Bodo saltans SSU-GFP-Neo cassette

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Abstract

This plasmid is designed to integrate within the small subunit ribosomal RNA gene region of *B. saltans*. It carries 1 kb of the *B. saltans* small subunit ribosomal RNA gene. To construct this cassette, we first sequenced the complete ribosomal operon of *B. saltans*, which has a total length of ~9 kb. The structure of the ribosomal operon of *B. saltans* is similar in the closely related kinetoplastida *Leishmania*, where the ribosomal RNA genes are arranged in head- to-tail repeats (Figure 4b and Yan et al., *Molecular and biochemical parasitology* 103, no. 2 (1999): 197-210). For the cassette construction, we used only a 1 kb region of the 18S ribosomal RNA gene, which is represented in the schematic as Homologous Recombination Region 1 (HR1) and Homologous Recombination Region 2 (HR2) each, of which is 500 bp . The GFP and Neo genes are located in the middle of the construct and their expression is controlled by the presence of the *B. saltans* tubulin intergenic region (Bs TUB IGR) at the 3’ and 5’ ends of each gene. As the transcription in *B. saltans* is polycistronic, the presence of the tubulin intergenic region is necessary for trans-splicing and polyadenylation.

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