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(54) METHODS FOR AMPLIFICATION OF NUCLEIC ACIDS ON SOLID SUPPORT

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(57) **ABSTRACT**

The present invention provides methods for amplifying a nucleic acid from a sample containing a mixture of nucleic acids utilizing a solid support. Methods are provided utilizing user-defined primer oligonucleotides for directional amplification that assists in further manipulation of the target nucleic acid, such as sequencing. Methods are also provided utilizing blocker and displacer oligonucleotides for generating amplified target nucleic acids of defined length. One of these methods provides a first oligonucleotide and a second oligonucleotide affixed to a solid support or separate solid supports. The first oligonucleotide is blocked to prevent extension from the 3'-terminus and has a sequence complementary to a first portion of a target nucleic acid. The second oligonucleotide has a sequence that is identical to a second portion of the target nucleic acid. In this method, a sample is applied to the solid support and the target nucleic acid within the sample binds said first oligonucleotide. The solid support is then washed to remove unbound nucleic acids. A primer sequence containing a target binding region and a polymerase promoter sequence is then annealed to the bound target nucleic acid and extended producing a first duplex nucleic acid. The target sequence is then removed leaving a first nucleic acid that can now bind the second oligonucleotide. The second oligonucleotide is extended to produce a second duplex nucleic acid that contains a second nucleic acid. The second nucleic acid is then amplified by adding a polymerase.

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FIGURE 1A

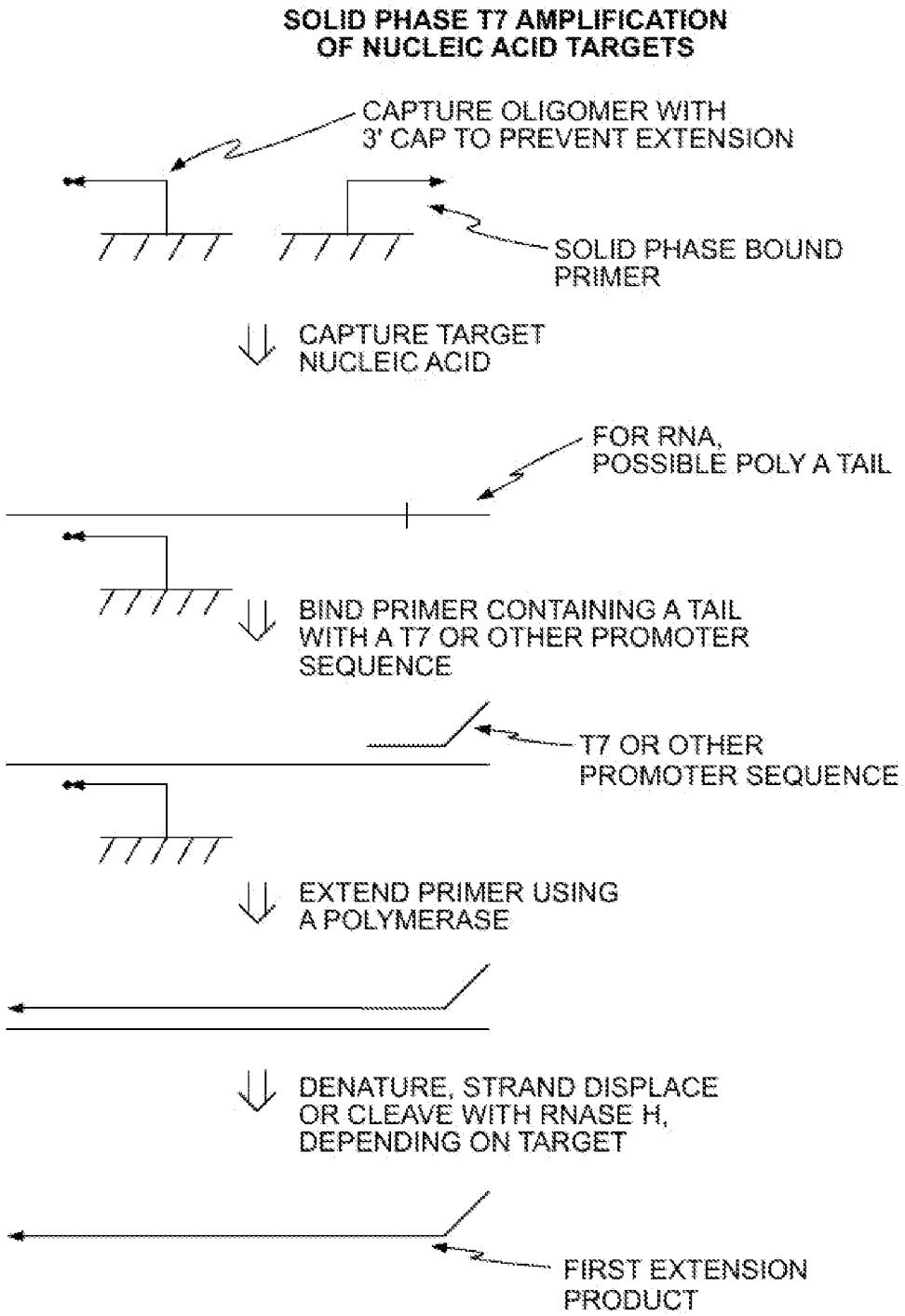

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FIGURE 1A CONT.

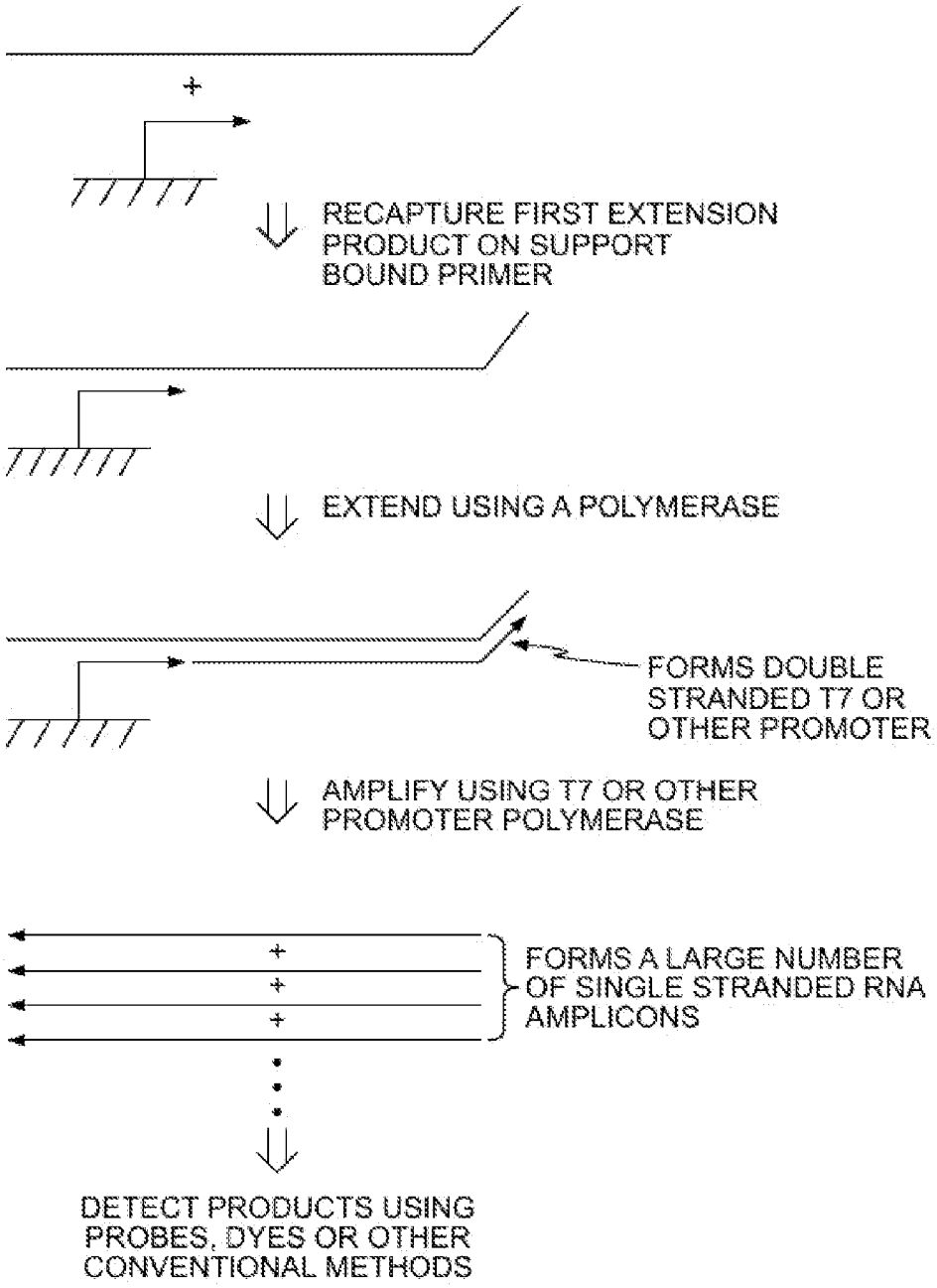

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FIGURE 1B

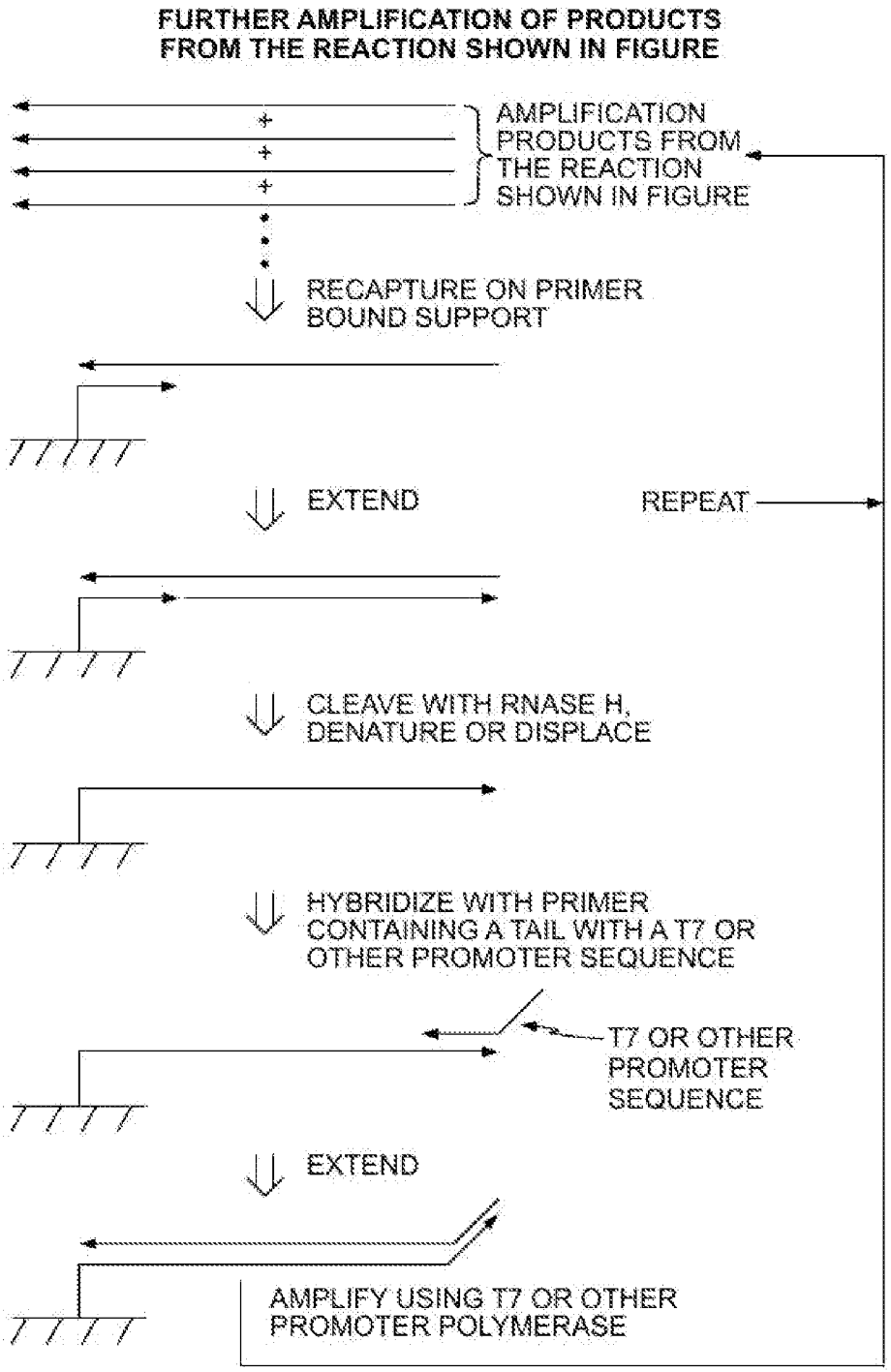

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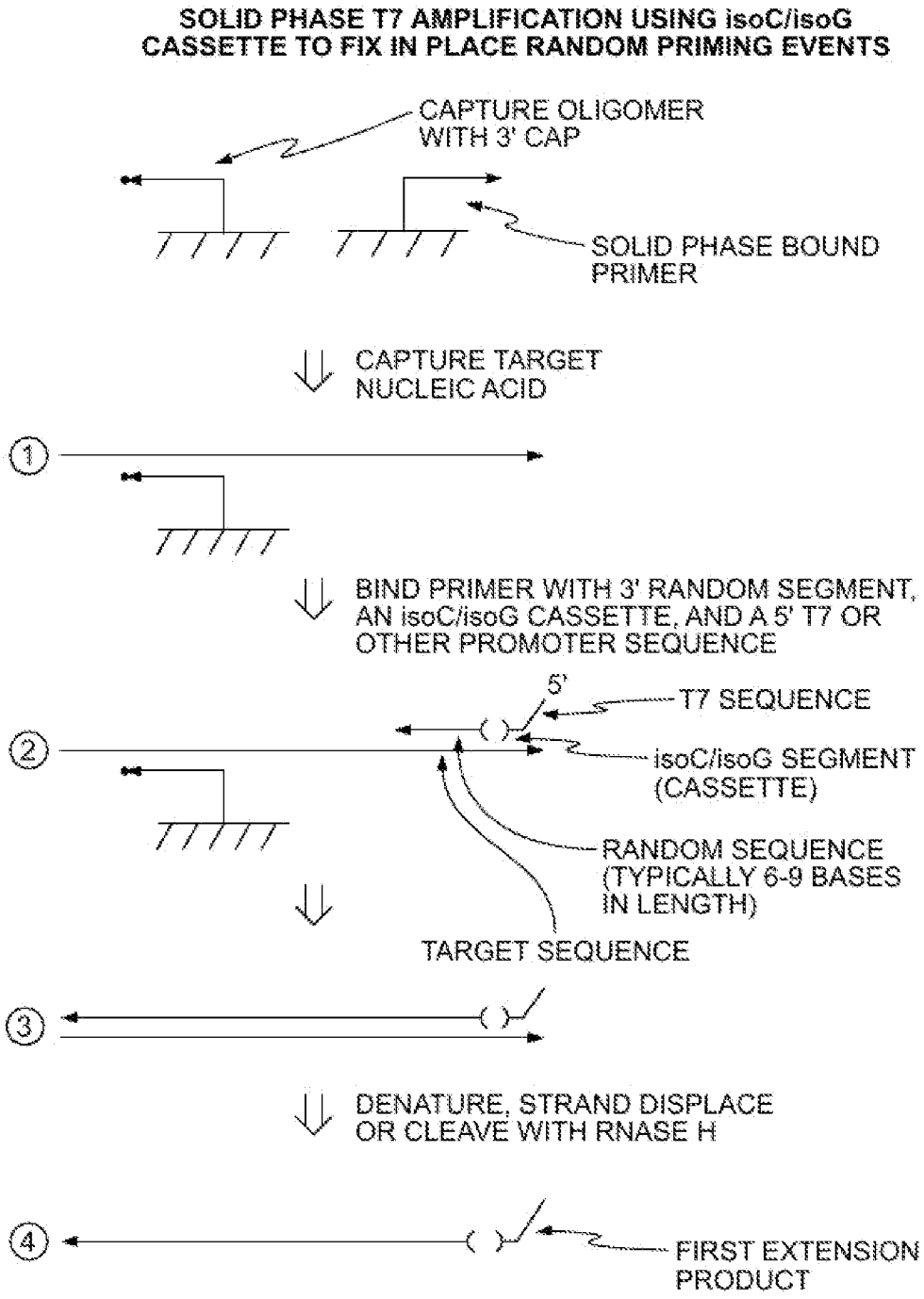
FIGURE 2



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