Watered down taste

Extreme weather with alternating periods of drought and heavy rainfall can negatively affect yield and quality of agricultural crops. Recent research into the impact of climate variations on the quality of tea have shown how dramatic the influence can be.

By Guido Deussing

Second only to water, tea is by far the most widely consumed beverage in the world. Apart from actual or imagined physiological effects, the main and most important aspects for a tea drinker are thought to be flavor and enjoyment. There may be regularly recurring trends towards consuming only the healthiest beverages, but generally, consumers' main concern is that their drinks taste good. However, sensory characteristics such as taste and aroma are exactly the qualities affected by weather, as Professor Albert (Al) Robbat, Director of the Tufts University Sensory and Science Center in Medford, Massachusetts explains to GERSTEL Solutions worldwide. As it turns out, this is especially the case when the harvest literally falls into the water.

Tea – an ideal model plant for climate effect studies

In cooperation with an interdisciplinary team of colleagues, Al Robbat investigated tea samples from Yúnnán province in Southwestern China, an area renowned for excellent tea. The aim was to determine why tea leaves picked during the rainy monsoon season are less aromatic and frequently have off flavor issues compared with tea leaves picked during the dryer spring season. This was found to be the case even when comparing leaves picked from the same plant. The chromatography expert explains that not only do tea drinkers suffer when exposed to inferior quality brews; tea farmers incur significant losses since they are forced to sell crops grown during the monsoon season at much lower prices compared with the spring crop.

There is an urgent need for more insight into causes and effects since the Yúnnán region is increasingly faced with extreme weather conditions that can affect quality and yield and in extreme cases lead to total loss of harvest. As Al Robbat explains, there has already been a noticeable shift in the start of the summer monsoon and its duration, which means that periods with ideal harvest conditions are becoming shorter. The scientists set out to determine the root causes for changes in quality and flavor of Yúnnán tea as a function of rainfall, temperature, and elevation. Their findings are reported in Journal of Chromatography A [1].

Preliminary investigations showed the way

Initial investigations of tea harvested in the mountains of Yúnnán from spring until the onset of the monsoon period brought the following results: With increasing

amounts of rainfall, the catechin compound concentrations dropped by more than 50 percent. Among these are: catechin, catechingallate, epicatechin-3-gallate, epigallocatechin, epigallocatechin-3-gallate, gallic acid, gallocatechin and gallocatechin-3-gallate. The same effect was found for methylxanthines (caffeine, theobromine and theophylline) Al Robbat reports [2]. Although catechins and methylxanthines, astringent bitter compounds characteristic of poor quality teas decreased in concentration, other polyphenolic compounds (also astringent and bitter) increased in concentration. "Initially, we assumed that the loss of quality was related to a kind of dilution effect, in other words that the plant growth would outpace the production of secondary metabolites to which the flavor compounds belong", the scientist explains. However, when they determined that the total concentration and activity of antioxidants in teas harvested in spring was lower than in comparable teas harvested during the monsoon period, Al Robbat and his colleagues concluded that the plant chemistry, i.e. metabolism and physiology had changed completely and had adapted to the change in precipitation. This had resulted in changes in metabolism and, therefore, in the associated flavor determining metabolites. The idea was obvious, Prof. Robbat explains, to not just focus on significant flavor compounds in tea, but rather look at the bigger picture, including determining the entire group of flavor relevant compounds and to generate a profile of as many metabolites as possible in order to understand how environmental conditions influenced tea plant chemistry.

A closer look at plant metabolism

Taste and aroma of tea are a result of complex interactions between hundreds of chemical compounds. Al Robbat: "Extending our knowledge of potentially sensory relevant metabolites and monitoring them over a period of time is key if we want to develop an understanding of how environmental and climate factors influence tea quality." We know that seemingly unimportant compounds can significantly influence the organoleptic quality. Most studies listed in literature have focused on seasonal changes of non-volatile compounds, but we now know that volatile organic compounds (VOCs) with low odor thresholds contribute significantly to the total sensory impression, Al Robbat points out. This realization had led the group towards using GC/MS as the technique of choice for monitoring metabolites.

Tea harvester in a Chinese highland region.

Flavor research needs GC/MS analysis, and GC/MS methods are widely used in metabolomics research (see page 9). In the case of tea plants, the sample matrix is highly complex and a huge number of potentially relevant flavor compounds are involved. Literature sources count around 600, according to Prof. Robbat. This is the reason that a sequential, multidimensional GC-GC/MS method was used in order to comprehensively determine all detectable volatile metabolites in spring- and monsoon teas. The acquired data were used to build a metabolite database that relies on a deconvolution software to solve highly complex analysis tasks very quickly using a standard GC-MSD system. This is even possible when multiple compounds co-elute and also have overlapping signals.

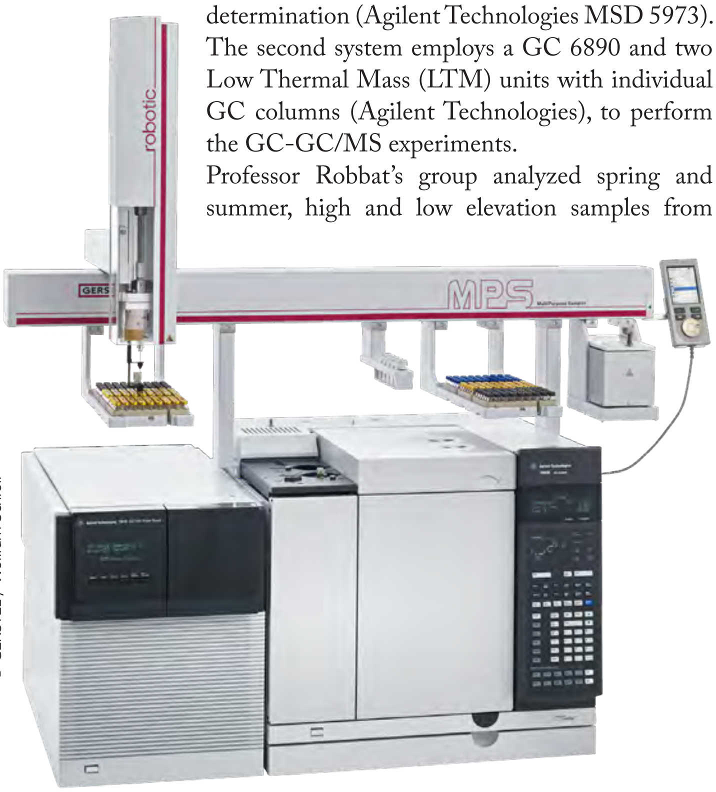
The deconvolution technology developed by Professor Robbat and his colleagues offers added value in comparison with conventional approaches. This includes the possibility to obtain and store retention times and spectra of neat metabolites that aren't skewed by matrix influence. It is also possible to identify perfectly co-eluting compounds. Professor Robbat and his group routinely measure the relative concentration of more than 400 compounds from infused tea leaves.

Technology that enables successful analysis

Initially the aim had been to use the spring harvested tea samples to find as many metabolites as possible and build a solid reference database. Al Robbat: "In order to build a comparative database, we instead analyzed tea samples consisting of buds and leaves that had been collected over three day periods in both spring and summer". In other words, the periods spanned both the dry season (spring) and the rainy season (monsoon). Collected tea samples were briefly microwaved in the field to stop enzymatic activity and subsequently sent to the laboratory in sealed plastic bags. In the laboratory, they were vacuum sealed, wrapped in aluminum and stored in a deep freezer awaiting analysis. GC-GC/MS analysis was performed on

extracts of the tea samples. The previously used classical extraction procedure was simultaneous distillation and extraction (SDE) using the Likens-Nickerson process on a tea infusion prepared using deionized water and concentrated under a nitrogen atmosphere over anhydrous sodium sulfate. For the work described here, the GERSTEL Twister® was used since it provides a simple, clean method to extract organic chemical compounds from the tea infusion requiring no or almost no use of organic solvent. The Twister is a glass coated magnetic stir bar fitted with a PDMS sorbent phase. While stirring the sample, the Twister efficiently extracts organic compounds. The Twister was also used to collect organics in the field by placing it directly under the leaf and holding it in place with a magnet [3]. For the tandem GC-GC/MS analysis Al Robbat and his colleagues used two 6890 GCs from Agilent Technologies. The first system was fitted with an FID; analyte separation was performed using a column with polar stationary phase. Sample introduction to the Cooled Injection System (CIS) PTV-type inlet was automated using the GERSTEL MultiPurpose Sampler (MPS). The second GC system was configured with a non-polar capillary column and a Cryo-Trapping System (CTS) in order to enable heart-cutting and cryofocusing of fractions separated on the first column and to deliver these to the second column as a narrow band for best possible quality of separation as well as MS determination (Agilent Technologies MSD 5973). The second system employs a GC 6890 and two Low Thermal Mass (LTM) units with individual GC columns (Agilent Technologies), to perform the GC-GC/MS experiments.

Professor Robbat's group analyzed spring and summer, high and low elevation samples from both Yunnan and Fujian Provinces, temperatures during sampling varied by up to 10 °C. The samples were taken to study locational differences over a 3-year period (2014-2016)[4]. The data analysis software developed by Dr. Robbat and sold through GERSTEL provided the means to analyze these samples by GC/MS and track compositional changes in tea under stress conditions [5].



Analysis results confirm sensory impression

Al Robbat: "Our analysis results confirm what tea farmers tell you: The spring tea is of much higher quality and has a sweet floral flavor compared with the monsoon tea, which is described as green or earthy. Using tandem GC-GC/MS analysis we succeeded in identifying hundreds of metabolites in the spring and monsoon teas". Professor Robbat's group found 169 metabolites that were common to both tea types and more than 100 compounds that were unique seasonal compounds. A further 163 compounds were detected, but could not be identified. In future, we will increasingly have to contend with extreme weather conditions. This means that plant research must develop tools that can monitor the influence not only on plant growth and yield, but also on taste, flavor and nutritional value of crops. "Our work has delivered a set of tools to monitor seasonal variations in the metabolism of tea plants", says Al Robbat, "and I'm certain these tools can be used for similar metabolomics studies of crops and human systems".

Using a comparable MPS-GC/MS system, Professor Al Robbat performed the analysis of tea plant extracts and determined secondary metabolites as described in this article.

Reference

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