


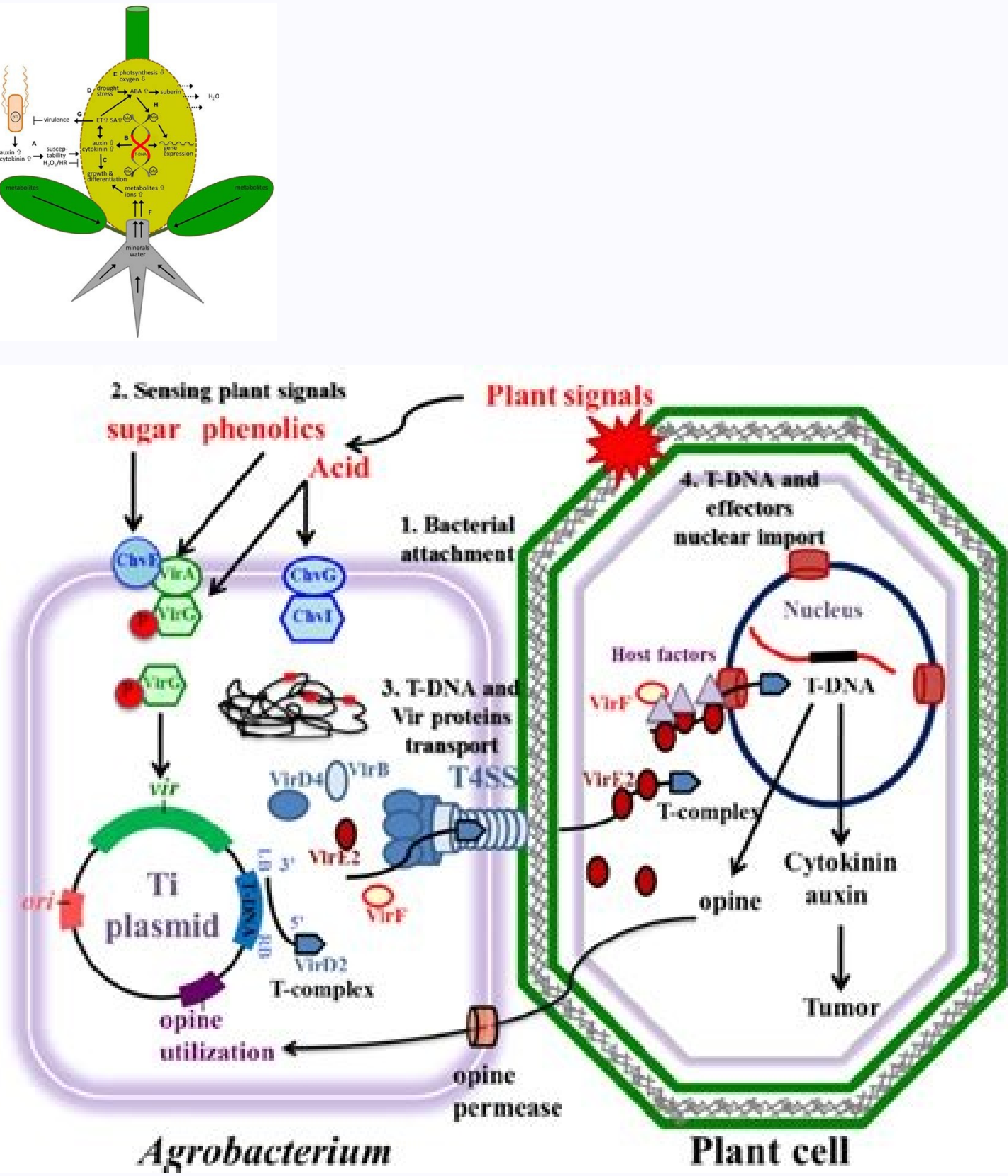
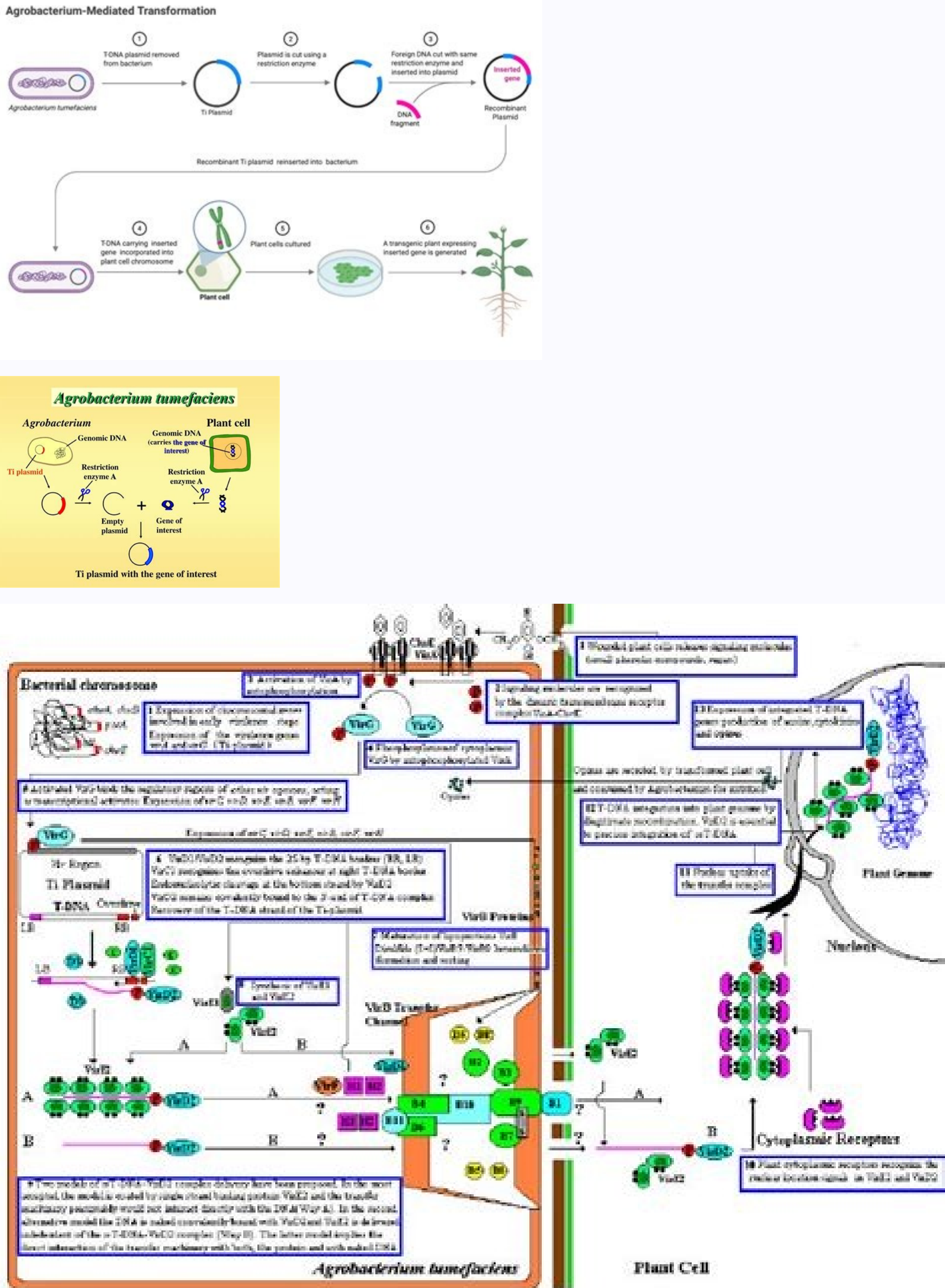
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Agrobacterium tumefaciens plant transformation



What is the role of agrobacterium tumefaciens in plant transformation. Agrobacterium tumefaciens-mediated transformation of the plant pathogenic fungus magnaporthe grisea. What is the role of agrobacterium tumefaciens in plant transformation class 12. What is agrobacterium tumefaciens. Agrobacterium tumefaciens a natural tool for plant transformation. How to grow agrobacterium tumefaciens. How to use agrobacterium tumefaciens in plants. Plant transformation using agrobacterium tumefaciens.

1 and ver. Indeed, the ethylene levels in the plant tissues during the transformation were reduced by the A. 166 (1), 175–176, 11, 89–95, pBBR1MCS5, pBBRacDS, pBBRgabt, and pBBRacSgabt were introduced into A. C-E: A. P., Nakamura, I., Mii, M. A., Lim, C. doi: 10.1093/pcp/pcr041PubMed Abstract | CrossRef Full Text | Google Scholar Sainsbury, J., Thuenemann, E. (2009). P., Yu, A., Renou, J. doi: 10.1126/jb.173.17.5260-5265.199PubMed Abstract | Google Scholar Sheip, B. torquaglin, D. (2010). For hybridization, a digoxigenin (DIG)-labeled DNA probe, specific for pTi (0.8 Kb), was used. doi: 10.1016/S0176-1617(11)80343-3CrossRef Full Text | Google Scholar Dehliens, R., Beyerle, B., De Greve, H., Deboeck, F., Schell, J., Van Montagu, M., et al. 1, ver. (2010). 110 (2), 678–683. Other negative factors in Agrobacterium-plant interactions, aside from ethylene and GABA, have been reported by previous studies (Liu and Nester, 2006; Yuan et al., 2007; Yuan et al., 2008; Anand et al., 2008). tumefaciens, GABA taken into bacterial cell would be degraded. R., Soto, C. doi: 10.1038/318624a0CrossRef Full Text | Google Scholar Stachel, S. N., Kang, L., Tang, Y., et al. lycopersicum “Micro-Tom” were observed. doi: 10.1073/pnas.1807482115 CrossRef Full Text | Google Scholar Komari, T. Therefore, the activities of AcDS and GabT in V4-E and V4-G, were enough to increase the T-DNA transfer frequencies in E. (A) Map of a plasmid for the expression of ACC deaminase (acdS) and GABA transaminase (gabT) in A. 139 (3), 309–312. J., Uchi, S., Watanabe, S., Ezura, H. lycopersicum “Micro-Tom.” Almost 100 explants of “Micro-Tom” were inoculated for each bacterial strain [(C-G), (V1-G), (V3-G), and (V4-G)]. 343, 15–41, 31, 805–813. tumefaciens and triggers the degradation of the quorum-sensing (QS) signal, resulting in the reduced horizontal gene transfer of the Ti plasmid and the aggressiveness of the plant host (Chevrot et al., 2006; Haudecoeur et al., 2009). bronchiseptica; pBBR1 Rep, protein for replication required by pBBR1 oriV, GmR, Gentamicin resistance gene. Front Microbiol. Biochem. Antibiotics were added at the following final concentrations: ampicillin at 100 µg/ml, gentamicin at 50 µg/ml, spectinomycin at 50 µg/ml, and kanamycin at 50 µg/ml. (2018). Plant-Agrobacterium interaction mediated by ethylene and super-Agrobacterium conferring efficient gene transfer. (1996). doi: 10.1007/s00299-018-2350-1PubMed Abstract | CrossRef Full Text | Google Scholar Hoshikawa, K., Ishihara, G., Takahashi, H., Nakamura, I. (B) GUS stained explants of S. HE and KN critically revised and approved the manuscript for publication.FundingThis research was supported by grants from the New Energy and Industrial Technology Development Organization (NEDO) to HE and from JSPS KAKENHI (Grant Numbers JP24780001 and JP19K05964) to SN. The vector maps were described in Supplemental Figure 1. tumefaciens strain with improved potential for transformation by imbuing it with the ability to remove ethylene and GABA, which are negative factors in the Agrobacterium-plant interactions. Plant Cell Rep. 1 increased the transformation frequency up to 3.2 and 2.8 times in E. 4 were evaluated in Erianthus ravenae, Solanum lycopersicum “Micro-Tom,” Nicotiana benthamiana, and S. tumefaciens strains were grown at 28°C in Luria Broth (LB) medium (1% bacto-tryptone, 0.5% yeast extract, and 0.5% NaCl). The ternary transformation system: constitutive virG on a compatible plasmid dramatically increases Agrobacterium-mediated plant transformation. Since both activities have different effective points in the transformation process, Super-Agrobacterium ver. 294 (5550), 2317–2323. The purified DNA was digested with HindIII, electrophoretically separated in 0.8% agarose gel, and transferred onto Gene Screen Plus nylon membranes (Roche Diagnostics, Basel, Swiss) with 20× saline-sodium citrate (SSC) buffer. lacP, lac gene promoter from E. Coomassie Brilliant Blue staining (bottom panel) is shown as an internal control. These results indicate that introducing the acdS and gabT at the same time in A. tumefaciens with GabT activity locally decreased GABA content in the plant calli and maintained higher shoot regeneration frequencies. P., Zarei, A., Deyman, K. doi: 10.5511/plantbiotechnology.12.0125aCrossRef Full Text | Google Scholar Hu, X., Zhao, J., DeGrado, W. This showed that in the N. P., et al. Bars represent the standard deviation (n = 3). A. 2 (5), 873–880. Third DWFI paralog in Solanaceae, sterol Δ24-isomerase, branches withanolide biosynthesis from the general phytosterol pathway. benthamiana and S. P. In all strains, the accelerated growth period began 10 h after culturing, and after 18 to 26 h, the logarithmic growth phases were observed (Figure 1B). tumefaciens GV2260 (pBBR1MCS-5, pEKH2), V1-E: A. B., et al. In this study, to further increase the transient and/or stable transformation frequencies, Super-Agrobacterium was updated to ver. C., Chakravarthy, S., Martin, G. with acdS and GabT activity at the same time, enhanced the stable transformation frequency approximately 3.6 times, compared with that of the original GV2260 strain. We succeeded in producing an A. tumefaciens GV2260 (pEAQ-GFP-HT) via electroporation. Figure 1 Effect of ACC deaminase and GABA transaminase activity on the transfer of T-DNA. L., Johnson, S., Gelvin, S. R., Ryu, C. V4-G showed the highest frequency of class 4, the frequencies were 3.9, 1.4, and 1.5 times higher than the C-G, V1-G, and V3-G, respectively. Agrobacterium tumefaciens recognizes its host environment using ChvE to bind diverse plant sugars as virulence signals. Transgenic Res. Applications, such as aminoethoxyvinylglycine (AVG), an ethylene biosynthesis inhibitor, and AgNO3 or silver thiosulfate (STS), ethylene perception inhibitors, were effective at improving the T-DNA transfer frequencies in tomato, melon, and bottle gourd (Davis et al., 1992; Ezura et al., 2000; Han et al., 2005; Nonaka and Ezura, 2014). S., Chin, D. (1961). Binary vectors and super-binary vectors. The cells were lysed on ice by sonication and centrifuged at 5,000 × g at 4°C for 15 min. doi: 10.1073/pnas.83.2.379PubMed Abstract | CrossRef Full Text | Google Scholar Sun, H. Infection of A. A plant cell factor induces Agrobacterium tumefaciens vir gene expression. (2008a). (2001). (D) Transient transformation via agroinfiltration methods on N. tumefaciens must be enlarged, and its transformation efficiency increased. For transformations, the pelleted bacterial cells were resuspended in liquid Murashige and Skoog (1962) (MS) containing 30 g/l glucose, and 500 µM acetosyringone at pH 5.2. The cell density was then adjusted to 0.4–0.5 at O.D.600. Table 1 List of A. 43 (4), 495–502. J., Bozzo, G. D., Mok, I. tumefaciens GV2260 (pBBRacDS, pEAQ-GFP-HT); V3-Q: A. Open and solid squares represent V3 and V4, respectively. 106 (34), 14587–14592. 191 (18), 5802–5813. The entire process for Agrobacterium-mediated stable transformation is divided into four steps: i) T-DNA transfer and integration into the plant genome, ii) calli induction, iii) the regeneration of the shoots, and iv) rooting. All Super-Agrobacterium strains increased the callus inductions compared with the C-G (Figures 3A–D). Open and solid circles represent A. The degree of staining was categorized into 4 classes (Figure 2B). A single colony was picked and cultured in 2 ml of LB medium at 28°C and 200 rpm for 2 days until the pre-culture reached the stationary phase. In the rooting step, there were no significant differences detected between them. doi: 10.5897/BJ09.057CrossRef Full Text | Google Scholar Khuong, T. Categorized into 4 classes: (Class 1) less than 5%, (Class 2) 5–10%, (Class 3) 10–20%, and (Class 4) more than 20%. benthamiana. AcDS activity did not improve the T-DNA transfer, but GabT activity was effective at increasing the T-DNA transfer. Then, the AcDS and GabT activity were measured, as described in previous studies (Nonaka et al., 2008a; Nonaka et al., 2017). On the other hand, the frequency is still not enough depending on the plant species and cultivars (Figures 3E, F). Differential accumulation of γ-aminobutyric acid in elicited cells of two rice cultivars showing contrasting sensitivity to the blast pathogen. 1 or ver. 6 (3), e23692. 3 were compared in E. In Super-Agrobacterium ver. 1, the expression of acdS gene was under the control of the lac promoter, which shows constitutive expression in A. Shoot regeneration ratios (shooting number / calli number) were increased with the inoculation of the V3-G and V4-G. Proc. GABA is a biologically active agent in animals, plants, and bacteria. This solution was applied to an Attune focusing analyzer (ABI, CA, USA), and 2n plants were selected. doi: 10.1111/plb.12165CrossRef Full Text | Google Scholar Guo, M., Ye, J., Gao, D., Xu, N., Yang, J. Therefore, the utilization AcdS activity seemed to be reasonable. Tissues were each subcultured for 10–14 days. Agro Infiltration Method. 72, 248–254. (1990). tumefaciens with AcDS activity (Nonaka et al., 2008a; Malambane et al., 2018). Agrobacterium tumefaciens is an α-proteobacteria that causes crown gall disease in many agriculturally and economically important species, such as those from the families Rosaceae (rose, apple, cherry, and pear), Vitaceae (grape), and the genus Juglans (walnut) (Kado, 2014). Therefore, it was difficult to detect the differences of GABA content. GABA is taken up into A. 6 (2), 271–282. Utilization of GabT activities increased the transient and stable transformation frequencies in tomato and grass plants (Nonaka et al., 2017) (Super-Agrobacterium ver. 3). Therefore, it would be difficult to compare between our and previous results. C., Memelink, J. . Ti plasmid vector for the introduction of DNA into plant cells without alteration of their normal regeneration capacity, ravenae and tomato with the tissue culture and co-cultivation methods. Therefore, the importance of T-DNA is integrated into the plant genome via complicated plant cell systems (Guo et al., 2019), and results in crown gall disease. doi: 10.1111/j.1467-7652.2009.00434.xPubMed Abstract | CrossRef Full Text | Google Scholar Sheehy, R. (2019). Therefore, replacing promoters would increase the transient transformations in S. ravenae were kindly provided by Prof. S., Chang, Y., Hsu, L., Ronzone, E., et al. 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Open. doi: 10.1128/AEM.02253-07PubMed Abstract | CrossRef Full Text | Google Scholar Nonaka, S., Yuhashi, K., Takada, K., Sugawara, M., Minamisawa, K., Ezura, H. Thus, V4 exhibited approximately 3.6, 1.6, and 1.6 times the stable transformation frequency of C-G, V1-G, and V3-G, respectively. There has been further effort to increase the T-DNA transfer frequency of A. Moreover, Super-Agrobacterium ver. 1 showed stronger inhibition of ethylene evolution and higher T-DNA transfer frequencies than chemical treatments in melon and wild water melon (Nonaka et al., 2008a; Malambane et al., 2018). Nature 318 (6047), 624–629. tumefaciens GV2260 carrying pEAQ-GFP-HT (Sainsbury et al., 2009) was grown in LB media, resuspended in 10 mM MgCl2, 10 mM MES, pH 5.6, 150 µM acetosyringone, and incubated for 3 h at room temperature. doi: 10.1111/j.1365-3113X.2010.04327.xPubMed Abstract | CrossRef Full Text | Google Scholar Planamente, S., Morera, S., Faure, D. 10, 1292. (2013). 3 and ver. 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Table 2 Effect of the Super-Agrobacterium ver. 1, ver. 3, and ver. 4 on plant regeneration and transformation of the “Micro-Tom” cotyledons. A. tumefaciens with AcDS and GabT was expected to cause reduced ethylene and GABA content in plants. Plant Biotechnol. coli (Accession No. CP040667); pBBR1 Rep, replication protein for the broad-host-range plasmid pBBR1 from Bordetella bronchiseptica; pBBR1 oriV, replication origin of the broad-host-range plasmid pBBR1 from B. (2005). J., Schilperoord, R. Open. doi: 10.1128/AEM.02253-07PubMed Abstract | CrossRef Full Text | Google Scholar Nonaka, S., Yuhashi, K., Takada, K., Sugawara, M., Minamisawa, K., Ezura, H. Thus, V4 exhibited approximately 3.6, 1.6, and 1.6 times the stable transformation frequency of C-G, V1-G, and V3-G, respectively. There has been further effort to increase the T-DNA transfer frequency of A. 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Open. doi: 10.1128/AEM.02253-07PubMed Abstract | CrossRef Full Text | Google Scholar Nonaka, S., Yuhashi, K., Takada, K., Sugawara, M., Minamisawa, K., Ezura, H. Thus, V4 exhibited approximately 3.6, 1.6, and 1.6 times the stable transformation frequency of C-G, V1-G, and V3-G, respectively. There has been further effort to increase the T-DNA transfer frequency of A. Moreover, Super-Agrobacterium ver. 1 showed stronger inhibition of ethylene evolution and higher T-DNA transfer frequencies than chemical treatments in melon and wild water melon (Nonaka et al., 2008a; Malambane et al., 2018). Nature 318 (6047), 624–629. tumefaciens GV2260 carrying pEAQ-GFP-HT (Sainsbury et al., 2009) was grown in LB media, resuspended in 10 mM MgCl2, 10 mM MES, pH 5.6, 150 µM acetosyringone, and incubated for 3 h at room temperature. doi: 10.1111/j.1365-3113X.2010.04327.xPubMed Abstract | CrossRef Full Text | Google Scholar Planamente, S., Morera, S., Faure, D. 10, 1292. (2013). 3 and ver. 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pathogen attack (Pate et al., 2010; Renault et al., 2011; Shelp et al., 2012; Forlani et al., 2014), but the action mechanisms of GABA in plants are still to be clarified, and the chemical compounds related with GABA perception or signal transduction in plants have not been identified. *torum*, respectively. It was found that some chemical compounds control GABA effect in animals, doi: 10.1093/oxfordjournals.pcp.a077982CrossRef Full Text | Google Scholar Park, D. R. tumefaciens has the ability to transfer T-DNA from bacterial cells to plant cells (T-DNA transfer), lycopersicumCalli of E. High efficiency transformation of maize (Zea mays L.) mediated by Agrobacterium tumefaciens, doi: 10.1093/pcp/cpi251PubMed Abstract | CrossRef Full Text | Google Scholar Takahashi, H., Sumi, M., Koshino, F. Cell Microbiol. (B) Growth curve of A. One-month after inoculation, the calli inductions were observed. Gamma aminobutyric acid: circulatory and respiratory effects in different species; re-investigation of the anti-strychnine action in mice. doi: 10.1128/JB.00451-09PubMed Abstract | CrossRef Full Text | Google Scholar Hiei, Y., Ohta, S., Komari, T., Kumashiro, T. tumefaciens GV2260 (pBBRacdSgabt). doi: 10.1073/pnas.0600366103PubMed Abstract | CrossRef Full Text | Google Scholar Malambane, G., Nonaka, S., Shiba, H., Ezura, H., Tsujimoto, H., Akashi, K. 13 (11), e0200972. After 3 days of co-cultivation, the number of blue spots were counted to evaluate the T-DNA transfer in E. C-G: A. Previous studies have demonstrated that GABA was independent of vir gene expression (Chevrot et al., 2006; Haudecoeur et al., 2009). Nucleic Acids Res. *torum*, the success of the V4-Q strain with the Agroinfiltration treatment was greater than that of the C-Q strain, but the same as that of the V1-Q and V3-Q strains (Figure 2E).Figure 2 Transient transformations in tomato via tissue culture and co-cultivation method. tumefaciens GV2260 (pBBRacdSgabt, pEAQ-GFP-HT).V4 was effective at the T-DNA transfer in E. C., Glick, B. T., Farris, M. doi: 10.1104/pp.107.111302PubMed Abstract | CrossRef Full Text | Google Scholar Bradford, M. These results mean that ethylene and GABA influence the T-DNA transfer frequencies at almost the same level in these plant species. Nakamura (Chiba University, Japan) and Prof. A. Roop, R. 307 (2), 185–190. J Vis Exp. The AcdS activity showed higher callus induction frequencies than the Gabt activity, whereas the Gabt activity induced higher shoot regeneration ratios than the AcdS. Salicylic acid and systemic acquired resistance play a role in attenuating crown gall disease caused by Agrobacterium tumefaciens. (2006) doi: 10.1113/jphysiol.1950.sp006178PubMed Abstract | CrossRef Full Text | Google Scholar Ezura, H., Yuhashi, K. ravennae is known for its high bio-mass production and is relevant for practical agriculture. C. GV2260 (pBBRMC51-5). V1. A. R. Hooykaas, P. Camy 355 P; Cauliflower mosaic virus 35S promoter, NosT; Nopalyn synthesis gene terminator, GFP; green fluorescence gene, RB; Right border sequence, LB; Left border sequence.Supplemental Figure 2 | Map of the pIG121-Hm vector and the southern blot analysis of the T0 generation. tumefaciens GV2260 (pBBRMC51-5, pEAQ-GFP-HT); V1-Q: A. tumefaciens GV2260 (pBBRgabt); V4. A. Red numbers indicate the transgenic lines with single copy. Metabolism of 1-aminocyclopropane-1-carboxylic acid. benthamiana. A. Ankenbauer, R. L. tumefaciens C and V1, respectively. virD genes are induced by acetosyringone at pH 5.2, which is the co-cultivation condition. L., Brikis, C. Moreover, because our system was the plasmid with acdS and gabt gene, it was used in combination with other strains, such as the EHA105, EHA101, LBA4404, MP90, and AGL1. 173 (17), 5260–5265. tumefaciens would be effective at increasing the T-DNA transfer. Plant Cell Physiol. S. (A) Map of pEKH2-nosPNPTII-ubiPGUS-35SPHPT. doi: 10.1074/jbc.M110.140715PubMed Abstract | CrossRef Full Text | Google Scholar Renault, H., El Amrani, A., Palanivelu, R., Updegraff, E. tumefaciens GV2260 (pBBRacdSgabt, pEKH2). (1962). The GUS stained areas were converted into numerical values by Image J (National Institutes of Health; and the percentage of GUS stained area for each explant was calculated). Evaluation of four Agrobacterium tumefaciens strains for the genetic transformation of tomato (Solanum lycopersicum L.) cultivar Micro-Tom. Organ. tumefaciens C58 strain, which was the original strain for Agrobacterium-mediated transformation, does not have the acdS gene or its activities (Wood et al., 2001; Nonaka et al., 2008a). (2011). 13 (13), 4777–4788. Construction and expression in tobacco of a β-glucuronidase (GUS) reporter gene containing an intron within the coding sequence. tumefaciens strains using the cell lysate at the 'Early stage' (O.D.600 0.7). GFP signals were used as indicators of transformation. The abilities of the Super-Agrobacterium ver. 103, 15–22. E., Messens, E., Van Montagu, M., Zambryski, P. The browning callus appearance was suppressed by A. A. A conserved mechanism of GABA binding and antagonism is revealed by structure-function analysis of the periplasmic binding protein Atu2422 in Agrobacterium tumefaciens. The C-G, V1-G, V3-G, and V4-G showed calli induction frequencies of 51.5 ± 0.6, 85.2 ± 8.8, 73.8 ± 2.03, and 91.8 ± 3.7%, respectively (Figure 3E, Table 2). N. S., Kim, C.-Q. A. (1983). Plant Sci. Biotechnol Adv. Plant Physiol. 3, and ver. Commun Integr Biol. NPTII probes were used in. doi: 10.1111/1574-6968.2010.01977.xPubMed Abstract | CrossRef Full Text | Google Scholar Haudecoeur, E., Planamente, S., Cirou, A., Tannieres, M., Shelp, B. 4 showed higher level of T-DNA transfer than GV2260 and Super-Agrobacterium ver. 64 (2), 318–330. These results imply that all of the lines we obtained were independent and did not contain a cloned plant. 1, but the level of T-DNA transfer was same in ver. ravennae calli. E., Honma, M., Yamada, M., Sasaki, T., Martineau, B., Hiatt, W. On the other hand, research has shown that the ethylene target points are involved with vir gene expression, and the ethylene perceiving plant would reduce vir gene inducers or release antagonists of the vir gene inducers (Nonaka et al., 2008b). One-way analysis of variance (ANOVA) and Tukey Kramer's multiple range test, with P < 0.01 or P < 0.05, were carried out to determine the significant differences. GABA controls the level of quorum-sensing signal in Agrobacterium tumefaciens. FEMS Microbiol. 8, 195. Therefore, the additional effort should have been required to adapt Agrobacterium-mediated transformation for a wide variety of plants. doi: 10.1385/1-59745-130-4:15PubMed Abstract | CrossRef Full Text | Google Scholar Kovach, M. 20 (4), 773–786. B. 193–194, 130–135. doi: 10.1093/pcp/pcn113PubMed Abstract | CrossRef Full Text | Google Scholar Anand, A., Uppalapati, S. doi: 10.1002/mbio.3.123CrossRef Full Text | Google Scholar Stachel, S. The AcdS activity was measured spectrophotometrically at 340 nm. doi: 10.1016/0003-2697(76)90527-3PubMed Abstract | CrossRef Full Text | Google Scholar Cangelosi, G. In our study, the transformation frequency was calculated using regenerated rooting shoot with diploid and a single copy per inoculated segment. tumefaciens with AcdS and Gabt increased the T-DNA transfer and stable transformation frequency. After regenerated diploid shoots (2n) were selected, the exogenous T-DNA was detected by PCR (data not shown) and Southern hybridization analysis (Supplemental Figure 2). Classification of GUS-stained cotyledon explants. doi: 10.1073/pnas.87.17.6708PubMed Abstract | CrossRef Full Text | Google Scholar Chetty, V. The bars indicate the standard deviation (n = 3). Biotechnol. The calli were induced directly from the seeds on MS medium, containing 1 g/l casamino acids, 2 mg/l 2,4-dichlorophenoxyacetic acid (2,4-D), 0.2 mg/l 6-benzylaminopurine (BAP), 30 g/l 4-O-α-D-glucopyranosyl-D-glycopyranose (maltose H) (Wako, Tokyo, Japan), and 0.3% Gelrite (Wako, Tokyo, Japan), were subcultured for 2 weeks before inoculation. Other steps showed different responses to AcdS and/or Gabt activity (Figure 3 and Table 2). tumefaciens GV2260 (pBBRgabt, pEAQ-GFP-HT). V4-Q: A. To evaluate the ability of the T-DNA transfer in C-G, V1-G, V3-G, and V4-G, the frequency of class 4 was compared. Different characters indicate values that were statistically different in the one-way ANOVA and Tukey-Kramer method, multiple comparison method (P < 0.01).Cells were collected and washed twice with 100 mM Tris-HCl (pH 8.5) and resuspended in 1.5 ml of lysate buffer. P., Okura, V. The sterilized tomato seeds were sown on MS medium, containing 15 g/l sucrose (Wako, Tokyo, Japan) and 0.3% Gelrite (Wako, Tokyo, Japan). An alternative strategy was the utilization of ACC deaminase (AcdS) activity, which cleaves the plasmid pBBRacdSgabt and did western blot analysis. 1-Aminocyclopropane-1-carboxylate deaminase enhances Agrobacterium tumefaciens-mediated gene transfer into plant cells. On the other hand, significant differences in GABA content during the co-cultivation were not observed between A. 4) with the ability to remove ethylene, suppressed the browning phenomena. After ultraviolet (UV) cross-linking, the membranes were hybridized in a solution containing 7% sodium dodecyl sulfate (SDS), 50% deionized formamide, 50 mM sodium phosphate (pH 7.0), 2% blocking solution, 0.1% N-lauroylsarcosine, 0.75 M NaCl, and 75 mM sodium citrate at 42°C overnight, doi: 10.1007/s11248-010-9458-6PubMed Abstract | CrossRef Full Text | Google Scholar Yuan, Z. The effect of γ-aminobutyric acid on blood pressure. Nature 303 (5913), 179–180. The transformation frequency, which might depend on the bacterial strain, binary vector and the selection method. 42 (10), 1825–1831. In brief, after 3 days of co-cultivation, tomato cotyledon segments were placed on a callus-induction medium [MS medium containing 0.3% Gelrite (Wako, Tokyo, Japan), 1.5 mg/l zeatin, 100 mg/l kanamycin, and 375 mg/l Augmentin (GlaxoSmithKline, London, UK)] for 4 weeks. Plant Biol. C., Lomonosoff, G. Next, we evaluated the abilities of Super-Agrobacterium V4 using S. A binary plant vector strategy based on separation of vir- and T-region of the Agrobacterium tumefaciens T-plasmid. J. Bacteriol. 4 was one third of that found in Super-Agrobacterium ver. From these results, the effect of the Super-Agrobacterium was found to be different, dependent on the plant species, thus the selection of the most suitable strain is important for the successful application of the technology.Even under conditions where the vir gene is sufficiently expressed, our Super-Agrobacterium strains could further improve T-DNA transfer. ACC deaminase was probed with an anti-ACC deaminase antibody. doi: 10.1038/srep42649PubMed Abstract | CrossRef Full Text | Google Scholar Nonaka, S., Sugawara, M., Minamiasawa, K., Yuhashi, K., Ezura, H. Efficient transformation of rice (Oryza sativa L.) mediated by Agrobacterium and sequence analysis of the boundaries of the T-DNA. The number of GUS-stained spots per 1 g of E. Natl. 5, 340. tumefaciens GV2260 (pEKH2-nosPNPTII-ubiPGUS-35SPHPT; pEKH2). A. coli, GeneBank accession #J01780). KanR; Kanamycin resistance gene. The ligated fragment was inserted into pBBRacdS (Nonaka et al., 2008a) and digested with EcoRI and XbaI (New England Biolabs, Hirschin, UK). doi: 10.3797/11292CrossRef Full Text | Google Scholar Wang, H., Lin, J., Chang, Y., Jiang, C. 83 (2), 379–383. tumefaciens was re-suspended in 100 μl of BugBuster Master mix (Novagen, MA, USA) for lysate preparation. New Phytol. Chem. 103 (12), 4658–4662. coli; acdS, ACC deaminase gene from Pseudomonas sp (Sheehy et al., 1991, Accession No. M73488); gabt, GABA transaminase gene from E. The plant signal salicylic acid shuts down expression of the vir regulon and activates quorumone-quenching genes in Agrobacterium. Cooperative Research Grant from Plant Transgenic Design Initiative (PTrad), Gene Research Center in Tsukuba Innovation Plant Research Center (T-PIRC), University of Tsukuba, Japan supported this research.Conflict of InterestThe authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.AcknowledgmentsWe appreciate the help of Prof. 38 (1), 75–84. ravennae calli were assayed histochemically with X-Gluc buffer containing 100 mM phosphate buffer, 10 mM EDTA, 2.5 mM potassium ferricyanide, 2.5 mM potassium ferrocyanide, 0.1% Triton X-100, and 0.5 mg/l X-glucuronide. Therefore, to expand plant spices and cultivars adapting Agrobacterium-mediated transformation, multiply suppress of these negative factors would be also effective.Data Availability StatementAll datasets GENERATED for this study are included in the manuscript/Supplementary Files.Author ContributionsSN designed the experiments, analyzed the data, and wrote the manuscript. doi: 10.1002/1460-2075.1983.tb01715.xPubMed Abstract | CrossRef Full Text | Google Scholar strain ACP gene encoding 1-aminocyclopropane-1-carboxylate deaminase. W., Setubal, J. Comparative transcriptome analysis of Agrobacterium tumefaciens in response to plant signal salicylic acid, indole-3-acetic acid and gamma-amino butyric acid reveals signalling cross-talk and Agrobacterium-plant co-evolution. (2017). J., Ceballos, N., Garcia, D., Narváez-Vásquez, J., Lopez, W., Orozco-Cárdenas, M. Each experiment was repeated three times.Ploidy AnalysisThe ploidy of the rooting shoots was checked with flow cytometry. P., Liu, P., Saenkanh, P., Banta, L. E., Elzer, P. I., Yasuta, T., Minamiasawa, K. However, some plants have significantly lower transient gene transfer rates, creating a limitation in plant species research that should be resolved by increasing the transient transformation (T-DNA transfer) frequency in a wide variety of plant hosts. Ethylene production in plants during transformation suppresses vir gene expression in Agrobacterium tumefaciens. To utilize this unique ability of A. Therefore, in N. lycopersicum "Micro-Tom." Additionally, we evaluated the ability of T-DNA transformation with V4 in the Agroinfiltration method. PLoS One, G., Nester, E. tumefaciens, resulting in increased T-DNA transformation frequencies (Super-Agrobacterium ver. M., Haring, M. Calli that formed segments were cultured on shoot-induction medium [MS medium containing 0.3% Gelrite (Wako, Tokyo, Japan), 1.0 mg/l zeatin, 100 mg/l kanamycin, and 375 mg/l Augmentin (GlaxoSmithKline, London, UK)] for 4 weeks. To measure the AcdS and Gabt activity, cells were collected by centrifugation, and the lysate was prepared. pEAQ: versatile expression vectors for easy and quick transient expression of heterologous proteins in plants. ravennae and S. *torum* for both transient and stable transformations.Materials and MethodsBacterial Strains, Vectors, and Culture ConditionsAll bacterial strains and vectors, which were used in this study, were listed up in Table 1. Masahiro Mii from Chiba University, Japan, 29, 87–93. 49 (9), 1378–1389. The shoots were then placed on rooting medium, which consisted of half-strength MS medium, 0.3% Gelrite (Wako, Tokyo, Japan), 100 mg/L kanamycin, and 375 mg/l Augmentin, for 2 weeks. If the target point of the ethylene was the reduction of the vir gene inducer, the effect of the Super-Agrobacterium ver. Different characters indicate values that were significantly different according to the one-way analysis of variance, multiple comparison method (P < 0.01). Cult. C. 3 was also effective in the agro-infiltration method (Hoshikawa et al., 2019). Knoch et al., 2019). Stable transformation techniques are important as they are used for breeding GM crops. Previous research compared the vir gene promoters (virB, virC, and virD) with the lac promoter activities; the vir gene promoters were found to show higher promoter activities than the lac promoter (Someya et al., 2013). Jpn. doi: 10.1016/j.plantsci.2012.06.001PubMed Abstract | CrossRef Full Text | Google Scholar Someya, T., Nonaka, S., Nakamura, K., Ezura, H. (2007). After the third wash, the seeds were kept in water for 2 days. A rapid and sensitive method for the quantitation of microgram quantities of protein utilizing the principle of protein-dye binding. (C) ACC deaminase activity in A. No differences were observed between the strains in E. W. R., Lineberger, R. Anal. doi: 10.3389/fmicb.2014.00340PubMed Abstract | CrossRef Full Text | Google Scholar Khoudi, H., Nouri-Khemakhem, A., Gouia, S., Masmoudi, K. Expressing multiple genes using the same promoter may reduce the expression levels of each gene. Optimisation of tomato Micro-tom regeneration and selection on glufosinate/Basta and dependency of gene silencing on transgene copy number. Hypothesis/review: contribution of putrescine to 4-aminobutyrate (GABA) production in response to abiotic stress. (E) Appearance of calli, (F) Frequency of regeneration, (G) Rooting ratio, (H) Frequency of calli regeneration. H. GUS stained tomato cotyledon explants were observed and images were taken using a stereoscopic microscope system (Leica: MX FLIII, DFC300 FX, Application Suite, Leica, Wetzlar, Germany). Although the function of GABA in plants needs to be clarified, there have been several studies regarding the functions of GABA as a signaling compound in plant growth and development. C., Haudecoeur, E., Faure, D., Kerr, K. (C) Appearance ratio Class 4 in tomato cotyledons. M. 14 (6), 745–750. NosP; Nopalyn synthesis gene promoter, UbpI; ubiquitin gene promoter from rice, NosT; Nopalyn synthesis gene terminator, nptII; neomycin phosphotransferase gene, uidA; beta-glucuronidase gene, hptII; hygromycin phosphotransferase gene, ORV; replication origin V (IncPα, plasmid RK2 from E. Comparative effects of ethylene inhibitors on Agrobacterium-mediated transformation of drought-tolerant wild watermelon. GUS stained tomato cotyledons were categorized depending on the stained area. To ascertain this, the T-DNA transfer abilities of Super-Agrobacterium ver. According to the results, GUS stained tomato explants were categorized into 4 classes: (Class 1) less than 5%, (Class 2) 5–10%, (Class 3) 10–20%, and (Class 4) more than 20%. tumefaciens GV2260 (pBBRacdS. Isolation, sequence, and expression in Escherichia coli of the Pseudomonas sp. Boscì Biotechnol Biochem. A., Hoge, J. tumefaciens GV2260 (pBBRacdSgabt, pIG121-Hm).References Akhiro, T., Koike, S., Tani, R., Tomimaga, T., Watanabe, S., Iijima, Y., et al. doi: 10.1007/s11240-010-9748-3CrossRef Full Text | Google Scholar Ohta, S., Mita, S., Hattori, T., Nakamura, K. 4, the enzymatic activity was one third of the Super-Agrobacterium ver. tumefaciens were effective at increasing the T-DNA transfer frequency (Nonaka et al., 2008a; Nonaka et al., 2017; Ntui et al., 2010; Hao et al., 2010), but it was not clear which was more effective in Agrobacterium-plant interactions. doi: 10.3389/fpls.2014.00681PubMed Abstract | CrossRef Full Text | Google Scholar Nonaka, S., Someya, T., Zhou, S., Takayama, M., Nakamura, K., Ezura, H. GABA accumulation causes cell elongation defects and a decrease in expression of genes encoding secreted and cell wall-related proteins in Arabidopsis thaliana. tumefaciens did not affect its bacterial growth. Mitsui (Tohoku University, Japan) for the gift of the pEKH2 plasmid and pBBR1MC5-5, respectively. 47 (3), 426–431. An efficient Agrobacterium tumefaciens-mediated genetic transformation of 'Egusi' melon (Colocynthis citrullus L.). To estimate whether these two genes affect bacterial growth or not, growth curves were compared for the four strains [(C), (V1), (V3), and (V4)]. Z. E., Miller, A. W., Zambryski, P. lycopersicum"). Effect of gamma-aminobutyric acid (GABA) on normotensive or hypertensive rats and men. J., Shen, J., Johnson, S., Lowe, B., Radke, S., et al. (I) Frequency of appearance for transgenic tomato plants which have single copy of T-DNA. (2014). doi: 10.1073/pnas.1215033110PubMed Abstract | CrossRef Full Text | Google Scholar Ishida, Y., Saito, H., Ohta, S., Hiei, Y., Komari, T., Kumashiro, T. J., Moréira, S., et al. RepB C-terminus mutation of a pR1-repABC binary vector affects plasmid copy number in Agrobacterium and transgene copy number in plants. Agrobacterium-mediated horizontal gene transfer: Mechanisms, biotechnological application, potential risk and forestalling strategy. Molecular basis of CheV function in sugar binding, sugar utilization, and virulence in Agrobacterium tumefaciens. doi: 10.1007/600299-013-1456-0PubMed Abstract | CrossRef Full Text | Google Scholar Knoch, E., Sugawara, S., Mori, T., Poulsen, C., Fukushima, A., Harholt, J., et al. When the O.D.600 of the culture reached 0.7 to 0.9, the cells were then centrifuged, collected, and checked for enzymatic activity. tumefaciens. EMBO J. Biol. I. doi: 10.1093/nar/13.13.4777PubMed Abstract | CrossRef Full Text | Google Scholar Elliott, K. tumefaciens were 0.3 at O.D.600 for N. 15, 473–497. Especially in tomato, this newly bred bacterium (Super-Agrobacterium ver. doi: 10.3389/fpls.2017.00195PubMed Abstract | CrossRef Full Text | Google Scholar Wood, D. The inoculated explants were cultured on co-cultivation medium (pH 5.2) containing MS salts, 30 g/l glucose, 500 μM acetosyringone, and 0.3% Gelrite (Wako, Tokyo, Japan) at 25°C, for 3 days in the dark. tumefaciens GV2260 (pBBRMC51-5, pIG121-Hm). V1-Q: A. doi: 10.1073/pnas.0600313103PubMed Abstract | CrossRef Full Text | Google Scholar Davis, M. PubMed Abstract | CrossRef Full Text | Google Scholar van der Fits, L., Deakin, E. 146, 70–84. Rep. tumefaciens GV2260 (pIG121-Hm). Q. A. (2012). The strength of the GFP signal was used as an indicator of the frequency of the T-DNA transfer. Biochemical mechanism on GABA accumulation during fruit development in tomato. K., Park, S. Some bacteria are known to harbor GABA transaminase (gabt), a GABA degradation enzyme. tumefaciens strains were then cultured on solid LB medium at 28°C for 2 days. 2 than in ver.1 (Someya et al., 2013).Gamma-aminobutyric acid (GABA), an amino acid, was determined to be another negative factor in Agrobacterium-plant interactions (Chevrot et al., 2006; Haudecoeur et al., 2009; Nonaka et al., 2017). The AcdS activity was measured according to a modified protocol based on that of Honma and Shimomura (1978).

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