PROFILES OF THE AROMA-RELATED PEPTIDES IN UNFERMENTED AND UNDERFERMENTED COCOA BEANS

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Introduction

The development of cocoa aroma requires that cocoa beans are fermented and dried and then roasted (Rohan, 1964 and Ziegleder and Biehl, 1988). Fermentation results in the production of aroma precursors, such as amino acids, reducing sugars and peptides. Cocoa specific aroma precursors are formed during fermentation by proteolytic prosesses (Ziegleder and Biehl, 1988). The proteolysis of seed proteins and the formation of aroma precursors are strongly dependent on the degree and the time of nib acidification during fermentation. Recent studies on the formation of cocoa-specific aroma precursors have shown that they are derived from the vicillin-class globulin of the cocoa seeds by the coordinate action of an aspartic endoprotease and a carboxypeptidase. In addition to the released (hydrophobic) free amino acids, hydrophilic oligopeptidases were shown to be essential precursors of the typical cocoa aroma. However, up to now, the aroma-related hydrophilic peptides have not been isolated and characterised. Therefore, this report describes aroma-related peptides in unfermented and underfermented cocoa beans.

Materials and Methods

Ripe cocoa pods of the PBC 140 variety were obtained from Perak, Malaysia. The pods were depodded and 45 kg of fresh beans were fermented using shallow wooden boxes (0.78 x 0.78 x 0.78 m) for duration of 72 h. The beans were turned every 24 h. Representative samples of 2 kg each were taken from fermenting box at 0 and 3 days during fermentation. Cocoa powders were prepared from 3 days fermented beans. Samples were defatted and powdered. Unfermented and underfermented cocoa powder (2 g each) were suspended in citric-phosphate buffer (pH 3.5 and 5.8). The suspensions were incubated at 45°C in a shaking water bath for 24 h. After incubation, methanol was added to a final concentration of 70% (v/v). The suspensions were stirred at room temperature for 1 h and centrifuged at 10 000 x g for 15 min. The supernatants were collected and the methanol was removed under pressure at 40°C by rotary evaporator. The aqueous solution was freeze-dried. Peptide mixtures were analysed by reversed-phase HPLC as recently described of Voigt et al. (1994).

Results and Discussion

Peptide patterns from unfermented and undefermented cocoa beans were analysed by reversed phase HPLC. The extract from underfermented cocoa beans obtained by autolysis at pH 3.5 revealed very similar HPLC chromatrogram with unfermented cocoa beans and autolysis at pH 5.8 obtained very similar with underfermented cocoa beans without incubation. However, the proportion of hydrophobic component at pH 3.5 was considerably increased as compared with autolysis at pH 5.8. Analyses of the peptide patterns revealed that the hydrophobic peptides generated during autolysis at pH 3.5 were transformed to considerably more hydrophilic components. The liberation of hydrophobic amino acids are due to the cooperative action of the aspartic endoprotease and the carboxypeptidase present in the ripe cocoa beans (Voigt et al. 1993; 1994). Therefore, the optimum fermentation conditions are dependent on the optimum pH values of the aspartic endoprotease (pH 3.5) and the carboxypeptides (pH 5.8) (Voigt et al. 1994).

Conclusions

The released (hydrophobic) free amino acids, hydrophilic oligopeptides were shown to be essential precursors of the typical cocoa aroma. Raw cocoa with high aroma potentials which are obtained from fermentations with moderate acidification (pH 5.5 - 5.0) contain considerably higher levels of predominantly hydrophobic free amino acids than raw cocoa beans from very acidic fermentations (pH 4.5 - 4.0).

References

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