Table SI. Terminus DNA end configuration of the NHEJ substrates employed in the study.

NHEJ substrate type	Restriction Enzyme(s)	Terminus sequence	
1) blunt ends	*Eco*RV	5'···GAT ATC···3' 3'···CTA TAG···5'	
2) cohesive 5'-PSS	*Eco*RI	5'···G AATTC···3' 3'···CTTAA G···5'	
3) non-matching 5'-PSS	*Not*I-*Eco*RI	5'···C AATTC···3' 3'···GCCGG G···5'	
4) blunt end + 5'-PSS	*Bam*HI-*Eco*RV	5'···G ATC···3' 3'···CCTAG TAG···5'	
5) cohesive 3'-PSS	*Pst*I	5'···CTGCA G···3' 3'···G ACGTC···5'	
6) non-matching 3'-PSS	*Sac*I-*Kpn*I	5'···GAGCT C···3' 3'···C CATGG···5'	
7) blunt end + 3'-PSS	*Sac*I-*Sma*I	5'···GAGCT GGG···3' 3'···C CCC···5'	
8) 5'-PSS + 3'-PSS	*Pst*I-*Xho*I	5'···CTGCA TCGAG···3' 3'···G C···5'	
9) non-matching 3'-PSS	*Pst*I-*Kpn*I	5'···CTGCA C···3' 3'···G CATGG···5'	

The single-cut digestion provided linear DNA molecules bearing blunt (1) or matching 4 nt protruding single strand ends (PSS) (cohesive 5'(2)- and 3'- PSS(5)). Cleavage at two different restriction site generated substrates with either non cohesive ends (non-matching 5' (3)- and 3'-PSS (6 and 9)) or ends with different polarity (blunt ends + 5'(4)- or 3'-PSS (7) and 5'-PSS + 3'-PSS (8)).