Poster session: **Coffee**

Poster Coffee

Coffee Poster 01

UHPLC-MS/MS method for rapid quantification of chlorogenic acids in roasted coffee

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Text Chlorogenic acids (CGAs) are a large class of esters formed between quinic acid and hydroxycinnamic acids. They are present in coffee as a complex mixture of positional and geometric isomers, where caffeoylquinic acids (CQA) are the most abundant, followed by dicaffeoylquinic acids (diCQA), feruloylquinic acids (FQA) and p-coumaroylquinic acids (p-CoQA)[1].
There's a growing interest for complete and unambiguous identification of this class of compounds that take part in the generation of color, flavor and aroma of coffee during roasting and that in the last decades were also object of numerous studies to evaluate their impact on human health. The aim of this work is to develop a new reliable and fast LC-MS/MS method for quantification of major chlorogenic acids in roasted coffee, optimizing the procedure described by the validated method[2] and following the AOAC guidelines[3]. UHPLC-ESI-MS/MS analysis was conducted on an Agilent 1290 HPLC, coupled with a Sciex Triple Quad 4500. A mixture of methanol/water was used for the ultrasound extraction of the target compounds, followed by filtration, where different filters were evaluated. Chromatographic separation of extracts was performed on a Acquity BEH C18 (Waters) column with a gradient elution of acetonitrile and aqueous formic acid (0,1% v/v); MS Electrospray Ionisation Source was operating in negative mode, acquiring in Multiple Reaction Monitoring (MRM). Operating conditions were optimized using a 5-CQA standard solutions. Specific compounds transitions were confirmed based on literature data and comparison with reference standard solutions. The developed method allows the simultaneous quantification of 12 chlorogenic acids in roasted coffee in 15 minutes and was validated in terms of specificity, linearity, concentration range, limit of detection (LOD) and limit of quantification (LOQ), precision and accuracy participating to a proficiency test on the same matrix.

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Poster session: **Coffee**

Coffee Poster 02

Effect of sugar addition on temporal dominance profiles of coffee- related sensory attributes and emotions

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Text Coffee is a popular beverage, not only due to its stimulating nature but rather because of the unique sensory properties. It is often enjoyed with various add-ins, such as sugar, which leads to the changes in the flavor profile of the product.
Therefore, the objective of the present study was to investigate the effect of sugar addition to the coffee beverages, prepared from Arabica and Robusta beans, on the temporal aspects of sensory attributes and coffee evoked emotions, selected by using the check-all-that-apply method (CATA). The samples were evaluated by 42 untrained individuals in multiple sip approach over 60 seconds.
The temporal dominance profiles of the sensory attributes and emotions were investigated by Temporal Dominance of Sensations (TDS) and Temporal Dominance of Emotions (TDE) methods, respectively.
When evaluating Arabica and Robusta coffee without sugar, the most dominant attributes during the entire testing time were "bitterness" and "astringency", both reaching the level of significance (5%). Addition of sugar showed an increase above the significance level of the dominance rate of "sweetness", shortly after each sip in both kinds of coffee.
Independent of the coffee species, the samples without sugar were associated with emotions like "disappointed" and "active". The addition of sugar resulted in a distinct decrease of the negative emotion "disappointed" and the panelists associated the samples rather with the emotion "calm" then "active". The emotion "satisfied" did not reach the level of significance at any time during the evaluation. However, it increased after every sip and correlated with the enhancement of "sweetness".
The results showed that the addition of sugar has a potential to change the perception of sensory attributes and emotions during coffee consumption.

Coffee Poster 03

Effectiveness of polyphenolic and trace element concentration in leaves of *Coffea arabica* for the chemometric discrimination of coffee leaf rust resistance

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Text Coffee (*Coffea*) is an important commodity and it involves about 500 million people from cultivation of the coffee trees to final consumption of infusions of the ground roasted coffee beans. In contrast to a considerable amount of research performed on green coffee beans, there are relatively few studies concerning the chemical constituents of coffee leaves[1].

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Chlorogenic acids, present in coffee leaves, act as antioxidants in plants and protect animals against degenerative, age-related diseases when present in the diet.
Hemileia vastatrix is a parasite, specific to coffee plants and causes coffee leaf rust – a very destructive disease. Some coffee plants have natural resistance which is mainly linked to a gene and specific host resistance response (H_2O_2 production). *Coffea arabica* is characterized by a low genetic diversity, which is reflected in its susceptibility to numerous diseases[2].
An increase in flavonoid production could be related to resistance to fungal disease, with the levels and flavonoid types being an early physiological response to rust infection.
We are of the opinion that there may be a strong correlation between various factors which render *Coffea arabica* plants susceptible to rust infection. These are (1). The concentration of trace elements (essential cofactors in the function of enzymes important in the deactivation of reactive oxygen species) such as PPO, superoxide dismutase and catalase (2). the concentration of major polyphenolic compounds.
To address this, coffee leaves harvested from Minas Gerais - Brazil (suceptible and resistant to rust) were extracted using the QUEChERS technique[3], and polyphenol and trace element content (Al, Cu, Mg, Mn, Fe, Sn, Zn) were evaluated. Principal component analysis (PCA) was then applied and we could establish the role of polyphenolic and trace element concentration in the leaves to rust infection resistance. To our knowledge this is the first study to attempt this approach.

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Coffee Poster 04

Exploiting low pressure liquid chromatographic systems in coffee analysis

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Text The development of low pressure liquid chromatographic flow systems is an emerging field of research that has reborn since the development of monolithic columns. These columns present as main advantages low back pressure and high separation capacity. These column's features make them suitable to be coupled into traditional flow systems and surpass one of the main handicaps of FIA systems: low selectivity and/or difficulty to perform multiparametric determinations.
In this communication, three case-studies will be presented that focused the determination of methylxanthines [1], niacin [2] and trigonelline [3] in coffee extracts, exploiting low pressure liquid chromatographic flow systems. All flow systems were simply comprised of a peristaltic pump, a low pressure injection valve, a 1-cm length C_{18} monolithic column, and the detection system. For methylxanthines determination, reversed phase elution and an UV-Vis detector were used. For niacin determination, the C_{18} monolithic column was previously coated with a surfactant (ion chromatography was therefore used) and a boron doped diamond electrode was used for amperometric detection. For trigonelline determination, an ion-pair reagent was added to the mobile phase to enable chromatographic separation of this compound, previously to electrochemical detection. These case-studies illustrate the several advantages of this approach, namely: (i) versatility concerning the different chromatographic separation modes that may be exploited; (ii) easiness of assembling, likewise traditional flow analysis systems; (iii) low implementation cost if compared to traditional HPLC systems; (iv) high sample rate analysis and easiness of operation.

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Coffee Poster 05

Evaluation of polyphenols and LMRP content in coffee extract with different levels of roasting and after the digestive vitro system simulation.

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Text Polyphenols are natural antioxidants, which play an important role in the human diet, contributing to the proper functioning of the body. Coffee is one of the natural products with a high content of polyphenols, especially hydroxycinnamic acids [[1]]. The concentration and profile of polyphenols are significantly modified during roasting, the content of free polyphenols is reduced due to binding with Maillard reaction products (MRP) and degradation to lactones, guaiacols and others. The state of the knowledge currently available covers only the changes of coffee polyphenols depending on the degree of roasting, but the effect of their binding to high molecular mass substances during roasting has not been characterized in terms of bioavailability and absorption [[2],[3]].
The aim of the study was to determine a profile of the polyphenols in coffee extracts from green, light and dark roasted beans and their fractions obtained by countercurrent partition chromatography regarding to the degree of esterification, before and after *in vitro* digestion and absorption.
In vitro digestion of coffee extracts was performed in the gastrointestinal tract model in order to obtain absorbed digested solutions from selected parts of the gastrointestinal tract. The solutions were analyzed by LC-ESI-MS technic. The conducted analysis showed that the extracts differed in terms of the polyphenol content and their bioavailability, which was increased after digestion, showing that the analysis of free polyphenols does not cover the whole available after digestion. The research was funded by National Center of Science (project No. UMO-2018/2019/N/NZ9/01160).

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Coffee Poster 06

Discrimination of genealogical groups of elite genotypes of *Coffea arabica* L. by the chemical composition of pulped coffees*

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Text The aim of this work was to use a model created by the partial least-squares discriminant analysis method (PLS-DA) to classify and discriminate elite genotypes of *Coffea arabica* L. submitted to the wet processing in regard to the chemical composition of the beans. We evaluated 31 elite genotypes of *Coffea arabica* L. in an experiment at Epamig Experimental Field in Patrocínio, Minas Gerais, Brazil, which were divided into groups according to their genealogical origin. The coffees were harvested selectively ripe and sent to the processing wet method obtaining the pulped coffee. The coffees were dried in screened sieves until they reached 11-12% water content (b.u.) and were then subjected to chemical analysis. The data were submitted to the chemometric analyzes, PCA and PLS-DA, to characterize and discriminate genealogical groups. The PCA identified the trends of most of the elite genotypes evaluated in relation to virtually all variables. The results of the PLS-DA model showed the variables that most influenced the classification of genealogical groups and identified the possible chemical markers for each group of elite genotypes processed by the pulp method. Sucrose, soluble solids and lipids were important markers for the Bourbon genotype group (GB). The polyphenols were identified as markers for the group of rust-resistant genotypes (GR) and the protein for the group Paraíso germplasm (GP). The PLS-DA model was effective in discriminating the genealogical groups from the chemical composition of pulped coffees.
***Support:** Fapemig and Consórcio Pesquisa Café

Coffee Poster 07

In vitro evaluation of proteolytic digestibility and degree of glycolysis of full coffee extracts and fractions containing various groups of fiber.

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Text Dietary fiber includes the group of polysaccharides which are not digested in the gastrointestinal tract, however has the function of stimulating the growth of beneficial

microbiota, which makes them an integral part of the human diet. The insoluble fiber passes through the entire digestive tract in an unchanged form, while soluble fiber passes through the small intestine relatively unchanged until it reaches the colon. The soluble dietary fiber gels affect the reduction of cholesterol absorption and lowering blood glucose, and the both fractions help to cleanse the body by expulsion of harmful substances found in the gut, what counteracts the pathogenesis of many diseases and allows the proper functioning of the digestive system [1].
Coffee extracts contain many bioactive substances, but are also characterized by a high content of dietary fiber, mainly in the form of polysaccharides such as arabinogalactans and galactomannans, but also as high molecular weight Maillard reaction products (MRP) that arise during the roasting of coffee beans and contain polypeptide and polysaccharide chains[2],[3].
The aim of the study was to determine the proteolytic and glycolytic digestibility of coffee extracts and their fractions containing MRP depending on the degree of roasting of coffee, in terms of dietary fiber content and properties. For this purpose *in vitro* digestion was used, using standard enzymes such as α-amylase, amyloglucosidase, protease, then the relative digestibility of proteins was determined using the trinitrobenzenesulfonic acid (TNBS) method, while the content of reducing sugars was determined using the 3.5-dinitrosalicylic acid (DNSA) method. The analysis provided information on the influence of roasting on the bioavailability of proteins and saccharides of coffee extracts and content of dietary fiber fractions. The research suggests that coffee extracts are a good source of both fractions of dietary fiber. The research was funded by scholarship from the Own Scholarship Fund TUL (project No. RNN/WFS/22/2018).

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Coffee Poster 08

Discrimination of genealogical groups of elite genotypes of *Coffea arabica* L. by the chemical composition of natural coffees*

Marcelo Ribeiro Malta[1], Antônio Carlos Baião Oliveira[2], Gilberto Rodrigues Liska[3], Larissa de Oliveira Fassio[4], Gladyston Rodrigues Carvalho[1], Antônio Alves Pereira[1], Ackson Dimas Silva[1], Laisa Nayara Alvaro[1], Diego Menez Mota[1]
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Text The aim of this work was to use a model created by the partial least-squares discriminant analysis method (PLS-DA) to classify and discriminate groups of elite *Coffea arabica L.* genotypes submitted to the dry process (natural coffees) in regard to the chemical composition of the beans. We evaluated 31 elite genotypes of *Coffea arabica* L. in an experiment at Epamig Experimental Field in Patrocínio, Minas Gerais, Brazil, which were divided into groups according to their genealogical origin. The coffees were harvested selectively ripe and sent to the processing dry method obtaining the natural coffee. The coffees were dried in screened bottom sieves until they reached 11-12% water content (b.u.) and were submitted to chemical analysis. The data were submitted to the chemometric analyzes, PCA and PLS-DA, to characterize and discriminate genealogical groups. The PCA identified the trends of most of the elite genotypes evaluated in relation to virtually all variables. The results of the PLS-DA model showed the variables that most influenced the classification of the genealogical groups and identified the possible chemical markers for the elite genotypes processed by the dry method. The variables lipids, soluble solids, and protein influenced the classification of the Bourbon group (GB). The variable sugar was the one that most influenced in the classification of the Paraiso germplasm group (GP) and the polyphenols variable was the one that most influenced the separation of the rust-resistant cultivars (GR). The PLS-DA model was effective in discriminating genealogical groups from the chemical composition of natural coffees.
***Support:** Fapemig and Consórcio Pesquisa Café

Coffee Poster 09

The impact of ground coffee particle size on the extraction of espresso coffee

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Text Espresso coffee (EC) is one of the most known and consumed types of coffee. EC extraction depends deeply on ground coffee particles produced by the grinding process. To achieve a tempting aroma and taste in EC, the grinding process is in fact a crucial step, because the taste and flavor change widely due to the particle size of ground coffee [1][2]. Particles size distribution (PSD) is essential for extraction kinetics and hydrodynamics of the coffee packed bed [3]. The extraction yield and the strength of the extracted coffee depend greatly on PSD. The same type of roasted coffee in three different PSD (coarse, medium and fine) can generate from sour to acidic (non-volatiles) and from woody/papery to rubbery (volatiles) notes, while maintaining the temperature and pressure constant (94^0C and 9 bar) [4][5]. This highlights that the porosity of the bed and the particle size have to be adjusted to obtain the desired flavor. The influence of PSD on the number of volatiles and non-volatiles in extracted coffee has not well been studied yet [6][7].
This research particularly aims to develop an innovative extraction process by altering PSD and by decreasing the amount of grinded coffee before extraction (from 14 g to 12 g for double Italian EC, at particle sizes between 200-400 and 400-1000 microns). Quantitative and qualitative analyses on bioactive compounds (caffeine, trigonelline and chlorogenic acids) are carried out with HPLC-VWD and GC-MS [8]. Extractions are performed in triplicate for each particle size. The first results confirm a good extraction efficiency of caffeine, which accounted for 170 mg (200-400 microns) and 90 mg (400-1000 microns), respectively. As a consequence, the extraction with smaller particles not only escalated the quantity of bioactive compounds, but also developed descriptive notes of espresso coffee. The final outcomes will give us the opportunity to study further different extraction processes and to develop more sustainable and economically affordable coffee of high quality.

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Poster session: **Coffee**

Massimo Ricciutelli, Gianni Sagratini, Veronica Sirocchi, Giacomo Tomassoni, Sauro Vittori, (2012), Optimization of espresso machine parameters through the analysis of coffee odorants by HS-SPME-GC/MS, Food Chemistry, 1127-1133

Coffee Poster 10

Effect of the repeated intake of coffee silverskin extract on the short-chain fatty acid profile of rat feces

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Text Background: Coffee silverskin (CS) is a thin tegument of the outer layer of the green coffee bean and contains bioactive compounds, such as dietary fiber (28 %), among others[1]. Dietary fiber is fermented in the large intestine by the microbiota leading to the generation of short-chain fatty acids (SCFAs) that contribute to human well-being.

Aim: The present investigation aimed to assess the effect of the repeated intake of coffee silverskin extract (CSE) and sex on the SCFAs profile of rat feces.

Methods: Male (n=15) and female Wistar rats (n=15) received CSE[2] (1 g/kg bw) or water (control group) by gavage once a day for 28 days. The total dietary fiber intake was calculated as the sum of the fiber contents of the diet and CSE (Table 1). Previous studies have demonstrated the safety of CSE at the dose hereby employed [3].

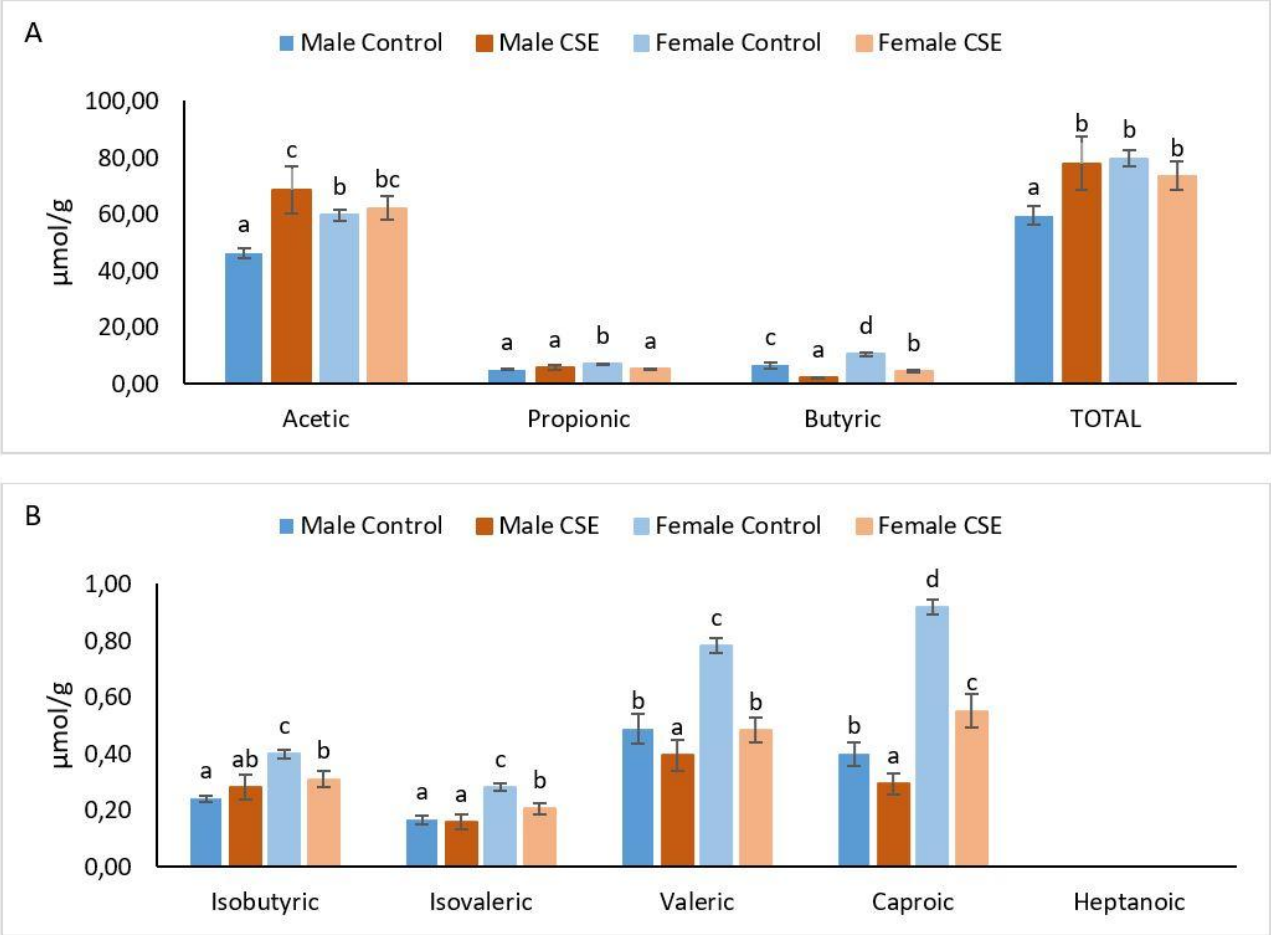
	Male		**Female**	
Fiber intake (g/rat/day)	Control	CSE	Control	CSE
From diet	0.87 ± 0.04 a	0.86 ± 0.00 a	0.65 ± 0.07 a	0.67 ± 0.07 a
From CSE	-	0.08 ± 0.00	-	0.06 ± 0.00
Total	0.87 ± 0.04 a	0.94 ± 0.04 b	0.65 ± 0.07 a	0.73 ± 0.07 b

Table 1. Fiber intake from diet (3.9%), coffee silverskin extract (CSE) and total of controls and rats treated with CSE (1 g/kg bw). Data represent the mean ± standard deviation. Different letters denote significant differences (T test, $p < 0.05$).

Feces from the last 24 h were collected from the treated and control groups and SCFAs were identified and quantified by using gas chromatography coupled to mass spectrometry (Agilent 7890A, Agilent 5975C).

Results: Rats treated with CSE had a significantly increased intake of dietary fiber ($p < 0.05$) (Table 1). The most abundant SCFAs found in feces were acetate, propionate and

butyrate (Figure 1). Total SCFAs (60 µmol/g and 78 µmol/g for control and treated rats) were significantly increased ($p < 0.05$) in feces when male rats were treated with CSE. However, this effect was not observed in female rats (79 µmol/g and 73 µmol/g for control and treated rats). In male rats, acetate significantly increased ($p < 0.05$) in feces after the repeated intake of CSE, but propionate and butyrate levels were decreased.



Majority (A) and minority (B) short chain fatty acids (SCFAs) in feces of controls and rats treated with CSE (1 g/kg bw). Data represent the mean ± standard deviation. Different letters denote significant differences (Tukey test, $p < 0.05$).

Conclusion: Sex seems to influence the SCFAs profile of rats' feces. CSE fiber undergoes significant fermentation in male rats. CSE might be considered as a sustainable functional food ingredient with effect on the gut microbiota and health promoting properties associated to SCFAs.

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Coffee Poster 11

Food and Beverage Fraud Prevention Using Isotope Fingerprints

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Text In this presentation the application of **stable isotope fingerprints in food and beverage fraud detection** is explored. Data are shown that show how stable isotopes offer conclusive answers on questions associated with origin, adulteration and correct labeling of food and beverage products. An overview of the interpretation of isotope fingerprints and the technology used is also provided.

The food and beverage industry suffers from fraudulent activities that include incorrect labeling of products and adulteration, which has a significant impact on food and beverage safety, brand names and reputation and the market economy. Preventing food and beverage fraud is a key challenge that requires a reliable, cost-effective analytical process that can detect whether the labeled product is authentic or if it has been changed after the final manufacturing process, or alternatively if it has been independently produced, using alternative ingredients, but labeled as an original product.

Detecting food and beverage fraud can be achieved using stable isotope measurements because stable isotopes can differentiate between food and beverage samples which otherwise share identical chemical composition: this is called the **isotope fingerprint**. Using **the isotope fingerprint of food and beverage products** is a reliable technique in food and beverage fraud prevention and food safety.

Coffee Poster 12

An analytical decision maker for routine controls of the incoming defective smoky cocoa beans.

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Text Cocoa smoky off-flavour generates from unappropriated or not well controlled artificial drying applied to beans to speeding up the post-harvest process to counteract the effect of unfavourable climate. Smoky off-flavour cannot be removed during chocolate production and it can heavily affects the quality of the finished chocolate products[1]. Tests to define the cocoa quality is often carried out on the liquor and not on the incoming

beans. Moreover, sensory tests require a trained and aligned panel, and it can seldom be implemented at-line for an immediate feedback and a critical objective evaluation. At the same time, a reference objective method to detect this off flavour on incoming raw material is not available. The aim of this study is to use diagnostic mass spectral fingerprints of the headspace sampled by SPME (HS-SPME)-*enose* combined with chemometrics in developing an instrumental prediction model to detect smoky defective beans to be used as an analytical decision maker for routine controls[2-3]. Fifty cocoa bean samples from crops of different years and origins were analysed and sensory evaluated from an internal panel. A supervised PLS-DA model classification built on a cross-validated (5 CV) training set (n=35) and applied on an external test set (n=12) of samples providing a 100% of correct classification. Results show that the HS-SPME-eMS fingerprints combined with chemometrics is a promising TAS (Total Analysis System) for a high throughput method to detect defective cocoa beans[4].

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Coffee Poster 13

Prediction of coffee sensory quality through MS-e-nose as analytical decision maker for routine controls: possibilities and limitations.

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Text Aroma is a primary hedonic aspect of a good coffee playing a fundamental role in coffee choice [1] and is a meaningful signature of the products [1-5]. The cup tasting is the most important criteria to define the coffee quality, nevertheless it requires trained and aligned panel, and it can seldom be implemented at-line for an immediate feedback . This study aims to apply diagnostic mass spectral fingerprints to develop an instrumental prediction model exploitable as an analytical decision maker for routine controls as a complement to define coffee sensory quality in cup. This method in combination with sensometrics, can be implemented in an automatic Total Analysis System (TAS) to provide a high throughput solution for coffee quality control.
Coffee samples were sensorially evaluated and analyzed by HS-SPME-MS and the resulting data elaborated with multivariate analysis. The panelscores were submitted to ANOVA and to a paired t-test between each expert. PLS-DA applied to the reprocessed MS spectral fingerprints enabled to select 36 (m/z) on 315 fragments suitable to correlate with high and low scores within a sensory attribute. The regression models was built with a training set of 146 objects and an external test set of 30. The leave-p-out cross-validation (20) method was used to select the number of components (fragments). Acid, bitter and woody notes were the most reliable. The mean error in the sensory scores prediction on

test set with these data was within the fixed limit of ±1.
The results show that the HS-SPME-MS fingerprints in combination with chemometrics is a promising approach to be used as a TAS system for a high throughput solution to define the coffee sensory quality in cup. This approach offers a reliable sensory scores prediction if, and only if, applied with a robust mathematical model derived from a high number of representative samples and an accurate alignment in the lexicon to rate the samples.

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Coffee Poster 14

Coffee sensory properties: a complementary data fusion to simulate odor&taste integration by instrumental approach. Possibilities and limits

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Text This work deals with the study of a flavoromics approach to describe and predict coffee sensory quality and its potential and limits in routine control.
The cup tasting is nowadays the most important criteria to define the coffee quality, although i) it is time-consuming requiring panel training and alignment, and ii) it cannot be easily implemented at-line for an immediate feedback and/or a critical objective evaluation.
Flavoromics is an “-omic” and “-holistic” approach focused on low molecular mass compounds (volatile and non-volatile) linking them to a sensorial perception through chemometric approaches[1-2]. Coffee cupping considers both aroma and taste and its evaluation considers several attributes at the same time[3-4]: the definition of the flavor quality therefore requires two complementary analytical techniques.
Chemical data of the investigated coffee samples (n=150) was obtained by analyzing both volatile and non-volatile fraction by HS-SPME-GC-MS and HPLC-UV/DAD. Sensory data were collected by an expert panel in a monadic way through a quantitative descriptive analysis. Multiple Factorial Analysis (MFA) was used to correlate the pool of observations (volatiles, non-volatiles, and sensory scores). PLS-DA was applied to select informative compounds from each analytical approach and PLS regression was used to correlate chemical and sensory data in prediction.
When focusing on bitter note, the sensory prediction models relative to targeted aroma and taste fingerprints displays good results in determining the sensory scores (Q^2 in prediction is 0.98 *vs* 0.95 with a prediction error of 0.6230 *vs* 1.0501 respectively for aroma and taste). However, coffee cupping is a multimodal perception involving different attributes at the same time[3-4] that can therefore be better represented by adopting an aroma and taste data fusion. This approach results in bitter scores prediction values of Q^2

0.89 with an error of 1.4007. A comparison of the Q model parameter with the other sensory notes highlights a different contribution of this taste fraction in their description since it has more impact in describing Acid, Bitter, Woody notes than Flowery and Fruity.

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Coffee Poster 15

PROLIFIC: Integrated cascades of PROcesses for the extraction and valorisation of proteins and bioactive molecules from Legumes, Fungi and Coffee agro-industrial side streams

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Text Agro-industrial residual biomass, side streams and food production by-products like legumes, fungi and coffee are potential sources of valuable ingredients even though the routes for their exploitation are still at an early stage. Pursuing the ambition of achieving prolific valorisation of untapped biomass streams, the project R+D+I activities and partners of **PROLIFIC** have been positioned around a central innovation cycle that is mainly driven by industrial end-users who exactly know the needs of their customers and the technical constraints and industrial environment in their respective sector.

In particular, coffee is the highest consumed beverage in the European Union after water. Europe however does not grow coffee but imports around 45% of the exported crop, worldwide. Coffee residues result from the roasting process and are available in EU all year long in considerable amounts. Two main by-products are considered in this project: *Silver skin* up to 5% of raw material and *not compliant green coffee* (defective beans) which may vary according to the quality standards of each coffee company.

The **PROLIFIC** project will apply a range of processing technologies to recover significant amounts of proteins/peptides and other value-added compounds such as phenols, fibres and caffeine from these industrial processing residues. The economically and environmentally sustainable extraction assisted either by enzymatic hydrolysis, ultrasound or microwave application and the conditioning techniques will be upscaled in an industrially relevant environment.

The extracts from biomass streams will be subjected to different treatments (such as

purification, concentration, hydrolysis, and stabilization) and converted into ingredients for food, cosmetic and feed formulations and components for polymer and composite formulations after safety and techno-economic analyses (Life-Cycle Assessment and Cost-Effectiveness Analysis).

This project has received funding from the Bio Based Industries Joint Undertaking under the European Union's Horizon 2020 research and innovation programme under grant agreement No. 790157.

Coffee Poster 16

Inhibition of growth and ochratoxin A production of *Aspergillus carbonarius* by lactic acid bacteria strains isolated from coffee cherries

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Text Coffee is one of the major crops consumed worldwide. Coffee beans sourced from Côte d'Ivoire are currently subject to ochratoxin A (OTA) contamination due to the post-harvest processing. This study aimed to improve the sanitary quality of Ivorian coffee beans by inhibiting the growth and OTA secretion of pathogenic molds strains using lactic acid bacteria (LAB) isolated from freshly harvested coffee cherries. Coffee sherries plated on potatoes dextrose agar (PDA) and Man Rogosa and Sharp agar (MRS) culture media have shown that potentially toxigenic fungal taxa mostly from genera *Aspergillus* and *Penicillium* are detected in Ivorian coffee. Results showed that five OTA-producing strains of *Aspergillus carbonarius* at levels ranging from 12,000 to 91,000 µg.kg^{-1} and 16 strains of LAB. Molecular identification showed a predominance of *Labacillus plantarum* strains among the BAL strains potentially have fungal antagonism. The fungal growth inhibition tests performed using the double-layer agar technique revealed ten (10) LAB strains with antibiosis potential ranging from 17 to 78 % and seven (7) LAB strains with total inhibition of *A. carbonarius* growth abilities. These observations suggest that *L. plantarum* could be used for growth control and prevent of OTA secretion of *A. carbonarius* in coffee cherries at the field level. Furthermore, cell suspensions and supernatants of tested LAB liquid culture medium reduced OTA production to about 90 and 60 % respectively. Results suggest a direct action of LAB or an indirect action of LAB metabolites on *A. carbonarius* growth, coupled with OTA consumption and / or binding by LAB strains. This study shows a promising biological control of OTA production and excellent abilities of tested LAB for OTA detoxification in coffee beans

Poster session: **Coffee**

Coffee Poster 17

Carbonyl volatile profile of green and roasted coffee to evaluate the quality of beans

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Text The volatile profile of roasted coffee beans has been widely studied and correlated with geographical origin, post-harvest conditions, roasting procedure, and the presence of defective beans. On the contrary, the volatile composition of green beans is less studied; although the study of the volatile profile of unprocessed natural products is of recognized utility for many purposes.
In green coffee batches, the presence of defective beans can compromise the organoleptic quality of the roasted coffee. Normally, the evaluation of green coffee is performed by visual inspection to detect defects, a process highly subjective and prone to misinterpretations. For this reason, the development of accurate and straightforward methodologies that can characterize the quality of the raw materials for the production of roasted coffee and identify the presence of defective beans is important to consider.
In this work, simple methodologies were developed to evaluate the volatile profile of green coffee beans. Carbonyl compounds were studied to evaluate beans with different defects, and the variation on the carbonyl compounds content with the roasting degree. The compounds were extracted from the headspace of the samples, derivatized with 2,4-dinitrophenylhydrazine, and analysed by high performance liquid chromatography with UV detection (HPLC-UV). The identity of the peaks was assessed by mass spectrometry (MS) analysis [1]. Around 30 carbonyl compounds were identified in green (healthy and defective) and roasted coffee beans. The developed methodologies showed to be promising to be used as simple, low cost and easy to perform strategies for the evaluation of the quality of green coffee batches. The identified volatiles can be correlated with coffee ageing, presence of defective beans, and degree of roasting, envisaging the potential use of some identified compounds as coffee beans quality markers.

Acknowledgements: J. R. Santos and I. M. Valente acknowledge the funding program (DL 57/2016 – Norma transitória) supported by Fundação para a Ciência e a Tecnologia I.P. This work was supported by UID/QUI/50006/2019 with funding from FCT/MCTES through national funds.

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Poster session: **Coffee**

Coffee Poster 18

Coffee Silverskin and Spent Coffee Ground investigation: A new analytical method for 30 bioactive compounds quantitation

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Text Nowadays, there is great political and social pressure to reduce the pollution arising from industrial activities, including coffee sector[1]. Coffee is one of the largest traded commodity worldwide since the consumption of coffee beverages is overtaken only by that of water and tea[2][3]. Hence, to satisfy the immense demand for coffee, coffee industry produces a huge quantity of byproducts which cannot be considered just as waste materials but should be considered as starting raw materials for other projects. The main coffee byproducts, produced during roasting process and both soluble coffee industry and coffee brewing, are Coffee Silverskin (CS) and Spent Coffee Ground (SCG), respectively[4]. The purpose of this project was to deepen the knowledge about CS and SCG in the perspective of their reuse as nutraceutical. A new analytical method for quantitation of 30 phenolic compounds by using UHPLC-MS/MS has been developed. Then, various extractions, i.e. magnetic stirrer and ultrasound-assisted extraction, using different solvents viz., hydroalcoholics, alcoholics and aqueous, were evaluated for their ability to take out target molecules from those matrices. From an analytical point of view, UHPLC-MS/MS studies were performed using an Agilent 1290 Infinity series and a Triple Quadrupole 6420 (Agilent Technology) equipped with an ESI source operating in negative and positive ionization mode. The separation was achieved using a Kinetex PFP analytical column (50 × 2.10 mm i.d., 2.6 µm) using a binary gradient of water (A) and methanol (B) both with 0.1% formic acid. The method was sensitive (LOQs for all compounds were from 1 to 100 µg kg^{-1}), linear (R^2 for all analytes was higher than 0.9907), accurate and robust. Results demonstrated that highest content of total phenolic compounds were found in ultrasound extraction with a mixture of EtOH:H_2O 70:30 v/v. In this case, the highest amount of total bioactive compounds concentration was found, followed by aqueous extraction with magnetic stirrer and methanolic ultrasound extraction. In the future, several biological activities, i.e., antioxidant, prebiotic and antimicrobial, will further be studied in the most promising extracts.
Acknowledgments: The authors are grateful to the University of Camerino for funding the FAR project (year 2018).

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Coffee Poster 19

Chemical and sensorial profile from differents roast profiles.

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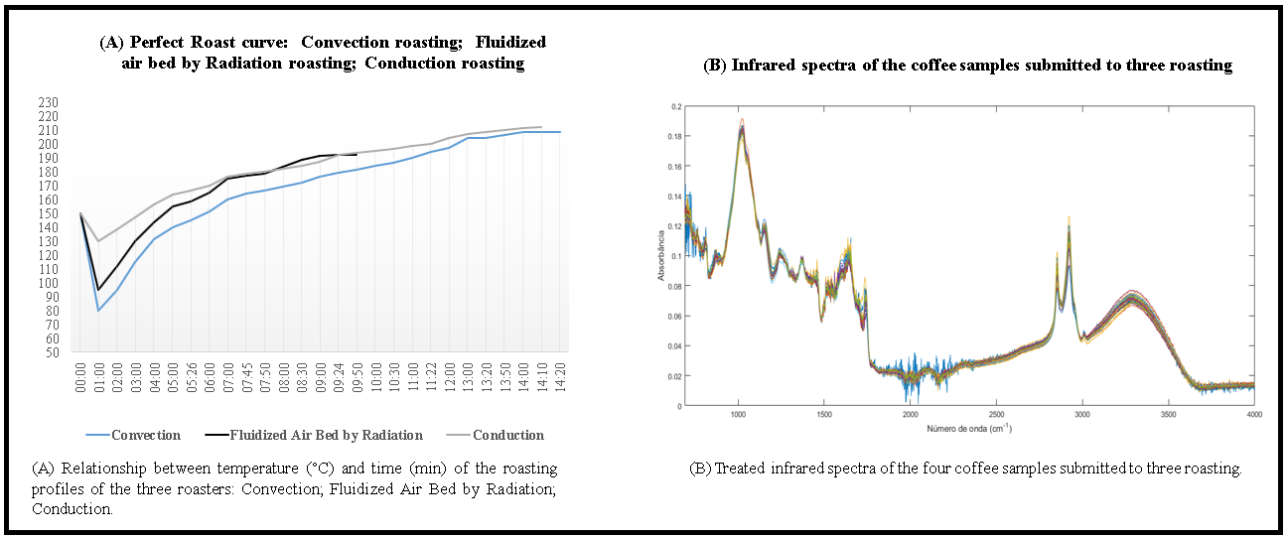
Text The process of roasting a coffee has a number of complexities depending on the procedures that are adopted. In recent years a diversity of technologies has been observed that are helping to improve the quality and / or control of this process. Thus, different roasting matrices have been employed in the industry for the purpose of improving quality levels. In this study, 3 different roast curves (light, medium and dark) were applied, associated with different roast matrices with purposeof identifying the best time of roast to improve the entire process. The experiments were realized, with *Coffea arabica* (pulped natural), were conducted in a randomized complete block design with 3 replications, in the plot scheme subdivided in time. As the plots consist of 3 roasters, Fluidized Air Bed by radiation and conduction and convection, and subplots composed of 07 roasting times, 2, 4, 6, 8, 10, 12 and 14 minutes.
For the statistical analysis, joint analyzes of experiments were performed, the means being compared by the Tukey test at 5% of probability, followed by dendrograms using the Mean Euclidean distance to measure the distances between the chemical and sensorial analysis groups by the method of complete hierarchical linkage. The infrared spectra in the middle region of the roasted and ground coffee samples were obtained on a Cary 630 FTIR spectrometer in an attenuated total reflectance (ATR) diamond accessory with reflection angle of 45 °, 1 mm diameter, 200 µm active area and approximately 2 µm penetration depth in the sample, using a Zinc Seleneto reflectance detector (ZnSe).
Preliminary results indicate that the Q-Graders have an acceptability for the medium roasts considered as optimal by the literature recommended by SCA and the chemical data from the infrared analysis indicates that the chemical structure undergoes changes in the groupings of the secondary amines (sugars) in the range of 3400 - 3350cm^{-1}, thus indicating a possibility to understand the behavior of the Maillard reaction based on the sugars present in the coffee fruits.



(A) Relationship between temperature (°C) and time (min) of the roasting profiles of the three roasters: Convection; Fluidized Air Bed by Radiation; Conduction.

(B) Treated infrared spectra of the four coffee samples submitted to three roasting.

(A) Relationship between temperature (°C) and time (min) of the roasting profiles of the three roasters: Convection; Fluidized Air Bed by Radiation; Conduction. (B) Treated infrared spectra of the coffee samples submitted to three roasting.

[1]

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Coffee Poster 20

Effect of different altitudes in the profile of the bacteria community in coffee grains

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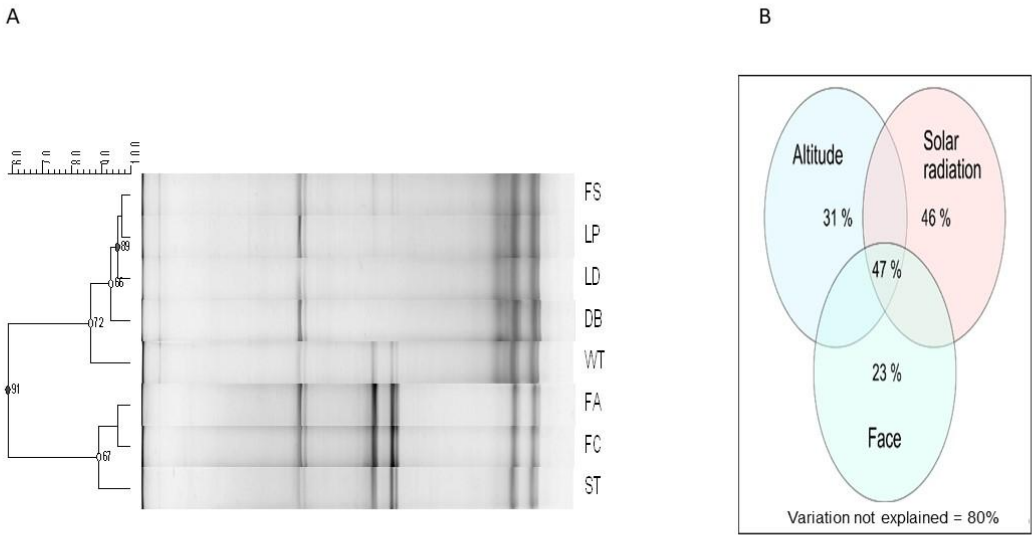
Text The coffee quality depends on environmental factors as shade, solar radiation and altitude, as well as technical factors such as processing, drying and roasting. If the environment affects the final quality, consequently both wet and dry processing significantly influence the microbial community structures. Thus, the aim of this study was to evaluate the Bacteria community profile present on fruits from *Coffea arabica L.* collected in 8 different farms with altitude ranging from 735 to 1,078m. The genomic DNA from the samples were extracted using the Nucleo Spin Soil Kit. Later, we have used the nested PCR-DGGE technique. The first PCR reaction was performed with the 27F and 1492R primers and then a Nested PCR reaction with the U968-GC and 1492R primers. From the product of the second reaction, 20 µL was applied on an 8% (w/v) acrylamide gel

with a denaturing gradient from 35% to 55% to obtain the community profile. The gel was subjected to vertical electrophoresis at 60V for 12h at 60°C, then stained for 40min with *Sybr Gold* (1x); the gel was then photographed under UV light on a Molecular Imaging System. The DGGE profiles, aligned based on the external markers, were analysed and compared using BioNumerics software.



A) Cluster analysis obtained from the DGGE banding pattern of the 16S microbial communities from coffee fruits. Part B) - Venn diagram.

Farmer	Altitude	Sun Face
FC	735.00	West
FA	799.17	East
ST	870.24	East
DB	969.00	South
LD	969.00	South
WT	1021.99	South
FS	1052.17	South
LP	1078.08	South

Experiments points, altitude and solar radiation face

Dendrogram analysis showed that two main groups with 55 % the similarity. The first group with 85 % the similarity (FC, LP, LD, DB and WT) involves the farms located in altitude ranging from 907 to 1.078 m belonging the face south, and the second with 90 % the similarity (FA, FC and ST) involves the farms located in altitude ranging from 735 to 870m, belonging in the face west and east. The Venn diagram showed that face, solar

radiation and altitude explain 47 % of the variation the profile of the bacterial community. This work showed the influence of the face, solar radiation and altitude in the community profile of bacteria and that other factors may also affect this profile.
[DeBruyn]

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Coffee Poster 21

Application of the CATA (check-all-that-apply) method to validate a panel for sensory analysis of coffee

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Text The training of a panel for the QDA analysis requires lot of time and energy, both during the training and the monitoring of the panel performance itself. Several rapid sensory methods have been proposed like Flash Profile[1] (FP), Free Choice Profiling[2] (FCP) and CATA (*check-all-that-apply)* method. While the application of FP and FCP is widely used with untrained subjects, CATA method has been used where trained panels has been employed[3].
The aim of this work was then to compare a QDA with a CATA analysis performed on nine espresso coffee samples.
In particular, comparison was performed only for odor and flavor sensory descriptors.
While the reproducibility of the panel used for QDA has been checked by using the analysis of variance considering Product, Judges, Repetition and their interaction as main factors, the reproducibility of the panel used for CATA has been checked using an average reproducibility index (R_i)[3].
Only judges who have shown a reproducibility index (R_i) > 0,5 have kept for following analysis. Moreover, in order to evaluate the agreement of panel in products evaluation, correspondence analysis (CA) for each product has been performed.
Once assessed the performance of both panels, PCA and CA analysis have been performed showing that while PCA explained the 45,52% of differences between products, the CA analysis explained the 59,18% of differences. Products were then described by QDA and CATA with same characteristics.
CA analysis has been then compared to the PCA analysis comparing sample configurations in the first and in the second dimension of each methodology using the Regressor Vector coefficient (RV)[4].

The RV coefficient between sample configuration was significant as it was 0,603.
This data shows that results obtained from both panels are comparable then the CATA methods can be an interesting alternative to QDA above all for the low time request for panel training.

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Coffee Poster 22

Identification of chlorogenic acid and lipid biomarkers for the differentiation of Arabica and robusta coffee beans

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Text Abstract
Coffea arabica and *Coffea canephora* (Robusta coffee) are the most commonly consumed coffee varieties globally. In this study, NMR and LC-ESI-MS^n techniques were employed to profile and quantify the most abundant chlorogenic acid in 54 different samples of the two coffee varieties from diverse origins of the world. Mono-caffeoyl quinic acids were found to show no variations if the two coffee varieties were compared. Significant variations were observed for feruloyl quinic acids, dicaffeoyl quinic acids and 5-sinapoylquinic acid. Additionally isomer ratios were explored as a potential marker for coffee authenticity along with a thorough statistical evaluation of rather extensive data set. Furthermore, the triacylglycerol constituents of the Arabica and Robusta coffee were profile to discriminate between the two coffee varieties by liquid chromatography coupled with mass spectrometry (LC/MS) and molecular ions fragmentation by tandem mass spectrometry (MS/MS). From the tandem-MS analysis, ammonium adducts [NH_4^+] of the TAGs were acquired and 18 TAGs were identified with their respective molecular weights and relative abundance (%) extrapolated from the data system with the aid of DAD detector. Some of these TAGs extracted from the coffee oil are being reported for the first time in this study and could be use to differentiate distinctly between the two coffee varieties.

Coffee Poster 23

Development of a functional home-made beverage based on silverskin

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Poster session: **Coffee**

Text Coffee is the most important food commodity in the world with 8.8 million tons of green coffee beans globally produced in 2015/2016. Currently, it is cultivated by 70 countries around the world, and for some of them, it represents the main agricultural export product. From the coffee cherry transformation into coffee brew a large amount of by-products are obtained but these materials contain appreciable amounts of bioactive compounds, mainly chlorogenic acids and antioxidant dietary fiber. Therefore, they represent an exciting opportunity to obtain new functional ingredients to be use as natural antioxidant, nutraceuticals, and preservatives in an enormous variety of food preparations with high nutritional value. Coffee silverskin (CS) is the thin tegument that cover the two green coffee beans and it represents the by-product coming from the roasting process. It represents about 4.2 % (w/w) of coffee beans hence, considering a production of 8.8 million tons of green coffee, a total of 0.37 million tons of CS is annually produced. This large amount of CS represents a great disposal cost for large-scale coffee roasters in consuming countries. Carbohydrates, ashes, proteins, fats and bioactive compounds such as chlorogenic acids, melanoidins and caffeine characterize the chemical composition of CS. In literature its use as structural, prebiotic, antioxidant, antimicrobial, anti-glycemic, anti-allergenic and anti-diabetic was then suggested. The aim of this work was to develop a home-made functional beverage using CS obtained by roasting process of Arabica, Robusta and decaffeinate beans. The beverages were prepared with two different dimension (250-500 µm and 1000-2000 µm) CS comparing four different home-made extraction techniques: infusion, moka, espresso and capsule. Obtained results shown that the better CS particle size was 250-500 µm and moka and infusion the best extraction techniques to get a beverage rich in polyphenols. Between the CS categories, decaffeinated CS beverages were the richest of all polyphenol compounds.

Coffee Poster 24

Chemical and sensory perception of Robusta Coffees under wet processing

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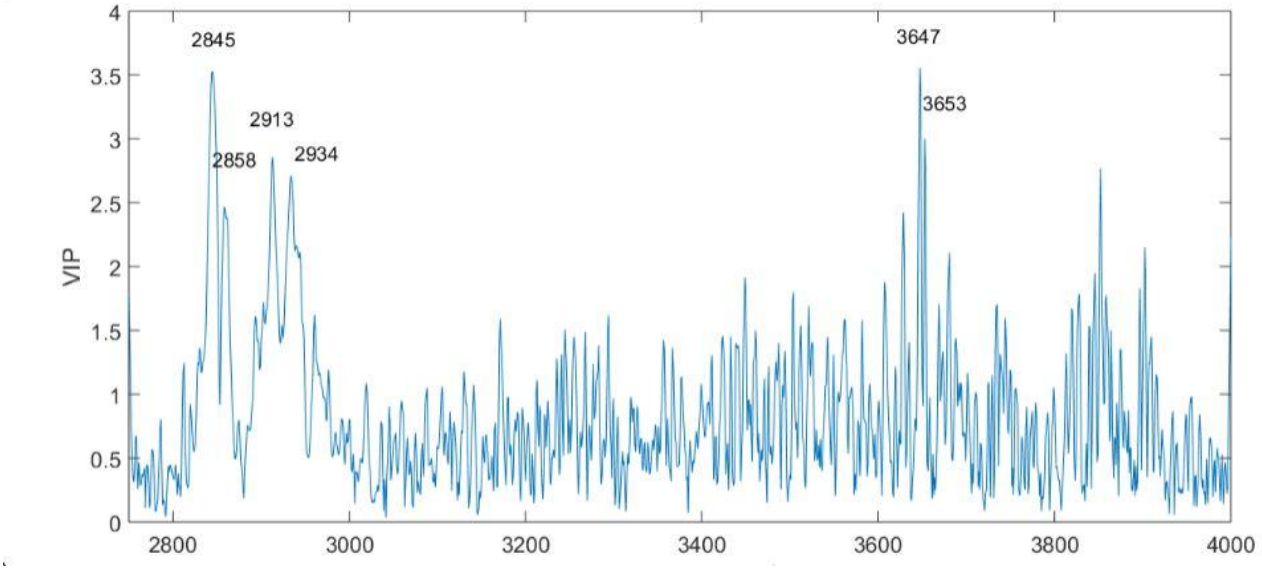
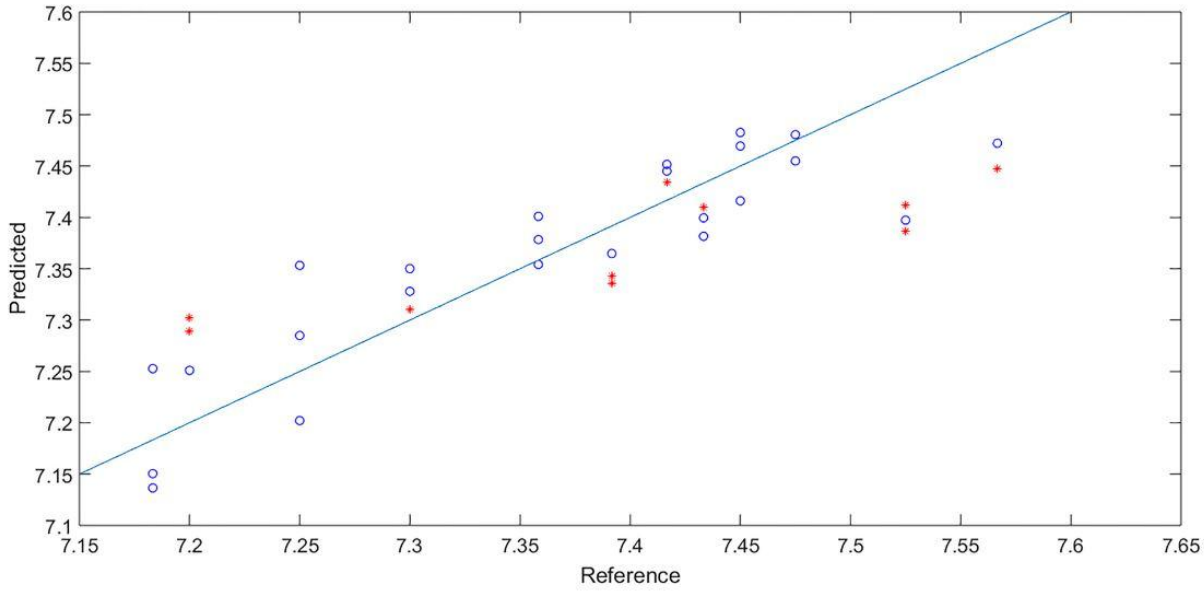
Text Robusta coffee represents almost 20% of all Brazilian coffee yield. However, in terms of quality, it shows less relevance in relation to arabica coffee. This study applied the use of spontaneous and induced fermentation with starter cultures (yeast) to evaluate the sensory potential from the impacts generated by the types of processes adopted. The experiments were conducted in a randomized block design with five replicates, with three fermentation times (24, 48 and 72 hours) and four wet processes: Washed, Yeast fermentation, Fully washed without yeast and Fully washed with yeast. The sensory analysis of the coffees was carried out by a panel of 6 cuppers, number proposed to

reduce the subjectivity of the analysis [1]. Infrared spectra were taken in the medium region (FTIR) of the coffee samples generated by the treatments. The method was used in the construction of chemometric models to predict sensory ratings given to the attribute ‘acidity’. The sensory results indicated a higher score for the acidity attribute, in dry fermentation with yeast at the time of 48 hours, indicating that the induced fermentation is able to act in the construction of sensory routes. The study also indicates that it is possible to estimate the score of the sensory attribute acidity of the robusta coffee submitted to different processes and fermentation times using infrared spectra in the medium region associated to the PLS regression. The selected band (2800 to 4000 cm-1) was able to capture the information related to the acidity attribute, which has the highest contribution wave numbers in the prediction highlighted in Figure 1.



Graph of the weight of the variables (VIP) in the constructed PLS model.

The considerable stability of the models allowed us to establish a correlation between the chosen spectral region and the sensory attribute acidity, since it is a chemical property with direct influence on the spectral profile, due to the presence of chlorogenic acids.



Relation between reference (o) and predicted (*) values of the score for the sensory attribute acidity, of the coffee samples.

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Coffee Poster 25

Free, Esterified, and Glycosylated Sterols in Arabica and Robusta Coffee Beans

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Text Over the past years phytosterols have achieved more importance due to their beneficial properties. In scientific literatures it was reported that the phytosterols reduce the cholesterol levels and have anti-carcinogenic effects. Coffee oil contains a number of sterols. In addition to 4-desmethylsterols, various 4-methyl- and 4,4-dimethylsterols have been identified. Cholesterol, campesterol, stigmasterol, ß-sitosterol, stigmastanol, Δ5-avenasterol, Δ7-stigmastenol, Δ7-avenasterol, citrostadienol, gramisterol, cycloartenol, and traces of 24-methylenecycloartenol were quantified in different coffee infusions, Scandinavian style coffee, espresso, and filtered coffee. The main sterol is ß-sitosterol with about 50%, followed by stigmasterol, campesterol, and Δ5-avenasterol. Sterols occur as free sterols and sterol fatty acid esters. In the latter C_{18}, C_{16} and $C_{18:1}$ are the main compounds with a proportional distribution similar to that reported in triglycerides.
Steryl glucosides were isolated and elucidated in green Arabica and Robusta coffee beans by our group for the first time [1]. The modified analysis scheme first developed by Oelschlägel et al. [2] allows for obtaining three sterol fractions: the first contained sterol fatty acid esters, the second free sterols and, the third fraction steryl glucosides. The sterol fatty acid esters were analysed after saponification and derivatisation, the free sterols after silylation by GC/FID and quantified via ß-sitosterol. The separation of the steryl glucosides was accomplished on a Phenomenex Synergi 4 µ Fusion 250 x 3 mm equipped with diode array detector. The quantification was carried out either with ß-sitosterol or with stigmasterol, depending on the number of double bounds in the molecule.
Between 8% and 12% of the total sterols were steryl glucosides**.** The contents of the main sterol glucosides in different green Arabica and Robusta coffees were presented as well as the percentage distribution of the sterol fractions (free, esterified and glycosylated) in relation to the total sterols.

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Coffee Poster 26

Effects of weed control methods in coffee crop inter rows on quality of coffee beverage

Elifas Nunes de Alcântara, Marcelo Ribeiro Malta; Carla Pádua Martins
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Text Abstract: Weed control is important cultural practices in the coffee crop management. Weeds compete with the coffee crop for light, nutrients and water, and interfere with growth and development; weed competition may reduce yield coffee by up to 77%. Coffee crop is more sensitive to weed competition for water on the dry season and for nutrients at the rainy season. This competition, in addition to affecting the production, affects also, other parameters. This study was made to determine whether different weed control methods in coffee inter rows, may affect the quality of coffee beverage. An experiment installed at the Agricultural Experimental Field of research at south of Minas Gerais - EPAMIG (Minas Gerais state research institution)- at São Sebastião do Paraíso, MG, Brazil, in an Oxisol clayey, with randomized block design using seven weed control treatments between on the lines using three replicates. The coffee cultivar used were, Paraíso MG2. Treatments were: mower, disk harrow, rotary tiller, post-emergence herbicide (glyphosate), pre-emergence herbicide (oxyfluorfen), manual weeding and no weed control. The grain samples were collected in the treatments and prepared separately during each year, from 2008 through 2018. The coffee grain samples were sent to coffee quality laboratory, for sensory analysis of tasters. From 2008 through 2010, analyzes were done in the BSCA (Brazilian Specialty Association) rating protocol, however, from 2011 through 2018 the SCAA (Specialty Coffee Association of America) protocol was used. In these eleven years, the results indicated that, the pre-emergence herbicide treatment, at coffee inter rows, presented in all evaluations scores over 80 points. In other words, this weed control method, supplanted all others due to its influence on coffee drink quality, that was classified as special coffee.

Key words: weed control methods, coffee drink quality.

Coffee Poster 27

Effects of weed control methods in coffee inter rows on coffee yield.

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Text Abstract: The coffee plant is very sensitive to weed competition. Yield losses due to weed competition, may reach to 77%. On the coffee inter rows weeds may be controlled by several ways. To compare effects of weed control at inter rows, an experiment was

installed in 2006, in randomized block design with seven treatments on coffee inter rows: mower, disk harrows, rotary tiller, post-emergency herbicide (glyphosate) at 720g a.i./ha pre-emergency herbicide (oxyfluorfen) at 720 g a.i./ ha, manual weeding and no weeding, using three replications, in a coffee cultivar Paraiso (MGH 419) planted in Oxisol soil spaced 4 by 0.7 m at EPAMIG at São Sebastião do Paraíso, MG. Yields of each treatment from 2008, through 2018, were assessed. Pre-emergency herbicide treatment, presented the highest production, and treatment without weed control at inter rows, presented the lowest yield. The mechanical methods produced 15% to 20% less than pre emergency herbicide treatment. Results shown that, these methods depend on the operational availability of weed control. Disk harrow and the rotary tiller treatment methods disseminated the bermuda grass [Cynodon dactylon (L.) Pers] and the purple nutsedge (Cyperus rotundus L.) at the area. Mechanical weed control methods, presented up to until 20% less yield than pre-emergency herbicide. Thus the results shown that pre-emergency herbicide treatment was the best weed control method for coffee crop.

Key words: weed control, coffee, yield.

Coffee Poster 28

Identification and profiling of human urinary metabolites using UHPLC-MS and ^{1}H NMR after roasted coffee consumption

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Text Diet-relevant phenolics of coffee, such as the caffeoylquinic acids (CQAs), the feruloylquinic acids (FQAs), as well as free cinnamic and benzoic acid derivatives, have been shown to possess a wide range of health benefits [1] [2]. To fully understand the fate of coffee phenolics and their metabolites within the human body, it is important to assess their metabolism and bioavailability after ingestion. The present study is aimed at the apparent human gut bacterial and liver metabolism of coffee phenolics, which can be reflected in the form of optimized compounds and their metabolites in the urine samples. A feeding trial experiment was conducted, where recruited volunteers (n = 9) consumed 4 cups of roasted robusta coffee for two days. Their urine and fecal samples (n=2) were collected at different time points over 48 h period.
Optimized methods using UHPLC-MS/MS and ^{1}H NMR techniques were implemented to identify and quantify the bioactive compounds of coffee in the volunteer urine samples. Moreover, untargeted and targeted metabolomics approaches using principal component (PCA) and partial least squares discriminant analysis (PLS-DA) were applied to the LC-MS and NMR data, to allow the identification of significant biomarkers responsible for discrimination of the metabolomes of various volunteers. The results herein indicated the bacterial and phase-II conjugated metabolites of CQAs, FQAs and other cinnamic acid derivatives could be the main contributors of coffee-related health effects, as well as contributing to the urinary metabolome variations between different individuals. Additionally, modulation of human primary and secondary urinary metabolites was also investigated.

Poster session: **Coffee**

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Coffee Poster 29

Elaboration of sensory profile of Coffea canephora processed with induced fermentation

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Text The use of starter cultures in the coffee fermentation is an interesting alternative to obtain a differentiated product. Using the correct microorganisms, the fermentation can contribute to formation of new sensory features, with the modification of the flavor profile of the coffee, raising the quality curve generating more flavor, aroma and texture. The present study aims, with different types of processing using yeast Saccharomyces cerevisiae as a start culture, to add more quality to the sensory profile of conilon coffee. The experiment was conducted in a randomized block design with 5 replicates and 5 treatments: Semi-Dry, Washed, Yeast Fermentation, Fully Washed Without Yeast, Fully Washed With Yeast and a 24-hour time factor. For the sensory analysis, the UCDA [1] methodology was used. Where the characteristics, balance, overall, uniformity, clean cup and total score were evaluated. Statistical analysis was performed with variance analysis and the means were compared by the Tukey test at 5% probability.

Treatment	Balance		Overall		Uniformity		Clean Cup		Total Score	
Semi-Dry	7.28	b	7.28	b	10.00	a	10.00	a	78.98	b
Washed	7.48	ab	7.49	ab	10.00	a	10.00	a	80.74	ab
Yeast Fermentation	7.47	ab	7.49	ab	10.00	a	10.00	a	80.22	ab
Fully Washed Without Yeast	7.48	ab	7.48	ab	10.00	a	10.00	a	80.42	ab
Fully Washed With Yeast	7.60	a	7.68	a	10.00	a	10.00	a	81.87	a
Média	7.46		7.48		10.00		10.00		80.44	

[1] Means followed by at least one letter vertically, do not differ from each other by the Tukey test at 5% probability.

Table 1 - Averages of the characteristics Balance, Overall, Uniformity, Clean Cup Total Score, evaluated in four treatments.

According to sensory results observed, for the attributes: group, balance, overall and total score, indicate that the use of yeast in the Fully Washed With Yeast treatment was statistically different from the Semi-dry treatment. However the treatments Washed, Yeast Fermentation, Fully Washed Without Yeast were not differentiated to 5% of probability by Tukey's test. Based on the sensorial descriptors and the total score, the best processing for the conilon coffee at the 24 hour fermentation time was the Fully Washed With Yeast, consisting of inoculating yeast for dry fermentation, without water, and washing the pulp after fermentation. Therefore, this processing can increase the sensory curve, causing producers to have the opportunity to optimize the coffee quality produced, introducing a new process technique with fermentation, to enhance the final quality of the beverage.

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Coffee Poster 30

Exploring the impact of spontaneous wet fermentation on microbiota and coffee quality

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Text Coffee (*Coffea arabica*) grows in tropical regions where its fruit, *i.e.* the coffee cherries, undergo a series of processing steps until the dried green coffee beans are ready for roasting. Thereby, coffee quality is influenced by many factors, e.g. genetic, climate, harvest, post-harvest-processing, and storage. In wet processes, spontaneous fermentation of the pulped coffee beans has a major impact on coffee quality [1]. The aim

of this work was to monitor two wet processes in Nicaragua, called “Reposo” and “Tradicional”. The main difference is the so called *Reposo* step, a resting of coffee cherries for 40 h before pulping. The general process is described as follows: coffee cherries (*C. arabica* var. Catuaí Rojo) were floated, either pulped directly followed by 14 h of fermentation in the ”Traditional” process or rested for 40 h during *Reposo*, followed by 10 h of fermentation in the “Reposo” process. Then, the coffee cherries were washed and sun dried. Lactic acid bacteria (LAB) and yeasts as well as pH were determined at different steps during the two processes. The initial cell counts were 4.06 log cfu/g for LAB and 4.42 log cfu/g for yeasts and the pH was 5.38. After 40 h *Reposo*, LAB and yeasts increased to 7.85 lof cfu/g and 6.26 log cfu/g, whereas pH decreased to 4.95. During the subsequent fermentation, LAB and yeasts slightly increased to 7.98 log cfu/g for LAB and 6.62 log cfu/g for yeasts in the “Reposo” process and to 7.63 log cfu/g and 6.46 log cfu/g, respectively, in the “Tradicional” process. The pH after fermentation was 4.84 in the “Reposo” and 5.23 in the “Tradicional” process. During 10 d of drying to a moisture content of 11 %, the microbial cell counts decreased steadily and reached levels of initial counts of coffee cherries. The metabolites produced by the microflora during fermentation were discussed in relation to expected cup quality. To conclude, an optimised and controlled fermentation seems promising for producing higher and more stable coffee quality.

Acknowledgement: This project was funded by Innosuisse (31983.1 INNO-LS) and done in collaboration with Kaffeemacher GmbH

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Coffee Poster 31

Solvent removal of the efluent of coffee fruit processing.

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Text This process involves a large volume of water to wash and depulp the fruits, resulting in an effluent with high concentrations of organic material and nutrients, which, if inappropriately discarded, have high pollution potential. The strategy of recirculating water in the processing units contributes to the reduction of the consumption and the effluent generated, it incorporates organic and inorganic material, changing its characteristics and

hindering its flow in the hydraulic system, where clogging may occur. It is necessary to remove part of the coarse settling solids into the processing unit. This removal can be done by industrialized solid separators, by homemade sieves or structures adapted for this purpose. This study aimed at quantifying the removal of waste from the coffee berry processing unit, throung a system consisting of settling boxes and sieves. A removal system of coarse settling solids made with three decanting boxes connected by PVC tubes and two static sieves tilted at 10% in the outlet of the third box was adapted. The experimental design got randomized blocks split in sections (4X5) subdivided in four replications, having the four collecting points in the sections (CP1, CP2, CP3 and CP4) and the five time periods (T10, T40, T70, T100 and T130 min.) Data were subjected to analysis of variance and the average results were compared to Tukey test at 5% probability. 39.420 L of Arabica coffee berries were processed and analyses of the concentration of total solids as well as measurements of electrical conductivity were performed, whereas the chemical analyses included the measurement of the hydrogenionic potential as well as the macro/micronutrients concentrations. The sensory characteristics of the beverage were analyzed using the methodology Specialty Coffee Association of America. We have concluded that: the recirculation of water for processing, came down from 2,2 L to 0,52 liters of water per liter of processed fruit; During the time of recirculation of wastewater from coffee; the concentrations of ST, EC, N, P, K, Ca, Mg, S, Cu, Zn, Mn, Fe and B increased, the pH decreased; the time of recirculation of water had no effect on the beverage quality of peeled grains.
[Ref1]
[Ref2]
[Ref3]

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Coffee Poster 32

Sensorial descriptors of robust coffee fermented by different forms of processing

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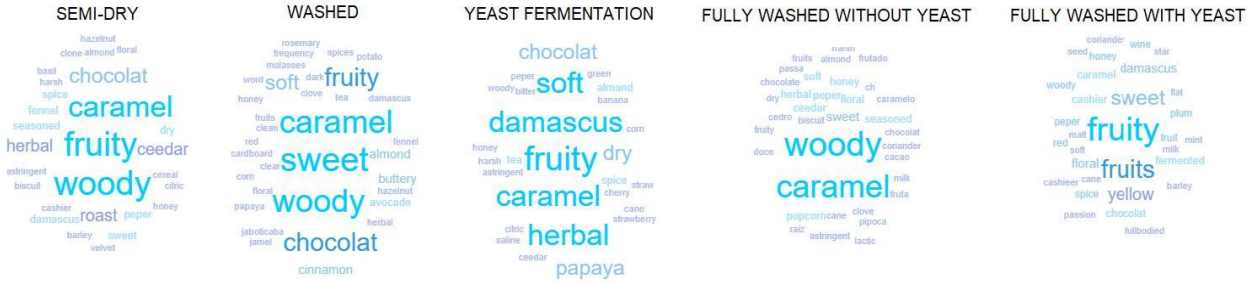
Poster session: **Coffee**

Text The production of special coffee / conilon coffee has been growing in the specialty coffee market. Thus, it is necessary to increase the sensorial quality, which can be do through post-harvest techniques, allowing greater process control, aiming at obtaining a product with special sensorial characteristics. This study evaluated different types of post-harvest processing, among them the inoculation of Saccharomyces cerevisiae as a start culture, with the objective of measuring the Q-Graders' sensorial descriptors. The experiment was conducted in a randomized block design with 5 replicates and 5 treatments: Semi-Dry, Washed, Yeast Fermentation, Fully Washed Without Yeast, Fully Washed With Yeast. In the sensory analysis, the Q-grader makes quantitative and qualitative analysis, in this the tasters describe the perceived aromas and flavors. In the Semi-Dry, Washed and Fully Washed Without Yeast processes, both without inoculation of Saccharomyces cerevisiae, the sensory attribute of woody is observed, this attribute indicates worst quality. In Yeast Fermentetion and Fully Washed With Yeast processing, both with yeast inoculation, the most used sensorial descriptors were: Fruity, yellow fruits, floral, papaya, damascus. Thus, it is observed that the sensorial attributes that indicate higher quality are potentiated with the yeast inoculation in the robusta / conilon Coffee.



Main sensorial characteristics of coffee submitted to different processing.

Coffee Poster 33

Chemical and structural properties of parchment from washed and natural coffees

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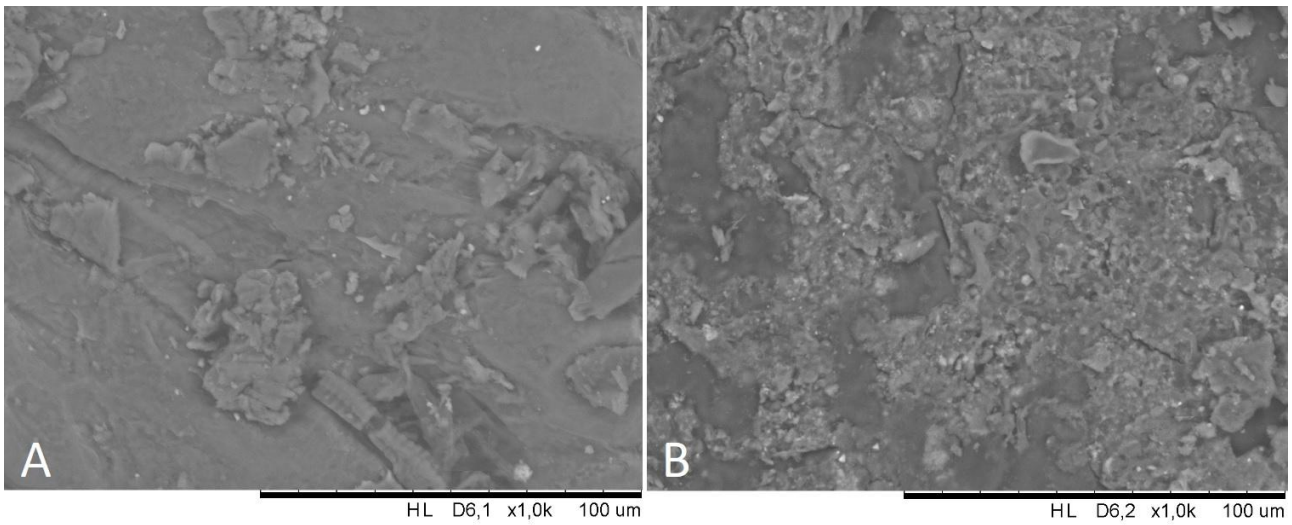
Text Coffee is one of the most consumed beverages worldwide. However, coffee cherries processing into coffee beans generates a plethora of waste, including parchment, whose destination remains without relevance. The two main coffee processing methods are wet (washed) and dry (natural) processes. In this sense, the investigation of these two types of

parchment will help to reduce coffee waste while generating a possibility of reuse within the concept of bioeconomy.
The present study main objective was to study the morphological and chemical properties of coffee parchment generated from washed and natural coffee processing. Samples of washed and natural processing were ground and sieved. The material was fractionated between the 20 and 80# mesh Tyler screens, corresponding to particle diameters between 180 and 850µm, respectively, according to the standard procedure for sample preparation reported by the National Renewable Energy Laboratory (NREL) (Hames et al., 2008).
Samples were subjected to the following analysis: surface morphology analysis by scanning electron microscopy, cellulose, hemicellulose, and lignin contents, total ashes, extractives, and total phenolics (Sluiter et al., 2008a,b, 2012).
Phenolics, cellulose, hemicellulose and lignin contents were higher in washed parchment, except for extractives content and total ashes, as displayed in Table 1.
Additionally, image analysis of coffee parchment obtained from washed and natural processes, revealed a more porous surface in the natural processed, as expected (Figure 1).
These attributes allowed the differentiation between parchment from washed and natural coffee. Their phenolic content and fiber contents are promising features to be used for different purposes, such as food, pharmaceutical or cosmetic industries.

Component	Washed process	Dry process
Acid gallic (mg.100-1)	423 ± 9	379 ± 13
Extractives content (%)	12 ± 0.5	21 ± 0.1
Cellulose content (%)	35 ± 0.2	26 ± 0.9
Hemicellulose (%)	22 ± 0.2	17 ± 0.2
Lignin (%)	21 ± 0.1	19 ± 0.6
Total ashes (%)	6 ± 0.01	7 ± 0.10

Total phenolics (acid gallic), cellulose, hemicellulose, lignin and total ashes content in coffee parchment obtained from washed or dry processes of coffee (average ± standard deviation)



Scanning electron microscopy of coffee parchment obtained from washed (A) dry (B)

processes of coffee cherries (x1.0k)

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Coffee Poster 34

Conventional and new consumption of coffee: Implications in antioxidant activity, bioactive compounds and aluminum content.

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Text Coffee is the most consumed non-alcoholic beverage around the world. This beverage is obtained from the infusion of the seed of the coffee fruit. In recent years, coffee consumption has increased significantly, due to the new extraction and consumption methods of this drink, such as espresso coffee capsules. Furthermore, in several studies the consumption of coffee and their beneficial effects on health have been published. The objective of this work was to investigate the effect of new coffee extraction and consumption methods on bioactive compounds composition and on antioxidant capacity of this beverage, and also to evaluate the possible migration of aluminum to coffee beverage in the new types of packaging and preparation of coffee. In order to reach this objective, the antioxidant capacity was determined, the analysis and quantification of bioactive compounds was carried out, the melanoidin content was determined and the aluminum concentration levels were measured in the different coffee samples obtained. The main results achieved in this study showed that the coffee beverages made with an espresso capsule machine presented a significant decrease in the antioxidant capacity and in the content of phenolic compounds compared to coffee beverages made with traditional mocha or filter coffee machines. The amount of chlorogenic acid, caffeic acid and caffeine were significantly lower in these new coffee brewing formats, and this fact could be related to the lower antioxidant activity perceived in capsules coffee samples. Regarding the aluminum content, it was found that the coffee beverage had lower concentrations of aluminum than tea, and surprisingly, decaffeinated coffees shown a

significant increase of the content of this metal. This study suggests that coffee made by capsule extraction method has fewer antioxidant compounds than coffee made by traditional methods. There is no increase in the concentration of aluminum in the coffee beverage made by the capsule method, but the industrial decaffeination methods of the green coffee bean should be studied deeply, due to the significantly elevated aluminum values of this type of coffee.

Coffee Poster 35

Coffee silverskin as active and plasticizing agent for starch films

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Text Coffee industry generates an excessive amount of residues caused by the high worldwide coffee consumption [1]. Coffee silverskin (CS) is a thin tegument of the coffee beans outer layer obtained during the coffee roasting. Its biomolecules-rich composition (cellulose, hemicelluloses, lignin, polyphenols, and fats [2]), makes it a sustainable feedstock for many applications as soil fertilization, cosmetic, dietary nutrition, and fuel production. On the other hand, environmental concerns derived from landfilled petroleum-based materials requires their replacement by alternative biobased and biodegradable formulations. Herein, starch, a polysaccharide with thermoplastic ability, has been used as precursor of bioplastic formulations [3], being most of the times blended with other constituents to improve their mechanical and barrier performance. In this work, different amounts of crude CS (1; 5; and 10% w/w) were incorporated into starch formulations and their effect on films mechanical, physicochemical and barrier properties was evaluated. Each film was produced through solvent casting. CS gave rise to yellowish starch films, being the color intensity directly related to the CS dosage. Starch/CS films showed an increased flexibility, surface hydrophobicity, antioxidant activity, UV radiation absorption, and low water vapor permeability, when compared to the pristine starch films. Therefore, the addition of CS to starch formulations revealed to be a promising strategy to produce UV-protective and antioxidant biobased films with improved mechanical and physicochemical performance, opening a new opportunity for CS valorisation.

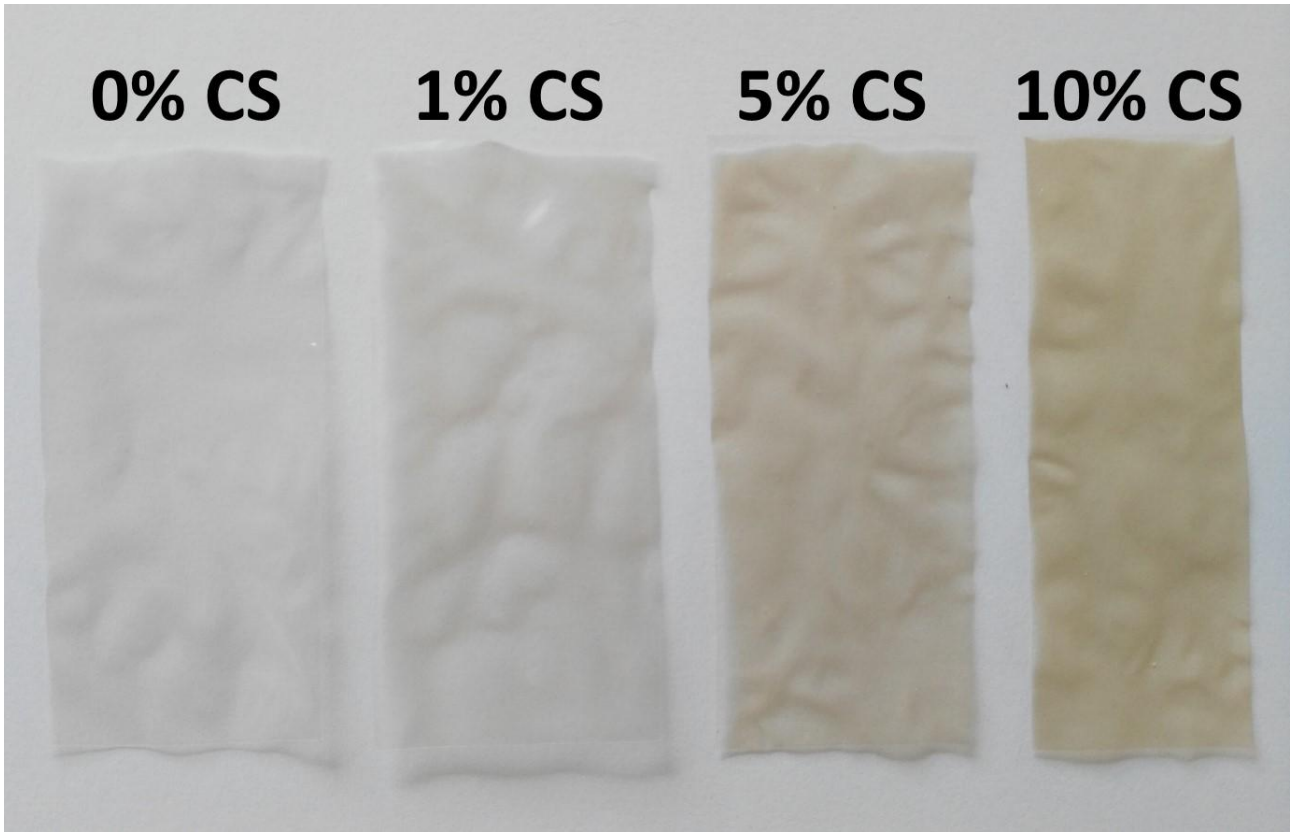

Fig. 1 - Starch films containing different CS amounts (0; 1; 5; and 10% w/w of the starch)

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Coffee Poster 36

Coffee Diseases and Their Management towards Sustainable Coffee Production: Experience from Ethiopian Coffee Industry

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Poster session: **Coffee**

Text The predominant role of coffee (*Coffea arabica* L.) in the Ethiopian economic, social and cultural dimensions dates back to several centuries as the country is the primary center of origin and genetic diversity of the plant. In its wild state, Arabica coffee is a forest plant restricted to the highlands of southwestern Ethiopia. Ethiopia can be considered as the biological and cultural home of coffee. Today, coffee is single source of income for 4.7 million small-holder farmers and about 15 million people directly or indirectly depends on coffee sector for their livelihoods. It contributes 29 – 31% of export earnings of the nation.

Despite its significant role, coffee production in Ethiopia has been threatened by a number of diseases. So far, fourteen registered coffee diseases are known to affect its production, among which coffee berry disease (C*olletotrichum kahawae*), coffee wilt disease (*Gibberella xylarioides*) and coffee leaf rust (*Hemileia vastatrix*) are economically important. The direct impact of coffee diseases includes rotting of berries, growth retardation of the trees, limited flowering and berry development leading to poor yield and unacceptable quality.

The development and release of 40 disease resistant varieties for commercial coffee cultivation in Ethiopia significantly reduced disease pressure. Moreover, Ethiopian coffee is mainly grown under shade trees, wherein optimum shade provides the right conditions for successful cultivation, by reducing daytime air and soil temperatures and preserving soil moisture. It also has key benefits for stabilizing natural ecosystems, including nutrient recycling, watershed preservation, enhancing pollination, temperature buffering, shelter from wind and heavy rainfall, and carbon storage. Therefore, coffee production system, diverse genetic resource availability and various growing conditions, with different cultivation practices in Ethiopia results in significant reduction of coffee disease pressure, while ensuring sustainable coffee production and environmental protection.

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Kifle Belachew, Demelash Teferi and Leggese Hagos. 2015. Coffee Thread Blight (*Corticium koleroga*): a Coming Threat for Ethiopian Coffee Production. Journal of Plant Pathology and Microbiology. 6(9): 1- 6

Coffee Poster 37

Evaluation of Coffee Genotypes against Coffee Wilt Disease (G*ibberella xylarioides* Heim and Saccus) at Southwest Ethiopia

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Text Coffee wilt disease (CWD) is the second major coffee disease in Ethiopia next to Coffee berry disease. It is essential to reduce coffee yield losses due to CWD in the

country through the development and use of genetically resistant coffee varieties. Longer-term prospects of successful management of coffee wilt disease depend principally upon employing resistant coffee cultivars. With this objective laboratory and field evaluations were conducted to screen some coffee genotypes against coffee wilt disease.

The study was conducted using 44 coffee accessions, which were collected from Bale area, South-eastern part of Ethiopia, in 2004, along with four controls (catimor J-19, catimor J-21, Gesha and 370). The laboratory and greenhouse studies were conducted using complete randomized design (CRD). Inoculation in green house started when the seedlings attained the full expanded cotyledon stage (seventy days after sowing). A representative isolate of *Gibberella xylarioides*, the causative agent of coffee wilt disease were multiplied and inculcated using standard procedure.

Disease severity or mean percent seedling death ranged from 0.00 to 89.96 %. Most of coffee accessions expressed resistant reaction against CWD pathogen compared to control treatments under laboratory test. Incubation period exhibited highly significant ($P<0.01$) variation among Bale coffee accessions. The mean incubation period among the tested accessions in days ranged from 0 to 77. Diseases symptom expression wasn't recorded on five accessions, namely, B64/04, B70/04, B104/04, B124/04 and B143/04. The result showed in lowest seedling death rate, long incubation period and high field survival rate of most accessions indicating resistant reaction to coffee wilt disease.

Thus present experiment implied that the potential of obtaining coffee wilt disease resistant coffee variety from these accessions. Moreover the study revealed alleles found in the coffee gene pool of Ethiopia may hold the key to the species long term survival providing the traits needed to cope with new diseases and climate change; this underline the importance of systematic evaluation of the coffee accessions and utilization of best performing accessions having high yielding, typical quality and disease resistant.

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Coffee Poster 38

The Potential of Oak Acorn as Coffee Substitute

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Text J. Zapp, K. Müller, Technische Hochschule OWL, Lemgo

The use of oak acorns in human and animal nutrition has a long tradition in many European countries. Furthermore, infusion beverages based on roasted acorns are

considered a promising alternative to coffee. However, the limiting factor in the consumption of acorn-based coffee substitutes is their high tannin content, leading to unpleasant taste attributes such as excessive bitterness and unattractive astringency. This study examines the influence of roasting conditions on the potential of oak acorns as coffee substitute. Roasting experiments were conducted on a small scale coffee roaster (Probatino, Probat-Werke GmbH, Germany).
Furthermore, we investigated the effects of different pretreatment procedures prior to roasting on the physical, chemical and sensory characteristics. Antioxidant properties and polyphenols composition were determined by means of HPLC. It could be shown that predrying results in a considerably improved roasting homogeneity. Soaking and leaching the acorns with water results in the partial depletion of polyphenols and improved sensory properties after roasting.

Poster session: **Cocoa**

Poster Cocoa

Cocoa Poster 01

Impact of total fat content on the sensory profiles of eleven *Theobroma cacao* cultivars

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Text Of the 4,000 *Theobroma cacao* accessions in international gene banks, only a few cultivated varieties (cultivars) have been characterized for their sensory attributes or total fat content [1][2][3][4]. None of these previous studies looked at cultivar total fat content (TFC) as a modulator of cocoa liquor sensory profiles. This highly-controlled study characterized eleven different *T. cacao* cultivars from up to three different harvests for their sensory properties, including taste, flavor, and mouthfeel attributes, to study how these attributes are modulated by TFC.

T. cacao cultivars were sourced from the Fundación Hondureña de Investigación Agrícola (FHIA) research institute in Honduras: Caucasia 39, CCN 51, EET 95, ICS 1, ICS 95, IMC 67, Pound 12, TSH565, UF 613, SCC-661, FHIA 74. Cacao pods were harvested from trees grown in the same plot in La Masica, Honduras to control for environmental and geographical factors. After cacao pods were harvested at maturity, cacao beans were fermented in a laboratory-scale fermenter and dried with a forced-air oven. Upon shipment to Penn State, beans were treated with infrared heat and then frozen to help in shell removal, winnowed, nib roasted and ground into cocoa liquor. The resulting 27 cocoa liquors were analyzed by a trained descriptive analysis (DA) sensory panel, which rated the intensities of each taste, flavor and mouthfeel attribute on 100-point line scales [5]. Total fat content (TFC) of all cocoa liquors was measured by time domain-nuclear magnetic resonance (TD-NMR) [6].

We found up to a 10% difference in TFC of these commonly-used cultivars and significant differences in cocoa liquor sensory profiles. Linear regression of TFC and statistically significant ($p < 0.05$) sensory attributes, as assessed by Analysis of Variance (ANOVA), demonstrate the modulating effect of TFC on several mouthfeel attributes, including astringency, mouthfeel, and viscosity.

This work provides insights on how fat can modulate astringency, a sensation that is often explained by a “lack-of-lubrication” mechanism. More broadly, this research contributes to the sensory science literature and demonstrates how a component found naturally in cocoa, such as fat, may counteract or synergistically affect flavor, taste, and mouthfeel components in a fatty food matrix.

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Cocoa Poster 02

Effect of trap colour on catches of brown cocoa mirid, *Sahlbergella singularis* Haglund (Hemiptera: Miridae), by sex pheromone traps in Cameroon

Hermine Claudine Mahot[1], Joseph Raymond Mahob[2], Hanna Rachid[3]
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Text The mirid bug, *Sahlbergella singularis,* is the most economically important insect pest of cocoa in Central and West Africa. In this study, the effect of colour on catches of male adult bugs in pheromone traps was investigated. The trial was conducted in seven villages in two localities, Ayos and Konye, located in Centre and South-West Regions of Cameroon, during two consecutive years. Catches were compared in white, green and purple rectangular sticky traps baited with the sex pheromone. Similar temporal patterns in trap catches were observed with all three colours, but catches were consistently higher in green traps (47% overall) than purple traps (32%). with lowest catches in white traps (21%). Measurements of reflectance of the trap materials showed typical peaks in both short and longer wavelengths for the purple, a typical peak at approximately 530 nm for the green and reflectance peaks exceeding 100% of the incident light in the 400-450 nm range from the white, presumably due to the presence of optical brighteners. This is an important result for development of pheromone traps for monitoring and control of cocoa mirids in that white traps have generally been used in previous work.

Cocoa Poster 03

Flavor of Pseudo-Cocoa Liquor: Effects of Polyphenols, Fat Content, and Training Method

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Text Current theories of astringency propose that this sensation is a result of delubrication in the oral cavity due to precipitation of salivary proteins [Reference1] [Reference2]. Astringency, commonly described as a drying or puckering sensation, is a main driving factor for rejection of certain foods [Reference3] [Reference4]. Previous studies have shown that fat plays a role in moderating astringency in foods. To investigate the role polyphenols and fat play in astringency perception, we used modified cocoa powders to produce pseudo-cocoa liquor systems that were rated for taste and flavor attributes on generalized Labeled Magnitude Scales by semi-trained consumers. Our results show significant differences among the cocoa liquors resulting from acetone-water extraction of free polyphenols (Figure 1) and fat content variation (Figure 2). No significant differences resulted from training with oil- vis-à-vis water-based reference solutions.

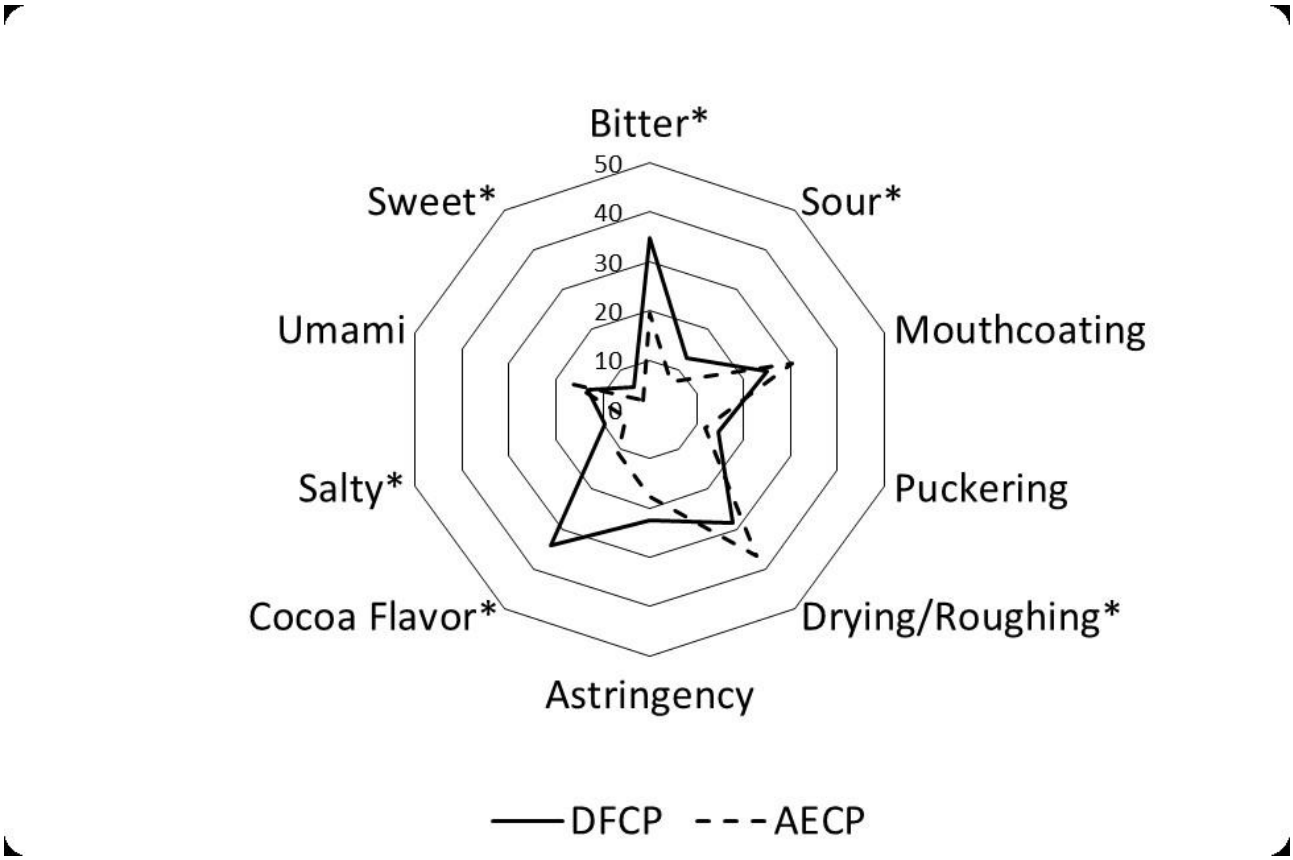

Figure 1: Mean attribute intensity ratings for defatted cocoa powder (DFCP) and acetone extracted cocoa powder (AECP). Attributes that are significantly different by t-test are marked with (*; $p < 0.05$).

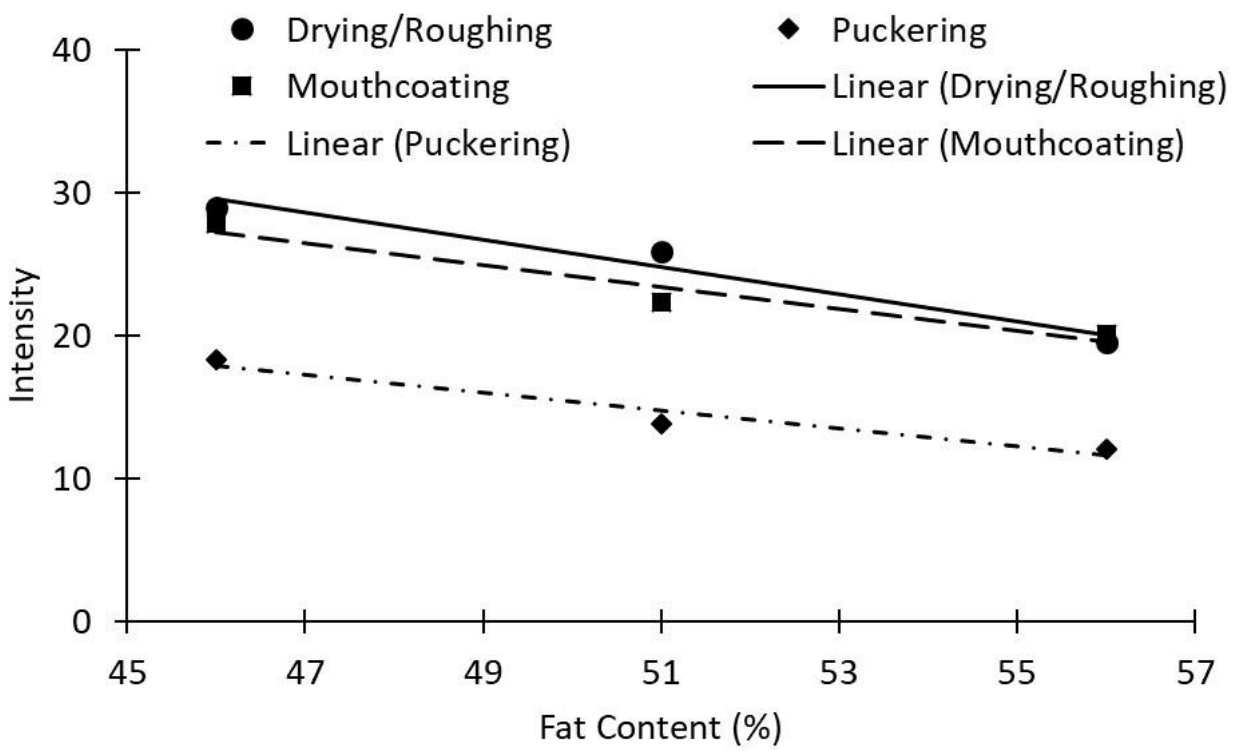

Figure 2: Mean attribute intensity ratings vs. fat content. Attributes shown have significant linear trend by regression analysis.

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Cocoa Poster 04

Application of Napping and SensoGraph procedure on sensory characterization of raw chocolate and cocoa beans

Giuseppe Zeppa[1], Olga Rojo-Poveda[1], Livia Reguengo[1], Chiara Roggero[1], Marta

Poster session: **Cocoa**

Bertolino[1], Daniela Ghirardello[1], David Orden[2], Letricia Barbosa-Pereira[1]
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Text Raw chocolate is a new type of chocolate produced from minimally-processed ingredients and using temperatures below 42°C during the all transformation processes, comprising fermentation and drying and avoiding the roasting step. It is made from raw-processed cocoa beans and cold pressed cacao butter. Also, several “raw” sweeteners, such as coconut sugar, agave or maple syrups, xylitol or stevia, may be used as replacers of cane or beet sugars that are not allowed.
The aim of this work was to define the sensory characteristics of nine cocoa beans (three raw, three traditionally fermented, dried but unroasted, and three roasted) and eight chocolates (four raw and four traditionally produced) from different origin applying two Projective Mapping procedures: Napping and its geometric counterpart SensoGraph[1]. With these techniques, it is possible to perform a fast sensory profiling based on a consensus map integrated by weighted connections between samples. Sensory analyses were performed by nine trained tasters during two sensory sessions. The results were preliminary subjected to a Multiple Factor Analysis in order to highlight the sensory profile of each sample. Significant differences for chocolates and beans according to their origin and treatment were obtained. The fifty identified sensory descriptors were divided into fifteen classes. The descriptors with a high sample discriminant capacity were spicy, fermented and dried fruits. The sample tablecloth positions provided by each taster were later clustered using the Gabriel graph and then merged into a single map using the algorithm of Kamada and Kawai, which provides relationships between samples. The results have underlined the interconnections between samples helping to understand the relation between the sensory profile and ingredients and/or process and/or origin inside the samples. In particular, the origin was a prominent factor in the sample discrimination while the treatment (raw or traditional) proved to be a weak factor.

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Cocoa Poster 05

Aroma fingerprints as an identity card of different comfort foods: cocoa and coffee products

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Text Tools to trace sustainable cocoa and coffee productions are necessary in particular in view of climate changes and political situations of production countries. Fingerprinting is a good approach to monitor and authenticate food[1]Food authentication is often based on the degree of similarity of the fingerprint between the investigated sample and a representative reference. This operation is known as food ‘Identitation’[2-3] and its reliability depends on its

correctness.
In this study, HS-SPME-GC-MS combined with chemometric tools was applied to a set of cocoa samples of different origin and commercial coffee blends to investigate their aroma chemical fingerprinting.
One hundred samples of cocoa beans and paste and twenty samples of two commercial coffees with characteristic sensory notes were analysed with the above approach.
*U*ntargeted and *T*argeted (*UT*) *fingerprinting* data from cocoa samples were used to extract relevant chemical information for origin discrimination and to cross-validate them in incoming raw material and in intermediate chain products. Results indicate coherent clear clustering of samples in function of their origin both in raw beans and in the cocoa pastes. Prediction of cocoa beans classification with the untargeted fingerprint on an external validation set has resulted in a very distinctive clouds with global model sensitivity of 75% and specificity of 100%, while the clouds of cocoa paste samples are less defined, but with a high specificity and sensitivity (78%).
Targeted fingerprints of coffee samples were used to study the encrypted information supporting their discrimination. PCA, PLS-DA and SIMCA were applied to the data set deriving from the analysis with the two methods. Information deriving from the aroma chemical fingerprints of coffee samples highlights the ability of the volatile fractions to discriminate the two set of commercial coffees based on chemical components that can be related to the different sensory notes and, within the blend, to the expiring date. The ability of PLS-DA to classify the two commercial blends based an external test set of coffees was 100%

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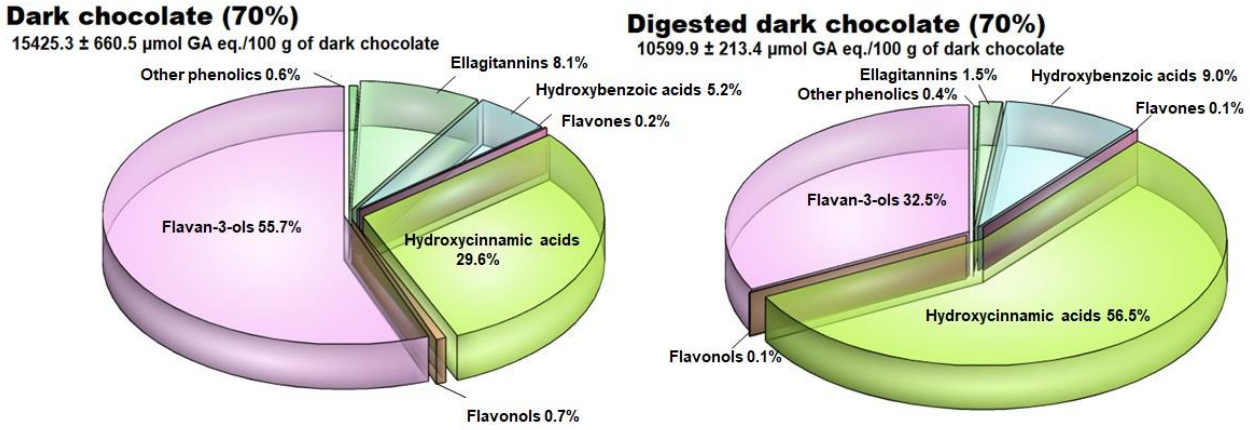
Cocoa Poster 06

Anti-proliferative activity and cell metabolism of bioaccessible dark chocolate phenolic compounds

Serena Martini, Angela Conte, Davide Tagliazucchi
University of Modena and Reggio Emilia, Department of Life Sciences, Reggio Emilia, Italy

Text Recently, chocolate and its polyphenols have gained much more attention for their important role in human health, especially in the treatment of cardiovascular diseases (1). However, a thorough evaluation of chocolate phenolic profile is still lacking. This study provides a comprehensive characterisation of dark chocolate phenolic profile, using non-targeted mass spectrometry identification (2). This approach allowed a tentative identification of 158 individual phenolic compounds: 67 were identified for the first time in chocolate, among these 38 were detected for the first time in cocoa and cocoa-derived products in general. Ellagitannins, never reported in cocoa or chocolate, represented about the 10% of the phenolic profile of dark chocolate. The enrichment of dark chocolate

with Sakura green tea leaves or turmeric powder influenced and modified the phenolic profile, leading to an increase in the phytochemicals concentration. In this way, this functionalization might maximize the beneficial effect of cocoa consumption as well as reducing the amount of sugars and calories introduced with chocolate. The bioaccessibility of phenolic compounds after *in vitro* gastro-intestinal digestion was further evaluated.



Percentage incidence of phenolic classes of dark chocolate identified and quantified after chemical extraction and in vitro digestion

Antioxidant activities increased after the gastric step and rose further at the end of the digestion. Furthermore, *in vitro* digested phenolic-rich fractions were tested for their anti-proliferative effect against two human colon adenocarcinoma cell models (Caco-2 and SW480). The metabolic fate, the major bio-transformation in the molecular structures and the impact on anti-proliferative activity of chocolate bioaccessible phenolic compounds were evaluated by mean incubation with the two cell lines. Our results provide evidences that Caco-2 and SW480 cell lines are able to metabolize phenolic compounds by means of phase I and II enzymes. The major metabolic reactions displayed pathways for phase I de-esterification and hydroxylation and phase II glucuronidation, sulphation and O-methylation. Simple hydroxycinnamates were released from quinic and aspartic-conjugates.

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Cocoa Poster 07

Dark chocolate protects meat lipids from oxidation during *in vitro* gastro-intestinal digestion

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Poster session: **Cocoa**

Text Recent studies suggest that meat consumption may be related to the onset of cardiovascular diseases and colorectal cancer (1). There is evidence that this risk may not be caused by meat *per se*, but may reflect high-fat intake, and/or compounds generated during cooking and processing (1-2). In particular, the oxidative phenomena involving polyunsaturated fatty acids occurring during meat cooking and gastro-intestinal digestion can result in the formation of lipid oxidation products, such as lipid hydroperoxides and advanced lipoxidation end-products, which might adversely affect human health (3). The aim of this work was to evaluate the effect of dark chocolate on the formation of lipid hydroperoxides during co-digestion with turkey meat. *In vitro* digestion of turkey meat resulted in an increase in lipid hydroperoxides formation. The addition of 1 g of dark chocolate to 5 g of turkey meat caused an inhibition in the lipid hydroperoxides formation by 56% at the end of the digestion. Indeed, dark chocolate showed a concentration-dependent inhibitory effect during co-digestion with turkey meat. Phenolic compounds were then extracted from chocolate and co-digested with turkey meat. The addition of the polyphenol-rich extract to turkey meat (at the same concentration as found in 1 g of chocolate) caused an inhibition in the lipid hydroperoxide formation of 79%. Once again, the inhibitory effect was concentration-dependent. These results suggest that polyphenols were responsible for the inhibitory effect. Successively, dark chocolate phenolic compounds were identified and quantified by using liquid chromatography coupled with mass spectrometry. Flavanols was the most abundant phenolic class and, in particular, epicatechin and procyanidin dimer B were the main phenolic compounds present in the analyzed dark chocolate. Co-digestion with epicatechin (at the same concentration as found in 1 g of chocolate) caused an inhibition in lipid hydroperoxides formation of 100%. In conclusion, our results suggest that the consumption of dark chocolate after a turkey meat-based meal may inhibit the formation of lipid hydroperoxides and that chocolate phenolic compound were responsible of the observed effect. Inhibition in hydroperoxide formation during digestion may lead to a reduction in their absorption at intestinal level, possibly protecting from the onset of chronic diseases associated with meat consumption.

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Poster session: **Cocoa**

Cocoa Poster 08

Roasting kinetics of cocoa nibs

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Wageningen University and Research, Wageningen University and Research, Wageningen, Netherlands

Text Cocoa beans are a commodity of huge economic significance in the world and the key raw material for chocolate manufacturing. Their seeds, commonly known as cocoa beans, are removed from the pods to be fermented and dried before commercialization. In cocoa Industry, the husks of the beans are often removed prior to roasting by exposing them very quickly to an extremely high temperature. The collected cocoa beans without husks are termed nibs. Very important quality and sensory characteristics of the final product depends on the next step, the roasting. During this procedure, the temperature rises to between 110 and 150°C, which is then maintained for between 15 min to 2 h until the moisture content is below 2%. This process helps to develop deep brown colours, characteristic chocolate flavours and reduce the bitterness and the astringency of the beans.

Beside these desired properties, the roasting process comes with certain food safety concerns caused by neo-formed contaminants produced in foods during heat treatment, such as 5-hydroxymethylfurfural (HMF). HMF is a furanic compound which forms as an intermediate in the Maillard Reaction and is considered as an indicator of heat damage during thermal process. HMF has been also reported as cytotoxic at very high concentrations.

Roasting effects of different varieties of cocoa beans have been widely studied using convective ovens in laboratory scale which can represent the industrial way in terms of long time roasting (LTR) and temperature. The fluidized bed roaster is a hot-air short time roasting (STR) method that has shown positive results in quality and nutritional characteristics in coffee. The objective of this research is to study the roasting kinetics of cocoa nibs by LTR and by STR at different temperatures in terms of moisture content, aw, colour and the formation of HMF. Cocoa nibs were roasted at 110, 120, 130 and 140°C for 30, 40, 50 and 60 min and for 2.0, 2.5, 3.0 and 3.5 min for LTR and STR respectively. Forastero cocoa beans from Ivory coast was used to obtain the nibs. Moisture content was determined by gravimetric method, aw by dew point hygrometer, colour by digital image analysis and HMF by HPLC. The upper is the temperature, the higher the HMF concentration. The time of roasting at any temperature is proportional to the brown colour intensity. Kinetics data of cocoa nibs roasting will be presented.

Cocoa Poster 09

Comparison of UHPLC-MS and DI-FTICR-MS for analysis of Cocoa Bean Fermentation

Britta Behrends, Roy N. D'Souza, Nikolai Kuhnert

Poster session: **Cocoa**

Jacobs University Bremen, Bremen, Germany

Text The classification of cocoa based on quality has been a major challenge in the chocolate industry, which has partially been addressed using several information-rich techniques, such as LC-MS and Near Infrared (NIR) spectroscopy. These methods have been shown to not only assess the degree of fermentation of a cocoa sample, but also discriminate between its origin. A time-consuming chromatography step has always been preferred before MS analysis of a sample as it mitigates several disadvantages of direct MS analysis, most importantly, matrix effects of ion enhancement and ion suppression. While direct infusion MS makes absolute quantification of target analytes generally unfeasible, recent advances in ESI sources have made significant reductions in matrix effects, by not only allowing for lower sample dilutions, but also smaller drop sizes. We have analyzed high-resolution direct-infusion FTICR-MS and UHPLC-MS data of a daily time course of fermented cocoa beans (0 – 7 days) in order to assess the equivalence of these methods in an untargeted fashion. Integrating an LC-MS pipeline within the cocoa processing chain has been avoided in the cocoa industry due to the requirement of expensive instrumentation, skilled operators, as well as the time-consuming sample preparations and analyses involved in LC-MS routines. A direct infusion MS would provide a rapid, cost-effective, information rich (orders higher than NIR), and convenient method to assesses cocoa quality within the industry.

Cocoa Poster 10

Effect of main Ivorian cocoa producing and fermenting regions on the formation of flavor coumpounds in raw cocoa and the sensory quality of chocolates produced thereof

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Text Cocoa fermentation is the core of post harvest processing beans. The purpose of fermentation is to improve and establish odor and distinctive flavor of chocolate. The factors that influence the quality of fermented cocoa beans and the development of volatile profile chocolate include probably the region of fermentation performance. This research purposed to determin the effect of main Ivorian cocoa producing regions on the flavor compounds fromation in cocoa beans and the organoleptic traits of chocolate produced thereof. The fermentation of cocoa beans extracted from the same cocoa pods variety was carried out in five mains cocoa regions according to the plastic box technique and the same duration. Analysis of flavor compounds of fermented cocoa beans were done by SPME-GC/MS technic and twelve judges who had been previously selected and trained,

performed sensorial analyses of the chocolates. Results shows 71 aroma compounds belonging to to 12 chemical families. The concentrations of cocoa beans in alcohols, aldehydes, ketones, esters, acids and pyrazines varied from 47.20±08.02 to162.00±28.52, 51.55±8.69 to 86.84±17.93, 119.27±32.30 to 242.27±69.51, 638.41±184.06 to 1695.23±208.29, 1244.55±307.43 to 2895.13 ± 458.34, 60.85±42.51 to 666.31±315.33 $\mu g.g^{-1}$ respectively. We measured highest contents of main flavor compounds such as pyrazines, esters, alcohols and ketones in cocoa beans that were fermented in Daloa. Fermented Cocoa beans from San Pedro presented low contents in acid, ester, alcohol and ketone compounds. Hence, chocolates resulting from cocoa fermenting in San Pedro were less acidic and astringent but with good overall quality traits such as sweet and dried fruits aromas. These differences in flavor compounds contents of beans and sensory quality of chocolate produced thereof could be due to the diversity of specific and endemic yeasts involved in the cocoa fermentation.

Cocoa Poster 11

Thermal degradation of asparagine containing cocoa peptides

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Text During thermal food processing, the chemical composition of the raw material changes dramatically with myriad new compounds being formed [1]. In most cases, the thermal decomposition products are desirable contributing significantly to the organoleptic properties of the final food product. However, food heat treatment is also considered responsible for compounds formation with adverse health effects such as polyaromatic hydrocarbons. The decomposition of asparagine residues, in the presence of reducing sugars, has been reported to form acrylamide, a possible carcinogen in foods [2]. We have investigated the thermal decomposition of 14 asparagine containing peptides found in cocoa. The peptides were heated under cocoa roasting conditions in the presence of sugars. Acrylamide formation has been examined and chemical mechanisms have been proposed.

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Cocoa Poster 12

Influence of *in vitro* gastrointestinal digestion on phenolic compounds and antioxidant activity of cocoa melanoidins

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Poster session: **Cocoa**

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Text The aim of the present study is to compare the effects of simulated gastric and intestinal fluid digestion on the composition and content of phenolic compounds of melanoidins isolated from cocoa beans of different *Theobroma cacao* L. groups, and to explore the change of phenolic compounds caused by simulated digestion on antioxidant activity. Samples were prepared using *in vitro* model simulating the physicochemical (pH, temperature and bile salts) and biological (pepsin and pancreatic enzymes) gastrointestinal conditions. The phenolic profiles of samples were evaluated by the determination of total phenolic content and UHPLC-DAD-ESI-HR-MS/MS analysis. The antioxidant properties were measured with four different methods after each digestion step. Digestion of cocoa melanoidins with simulated gastric and intestinal fluid causes the release of bound phenolic compounds, mainly flavonoids and phenolic acid derivatives. The highest amounts of phenolic compounds were observed after digestion with simulated intestinal fluid. The simulated intestinal fluid treatment significantly reduced the 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical scavenging activity of cocoa melanoidins. However, both simulated gastric and intestinal fluid digestion increased their ferrous ion chelating ability and ferric reducing antioxidant capacity (FRAP). The release of phenolic compounds and antioxidant activities of cocoa melanoidins were found to be dependent on the digestion phase, the phenolic group and the antioxidant activity assay. The results of this study indicate that melanoidins isolated from cocoa beans and products obtained as a result of its processing are a good source of bioaccessible phenolic compounds and can be considered as functional food ingredients or dietary supplements.

Cocoa Poster 13

Application of mathematical modelling for cocoa bean spontaneous fermentation processes

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The process of cocoa bean fermentation represents a key step in cocoa processing in terms of development of chocolate's flavour and aroma. Despite its high industrial relevance, there are hardly any attempts of constructing a mathematical model of cocoa bean fermentation. Here, a quantitative model of cocoa bean fermentation is constructed based on available microbiological and biochemical knowledge. The model is formulated as a system of coupled ordinary differential equations (ODE) with two different types of state variables: (1) Metabolite concentrations of glucose, fructose, ethanol, lactic acid and acetic acid, and (2) Population sizes of yeast, lactic acid bacteria and acetic acid bacteria. In total, the model comprehends 25 unknown parameters that were estimated using the Markov chain Monte Carlo No-U-Turn sampler RStan. The ODEs were specified and solved by the built-in mechanism of `Stan'. Thereafter, we demonstrate that the model can quantitatively describe existing fermentation series and that the estimated parameters can be used to extract and interpret differences in environmental conditions. Thus, the proposed model is a valuable tool towards a mechanistic understanding of this complex biochemical process.

Cocoa Poster 14

Origin and varietal based diversity of proteomes and peptidomes in *Theobroma cacao*

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Text A comprehensive analysis of cocoa proteins and peptides from unfermented and fully-fermented beans, respectively, from a wide range of geographic origins was carried out to catalogue systematic differences based on their origin, variety as well as fermentation status. Protein quantities as well as their profiles derived from two-dimensional gel electrophoresis, showed striking differences for unfermented beans depending on their geographical origin. Degradation products of proteins produced during fermentation are believed to be the key precursors of a range of Maillard reactions that deliver the characteristic flavor and aroma of cocoa and chocolate. We have utilized UHPLC-ESI-Q-q-TOF to relatively quantify and annotate the cocoa oligopeptides based on their characteristic fragmentation pattern. With the identification of more than 800 unique oligopeptides across 25 different samples, we reported the largest collection of cocoa oligopeptides ever observed and identified. Our study enabled a comprehensive analysis of the attributes that characterize storage protein degradation in cocoa during microbial fermentation. We demonstrated that major differences in protein content of non-fermented cocoa beans were predominantly attributed to the geographic origin in terms of continental regions. Furthermore, our study suggested that beans can be classified based on their fermentation status using their peptide profile. However, unlike the protein profile, this classification was independent of bean origin and rather depended on the fermentation method applied in the country of origin ultimately indicating diversified proteolytic activities. Hence, the peptide profile of a bean can serve as a scientifically reliable indicator for the degree of fermentation. We propose to use the observed peptide number as a novel chemical classification tool to assess cocoa quality in terms of fermentation status.

Cocoa Poster 15

Adding beneficial polyphenols and lipids from pistachio to chocolate products

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Text Nuts are nutrient foods with complex matrices, rich in unsaturated fats and other bioactive compounds and provide rich sources of food lipids, up to 75% on a wet weight basis and the oil contained 98.4% triacylglycerol rich in unsaturated and polyunsaturated fats [1].
Pistachio (*Pistachio Vera L.)* is one of the most important tree nuts. Iran, USA and Turkey are the main producers of pistachio nut and the global production increases steadily. A tendency for increasing nut consumption is attributed to their nutritional components,

including sterols, vitamins, minerals, fatty acids and phenolic compounds and their antioxidant properties. Moreover, they are appreciated for benefit on human health since they regulate cholesterol levels and have a positive effect on cardiovascular disease for their antioxidant concentration. Composite products such as pralines, filled chocolates dominate the chocolate confectionery market, where a chocolate coating layer is in direct contact with a fat-based cream, biscuit, nut or nut paste [2].
Nowadays, commercial dark chocolate typically contains between 25% and 40% cocoa butter with the rest being variable amounts of sugar and solid cocoa powder. The triacylglycerol in nut-based fillings, such as triolein (OOO), LOO, LLO, POO, and SOO, where L stands for lauric acid, are predominately liquid at room temperature. [3].
In this work, pistachio samples were analysed by HPLC-ESI-MS and their chemical profile was compared to cocoa lipid profile. Our final aim was to examine whether lipids from nuts are complementary to cocoa butter.
Our results show that both plants demonstrate similar lipid profile, which bring a new application in cocoa industry.
Key words: pistachio health benefits - cocoa lipids – nut chocolate – lipid profile of pistachio

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Cocoa Poster 16

Effect of chemically driven transformation of cocoa seeds to cocoa beans in the generation of phenolic compounds

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Text Chemically driven transformation has recently been carried out in order to achieve higher control over the seeds-to-cocoa beans transformation process, with the aim to improve the desired flavor profile in cocoa products [1]. To assess the impact of organic acids on polyphenols levels of the seeds during their transformation to cocoa beans, these were subjected to several chemically driven transformations in a shaker incubator at 37,5 °C and stirring at 150 rpm, employing acetic, lactic and citric acids at a concentration of 15 g/L, without microorganisms. After 6-days incubation, solid-liquid extractions on both treated and untreated cocoa beans, were carried out with several solvents at increasing

polarity order. The respective chemical profile of each resulting extract was then analyzed by RP-HPLC-ESI-MS. The chromatographic data were subsequently used to be analyzed upon multivariate statistical analysis in SIMCA 13.0 program. The main compounds identified in the obtained with ethyl acetate extracts were procyanidin C1, procyanidin B, epicatechin and caffeine. However, according to the organic acid used in the transformation, some epimers of procyanidin B and procyanidin C1 were additionally identified, which were not found in the untreated cocoa seed. Results indicate that the chemically driven transformation could be used to increase the concentration and/or number of polyphenols in cocoa beans raw material, which have demonstrated interesting beneficial effects in human health [2].

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Cocoa Poster 17

Synthesis of symmetrical and unsymmetrical triacylglycerols (TAGs) for quantification of triacylglycerols (TAGs) in cocoa butter and coffee oil.

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Text A number of symmetrical and unsymmetrical triacylglycerols (TAGs) have been synthesized using Hassner esterification method with further modifications in chemical reagents. For synthesis of unsymmetrical triacylglycerols (TAGs) multistep chemical synthesis has been developed.
These cocoa related TAGs, have been used as authentic standard for food absolute quantification.
Triacylglycerols (TAGs) composition in cocoa butter has been shown to differ significantly according to the origin and ways of fermentation. Triacylglycerols (TAGs) profile in coffee oil can additionally be employed to differentiate between Robusta and Arabica coffee beans. Triacylglycerols (TAGs) profiles have been quantified in cocoa butter and coffee oil by our group by HPLC-ESI. Non aqueous reversed phase liquid chromatographic methods have been developed for cocoa and new classes of TAGs have been identified in cocoa.
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Cocoa Poster 18

Solid versus liquid: A new approach to aroma profile testing of cocoa liquors

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The identification of flavor and aroma relevant components in raw cacao, cocoa liquors and cocoa based products is a key point in the field of cocoa research. There is strong evidence that the genotype, the fermentation conditions, and the post-harvest processing have a significant impact on the flavor development in cocoa.
To further enlighten these correlations the CORNET project “Quality improved Cocoa and Cocoa-based Products with Flavor Profiles on Demand – From Farm to Chocolate Bar” (*Federal Funding Advisory Service on Research and Innovation* (IGF) Project No. 169EN/2 funded by the *Federal Ministry for Economic Affairs and Energy* (BMWI) through the *German Federation of Industrial Research Associations* (AiF) represented by the *Research Association of the German Food Industry* (FEI)) aims at investigating the sensory profiles of both, established and novel cocoa genotypes from selected Peruvian cocoa farms that are exposed to different fermentation conditions and post-harvest treatments. In this context, one of the research approaches in the field of cocoa sensorics deals with the question if it is possible to replace the hitherto prevalent tasting of liquid

cocoa liquors with a better standardized tasting of molded liquors.
Methods: To ensure a high level of validity and reliability of the results, a sensory panel was recruited and trained on the basis of a newly developed standardized protocol for molded liquors. Established protocols normally focus on the evaluation of liquid liquors only. Three different cocoa liquors of both, molded and liquid samples are presented to the panel and are evaluated in profiling sessions as well as in discrimination tests.
Expected Results and Discussion: The project aims to generate a more easy sensory evaluation procedure and a related assessment scheme for cocoa and cocoa products. These tools can be used to guarantee a quality-based cocoa and chocolate production with flavor profiles on demand from bean to chocolate. The new protocol enables experts to perform sensory analyses all over the world with a higher comparability between the results. A key factor for that is, besides a standardized protocol, the tasting of molded cocoa liquors.

Cocoa Poster 19

Occurrence of filamentous fungi during cocoa post-harvest processing in Honduras and potential aflatoxin production

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Text Cocoa beans are the major raw material for chocolate production. The fermentation of cocoa beans is the first step in cocoa post-harvest processing and is characterized by a succession of yeasts, lactic acid bacteria (LAB), and acetic acid bacteria (AAB) [1]. Dependent on surrounding conditions e.g. humidity due to rainfall, filamentous fungi are often observed with predominance in the late phase of fermentation, during drying, and storage. Fungal presence in cocoa is generally regarded as undesirable and often related to the formation of off-flavors, spoilage, and mycotoxin accumulation [2]. In this study, 516 filamentous fungi were isolated from cocoa beans during fermentation (n=29), drying (n=216), and storage (n=271) in Honduras. A selection of 206 isolates was identified using a multiphasic sequencing approach. The remaining 310 were indirectly identified by means of macroscopic characterization after growth on different fungal agars. In an overview, filamentous fungi of the genera *Aspergillus* (n= 218, corresponding to 42.2 %), *Penicillium* (n=85, 16.5 %), *Lichtheimia* (n=69, 13.4 %), and *Fusarium* (n=10, 1.9 %) were identified concomitant with other genera (n=26, 5%), yeasts (89, 17.2 %), and a share of 3.7 % unidentified isolates (n=19). 148 isolates of the section *Flavi* were further characterized regarding aflatoxin production potential. After growth on yeast extract sucrose agar with β-cyclodextrine and sodium deoxycholate (YCSD) and coconut cream agar (CCA), 41 (28%) *Aspergillus* strains showed aflatoxin production on both agars, 24 (16%) only on one agar, and 46 (31%) on no condition. The remaining 61 (41%) isolates showed no aflatoxin production on at least one agar, whereas the second agar did not reveal clear results. A PCR approach targeting *alfP*, *alfO*, and *alfD* confirmed the phenotypic production of aflatoxin in 8 strains of 10 *A. flavus/A. oryzae* genotypically. This study revealed a high risk of aflatoxin producing fungal strains present during post-harvest processing of cocoa

beans. A potential production of further mycotoxins is currently under investigation.

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Cocoa Poster 20

Supervised and Unsupervised Classification of Cocoa Bean Origin and Processing using LCMS

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Text Identifying the country-of-origin of cocoa beans (*Theobroma cacao*) based upon their chemical composition holds potential for multiple applications in the chocolate industry: from development of single origin chocolates to fine tuning of desired flavors [1,2]. Here we use a large data set comprising of 213 samples of fermented (113) and unfermented (77) cocoa beans, as well as selected cocoa liquors (23), sourced from various countries (8) and belonging to different stages of biochemical transformation in the cocoa processing pipeline. We study the classification of bean origin using an unsupervised and a supervised method of learning, PCA (Principal Component Analysis) and LDA (Linear Discriminant Analysis), respectively. We observe that while PCA can only provide a limited separation in bean origin (see [1]), the separation becomes better using LDA as a supervised method, as expected. In this case, a simple filtering criterion of compounds based on a Gaussian distribution of intensities (Gaussian Feature Stability Requirement) dramatically enhances the classification of the samples according to bean origin. Essentially this method is capable of removing conflicting signals from the LCMS data, which limit the classification. In this form, the supervised learning holds the possibility to identify potential markers of a specific origin.
Going from classification to prediction, we show how well the country can be predicted from the LCMS data for a sample. Furthermore, we employ a range of methods for extracting the minimal set of compounds required for such a prediction.
On a more theoretical level, we test the classification using a binarized version of LC-MS profile representing presence (1) or absence (0) of compounds rather than their actual intensity values. We observe that the mere presence or absence of compounds can still provide a good amount of classification, which suggests alternative strategies of biomarker search.

Poster session: **Cocoa**

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Cocoa Poster 21

Identification and profiling of human urinary metabolites using UHPLC-MS and ^{1}H NMR after cocoa consumption

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Text *Theobroma cacao* is a rich source of dietary polyphenols[1] and is associated to numerous health benefits [2]**.** However, very limited studies have been conducted to determine the fate of cocoa phenolics and their metabolites within the human body after ingestion. The current study aimed to investigate the rate and extent of gut bacterial and liver metabolism of cocoa phenolics after human intake. A feeding trial experiment was conducted, where recruited volunteers (*n* = 8) consumed 200 g of cocoa (for two days) and their urine and fecal (*n* = 4) samples were collected at different time points over 48 h period. The phenolic metabolic profile including both bacterial and phase II conjugation were determined by combination of UHPLC-MS/MS & ^{1}H NMR techniques.
Moreover, an untargeted & targeted metabolomics approach using principal component (PCA) and partial least squares discriminant analysis (PLS-DA) were applied to the LC-MS and NMR data, to allow the identification of significant dietary biomarkers and to visualize the discriminatory putative biomarkers between the metabolomes of various volunteers. The results herein indicated a significant increase of catechin bacterial metabolites (i.e. lactones and phenylacetic acids) and their phase-II conjugated forms detected in urine could be the contributing biomarkers of cocoa health benefits. Additionally, modulation of human primary and secondary urinary metabolites was investigated.

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Poster session: **Cocoa**

Cocoa Poster 22

Selecting stress-tolerant anti-fungal lactic acid bacteria-yeast co-cultures to limit growth of mycotoxigenic moulds during cocoa bean fermentation

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Text The spontaneous fermentation of cocoa beans is dominated by a succession of yeasts, lactic acid bacteria (LAB), and acetic acid bacteria. The occurrence of filamentous fungi is of great concern due to product quality deterioration and the potential accumulation of mycotoxins in cocoa beans. The aim of this study was to develop anti-fungal LAB-yeast co-cultures that limit the growth of filamentous fungi during cocoa bean post-harvest processing. LAB-yeast co-cultures were selected from 26 anti-fungal LAB and 13 anti-fungal yeast strains based on stress tolerance, growth inhibition of mycotoxigenic filamentous fungi, and their influence on cocoa bean quality in application trials. LAB strains were able to grow in cocoa pulp simulation medium in the presence of 10% ethanol, 0.7% lactic acid, and 1.4% acetic acid and at 45 °C and yeasts showed high tolerance towards 10% ethanol and 1.5% lactic acid. Three selected anti-fungal strains, *Lactobacillus fermentum* M017, *Lb. fermentum* 223, and *Saccharomyces cerevisiae* H290, inhibited the growth of the aflatoxin-producing strain, *Aspergillus flavus* S075, at 100% on the surface of 20 g cocoa beans (Fig. 1). When applied as LAB-yeast co-cultures to 1-kg cocoa bean fermentations, the strains led to cocoa beans, on a scale from 0 to 10, with 0.9-1.5 units lower bitterness, astringency, and off-flavours than spontaneously fermented beans. The co-culture *Lb. fermentum* 223/*S. cerevisiae* H290 even led to 1.2 and 1.5 units higher fine and cocoa flavours, respectively, and is suggested for future applications based on this positive influence on cocoa flavour, its anti-fungal activity against *A. flavus* S075, and the stress tolerance of both strains.

Figure 1. Inhibition of aflatoxin-producing strain A. flavus S075 by anti-fungal strains after 4 days of incubation at 30 °C on the surface of cocoa beans.

Poster session: **Cocoa**

Cocoa Poster 23

Assessment of the threshold tolerance of cacao fruits of different cacao genotypes to *Sahlbergella singularis* Haglund attacks under field conditions in the Southern Cameroon (Central Africa)

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Text *Sahbergella singularis* Haglund, 1895 is one of the major pests in cacaoculture in Africa. Cocoa production losses due to this species have been widely documented in West Africa. However, their impact on cocoa production is unknown in Central Africa, especially in Cameroon. Moreover, no data is available on the threshold level of fruits tolerance to mirid attacks. For these reasons, we assessed the effect of *S. singularis* on the productivity of ten cacao genotypes as well as the threshold number of the lethal feeding punctures to fruits under a randomized experimental design. Observations were made three categories of fruits, viz: cherelles, immatures and matures. A control trial was also set up per batch. The overall results showed that 68.0% and 0.4% of fruits aborted respectively in mirid and control trials. The percentages of aborted fruits were significantly (*p<5%*) different between cacao genotypes, and ranged from 20 to 100%. Bonferroni test revealed six homogenous groups for cacao genotypes susceptibility to mirid attacks; SNK52 proved to be most tolerant/resistant whereas two genotypes (UPA138 and SNK67) revealed more sensitive. In contrast, six genotypes (SNK07, IMC60×SNK417, T60/887×PA7, T79/501×SNK479, UPA143×ICS84, UPA143×NA33) displayed similar sensitivity to mirid attacks. ANOVA showed that the threshold tolerance of tested fruits, expressed by the mean numbers of lethal feeding punctures, to *S. singularis* attacks were comparable between cacao genotypes. This new quantitative database improves our knowledge on the (i) threshold tolerance of fruits to *S. singularis* attacks and (ii) economic impact of this pest on cocoa production in Cameroon.

Poster session: **Tea**

Poster Tea

Tea Poster 01

Evaluation of Quality and Intensity of Astringent Taste of Green Tea Based on Mass Spectrometry-Based Targeted Metabolic Profiling of Phenolic Compounds

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Text Abstract
It is well known that the phenolic compounds are the predominant factors for astringent taste of tea beverage. However, the correlation between astringent taste and phenolic compounds of tea beverage remains limited understanding. Here, the aforementioned correlation was investigated via twice partial least-squares (PLS) analyses using 47 green teas as samples. On the basis of the first round of PLS analysis, the samples were assigned to four groups for different astringent qualities. Targeted metabolic profiling analysis enabled by the MRM mode of UPLC-QQQ-MS/MS revealed that compared with the samples in the fourth sample group (i.e., little astringent), the following metabolic pathways were upregulated: in the first sample group (i.e., coarse and astringent), proanthocyanidin condensations, myricetin, and quercetin glycosylations; in the second sample group (i.e., grassy and astringent), phenolic compound acylations and kaempferol glycosylation; in the third sample group (i.e., bitter and astringency), myricetin and quercetin glucosylations. Based on the second PLS analysis of each group sample, the key metabolic pathways contributing to the astringent intensity of the first, second and fourth groups samples were confirmed as the phenolic acids acylations. In the third group sample, several metabolic pathways together affect the astringent taste intensity. In this study, the biochemistry basis of astringency qualities and intensity of green tea were elucidated.

Keywords: Astringency; Green teas; Mass spectrometry-based targeted metabolic profiling; phenolic compounds.

Tea Poster 02

RNA-Sequencing Analysis Reveals L-Theanine RegulatingTranscriptional Rhythm Alteration in Vascular Smooth Muscle Cells Induced by Dexamethasone

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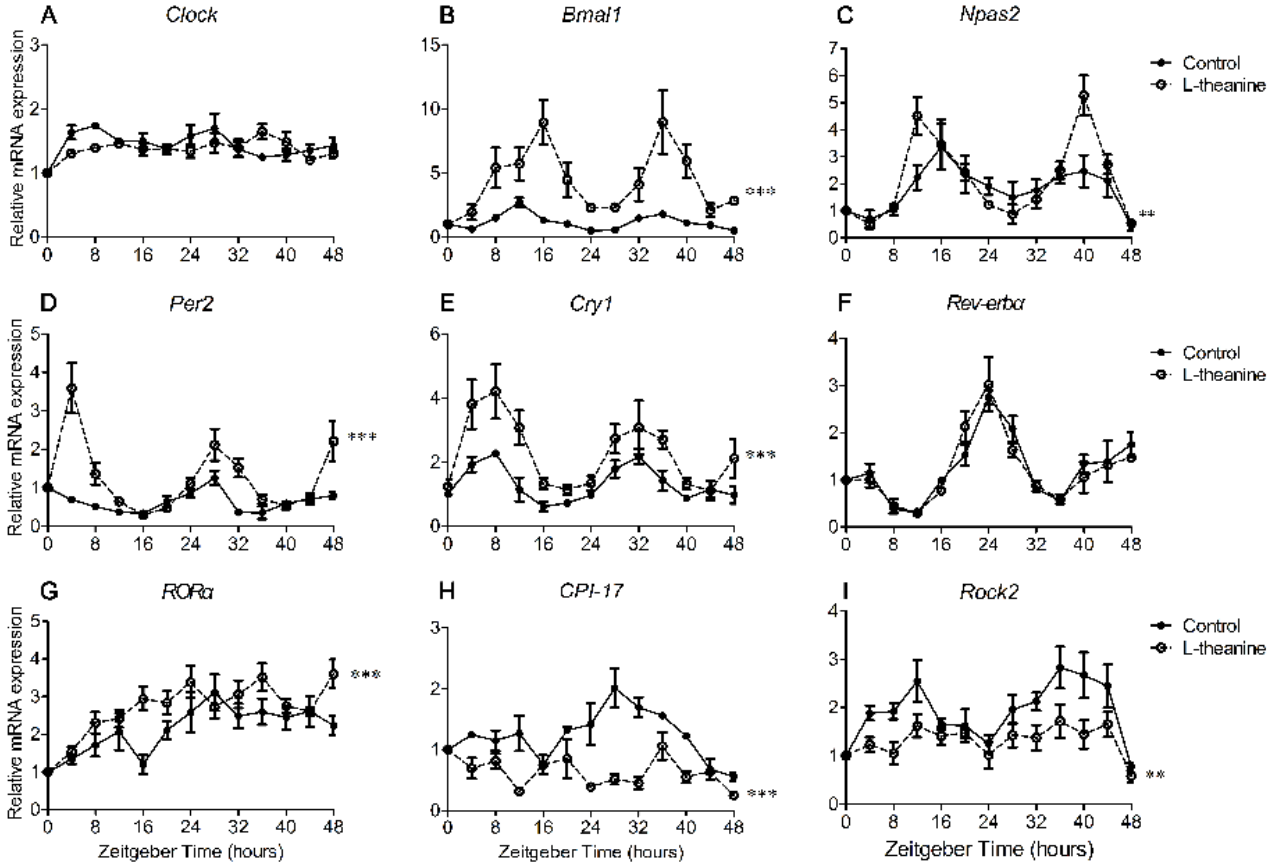
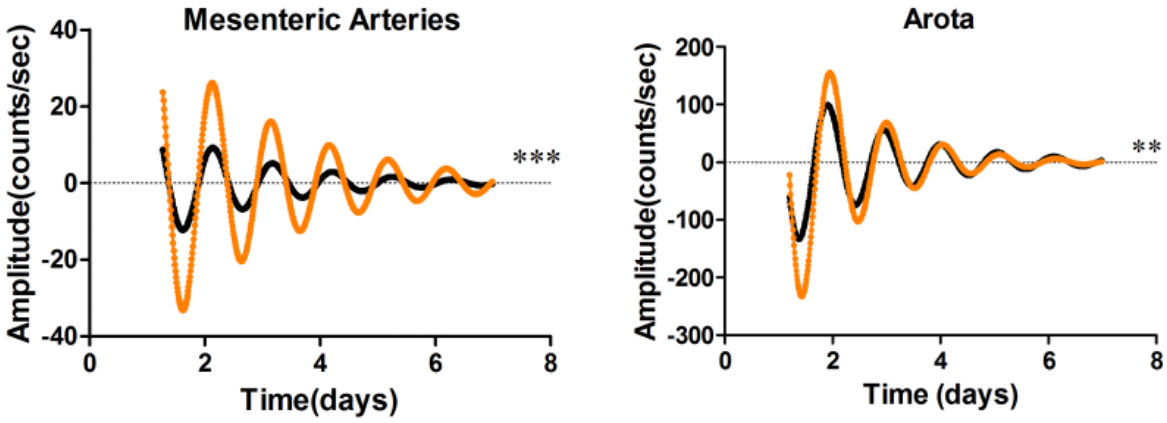
Text L-Theanine, a unique amino acid in tea leaves, is known to have beneficial effects on stress relief, tumor suppression, and prevention of hypertension and cardiovascular diseases (CADs). The disruption of the circadian rhythm has been implied in the pathogenesis of CADs. However, it is unknown whether L-theanine has a modulatory effect on the vascular circadian rhythm. In this research, we have established a circadian gene expression model in rat vascular smooth muscle cells by dexamethasone induction. L-Theanine treatment enhanced the expression amplitude of clock genes mRNA and proteins, including *Bmal1*, *Cry1*, and *Per2.*



The effect of L-theanine on genes expression amplitude

In addition, pairwise comparisons of the RNA-sequencing data showed that L-theanine is able to upregulate a ray of the rhythm genes and differentially expressed genes that are

involved in vasoconstriction and actin cytoskeleton regulation pathways. Moreover, the results from fluorescence detection showed that L-Theanine increase the amplitude and also advanced the rhythm phase of the PER2 expression in the aorta and mesenteric artery isolated from the PER2::LUC mice.



Peipheral tissues amplitude increased by L-theanine

The current study is the first to report that L-theanine enhances the amplitude of some major circadian gene expressions and decreases the amplitude of vasoconstriction gene *RhoA*, *Rock2*, and *CPI-17* expressions in primary cultured VSMCs induced by dexamethasone. Our findings implicated that L-theanine may have an important role in modulating the vascular circadian system and, thus, preventing circadian gene-related metabolic syndrome and CVDs.
[01][02]

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[02] Seira Narishige, Mari Kuwahara, Ayako Shinozaki, et al., (2014), Effects of caffeine on circadian phase, amplitude and period evaluated in cells in vitro and peripheral organs in vivo in PER2::LUCIFERASE mice. , Shigenobu Shibata, British Journal of Pharmacology, Tokyo, 5858-5869, 171(24), DOI:10.1111/bph.12890, 2014-08-16, Laboratory of Physiology and Pharmacology, School of Advanced Science and Engineering, Waseda University.

Poster session: **Tea**

Tea Poster 03

Characterisation of Principal Chemical Constituents, Vitamin and Mineral Elements of Nigerian Tea Clones

Lukumon Odunmbaku[1], Taofeek Shittu[2], Samuel Oluwole Babajide[1], Samsideen Aroyeun[3]
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Text ABSTRACT
Agronomic traits have been used severally to assess the quality of commercially grown tea plants (*Camellia sinensis*) in Nigeria but there is a dearth of information on the principal chemical constituents of Nigerian Tea leaves for industrial use. Thus, this study evaluated the chemical composition of some tea clones grown in Nigeria. Ten (10) Clonal genotypes of Tea plant, Camellia sinensis (L.) O. Kuntze grown in Nigeria were analysed for epigallocatechin gallate (EGCG), epigallocatechin (EGC), epicatechin gallate (ECG), epicatechin (EC), Caffeine, and vitamin (water and fat soluble) contents using High Performance Liquid Chromatography while selected mineral elements were determined using Atomic Absorption Spectrometry. Results showed that Nigerian tea clones contain 11.78-64.75, 0.09-1.25, 0.6-6.67, 1.06-4.66 and 0.60-2.51 (mg/g) EGCG, EGC, EC, EGC and caffeine respectively. Vitamin C, B_1, B_2, B_3, B_6, B_9 and B_{12} content also ranged between 2.00 and 3.99, 2.80 and 3.59, 18.73 and 40.87, 1.80 and 11.48, 1.97 and 3.77, 30.95 and 60.56 and 2.57 and 8.94 mg/g, respectively. Vitamins A, D and K were below 0.1 mg/g while vitamin E ranged between 0.26 and 0.27 mg/g. Cu, Mn, Ca, Mg, Na and K ranged from 0.22 to1.03, 0.08 to 0.29, 4.59 to 10.44, 12.67 to 155.60, 9.39 to 12.02 and 0.91 to 0.99 (mg/g) respectively. This study revealed the principal chemical constituents of Nigerian tea clones.
Key words: Tea, Catechins, Polyphenols, EGCG, EC, EGC, Caffeine

Tea Poster 04

Effect of Process Variables on the Sorption Characteristics of Nigerian Green Tea

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Text Moisture sorption isotherm information is required for storage of tea and agricultural products. This study therefore evaluates the effect of steaming time, drying temperature and drying time on the moisture sorption isotherms of optimized Nigerian green tea samples at 27, 35, and 40^{o}C using the standard gravimetric static method over a range of relative humidity from 10% to 80%. Plots of equilibrium moisture content (EMC) and water activity (a_w) was generated for isotherm curves. The experimental sorption curves were fitted by seven equations: Peleg, Henderson, GAB, BET, Caurie, Oswin, and Smith equation. The sorption isotherms of Nigerian green tea indicated a slight decrease in the

amount of water activity with increase in temperature at constant Equilibrium Relative Humidity (ERH). The characteristic of the obtained sorption isotherm was a type III sigmoidal curve according to Brunauer's classification. The sorption isotherm results indicates that the Peleg model gives better fit to the experimental data with lowest values of MRE and SEE than other models. The GAB monolayer moisture content at 27, 35 and 40^{o}C ranged between 1.94 and 6.83, 2.94 and 6.47 and 3.31 and 6.65%, respectively.

Tea Poster 05

Three tea catechins inhibit contraction of vascular smooth muscle

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Anhui Agricultural University, state key laboratory of tea plant biology and utilization, Anhui Agricultural University, Hefei, Anhui, China

Text Hypertension is a progressive cardiovascular syndrome caused by many factors. Many studies have demonstrated that consumption of tea can prevent hypertension. Tea is rich in polyphenols, which catechins constitute approximate 80% of the total amount of polyphenols. Recently, some methylated catechins have been found and isolated. The methylated catechins are relatively stable, and showed anti-inflammatory and anti-allergy effects. However, little is known for its vasodilation effect. The current study investigates the vasodilation function of newly isolated 3" methylated ECG as well as ECG and EGCG by using rat mesenteric artery.

Our results revealed that all three polyphenols have vasodilation function, and this function shows a concentration dependent manner. Using norepinephrine induced contraction on the second branch of mesenteric artery, 20μM and 40μM 3" methylated ECG significantly decrease the contraction of mesenteric artery compare to ECG. Moreover the relaxation effect of 3" methylated ECG on the artery is similar to the EGCG. Mechanism study has shown that this three polyphenols inhibit angiotensin II (AngII) induced phosphorylation of protein kinase C-dependent phosphatase inhibitor of 17kDa (CPI-17) and myosin light chain (MLC-20) in cultured rat vascular smooth muscle cells (VSMC), which cause vaso-contraction. Furthermore, the three polyphenols also inhibit NE induced phosphorylation of CPI-17 and MLC-20 in rat aorta. AngII and phenylephrine can activate G-protein coupled receptors and then activate ROCK which enhances SMC contraction. MYPT1 (myosin phosphatase targeting subunit 1) is directly phosphorylated by Rho kinase (ROCK). Our result indicated that the three catechins significantly decreased NE induced phosphorylation of MYPT1 in rat aorta. Taken together, the function of vascular relaxation of ECG3"Me, ECG and EGCG may achieve by regulating Rho-ROCK pathway.

Poster session: **Tea**

Tea Poster 06

Dietary Supplementation of Green Tea Preventing Hypertension of Older C57BL/6 mice induced by Deoxycorticosterone acetate and Salt

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Text Senior hypertension affects the life quality of aged population. There are a few reports about the effects and mechanisms of green tea supplementation preventing aged related hypertension. The current study investigated the effect and mechanism of green tea diet on prevention of hypertension induced by deoxycorticosterone acetate (DOCA) and salt in C57BL/6 old mice. The mice were pre-treated with or without the tea diet(2.5%(L), 5.0%(M) and 7.5%(H))for 6 weeks. Then mice were implanted DOCA pellets subcutaneous and given salt drinking to induce hypertension. Blood pressure (BP) was recorded by tail-cuff method, and the contraction of mesenteric arteries were monitored by the Myograph System. RT-PCRs were performed to detect the gene expression of some inflammatory factors and vasoconstrictor genes. Immunohistochemical analysis was used to detect macrophage infiltration in kidney. IL-6 and NGAL were detected in plasma also. The results showed that DOCA plus salt induced systolic blood pressure (SBP) and diastolic blood pressure (DBP) increasing remarkably, and dietary supplements of green tea significantly reduced SBP with dose-dependent (137mmHg model group vs 132mmHg of L group, 128mmHg for M group, and and 125mmHg for H group at 51 week old mice). And green tea diet significantly reduced the contraction of mesenteric arteries stimulated by 143mM potassium chloride, 100nM angiotensin II, 5-hydroxytryptamine, U46619 and norepinephrine with dose-dependent in old mice, respectively. In aorta, inflammatory factors (*IL-6*, *IL-1β*, *TNF-α* and *MCP-1*) and vasoconstrictor (*CPI-17*, *Rock1*, *Rock2* and *RhoA*) genes expression were suppressed observably by tea diet compared with model group. HE staining showed the remodeling of muscular layer of aorta in model group and it was improved by M and H tea diet supplement. In kidney, *IL-6*, *IL-1β*, *TNF-α* and *MCP-1* were suppressed observably by tea diet compared with model group. Furthermore, tea diet inhibited the injury of glomeruli and tubules and macrophage infiltration caused by DOCA plus salt. IL-6 and NGAL level in plasma were decreased by M and H tea diet also compared with model group. In conclusion, dietary supplement of green tea inhibited inflammation and improved the injury of kidney and aorta to prevent hypertension of old mice. Our data indicated that the dietary supplement of green tea has potential as a food additive for preventing hypertension of aged people.

Poster session: **Tea**

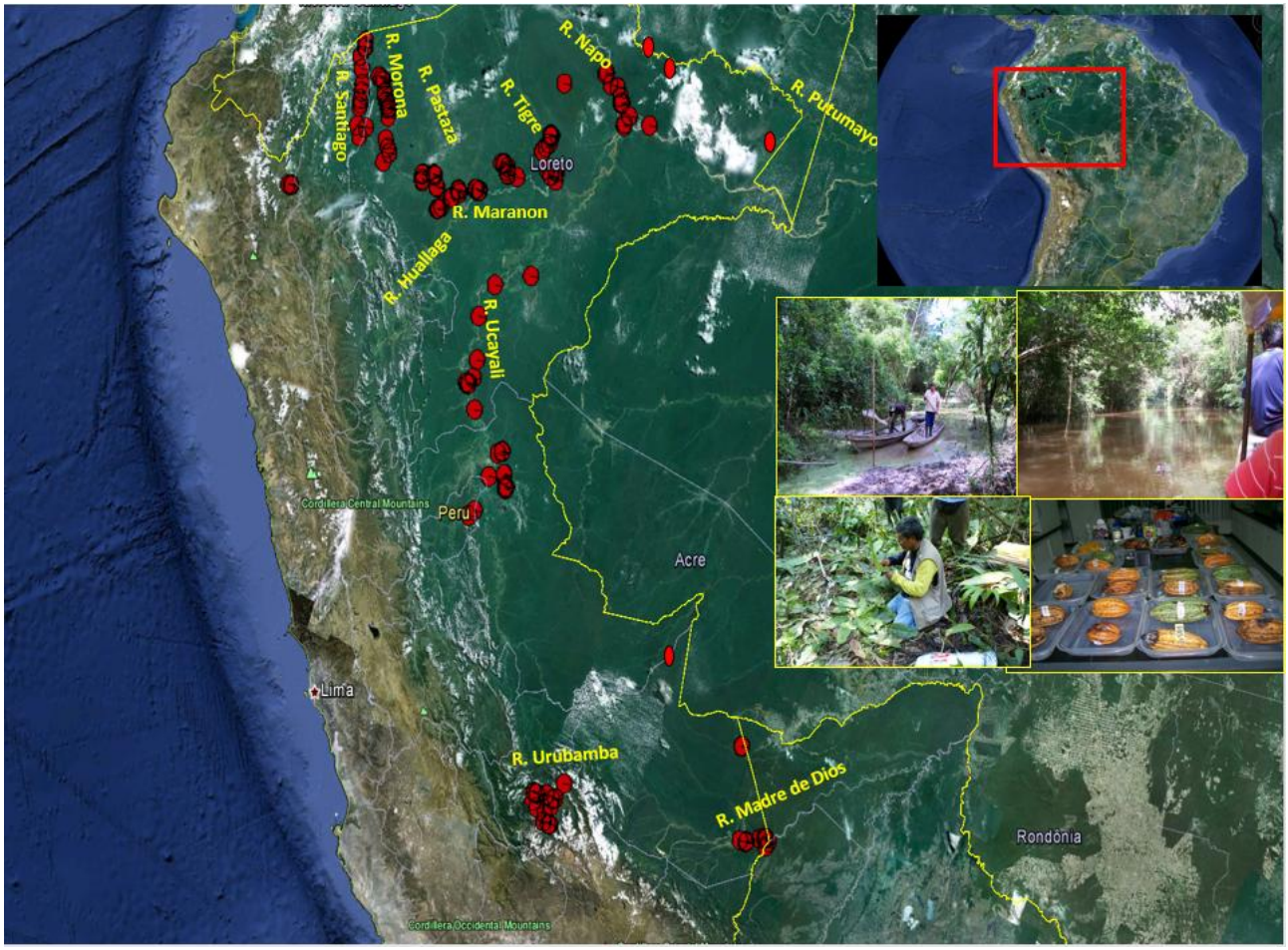
Tea Poster 07

Classification and botanical authentication of cacao (*Theobroma cacao* L.) and tea (*Camellia sinensis* [L.] Kuntze) using nanofluidic SNP fingerprinting

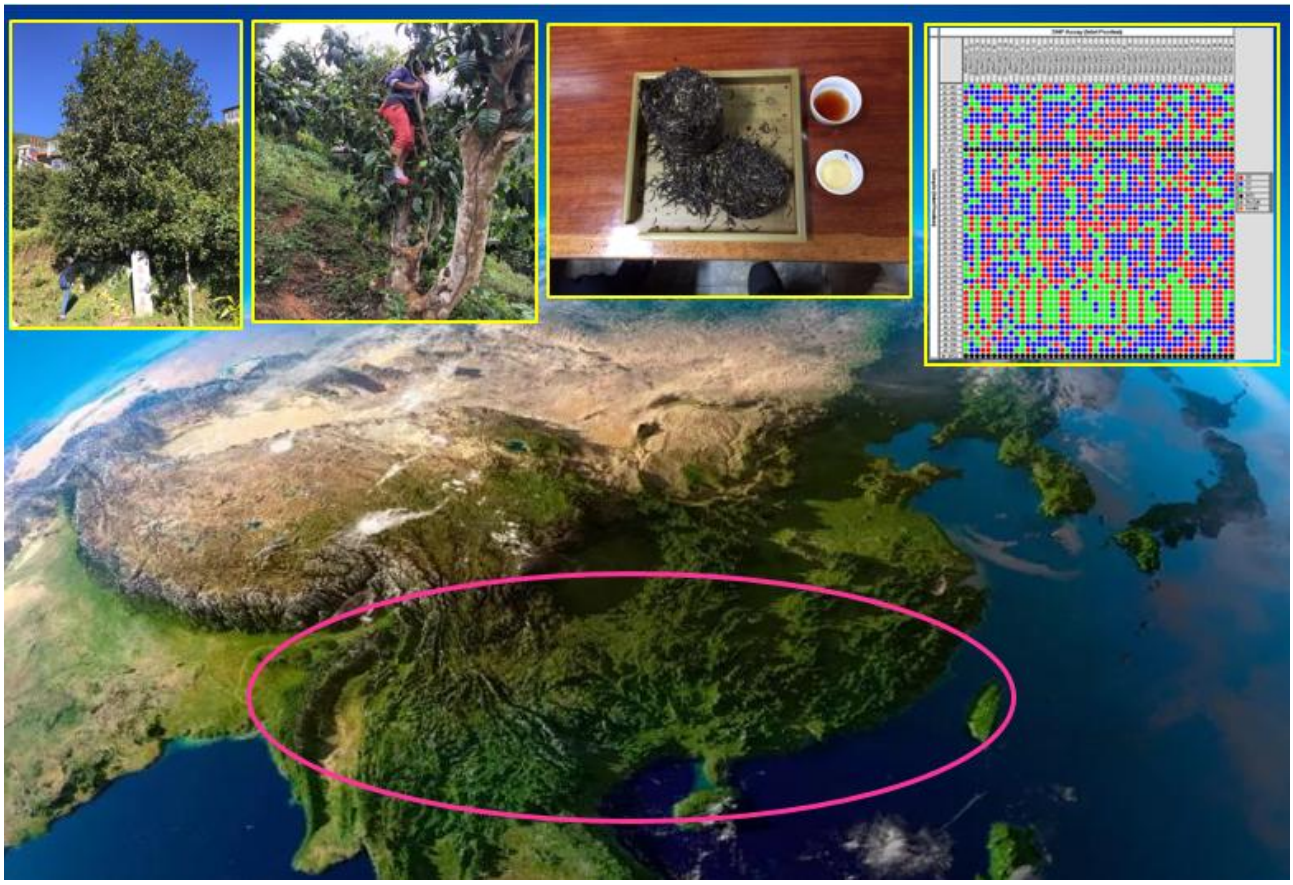
Dapeng Zhang[1], Enrique Arevalo[2], Wanping Fang[3], Borthakur Devajit[4], Boyi Wang[5], Lin Zhou[6], Lambert Motilal[7], Lyndel Meinhardt[8]
[1]USDA-ARS, Sustainable Perennial Crop Laboratory, Beltsville, United States, [2]Instituto de Cultivos Tropicales (ICT), Tarapoto, Peru, [3]Department of Tea Science, Nanjing Agricultural University, Nanjing, China, [4]Tocklai Tea Research Institute, Jorhat-785008, Assam, India, [5]Yunnan Forestry Tech. College, Kunming, China, [6]Shanghai Academy of Agricultural Sciences, Shanghai, China, [7]Cocoa Research Centre, The University of the West Indies at St. Augustine, St Augustine, Trinidad and Tobago, [8]USDA-ARS, Sustainable Perennial Crop Laboratory, Beltsville, MD, United States

Text Cacao (*Theobroma cacao* L.) and Tea (*Camellia sinensis* [L.] Kuntze) are important perennial crops in tropical and subtropical regions. Rapid market segmentation has resulted in a strong demand for specialty cocoa and tea products. One problem with the premium market is contamination with off-types, adulterating raw premium material. Accurate determination of genetic identity for a single cacao bean or a single tea bud is essential for ensuring varietal authentication. Through numerous collecting expeditions in the primary gene pools of cacao and tea in the Americas and Asia, we sampled a large number of wild populations and landraces of cacao and tea. Using a nanofluidic single nucleotide polymorphism (SNP) genotyping system, we analyzed the collected samples and classified the wild cacao and tea genetic resources into different genetic groups, which provides a baseline information regarding the geographical distribution of genetic diversity for these two species. We then generated SNP fingerprints for small quantities of DNA extracted from any given single cacao bean or a single tea leaf. Based on the SNP profiles, the adulterant varieties can be unambiguously distinguished from the authentic ones by multi-locus matching. For varieties with unknown genetic identities, assignment tests based on both Bayesian clustering analysis and allele frequency can be applied to separate the authentic from the non-authentic samples. The nanofluidic SNP protocol, together with forensic statistical tools, is sufficiently robust to establish authentication and to verify gourmet cacao and tea varieties. This method shows significant potential for practical application to the chocolate and tea industries.

Poster session: **Tea**



A map showing the wild cacao collecting sites in the Peruvian Amazon. Each red dot represents a collecting site.



A map showing the geographical region where genetic diveristy in the primary gene pool of Camellia sinensis was analyzed using SNP markers

[1]

References:

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Poster session: **Tea**

https://doi.org/10.1007/s11295-019-1316-y

Tea Poster 08

Green tea polyphenol EGCG alleviates metabolic abnormality and fatty liver by decreasing bile acid and lipid absorption in mice

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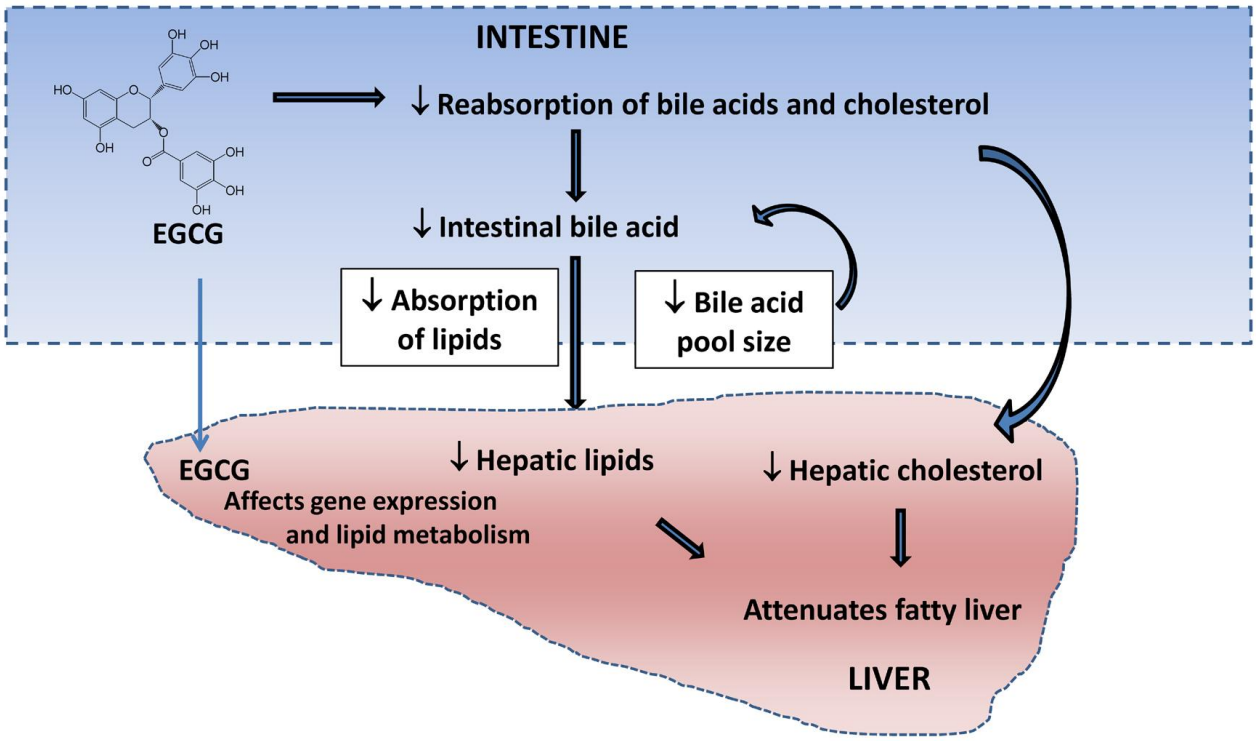
Text



The beneficial effects of EGCG are proposed to be due to the decreased intestinal bile acid reabsorption, decreased lipid absorption, reduced hepatic lipid load and altered hepatic and lipid metabolism.

The tea polyphenol (-)-epigallocatechin-3-gallate (EGCG) has been shown to ameliorate metabolic abnormalities and fatty liver induced by high-fat diets. In the present study, we investigated the time-dependent effects and the mechanisms of actions of EGCG on bile acid homeostasis and lipid metabolism in male C57BL/6J mice fed a high-fat western-style diet. We found that EGCG (0.32% in the diet) significantly reduced body weight gain, mesenteric fat mass, fasting blood glucose, insulin resistance, serum cholesterol and severity of fatty liver after treatment for 17 weeks. However, most of these effects were decreased or not observed after treatment for 33 weeks. Therefore, we used samples harvested at week 17 for biochemical characterizations. EGCG treatment significantly elevated the mRNA levels of cholesterol 7α-hydroxylase, HMG-CoA reductase, low-

density lipoprotein receptor and scavenger receptor B1, and partially normalized the lipid profile as revealed by lipidomic analysis. The intestinal bile acid content was significantly decreased, while fecal excretion of bile acids, cholesterol and total lipids were increased. We propose that EGCG decreases bile acid reabsorption, resulting in lower intestinal bile acid levels, which further decrease the absorption of lipids. These actions contribute to the alleviation of metabolic abnormalities and fatty liver disease caused by a high-fat diet.

Tea Poster 09

The Smell Intensity at the Final Shaking is a Key Clue to The End Oxidation Volatiles in Oolong Tea

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Text Bao-chung oolong tea is a typical partial-fermented tea in Taiwan. Its smooth and elegant flavor makes it worthy to promote to the world. However, the manufacturing process of oolong tea still remains low automation and depends on experience. The key operation of good quality is the cyclic shaking and setting steps. Tea leaves are oxidized during the shaking and setting in the intervals. We defined the smell to 3 intensities: (1) the strong odor group (H); (2) the moderate odor group (M); and (3) the weak odor group (L). We separated the tea leaves into 3 groups, H-, M-, L-group, according to the smell intensity before the second shaking. The timing decision of the third shaking was followed by the same classification of the second shaking to manipulate. In the final shaking (the 4th shaking), we separated the tea leaves into 3 groups again. The combinations of different intensities of shaking timing were 9 treatments (3 in the 2nd shaking Í 3 in the 4th shaking). The results show the variations of peak areas from every single compound were not regular in 3 levels of smell intensity. However, the smell intensity could be represented by the sum of peak areas for all detected volatiles (Fig. 1). There were low amounts of detected peaks in all L-group of the 4th shaking (HL, ML, LL), and the high level in all H-group (HH, MH, LH). These results suggest the variations of total peak areas may link to the smell intensities when deciding the shaking operation. Furthermore, volatiles quality performance index of the end oxidation, the ratio of methyl salicylate to trans-2-haxenal, which reveals as floral odor compound to the green odor compound were significantly related to the smell intensities of the last shaking (Fig. 2). By our results, the amounts of volatiles before the final shaking would be used as an index for shaking decision according to the ideal aroma of tea product that manufacturers' wish.

References:
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Poster session: **Tea**

Tea Poster 10

Green tea polyphenols and epigallocatechin-3-gallate attenuate perfluorodecanoic acid-induced liver toxicity in mice by inhibiting NLRP3 inflammasome activation

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Text Perfluorodecanoic acid (PFDA) is a is a highly toxic, persistent environmental pollutant that is extensively used in food applications and found globally in the environment, wildlife, and humans. PFDA was categorized as a persistent organic pollutant by the United Nations Environment Program in 2009. Human beings are exposed to PFDA through the ingestion of contaminated food and water as well as the inhalation of polluted air. In Western countries, the plasma concentration of PFDA in the general population was observed to be 0.8 ng/mL. However, molecular mechanisms underlying PFDA-induced liver toxicity have not been fully elucidated. Green tea polyphenols (GTPs), particularly epigallocatechin-3-gallate (EGCG), are natural antioxidants and effective in preventing various types of liver damage. Therefore, we investigated the protective role of GTPs and EGCG against PFDA-induced liver toxicity in mice. A PFDA-induced liver toxicity model was established by giving mice drinking water containing different concentrations of PFDA. GTPs or EGCG (0.32%, w/v) were co-administered to mice exposed to PFDA in drinking water. We found that (i) GTPs and EGCG reduced the mortality of mice who received a lethal dose of PFDA; (ii) GTPs extended survival time and inhibited weight loss among mice who received with a lower toxic dose of PFDA; (iii) GTPs and EGCG ameliorated liver toxicity, oxidative damage, and histological changes caused by a moderately toxic dose of PFDA; and (iv) GTPs and EGCG reduced hepatic inflammation and NLRP3 inflammasome activation caused by a moderate toxic dose of PFDA. Taken together, these results suggest that GTPs or EGCG supplements (or green tea intake) can be beneficial for people exposed to PFDA. This study is the first to reveal a mechanistic basis of GTPs and EGCG protection against PFDA-induced liver toxicity.

Tea Poster 11

Comparative study of Freeze drying and Vacuum drying on Antioxidant, Activity and Color of *Camellia assamica from Assam tea estate, India*

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Text Freeze drying method was reported to be more effective in drying of tea for better quality compare to natural drying method used in tea processing. The previous study on various tea types other than *Camellia assamica* showed that freeze dried tea contained higher amount of polyphenols and flavonoid compare to other drying methods. Till now the comparative study of freeze drying study and advances vacuum drying method on

Camellia assamica has not fully explored yet. Thus the object of this study was to compare both the methods of drying and investigate the effect on antioxidant and activity as well as colour change of *Camellia assamica*. To determine the antioxidant activity DPPH FRAP and ABTS assays are used. Color of the tea samples was measured with Hunter Lab colorimeter. the highest antioxidant and antiradical activities with an average FRAP value of 2.48 mmol/g and IC_{50} of 12.8 and 8.32 mg/ml for DPPH and ABTS assays, respectively are obtained for Vacuum drying samples which was found to have a very less difference with freeze drying method . In the color analysis for the vacuum dried sample it was found to be darker with $\Delta L^*$is 12.6, and the chroma ΔC^* is 2.6 CIE units. Analytically cholorophyll *a* is 1.48 mg./gm, cholorophyll *b* is 1.10 mg./gm and carotenoids 0.68 are determined in the tea samples.

Key Words: Tea, Vacuum drying, Freeze drying, Antioxidant activity, Color

Tea Poster 12

Mate Tea and Mate Tea Based Beverages – Analysis and Quality

Isabelle Kölling-Speer, Magdalena Krause, Karl Speer
Food Chemistry, Technische Universität Dresden, Dresden, Germany

Text Yerba Mate tea, an infusion made from the leaves and small stems of the tree *Ilex paraguariensis,* is a widely consumed nonalkoholic beverage in South America, especially in Argentina, Brazil, Uruguay and Paraguay, which is gaining rapid introduction into the world market, either as tea itself or as ingredient in refreshments.
Yerba mate has been shown to be hydrocholesterolemic, hepatoprotective, central nervous system stimulant, diuretic, and to benefit the cardiovascular system. The bioactive compounds protect DNA from oxidation and in vitro low-density lipoprotein lipoperoxidation and has a high antioxidant capacity. Numerous phytochemicals have been identified in mate tea that may be responsible for its health benefits. Among them, the polyphenols (chlorogenic acids) and xanthines (caffeine and theobromine) are important as well as the bitter and highly water-soluble saponins.
In Germany, mate has become more and more popular in recent years, but not as a traditionally prepared tea beverage, but as a caffeinated soft drink. Due to its stimulating effect the beverage was initially popular in the hacker scene, later it was also consumed in the Berliner techno- und hipster-scene. Meanwhile, mate based beverages from various manufacturers are offered everywhere, but still mainly in university towns.
The contents of the xanthines, the chlorogenic acids, and the saponins of mate tea have been analyzed and reported by various authors. However, only one paper describes the simultaneous determination of the three compound classes using LC-MS.
In our working group, an HPLC-DAD-ELSD method was developed that allows determining the xanthines and chlorogenic acids as well as the saponins of mate and mate based beverages in one run.
Using the new method different mate teas and mate tea based drinks of the trade were analyzed and jugded regarding their mate content.

Tea Poster 13

Characterization of Teas using HS-SPME-GC/MS

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Food Chemistry, Technische Universität Dresden, Dresden, Germany

Text Next to water, tea (*Camellia sinensis* L.) is one of the most widely consumed beverages in the world and is cultivated in more than 30 countries. The growing seasons, geographical regions, processing, and fermentation methods create many varieties that contribute to each tea`s uniqueness, especially to the tea aroma. Consequently, the volatile compounds are an important criterion in the quality control of tea.
The volatile compounds in the white, green, and black teas from different countries were analyzed and compared with regard to their composition. The concentrations of the volatile compounds measured in the samples varied considerably from each other and thus allow a classification of each tea species.

Tea Poster 14

Identification and profiling of human urinary metabolites using UHPLC-MS and ^{1}H NMR after black tea consumption

Mihella Berhanu Retta[1], Nikolai Kuhnert[2], Inamullah Hakeem Said[2], Sara Haka[2]
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Text Black tea is one of the most globally consumed beverages, which is rich in phenolics [1] and is considered to possess various beneficial health effects [2]. To systemically understand the bioavailability and metabolism of tea phenolics, it is significant to assess the fortune of tea phenolics after ingestion. The presented research focused at the apparent human gut bacterial and liver metabolism of catechins monomers, dimers and their oligomeric compounds after black tea intake.
The designed experiment comprised of various volunteers (n = 9) recruitments, who consumed 4 cups of black tea (for two days) and their urine and fecal samples (n=2) were collected at different time points over 48 h period. An optimized method using UHPLC-MS/MS and ^{1}H NMR techniques were implemented to identify and quantify the bioactive compounds of tea in volunteer's urine samples.
Moreover, an untargeted & targeted metabolomics approach using principal component (PCA) and partial least squares discriminant analysis (PLS-DA) were applied to the LC-MS and NMR data, to allow the identification of significant biomarkers responsible for discrimination of the metabolomes of various volunteers. Finally, the results indicated a significant amount of catechin bacterial metabolites (i.e. lactones and phenylacetic acids) and their phase-II conjugated forms detected in urine could be the contributing biomarkers of black tea positive health impacts.
Additionally, modulation of human primary and secondary urinary metabolites was also

investigated.

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Tea Poster 15

Aroma Characterisation of Pu-Erh Teas

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Text Pu-Erh tea is used in China as a health drink and is known in Chinese medicine for stimulating the metabolism and strengthening the immune system [1]. To produce Pu-Erh Tea, the dried leaves of Camelia sinensis are pressed and fermented by microorganisms [2]. The aroma composition of Pu-Erh teas was already analysed by Gas Chromatography-Olfactometry/Mass Spectrometry (GC-O/MS) in combination with Solid Phase Micro Extraction SPME [3, 4]. However, dilution to odour threshold techniques such as aroma extract dilution analysis (AEDA), as applied for the aroma characterisation of black and green tea infusions [5], have not been performed till now in order to investigate the key molecules responsible for the Pu-Erh tea aroma. The aim of this investigation was to identify the key odorants in the infusions of two Pu-Erh tea varieties, namely Shu and Sheng. For this reason, SAFE distillates of the Pu-Erh tea infusions were prepared and analysed afterwards by GC-O/MS in combination with AEDA. The results of this study clearly showed differences in the aroma profiles of the two investigated Pu-Erh teas on molecular level.

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Poster session: **Tea**

Tea Poster 16

Tea Plantation in Bangladesh: A Historical Analysis

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Text ABSTRACT

The purpose of this presentation is to disseminate the results of my recent research and some interesting findings about the history of tea plantation in Bangladesh. It is a non-technical presentation intended for lay audience.
Tea plantation began in the Northeastern region (Sylhet) of Bangladesh in the early 19th century. At that time, Sylhet was an integral part of Assam that has been famous for growing the world's finest varieties of tea. Tea plantation began in Assam and Sylhet at the initiative of the then colonial British government and the British companies like Duncan Brothers and James Finlay.

In order to unfold the origin of tea plantation in Bangladesh, I basically depended on Internet sources. This presentation will unfold some interesting aspects of early tea plantation with regard to the adventurous and entrepreneurial initiatives of the British governmental officers mobilizing tea-laborers from Orissa, Bihar and other parts of India. It will also highlight some economic, social, political, and moral issues pertaining to the history of tea plantation and the current state the tea industry in Bangladesh.

Tea Poster 17

Lemongrass agro-industrial by-product as a source of essential oil and carbohydrates with potential biological activities

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Text

Herbal preparations of lemongrass *(Cymbopogon citratus*) contains several bioactive compounds in its decoction, infusion, and essential oil extracts [1]. Hydrodistillation of lemongrass essential oil produces an aqueous waste which is usually discarded. A comparative study between this aqueous waste and an infusion was shown a similar phenolic compounds profile with antioxidant and anti-inflammatory potential [2]. The aromatic plants industry only commercialized the leaves with high quality standard. In the beginning of the season a first harvest is performed, cutting the leaves and discarded them

as by-product because they present some parts dried/brownish. After this practice, lemongrass plant develops leaves with high quality for infusions. It is possible that the essential oils (EOs) present in these by-products represent a source of valuable compounds with antimicrobial activity [1], as observed for the leaves used for infusion. In a circular economy concept, the aqueous waste produced during the hydrodistillation was also recovered as a decoction extract. The EOs yield was 0.41% and the decoction yield was 15%, dry weight. A total of 15 compounds were identified and quantified by GC-MS in EOs being the geranial and neral the major compounds present with 309 and 212 µg/mg, respectively. The composition of the decoction obtained during hydrodistillation contains 32% of carbohydrates, manly composed of glucose (83%), followed by fructose (5%) and galactose (4%). Carbohydrates presented also 10% of free sugars, mainly composed of glucose (46%). The glucose present in this water extract seems to be derived from glycosylated luteolin and apigenin derivatives [2]. This study shows that lemongrass by-products can be a profitable source of EO due to their high content of geranial and neral, showing potential to be incorporated as bioactive compounds.

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