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BIOACTIVE INGREDIENTS IN COCOA AND CHOCOLATE PRODUCTS AND THEIR HEALTH PROMOTING PROPERTIES

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Introduction

There is a growing interest in the food industry and nutritional science as well as in preventive health care in the evaluation and development of natural antioxidants and radical scavengers from plant materials. Thereby, considerable interest is focused on plants rich in polyphenol compounds which are well-known for their significant antioxidant and radical scavenging activity resulting in multiple beneficial nutritional-physiological and health effects in the human organism.

Cocoa and chocolate products are popular consumer goods. They had been identified as a rich source of dietary polyphenols which gained much interest recently due to their antioxidant capacity and their health benefits. It has been shown that chocolate especially dark chocolate with high contents of cocoa is one of the most polyphenol-rich foods along with tea and wine. Numerous studies have already been performed on different aspects of the antioxidant properties and health benefits of cocoa and cocoa products in regard to cardiovascular diseases, plasma antioxidant activity, low density lipoprotein (LDL) oxidation, blood pressure, arterial flow mediated dilation and platelet aggregation.

The objective of this study was to quantify the total polyphenol compounds, flavonoids, catechins as well as proanthocyanidins and to evaluate the antioxidant and free radical scavenging properties of cocoa powder and various chocolate products with different cocoa fractions ranging between 32% and 85%.

Materials and Methods

Cocoa and chocolate products which were manufactured and distributed in Austria were analyzed for their content of total polyphenols, flavonoids, catechins and proanthocyanidins by standardized photometric methods. The antioxidant activities were determined with the DPPH\(^+\) radical scavenging method (Inhibitory Concentration IC\(_{50}\)) as well as with the ABTS-radical assay (Trolox Equivalent Antioxidant Capacity TEAC).

Cocoa and chocolate products:
Cocoa powder
Chocolate powder (32% cocoa)
Baking chocolate (59% cocoa)
Semi-sweet chocolate (62% cocoa)
Bitter sweet chocolate (85% cocoa)

Preparation of extracts
The cocoa and chocolate products were defatted with n-Hexan and extracted with water/methanol (80:20) for analysis of the polyphenol substances and antioxidant activity.
**Determination of total polyphenols**
The content of total polyphenols was determined photometrically according to the Folin-Ciocalteu method and expressed as gallic acid equivalents (1).

**Determination of flavonoids**
The content of flavonoids was determined photometrically as aluminium chelate complex using quercetin as reference (2).

**Determination of catechins**
The content of catechins was determined photometrically after reaction with DAC using (+)-catechin as reference (3).

**Determination of proanthocyanidins**
The content proanthocyanidins was determined photometrically after depolymerisation in an acidic environment and expressed as cyanidin equivalents (4).

**Determination of antioxidant activity and radical scavenging capacity**
The antioxidant activities and radical scavenging properties were determined with the DPPH* radical scavenging method in terms of their Inhibition Concentration IC₅₀ representing the amount of antioxidant necessary to decrease the initial DPPH* concentration by 50%. Low IC₅₀ values are indicating high radical scavenging activity (5).

Furthermore, TEAC (Trolox Equivalent Antioxidant Capacity) values were determined using the ABTS method (6). High TEAC values are indicating high antioxidant activity.

**Results and Discussion**

**Content of polyphenol substances**

In the different cocoa and chocolate products the content of total polyphenols was determined in the range of 6.9 to 23.1 mg/g with the highest amounts in cocoa powder followed by the different chocolate products in the order of their cocoa fraction percentage. The same trend was observed regarding their content of flavonoids (2.0 – 7.9 mg/g), catechins (1.0 – 2.6 mg/g) and proanthocyanidins (1.5 – 3.1 mg/g). Generally, there is a good correlation between the cocoa content in the products and the content of polyphenol substances (Fig. 1).

**Antioxidant and free radical scavenging capacity**

All the cocoa and chocolate products showed significant antioxidant and free radical scavenging activity which was calculated by the respective IC₅₀-value as well as the TEAC-value. Low IC₅₀-values and high TEAC-values are indicating high antioxidant activity. The highest antioxidant capacity was observed in pure cocoa powder (IC₅₀: 59 mg/l; TEAC: 396 μmol Trolox/g) followed by the chocolate products in order and correlation of their cocoa fraction percentage (IC₅₀: 104-207 mg/l; TEAC: 120-190 μmol Trolox/g) as shown in Fig. 2 and Fig.3.
Figure 1: Total polyphenols, flavonoids, catechins and proanthocyanidins in cocoa and chocolate products

Figure 2: IC₅₀ values of cocoa and chocolate products
Conclusion
Pure cocoa powder and chocolate products with high cocoa fraction percentage are consumer goods with a high content of total polyphenol substances including flavonoids, catechins and proanthocyanidins.

The cocoa and chocolate products show significant high antioxidant activity and radical scavenging capacity. In correlation to the content of polyphenol substances pure cocoa powder shows the highest antioxidant potency followed by the chocolate products in the order of their cocoa fraction percentage.

There is a good correlation between the antioxidant activities and radical scavenging properties determined by the DPPH-method expressed as IC_{50} and by the ABTS-method expressed as TEAC-values.

The results of this study indicate that cocoa and chocolate products are a major source of dietary antioxidants and consumption of these products offer a potential beneficial impact in maintaining and promoting human health.

References