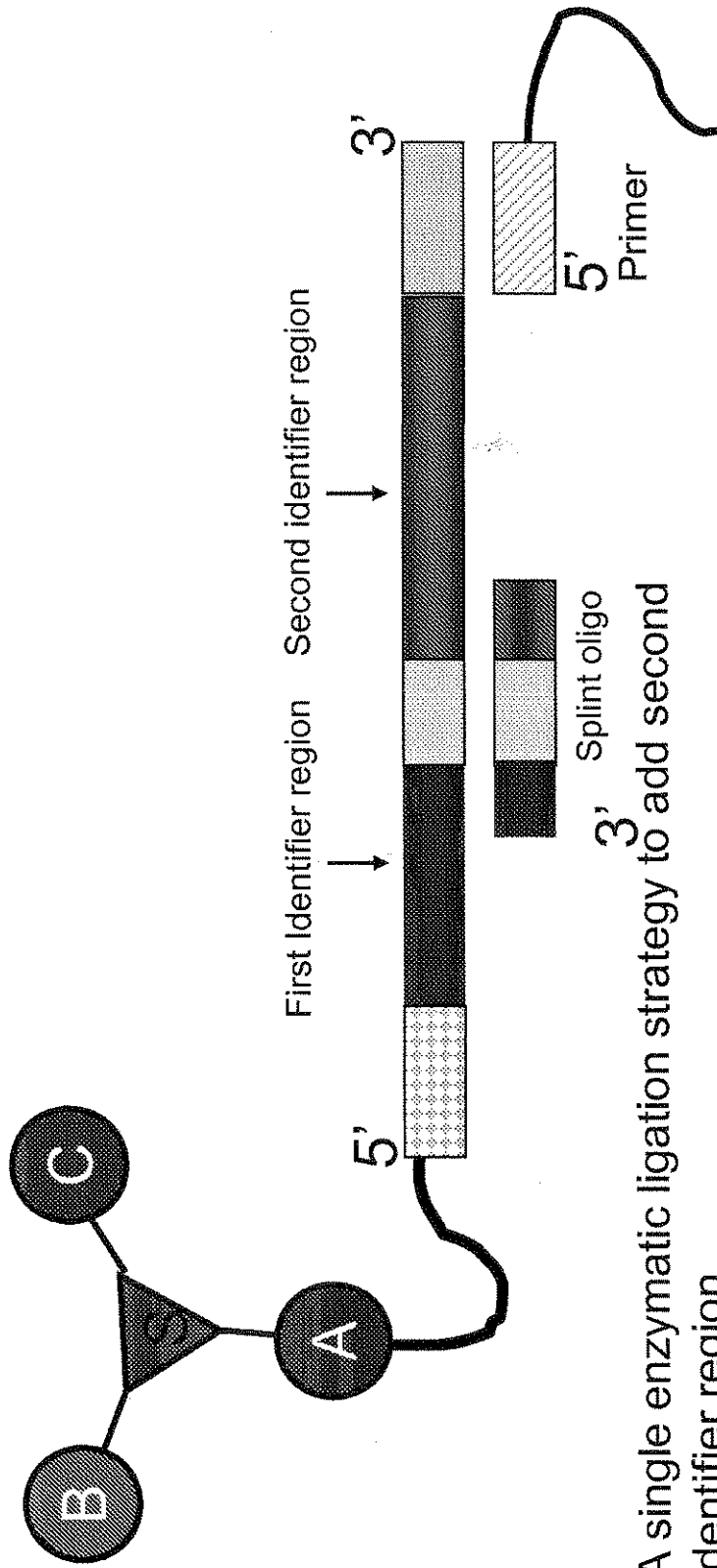
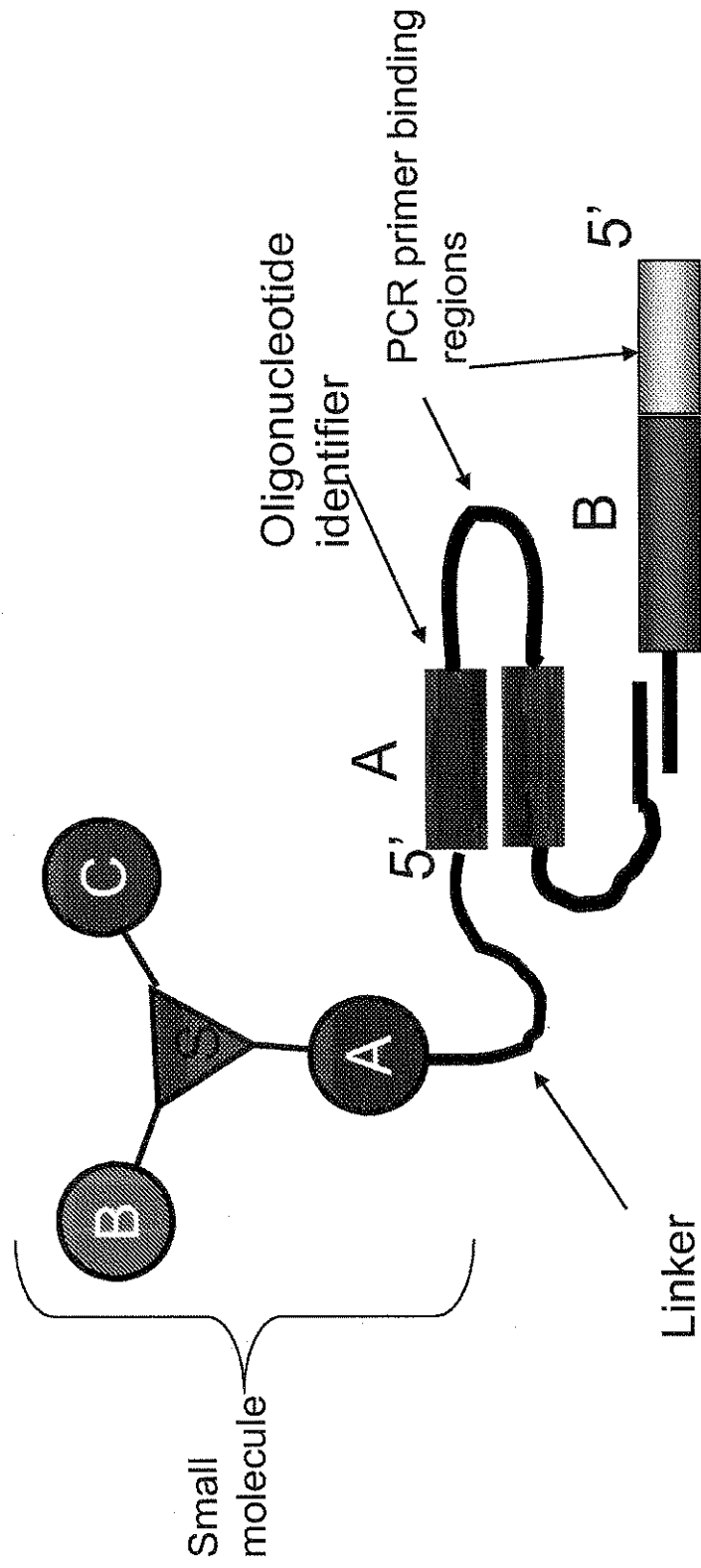


Figure 1



- A single enzymatic ligation strategy to add second identifier region
- Potential that exposed bases will be damaged during chemical synthesis; seek to minimize through double-stranded approaches

Figure 2

Mode 1

Polymerization to add second identifier region

Figure 3

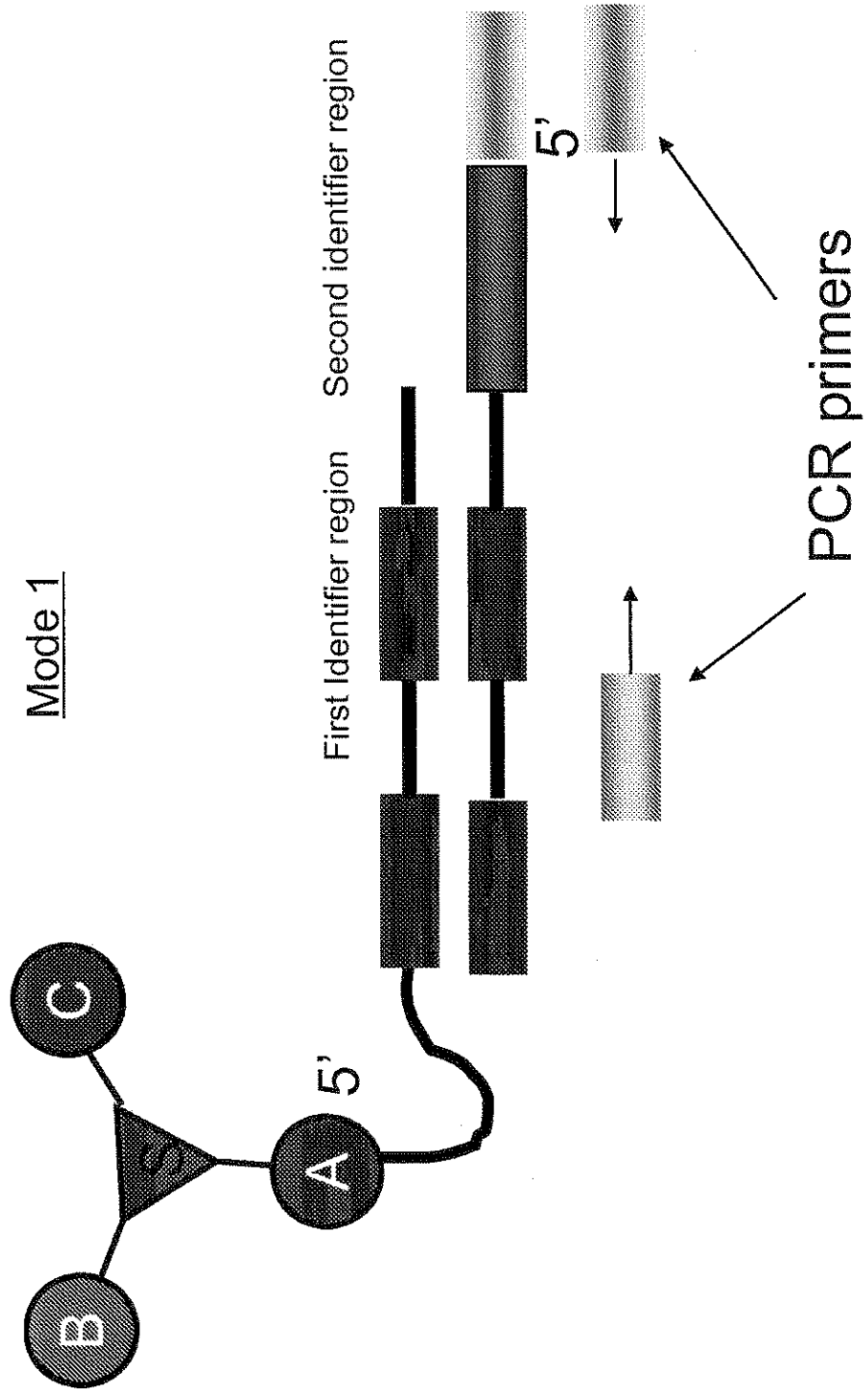
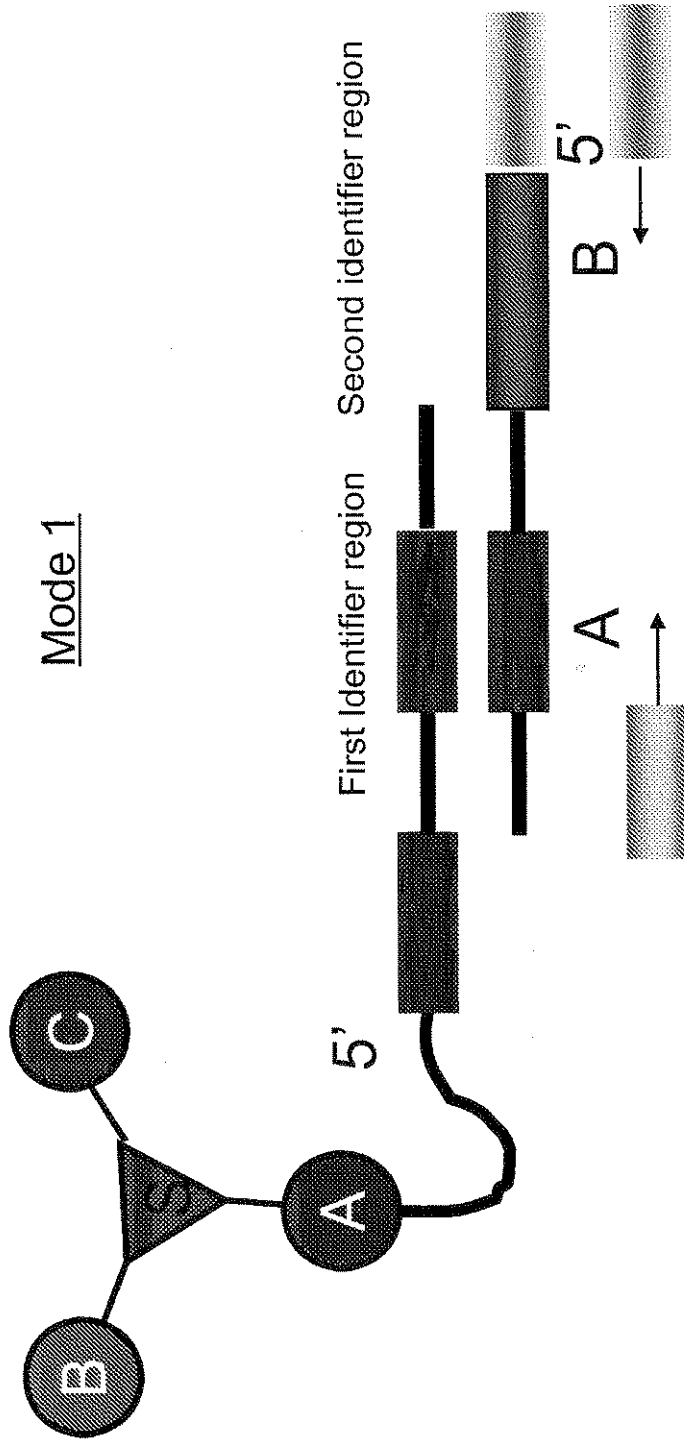


Figure 4



Achieved by hybridization of tags (non-enzymatic)
 Could introduce psoralen crosslink
 Or polymerization

Figure 5

Mode 1

- Use of a hairpin to protect DNA bases during synthetic steps, followed by enzymatic ligation of second identifier region

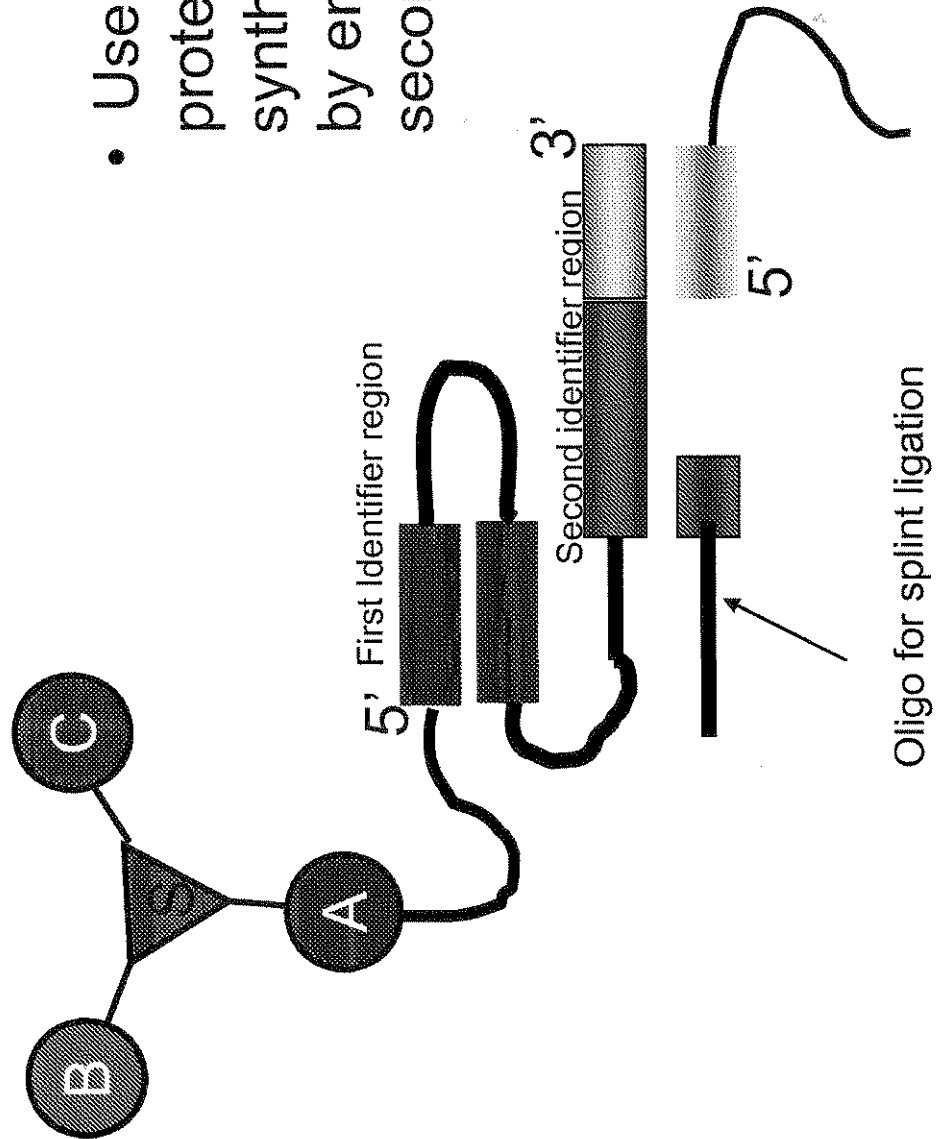
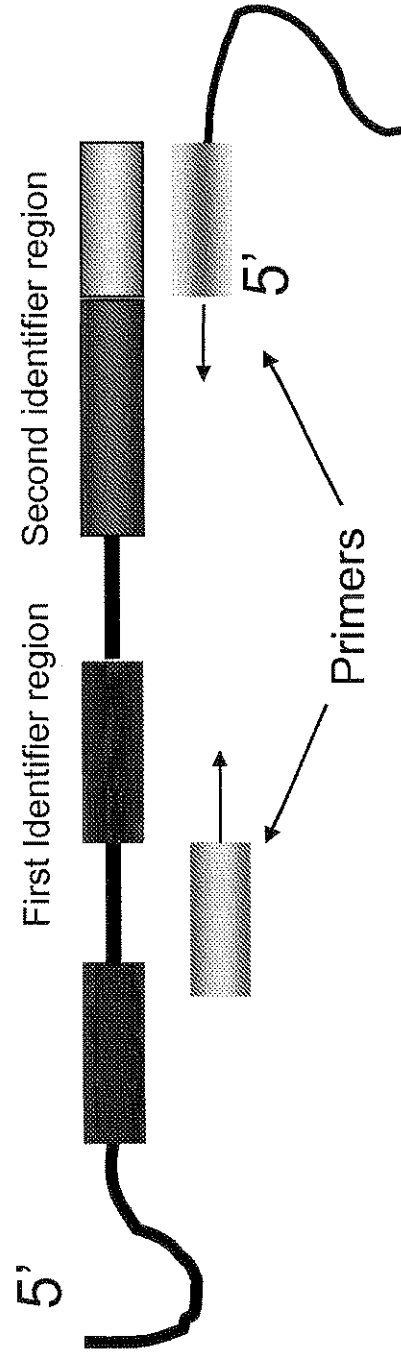
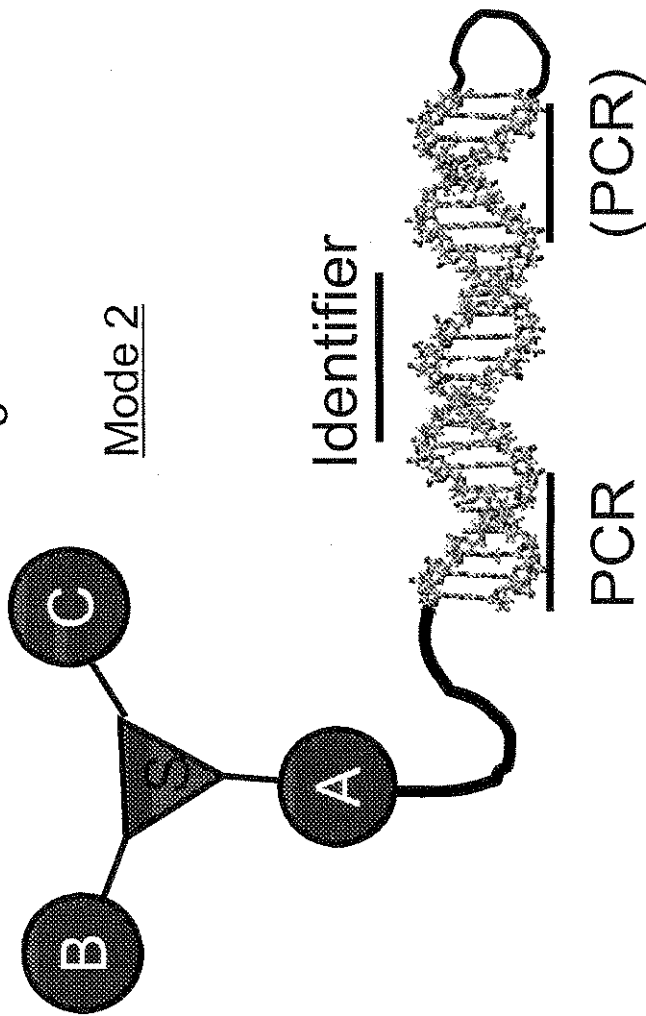


Figure 6

Mode 1

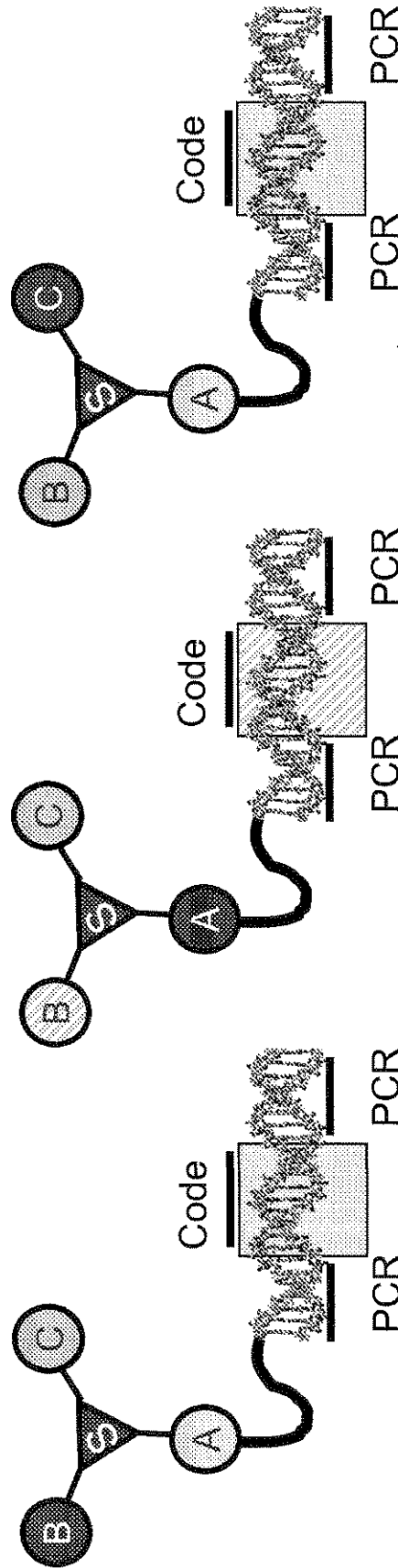
From hairpin model, amplification of identifier regions

Figure 7

Mode 2

- Small molecule tethered to ss or dsDNA
 - DNA could be hairpin on opposite end
- Linkers should vary in size and makeup (typically PEG-like, 20 to 200 atoms)
- Identifier region can be any size, but 5 nt - 20 nt preferred
- Identifier region only codes for A or B and in some schemes, S

Figure 8

Mode 2

- A is tagged
- B is "methyl"
- C is not pooled
- A x C interactions (1000 x 1000 = 1,000,000)
- A is "methyl"
- B is tagged
- C is not pooled
- B x C interactions
- A is tagged
- B is not pooled
- C is "methyl"
- A x B interactions

- 3,000,000 combinatorial fragments examined

Figure 9

Mode 2: Screening and Deconvoluting

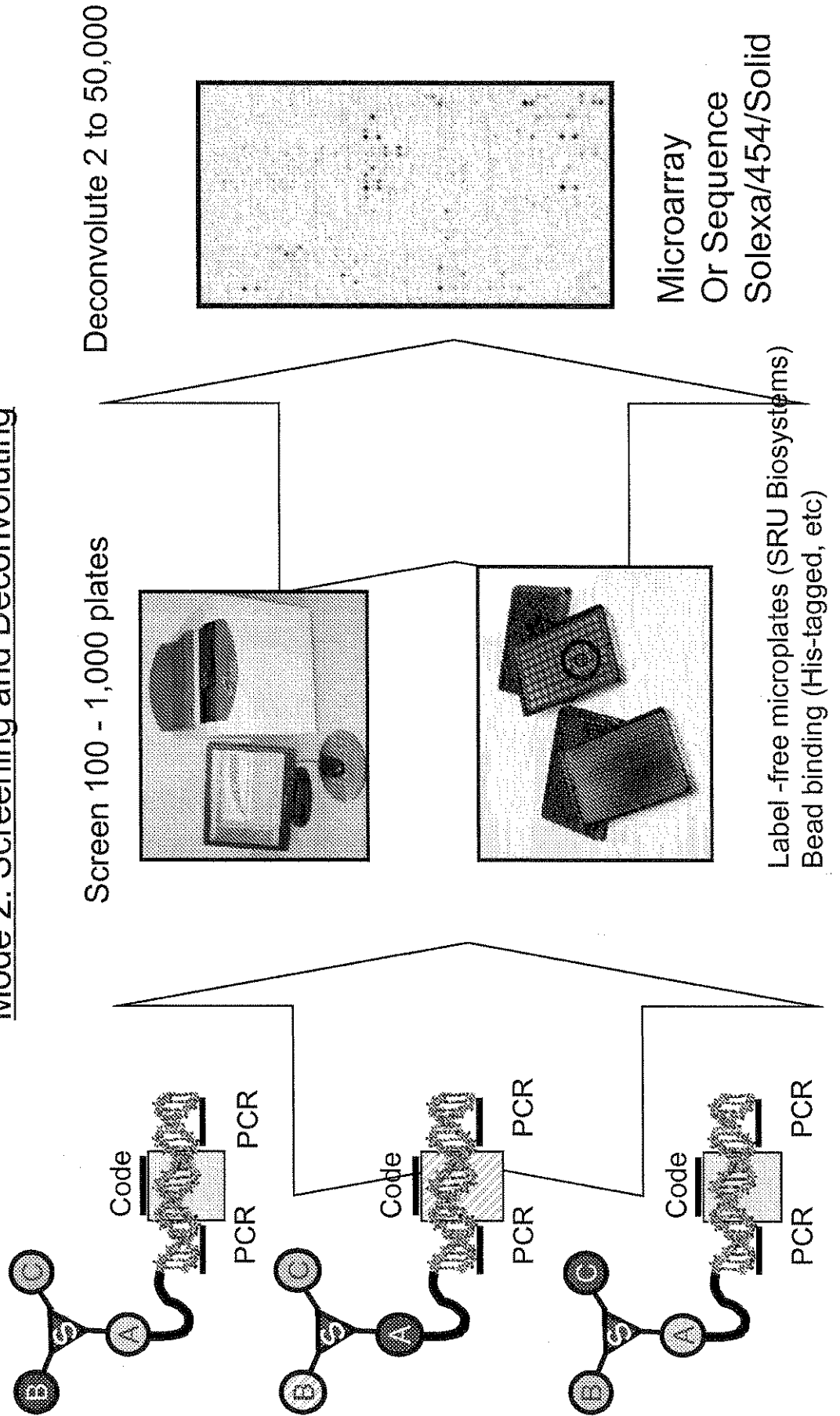


Figure 10

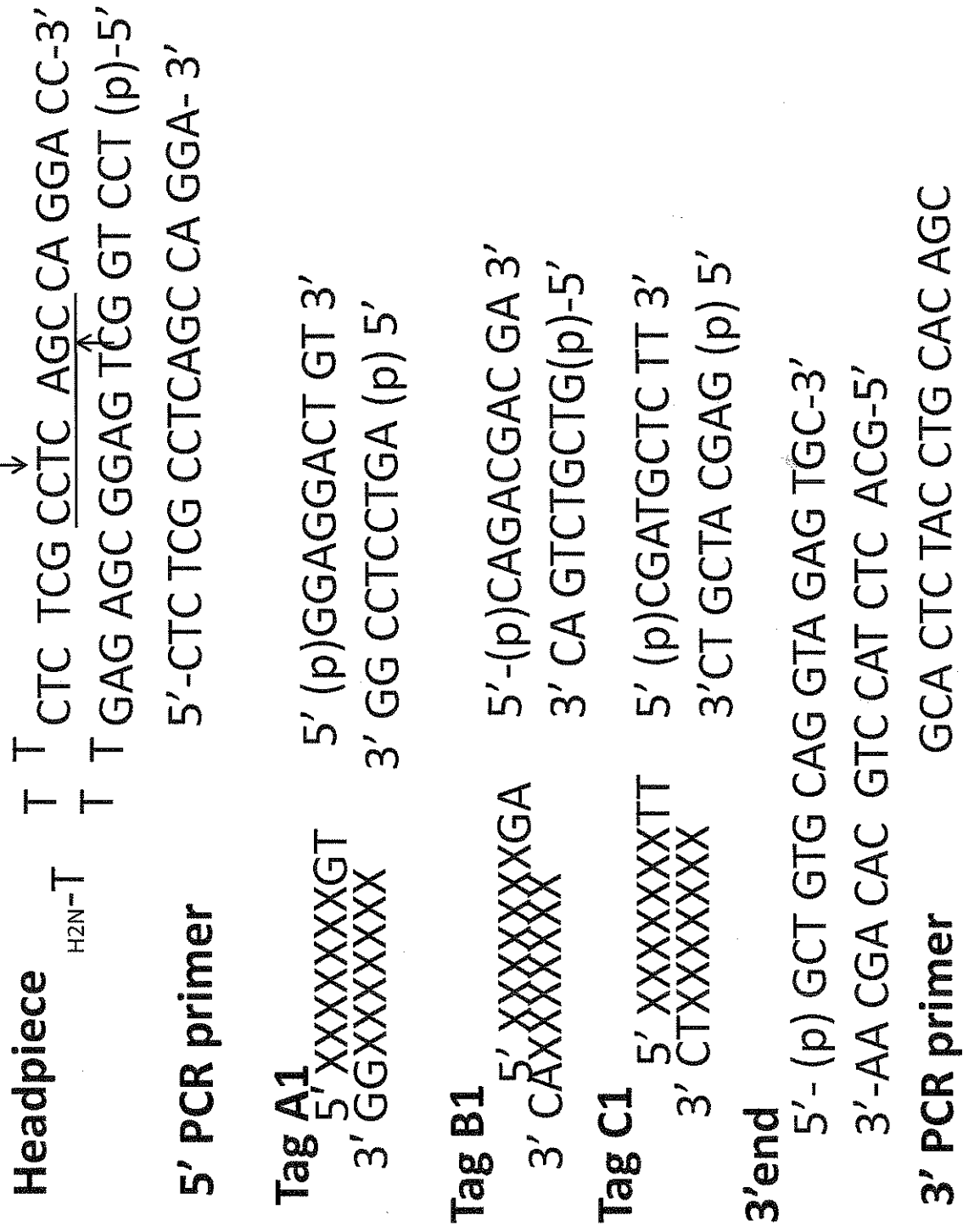


Figure 11

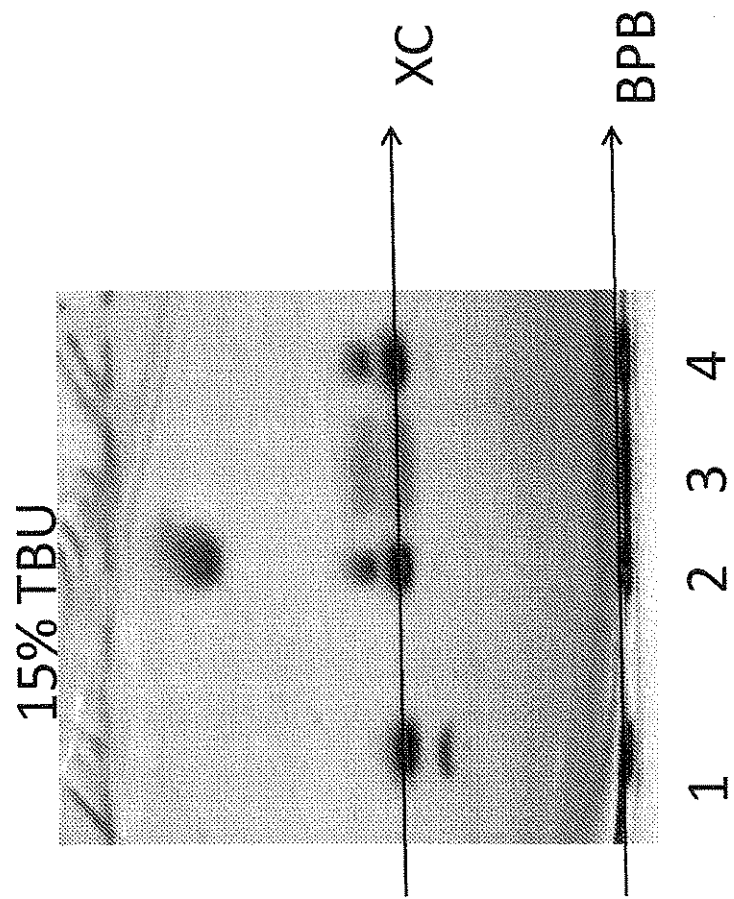


Figure 12

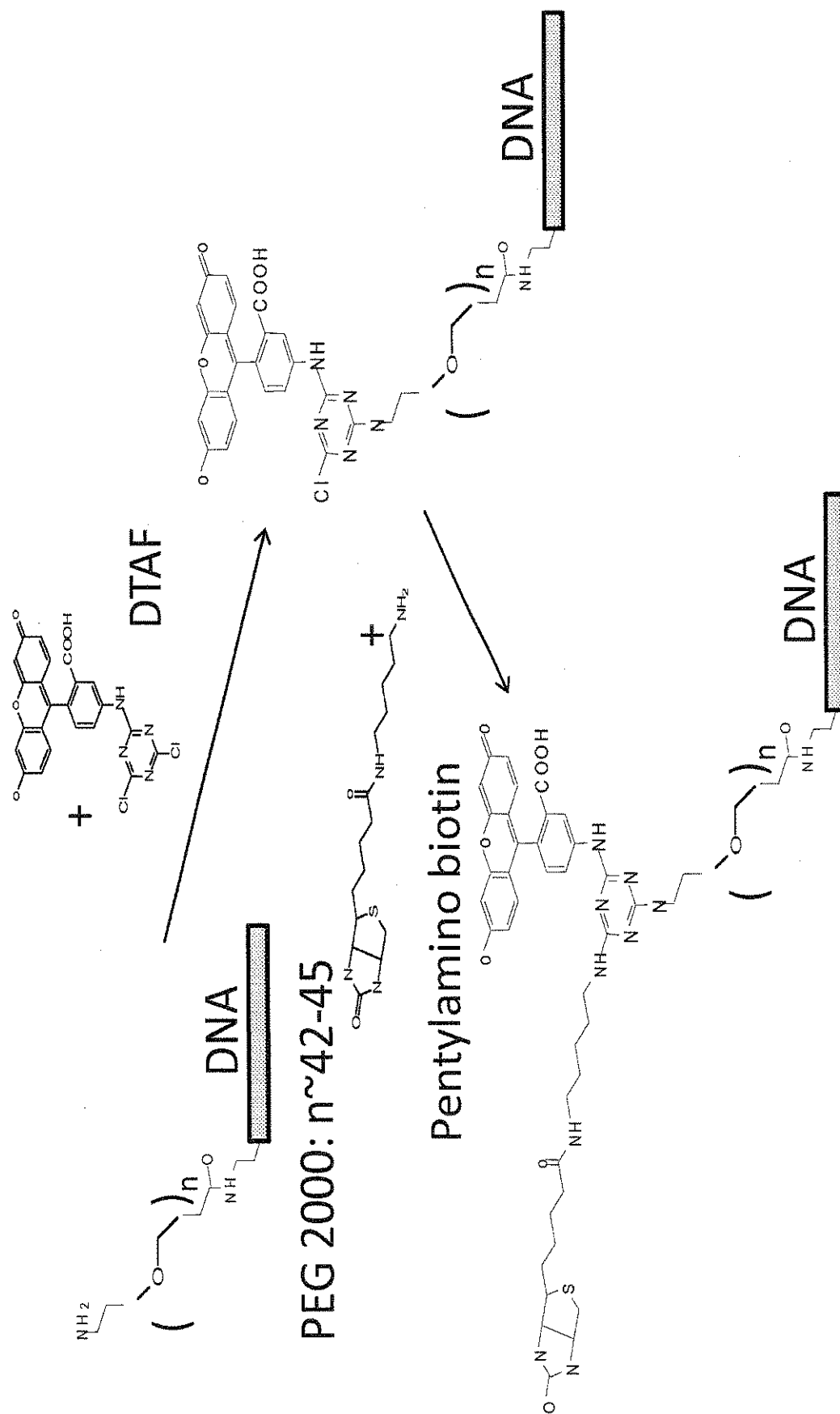
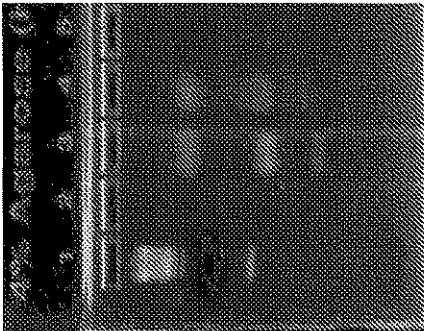


Figure 13

A. Ligation of DNA Tags

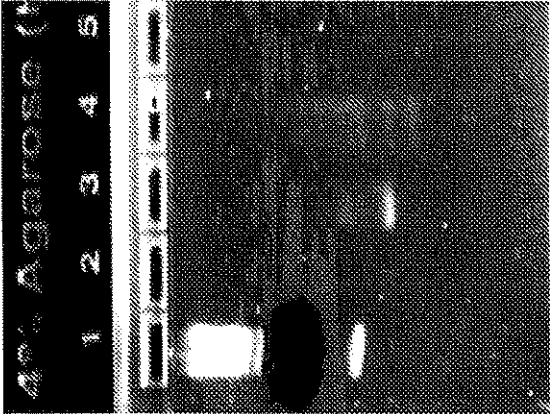
HP		A	B	C	3' end
H2N-T	T T	CTC TCG CCTC AGC CA GGA CC-3'	5'-(p)CAGACGAC GA 3'	5'-(p)CGATGCTC TT 3'	5'-(p) GCT GTG CAG GTA GAG TGC-3'
	T T	GAG AGC GGAG TCG GT CCT (p)-5'	3' CA GTCTGCTG(p)-5'	3'CT GCTA CGAG (p) 5'	3'-AA CGA CAC GTT CAT CTC ACG-5'

B



M 1 2 3

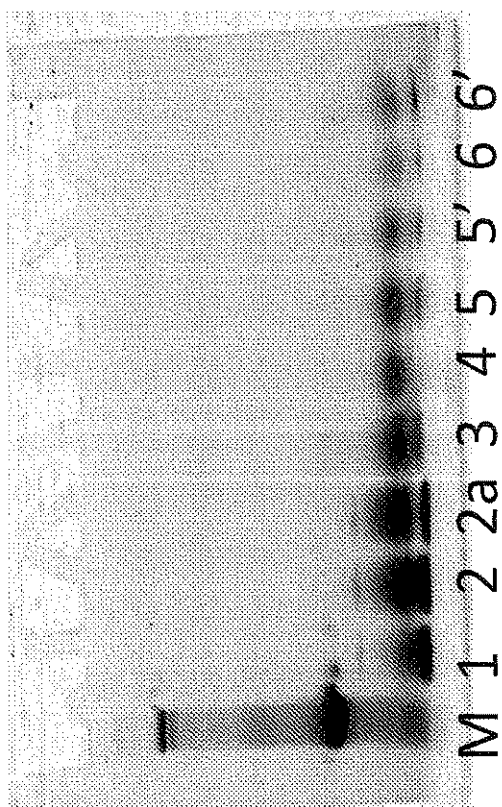
C



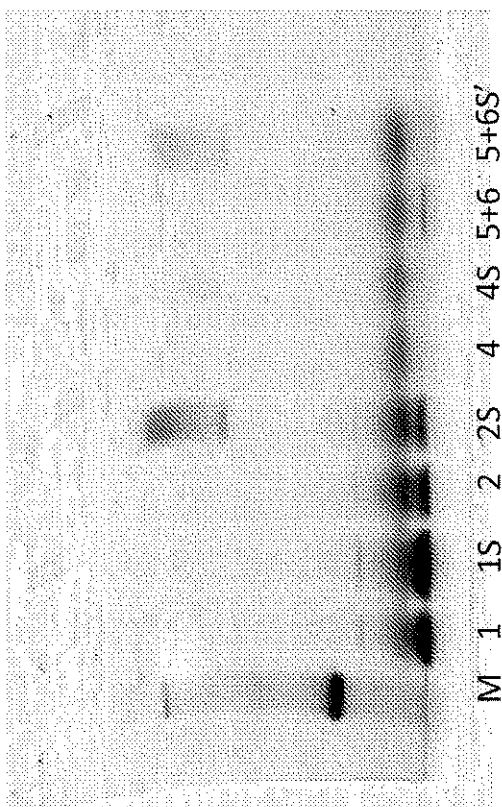
M 1 2 3 4

Figure 14

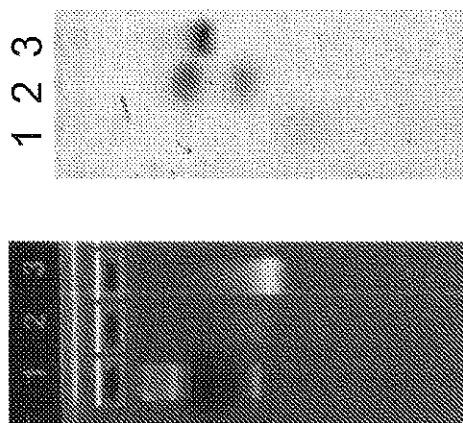
A



B



C



D

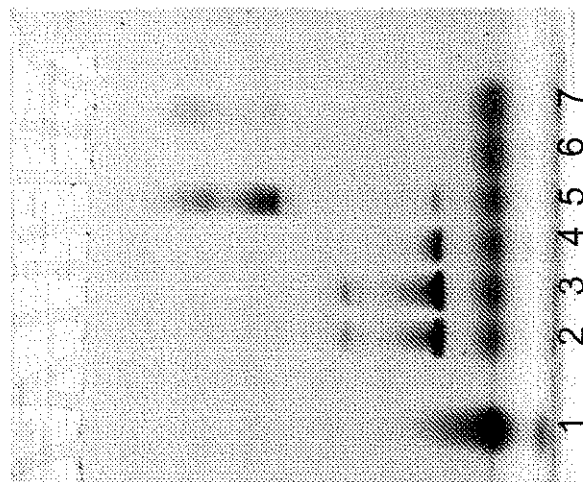


Figure 16

