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Short communications

Polyphenol antioxidants in commercial chocolate bars: Is the label accurate?

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ABSTRACT

Polyphenols in cocoa have been shown to be responsible for beneficial health effects, especially in the cardiovascular area. Many people consume their chocolate in the form of bars, readily available in retail stores. The only information that may be useful to the consumer in choosing a healthier bar, with the exception of the nutrients, is the % cocoa solids on the label. We have examined the polyphenols in commercial bars by use of two antioxidant assays and corrected that value for non-fat cocoa solids, the source of the polyphenols in the chocolate. We also separated and analyzed by HPLC the two major monomeric polyphenol antioxidants, epicatechin and catechin. We found a significant and linear relationship between label % cocoa solids and the antioxidant assays as well as the sum of the monomers. Consumers can thus rationally choose chocolate bars based on % cocoa solids on the label.

1. Introduction

Until the 1990s, chocolate had been regarded as nothing more than an unhealthy sweet. A 1994 publication showed that the saturated fat stearic acid in chocolate did not raise serum lipids (Kris-Etherton & Mustad, 1994). In 1999, a blinded analysis study showed that dark chocolate bars contained significantly more of the health-promoting polyphenols than milk chocolate (Vinson, Proch, & Zubik, 1999). In 2006, an article was published that showed that cocoa inhibited atherosclerosis in an animal model and cocoa was found to be the third leading source of polyphenol antioxidants in the US diet (Vinson et al., 2006). Consuming polyphenols in a cocoa muffin and dark chocolate bits produced a post-prandial human *in vivo* antioxidant effect in spite of the presence of fat and sugar prooxidants in the products (Vinson et al., 2006). Now people are thinking of chocolate as a functional food.

There has been a recent spate of research regarding the cardiovascular health benefits of chocolate, as a current PubMed search found over 4700 citations for "chocolate". A meta-analysis of 114,000 subjects found that the highest chocolate consumption reduced the risk of heart disease and stroke deaths by over 30% (Buitrago-Lopez et al., 2001). Furthermore, a clinical study showed that even a very small dose (6 g) of dark chocolate produced a small but significant decrease in

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blood pressure after 18 weeks in subjects with pre- or stage 1 hypertension (Taubert, Roesen, Lehmann, Jung, & Schömig, 2007). Another investigation found that 10 grams of a 70% cocoa solid chocolate for 1 month induced multiple cardiovascular benefits for young healthy subjects, including improvements to blood pressure, flow mediated dilation, pulse wave velocity, and arterial stiffness index (Pereira et al., 2014). A different group investigating the same cardiovascular benefits proved that polyphenols in cocoa, not nutrients, were responsible for the beneficial changes (Grassi et al., 2014). They showed in a double-blind crossover study with healthy subjects that a low calorie 10 g dose of cocoa containing increasing levels of polyphenols caused a dose–response improvement in the parameters. Chocolate has also been linked to improvement of chronic diseases. For instance, dark chocolate (but not milk chocolate) increased nitric oxide and improved walking autonomy in patients with peripheral artery disease (Loffredo et al., 2014). Only dark chocolate increased serum epicatechin and its metabolites, which are the putative bioactives. Chocolate consumption was also found to decrease mortality following an initial myocardial infarction (Janszky et al., 2009). The flavanols contained in chocolate were reviewed as mood enhancers (Smith, 2013). Another review highlighted the mechanisms of the flavanol metabolites in the brain which slow the cognitive decline in aging and improve cognitive function in healthy subjects (Sokolov, Pavlova, Klosterhalfen, & Enck, 2013).

The increased publicity of these studies and the possibility that chocolate consumption can affect surrogate markers of heart disease may ultimately lead manufacturers to a health claim for chocolate. A chocolate bar, rather than cocoa, is the preferred product consumed in many cultures. What chocolate bar should the informed consumer purchase, given the large number of domestic and foreign chocolates available in the marketplace? At present, the labeling on many chocolate bars lists the % cocoa solids. Since cocoa solids represent the source of bioactive polyphenols (flavanols) in the bar, the number may indicate the level of these compounds in the product. We hereby present evidence that this is the case.

2. Materials and methods

Commercial chocolate bars (n = 46), both labeled and unlabeled with % cocoa solids, were obtained from local markets and the internet. Five manufacturing countries (four from Europe and the USA) and seven single countries of origin were represented. Fat varied from 28 to 64%, while cocoa solids listed on the label ranged from 30 to 100%. There are two common methods available to analyze the polyphenols in chocolate. We measured the total amount of polyphenol antioxidants in chocolate in duplicate as catechin equivalents using two different antioxidant assays, the single reagent Folin method (Vinson, Proch, & Bose, 2001) and a free radical antioxidant power (FRAP) method using catechin as the standard. The antioxidant concentration was corrected for label fat content since nonfat cocoa solids are the origin of the polyphenol antioxidants in chocolate bars. Another more complicated and expensive method is the HPLC assay of the individual compounds, especially the putative bioactive flavanols catechin and epicatechin, which we used on 17 of the bars. For the HPLC assay, we used our sample preparation method (Vinson et al., 1999), followed by gradient elution at 1 ml/min from 100% water/1% acetic acid to 100% methanol/1% acetic acid over 30 min with a Phenomenex 25 cm Luna C18 column (Torrance, CA) at 280 nm.

3. Results

The labeled chocolate bars (n = 33) ranged from 30 to 100% cocoa solids containing 21.6–90.4 μmol/g of Folin catechin equivalents and 24.6–189 μmol/g FRAP catechin equivalents. Dark chocolate contained significantly more polyphenol antioxidants than milk chocolate, $p < 0.001$, as previously shown (Vinson et al., 1999). Products with % cocoa solids labeling had significantly more antioxidants than those without labeling (n = 13), $p < 0.001$. For single manufacturers with products of differing % cocoa solids, the antioxidant content always increased with the % cocoa solids. Independent of the type of chocolate, manufacturer, or country of origin, there was a positive linear correlation between polyphenol concentration and % cocoa solids for 31 products for both the Folin and FRAP data ($p < 0.001$). One chocolate manufactured by a patented processing technique with 72% cocoa solids contained the most total polyphenols of all chocolate bars. Other published research found linear relations with Folin but used a complicated analyzed value for non-fat cocoa solids (Cooper et al., 2008; Miller et al., 2006). The label % cocoa solids were also highly correlated with the sum of the major flavanols catechin and epicatechin as measured by HPLC (0.680, $p < 0.001$) (Cooper et al., 2008; Miller et al., 2006).

A choice the consumer can make would be whether to buy a foreign or domestic produced chocolate, with the latter probably being less expensive. A comparison of 13 labeled domestic *versus* 20 foreign chocolate bars showed that there was a statistically significantly greater amount of Folin and FRAP antioxidants in the foreign products ($p < 0.01$). A paired comparison of domestic and foreign bars with identical % cocoa solids, 25, 60 and 72% also showed a significantly greater amount of antioxidants in the foreign products ($p < 0.05$).

For consumer information, the data in Fig. 1 with a slope of almost 1 show doubling the % cocoa solids will double the amount of antioxidants found in the chocolate bar. While consumers need only look for a dark chocolate with % cocoa solids to get a high polyphenol antioxidant chocolate, they should also pay attention to the caloric content. A recent long-term supplementation study showed a slight but significant weight gain at the high dose (25 g) of chocolate, but a beneficial blood pressure lowering effect of the low dose (6 g) with no effect on weight gain (Desch et al., 2010). In addition, a 6 year epidemiological study found that habitual chocolate consumption may increase body weight in a dose–response manner (Greenburg & Buijsse, 2013). It is important for the consumer to know that dark chocolate consumption promotes more satiety, lowers the desire to eat something sweet, and suppresses energy consumption compared with milk chocolate (Sørensen & Astrup, 2011).

Although detrimental effects, such as weight gain, may arise from frequent chocolate consumption, there are also many