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PHYSICAL CHEMICAL STUDIES REGARDING CIDER STABILITY STORED UNDER AMBIENT CONDITIONS

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Abstract

Multiple cider samples obtained from United Kingdom market were analyzed for their parameters before opening and after 24/48 hour storage on ambient conditions.
We aimed to study the basic parameters such as color, turbidity and sulfites contained in different type of ciders.
The first step to achieve our general aim was to select from the diversity of cider market and to assess the parameters used to conduct this study.
The physical and chemical studies allow us to evaluate not only the diversity of the samples but also the how they behave in different ambient conditions.
This information is essential for the selection of cider consumption and for better understanding of the selected parameters for further beverage studies, especially apple and pear made ones.
Deviant variation from standard was found in more than half from twenty five samples analyzed. The results of previous studies showed that the defective storage conditions of cider might affect not just the bacteria contaminations, but that include also defective physical and chemical composition.
The experiments exposed that weaknesses of the opened product that might become a quality, or more than that, a safety issue through its flawed parameters when these common conditions exist.

Key words**:** *cider, color, sulfites, physical, turbidity.*

INTRODUCTION

The formative taste suggestion is almost regularly reflected through the eyes. But there are various elements to a food article, optical aspect and color are the most distinct provocation. In attributing anticipation to responsive facts, humans are observational creatures.
Set incertitude or clash in the perceptive suggestion we get, we facilitate to depend most on what we observe.
Furthermore we might appraise that the feel of smell and ingest should overcome in conditions of food reflection, it is alerting, that a perceivable deviation could also be commanding. This is due to primal choices of foods and beverages frequent trust on receptions.
Our acuity to detect regular everyday particularities is surely faulty, so appearance may sometimes be an increased trustworthy inception of guidance. Visible briefing of foods gives effectual clues as to other sensitive ranges, emerging wishes about what we are about to consume.
On the other hand what happens is that the sulfites reduce the development of spoilage yeasts and bacteria, however allowing the looked-for fermenting yeasts (like Saccharomyces cer. or others) to grow and to lead the transformation to alcohol.
Fluctuating the color of cider can be caused by some metals. Darkening may give escalation to the reactions between metal (industrial tools or machines) and tannin of cider, helped by atmospheric oxygen. Such changes occur only after unscrewing cider's bottle and contact with air oxygen.
There are fears about the addition of sulfites as a secondary factor beyond measure resulting from the use there of in phases bottling but also because some people are allergic and can be considered a health hazard.
Haziness can be determined by microbial activity, especially in those cases when the fermentation went wrong.
But even after opening the bottles spoilage may occur, especially if the bottles are not stored at proper temperature. Massive infestation and

alteration of ciders can be observed many times with naked eye.
Sometimes, even the product is filtered, the cloudiness can be induced by pectin which is not soluble in alcohol.
An important factor is the origin of cider, the appearance of fresh ciders made from raw apple/pear juice that was not put throughout a filtration process to remove granular elements of pulp or deposit.
The second origin is the pear/apple juice which is already filtered to eliminate bigger elements and is pasteurized to be preserved for a while till will be used in the fermenting process.
Using different analyzers and methods helped to get and collect accurate data regarding cider samples and provided the results of indicators that impacted product physical and chemical changes.
Considering the diversity of the cider on the market, variations of their properties we selected couple of parameters for the study.
The absorption spectrum is influenced by strength: such as the highest values can be found at higher concentrations. Other factors that might influence the absorption are temperature, pH etc.
Chemical stabilizers or inhibitors like sulfites and addition of artificial colors, flavors are frequently inputted in pear/apple juices to prolong their shelf life and make them look have natural properties. Nevertheless these days market demand for fresh and harmless nutrients deprived of artificial additives and presservatives conduct to rise the awareness in consuming of nutrient additives from natural bases.

MATERIALS AND METHODS

Equipment determining total sulfites:
1. Distillation apparatus scrubber
A. 250 ml round bottom flask,
B. Claisen adapter
C. Pasteur pipette with a rubber stopper
D. Graham condenser 300 mm
E. adapter Connection Vacuum adapter
F. 50 ml flask seem G. H. Adapter Reduction
I. adapter connection
J. socket adapter
2. burette 10 ml / 100 ml; 3. vacuum cleaner source/water; 4. 5, 10 and 20 ml volumetric pipette; 5. tube connection to water and vacuum

Photo no.1 - Sulfites determination

Samples must be cold when they are used for analysis to reduce degradation of labile sulfites. In the same time samples should be used in the manner to minimize disclosure to air and heat. After titration the sulfite content, expressed in µg SO2/g food (ppm), as follows:

SO_2 (ppm) = (32.03)(V_{Corr})(N)(1000) / W_{Sample}, Where

32.03 = milliequivalent weight of SO_2
N = Normality of NaOH titrant.
V_{Corr} = volume (mL) of NaOH of normality N required to reach end point, minus the NaOH volume required to titrate the reagent blank.
1000 = factor to convert milliequivalents to microequivalents.
W_{Sample} = Sample weight, in grams.

Formula no.1 - Titration Quantitation (source U.S Department of Agriculture Food Safety and Inspection Service 2016, February)

ALCOLYZER ANALYZING SYSTEM – ALCOHOL DETERMINATION

It is a high precision measurement machine and it was used for the research to get accurate indicators results. It has a pack of common density, concentration tables stored in the software, and new substances are easily introduced as a table or a polynomial. It determinates the sugar content (<0.01 Brix, g/L), alcohol content (<0.01 %v/v, <0.02 Proof); Determination of extract content (Plato, Balling) Quality control of soft drinks (< 0.01 Brix)

Photo no. 2 - Alcolyzer Analyzing System

Sample analysis is performed completely in one measurement cycle that establishes alcohol content, density, sugar content, degree of fermentation, calories.
Measurement of turbidity, color and pH values were delivered in the same time.
Level of measurement accuracy is up to an alcohol content of 12% v /v (but values are revealed up to 30%v /v). It has stoppage due to removal-cleaning between samples and can be adjusted and calibrated really simple (with water and water-alcohol solution). Time for each sample evaluation is four minutes (Anton Paar GmbH, 2016).

EPA, TURBIDIMETER 2100AN 115VAC
Hach 2100 series laboratory turbidimeter is planned to provide the best accuracy and sensitivity in any application.
The 2100AN is prepared with a stable halogen-filled tungsten filament lamp and is ideal for testing higher ranges of turbidity up to 10,000 NTU (Hach Company, 2013).
Additional absorbance, transmittance, and color detection modes make the 2100AN one of the most flexible of any bench to turbidimeter.
It can be used with exchangeable color filters and optional adapters for cell to measure smaller sample volumes. It had built-in printer which provides a data record of calibration and measurement. Turbidity can be interpreted as a measure of the relative clarity of water.

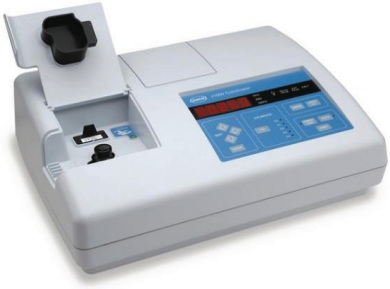


Photo no. 3 – Haze determination

SPECTROPHOTOMETER CECIL
The benefits of using for the research of this spectrophotometer were due8n M optical bandwidth, automatic lamp change, the light reduced for greater precision calibration test automatically at startup.

In order to perform proficiently, thin films must have the appropriate thickness.
Film thickness is often measured, both during and after thin-film coating.
Spectral reflectance measures the amount of light reflected from a thin film over a range of wavelengths, with the incident light at a known angle to the sample surface.
Ellipsometry is similar, except that it measures reflectance at non-normal incidence and at two different polarizations.
Thickness of a few microns can be measured by ultra-violet/visible spectrophotometers, using a specular reflectance accessory.
This method was described by Cori and Wimpfheimer (1999) and may be performed on a sample area as small as 2 mm diameter.

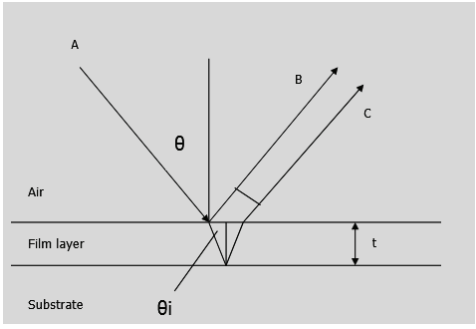


Photo no. 5 – Reflection of light in a thin layer

An incident beam of light, A, hits the surface of the film at an angle θ, from the normal.
Part of the beam is reflected at the front surface as beam B.
Whilst some of the remainder is refracted by the film layer, travels the film, is reflected at the interface of film layer and substrate, and then finally emerges from the film as beam C.
Beams B and C arrive at the detector of the spectrophotometer.
The combined intensity of beams B and C, at any given wavelength, is a function of the phase variance, if any, between the two at that given wavelength.
If the wavelength of the incident beam is continuously varied, as when scanning a spectrum, the result will be a continuous series of maxima at wavelengths, where constructive interference (reinforcement) occurs and minima, at wavelengths where destructive interference (cancellation) occurs.

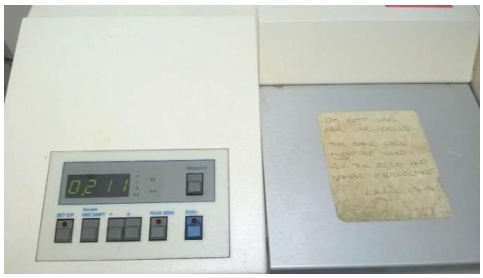


Photo no.4 - CT-310 Chroma Meter Konica

The Chroma Meter CT-310 is a small-sample tri-stimulus colorimeter for measuring the transmittance and color of fluids (Konica Minolta specifications, 2016).
It has three different sample cells (with optical path lengths of 2mm, 10mm, and 20mm) and two sensitivity settings enable measurements of fluids with a wide range of densities.
Other versatile functions include data memory, printout statistical calculations, and data communication. It minimizes the differences in readings between multiple instruments.
Light from the pulsed xenon arc lamp is thoroughly diffused by passing through two diffusion plates and into a mixing chamber, and then enters two optical fiber cables.
Optical fiber cable 1 transmits the light to the enlighten in gend face to illuminate the specimen for measurement; optical fiber cable two transmits light directly to sensors for monitoring the light double-beam feedback system.

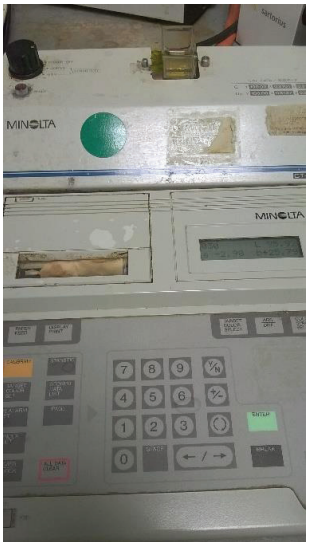


Photo no. 6 – a* and b* determination

METHOD USED

Nowadays there are many methods for the determination of alcoholic beverages, among them the turbidity measurement is the most important because it is a simple and very important indicator in changing the properties of a drink, regardless of technology or final product stage.
Sudden change in turbidity may indicate the source of contamination (biological, chemical, etc.) or can indicate a problem in the manufacturing process.
The aim of this study was to determine differences and to compare the results of different types of cider and how they behave after they are no longer airtight.
A complete study of the composition of the apple juice, fermentation or final product (cider) includes determining data other than those derived exclusively from chemical analysis, buta study of the characteristics of debt, including sensory analysis, all executed by certain rules precisely.
These studies will contribute data to guide students, researchers for future experiments and analyzes in this field.

TURBIDITY

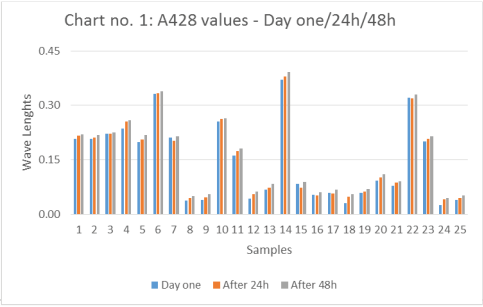
Preparation of sample: cider sample must be degassed prior to testing, otherwise they influence of the bubbles of gas, and will result in higher yields. Decant part of cider in a beaker and mix until all the gas has been released. You can also use ultrasonic waves degassing device. Turbidity measurement parameters used or this study, but general industry and wine, beer, cider are:
EBC - "European Brewery Convention".
NTU - "Nephelometric Turbidity Unit".

Measurement of absorption spectrum- the color Togauge Konicahave used the values and CIEL*bre presenting a* b*(CIELAB) color space is specified by: French international de l'Eclairage Commission, hence the initials.
It defines all the colors perceptible to the human eye and was made to serve as a device independent model to be used as a reference.
The three coordinates of CIELAB represent the brightness of the color (L* = 0 basic black and L* = 100 indicating white diffuse), and its

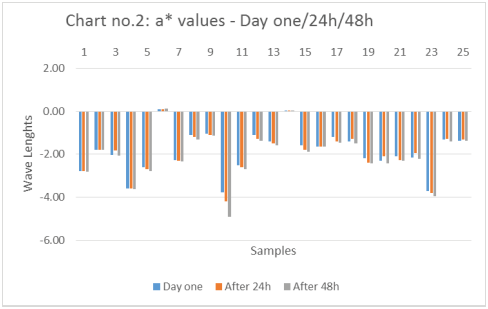
positions between red/magenta and green (a*, negative values indicate green and positive indicated purple) and positions between yellow and blue (b*, negative values indicate blue and positive indicating yellow). For Cecil measuring device was used to measure color related parameters.
Values were expressed in nanometers using wavelength spectrum from 280 to 500 nm, depending on the sample or specification of each product. Before the samples were used for analysis the machine analysis (Cecil) was calibrated according to the wave lengths standard specifications.

RESULTS AND DISCUSSIONS

Experiments in this chapter focused on the differences between the parameters of bottled cider, which occurred after opening and keeping both at ambient room temperature for24 – 48 hours.
They were covered by the sulfur differences and different properties which may affect flavor, aroma, appearance of the product (especially color etc.). The samples used for this study cider were taken from local markets bottled and cans too.
Other further studies will try to reveal pH, acidity, ABV trend for the same types of ciders.
In the chart number one are represented the results of twenty-five assessed values cider samples color spectrum length A428.
These samples were taken in the first working lab for measuring parameters in one day, and for subsequent determinations, respectively 24hours, 48 hours were held at an ambient temperature of 19°Celsius.

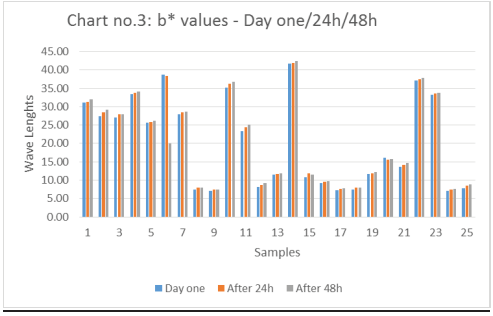


On the day one a twenty samples had values more than 0.04 (on the A428 measurement scale).
A number of thirteen cider samples, representing more than half of total samples tested, had A428 values increased after 24 hour from the opening moment (as the average for total samples was 0.1514).
Go after 48 hour storage a number of twelve sample had a significant increase of the value with an average of 0.256 (A428 wave length).

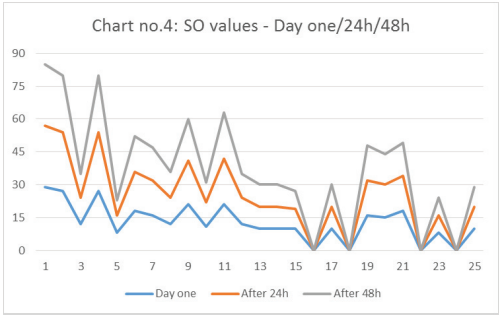


In the second chart are represented the results of the same ciders, for a different parameter (a* values).

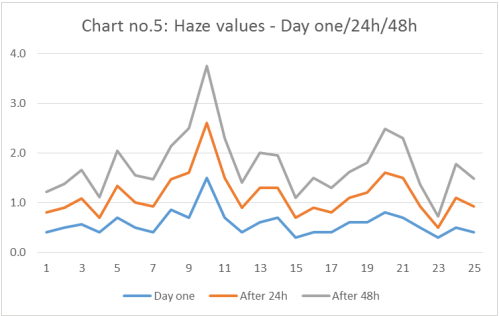
A number of thirteen sample kept the increasing trend from day one to the end (48 hours), with just two positive values, and eleven sample with values between -4.2 and -1.2 after twelve hours of storage.
Six samples (24h measurement) were under day one values with the average values of -1.63333.
After 48 hours just three samples kept the same value like day one result and twenty two samples had increased values with an average of -2.08227.



For the b* measurements almost all the results increased, the highest value was a peak of 11.5 representing a 6.48% growth from day one value. Two sample values have increased from day one to 24/48 hours, but the values were equal from 24 to 48 hours. Between all samples the lowest value is 7.33, representing a dry cider and the highest is 42.06 representing a cloudy cider.



Sulfites values shown on y axis (measured in ppm).
The samples sixteen and twenty-two values were zero since they are canned cider.
In this case is not allowed to use sulfites, which are considered corrosive agents, they might be dangerous through the explosion of the aluminum cans.
There are four results above the value of twenty, due to the addition of sulfites in the fermentation process to help specialy east to end the process, but also to inhibit other microbial forms that may harm the process itself (by altering the taste, smell, appearance, wrong fermentation etc.).
Fourteen samples went from day one to 48 hours of storage values with a small decrease of the amount of sulfites since they were wasted into physical – biological process activities of the cider.
The average of the decrease is 1.428571 (for fourteen samples).
Other ten samples kept a constant value from day one to the end of the experiments.
Turbidity values shown on y axis (measured in Nephelometric Turbidity Unit– NTU).
The haze measurements revealed that a number of seven samples had an average percentage decrease of 22% from initial value of haze from day one to 24 hours and an average 16% from day one to 48 hours, which reveals a small increase of the value from 24 hours to 48 hours measurement based on cider's ongoing biological activity.



CONCLUSIONS

When the temperature is close to refrigeration, the optimum pH is kept, even if the bottle or the can has been opened to about 24hours.
Sulfur dioxide value depends critically on the pH apple juice even is used only in small quantities.
Cider apple/pear juice, when is pressed, it covers a large number of yeasts, molds and some bacteria that are not useful, and only very few of those useful.
Some cider manufacturers in the UK have set a maximum limit of total SO210mg/L, which was challenging to maintain product within these limits.
One reason was the occurrence of smell-urine mouse taste, especially in the mixing phase (for low levels of sulfites).
Although not everybody can spot this taste, there is a risk that those who drink this taste not ably in cans.
For cans the total allowable sulfites that might come into contact with the protection lacquer is around 25 ppm, but not more than this value.
Otherwise the metal of which it is made can may be corroded if the can lacquer cannot protect the result antaction of hydrogen sulphide generated inside.
Some brands of bottled cider in cans are often specially fermented in the total absence of sulfites, exactly what our results showed throughout the experiments.
However, in the study were detected several types of cider, especially cloudy ones (unfiltered type, or traditional recipe), which had sulfites levels over 25 ppm.

This is not necessarily a factor that might increase the risk of corrosion of aluminum cans because some manufacturers choose a double or triple degree of varnishing depending on the action of bottled liquid.
Sulfites levels in the study were determined on the basis of laboratory research and most are normal with standards, with a much lower chance of taste and flavors to the finished product.
However for higher values, this is somewhat normal for products classic unfiltered, cloudy etc. Also, several acts yeast during the fermentation, naturally reducing the content of sulfur dioxide.
Adding the sulfites, after fermentation it is also sometimes used after the fermentation has been completed, products in the shelves, storage and packaging.
This is partly antimicrobial action but also to act as an antioxidant. Or rather, destroy oxidation initiators such as hydrogen peroxideoraldehydes, not leading to unwanted flavors.
In these cases is usually added a fixed quantity of 50 ppm each time (up to the legal limit of 200 ppm for all additions summed together), to achieve a residual result of 30 ppm SO free for the day.
This is due to the fact that the antioxidant properties of sulfur dioxide are not affected by pH.

THE COLOR AND TURBIDITY

The difference is noticeable between a cloudy and a pasteurized cider color. Enzymes change hue apple cider in a dark brown color which is inactivated by heat. For direct apple juice pasteurizing when is obtained, this lead to the color change.
But after pasteurization the process of recovering color, clarity and turbidity makes changes to the product.
The values resulting from the study confirmed that both color values a* and b*, A 428 and verified turbidity cider were closer to the market demands, no significant changes were reported.
We noticed some increased values just between day one and 48 hours storage, due to ambient temperature and oxygen intake that leaded to microbiological and biochemical changes of color turbidity, higher than if the products were kept cold and unopened.
Relationships between color and the cider types were observed. There was an undesirable but not essential relationship between color and cloudiness results as well a helpful but not significant relationship was observed between color and sulfites results. Pear and apple ciders change their color into darken cue because natural enzymes react with oxygen. This might be an indicator because it shows the aroma of the cider in the end.
Further research about cider will try to connect the information gathered until now with experiments regarding pH, acidity. The correlation can be made also with the microbial activity of the cider after pasteurization and how different conditions can affect its own characteristics like ABV, sugar content and many others.
Other researchers observed Significant changes in pH, Brix and viscosity only for cider microfiltered with 0.45 μm (Evonne Lau and col., May 2012). They suggested that same type of particles were present in their cider samples and just the concentration was different for different types of ciders.
Their conclusions were the composition of apples is subject to inconsistency due to season and raw material variations. This might bring unpredictability in the composition and turbidity of pear/apple cider and as a result in the effect of the microfiltration process.
The results of this work indicate that color, turbidity and sulfites content are deeply correlated with the process of various raw materials, additives and with the ciders type. More than that ambient conditions of the unopened bottles accelerated all the biochemical processes.

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