

Aar1 digestion protocol

How to combine parts from a donor vector into an acceptor vector.

Start with standard 5ml culture miniprep, eluted with 50ul.
(should yield ~250 ng/ul)

FOR VECTORS and INSERTS:

Digest 20ul (~5 ug) of miniprep (use Aar1 specific buffer):

Water	30.6
Buffer	6
Oligo	0.9
DNA	20
Enzyme	2.5

3 hrs at 37 deg.

For VECTORS, PCR purify this reaction, and elute with 44ul.

PCR purification	eluate
Buffer 4	5
BSA	0.5
Xma1	1.5

Digest 2 hrs at 37deg. Add 1ul CIP, incubate one more hour.

Gel Purify vectors and inserts. For weak bands, elute with 30ul, for strong use 50ul.