

**Quality improved raw cocoa and cocoa-based products with flavour profiles on demand - From farm to chocolate bar (CocoaChain)
– CORNET –**



Coordination:	Forschungskreis der Ernährungsindustrie e. V. (FEI), Bonn
National Agencies:	AiF – Arbeitsgemeinschaft industrieller Forschungsvereinigungen „Otto von Guericke“ e. V., Köln/Deutschland FONDECYT - Fondo Nacional de Desarrollo Científico, Tecnológico y de Innovación Tecnológica, Peru
Research Association:	La Asociacion Peruana de Productores de Cacao (APPCACAO), Lima/Peru
Research Institutes:	Universität Hamburg Hamburg School of Food Science Institut für Lebensmittelchemie AK Prof. Dr. Bernward Bisping Prof. Dr. Bernward Bisping/Prof. Dr. Sascha Rohn Hochschule für Angewandte Wissenschaften Hamburg Fakultät Life Sciences, Department Ökotröphologie Professur für Lebensmittel-Mikrobiologie und -Toxikologie Prof. Dr. Katharina Riehn Julius-Kühn-Institut (JKI) Bundesforschungsinstitut für Kulturpflanzen, Berlin Institut für ökologische Chemie, Pflanzenanalytik und Vorratsschutz AG Spektroskopie von Pflanzeninhaltsstoffen Prof. Dr. Frank Ordon/Dr. Andrea Krähmer Universidad Peruana Cayetano Heredia, Lima/Peru Dr. Fabiola Leon Velarde/Dr. Rosario Rojas Universidad Nacional Agraria La Molina, Lima/Peru Dr. Carmen Velezmoro Sánchez/Ing. Wilfredo Lévano Carnero
Industrial Branch:	Confectionary Industry
Projektkoordinator (deutsches Teilprojekt):	Dr. Daniel Kadow August Storck KG, Berlin
Duration:	2016 - 2020
Volume:	€ 1.087.480,-- (total)

Initial Situation

With 1,3 million tons per year, accounting for about 40 % of the total European production, Germany is one of the largest chocolate producers worldwide. The average European consumer ate 5,9 kg of chocolate in 2012. Germans are the biggest consumers of chocolate in Europe with 11,7 kg per year and person. The key raw material in chocolate manufacturing are the seeds of the cocoa tree (*Theobroma cacao* L.). Peru, being one of the countries that cocoa is native of, has cocoa producing regions with great genetic diversity and a strongly growing cocoa sector.

Especially for high quality dark chocolates cocoa of high sensory quality is required. The sensory quality of cocoa mainly depends on the cocoa tree genotypes planted and the post-harvest processing (i.e. fermentation and drying). During the latter the precursors of chocolate flavor are developed and the amount of bitter tasting components is reduced. Aberration in post-harvest process may result in off-flavor formation and mycotoxin contamination of the cocoa beans. Nevertheless, cocoa production still relies on an uncontrolled, spontaneous fermentation process, which does not permit proper process control. This spontaneous fermentation process is one of the main reasons for unsatisfactory great heterogeneity of cocoa and for frequent complaints on sensory attributes. In addition, in the last years the global cocoa production did not meet the manufacturers demand several times and deficits of up to 300.000 tons (2006/2007) occurred.

Because of these circumstances, chocolate manufacturers encounter increasing problems with the sourcing of cocoa with high and stable sensory quality. At the same time, cocoas with distinct flavor profiles, not contaminated by mycotoxins and from traceable origins gain more and more importance - in particular for small and medium enterprises.

Cocoa flavor is multi-dimensional. Beside the typical cocoa aroma, bitter and sour, floral, fruity and nutty notes may occur. Characteristic is also the astringency. The potential to develop specific flavor notes mainly depends on the genetic background of the cocoa trees. However, depending on the climatic conditions flavor intensity may vary. During fermentation and drying bitterness and astringency are decreased. In contrast, cocoa aroma intensity increases and may finally overlay fruity, floral and nutty notes. Through application of adapted fermentation and drying protocols distinct aspects of the flavor potential may be emphasized. Hereby, cocoas with distinct flavor profiles may be produced from one and the same starting material. The course of fermentation and thus also the development of the biochemical and the sensory profile strongly depends on the physiological status of the cocoa seeds. Besides seed endogenous substances, apparently also compounds from the fruit pulp may contribute to the biochemical and sensory profile of cocoa seeds. The respective substances are produced in the fruit pulp during fruit ripening but, may also be formed by yeast strains during fermentation. The impact and in particular the interplay of the mentioned factors on the development of specific biochemical and sensory profiles is not fully understood. The potential contamination of cocoa with mycotoxins, especially with ochratoxin A, apparently mainly results from aberration during fermentation and drying permitting the establishment of the respective fungi.

Aim of the project was to establish a model processing chain, comprising all processing steps from the field to the final chocolate product („From Farm to Chocolate Bar“) will be. Within this chain, impact and interplay of cultivation conditions, cocoa genotype, cocoa seed physiology, fruit pulp, fermentation with selected starter cultures and drying on biochemical and sensory profile development in cocoa and chocolate are studied exemplarily.

Research Results

For the genetic characterization of the original cacao trees, sample material was collected in the Peruvian regions of Cusco, Huánuco and San Martin. It could be shown that the planting distance between cacao trees has a significant effect on the infestation of the fruit with insects. The fruits of densely planted cacao trees were more frequently infested with insects. It was also shown that the fruits of the original cacao varieties were significantly less infected than the fruits of the cacao clone CCN-51. The caffeine content of the original cacao from Cusco was significantly higher than that of the other cacao varieties examined. The genetic data analysis shows that the cacao samples from San Martin and Huánuco differ significantly from the samples from Cusco. Some cacao properties, such as the catechine, cyanidine arabinoside and caffeine content, as well as the theobromine/caffeine ratio, are significantly dependent on the region of origin.

In addition, yeasts isolated from spontaneous cacao fermentations done in various countries were characterized with regard to their growth behavior and the metabolites formed in order to make a selection of yeast strains suitable for cacao fermentations. An analysis of the secondary metabolites was carried out using gas chromatography coupled with mass spectrometry (GC-MS) in order to find yeast starter cultures for cocoa flavor as required. Since the microorganisms isolated in Peru could only be imported into Germany at the beginning of October 2019 due to bureaucratic obstacles (Nagoya Protocol), the goals were only partially achieved and no final starter culture could be created to influence the later developed cocoa flavor.

In the Peruvian regions of Cusco (Ivohote and Quillabamba), San Martin (Tarapoto) and Piura (Piura), spontaneous cacao fermentations were characterized and yeasts, lactic and acetic acid bacteria were isolated from these fermentations. In Quillabamba the regional fine cacao "Chuncho" was used for fermentation, in Tarapoto the consumer cacao clone CCN-51 and in Piura a mixture of white and purple cacao clones (70 %/30 %) were used for fermentations. To assess the influence of starter cultures, a cacao fermentation was inoculated with a commercially available yeast strain for winemaking and compared with a spontaneous fermentation. It could be shown that the use of a starter culture leads to this starter culture prevailing during fermentation. The isolated microorganisms were stored until identification (after October 5th, 2019) at 4 °C in Lima at the Universidad Agraria La Molina (UNALM). The yeasts were characterized by means of FTIR (Fourier Transform Infrared Spectroscopy). The isolated lactic and acetic acid bacteria were identified using MALDI-ToF/MS from the beginning of 2020 after they were recultivated. Because of the long storage of the bacterial strains in Lima and because of the closure of the laboratories of the University of Hamburg as a result of the COVID 19 pandemic, only about 10 % of the original bacterial isolates could be used again as live culture.

An international workshop on cacao processing and sensory evaluation of cocoa and cocoa products was held from March 2017 to June 2017. From January 2019 to December 2019, cocoa masses in liquid and solid form were tasted by a trained cocoa sensory panel and compared with one another. The results suggest that sensory tests of solid cocoa masses with appropriate training of the panel participants provide results comparable to those of conventional tests with liquid cocoa masses.

In an extensive cutting test on Peruvian beans, the bean halves were rated according to the categories slate, slate-purple, purple, purple-brown, and brown. At the same time, photos and UV/VIS hyperspectral image data were recorded. A new way of displaying the cutting tests in a ternary diagram enables fermentation processes to be shown and the fermentation index to be read off.

Three different roasting protocols (low, medium, heavy roasting) for cacao nibs have been developed on the basis of literature data and in conjunction with a test design. Work was carried out on establishing an overall analytical platform that combines the chemical-analytical as well as the sensory characterization of biochemical and physicochemical quality parameters during cacao processing and chocolate production.

A strategy for creating field sampling plans for cacao and sample division in the laboratory was developed, which allows both biological variability and random uncertainty along the analysis process to be roughly quantified and thus to ensure the validity of the results.

After long negotiations and a great deal of bureaucratic effort (beginning of 2017 to September 2019), it was finally possible to obtain the export license for the isolated strains of yeasts and bacteria and for the leaves of cacao trees, so that this genetic material could be exported at the beginning of October 2019.

Economic Impact

The German confectionary industry generates sales of approximately 12,5 billion € per year. A total export volume of 3,3 billion € made Germany the largest exporter of chocolates in 2011. The number of employees is more than 50.000. The German Confectionary Industry is strongly embossed by small and medium enterprises (SME) and the market for high quality chocolates produced by small and medium enterprises is highly developed.

The essential basis for high quality dark chocolates is cocoa of high and stable sensory quality. Based on the project results an optimized processing protocol (handbook) for cocoa will be provided. Application of the protocol will permit to obtain specific and distinct flavor profiles („Flavor on Demand“). The production of cocoa with manufacturer-specific flavor profile and quality assures the availability of raw materials for already existing products. Moreover, it provides new options for product innovation. NIRS, MIRS and chromatography-based quick methods are implemented for verification of this specific quality after each of the processing steps. Hereby, new options for quality assurance are provided. A strategy for minimization of mycotoxin contamination through starter culture-based stabilization of the fermentation process increases product safety. Finally, recommendations based on the model processing chain, will permit improved access to the origin and will hereby contribute to the stabilization of the value added chain.

Publications (Selection)

1. Final Report 2020.
2. Beleites, C., Glitschka, M., Böttcher, C. & Krähmer, A.: NIR spectroscopy for cacao bean quality measurements. OCM 2019: Proc. 4th Intern. Conf. Opt. Charact. Mat. Karlsruhe, March 13-14, 13-22, (2019).

Weiteres Informationsmaterial

Hamburg University
Hamburg School of Food Science
Institute of Food Chemistry
WG Food Microbiology und Biotechnology
Prof. Dr. Bernward Bisping/Prof. Dr. Sascha Rohn
Ohnsorgstraße 18, 22609 Hamburg
Tel.: +49 40 42816-642
Fax: +49 40 42618-643
E-Mail: Bernward.bisping@uni-hamburg.de

Hamburg University of Applied Sciences
Faculty of Life Sciences
Department of Nutritional Science
WG Food Microbiology and Toxicology
Prof. Dr. Katharina Riehn
Ulmenliet 20, 21033 Hamburg
Tel.: +49 40 42875-6368
Fax: +49 40 42875-6499
E-Mail: Katharina.riehn@haw-hamburg.de

Julius-Kuehn-Institute (JKI)
Federal Research Institute of Nutrition and Food, Berlin
Institute of Ecological Chemistry, Plant
Analytics and Stored Product Protection
Dr. Hartwig Schulz/Dr. Andrea Krähmer
Königin-Luise-Straße 19, 14195 Berlin
Tel.: +49 30 8304-2210
Fax: +49 30 8304-2503
E-Mail: andrea.kraehmer@jki.bund.de

Research Association of the German Food Industry
GFPI/FEI EU-Office
Dr. Jan Jacobi
47-51, Rue du Luxembourg, B-1050 Brussels
Tel.: +49 172 2643357
Fax: +32 2 2820841
E-Mail: gfpi-fei@bdp-online.de

Universidad Peruana Cayetano Heredia
Dr. Fabiola Leon Velarde/Dr. Rosario Rojas
Av. Honorio Delgado 430, 06484 Lima/Peru
Tel.: +51 1 3190000
E-Mail: fabiola.leon-velarde@upch.pe

Universidad Nacional Agraria La Molina
Dr. Carmen Velezmoro Sánchez/
Ing. Wilfredo Lévano Carnero
Av. La Molina s/n, 12056 Lima/Peru
Tel.: +51 1 6147121
Fax: +51 1 6147121
E-Mail: cevs@lamolina.edu.pe

Funding reference

... ein Projekt der **Industriellen Gemeinschaftsforschung (IGF)**

gefördert durch/via



Das o. g. IGF-Vorhaben der Forschungsvereinigung Forschungskreis der Ernährungsindustrie e. V. (FEI), Godesberger Allee 125, 53175 Bonn, wird/wurde über die AiF im Rahmen des Programms zur Förderung der Industriellen Gemeinschaftsforschung (IGF) vom Bundesministerium für Wirtschaft und Energie aufgrund eines Beschlusses des Deutschen Bundestages gefördert.

First page's photo credit: © ExQuisine – fotolia.com

Stand: 6. Januar 2022