Improved yield of helper P-element by switching its vector from pBR322 to pBluescript

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A helper P-element plasmid, p π 25.7wc, (ref. 1) is commonly used for germline transformations in *Drosophila*. The yield of this plasmid (obtained from W. Bender at Harvard) is not good in our hands regardless of *E. coli* hosts. Since we have had a good luck with pBluescript vector (Stratagene) in the JM109 host, we decided to subclone the relevant portion containing the P-element into this vector.

p π 25.7wc was digested first completely with BamHI and EcoRI, then partially with Sall. The resulting 2.8kb BamHI-EcoRI and 1.2kb EcoRI-Sall fragments were isolated and ligated into BamHI-Sall digested pBluescript. Ca⁺⁺-treated JM109 was transformed with the ligated DNA. The resulting plasmid was named pBL π 25.7wc.

The gel picture below shows comparative yields of p π 25.7wc and pBL π 25.7wc both in JM109 prepared in an identical manner. Minipreparations (2) using a 10 ml culture routinely give 50 to 100 ug of pBL π 25.7wc, which is 10 to 20 fold greater than that of p π 25.7wc. Digestions with BamHI, EcoRI and Sall reveal all the relevant fragments of the P-element (see arrows). This new pBL π 25.7wc was used successfully as a helper P-element for transformations of *Drosophila* in our laboratory (unpublished observations).

a; HindIII digested lambda DNA marker
b and d; undigested and digested p π 25.7wc
c and e; same as above with pBL π 25.7wc

References

1. Karess, R.E. and Rubin, G.M. (1984) *Cell* **38**, 135-146.
2. Ish-Horowitz, D. and Burke, J.F. (1981). *Nucleic Acid Res.* **9**, 2989-2998.