Measurements of Beer Colloidal Stability

XIII[th] De Clerck Chair

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Beer Colloidal Stability

1. **Introduction**
2. Definitions and Basics
3. Goal
4. Management of the Colloidal Stability
5. Follow up of the Colloidal Stability
6. Measurement of the Colloidal Stability
7. Conclusions

Colloidal Stability

1. Introduction

To a certain extent,

colloidal haze does not affect the taste and should not affect beer quality;

but:

"*Most Consumers "drink with their eyes". They are often more willing to accept a glass of beer which does not taste quite right, over a glass of beer which is hazy*" (T. O'Rourke, 2002)

Indeed, the brilliance of a beer is probably the most "objective" parameter.

De gustibus et de coloris non est disputandum

So, most of the consumers will simply reject a beer that is not bright, that is hazy.

Colloidal Stability

1. Introduction

Because most of the consumers “*drink with their eyes*” …

Exceptions:

Some German Wheat beers

Belgian White beers

Abbey beers refermented in the bottle

→ overall if yeast is added in the glass ;-)

Some non filtered organic beers,
see current infatuation for “natural wines and beverages”

For some of the these beer, haze stability can be an issue

Colloidal Stability

1. Introduction

Because most of the consumers “drink with their eyes” …

- Exportation ↗

- Beer travels more and more: Transport and Storage conditions

- Rotation period in these distribution channels is longer

- More light-coloured beers; haze is more visible than in coloured beers

- **Commercial Shelf-life** for lagers: 3 months up to 1 year or more

- A non stabilized (full) malt beer does not achieve these targets easily

Therefore Beer **Colloidal Stability** is an important parameter to have under control

Beer Colloidal Stability

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Colloidal Stability

2. Definitions and Basics

2.1. Beer Stability

"The degree to which a beer tastes and looks as good at the end of its ***shelf life*** *as it did when it was first packaged"* (T. O'Rourke, 2002)

Beer Stability relates to "no" changes in sensory characteristics

- Flavour (taste - odour - aroma - texture - C02)
- Colour
- Foam
- Brilliance - clarity - brightness

or more pragmatically, it relates to "acceptable" changes …

Colloidal Stability

2. Definitions and Basics

- Brilliance – clarity – brightness

Beer is generally packaged bright, if haze is measured it is generally due to:

Biologic causes: yeast, bacteria, …

Non-biological causes: carbohydrates (starch, dextrin, glycogen, β-glucan, pentosan), mannoproteins, **polyphenols-proteins**, iron, Ca-oxalate,…,

…

But during storage, beer haze almost always increases. There is a Visual Brightness Instability : if non biological, the reasons are mostly due to

Colloidal Haze

formed by polyphenols and proteins interactions

Colloidal Stability → Focus of this lecture

Colloidal Stability

2. Definitions and Basics

2.2. Beer Colloidal Stability – Visual Brightness Stability

*"The **time** during which the brightness of a beer remains **as good** as it did when it was first packaged"*

Beer Colloidal Stability relates to "no" changes in visual appreciation of Haze

Again, more pragmatically, it relates to "acceptable" changes

Two dimensions: Time: Months
Haze: °EBC

Colloidal Stability

2. Definitions and Basics

2.3. Commercial Shelf Life

On most of the packaged beers, a date is labelled

"Period of time during which the beer should keep its intrinsic characteristics: best before date"

This is the → **Commercial or labelled Shelf-Life (CSL)**

→ Marketing decision

→ One dimension: Time

Colloidal Stability

2. Definitions and Basics

2.4. Technical Shelf Life

Technical or actual Shelf Life (TSL)

"Period of time during which the beer actually keeps its intrinsic ("visual") characteristics under specified storage conditions (for instance at 20°C)"

Different from CSL:

→ Scientific measurement

→ Two Dimensions (Time – Haze intensity)

Colloidal Stability

1. Definition and Basics

Some typical CSL values:

keg – PET lager beers: 3 months
domestic packaged lager beers: 3 to 12 months
export packaged beers: up to 12 months or more
special beers: up to 5 years

TSL specification: **time** before the **chill haze (CH)** reaches a **specific value** measured at a fixed temperature and angle, in specific storage conditions (20°C)

Specific CH value has to be defined:

beer haze in BBT: 0,4 – 0,8 °EBC

CH value: generally some tolerance; 1,5 or 2 °EBC for pale lager beers (influence of colour)

Colloidal Stability

2. Definitions and Basics

2.5. Permanent haze

Haze present in a beer at room temperature

- haze generally dissolves when the beer is heated. But with time, chill-haze changes into permanent haze which no longer disappears at these higher T°C

- is a haze in which polymerised (or oxidized) polyphenols react with proteins and where bindings between protein and polyphenols are irreversible

- particle sizes range from 1.0 to 10 µm

- the time before it appears can vary enormously

- is important for beers with a long shelf-life

- is measured at 20°C

Colloidal Stability

2. Definitions and Basics

2.6. Chill Haze

Haze formed when a beer is chilled (down to 0℃)

- It generally dissolves again when the beer is warmed up to 20℃

- Haze produced when low molecular weight polyphenols cross-link with proteins through weak and reversible interactions such as hydrogen bonds.

- particle sizes range from 0.1 to 1.0 µm

- precursor of permanent haze

- is measured at 0℃ *(conventionally, measured after a storage period of 24 hours at this T℃*

- is measured at a 90° angle

Beer Colloidal Stability

Colloidal Stability

3. GOAL

To ensure beer colloidal stability throughout its shelf life (sales period)

From a “Quality” perspective, the goal of the management of beer colloidal stability should be to bring the TSL close to the CSL

But, brewers being very pragmatic; very often: CSL > TSL

Cases where TSL > CSL also exist , money wasted...

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Colloidal Stability

4. Management of Colloidal Stability

To achieve a TSL close to CSL, colloidal stability improvements are needed to limit haze formation along storage

Many process parameters have an effect on colloidal stability (raw materials, O^2, …)

Suppliers propose more and more alternatives to stabilize the beer

Need for:

A better understanding and management of parameters related to beer colloidal stability to implement process improvements

More effective colloidal stabilisation treatments (CSL ↗)

More cost effective colloidal stabilisation treatments (Pressure on costs)

Colloidal Stability

4. Management of Colloidal Stability

Need for a better Colloidal Stability:

→ Need for adequate methods of evaluation of changes effectiveness

→ Need for adequate specifications

Specifications will depend on:

→ Defined Commercial Shelf Life (3, 6, 12, … months)
→ Defined Technical Shelf Life
Accepted haze intensity (°EBC)
Months

Offer the stability desired by the consumer, not more!

Colloidal Stability

4. Management of Colloidal Stability

So, for a defined Commercial Shelf Life

Define TSL: 2 dimensions (Time + Haze) + *Conditions*

Choose adequate evaluation method for colloidal stability

Set adequate specifications to this method

Estimate brand colloidal stability

Choose adequate methods of evaluation of the effectiveness of treatments or process changes (pilot scale, industrial trials, …)

Test and choose best treatments or process changes (cost and effectiveness)

Constraints: raw materials availability - brewery facilities - regulations, …

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Colloidal Stability

5. Follow-up of Colloidal Stability

Colloidal stability

- During storage, beer (even pasteurized) becomes hazy due to colloidal haze formation

This turbidity is caused predominantly by interacting substances such as **polyphenols** and **proteins** that can form visible colloids

Haze development is observed at low T°C (chill haze) and high T°C (permanent haze)

Follow-up of colloidal stability is defined here as the follow up of the chill haze development (°EBC) during storage (months)

Storage at 20°C (arbitrary)

Colloidal Stability

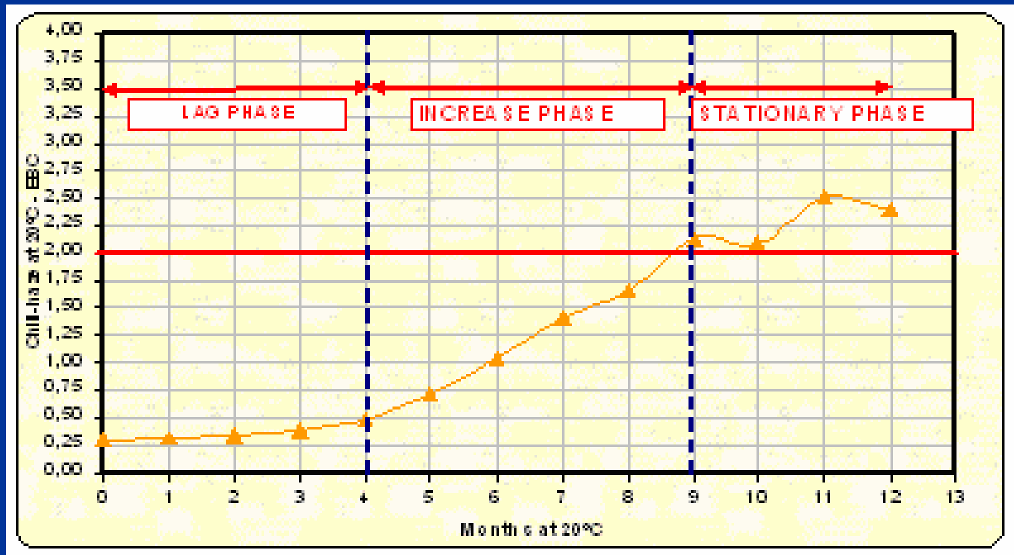
5. Follow-up of Colloidal Stability

Typical curve :

Lag phase:
No haze increase

Increase phase:
Linear haze increase

Stationary phase
No more haze increase



Colloidal Stability

5. Follow-up of Colloidal Stability

First phase: Lag or latency phase

Corresponds to the first weeks or preferably, months, of storage of the beer

Stored at 20°C, there is almost no haze increase when the beer is chilled to 0°C, no chill haze formation

Depending on flavanoids content in the raw materials and process conditions, both simple flavan-3-ols and tannoids can be present over a rather broad range of concentration in the fresh beer.

These differences between beers could account for the broad variability of lag phases between brands.

Which changes from flavanoids to tannoids do occur…

Colloidal Stability

5. Follow-up of Colloidal Stability

Second Phase: Increase Phase

The chill-haze starts to develop rapidly and apparently linearly

The rate of its formation is beer type dependent.

The polymers of flavanoids have reached a certain size and weight;

They are large enough to link several sensitive proteins together through reversible bonds to form larger aggregates.

Chill-haze is formed on beer cooling but partly re-dissolves when warmed up

Colloidal Stability

5. Follow-up of Colloidal Stability

Third and last phase; Stationary Phase:

Chill-haze development rate slows down or reaches a maximum value

This value is also beer type dependent.

The tannoids complete their polymerisation to form tannins after several months of beer storage at room temperature.

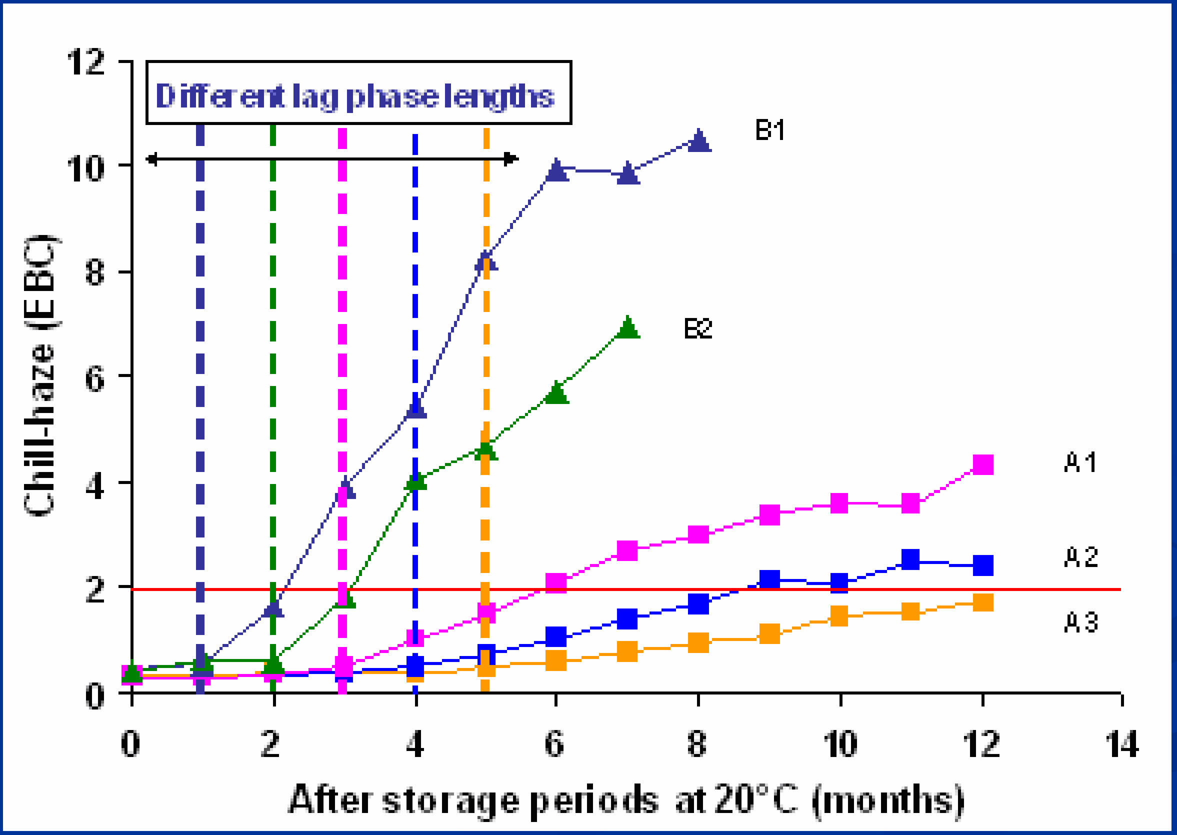
These tannins form complex irreversible bonds with proteins

These bonds do not break down completely when beer is heated up and result in a permanent haze.

Colloidal Stability

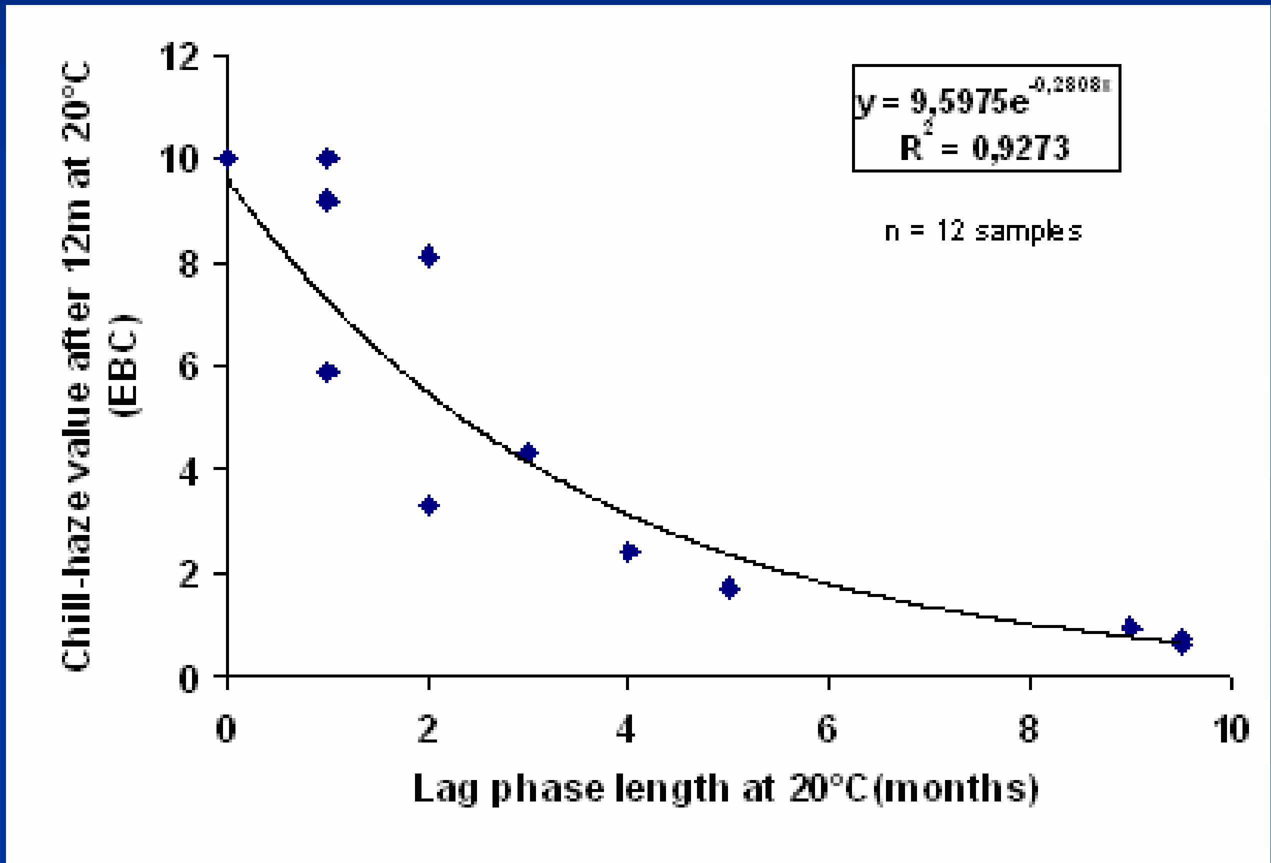
5. Follow-up of Colloidal Stability

It seems that the longer the lag phase, the lower the final chill-haze value.



Colloidal Stability

5. Follow-up of Colloidal Stability



Colloidal Stability

5. Follow-up of Colloidal Stability

It makes no difference for the consumer if the beer records 3 or 5 °EBC

At this level, for him, it is hazy

So what is important is the period of time during which the beer remains sufficiently clear or bright

→ So the longer the latency phase,

the better the colloidal stability of the beer will be

The best colloidal stabilisation treatment will be the one which stretches the lag phase close to the technical shelf-life of the beer.

The colloidal stability corresponds to the period of time during which the beer is still acceptable; so the lag phase period + some tolerance.

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Colloidal Stability

6. Measurement of Colloidal Stability

Methods available to “measure” colloidal stability can be divided into 3 groups:

6.1. Actual Colloidal Stability Follow-up

6.2. The Predictive Forcing Tests

6.3. The Indicative Tests

Colloidal Stability

6. Measurement of Colloidal Stability

6.1. Actual Colloidal Stability Follow-up

keeping the beer at ambient temperature (usually 20 +/-1°C)
measuring chill-haze each month until the end of its stated shelf-life or best before date

The complete curve of chill-haze formation is determined

The length of the most important phase, the lag phase is measured

Lag phase: Defined as the time (weeks or months) before chill haze value reaches a specified value

For instance: 2°EBC for lagers
5°EBC for brown beers (depending on beer colour)

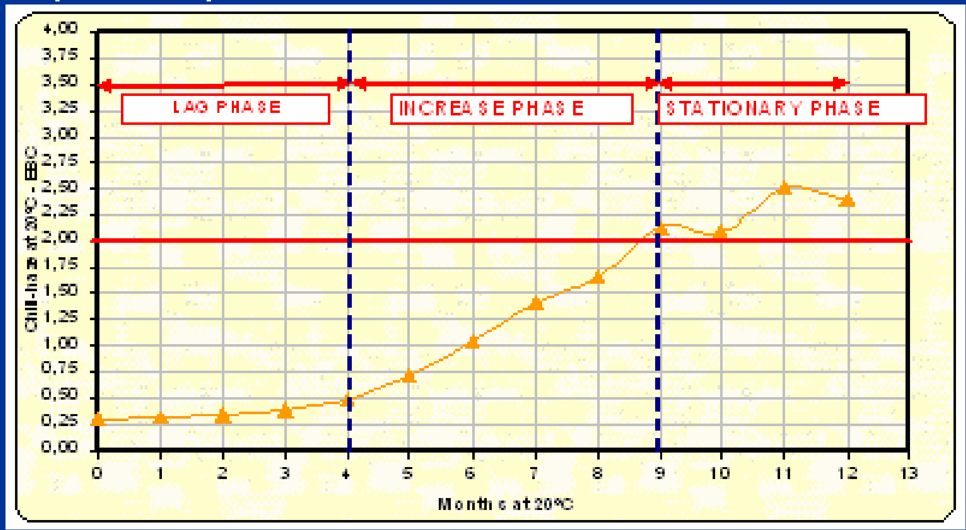
Colloidal Stability

6. Measurement of Colloidal Stability

6.1. Actual Colloidal Stability Follow-up

Advantage

Closer to real life situation
Measures the most important parameter (lag-phase)
Allows to calibrate quicker predictive tests



Disadvantage

Real life can be very different (transport, temperature)
Time before getting the results (but not time consuming)

Colloidal Stability

6. Measurement of Colloidal Stability

6.2. The Predictive Forcing Tests

Objective:

Assess quickly the colloidal stability

How?:

Accelerating beer ageing, particularly haze formation, by subjecting the packaged beer to storage at elevated temperature for a shorter time: months at 37°C, days at 50, 60°C, …

Colloidal Stability

6. Measurement of Colloidal Stability

6.2. The Predictive Forcing Tests

2 possibilities:

Follow-up of chill haze development and determination of the lag phase (spec on Time)

Measurement of chill haze value after several days (2, 3, 6, …) (spec on Haze)

The measured chill-haze or lag phase should be calibrated to the one found in the same beer after a prolonged storage at room temperature (related to real shelf life measurement).

Generally, each lab has its own preferred method, if not his own one…

Colloidal Stability

6. Measurement of Colloidal Stability

6.2. The Predictive Forcing Tests

Results expressed as a chill haze value

Applied FT	Nbr of days @ x °C	Temperature (°C)	+ 24h @ 0°C	Initial Chill-haze (0°C) in fresh beer
Analytica-EBC	**2**	**60**	yes	**Yes**
ASBC	**7**	**40/50/60**	yes	**Yes**
IOB**	**1**	**40/60 repeated 4x → 5 readings**	yes	**No (measure after 24h @ 20°C)**

**Alternate cycles of heating & cooling

Colloidal Stability

6. Measurement of Colloidal Stability

6.2. The Predictive Forcing Tests

Results expressed as a period of time

Applied FT	**Nbr of days @ x °C**	**Temperature (°C)**	**+ 24h @ 0°C**	Initial Chill-haze (0°C) in fresh beer
MEBAK	x	**60 → up to 2° EBC reached (measured each day)**	**yes**	**Yes**
Others (Leemans, Dublin 2003)	x	**37°C → up to 10 weeks depending on expected shelf life (measured each week)**	**yes**	**Yes**

Colloidal Stability

6. Measurement of Colloidal Stability

6.2. The Predictive Forcing Tests

Critical review

General

- Measurement of Chill Haze at 0°C?

  Why not at consumption T°C (+/- 5°C) , not sure the difference is negligible

- Which spec at the start in BBT?

  0,4° to 0,8 °EBC can make a big cost difference

- Measurement of a 90° angle, why not 25° angle?

  Overestimation of the "invisible" haze due for instance to the presence of glycogen, more critical for BBT specs than for colloidal specs

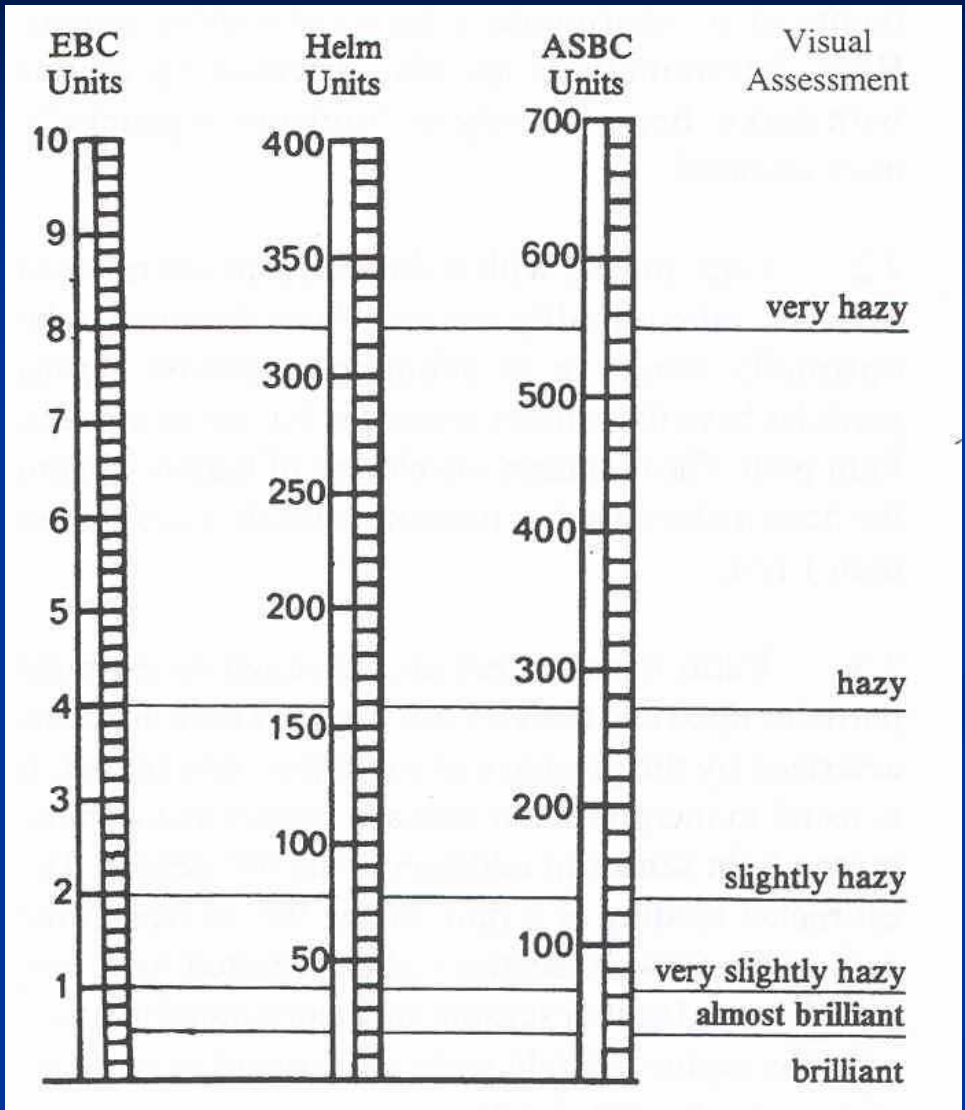
- General agreement for a limit at 2°EBC?

  Haze is easily noticeable at this level…

  Why this big difference (0,4 to 2 °EBC)?

  Why not recording time when significantly deviated from the initial value?

Colloidal Stability



This Figure gives qualitative statements for visual assessment of hazes in beer, compared to instrumental readings.

Colloidal Stability

6. Measurement of Colloidal Stability

6.2. The Predictive Forcing Tests

Critical review

Lagers	*Forced CH 3 days/60°C (EBC)*	*Forced CH 6 days/60°C (EBC)*	*CH end shelf-life (EBC)*
A	1.80	2.84	3.32
B	0.94	3.69	3.01
C	0.54	0.51	0.95
D	0.43	0.41	0.62
E	7.90	9.60	8.21
F	0.41	1.96	3.30
G	0.47	1.83	2.40
H	0.89	2.81	4.33
I	0.51	6.12	5.72
J	**2.59**	10.0	11.5

Reliability of the forcing tests of 3 and 6 days at 60°C for the prediction of beer colloidal stability during shelf-life (Leemans *et al*, EBC Dublin, 2003)

Colloidal Stability

6. Measurement of Colloidal Stability

6.2. The Predictive Forcing Tests

Critical review

Methods that record a haze value

EBC: *2 days: too short, do not discriminate between beers, measurement performed is still often in the lag phase; underestimation of the final chill haze value, overstimation of the colloidal stability. However, EBC specifies that other define conditions can be adopted by each brewer to suit each type of beer. 6 days is closer to the reality, 7 days is better for a one year TSL. You can also play on the Haze spec value.*

IOB: *Does it mimic correctly the real life? Difficult interpretation, should be calibrated against storage follow-up*

ASBC: *Period of time should be sufficient to be out of the lag phase, on the other hand, haze development could already be in the stationary phase*

60°C can be too high for some glass bottle resistance

Colloidal Stability

6. Measurement of Colloidal Stability

6.2. The Predictive Forcing Tests

Critical review

Methods that record a period of time

They need a calibration

Weekly follow-up at 37°C: *Timing before getting the results (but which are the projects that last less than 10 weeks.. ;-).*

--> R&D tool to be used when process performances have to be evaluated:

change in the raw material composition
modification of a process step which could affect beer colloidal stability
optimisation of the colloidal stabilisation treatment currently in use
evaluation of a new stabilizing agent
when beer's shelf-life has to be modified
when the real beer's shelf-life is unknown
when a new beer is developed and its real shelf-life is unknown

MEBAK style method for quality control as routine control tool

Colloidal Stability

6. Measurement of Colloidal Stability

6.2. The Predictive Forcing Tests

Critical review

Spec: depending on desired shelf-life

some clue: if period of time

Lag Phase Length at 20°C (in months)
= 1.2 x Lag Phase Length determined at 37°C (in weeks)
= x lag Phase at 50 or 60°C (in days)

some clue: if chill haze value, but not advised here…

Labelled Shelf-Life of Lager	Maximum Gray Limit of Chill-haze after Forcing Test
3 months	2.5 EBC
6 months	2.0 EBC
12 months	1.5 EBC

Colloidal Stability

6. Measurement of Colloidal Stability

6.3. The Indicative Tests

These tests are measurements of indicators, usually related to the protein or polyphenol contents of fresh beer

Pre- stabilisation measurements to evaluate process impact and/or deciding on the treatment level

Pre- or post-packaged measurement to monitor the effectiveness of stabilisation treatments

Consequently they enable to estimate the probable rate of haze production during beer real shelf-life.

Not an exhaustive review…

Colloidal Stability

6. Measurement of Colloidal Stability

6.3. The Indicative Tests

Polyphenols: Total Polyphenols

Measurement of all polyphenol species using the EBC method

Does not measure specifically polyphenols involved in the beer colloidal stability

Colloidal Stability

6. Measurement of Colloidal Stability

6.3. The Indicative Tests

Polyphenols: Flavanoids

Simple polyphenols (or flavanoids), such as (+)-catechin, (-)-epicatechin, procyanidin B3 and prodelphinidin B3, can be determined by

- Direct-injection High Performance Liquid Chromatography using both UV-absorbance and Electrochemical Detection (HPLC-ED) systems *(McMurrough & O'Rourke, 1997 - McMurrough, Madigan, D. & Kelly, 1997)*

- Spectrophotometry: colorimetric reaction,

Reference: - European Brewery Convention – Analytica-EBC: Beer-9.12

Delcour

Colloidal Stability

6. Measurement of Colloidal Stability

6.3. The Indicative Tests

Polyphenols: Tannoids

This is a nephelometric titration of soluble PVP (polyvinyl pyrollidone) solution *(O'Rourke, April 2002).*

PVP has a similar structure to a protein molecule and easily forms an insoluble precipitate with polyphenols, particularly with tannoids (medium size molecular weight polyphenols), which are known to be haze active.

When PVP is titrated in beer a haze is formed. These latter increases to a maximum and then decreases by a dilution effect as PVP addition continues.
The peak value gives a measure of the tannoids which can be correlated with chill-stability.

Importance of the shape of the curve (same max but narrow or large shape)?

Colloidal Stability

6. Measurement of Colloidal Stability

6.3. The Indicative Tests

Polyphenols: Tannoids

The tannoids tare defined as the fraction of polyphenolic compounds which can be precipitated by polyvinylpyrrolidon (PVP). They include the low and medium molecular polyphenols, the polymers of catechin and anthocyanogens. Tannoids come from the malt and hops. Although present at relatively low concentration, they play an important part in the colloidal and taste stability of beers. On the one hand they can precipitate the sensitive proteins of the wort and beer, on the other hand they act as protective agents. The tannoid content of beer, wort, barley, malt and hop extracts can be determined by precipitation with PVP. This polymer which can be considered as a structural analog of proteins binds with tannoids by hydrogen bonding to give an insoluble complex which leads to the formation of a haze. On continous injection of a solution of PVP into the sample a haze develops until all the tannoids are bound. Beyond this point an excess of PVP brings about a progressive dissolution of the haze.
The amount of PVP necessary to reach the maximum is proportional to the tannoid content of the sample. The Tannometer measures the amount of haze formed versus the amount of PVP injected and expresses the result in mg PVP/litre. The curve and the result are printed by the inbuilt plotter.
(literature: Der Begriff Tannoide, Monatsschrift für Brauwissenschaft, No. 7/8 93, page 263-279)

Colloidal Stability

6. Measurement of Colloidal Stability

6.3. The Indicative Tests

Proteins: Sensitive Proteins

This is a nephelometric titration of a tannic acid solution

Tannic acid is a “super” polyphenol which easily forms insoluble complexes with proteins.

A given amount of tannic acid is titrated against a given volume of beer to give haze measurement which relates to its stability.

Reference: - European Brewery Convention – Analytica-EBC: Beer-9.40

Colloidal Stability

6. Measurement of Colloidal Stability

6.3. The Indicative Tests

Saturated Ammonium Sulphate Precipitation Limit (SASPL) Test

The estimation of beer haze stability by determining the amount (ml) of saturated ammonium sulphate [$(NH_4)_2 SO_4$] solution needed to cause precipitation of haze material.

The more salt is needed to bring out protein, the less such precipitable protein is in solution. Consequently, the greater beer colloidal stability is expected.

Reference: - Institute of Brewing (IOB) – Methods of Analysis: Beer-9.39

Colloidal Stability

6. Measurement of Colloidal Stability

6.3. The Indicative Tests

Alcohol Chill - Haze

This is the alcohol chilling test from *Chapon (J. Inst. Brew., 1993)*

The haze stability is estimated by measuring the haze increase after the addition of alcohol and chilling.
The addition of alcohol, to 6% (vol/vol), in beer decreases the solubility of the protein-polyphenol complexes and increases the chill-haze.
The beer temperature is lowered to -8°C and the chill-haze is forced out within a total rest time of 40 minutes.

The lower the chill-haze the greater the colloidal stability. The chill-haze estimated is principally the protein fraction.

Reference: - Institute of Brewing (IOB) – Methods of Analysis: Beer-9.38

Colloidal Stability

6. Measurement of Colloidal Stability

6.3. The Indicative Tests

Some of these tests are automated

Tannometer is a microprocessor-controlled device that measures the turbidity of a liquid sample in the range of 0 to 300 EBC in units of 0.01 EBC.
It works on transmitted light at 510 nm and it can control the sample temperature including cooling it down to -8°C *(Chapon, L., 1993; McMurrough, I., et al, ASBC, 1997)*.

The Tannometer can produce automated results for:

- Tannoid content
- Sensitive Proteins
- Alcohol Chill-haze
- SASPL

This instrument is manufactured by Pfeuffer GmBH (Germany),
Another tool, PT standard is also available

Colloidal Stability

6. Measurement of Colloidal Stability

6.3. The Indicative Tests

A critical review (Leemans, EBC Dublin 2003)

For some of these parameters, correlations between the length of the lag phase during storage at room temperature and the corresponding tannoids, sensitive proteins and total polyphenols in fresh beer were studied.

Colloidal Stability

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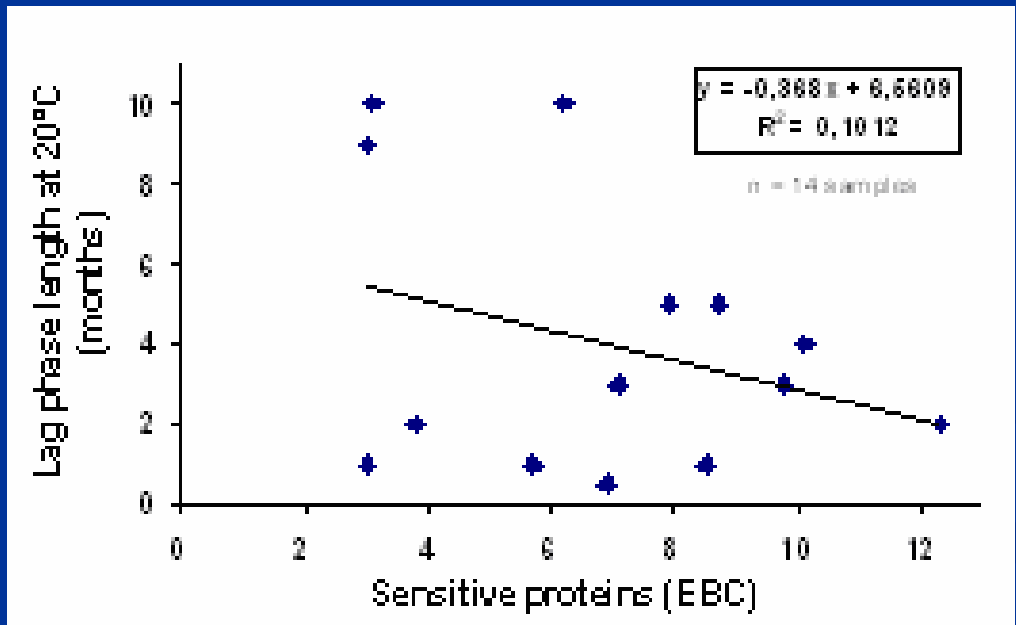
6.3. The Indicative Tests

Lag phase versus sensitive proteins



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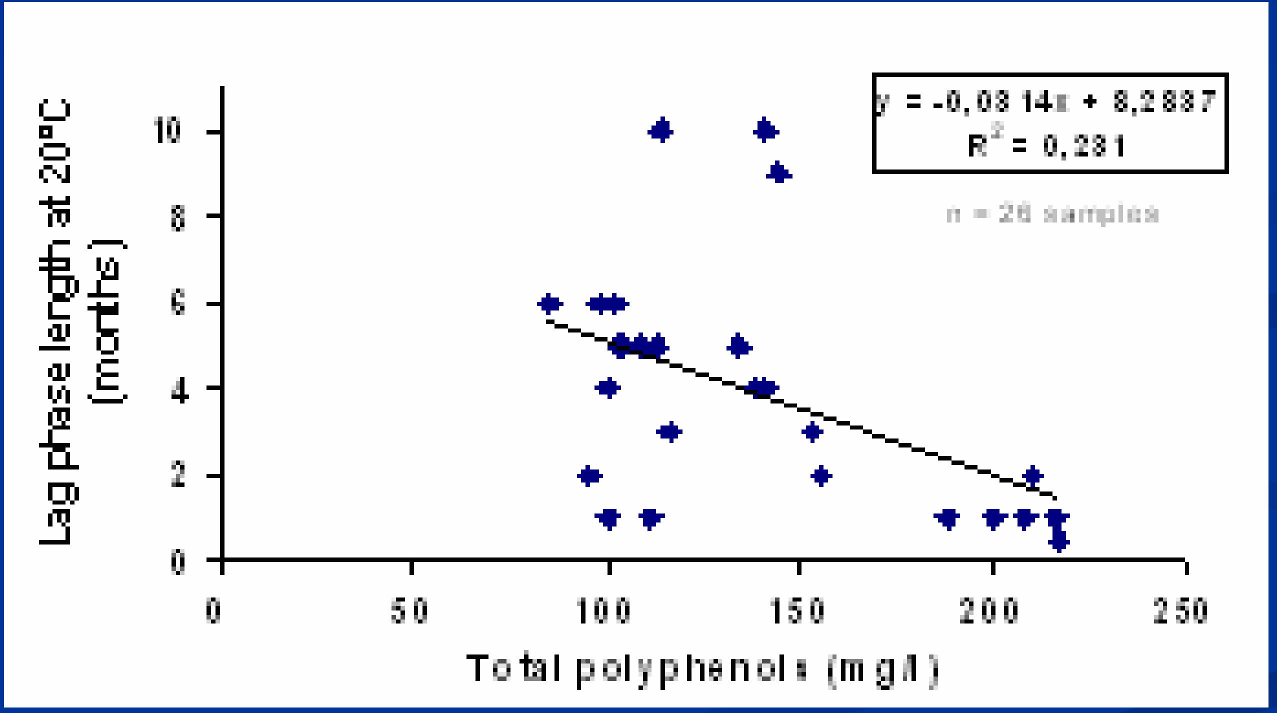
Lag phase versus Total Polyphenols



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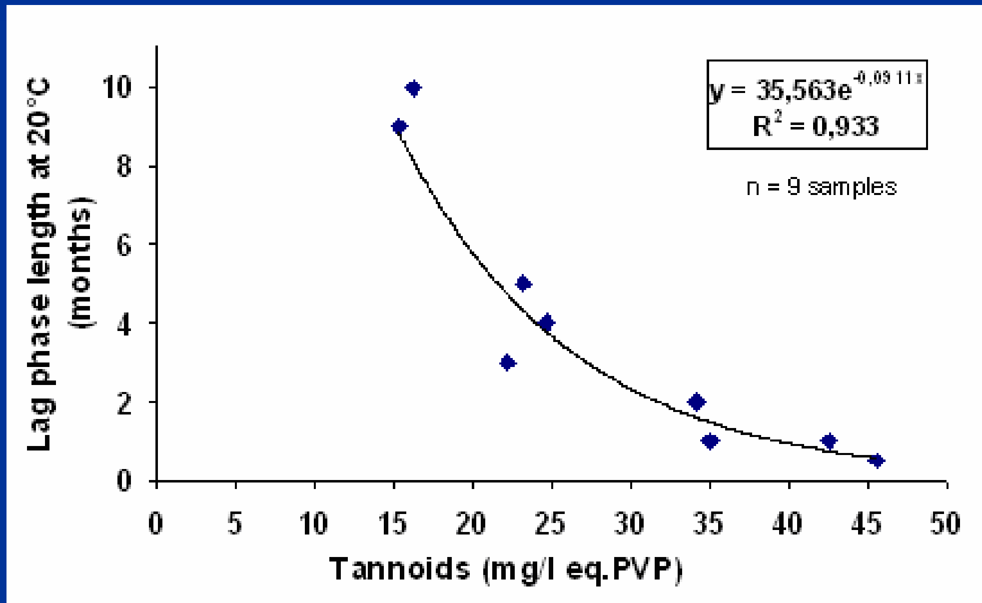
Lag phase versus Tannoids



Colloidal Stability

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6.3. The Indicative Tests

An exponential correlation between the lag phase and the tannoids level of fresh beer was found. According to these results, it seems that when tannoids concentration is higher than 25 mg/l, the lag phase would not exceed 1 to 2 months.

No correlation with sensitive proteins or total polyphenols was found.

This points out that sensitive proteins would be less critical for the colloidal stability than tannoids

This points out that it is not the level of total polyphenols but rather the tannoids fraction in the fresh beer that is important for colloidal stability

This is partly confirmed by the experience that PVPP leads to longer colloidal stability than Silicagel.

The analysis of tannoids could therefore represent a better and easy way to help predicting the colloidal stability.

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7. Conclusions

- Need for a “cost” effective management of colloidal stability

- Defining the goal to be achieved is a prerequisite to any measurement (CSL-TSL)

- Choosing the right method and specs is mandatory

- Colloidal Stability is better estimated by the measurement of the lag phase of CH development

- Depending on the TSL to be achieved, routine control is better performed by following CH at 60°C during 6 to 7 days

- Tannoids seem to be a good indicator of beer colloidal stability

- Still need for quicker methods, and more accurate predictive indicators

Colloidal Stability

Some Key References

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