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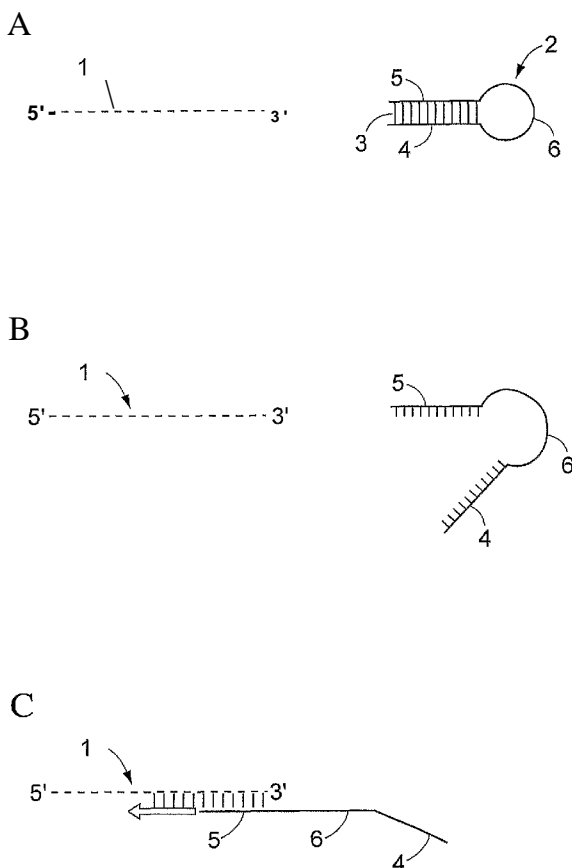
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[Continued on next page]

(54) **Title:** HOT START REVERSE TRANSCRIPTION BY PRIMER DESIGN



(57) **Abstract:** The present teachings provide methods, compositions, and kits for performing primer extension reactions. In some embodiments, a reverse transcription reaction is performed on a target polynucleotide with a hot start primer comprising a blunt-ended self-complementary stem, and a loop, and extension products form at high temperatures but reduce extension product formation at low temperatures.

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A CLASSIFICATION OF SUBJECT MATTER IPC(8). C07H 21/04 (2007 01) USPC 536/24 33 According to International Patent Classification (IPC) or to both national classification and IPC																			
B FIELDS SEARCHED Minimum documentation searched (classification system followed by classification symbols) USPC 536/2 ₄ 33 Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched Electronic data base consulted during the international search (name of data base and, where practicable search terms used) Database(s) searched WEST and Google Scholar Term(s) searched per, loop, stem, quencher, micro RNA, hot start primer, hybridize, temperature																			
C DOCUMENTS CONSIDERED TO BE RELEVANT																			
<table border="1"> <thead> <tr> <th>Category*</th> <th>Citation of document with indication where appropriate, of the relevant passages</th> <th>Relevant to claim No</th> </tr> </thead> <tbody> <tr> <td>Y</td> <td>US 5,837,442 A (TSANG) 17 November 1998 (17 11 1998), col 4, ln 59-62, col 6 ln 2-13, col 6, ln 45-47, col 6, ln 55 - col 7, ln 9, col 9, ln 13, and col 9, ln 8-21</td> <td>1-27</td> </tr> <tr> <td>Y</td> <td>US 6,365,729 B1 (TYAGI et al) 2 April 2002 (02 04 2002), col 4, ln 1-6, col 5, ln 17-25, col 6, ln 5-17, col 9, ln 12-14 col 9, ln 30-32, col 9, ln 64 - col 10, ln 25, col 10, ln 44-50, col 14, ln 60 - col 15, ln 4, col 19, ln 45 - col 20, ln 24, and col 20, ln 35-37</td> <td>1-27</td> </tr> <tr> <td>Y</td> <td>US 2004/0171039 A1 (BRUCHEZ et al) 2 September 2004 (02 09 2004), para [0048], [0072], [0130], [0139], [0140], [0142], [0145], [0188], [0220], [0226]-[0230], and [0337]</td> <td>1-27</td> </tr> <tr> <td>Y</td> <td>US 2005/0153916 A1 (MCSWIGGIN et al) 14 July 2005 (14 07 2005), para [0218]</td> <td>4, 5, 15 16 27</td> </tr> <tr> <td>Y</td> <td>US 2004/0142369 A1 (ALAJEM et al) 22 July 2004 (22 07 2004), para [0184]</td> <td>8 19</td> </tr> </tbody> </table>	Category*	Citation of document with indication where appropriate, of the relevant passages	Relevant to claim No	Y	US 5,837,442 A (TSANG) 17 November 1998 (17 11 1998), col 4, ln 59-62, col 6 ln 2-13, col 6, ln 45-47, col 6, ln 55 - col 7, ln 9, col 9, ln 13, and col 9, ln 8-21	1-27	Y	US 6,365,729 B1 (TYAGI et al) 2 April 2002 (02 04 2002), col 4, ln 1-6, col 5, ln 17-25, col 6, ln 5-17, col 9, ln 12-14 col 9, ln 30-32, col 9, ln 64 - col 10, ln 25, col 10, ln 44-50, col 14, ln 60 - col 15, ln 4, col 19, ln 45 - col 20, ln 24, and col 20, ln 35-37	1-27	Y	US 2004/0171039 A1 (BRUCHEZ et al) 2 September 2004 (02 09 2004), para [0048], [0072], [0130], [0139], [0140], [0142], [0145], [0188], [0220], [0226]-[0230], and [0337]	1-27	Y	US 2005/0153916 A1 (MCSWIGGIN et al) 14 July 2005 (14 07 2005), para [0218]	4, 5, 15 16 27	Y	US 2004/0142369 A1 (ALAJEM et al) 22 July 2004 (22 07 2004), para [0184]	8 19	
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D Further documents are listed in the continuation of Box C																			
<ul style="list-style-type: none"> * Special categories of cited documents A document defining the general state of the art which is not considered to be of particular relevance F earlier application or patent but published on or after the international filing date L document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified) O document referring to an oral disclosure use exhibition or other means P document published prior to the international filing date but later than the priority date claimed 	<ul style="list-style-type: none"> T later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention X document of particular relevance the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone Y document of particular relevance the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents such combination being obvious to a person skilled in the art & document member of the same patent family 																		
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Name and mailing address of the ISA/US Mail Stop PCT, Attn ISA/US, Commissioner for Patents P O Box 1450, Alexandria, Virginia 22313-1450 Facsimile No 571-273-3201	Authorized officer Lee W Young PCT Helpdesk 571 272 4300 PCT OSP 571 272 7774																		