

Practical Application of Multivariate Statistical Analysis for Evaluation of Sensory and Process Data from Full Scale Production

Olav Vind Larsen^a, Ian Williams^a, Anne Cathrine Lillelund^a, Sten Aastrup^a & Derek V. Byrne^b
^aAlfred Jørgensen Laboratory Ltd., Copenhagen, Denmark, www.ajl.dk and ^bSenseData Guidance International Ltd. Cork, Ireland

Introduction

Experience shows that improvements in flavor and flavor stability are often a result of several minor process adjustments, and it can be difficult to measure the influence of individual changes from full-scale production. A powerful tool to overcome this problem is to use Multivariate Statistical Analysis to analyze data with the purpose of gaining a basis for taking qualified decisions regarding changes in process and on raw material specifications. Here we present a data collection and analysis methodology that allows for systematic gathering of data from sensory panels, from laboratory analysis and the process, all from full scale production (2200 hl), such that these data can be analyzed using a Multivariate Analysis (MVA) Software Package (the Unscrambler). Data was collected from routine beer production and trials, and the data has been analyzed to determine relationships between sensory data, analytical data and process data, allowing for informed decisions regarding process changes to be made. The ultimate aim is the achievement of a marked improvement in the flavor stability of the beer produced.

Approach

A series of batches of full-scale beer are produced for a particular brand and three aspects of the beer are recorded in the same database:

- Sensory score data (from the fresh sample and forced aged sample (7 days at 38°C))
- Chemical analytical data (from the fresh sample and a selection from the aged sample)
- Production data gathered by the brewery

Firstly, approximately 5 normal beers are produced and the data is analyzed by MVA in order to assess the brands flavor and flavor stability status, as determined by the sensory and chemical analysis. Secondly, approximately 10 trials are conducted in an attempt to improve the flavor and flavor stability of the brand. For each of these trials, a single process change is made that may benefit flavor and flavor stability. Such changes could involve increasing the mashing-in temperature, improving the pitching yeast vitality, changing processing times, adding anti-oxidant in the process or any other aspects that warrant attention and may be of interest for the brewery to perform. After this step MVA is conducted again to see which trials had the most beneficial effect on flavor and flavor stability. Finally, a third set of approximately 5 trials are conducted, however in this instance a combination of several process changes per trial are introduced, again in an attempt to produce beers with an optimal flavor and a high resistance to staling.

As the total data collected for each trial is a rather large set of data, and because brewing is a multivariate process, it is necessary to use multivariate analysis to determine which sources of data are significant for the flavor stability of the beers. This approach could also be used by the brewery as part of a continuous improvement plan, as shown in figure 1.

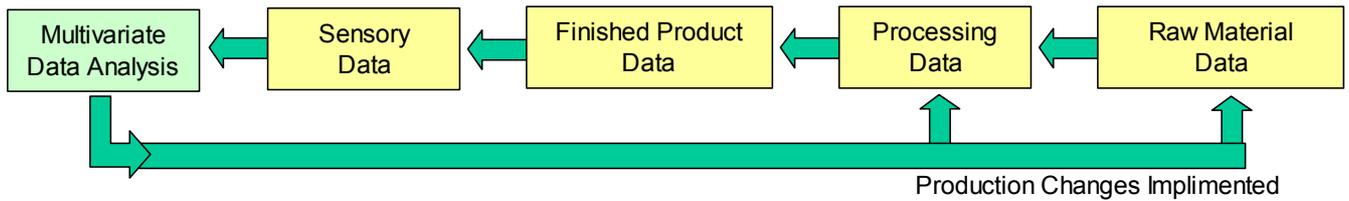


Figure 1: A Multivariate Data Analysis-Based Control Strategy in full-scale beer production to achieve improvements in brand flavor and flavor stability

The main goal in such a project is to reduce stale flavor development in full-scale production beers. This will occur in the best single process change trials (Step 2) and even more so in the combined process change trials produced in the 3rd step (see Figure 2)

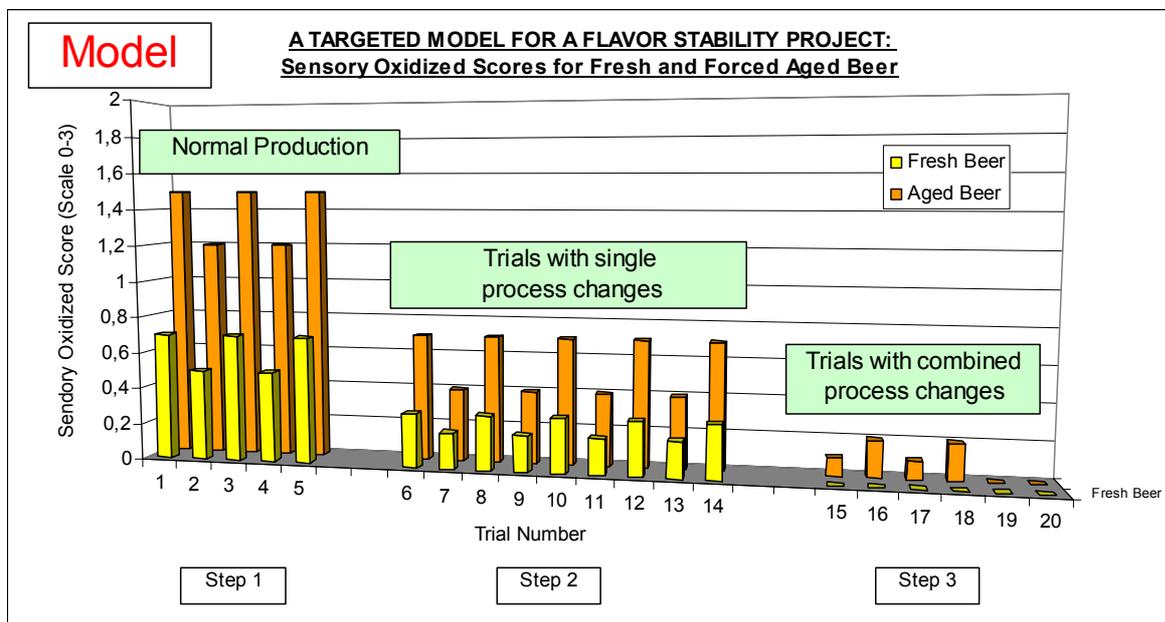


Figure 2: The aim of the project is to have reduced the brands susceptibility to oxidation by the final trial, 20 in this example.

However it should be noted that the suggested approach can also be implemented for other problem areas within the brewery operation.

Results and Discussion

In the project described below, a total of 16 full-scale trials were produced (2200 hl) from the full set of 20. The final set of 4 results for the project being reported here were not available when MVA was carried out.

Analysis of Trials grouped by sensory staling attributes

For the multivariate analysis (using Unscrambler® software), the first step was to analyze the grouping of the trials against sensory staling as measured by the taste panel.

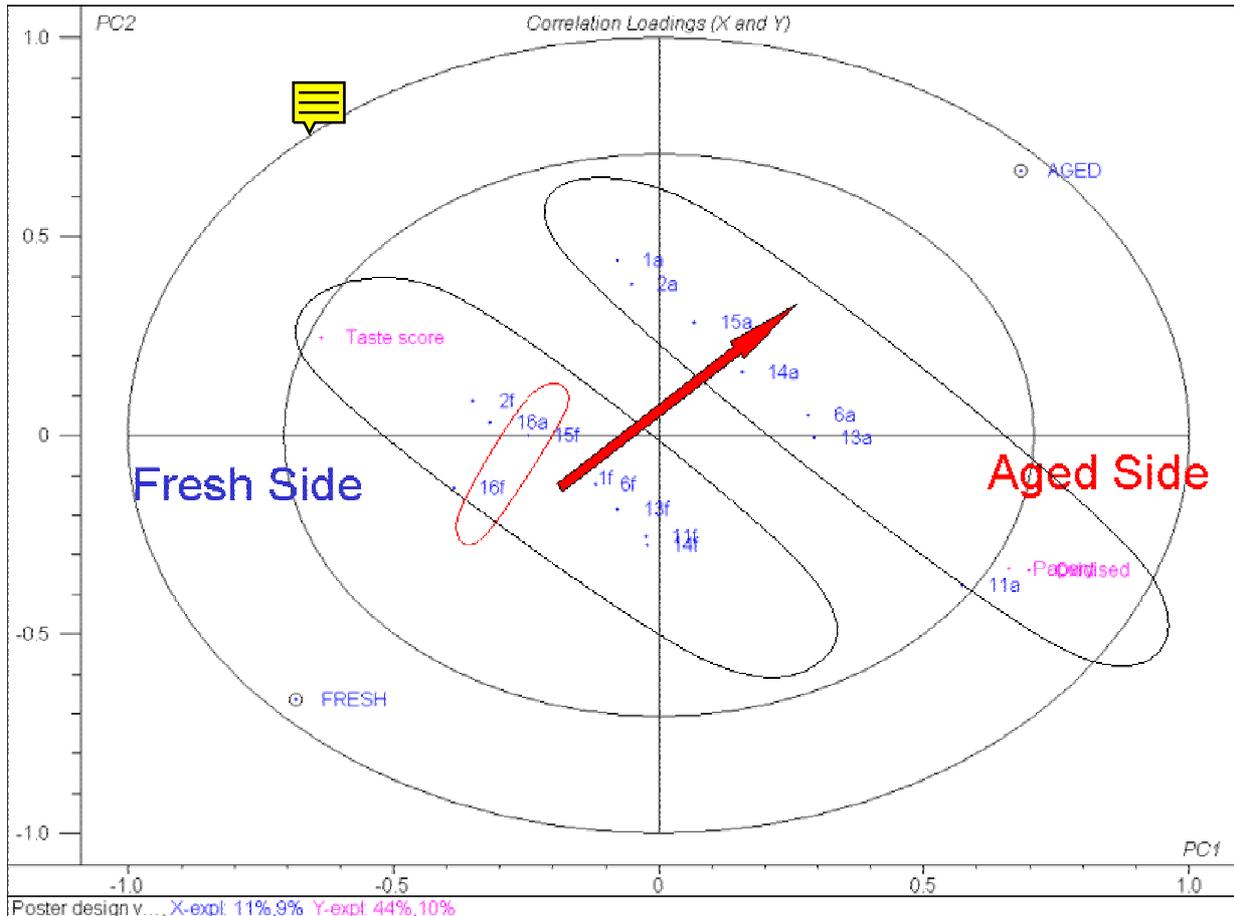


Figure 3: Multivariate Partial Least Squares Regression (PLSR) correlation loadings plot. Main design variables (Fresh and Aged) and beer trial indicators (selection of trials 1 to 16, f = fresh and a = aged) in the X-matrix and sensory terms in the Y-matrix. Ellipses represent $r^2 = 50$ and 100 % explained by the model. Significance at the 5 % level is indicated by circled variable points.

Figure 3 shows a MVA correlation loadings plot of Principal Component 1 (PC1) versus Principal Component 2 (PC2) for some of the trials. In this case, PC1 (the x axis) displays the main source of variation in the trials. This is freshness on the left hand side (see Taste score label) to staling on the right hand side (see the oxidized and papery labels). It is clear that the fresh samples (indicated with a "f", e.g. trial 6f) correlate with the fresh side and the aged samples (indicated with a "a", e.g. trial 6a) with the aged side. This analysis in itself is a validation that the taste panel is sensing the stale flavors present in the beers through differentiating the beers into fresh and aged groups. It can be noted however that some trials were more resistant to ageing than others. Trials 1, 2, 15 & 16 are shown to

have the greatest distance to the papery and oxidized flavors as indicated on Figure 3 (to the left side of the arrow). Trials 15 and 16 were produced using the knowledge gathered throughout the project with the purpose of making a very flavor stable beer. Trial 16 is shown in this plot to be especially resistant to oxidation as the fresh sample 16f is placed even further to the left than the fresh group and the aged sample 16a is actually described by the plot as being in the fresh group.

Correlations between sensory attributes and staling components

A second step is to plot the sensory results against corresponding staling chemical results, to serve as an additional level of validation of the taste panel's ability to discriminate between fresh and stale beer. It also serves to clarify which of the chemical components measured can be considered main indicators for staling for the particular brand.

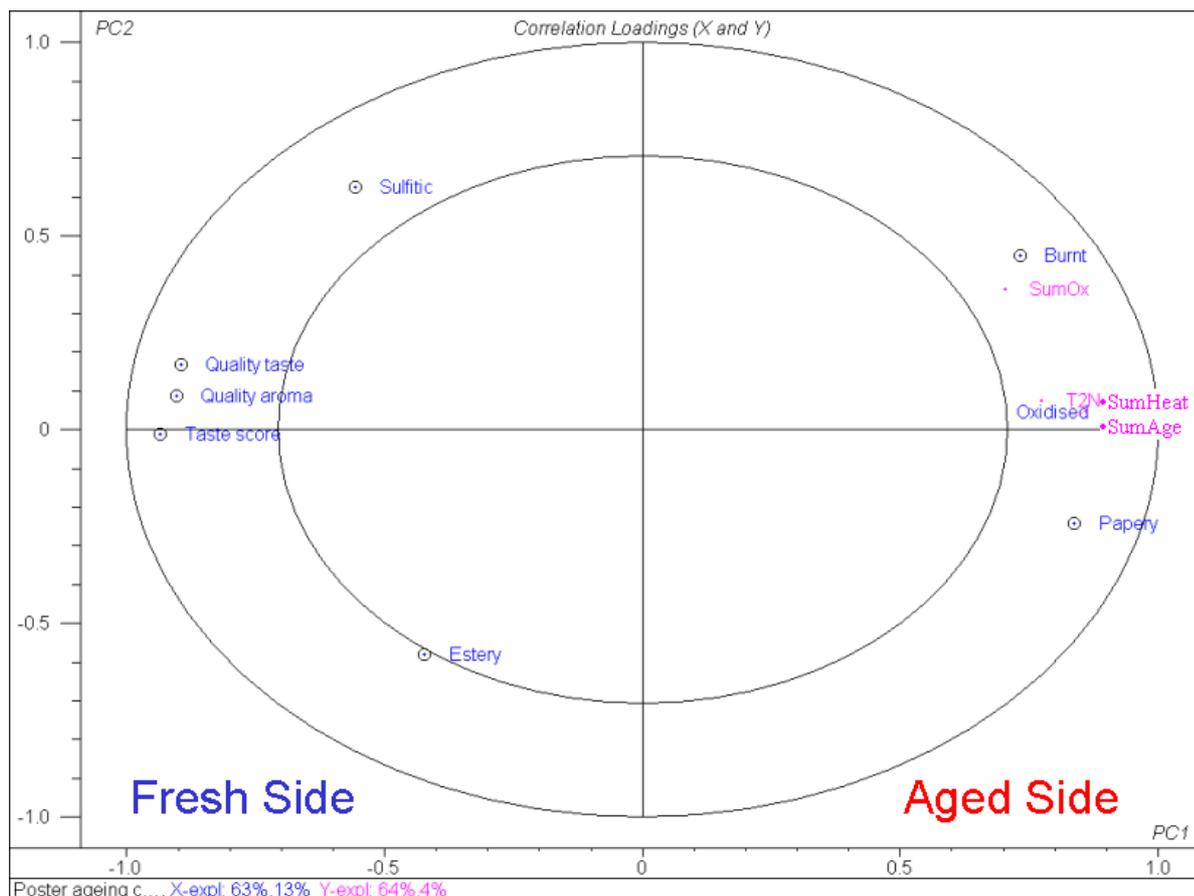


Figure 4: Multivariate Partial Least Squares Regression (PLSR) correlation loadings plot. A selection of sensory descriptors in the X-matrix versus Trans-2-Nonenal, and the sum of oxidation, heat and ageing chemicals (for 16 full-scale trials) in the Y-matrix. Ellipses represent $r^2 = 50$ and 100 % explained by the model. Significance at the 5 % level is indicated by circled variable points.

Figure 4 displays once again that PC1 contains the main source of variation, fresh to staling/ageing. It can be seen on the right hand side of the plot that the chemicals trans-2-nonenal (T2N) and the sum total of various oxidation related chemicals (e.g. benzaldehyde), heat load related chemicals (e.g. 2-furfural) and ageing related chemicals (e.g. 2 acetyl furan) in the beers, correlate with the sensory descriptors burnt, oxidized and papery as determined by the taste panel. It can also be seen that the “fresh” descriptors

(Quality taste, Quality aroma and Taste score) are grouped closely together on the far left of the plot (the fresh side).

This is an additional level of validation that the taste panel has sensed the stale flavors present in the beers. Moreover, and more importantly it indicates which chemical measurements can be used to predict the potential ageing stability of the beers as they are produced with respect to ultimate sensory effects.

This highlights another benefit of MVA. Through the determination of correlated measurements, excessive measurements can be reduced, which removes duplication and saves costs (e.g. if two chemicals correlate, then just measure one in the future).

Correlations between trials conducted and staling indicators

A third step of analysis was conducted to determine which trials and therefore, processing parameters were significant in reducing the stale flavors in the aged beers in the present investigations.

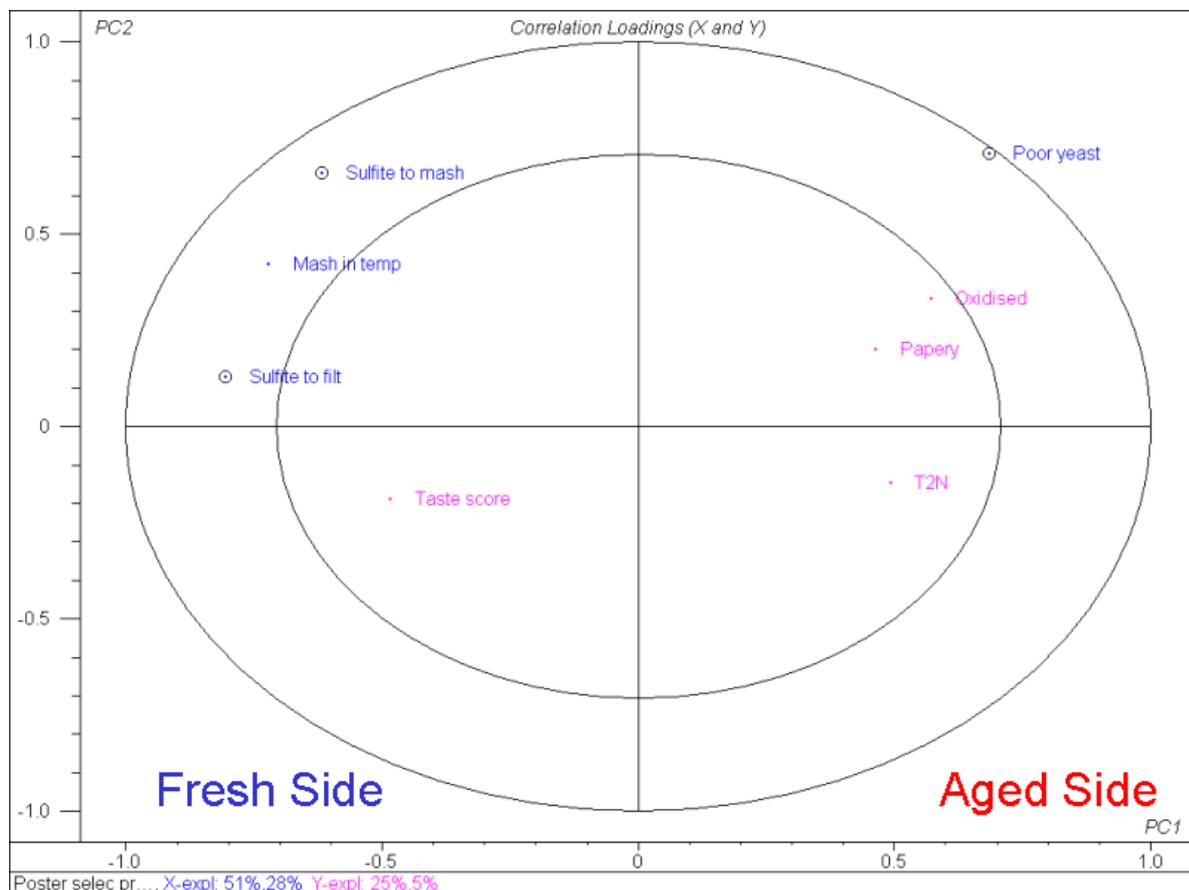


Figure 5: Multivariate Partial Least Squares Regression (PLSR) correlation loadings plot. Four important process parameters (1.sulfite added to mash, 2.higher mashing in temperature, 3. sulfite added to filtered beer , 4. poor yeast vitality and viability) in the X-matrix versus selected fresh and ageing indicators (taste score, oxidized, papery and T2N) in the Y-matrix. Ellipses represent $r^2 = 50$ and 100 % explained by the model. Significance at the 5 % level is indicated by circled variable points.

Figure 5 shows that PC1 (the x-axis) reflects staling to the right (where oxidized and papery flavors and T2N are positioned) and freshness to the left (where a good Taste

score is positioned). The four process parameters that correlate with, and thus result in, a higher level of freshness in the aged beer samples, are:

- Increasing the mashing-in temperature (to 60°C) (label “Mash In temp” on the plot)
- The addition of sulfite to the mash (label “Sulfite to mash” on the plot)
- The addition of sulfite to the filtered beer (label “Sulfite to Filt” on the plot)
- And using yeast of a high vitality and viability (label “Poor yeast” on the plot)

In this project it was clearly seen that poor yeast health had a major influence on flavor stability in the present example. Thus, of the many known actions possible to improve flavor stability, it would be most beneficial for this particular brewery to focus on ensuring better yeast health through improved processing methods, to achieve better flavor stability.

Correlations between yeast health process parameters and staling indicators

A further step is to analyze the general process data against both sensory and chemical staling indicators. The total process data will consist of data sets from raw materials, brewhouse, pitching yeast, fermentation, storage, filtration and packaging. These different data sets may be analyzed against the staling indicators separately, in order to assess their effect on the staling of the beers.

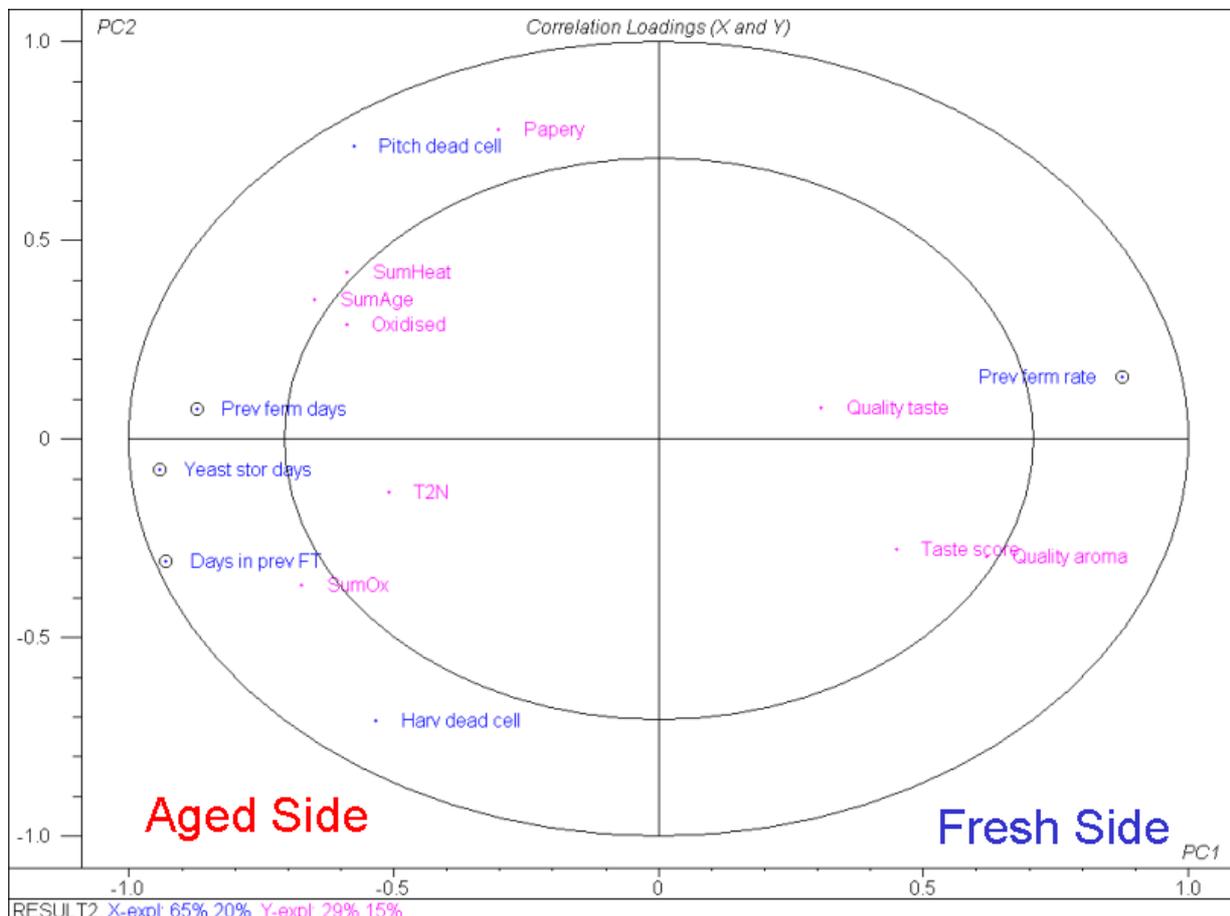


Figure 6: Multivariate Partial Least Squares Regression (PLSR) correlation loadings plot. Yeast vitality and viability in the X-matrix (Pitching yeast dead cells%, Fermentation and Total days in the tank the pitching yeast is coming from, Yeast storage day, harvest % dead cells) versus selected fresh and ageing indicators in the Y-matrix (Taste score, quality of aroma and taste, T2N, Sum of oxidation, heat load and ageing related chemicals, oxidized and papery flavor). Ellipses represent $r^2 = 50$ and 100 % explained by the model. Significance at the 5 % level is indicated by circled variable points.

Figure 6 shows an example of plotting a set of process data against sensory and chemical staling descriptors. PC1 (the x axis) indicates aged characters to the left (both flavors and chemicals) and freshness and good quality to the right. Correlating with an increase in ageing chemicals and aged flavors are:

- A higher % of dead cells in the pitching yeast
- A longer fermentation time in the fermenter the pitching yeast is coming from
- A longer period of waiting time before harvest in the fermenter the pitching yeast is coming from
- A longer yeast storage time in the yeast storage tank
- A higher level of harvest dead cells from the fermenter the beer is brewed in

Correlating with freshness is:

- An adequate fermentation rate in the fermenter the pitching yeast is coming from

This plot shows that this particular brewery has a fluctuating yeast vitality and viability and that poor yeast health is having a detrimental effect on flavor stability.

Trials 15 and 16

From the knowledge gained from the results of the first 14 trials, two more trials (trials 15 and 16) were conducted with the following process changes:

- Increasing mashing-in temperature from 45°C to 60°C
- Sulfite addition to the mash
- Pitching with very vital and viable yeast
- Sulfite addition to the filtered beer

In addition, Trial 16 also had

- Slightly lower wort aeration than normal (7.5ppm O₂ instead of 10ppm)

Both trials were fermented with the same yeast so as to be able to compare the effect of the lower wort aeration. The effect on the oxidized flavor for the 16 trials can be seen in Figure 7.

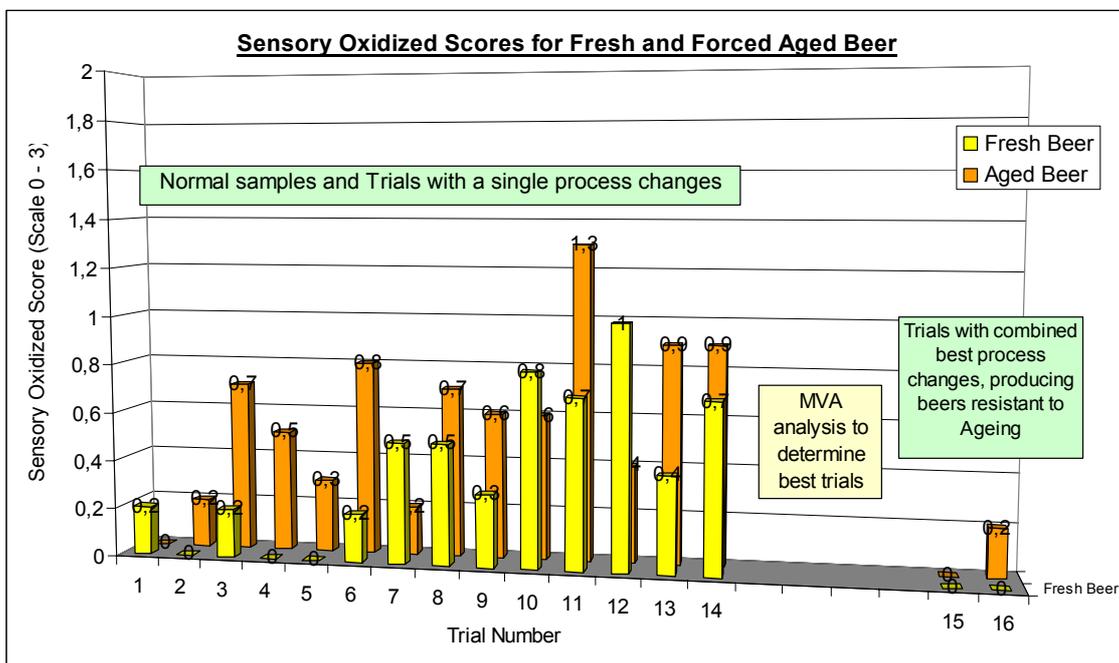


Figure 7: The sensory oxidized flavor in fresh and forced aged samples of trials 1 to 16.

In Figure 7 it can be seen that Trials 15 and 16 were very resistant to oxidation, as measured by the taste panel. A similar result was seen for the papery flavor and staling chemicals.

Overall, the multivariate analysis indicated that trial 16 was the best trial (see Figure 3) as MVA takes into account many aspects of the nature of the beer simultaneously (e.g. several staling off-flavors and chemicals). The pattern shown in Figure 7 plot for sensory oxidation levels in the trials, is useful to show that the methods used in this project significantly improved the flavor and flavor stability in this beer brand made in full-scale production.

Repeated trials to confirm the obtained results are correct are being carried out in Autumn 2002.

Conclusions

- Correlating flavor results with chemical and process data is a valuable methodology in the improvement of flavor and flavor stability, in full-scale production.
- Multivariate Analysis is necessary to analyze the data, as brewing is a multivariate process. The results provide the basis for making qualified decisions in changing process parameters to improve flavor and flavor stability.
- Through the determination of correlated measurements using MVA, excessive measurements can be reduced, which removes duplication and saves costs (e.g. if two chemicals correlate, then just measure one in the future).
- In this project it was confirmed that the taste panel's aged flavor descriptors correlated strongly with many staling indicators such as trans-2-nonenal and other oxidation, heat and ageing chemicals. The Multivariate analysis highlighted that a main contributor to the brand's unsatisfactory flavor stability was poor yeast handling. This has allowed the brewery to efficiently focus its attention and resources into this process area, in order to achieve the most substantial improvements in flavor stability. The health of the pitching yeast was dependent on:
 - An adequate fermentation rate in the fermenter the pitching yeast was coming from
 - A low number of total days in the fermenter the pitching yeast was coming from,
 - A minimum number of yeast storage days
 - A minimum number of dead cells in the yeast storage tank.
- Furthermore, full-scale beer with a high resistance to staling (Trials 15 & 16) were produced by the following process changes:
 - Increasing mashing-in temperature from 45°C to 60°C
 - Sulfite addition to the mash
 - Pitching with very vital and viable yeast
 - Sulfite addition to filtered beer
 - Slightly lower wort aeration than normal (7.5ppm instead of 10ppm)

More trials are in progress to confirm the obtained results.