M. Gastl, S. Hanke and W. Back

Analytical investigations to evaluate bitter sensation using a taste sensing system

The bitter impression created by hops is a special characteristic of beer. The range of bitter units in beer is normally between 20 and 50 IBU (International Bitter Units). Apart from their preservative effects hops are used to give the beer the right bitterness and aroma. [1] The influence of hops to the aroma and flavour of beer can be enhanced by the use of hop products and pre-isomerized hop products in the brewhouse or after fermentation.
Iso-alpha-acids are intensely bitter and are the main origin of bitter flavour because they are responsible for more than 75 % of beer bitterness. [2] Beside the influence of iso-alpha-acids there are many other influencing factors, which affect the sensation of bitterness. The beer matrix is very complex and as a result of those various ingredients causes matrix effects and interactions between the ingredients. The matrix effects are able to cover the bitter sensation. This is already well known by the sensorial evaluation of flavour stability and ageing effects of beer. In this case for example the hop flavour could have a positive masking effect.
A search of the literature reveals that humans have an inherent preference for sweet and umami and an inherent dislike for bitter and sour. The sensory perception is not linear nor is bitterness a taste attribute which is extremely well developed in human beings (compared to herbivorous animals) and is significantly influenced by matrix effects and therefore with regard to sensory aspects there is always a discrepancy in analytical measuring and sensory evaluation. [1] In cooperation with "TecLabS" we are testing an electronic tongue (Taste Sensing System SA402B, Intelligent Sensor Technology, Inc.) with regard to bitter sensation.

Descriptors: taste sensor, beer matrix, matrix effects, iso-alpha-acids, drinkability

1 Introduction

The human gustatory system is able to detect very complex matrices and detects the taste as harmonic or inharmonic. The bitter sensation is a typical characteristic of beer and hopped beverages. Sensory evaluation in the food industry is usually done by a human taste panel. The problem with human panels is the motivation of the tasters, the availability and the physiological situation of the tasters. The project to test an analytical method to evaluate the bitter sensation against the sensory evaluation by a human taste panel was investigated together with the German company TecLabS using a Taste Sensing System which is already established in Japanese breweries. The Taste Sensing System mimics the human gustatory sensory system. This publication shows first results comparing the information from the Taste Sensing System with a human taste panel with respect to bitter taste sensations in aqueous solutions of different compositions. The investigations showed good correlation between the measurement of iso-alpha-acids and the human sensory panel. These trials are limited to the bitter sensation caused by iso-alpha-acids. Following this work the plan is to transfer the results in aqueous solution and to repeat the trials in beer matrix to get more information on matrix effects. It is noticeable that matrix effects have a great influence on the perceived bitter sensation.

In beverage industry the term 'drinkability' is commonly used. Nevertheless there is no uniform definition for the true meaning of the term 'drinkability'. Many different influencing factors are known, which are important for good drinkability. However drinkability is linked to the matrix composition of beer definitely. The composition of the matrix, especially a specific harmony, seems to be most important. In this context the bitter impression created by hops has to be considered and the influence of matrix effects on the sensorial bitter sensation. The use of an electronic tongue could potentially be used to help understand the complex subject of drinkability with regard to bitter sensation.

2 Materials and methods

2.1 Human taste and taste sensor

The human taste sensory system is very complex and includes the:

- gustatory,
- olfactory,
- haptic,
- optic sensory system (light, colour).

A taste sensor with global selectivity consists of several kinds of lipid/polymer membranes for transforming the information from taste substances into electric signals. [3] Non-volatile compounds form the basis of taste perception. Taste consists of five principal attributes: quality, intensity, temporal dynamics, spatial topography and hedonics. Sensation is composed of 5 basic categories (sweet, bitter, sour, salty and umami). Intensity increases with rising concentration of the tastant. [4] Responses in gustatory receptors of many animals increase linearly with the concentration.

The human taste sensing system is very sensitive and multifaceted. That is why it is so difficult to simulate human taste. Beside

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the 5 basic taste perceptions the human sensory system has the ability to recognize:

- texture, body, mouthfeeling,
- ratio of components,
- aroma,
- balance,
- harmony,
- development of taste impression,
- off-flavours (e. g. diacetyl).

2.2 Taste Sensing System

The principal of the Taste Sensing System (Taste Sensing System SA402B, Intelligent Sensor Technology, Inc.) was presented at the EBC Symposium where the phenomenon of 'drinkability' was discussed. The results from this Taste Sensing System are outlined below. [5]

The taste sensor would appear to be a useful tool for process monitoring and the quality control of foods. Transformation of the sensor output of the Taste Sensing System into taste information is based on Weber's law. Webers law (1834) simply says that the size of the difference is proportional to the original stimulus value. The "difference threshold" or "just noticeable difference" is the minimum amount, by which stimulus intensity must be changed in order to produce a noticeable variation in sensory experience. The difference is 20 %. Weber's law can be applied to a variety of sensory modalities (e. g. brightness, loudness, mass etc.). [5] Figure 1 shows the prinicipal of Weber´s law.

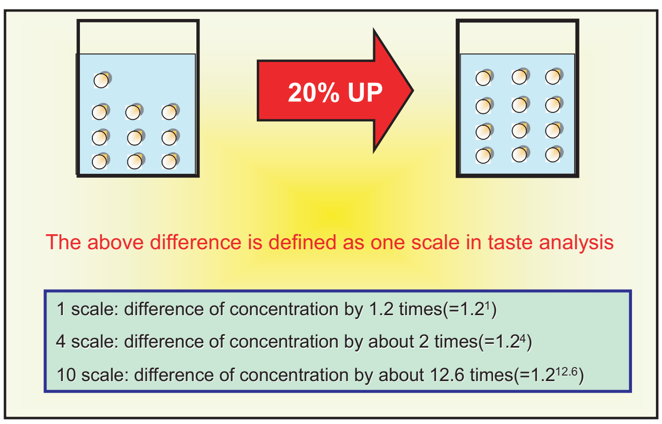

Fig. 1 Taste information based on Weber's law

In the 1990's Toko and co-workers developed the first nonspecific membrane based sensor [6], which was improved in following years. [3, 7] Figure 2 shows the Taste Sensing System SA402B from Insent Intelligent Sensor Technology, Inc. Japan [3] with its sensors.

Toko et al. give the system global selectivity, which means the system has the ability to classify different substances in different groups by taste similar to the human gustatory system. Taste is an objective factor. [3, 7] The taste sensor does not measure a molecule or a substance, but it expresses objective taste information qualitatively. [8] Figure 3 explains the composition of the electrodes of the Taste Sensing System.

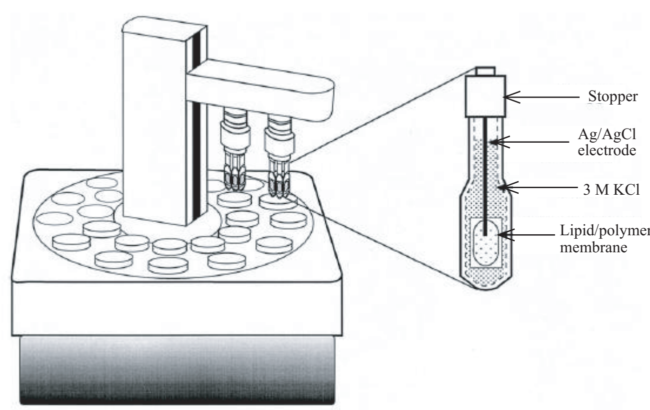

Fig. 2 Taste Sensing System SA402B [3]

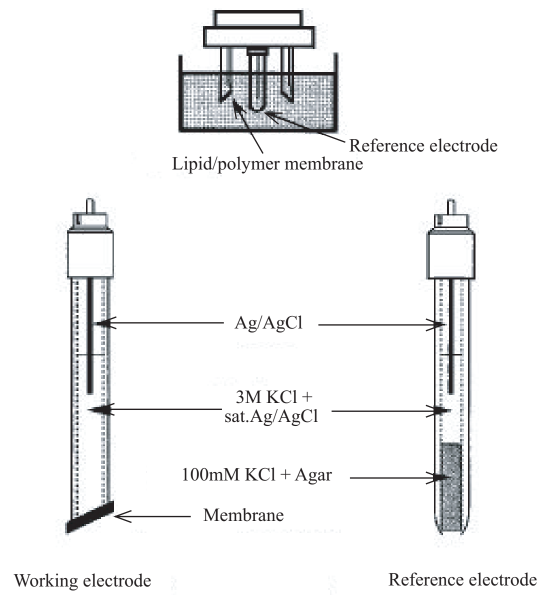

Fig. 3 Electrodes of the Taste Sensing System [9]

2.3 Principle of the measurement procedure:

Figure 4 shows the different steps and results of the measurement procedure.

1. Washing: Sensors are washed by washing solution.
2. Sensor check: Sensors are immersed in the reference solution, sensor outputs are measured afterwards.
3. Measure relative value: Sensors are immersed in the sample solution and then the sensor outputs are measured. As a result, the "relative value" is acquired.
4. Slight washing: Sensors are simply washed by washing solution.

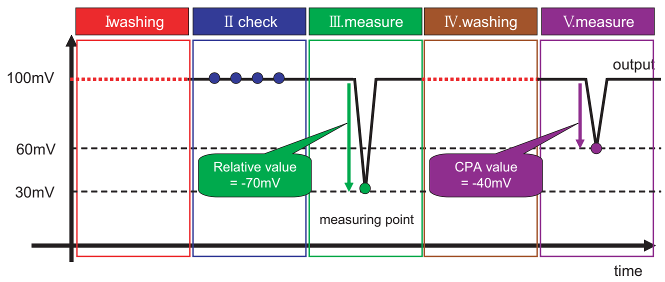

Fig. 4 Measurement procedure [Kobayashi, Y.; Intelligent Sensor Technology, Inc.; personal communication]

5. Measure CPA value: Sensors are immersed in the sample solution; thereafter sensor outputs are measured. As a result, the "CPA value" is acquired and the specific aftertaste calculated.

The System contains different sensor types with different sensor properties. Sensors "AE1" and "C00" exemplify the sensor properties for astringency and bitterness in Table 1. Aftertaste-A describes the aftertaste derived from astringency and aftertaste-B the aftertaste from bitterness (see Figure 4 step IV and V).

Table 1 Property of sensors

sensor	relative value
AE1	slight astringency from astringent substance (tannic acid)
C00	slight bitterness from acidic bitter substances (iso-a-acids)

sensor	CPA value
AE1	aftertaste from astringent substance (tannic acid)
C00	aftertaste from acidic bitter substance (iso-a-acids)

The Taste Sensing System is able to express the results as 2D scatter plot or radar chart (spider web). The taste sensor offers 8 types of taste information, which are pictured in Figure 5 as radar chart.

3 Experimental setup

The intention of this work was simulating the composition of beer ingredients in an aqueous solution and measuring the effect of the bitter taste impression by a human sensory panel as well as a Taste Sensing System. The Taste Sensing System mimics the human gustatory sensory system. In the focus of these investigations is to point out if this system could potentially be used in this field.

One of the main factors to consider is the sensory cognition of different compositions of beverage ingredients (the composition of the basic matrix: e. g. sugars, ethanol, bitter substances). The following trials were done at the Lehrstuhl für Technologie der Brauerei I to check the Taste Sensing System SA402B in cooperation with the TecLabs Company to find an analytical method to measure the influence of beer matrix on the impression of taste bitterness (iso-alpha-acids) in comparison with the bitter perception of a human taste panel. In the first part of the investigations the influence of the matrix composition of the basic non-volatile ingredients (simulating the composition of beer ingredients: variables are ethanol, dextrines, pH) is presented and how the bitter impression is being influenced by the matrix effects were checked in aqueous solution.

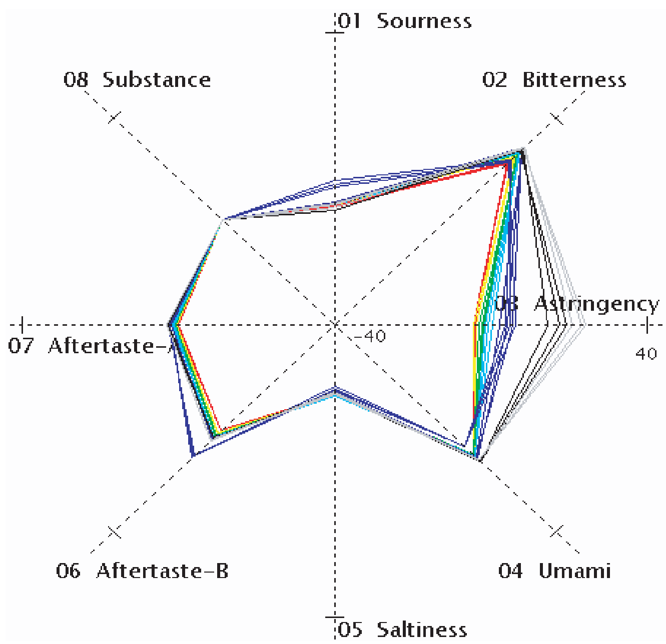

Fig. 5 Taste Information of the Taste Sensing System

This suggests that the impression of taste bitterness depends on the composition of the defined matrix. Bitterness and the perception of it have a great influence on the hedonic value of beer. On the one hand hop derived bitterness will be perceived very intensely in water, but on the other hand it behaves differently in beer. [10-13] This leads to the conclusion that there are some beer constituents which change the bitterness perception. Ethanol has an increasing effect on the perception of bitterness. [10, 11, 14, 15] Dextrins [16], carbohydrates [12], the pH-value [17], different rates of dissociated isohumulones [2] and phenolic compounds (catechine, epicatechine) [18] are also believed to effect different perceptions of bitterness.

Trials were done to investigate the influence of the main substances of beer matrix on the impression of taste bitterness using iso-alpha-acids (0–40 mg/l in 5 mg/l steps termed IA1 – IA8, Isohop 30 %, Joh. Barth & Sohn GmbH & Co., Nuremberg /Germany). To investigate how the bitter impression is influenced by the matrix effects, different aqueous matrix solutions are used. All chemicals used (substances etc.) are p.a. analytical grade: Dextrine (Maltrin M150 and Maltrin M180, Grain Processing Corporation, Muscatine/USA), citric acid monohydrate (p.a., Sigma-Aldrich, Seelze/Germany), ethanol (p.a., Riedel-De Haen, Seelze/Germany) and solved in ultrapure water (Milli-Q plus, Millipore, Billerica/USA).

The analysis of bitter substances with HPLC and photometric methods were done according to MEBAK and EBC standards. [19, 20] Sensory evaluation of the solution was carried out by the taste panel of Lehrstuhl für Technologie der Brauerei I. The intensity of the bitter taste was scored from 1 to 10 according to modified Kaltner schema. [23] Table 2 presents the experimental setup.

3.1 Experimental design

Table 2 Experimental setup

variable	matrix	steps per trial
iso- α-acids 0–40 mg/l 5 mg/l steps	ultra pure water	9
	+5 % dextrins	9
	pH 4.5	9
	pH 7	9
	+5 % ethanol	9
	+0.5 % EtOH	9
	+5 % dextrins, pH 4.5 (citric acid)	9
	+5 % dextrins, pH 7	9
	+5 % dextrins, 5 % EtOH	9
	+5 % dextrins, 0.5 % EtOH	9
	+5 % dextrins, pH 4.5+ 5 % EtOH	9
	+5 % dextrins, pH 7+ 5 % EtOH	9
	+5 % dextrins, pH 4.5+ 0.5 % EtOH	9
	+5 % dextrins, pH 7+ 0.5 % EtOH	9
	total sum	**126**

4 Results and discussion

To check the sensitivity of the Taste Sensing System samples of an increasing iso-alpha-acid concentration (0–40 mg/l = Loesung 1–8) in aqueous solution (pH 4.5) related to a standard solution (Standardloesung) were analysed. The signal of the Taste Sensing Systems showed a good correlation with increasing concentrations and agreed with the human taste sensing panel regarding the impression of bitterness. The Taste Sensing System showed good analytical results for the measurement of iso-alpha-acids and also quinine, which was investigated at the same concentrations. Figure 6 shows the results in 2D scatter plot of iso-alpha-acids.

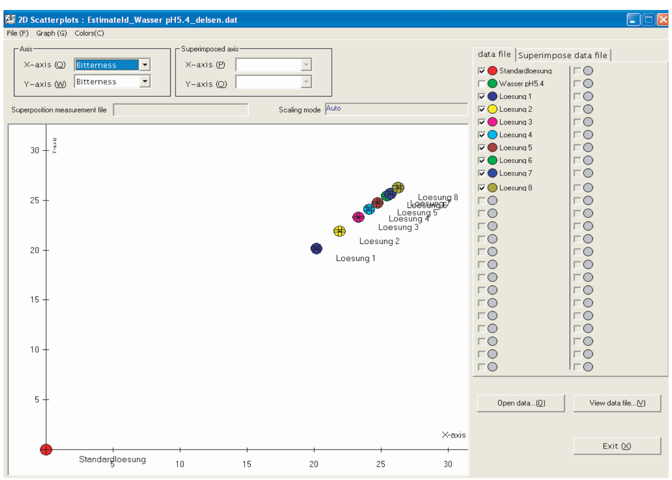

Fig. 6 Iso-alpha-acid concentration (0–40 mg/l)

The results were controlled in a 5 % dextrin solution and verified with photometric and HPLC methods. Figures 7 and 8 show a good correlation between the photometric measured IBU and the measured concentration of iso-alpha-acids and the bitterness signal of the Taste Sensing System in a 5 % dextrin solution.

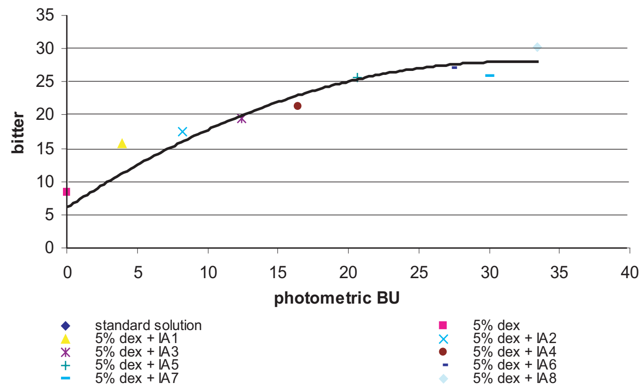

Fig. 7 Correlation between the photometric measured BU and the bitterness signal of the Taste Sensing System in a 5 % dextrin solution

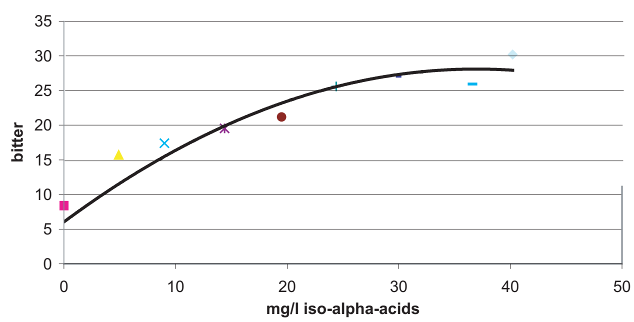

Fig. 8 Correlation between iso-alpha-acids in mg/l and the bitterness signal of the Taste Sensing System in a 5 % dextrin solution

These trials followed investigations of different matrix solutions (5 % dextrins + 0.5 % EtOH + pH 4.5; 5 % dextrins + 0.5 % EtOH + pH 8.0 and 5 % dextrins + 0.5 % EtOH + pH 4.5 and 5 % dextrins + 5 % EtOH + pH 4.5, etc.) The results of the Taste Sensing System showed that the bitterness sensation was lowered by a lower pH value. This was confirmed by our experience of tasting. The results are confirmed if the signals are based on the standard solutions. An increase of ethanol as well as higher pH levels leads to a higher astringent impression and a higher "aftertaste B".

Figure 9 contains the data from the bitterness signal of the different measured matrices samples. The plot shows a correlation between the photometric BU and the signal of bitterness from the Taste Sensing System (n = 135) of the different measured aqueous matrix solutions in principle. The distribution of the measuring points at 0 IBU derived from the different basic bitterness of the aqueous solutions (e. g. ethanol tastes bitter [21]). It has to be mentioned that normally the bitter sensation of a human taste panel perceived similar over a threshold level of about 25–30 IBU. The data underline that the Taste Sensing System is suitable for measuring the bitterness caused by iso-alpha-acids in different matrix compositions. Therefore the Taste Sensing System could be used for the measurement of bitterness in the brewing industry. The figure point out that there is generally a correlation between the photometric BU and the bitter signal, but only measured by one bitter sensor and caused by iso-alpha-acids. Normally the bitter sensation in beer is caused by many other factors (e. g. proteins, phenolic compounds, ethanol), too. This investigation should display a starting point for the analytical measurement of the bitter sensation.

By comparing the different solutions a good correlation was found between bitter taste, astringent taste and aftertaste A and B and the concentration of isohumulones. The concentration of isohumulones was measured as described previously.

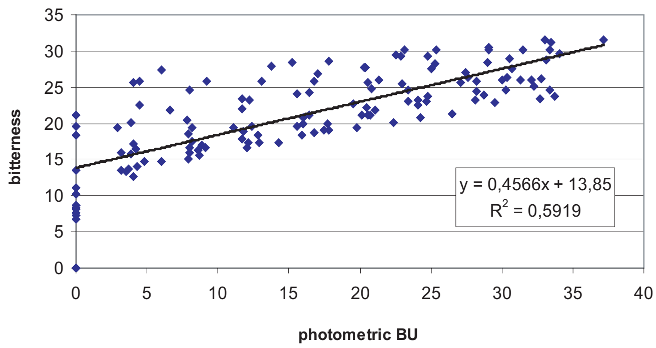

Fig. 9 Correlation between photometric BU and bitterness measured by the Taste Sensing System in aqueous solutions (n = 135)

For example Figure 10 shows the dependence of the bitter signal aftertaste-A and the photometric measured BU in a 5 % dextrin solution. Other taste impressions which are linked with bitterness (aftertaste and astringency) respectively correlated to a human bitter impression are also positive correlated to the concentrations of the isohumulones.

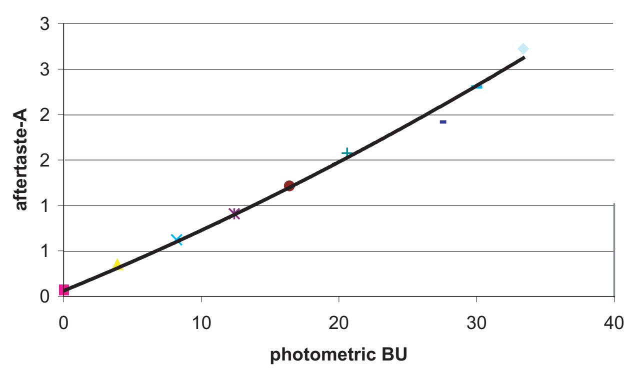

Fig. 10 Correlation aftertaste A and photometric BU

These results were similar in all experiments. In consequence the Taste Sensing System is able to taste isohumulone derived bitterness depending on the matrix. The output of the Taste Sensing System correlates very well with the experience of our taste panels. In consequence the Taste Sensing System is useful for comparing different products, but up to now only aqueous solutions are presented with a well-known matrix.

To evaluate the signal output for bitterness, astringency, aftertaste-A and aftertaste-B in detail and for comparison with each other the trials were always done with the standard solution in position A and in position B the solution without any added isohumulones. Therefore all samples referred to the standard solution and matrix without addition. The difference in the output pattern is shown in the following two charts (Figures 11 and 12). In Figure 11 the taste impression from the Taste Sensing System is related to the 5 % dextrin solution (matrix) and in Figure 12 the taste impressions are related to the standard solution (30 mM KCl + 0.3 mM tartaric acid). This difference shows that the system is capable of measuring effects caused by different beverage ingredients. To compare different matrices all measurements were referenced to standard solution. The isohumulones gave an intense bitter taste and also an astringent taste. But the aftertaste is mainly bitter and not astringent. This again correlates well with our tasting experience. A good beer shows a fine bitterness and no lingering aftertaste.

After the pre-examinations in aqueous solution Figure 13 shows first results of the correlation between the bitter signal and the perceived bitter intensity of the human taste panel measured in different beer samples. The results affirm that there is also a correlation between bitter sensation and bitter signal of the Taste Sensing System in beer matrix.

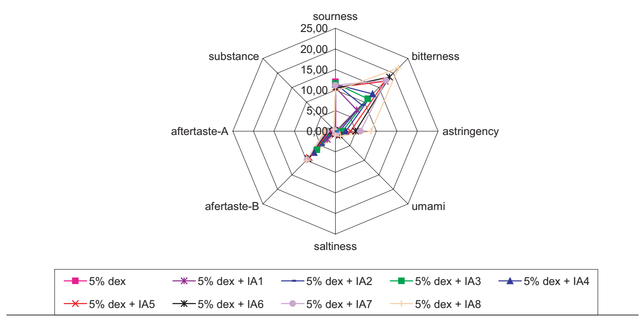

Fig. 11 Increasing iso-alpha-acid concentrations in 5% dextrin solution

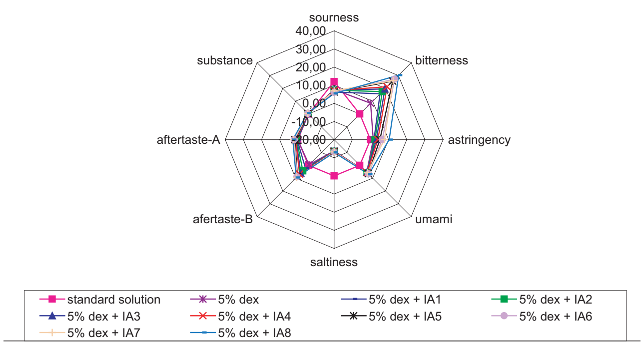

Fig. 12 Increasing iso-alpha-acid concentrations in 5% dextrin solution related to standard solution

5 Bitterness as a key factor affecting drinkability

With the trend towards high quality beers and many new beverage products, drinkability is increasingly important to the beverage industry. The production of beverages with high drinkability provides an opportunity to acquire customers, because high quality products with a high drinkability are in demand by the general public.

It is not easy to understand the meaning of the term 'drinkability', because drinkability is one word to describe many attributes like a good harmony, balance, digestibility, acceptability, moreishness, preference etc. Mattos and Moretti described the drinkability of beer as follows: "A beer that has a good drinkability is one that invites the drinker to have another glass. Drinkability is better used to represent an affective attribute rather than a descriptive attribute of beer". [22] A more extensive definition for drinkability also implies the perception and "well-being" of the drinker and distinguishes drinkability clearly from the misleading, but often used term "preference". Drinkability means a specific harmony and balance of a beverage as well as the stimulation to consume another drink. It is the characteristic of a tasty beverage that it invites the consumer to drink another glass. [5, 22] So it is clear that the composition of the matrix, especially the harmony of the matrix composition, affects drinkability. The bitterness created by hops or proteins is a key factor for the harmony of a beverage and in consequence for the drinkability.

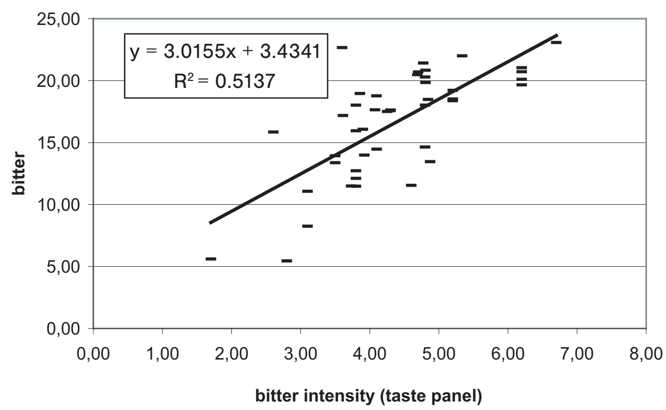

Fig. 13 Correlation between the bitter signal measured by the Taste Sensing System and the perceived bitter intensity by the taste panel in beer (n = 44)

Figure 14 shows the main influencing factors on drinkability due to the composition of the matrix. There are several relevant factors (matrix ingredients) in the beverage matrix that influence drinkability.

The balance and harmony of the beverage matrix seems to be the basis of good drinkability. Some key data like "ratio" are already well known in the non-alcoholic beverage industry to describe the ratio of sweetness to sourness (sugar content [g/l]/total acid content (calc. to citric acid) [g/l]), which contribute to the harmony and balance of the beverage. For the analytical evaluation of drinkability the starting point is to detect individual matrix ingredients and substances, which are positive or negative for drinkability. In addition, the harmony and balance of a beverage can be evaluated by the ratio of the different ingredients (proteins, sugars, ethanol) or substances (e. g. chemical or aroma substances, bitter substances), especially the influence of the volatile aroma compounds on the sensory perception of a beer have to be considered. Some substances (for example proteins) or aroma compounds are more responsible for good drinkability than others. For example the aroma compound diacetyl intensifies the impression of fullness and body. The composition of the matrix contributes to a high drinkability with a good balance and harmony of the components interacting with each other. In addition, matrix effects depend on the interactions between food ingredients. Beside masking and suppression these interactions could lead to synergistic or increasing effects, which influence drinkability. In this context it would be interesting to investigate more key data like "ratio". In summary, there are many different factors that influence drinkability; especially the beer matrix or rather the composition of it. Bitter sensation is significantly influenced by the beer matrix.

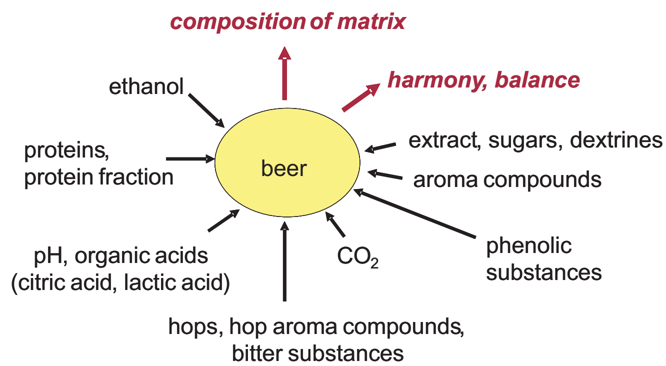

Fig. 14 Influence factors on drinkability

As well as the so called composition of the chemical compounds or matrix, some other positive or negative criteria on drinkability like the visual and sensory impression of the beverage, location of the consumption, characteristics of the customers and ingestion (stomach fullness is often assumed as key factor) have to be considered when evaluating the drinkability of a beverage. But drinkability is, like flavour stability, a complex topic, so it is not expedient to find a standard evaluation for drinkability. Analysis of these substances and the interaction between them seems to indicate a relationship between the beverage matrix and the sensory test.

With regard to beer, sensory evaluation (taste of the fresh and forced beer), flavour stability (analytical and sensory evaluation) and drinkability are closely connected parameters. In the brewing process these characteristics can also be influenced by many technological parameters such as:

- malt quality (e. g. Kolbach index),
- avoidance of thermal stress,
- avoidance of oxidation,
- hopping,
- biological acidification,
- fermentation by-products,
- good yeast quality,
- yeast management.

Because of the complexity of the human taste sensing system many attributes have to be considered such as the visual impression of the beverage and the psychological and physiological aspects of the product.

6 Conclusion

Of the many characteristics of beer, the matrix composition of the product contributes significantly to improving the harmony of the beverage. Until now, there have been few analytical methods available to evaluate a beverage objectively and to evaluate matrix effects analytically.

The results presented here describe the first trials of one analytical method designed to measure the sensation of bitterness with a Taste Sensing System. The bitter signal of the taste sensor correlated with the analytically measured bitter units/concentration of iso-alpha-acids. The aim is to establish an application of an analytical instrument for the evaluation of one key influencing factor affecting the harmony of beer. If it is possible to develop such an instrument, it could be used in addition to a human taste panel. A taste sensor gives the possibility to evaluate a beverage matrix without psychological and physiological aspects. Based on the results in aqueous solutions further trials with different matrix compositions are planned and will be carried out on beer. The results of this paper show first exploratory analysis. Based on these results we can conclude that this Taste Sensing System can be developed into an alternative measuring technique for the bitter sensation in beer. So it constitutes new possibility to measure the bitter sensation in beer.

7 Acknowledgement

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