RNAI BLOCKAGE OF DESOXYHEMIGOSSYPOL-6-OMT DECREASES THE AMOUNTS OF METHYLATED SESQUITERPENOIDS IN TRANSGENIC COTTON ROOTS

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<u>Abstract</u>

After pathogen attack, plants produce compounds, called phytoalexins that are toxic to microorganisms. In the case of the cotton plant, these include desoxyhemigossypol (dHG), desoxyhemigossypol-6-methyl ether (dMHG), hemigossypol (HG), and hemigossypol-6-methyl ether (MHG), written in the order of toxicity. Non-methylated compounds are more toxic to fungi than their methylated counterparts. We previously identified, cloned and sequenced the desoxyhemigossypol-6-O-methyltransferase (dHG-6-OMT) gene from Gossypium barbadense that is responsible for the conversion of dHG to dMHG. Our objective is to suppress expression of dHG-6-OMT using an RNAi construct, and determine the disease resistance of these RNAi lines. Eight plants from 6 independent transformation events were generated. Analysis of a small amount of roots from these healthy plants indicated that the methyl transferase was suppressed. Between 32 and 45% of total sesquiterpenoids are methylated in wild type roots. In contrast, only 0 to 20% of the total sesquiterpenoids are methylated in the roots of T_0 RNAi plants. Healthy roots from the F₁ progeny of one of these lines have been tested, and the RNAi transgene co-segregates with reduced root sesquiterpenoid methylation in the F_1 progeny. 1.4% of sesquiterpenoids are methylated in the RNAicontaining plants compared to 27.2% methylation in the WT siblings. The loss of methylated sesquiterpenoids in the roots of RNAi-containing plants leads to a reduction in total terpenoids (68% of WT levels). In the future, the RNAi-containing lines will be tested for pathogen resistance, and the roots will be analyzed for increased levels of sesquiterpenoids compared to the WT.