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Analysing Complex Polyphenols from Roasted Coffee and Black Tea by Mass Spectrometry

Nikolai Kuhnert

Jacobs University Bremen,Campusring 8, 28759 Bremen, Germany,
E-mail: n.kuhnert@jacobs-university.de

Roasted Coffee

Introduction

The coffee beverage constitutes the major source of dietary polyphenols accounting for an estimated total daily intake of 30 % of dietary phenols at an amount of 200-350 mg per cup of coffee for most humans. The major polyphenolics in coffee are chlorogenic acids (CGAs) accounting for an estimated 10 % of the dry weight of a green coffee beans [1]. Within the last decade around 100 different CGAs have been unambiguously identified in the coffee plant [2,3] including various monoacyl **1-3** and diacyl quinic acids **4-8**. During coffee roasting an estimated 50 % of CGAs disappear if compared to the green coffee bean, forming in a Maillard-type reaction a material referred to as melanoidines [4].

In this contribution we describe our efforts to characterize low molecular weight roasted coffee melanoidines with a particular emphasis on products formed from CGAs. We employed two distinct analytical approaches for melanoidine characterization. In the first approach we employed a targeted analytical strategy providing a hypothesis of likely reaction pathways of CGAs during roasting, including acyl migration, *trans-cis* isomerization of cinnamate moieties, quinic acid epimerization, dehydration to form quinide and cyclohexene derivatives and dimerizations of cinnamoyl moieties [5]. In a second non-targeted approach FT-ICR-MS data were obtained from low molecular weight melanoidine revealing molecular formulas for compounds excluded from the targeted approach.

Results and Discussion

Using our targeted approach we could show, by comparison of LC-MS data of authentic reference materials obtained through chemical synthesis with actual roasted coffee samples, that roasting products of chlorogenic acid **1** include chlorogenic acid lactone **2**, cyclohexene derivatives including shikimic acid derivatives **3** , epimers of chlorogenic acids **5** and acyl migration product **8**. In addition *cis*-cinnamoyl derivative **6** and water addition products **4** which are absent in the roast, however, are produced in the brewing process (see Figure 1). Using this approach around 60 chlorogenic acid derivatives could be unambiguously identified so far in a processed coffee brew. Extrapolation of tandem MS data allows a tentative structure assignment for at least 100 further derivatives.

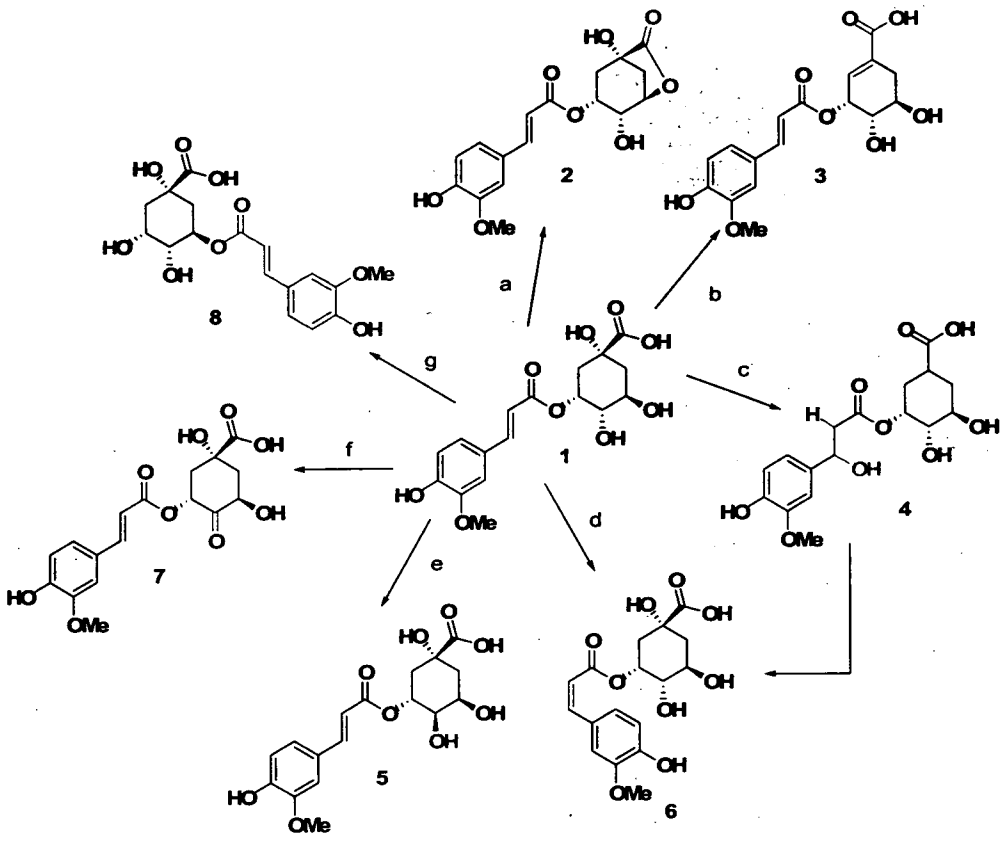

Figure 1: Derivatives of chlorogenic acid (here 3-caffeoyl quinic acid **9** shown as one example) formed during coffee roasting and brewing.

Using a non-targeted approach around 4 000 distinct molecular ions can be resolved in an ESI-FT-ICR-MS experiment of a roasted coffee melanoidine fraction. The data, if compared to model roast systems, in which CGA fractions and typical coffee carbohydrate fractions were subjected to typical roasting conditions show that only a small number of products arise from cross reaction. Hence compounds obtained from CGAs and carbohydrates and those obtained by their reaction with each other and with proteins or lipids can be unambiguously identified.

This suggests that compartmentalisation of individual compound classes take place in the bean during roasting or that an inherent lack of chemical cross reactivity exists. Additionally petrolomics style data interpretation reveals further tentative structures formed in the roasting process including oligomeric saccharides and their dehydration products and further interesting hydroxycinnamate derivatives. Tentative structures were subjected to further tandem LC-MS analysis and thus their structure confirmed.

Black Tea Thearubigins

Thearubigins are the most abundant group of phenolic pigments found in black tea accounting for an estimated 60 % of the solids in a typical black tea infusion. 50 years ago the term thearubigins was first introduced and up to now the chemical nature of the thearubigins remains largely unresolved if not mysterious despite many efforts clarifying their structures. This presentation summarises some of our attempts to clarify and elucidate the chemical

nature of the thearubigins, present in 15 commercially representative teas by analyzing the data obtained using combustion analysis, IR-spectroscopy, NMR spectroscopy, Diffusion NMR spectroscopy, UV-VIS spectroscopy, Circular Dichroism spectroscopy and Atomic Force Microscopy, MALDI-TOF-MS and ESI-FT-ICR-MS and tandem MS. The thearubigin fractions from these 15 teas are remarkably similar with respect to their spectroscopic fingerprint. The data obtained are consistent with the thearubigins being structures of not more than 2000 daltons comprising more than 10 000 individual chemical entities as detected by FTICR-MS. By applying petrolomics style data interpretation strategies and by developing novel data interpretation strategies on the data, a structural model for the thearubigins was developed and structures of around 400 thearubigin components further confirmed by targeted tandem MS experiments.

Conclusions

In conclusion our work provides for the first time a comprehensive picture about melanoidine derivatives formed in coffee roasting from chlorogenic acids and black tea thearubigins. Modern mass spectrometry methods offer an unprecedented insight into such extraordinarily complex materials containing thousands of analytes. Mass spectrometry offers the best of all methods with ultimate resolution combined with high sensitivity, selectivity through selective ion monitoring and multidimensional specificity by hyphenation to chromatography and other spectroscopic techniques.

References

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