Utilizing Genome Editing Technologies to Develop Ultra-low Nicotine Tobacco Lines

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The alkaloid nicotine is the most distinguishing feature of tobacco plants and comprises approximately 90% of the total alkaloids found in most tobacco plants. Due to the addictiveness of nicotine, the US Food and Drug Administration (FDA) is considering imposing regulations that would require the nicotine content of future cigarette products to be much lower than what is currently on the market. The level of nicotine being considered by the FDA for their proposed mandate is 0.4 mg/g within the tobacco filler, an amount that is approximately 95% lower than exists in cigarettes currently on the market. Although naturally occurring genetic mutations exist that can reduce the nicotine content of tobacco plants, it would be difficult to develop nicotine varieties through traditional breeding that meet this stringent ultra-low nicotine standard. In this research project, genome editing technologies were applied to mutate key steps in the nicotine biosynthetic pathway to explore new alternatives for generating ultra-low nicotine tobaccos. Of the various nicotine reducing strategies tested, combining mutations in genes encoding methylputrescine oxidase (MPO) activity with mutations in genes encoding berberine bridge-like (BBL) enzymes holds the greatest promise as means for obtaining ultra-low nicotine tobacco cultivars that retain acceptable agronomic properties.