Edible plants as oral “vaccines”

Description

Abstract

Plants are promising candidates as bioreactors for the production of oral recombinant proteins in the biopharmaceutical industry. As an initial step toward provision of an oral vaccine against the severe acute respiratory syndrome coronavirus (SARS-CoV), we have expressed a partial spike (S) protein of SARS-CoV in the cytosol of nuclear-transformed plants and in the chloroplasts of plastid-transformed plants. In the construction of both nuclear and plastid transformation vectors, a 2-kilobase nucleotide sequence encoding amino acids 1-658 of the SARS-CoV spike protein (S1) was modified with nucleotide changes, but not amino acid changes, to optimize codon usage for expression in plants. To investigate the subcellular localization of S1 during transient expression in tobacco leaves, a translational fusion consisting of S1 and the green fluorescent protein (GFP) was generated. Following agroinfiltration of tobacco leaves, analysis by laser confocal scanning microscopy revealed that the S1:GFP fusion protein was localized to the cytosol. In stable transgenic tobacco plants and lettuce plants generated by Agrobacterium-mediated transformation, tobacco and lettuce leaves were observed to express the S1 at high levels from the Cauliflower Mosaic Virus 35S promoter with Northern blot analysis. When the S1 was expressed in transplastomic tobacco, S1 messenger RNA and its corresponding protein were detected on Northern and Western blot analyses, respectively. Our results demonstrate the feasibility of producing S1 in nuclear- and chloroplast-transformed plants, indicating its potential in subsequent development of a plant-derived and safe oral recombinant subunit vaccine against the SARS-CoV in edible plants.

Li, H.-Y., Ramalingam, S., & Chye, M.-L.. (2006). Accumulation of Recombinant SARS-CoV Spike Protein in Plant Cytosol and Chloroplasts Indicate Potential for Development of Plant-Derived Oral Vaccines. Experimental Biology and Medicine

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“Plants are promising candidates as bioreactors for the production of oral recombinant proteins in the biopharmaceutical industry. as an initial step toward provision of an oral vaccine against the severe acute respiratory syndrome coronavirus (sars-cov), we have expressed a partial spike (s) protein of sars-cov in the cytosol of nuclear-transformed plants and in the chloroplasts of plastid-transformed plants. in the construction of both nuclear and plastid transformation vectors, a 2-kilobase nucleotide sequence encoding amino acids 1–658 of the sars-cov spike protein (s1) was modified with nucleotide changes, but not amino acid changes, to optimize codon usage for expression in plants. to investigate the subcellular localization of s1 during transient expression in tobacco leaves, a translational fusion consisting of s1 and the green fluorescent protein (gfp) was generated. following agroinfiltration of

tobacco leaves, analysis by laser confocal scanning microscopy revealed that the s1:gfp fusion protein was localized to the cytosol. in stable transgenic tobacco plants and lettuce plants generated by agrobacterium-mediated transformation, tobacco and lettuce leaves were observed to express the s1 at high levels from the cauliflower mosaic virus 35s promoter with northern blot analysis. when the s1 was expressed in transplastomic tobacco, s1 messenger rna and its corresponding protein were detected on northern and western blot analyses, respectively. our results demonstrate the feasibility of producing s1 in nuclear- and chloroplast-transformed plants, indicating its potential in subsequent development of a plant-derived and safe oral recombinant subunit vaccine against the sars-cov in edible plants."
Pogrebnyak, N., Golovkin, M., Andrianov, V., Spitsin, S., Smirnov, Y., Egolf, R., & Koprowski, H.. (2005). Severe acute respiratory syndrome (SARS) S protein production in plants: Development of recombinant vaccine. Proceedings of the National Academy of Sciences

, 102(25), 9062–9067.
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"In view of a recent spread of severe acute respiratory syndrome (sars), there is a high demand for production of a vaccine to prevent this disease. recent studies indicate that sars-coronavirus (cov) spike protein (s protein) and its truncated fragments are considered the best candidates for generation of the recombinant vaccine. toward the development of a safe, effective, and inexpensive vaccine candidate, we have expressed the n-terminal fragment of sars-cov s protein (s1) in tomato and low-nicotine tobacco plants. incorporation of the s1 fragment into plant genomes as well as its transcription was confirmed by pcr and rt-pcr analyses. high levels of expression of recombinant s1 protein were observed in several transgenic lines by western blot analysis using specific antibodies. plant-derived antigen was evaluated to induce the systemic and mucosal immune responses in mice. mice showed significantly increased levels of sars-cov-specific iga after oral ingestion of tomato fruits expressing s1 protein. sera of mice parenterally primed with tobacco-derived s1 protein revealed the presence of sars-cov-specific igg as detected by western blot and elisa analysis."
Li, H.-Y., & Chye, M.-L.. (2009). Use of GFP to Investigate Expression of Plant-Derived Vaccines. In Methods in Molecular Biology

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"Plants are low-cost bioreactors for the production of various biopharmaceuticals including oral vaccines. plant-derived oral vaccines are potentially useful in combating viral infections involving mucosal immunity. transgenic plants have been generated to successfully produce mucosal vaccines against cholera, hepatitis b, foot-and-mouth disease, and norwalk virus. as a first step toward the generation of oral vaccines against the severe acute respiratory syndrome coronavirus (sars-cov), we

have expressed a recombinant s1 protein of the sars-cov in transformed tobacco. since plant transformation and regeneration of stable transformants require considerable time, we initially used a green fluorescent protein (gfp) to tag the antigen in transient expression. gfp was fused to the carboxy-terminus of s1 for expression of s1-gfp to show expression of recombinant s1 by agroinfiltration of tobacco leaves. the gfp tag enables a relatively quick confirmation of antigen expression in plant cells by fluorescent microscopy. such analysis using gfp that precedes stable plant transformation will enable the rapid screening of multiple constructs to attain optimal recombinant protein expression. furthermore, this approach determines the subcellular localization of the recombinant protein in plant cells, providing information on optimal subcellular targeting for production in plant bioreactors. © 2009 humana press, a part of springer science + business media, llc."

Category

1. General

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