Linearizing DNA for Transfection

Digestion:

20 ul DNA (1ug/ul)
20 ul ScaI Buffer
5 ul ScaI Enzyme
155 ul dd H20
200ul

Incubate @37C for 2 hours

Add 2ul ScaI enzyme. Incubate 2 more hours at 37C

Add 1ml (5 volumes) of buffer PB to each rxn.

Add sample to QIA-quick column (2X, 600ul each)

Spin 1 min @ max speed, 4C. Discard flowthrough

Wash column with 750ul buffer PE. Spin 1min @ max speed, 4C. Discard FT

Spin 1 additional minute to get rid of residual EtOH

Place column in a fresh eppendorf tube and add 40ul Elution Buffer to center of column.

Let sit for 1 min. Spin for 1 min.

Store tube at –20C. Concentration is at 0.5 ug/ul