



Molecular and metabolite analyses of *Coffea arabica* L. fruits to identify candidate genes for diterpene biosynthesis

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Coffee is consumed by more than 800 million people who enjoy its taste, flavor, and health benefits. Cafestol (CAF) and kahweol (KAH) are two diterpenes found exclusively in the *Coffea* genus. It has been suggested that CAF/KAH are initially produced in the perisperm of the developing fruit, and after maturation they accumulate in the endosperm. Several nutraceutical effects on human health are described for CAF/KAH, such as antioxidant, anticarcinogenic and anti-inflammatory properties. However, studies also suggest that CAF may also increase cholesterol levels in blood, highlighting the importance of understanding their biosynthesis and functions. We used an integrated analysis of CAF/KAH metabolite accumulation and gene expression analyses during fruit development to identify the diterpene synthases (diTPS) and cytochrome P450 (P450) genes responsible for CAF/KAH biosynthesis in *Coffea arabica* L. The expression of several class I and II diTPS and P450 families showed a strong correlation with CAF/KAH biochemical profile and were selected for cloning, and functional characterization using heterologous protein expression and GC-MS technologies. This study aims to unravel the genes involved in CAF/KAH biosynthesis and provide the tools for manipulating CAF/KAH levels in coffee beans. It will also open the possibility to develop plants with desirable content of CAF/KAH and improve beverage quality focusing on human health.