Evidence for the Effect of the Cocoa Bean Flavour Environment during Fermentation on the Final Flavour Profile of Cocoa Liquor and Chocolate

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Abstract

Preliminary empirical studies have suggested a likely relationship between cocoa pulp flavour attributes and the expression of fruity and/or floral flavours in "fine" or "flavour" cocoa genotypes. In the current study, the aromatic pulp of two tropical fruit species (*Theobroma grandiflorum* and *Anona muricata*) was added to Amelonado cocoa beans during the fermentation process to further assess the possible effect of the cocoa bean flavour environment on the flavour attributes of cocoa nibs and of chocolates. The fruit pulps were added in equal quantities to wet Amelonado cocoa beans in micro-fermentation bags placed in the middle of an Amelonado fermentation mass one or two days after the start of the fermentation process.

Organoleptic analysis of the fermented and dried cocoa nibs, and of cocoa liquors and chocolates made with beans from the mixed micro-fermentations revealed the presence of flavour attributes from *T. grandiflorum* and *A. muricata*. This suggests that the cocoa cotyledon absorbed flavours from the aromatic pulps surrounding the cocoa beans during the fermentation process. The astringency of the chocolates obtained from the mixed fermentations also increased compared to the control treatment. This could be due to transfer of astringency present in the aromatic fruit pulps to the cocoa beans or to the modified micro-fermentation conditions. Evaluation by an amateur public in Montpellier, France, showed levels of preference for the chocolates that had acquired flavours of *T. grandiflorum* and *A. muricata* above that of the control treatment.

These findings constitute indirect evidence that the cocoa cotyledons are capable of absorbing fruity flavours from the cocoa mucilage surrounding the beans during the fermentation process, thus affecting the flavour attributes of the cocoa nibs, of cocoa liquors and of chocolates. This suggests that selection of cocoa genotypes for enhanced fruity flavours can be effectively done through rapid sensory evaluation of cocoa pulps for such flavours.

Furthermore, our findings constitute direct evidence that the sensory quality of cocoa nibs, cocoa liquors and chocolates can be modified substantially by adding aromatic substances to wet cocoa beans during the fermentation process. As far as known to the authors, these results constitute the first published evidence that the cocoa bean flavour environment (mucilage surrounding the beans) can affect expression of flavours in cocoa liquors and chocolates. Further development and testing of this innovative way to produce chocolates with diversified flavour profiles might possibly lead to commercial applications.

Scope

Cocoa flavour quality has been related to genetic and environmental factors (*e.g.* by Clapperton 1992, Lockwood and Eskes 1996, Sukha and Butler 2006). Clapperton *et al.* (1994) reported significant variations among cocoa genotypes and significant parent-offspring correlations for cocoa flavour, astringency, fruity flavour and viscosity. Further studies suggested that there is no effect of the pollen donor (Clapperton 1994) or of the cocoa fermentation environment, as provided by three contrasting cocoa masses (Clapperton *et al.* 1994), on the intensity of cocoa flavour. Recent work on pollen donor

effects by Sukha (2008) on a wider set of flavour attributes failed to detect xenia effects for most of the important flavour attributes *i.e.* cocoa flavour, acidity, fruitiness and floral flavour, but identified small pollen donor effects on astringency, nutty and 'other' flavours. Unpublished experiments undertaken in Brazil carried out in 1998 have shown that depulped cocoa beans, when fermented within a mass of cupuaçu (*Theobroma grandiflorum*) pulp and beans, are capable of absorbing flavours from the cupuaçu pulp (Smilja Lambert pers. comm. 2008).

No studies have been published yet on a possible direct effect of the flavour traits of the cocoa pulp (mucilage surrounding the cocoa beans) on the flavour of the fermented and dried cocoa beans. However, Eskes *et al.* (2007) have reported that significant differences exist in cocoa pulp flavours among genetically different cocoa origins and that human preference for cocoa pulp appeared to be positively correlated with the presence of fruity and/or floral flavours in the pulp and also with the level of sweetness of the pulp. Furthermore, the cocoa genotypes with pulp that was preferred by human beings also appeared to be the cocoa types that are known for their fruity or floral flavours in chocolates made with beans from these cocoa types. This is indirect evidence that the cocoa bean flavours in cocoa liquors and chocolates.

In this study, the direct effect of the cocoa bean flavour environment on the flavour profile of cocoa nibs, of cocoa liquors and of chocolates was assessed through the addition of aromatic pulp of two tropical fruit species to the wet beans of Forastero cocoa (Amelonado) during the fermentation process.

Materials and methods

Pulps obtained from freshly harvested ripe fruits of two tropical fruit species, *T. grandiflorum* (cupuaçu) and *Anona muricata* (sour sop, graviola or guanabana), were used in mixtures with cocoa beans under micro-fermentation conditions. The sweet and acid pulp of cupuaçu is concentrated (creamy), and its typical strong, aromatic, fresh fruit flavour is noticed right from the beginning to the end of tasting. The pulp of sour sop is more liquid, slightly acid, with "green" notes in the beginning and its typical delicate flavour coming through more towards the end. The pulps of these fruits are used in Brazil and elsewhere to produce aromatic ice-creams and fruit juices. Five treatments of micro-fermentation were applied using a fermentation mass made up of wet cocoa beans of the Amelonado cocoa type ("Comun" variety) growing in the farm "Boa Sorte" in Uruçuca, South Bahia, Brazil. The Amelonado variety (Lower Amazon Forastero) is known to produce "bulk" cocoa quality, with intense cocoa flavour, but generally with little or no fruity flavour (International Trade Centre, 1991).

The fermentations were carried out in Ilheus, January 2007, in a wooden box of 50×50×50cm that was filled with healthy, wet cocoa beans. The fermentation mass was turned 48, 72, 96 and 120 hrs after the initiation of the fermentation process and total fermentation duration was 144 hrs (6 days). The micro-fermentations were carried out by placing one kg wet cocoa beans, alone or mixed with an equal quantity of aromatic pulp of cupuaçu and of sour sop, in polystyrene netted bags in the middle of the fermentation mass permitting good exchange of fluids between the fermentation masses inside and outside the bags. The following treatments were applied, each with two replicates:

Treatment 1 (control). Micro-fermentation of a sample of one kg of cocoa beans without addition of aromatic pulp. The cocoa bean sample was obtained by putting beans from the Amelonado fermentation mass into two micro-fermentation bags (replicates) 24 hrs after the beginning of the fermentation process. The micro-fermentation bags were then placed in the middle of the fermentation mass.

Treatment 2. Micro-fermentation of cocoa beans, obtained from the fermentation mass 24 hrs after the start of the fermentation, mixed with fresh cupuaçu pulp. The mixture was placed in two micro-fermentation bags (replicates), each containing one kg of cocoa beans and one kg of cupuaçu pulp. The bags were placed in the middle of the fermentation mass 24 hrs after the start of the fermentation process.

Treatment 3. Micro-fermentation of a mixture of cocoa beans and cupuaçu pulp, as in treatment 2, but with addition of the cupuaçu pulp to cocoa beans taken from the fermentation mass 48 hrs after the start of the fermentation process and with initiation of the micro-fermentation at the same time.

Treatment 4. As treatment 2, but with addition of pulp of sour sop, instead of cupuaçu pulp.

Treatment 5. As treatment 3, but with addition of pulp of sour sop, instead of cupuaçu pulp.

The micro-fermented beans were sun-dried for seven days attaining 8 to 10% moisture content. The cocoa bean samples produced by the five treatments were sent to CIRAD, Montpellier, France, in May 2007, where nib flavour attributes were assessed in May 2007 by the first and third authors. Cocoa liquors and chocolates (65% cocoa and 35% sugar) were prepared by the Guittard Chocolate Company, California, USA, in September/October 2007 and chocolates were sent to Montpellier in November 2007. The cocoa liquors were evaluated by the fourth author in the following treatment sequence: two times tasting of treatment 1 (control), followed by treatments 2, 3, 4 and 5, and than again treatment 1. Chocolate samples were described by the first and third authors without a specific sequence. Overall preferences for the chocolates made with the beans of the five treatments were provided early 2008 through blind testing of the five treatments by approximately 20 chocolate amateurs in Montpellier.

Results

Evaluation of cocoa nibs

The evaluation (sniffing and tasting) of the nibs concentrated on fruity flavours. The nibs of treatment 1 were similar in taste to that of nibs from normal Amelonado-type beans (low acidity, quite bitter and no fruity notes). Fruity flavours were detected in the nibs of treatments 2, 3, 4 and 5 with treatments 2 and 3 demonstrating fresh-fruit flavours with strong resemblance to that of the cupuaçu pulp. The fruity flavour of the beans of treatments 4 and 5 was less pronounced and more delicate, and showed resemblance to that of the fresh sour sop pulp. There was a tendency of the fruity flavours to be more easily detectable in the nibs from the micro-fermentations initiated at 48 hrs after the beginning of the fermentation process (treatments 3 and 5) than at 24 hrs (treatments 2 and 4). The flavour traits appeared to be of similar nature and intensity for each of the two replicates of the treatments. Therefore, only beans from one replicate were used to produce cocoa liquors and chocolates.

Evaluation of cocoa liquors

The flavours found in liquors, as evaluated at the Guittard Chocolate Company by the fourth author (Table 1), show the presence of quite different flavour attributes for the treatments 1 to 5. Treatments 2 and 4 (cupuaçu and sour sop at 24 hrs) do change the flavour profile consistently with the addition of the aromatic pulps. Furthermore, these treatments also seem to alter the overall course of the normal fermentation to yield lower acidity and astringency, and to reduce the woody/bark notes present in the control. Treatments 3 and 5 (cupuaçu and sour sop at 48 hrs) also change the flavour profile, but in a rather in negative way. Acidity has become accentuated and shifted to a more "dirty", astringent, metallic character as well acetic (vinegar-like). Both liquors also lost the original Bahia woody/bark notes. A mild brown fruit flavour was recognised in the liquors of treatment 2 and a fresh floral note was observed with treatment 4. The panellist was not familiar with the specific flavour characteristics of the added pulps.

Evaluation of chocolates

The sensory evaluation of the chocolates carried out at CIRAD in France (Table 1) also revealed major variation for flavour traits. Cocoa flavour was more intense in treatment 1 in comparison to treatments 2 to 5. Strong astringency was present in the chocolates from treatments 3 and 5 (as noted with the liquors). A very low intensity, but complex fruity flavour was identified in the chocolate made from the beans subjected to treatment 1. A much stronger fruity flavour, resembling the typical cupuaçu pulp flavour (as identified by the first author), was noted in treatments 2 and 3, with this type of flavour being described as "yellow prune/cherry like" by the third author (Table 1). The typical sour sop flavour was recognised by the first author in the chocolates of treatments 4 and 5, and this type of flavour was described as "ripe yellow fruit" by the third author. The fruity flavours were perceived as intense in the chocolates derived from the micro-fermentations initiated 48 hrs after the beginning of the fermentation process (treatments 3 and 5) than at 24 hrs (treatments 2 and 4), despite the higher astringency in the chocolates for treatments 3 and 5, and the presence of sour notes for treatment 3.

Chocolate preference of an amateur public

The amateur public in Montpellier, to which the chocolates with cupuaçu and sour sop flavours were presented, generally expressed a preference for the chocolates that contained enhanced fruit-flavour attributes (treatments 2 to 5) in comparison to the chocolate prepared with beans of treatment 1 (control). Persons familiar with the cupuaçu and sour sop fruits recognised the flavours present in the respective treatments carried out with pulp of these fruits. A minority of persons preferred the low-intensity complex fruity flavour in the chocolate of treatment 1.

Discussion

Effect of the pulp flavour environment

The addition of aromatic pulp to the micro-fermentation treatments 2, 3, 4 and 5 had a striking effect on the flavours of the cocoa nibs, liquors and chocolates. The similarity in the flavours of the cupuaçu and sour sop pulp and the fruity flavours observed in the beans and chocolates, as recognised by persons that were familiar with these fruity flavours, suggests that the fruity flavours present in the cocoa pulp environment migrate into the cocoa beans during fermentation. The intensity of the cupuaçu pulp flavour in the beans was perceived to be stronger that that of the sour sop pulp. This is in agreement with the intensity of the typical flavours of these fruit species, with the sour sop flavours being milder than the cupuaçu flavours.

The low intensity of complex fruity flavour identified in the chocolate of treatment 1 (control, beans of Amelonado without fruit pulp) was a surprise, as such is not usually associated with pure Amelonado quality. It might have been caused by some exchange of fluids from the micro-fermentation bags containing cupuaçu and sour sop pulp to the micro-fermentation bags with the beans of the control treatment.

Comparison of liquor and chocolate flavour profiles

The sensory evaluation of the liquors showed the presence of fruity and floral flavours for the treatments 2 and 4, respectively, but not for treatments 1, 3 and 5. On the other hand, with the evaluation of the chocolates fruity flavours were detected for all treatments, and these were strongly expressed in the treatments 2 to 5. To explain the stronger expression of fruity flavours in the chocolates as compared to the liquors, it is hypothesised that the presence of sugars in the chocolates may boost the expression of fruity flavours perceived during sensory evaluation. If this is true, fruity flavours may in general be more easily detected in chocolates than in cocoa liquors.

Effect of the timing of the micro-fermentations

The slightly stronger fruity flavour intensity observed in the nibs from mixed pulp microfermentations initiated 48 hrs after the beginning of the fermentation process (treatments 3 and 5), in comparison to those initiated at 24 hrs (Table 1), might possibly be related to the germination process of the cocoa beans (Rohsius *et al.* 2006). As suggested by these authors, the speed of uptake of soluble compounds by the cotyledon during fermentation is possibly affected by the opening of the testa, as caused by the initiation of germination of the cocoa beans that occurs during the first two days of the fermentation process.

The addition of aromatic pulps during fermentation appeared to reduce cocoa flavour intensity and to increase the acidity and astringency of the liquors and the chocolates, especially so for the micro-fermentation treatments initiated 48 hrs after the start of the fermentation process. The increased acidity and astringency in the 48 hr treatments seem to have masked the expression of the fruity flavours in the cocoa liquors. The 48 hr treatments are apparently interfering more strongly with the normal cocoa bean fermentation process than the 24 hr treatments. These effects may have been caused by the transfer of astringency from the aromatic pulps to the cocoa beans or by a modification in the fermentation conditions caused by the addition of the aromatic pulps. The added pulps will have caused an increase in fermentable sugars, thus increasing the acidity around and in the cocoa beans and thereby probably reducing the activity of polyphenoloxidase and also of the proteolytic enzymes involved in the development of cocoa flavour precursors (Biehl *et al.* 1985; Lee *et al.* 1991). High levels of astringency and low intensity of cocoa flavour have been found to be often associated (*e.g.* Clapperton, 1992) in fermentation experiments.

Prospects for applications

The results suggest that cocoa pulp flavour traits (at least the fruity flavours) can migrate from the pulp to the cotyledons during the fermentation process. This finding opens the door for the possibility to select cocoa varieties for fine/flavour quality based on the evaluation of cocoa pulp flavour profiles, which are known to vary widely among cocoa genotypes (Eskes *et al.* 2007). Such would permit a substantial improvement in the selection efficiency for cocoa flavour traits and enhancement in the capacity of cocoa breeders and farmers to identify new flavours that can possibly be of economic value for the fine/flavour cocoa industry.

As far as known by the authors, our results constitute the first published demonstration of modifications of flavours in cocoa nibs and chocolates through the addition of aromatic fruit pulps to the cocoa beans during the cocoa bean fermentation process. These findings deserve more detailed investigation, especially so with regard to any negative side-effect of the aromatic pulp additions on the outcome of the fermentation process (as appeared to be the case in our study particularly with treatments 3 and 5 as compared to treatments 2 and 4). Once proven effective and consistent, the addition of aromatic substances to the cocoa beans during the fermentation process could open up a vast array of new opportunities to develop cocoa liquors, chocolates or other cocoa-based products with very diverse and special flavour profiles.

The effect of the addition of the cupuaçu and sour sop pulps on the flavours of the chocolates was perceived to be positive by an amateur chocolate public in Montpellier, France. This would mean that the addition of fruit pulps, as well as possibly of other aromatic substances, during the fermentation process may have a potential to positively affect the quality of chocolate and other cocoa products. This suggests that our findings, once tested further, could have implications of commercial interest.

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Table 1. Results of sensory evaluation of cocoa liquors, at the Guittard Company, California, and of chocolates at CIRAD, Montpellier, obtained from Amelonado cocoa beans fermented alone (Treatment 1) or in mixture with pulp of cupuaçu (Treatments 2 and 3) and with pulp of sour sop (Treatments 4 and 5) added to the Amelonado cocoa beans 24 or 48 hrs, respectively after initiation of the fermentation process.

	Flavour description	
Treatment	Cocoa liquors (Guittard evaluation)	Chocolates (CIRAD evaluation)
1	Moderately sour and acetic acid character, mild cocoa notes, moderate bitterness and astringency, with woody notes that persist to the end.	Slight acidity and presence of a low- intensity mild and complex fruity character. Moderate cocoa flavour, bitterness and astringency.
2	Lower front and throughout acidity. Deep flavour profile character with a more astringent, dark wood character. Presence of mild browned fruit notes (like raisins, but with more dried browned fruit).	Pronounced fresh-fruit character, with notes of yellow prunes and slightly fermented cherries. Low cocoa flavour and mild astringency. Slightly alcoholic taste mixed with mild acidity. The fruity character persists in the after-taste and then also develops spicy notes.
3	Sharp acetic/metallic taste upfront and strong astringency. Low in cocoa with some dirty bark notes that persist in the aftertaste.	As treatment 2, but with a stronger fresh- fruit character, in the presence of acidity and even sourness (pungency). Presence of strong astringent end-notes that mix with a persistent fruity, spicy and slightly sour after-taste.
4	Mild sour notes with background acidity and mild cocoa notes. Presence of a fresh, almost white floral note in the centre taste. Astringency comes back in at the end, but is mild without a bark wood character.	Low and pleasant acidity with moderate cocoa flavour and some astringency. Delicate and well-balanced ripe yellow- fruity character that is present from the beginning and persists in the after-taste, developing into a mild 'bouquet' of burnt caramel/fruity/spicy notes.
5	Mild sour notes with early astringency that builds up and becomes dominant at the end. Low in cocoa with some dirtiness in the end and in the aftertaste. No fruit or floral notes as in treatment 4.	As treatment 4, but with stronger acidity and more pronounced astringency. Low cocoa flavour. The fruity character develops together with increasing astringency.