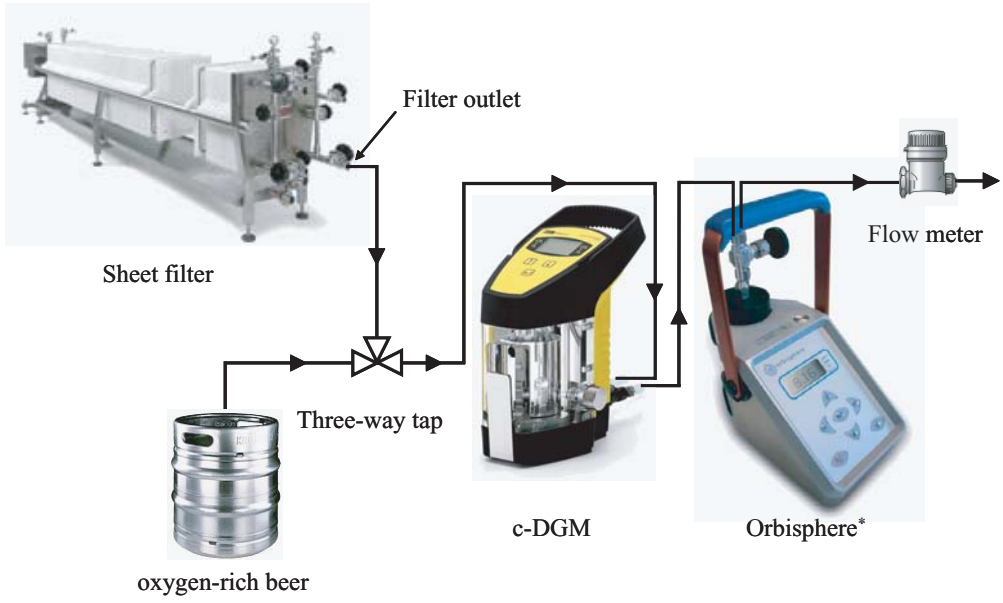


Figure 1 **Luminescence principle for oxygen concentration in liquids**
1.) Electron is irradiated with blue light.
2.) Electron gets into an excited state.
3.) Electron falls back to normal energy level.
4.) The emission of red light takes place [8].



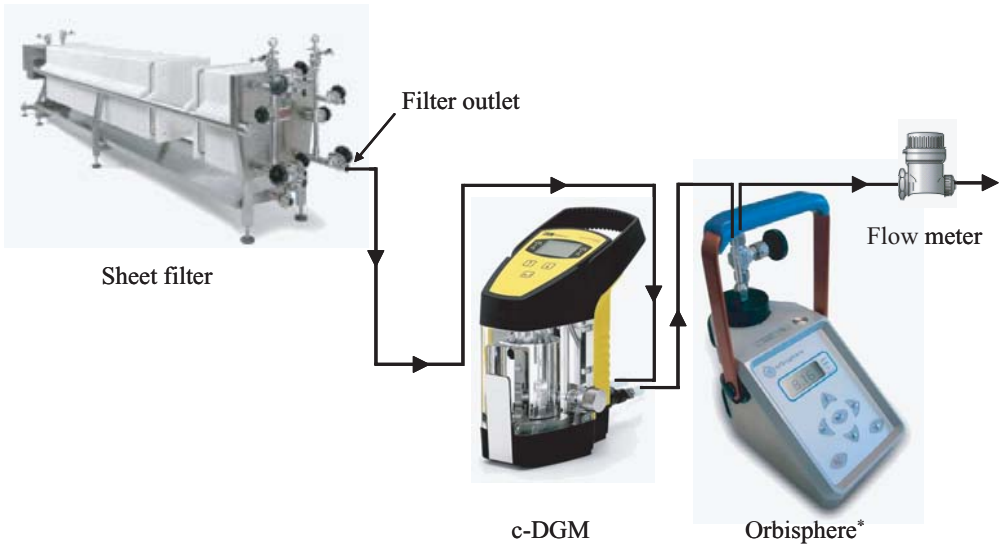
*Source: http://www.jojorama.de/deutsch/portfolio/digox/digox3.jpg
http://www.thiedig.de

Figure 2 Experimental set-up (in-line connection) for comparison of the c-DGM with DIGOX during long-term tests.



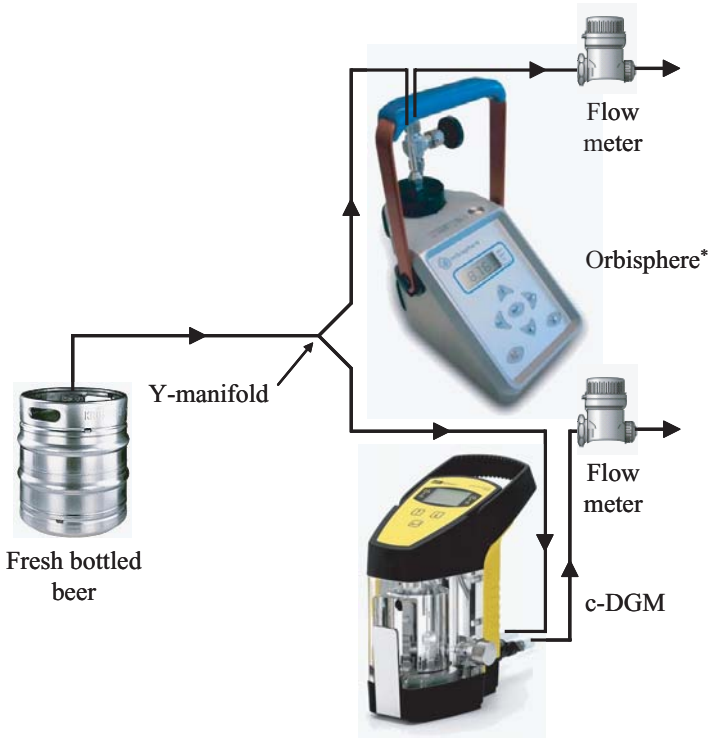
*Source: http://www.analytical-systems.co.nz/images/products/image002.gif
http://www.hachultra.com

Figure 3 Experimental set-up on location in a brewery for comparison of the c-DGM with the Orbisphere during long-term tests (in-line connection).



*Source: http://www.analytical-systems.co.nz/images/products/image002.gif
http://www.hachultra.com

Figure 4 Experimental set-up for comparison of the c-DGM with Orbisphere (in-line connection)



*Source: http://www.analytical-systems.co.nz/images/products/image002.gif
http://www.hachultra.com

Figure 5 Experimental set-up for comparison of the c-DGM with Orbisphere (parallel connection)

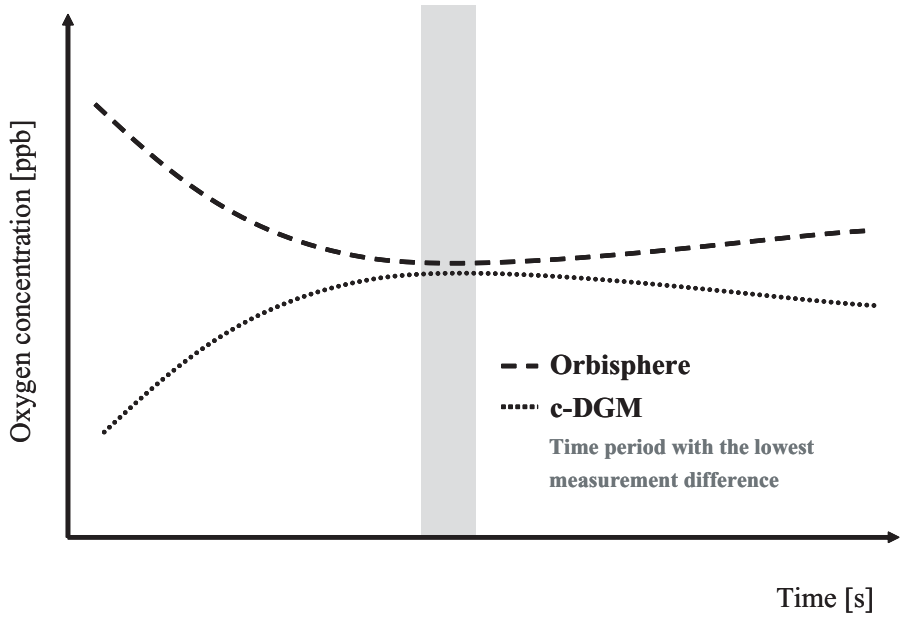


Figure 6 Scheme of the measurement curves of the c-DGM and Orbisphere obtained during the determination of oxygen concentration in beer sampled from 0.5 l bottle

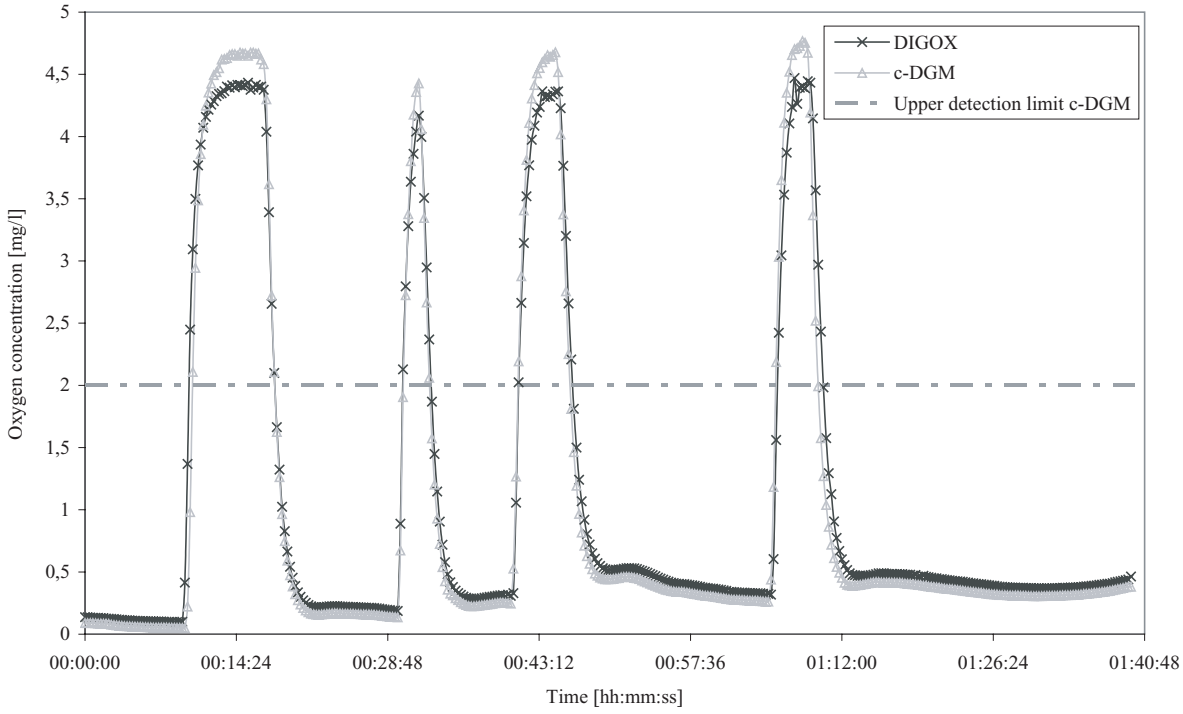


Figure 7 Measurement curves of oxygen over time obtained with using the c-DGM and the DIGOX instruments

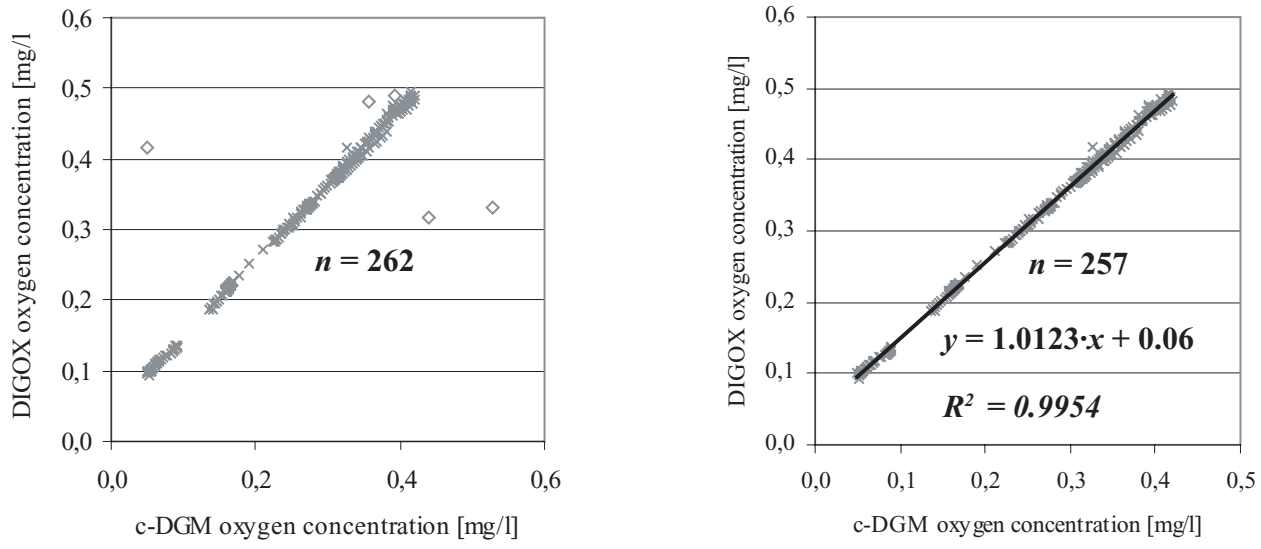


Figure 8 Correlation between the c-DGM and DIGOX according to the single measurement results below 0.05 mg/l and without outliers

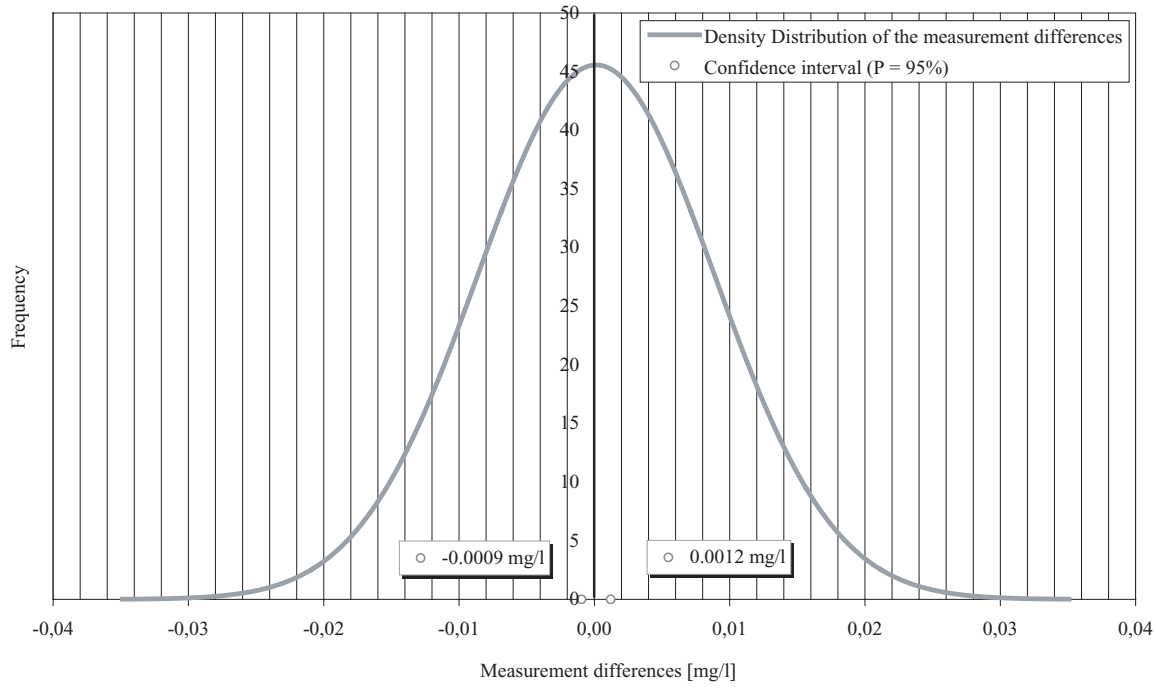


Figure 9 Density Distribution of differences obtained from the measurements of the c-DGM (corrected) and DIGOX

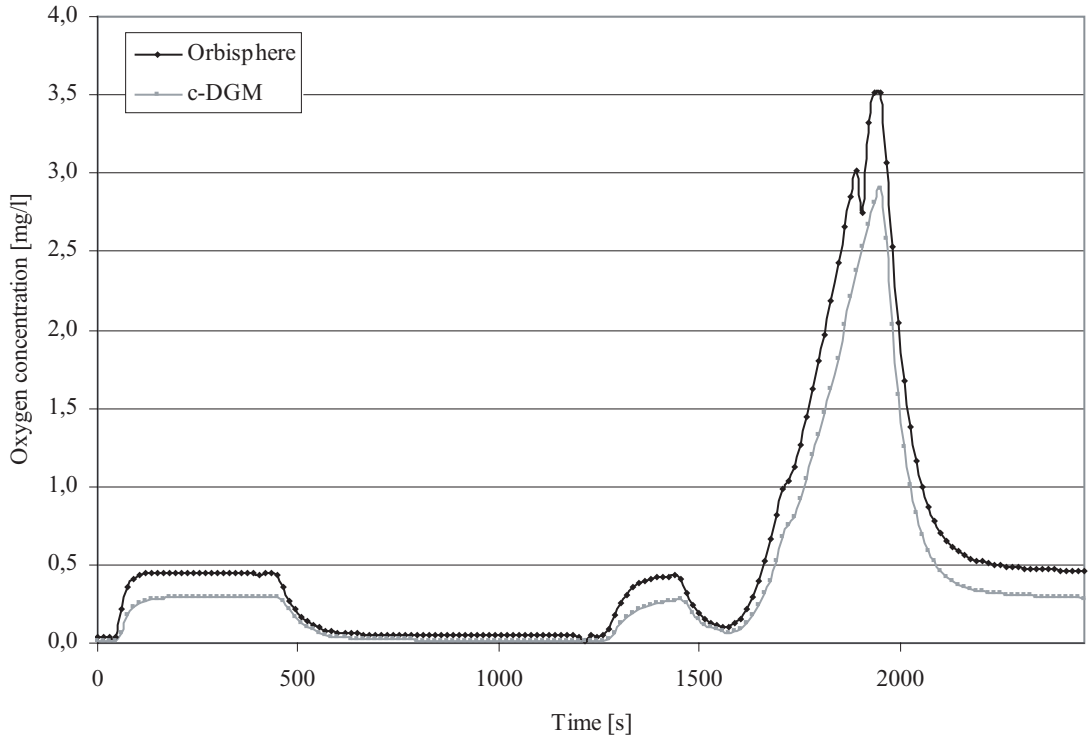


Figure 10 Measurement curves obtained with the c-DGM and the Orbisphere

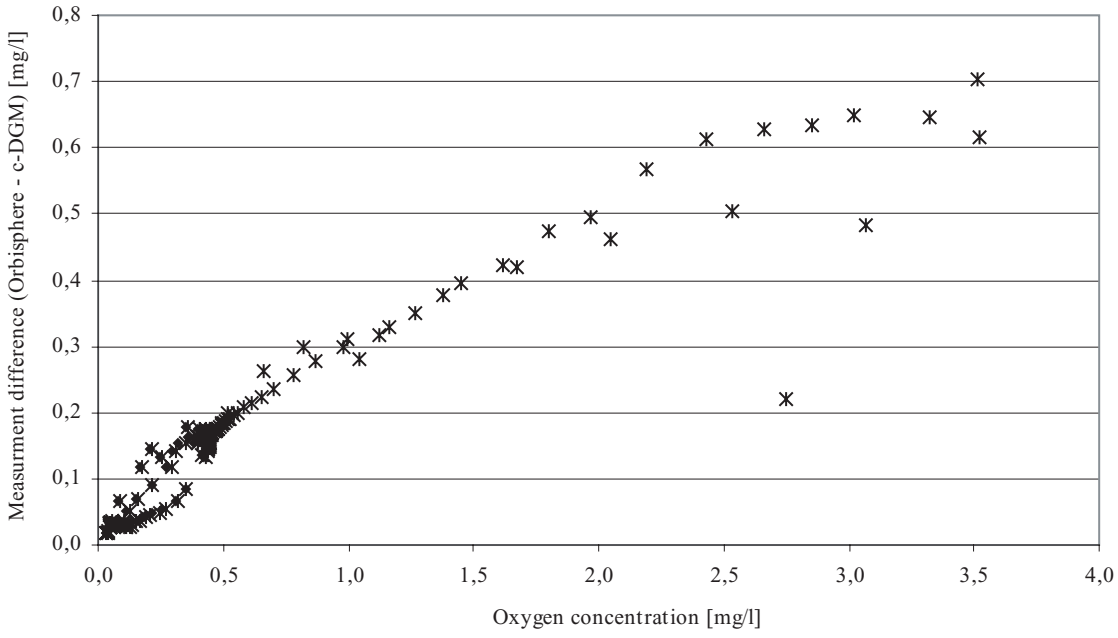


Figure 11 Measurement differences between the c-DGM and the Orbisphere in dependence of the oxygen concentration

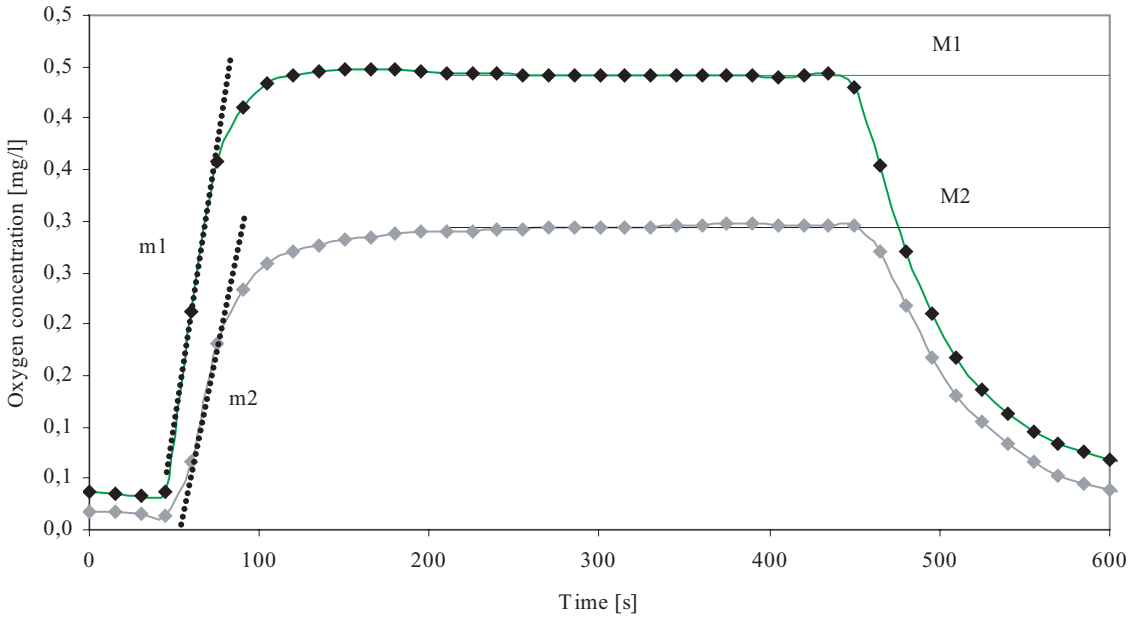


Figure 12 Extraction from Figure 10, showing the first switchover to the beer sample rich in oxygen

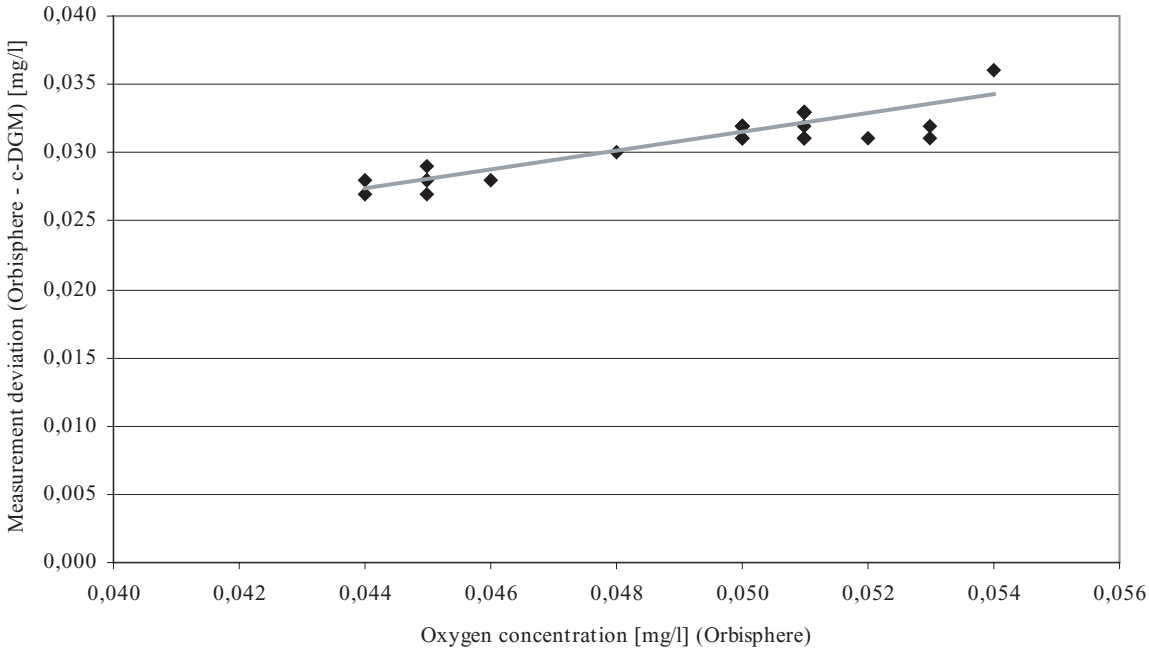


Figure 13 Measurement deviations between the Orbisphere and the c-DGM

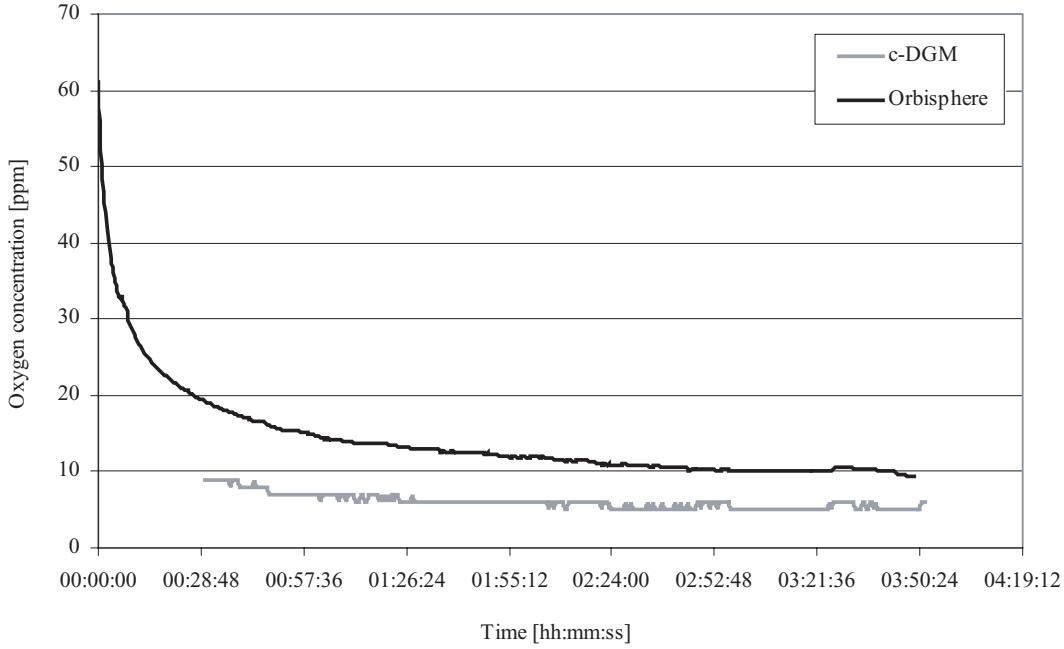


Figure 14 Oxygen measurement curves for the beer sample – Orbisphere vs. c-DGM

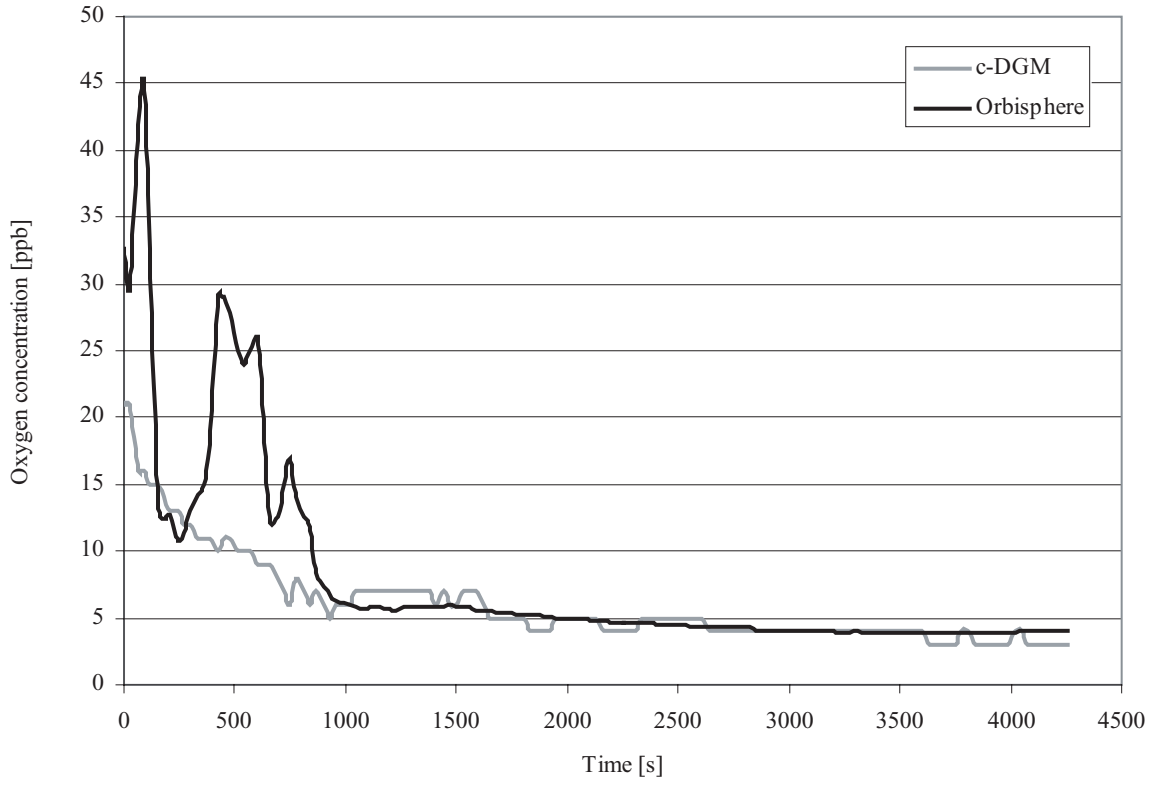


Figure 15 Oxygen measurement curves of the beer sample obtained under equal conditions – Orbisphere vs. c-DGM