

**Table S2 Strains and plasmids used in this study**

Strains or plasmids	Relevant characteristics	Sources
<b>Bacterial strains</b>		
<i>S. spinosa</i> ATCC49460	Wide-type strain	CGMCC
<b><i>Streptomyces</i></b>		
<i>S. avermitilis</i> ATCC31267	Wide-type strain	CGMCC
<i>S. coelicolor</i> M145	Wide-type strain	CGMCC
<i>S. coelicolor</i> M1154	$\Delta act, \Delta red, \Delta cpk, \Delta cda, rpoB[C1298T], rpsL[A262G]$	[1]
<i>S. albus</i> ATCC39012	Wide-type strain	CGMCC
<i>S. lividans</i> TK24	<i>RpsL</i> (S <sub>mr</sub> ) <i>Act</i> + <i>Red</i> <sup>+</sup>	John Innes Centre, Norwich, UK
<i>S. venezuelae</i> ATCC 15439	Wide-type strain	CGMCC
<i>S. avermitilis</i> ATCC 31267 BAC- <i>spn</i> -152- <i>rha</i>	<i>S. avermitilis</i> ATCC 31267 containing BAC- <i>spn</i> -152- <i>rha</i>	Present study
<i>S. coelicolor</i> M145 BAC- <i>spn</i> -152- <i>rha</i>	<i>S. coelicolor</i> M145 containing BAC- <i>spn</i> -152- <i>rha</i>	Present study
<i>S. lividans</i> TK24 BAC- <i>spn</i> -152- <i>rha</i>	<i>S. lividans</i> TK24 containing BAC- <i>spn</i> -152- <i>rha</i>	Present study
<i>S. venezuelae</i> ATCC 15439 BAC- <i>spn</i> -152- <i>rha</i>	<i>S. venezuelae</i> ATCC 15439 containing BAC- <i>spn</i> -152- <i>rha</i>	Present study
<i>S. albus</i> ATCC39012 BAC- <i>spn</i> -152- <i>rha</i>	<i>S. albus</i> ATCC39012 containing BAC- <i>spn</i> -152- <i>rha</i>	Present study
<i>S. coelicolor</i> M1154 BAC- <i>spn</i> -152- <i>rha</i>	<i>S. coelicolor</i> M1154 containing BAC- <i>spn</i> -152- <i>rha</i>	Present study
<b><i>E. coli</i></b>		
DH5 $\alpha$	<i>recA1, endA1, hsdR17, gyrA96, relA1,</i>	Stratagene
ET12567 (pUZ8002)	Nonmethylation strain ( <i>dam</i> <sup>-</sup> <i>dcm</i> <sup>-</sup> <i>hsdM</i> <sup>-</sup> ), TetR, CmR	[2]
BW25113	K-12 BD792 derivative: <i>lacIq, rrnBT14, <math>\Delta lacZ</math>WJ16, hsdR514, <math>\Delta araBADAH33, \Delta rhaBADLD78</math></i>	[3]
<b>Plasmids</b>		
pJL117	pIJ2925 derivative carrying the <i>ermE</i> *P from pIJ4090	[4]
pKD10	Derived from pKD20, Amp <sup>r</sup>	[5]
pCC1BAC BamHI	BAC cloning vector	<u>Epicentre</u>
pSET152	Integrative vector	[3]

BAC- <i>spn</i>	pCC1BAC with ~100 kb DNA insert containing the whole <i>spn</i> gene cluster	Present study
BAC- <i>spn</i> -152	BAC- <i>spn</i> derivative containing the 4.3 kb fragment of pSET152 containing <i>aac3</i> (IV), <i>oriT</i> , <i>attP-int</i>	Present study
BAC- <i>spn</i> -152- <i>rha</i>	BAC- <i>spn</i> -152 derivative integrated with <i>gtt</i> , <i>gdh</i> , <i>kre</i> , <i>epi</i>	Present study

#CGMCC: China General Microbiological Culture Collection Center.

## References

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- [2]MacNeil DJ, Gewain KM, Ruby CL, Dezeny G, Gibbons PH, MacNeil T: **Analysis of *Streptomyces avermitilis* genes required for avermectin biosynthesis utilizing a novel integration vector.** *Gene* 1992, 111(1):61-68.
- [3]Datsenko KA, Wanner BL. **One-step inactivation of chromosomal genes in *Escherichia coli* K-12 using PCR products.** *Proc Natl Acad Sci USA* 2000, 97(12): 6640-6645.
- [4]Zhao JL: **Regulation mechanism of avermectin biosynthesis.** *PhD thesis*. China Agricultural University, Microbiology and Immunology Department; 2006.
- [5]Bierman M, Logan R, O'Brien K, Seno ET, Rao RN, Schoner BE: **Plasmid cloning vectors for the conjugal transfer of DNA from *Escherichia coli* to *Streptomyces* spp.** *Gene* 1992, 116(1):43-49.