

Title: Purification and characterization of 3-O-methyl-D-glucose from the seed coat of *Vigna mungo* (L.) Hepper

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Abstract: *Vigna mungo* L (Black gram) belonging to the [Leguminosae](#) family is a most abundant source of protein for human and animal diets. Over 8.5 million tonnes of black gram are being produced worldwide, and after confiscating the [endosperm](#) for the protein source, the seed coat is dumped as agro-waste and used as feed for the cattle. This study aims to investigate and profile the metabolites in three distinct parts of *V. mungo* seeds: coat, [cotyledon](#), and intact seed. Also, it has been hypothesized that sugar [methylation](#) plays a pivotal role in facilitating black gram [seed germination](#) by serving as an energy source during metabolic processes. In our study, A total of forty-nine compounds belonging to the class such as fatty acids, saccharides, [triterpenes](#), sterols, and vitamins were detected in various parts of *V. mungo* seed by gas chromatography-mass spectrometry (GC-MS). Most interestingly, 3-O-methyl-D-glucose (3-OMG) was commonly detected in methanolic extract of coat, intact seed, and [cotyledon](#) with peak area percentages of 3.076, 1.157, and 1.100 %, respectively. Based on the highest peak area percentage in the coat, this compound was purified by using a [silica gel](#) column with a mesh size of 60–120 followed by Biotage SNAP cartridge KP-C18-HS (12 g). The yield and purity of the purified fractions were found to be 11.78 & 94.19 %, respectively by high-performance [liquid chromatography](#) (HPLC) analysis. Further, the mass confirmation of purified fraction containing 3-OMG (m/z - 217.06 g/mol) was done by liquid chromatography-mass spectrometry (LC-MS/MS) and reconfirmed with matrix-assisted laser desorption/ionization time-of-flight [mass spectrometry](#) (MALDI-TOF/MS). The MS/MS analysis of the fragment showed m/z 206.91 for 3-OMG which corresponds to the association of the [sodium](#) adduct. The concentration of 3-OMG was quantified as 332.65 µg/g of dry weight in the coat of *V. mungo* by HPLC analysis. Also, among the antioxidant assays conducted, such as [DPPH](#), [ABTS](#), FRAP, and [NOS](#), it was observed that the methanolic extract of coat exhibited the most significant [antioxidant activity](#). The 3-OMG is commonly used as a non-metabolized analogue of glucose by regulating glucose transporter, especially GLUT-4 that regulates the absorption of glucose by insulin into muscle and fat cells. This study supports the black gram seed with coat as an alternative medicine to regulate [blood glucose](#)

Key words: *Vigna mungo*, Black gram, Seed coat, Metabolite profiling, 3-O-methyl-D-glucose (3-OMG)

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