

BIOCHEMICAL STUDIES OF COCOA SWEATINGS - 1. AMINO ACIDS, MINERALS AND SUGAR ANALYSIS OF THE SWEATINGS

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ABSTRACT

Glucose, fructose and sucrose were the free fermentable sugars found in cocoa sweatings with the total concentration of fermentable sugars ranging from 11.32 - 13.82%. Fifteen free amino acids were identified and the concentrations for most of them were determined. The mineral analysis showed a high potassium level as compared to sodium and calcium. Other elements present were Mg, Zn, Fe, Co, and Mn.

Keywords:

fermentable sugars, amino acids, minerals, cocoa sweatings, chromatography.

INTRODUCTION

The mucilaginous pulp that surrounds fresh cocoa beans yields a sugary juice quite readily on exposure to the atmosphere. This juice called cocoa sweating has not as yet been put to any proper industrial use. Some reports have been made on analysis of the pulp (1,2,3) but a more

comprehensive analysis of the sweatings is lacking. This paper reports a study on the composition of free sugars, amino acids and minerals in the sweatings.

MATERIALS

Fresh cocoa pods of Amazonia, Hybrid and Amelonado varieties were harvested from the plantation of Kwadaso Agriculture Station, Ghana, and used within one hour of harvesting. Only fresh and healthy beans from greenish-yellow pods were used for the collection of the sweatings. The sweatings were collected by a modification of a laboratory apparatus designed by Maclean and Wickens (4). The determination of free sugar, amino acids and minerals was carried out on samples of sweatings collected within the first 30 minutes of the sweating period.

DETERMINATION OF FREE SUGARS

Soluble sugars were extracted from the sweatings in 80% ethanol, concentrated under vacuum, and desalted by re-extracting in pyridine. Pyridine was removed under vacuum and the residue taken up in water. Free sugars were determined by paper chromatographic analysis on Whatman No. 1 papers using Phenol-Ammonia (5) and Butanol-Acetic Acid-Water (12:3:5;v/v/v) (5) as solvents. Sugar spots were identified by their characteristic colours with aniline-diphenylamine, by their R_fs and by comparison with reference sugars.

Ascorbic acid in the sweatings was removed by the addition of ascorbic acid oxidase. Free glucose was then determined by the glucose oxidase method of Hugget and Nixon (6). Sucrose was estimated by the increase of glucose concentration after inversion. Fructose content was calculated as the difference of the total sugar content as determined by the anthrone method of Morris (7) and the sucrose and glucose contents.

DETERMINATION OF AMINO ACIDS

Free amino acids were estimated by automated

ion-exchange chromatography by the method described by JEOL (8). Amino acids were extracted into 80% ethanol solution according to the method of Draper *et al* (9). Separation of the amino acids was carried out employing a 5.8 hour run on a JEOL Amino Acid Analyzer (type JIC-6AH) with a 45x 0.8 cm column of JEOL resin IC-R-2. From the retention time and peak areas of the sample and standard analyses the identification and concentration of amino acids in the sample were determined.

MINERAL ANALYSIS

Organic matter in the sweatings was destroyed by a wet oxidation method using nitric and sulphuric acids (10). Minerals were determined by atomic absorption (Cu, Co, Fe, Mg, Mn, Zn) using a Hilger and Watts Atomspok atomic system and by flame photometry (Ca, K, Na) using a Gallenkamp Flame Analyzer FH-500.

RESULTS AND DISCUSSION

Glucose, fructose and sucrose were identified as the only free sugars present in the sweatings. The free sugars in the sweatings are therefore the same as the pulp sugars (glucose, fructose, sucrose) as reported by Chatt (11). The concentration of the sugars are shown in Table 1. The total free sugars (all fermentable) obtained from the 3 varieties of cocoa were; Hybrid 13.82% and Amelonado 11.32%.

TABLE 1:

Sugar composition of cocoa sweatings from different varieties of cocoa

| Cocoa Variety | % Free Sugars in Cocoa Sweatings | | |
|---------------|----------------------------------|----------|---------|
| | Glucose | Fructose | Sucrose |
| Hybrid | 4.21 | 4.31 | 5.30 |
| Amelonado | 2.71 | 5.41 | 3.43 |
| Amazonia | 3.82 | 5.55 | 4.07 |

These values are slightly short of the total pulp sugar content reported by Minifie (3) to be 14.07%. This relatively high sugar content places cocoa sweatings in the category of fruit juices that exhibit a significant potential as a large-scale fermentation material.

Twenty-one peaks were observed and recorded on the analysis of the amino acids. Thirteen of these peaks were identified as amino acids and quantified and the results are shown in Table 2. The eight unidentified peaks were very small and may therefore be assumed to be

amino acids and/or peptides present in trace amounts. Forsyth (2) reported the presence of three amino acids in the pulp. The presence of asparagine determined by paper chromatography confirms the report of Forsyth.

TABLE 2

Amino acid composition of cocoa sweatings (Amazonia variety).

| Amino acid | Concentration (U mole/ml) |
|---------------|------------------------------|
| Aspartic acid | 0.697 |
| Serine | 2.503 |
| Glutamic acid | 0.649 |
| Proline | 0.277 |
| Glycine | 0.015 |
| Alanine | 0.081 |
| Cysteine | 0.006 |
| Methionine | 0.006 |
| Isoleucine | 0.006 |
| Leucine | 0.022 |
| Phenylalanine | 0.036 |
| Histidine | 0.009 |
| Arginine | 0.058 |

The results of the mineral analysis given in Table 3 showed a higher content of potassium than sodium and calcium which is typical of some plant materials. Bediako (12) and Ankrah (13) have reported a very high potassium content (19000-37000 ppm) in cocoa husks another waste product from the cocoa industry.

TABLE 3:

Mineral composition of cocoa sweatings (Amazonia variety)

| Mineral | Concentration (ppm) |
|---------|---------------------|
| K | 2633.0 |
| Mg | 288.0 |
| Na | 38.0 |
| Ca | 25.0 |
| Fe | 4.7 |
| Zn | 5.0 |
| Co | 3.2 |

| | | | |
|----|-----|-----|-----|
| Mn | ... | ... | 0.8 |
| Cu | ... | ... | 0.0 |

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