



Supplementary Material

# Peroxidase-like DNA machines (PxDM) for visual detection of stem-loop primer amplification (SPA) product

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## S1. Material and Methods

Table S1. Sequence of primers

Oligonucleotide name	Sequence 5'-3'
PCR E.coli primers	
F3	GCCATCTCCTGATGACGC
B3	ATTACCGCAGCCAGACG
LAMP E.coli primers	
FIP	CTGGGGCGAGGTCGTCCTATTCCGACAAACACCACGAATT
BIP	CATTTTGCAGCTGTACGCTCGCAGCCCATCATGAATGTTGCT
F3	GCCATCTCCTGATGACGC
B3	ATTACCGCAGCCAGACG
SPA E. coli primers	
FrSL_AA	TTTATATAATATATAAAAGGTTGTTACAAAGG
BvSL_AA	TTTATATAATATATAAACGACCTCGCCC
FrSL_CC	CCTATATAATATATAGGAGGTTGTTACAAAGG
BvSL_CC	CCTATATAATATATAGGCGACCTCGCCC
Fr	AGGTTGTTACAAAGG
Bv	CGACCTCGCC

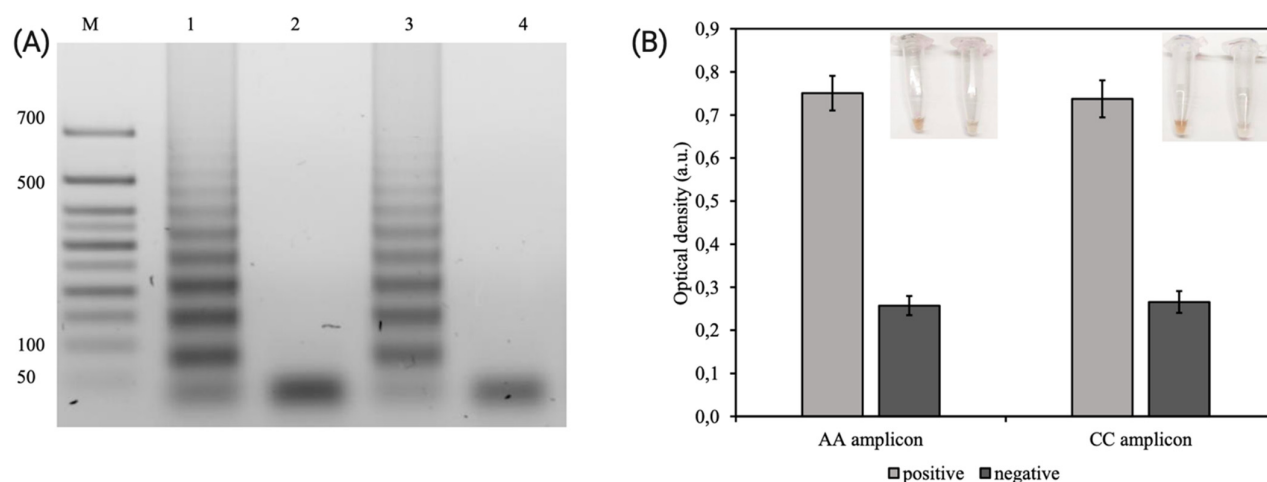
SPA <i>St. agalactiae</i> primers	
Sagal_Fr	CCGGTAGAGCTATTACC
Sagal_Bv	AATGTGTTATCCCACTTC
Sagal_FrSL	TTTATATAATATATAAAA CCGGTAGAGCTATTACC
Sagal_BvSL	TTTATATAATATATAAAA AATGTGTTATCCCACTTC
SPA Epstein-Barr virus primers	
EBV_Frw	GTCTCGCCGGACTATGGCCT
EBV_Rw	TGTTCCAGCCACCCGCCATC
EBV_FrwSL	TTTATATAATATATAAAA GTCTCGCCGGACTATGGCCT
EBV_RwSL	TTTATATAATATATAAAA TGTTCCAGCCACCCGCCATC

**Table S2. Sequence of the PxDMs**

Oligonucleotide name	Sequence 5'-3'
Target E.coli DNA	ATAGTCAGCCCATCATGAATGTTGCTGTCGATGACAGGTTGT TACAAAGGGAGAAGGGCATGGCGAGCGTACAGCTGCAAAA TGTAACGAAAGCCTGGGGCGAGGTCGTGGTATCGAAAGATA TCAATCTCGATATCCATGAAGGTGAATTCGTGGTGTGTTGTCG GACC
Target EBV DNA	TGGGCGGGCGTCTCGCCGGACTATGGCCTCGGCACGCTCGG CGTCGATGGCGGGTGGCTG GAACAGGCGGGCGAATGTGTAATCCCGGA
E.coli PxDM T1	CATATAAGGAATAGCAGAAGGCGGCTAG/HEG/CGTTACATT TTGCA/HEG/ <b>GGGTAGGG</b>
E.coli PxDM T2	GACCTCGCCCCAGGCTTT/HEG/CTAGCCGCCTTCTGCTATTC CTTATATG/HEG/TGCCCTTCTCCCTTTGTAA
E.coli PxDM F7	<b>GGGTTGGG</b> /HEG/GCTGTACGCTCGCCAACC
F7 SNP	<b>GGGTTGGG</b> /HEG/GCTGTAC <b>A</b> CTCGCCAACC
EBV PxDM f7	<b>GGGTAGGG</b> tttt GCGTGCCGAGGCcc
EBV PxDM T1	ATTCGTAATGCTGACTAGATAGATttttccCGCCATCGACGCCGA tttt <b>GGGTTGGG</b>
EBV PxDM T2	TGTTCCAGCCACCtttttATCTATCTAGTCAGCATTACGAATttttC ATAGTCCGGCGAGAC

/HEG/- hexaethylene glycol linkers. Nucleotide linkers between different functional parts of PxDM are shown in low case. Bold nucleotides form G-quadruplex. Sequences of synthetic analytes corresponded to the sequences of amplicons. Red letter is a mismatched nucleotide introduced to the sensing arm of the sensor.

### 3. Analysis of EBV amplification products



**Figure S1.** (A) An analysis of the EBV amplification products in 2% agarose. Analysis of Epstein-Barr virus amplicons obtained with AA and CC Epstein-Barr virus primers: M – marker 50 bp+, 1 – AA Epstein-Barr virus primers, 3 – CC Epstein-Barr virus primers, 2 and 4 – negative controls. (B) Analysis of Epstein-Barr virus amplicons obtained with AA and CC Epstein-Barr virus primers by Epstein-Barr virus PxDM. Colorimetric analysis of reaction buffer in the presence (positive) or absence (negative) of 300 ng of purified SPA product. Strand concentrations: 1  $\mu$ M of T1, 1  $\mu$ M of T2, 1  $\mu$ M of F7. Inset: The corresponding photograph of the color change.