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CORRECTION

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# Correction: Review: a comprehensive summary of a decade development of the recombinase polymerase amplification

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Correction for 'Review: a comprehensive summary of a decade development of the recombinase polymerase amplification' by Jia Li *et al.*, *Analyst*, 2019, **144**, 31–67.



The authors regret that the original version of the review contained some incorrect data. Corrections to the original article are listed as follows.

Some of the values in Table 5 were incorrect. The corrected version of Table 5 is presented below.

In section 2.3, data from ref. 41 were not cited correctly and should be removed. The respective passage should read as follows: "However, shorter amplicons (79 nucleotides;[37] 94 nucleotides[38–40]) and longer amplicons up to 1500 nucleotides[6] have also been reported."

In the Fig. 2 caption, "(*Bsu* or *Sau*)" should be removed after "recombinase" and inserted after "polymerase". The corrected passage should read as follows: "The recombinase disassembles from the nucleoprotein filament once the strand exchange is performed, and will be available for the next pair of primers. Next, the DNA polymerase (*Bsu* or *Sau*) extends from the 3′ end of primers."

In section 2.5, ref. 76 is not relevant as it is the same as ref. 77, and should be disregarded. The respective passage should read as follows: "However, several research groups have studied RPA reaction temperatures that lie outside of the recommended range.[38,44,45,60,62–75,77,78] The largest temperature range was tested between 15 °C and 50 °C;[62,64,69,70,77] and results indicated the marginal reaction temperature to produce a positive result should be greater than 30 °C.[62–64,66,67,69,71,74,77]"

In section 2.5, ref. 63 was not interpreted correctly. The corrected text should read: "Moreover, Lillis *et al.*[63] showed that the ambient temperature also had an effect on RPA reaction: the RPA reaction was unstable if the ambient temperature was below 30 °C, even at extended reaction time."

In section 2.8, ref. 107 is not required and should be deleted. The corrected passage should read: "For the TwistAmp® nfo kit, however, two types of amplicons are generated... (note that only the dual-labelled product will generate a positive signal in the test zone of a lateral flow strip detection based on a sandwich assay).[105,106,108]"

In section 2.8, the problems reported in ref. 117 were not sufficiently reflected in the original version of the review. Ref. 117 should be reported separately and the passage should read as follows: "As with lateral flow strip detection, direct usage of RPA amplicons is possible, but it is recommended to dilute the amplicons with the running buffer (*e.g.* 1/100 dilution) before running on the strip to (1) improve its wicking performance[114] and (2) avoid "faint ghost band" effects.[45,54,115,116] However, the dilution of the amplicon does not always prevent the appearance of a faint band, which can lead to specificity problems in the assay.[117]"

In section 3.2, ref. 166 was not cited correctly. The corrected version should read: "Results suggest that electrochemical detection could be up to 10-fold more sensitive than optical detection (by enzyme linked oligonucleotide assay).[166]"

In section 3.2, a reference was not provided for the sensitivity of the GeneXpert MTB/RIF assay. A reference should be added to the end of the following passage: "This ruthenium compound-based electrochemical detection achieved 11 CFU $mL^{-1}$ of

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*Mycobacterium tuberculosis* analytical sensitivity, which is even more sensitive than the GeneXpert MTB/RIF (Cepheid Inc.) detection (a World Health Organisation recommended tuberculosis diagnostic system that employs PCR real-time fluorescent detection; 131 CFU $mL^{-1}$)." The added reference is shown below as ref. 1.

In section 3.4, ref. 113 was not cited correctly. Instead of *Mycobacterium bovis*, *Mycobacterium tuberculosis* was used for demonstration. The corrected sentence should read: "Liu *et al.*[113] demonstrated a duplex detection of *IS6110* and *IS1081* insertion sequences of *Mycobacterium tuberculosis* using RPA-SMR assay, and achieved 3.2 and 12 genomic DNA copies per reaction analytical sensitivity respectively."

In section 4.1, ref. 182 and 183 were not cited precisely. The corrected version of the text should read: "The "microcliff" structured microchip demonstrated by Yeh *et al.* encased 200 to 1500 wells (30–100 nL per well),[182] and 224 wells (100 nL per well),[183] which allowed detection of $10^3$–$10^5$ and 10–$10^5$ copies per μL of MRSA DNA, respectively."

In section 4.2, ref. 194 should be deleted after the following sentence: "For the latter, one demonstration is on the digital video disk (DVD) by Maquieira research group, and the resulting signals can be detected by a DVD player (Fig. 12B).[188,191]"

Ref. 101 in the original article was incorrect and should be replaced with the correct reference, shown below as ref. 2.

Ref. 107 was not cited in the original article and should be disregarded.



**Table 5** RPA literature describing clinical/field trials

| Analyte(s) | Detection method | Limit of detection | Clinical/field sample(s) | Clinical sensitivity | Clinical specificity | Benchmark method | Limit of detection of benchmark method | Clinical sensitivity of benchmark method compared to RPA | Clinical specificity of benchmark method compared to RPA | Ref. |
|---|---|---|---|---|---|---|---|---|---|---|
| Nucleocapsid (N) gene of bovine coronavirus | Real-time fluorescent detection | 10 to 100 RNA copies (19 RNA copies by probit analysis) | 16 fecal and 14 nasal swab specimens collected from cattle showing intestinal and/or respiratory manifestations | 100% | 100% | Real-time RT[a]-PCR | 1000 RNA copies | The same | The same | 125 |
| Chlamydia trachomatis CDS2 gene | Lateral flow strip detection | 5–12 pathogens/ reaction | 70 self-collected first void morning urine samples from young adults (19 males and 51 females) | 83% | 100% | Roche Cobas Amplicor CT assay | — | Higher | The same | 92 |
| cAMP factor (*cfb*) gene of group B streptococci | Real-time fluorescent detection | 98 genome copies | 50 vaginal/anal samples collected from women | 96% | 100% | Real-time PCR | — | Higher | The same | 126 |
| DNA target sequence specific to *Cryptosporidium* spp. | Lateral flow strip detection | 100 oocysts per mL stool | A total of 10 human stool samples clinically verified to contain cryptosporidium by a reference laboratory and 11 stool samples from healthy volunteers presumed to be uninfected | 100% | 100% | Real-time PCR | — | Lower | The same | 127 |
| 5′-Untranslated region of Yellow fever virus (YFV) | Real-time fluorescent detection on the tube scanner | 44 genomic copies/ reaction in YFV RNA extracts; 21 genomic copies/reaction of YFV-spiked human plasma samples | 34 samples of monospecific pools of wild-caught mosquitoes collected from Kedougou, southern Senegal | 80% | 100% | Real-time RT[a]-PCR | 8 genomic copies/ reaction in YFV RNA extracts | Higher | The same | 117 |
| | Real-time fluorescent detection on the microfluidic platform | | 27 RNA samples of mosquito pools | 71.4% | 100% | | | Higher | The same | |
| *IS6110* gene of *Mycobacterium tuberculoss* (MTB) | Real-time fluorescent detection | 6.25 fg | 121 specimens including induced and expectorated sputum ($n$ = 119) and respiratory washes (bronchial and tracheal, $n$ = 2) collected from a total of 101 tuberculosis suspect cases (no more than 3 specimens/individual were tested) | 87.5% | 95.4% | Culture | — | Higher | Higher | 128 |
| *IS1081* gene of *Mycobacterium tuberculoss* | | 20 fg | | 91.4% | 100% | | | Higher | The same | |
| *Giardia* beta giardin gene | Lateral flow strip detection | $10^3$–$10^{3.5}$ cysts per mL of stool | 104 clinical stool samples | 73% | 96% | Real-time PCR | $10^{2.5}$ cysts per mL of stool | Higher | Higher | 129 |

**Table 5** *(Contd.)*

| Analyte(s) | Detection method | Limit of detection | Clinical/field sample(s) | Clinical sensitivity | Clinical specificity | Benchmark method | Limit of detection of benchmark method | Clinical sensitivity of benchmark method compared to RPA | Clinical specificity of benchmark method compared to RPA | Ref. |
|---|---|---|---|---|---|---|---|---|---|---|
| *IS6110* gene of *Mycobacterium tuberculoss* | Real-time photonic detection | $10^{-6}$-Fold diluted MTB sample | 42 clinical samples including 13 smear and culture positive samples and 22 smear and culture negative samples | 86% | 95% | Real-time PCR | — | Higher | Higher | 130[b] |
| A highly conserved 3′-untranslated region that cover DENV 1–4 | Real-time fluorescent detection | DENV serotype 1: 237 RNA copies; DENV serotype 2: 618 RNA copies; DENV serotype 3: 363 RNA copies; DENV serotype 4: 383 RNA copies | Inactivated DENV 1–4 spiked plasma and 31 DENV positive samples in Kedougou region in Senegal<br>RNA of 90 plasma samples extracted and tested between 2012–2013 by RT[a]-PCR in Bangkok (Thailand) | 98%<br>72% | —<br>— | Real-time RT[a]-PCR | — | Higher<br>Higher | —<br>— | 131 |
| 47 kDa gene sequence from the Karp strain of *Orientia tsutsugamushi* (47-RPA) and the 17 kDa gene sequence from the Wilmington strain of *Rickettsia typhi* | Lateral flow strip detection | 47 kDa gene: 53 DNA copies/reaction<br>17 kDa gene: 20 copies/reaction | 10 positive and 10 negative human samples<br>— | 80%<br>— | 100%<br>— | Real-time PCR | 47 kDa gene: 10 DNA copies/reaction<br>17 kDa gene: 6 DNA copies/reaction | Higher<br>— | Higher<br>— | 95 |
| Ribosomal 18S DNA of *Entamoeba histolytica* | Lateral flow strip detection | 2.5 fg from serial dilutions of pure DNA extracted from parasites; 40 parasites from spiked stool sample | 32 samples of DNA extracted from clinical samples | 100% | 100% | Real-time PCR | 2.5 fg from serial dilutions of pure DNA extracted from parasites | The same | The same | 132 |
| A sequence designed based on ITS sequences of the *Madurella mycetomatis* type strain CBS 109801 | Gel electrophoresis detection | 0.23 ng of DNA | 12 patient biopsy specimens | 100% | 100% | Conventional PCR | — | The same | The same | 133 |
| Ebola virus (EBOV) nucleocapsid sequence | Real-time fluorescent detection | 5 genomic copies/reaction of a molecular RNA standard;<br>15 genomic copies/reaction in EBOV-spiked human plasma samples | 928 post-mortem swab samples | 100% | 100% | Real-time RT[a]-PCR | — | The same | The same | 134 |

Table 5 *(Contd.)*

| Analyte(s) | Detection method | Limit of detection | Clinical/field sample(s) | Clinical sensitivity | Clinical specificity | Benchmark method | Limit of detection of benchmark method | Clinical sensitivity of benchmark method compared to RPA | Clinical specificity of benchmark method compared to RPA | Ref. |
|---|---|---|---|---|---|---|---|---|---|---|
| Orf virus (ORFV) DNA polymerase gene segments | Real-time fluorescent detection | 100 DNA copies | 22 samples collected from suspected cases of Orf, 8 nasal swabs collected from experimentally infected sheep and 5 samples obtained from healthy goats | 86% | 100% | Real-time PCR | — | Higher | The same | 135 |
| Leader peptidase A (*LepA*) gene of *Streptococcus pneumoniae* | Real-time fluorescent detection | 4.1 genome equivalents/reaction | 15 blood samples including 11 confirmed culture positive and 4 confirmed culture negative for *Streptococcus pneumoniae* | 100% | 100% | Real-time PCR | 5.1 genome equivalents/reaction | The same | The same | 97 |
| Orf virus (ORFV) DNA polymerase gene segments | Lateral flow strip detection | 80 copies/reaction of DNA plasmid | 24 ORFV-spiked tissues samples, 53 samples collected from goats with suspected ORFV infection, 8 nasal swabs samples and 5 tissues samples from healthy goats | 100% | 100% | Real-time PCR | — | The same | The same | 64 |
| *Leishmania donovani* (LD) kinetoplast minicircle DNA | Real-time fluorescent detection | 100 DNA copies applying the LD DNA linearised plasmid; 1 genomic DNA copy | 96 buffy coats and skin biopsies collected from visceral leishmaniasis, asymptomatic and post-kala-azar dermal leishmaniasis | 100% | 100% | Real-time PCR | — | The same | The same | 121 |
| Highly pathogenic porcine reproductive and respiratory syndrome virus (HP-PRRSV) NSP2 gene | Real-time fluorescent detection | 70 RNA copies/ reaction | 68 tissue samples and 10 serum samples collected from suspected pigs of HP-PRRSV, 35 serum samples and 12 tissue samples collected from healthy pigs | 97.6% | 100% | Real-time RT[a]-PCR | — | Higher | The same | 136 |
| 100% conserved sequence of a major capsid protein gene of all cyprinid herpesvirus 3 strains | Gel electrophoresis detection | 10 copies of genomic DNA | 12 confirmed latently infected fish and 1 confirmed uninfected fish | 100% | 100% | Real-time PCR | — | Lower | The same | 66 |
| cAMP factor (cfb) gene of group B streptococci | Real-time fluorescent detection | 6.25–12.5 genome equivalents | 124 clinical samples | 100% | 100% | Real-time PCR | 3.1–6.25 genome equivalents | The same | The same | 137 |
| Non-structure protein 1 (nsP1) of Chikungunya virus (CHIKV) | Real-time fluorescent detection | 80 genome copies of extracted RNA from CHIKV isolate LR strain | 58 suspect Chikungunya fever cases | 100% | 100% | Real-time RT[a] PCR | 80 genome copies of extracted RNA from CHIKV isolate LR strain | The same | The same | 87 |
| A sequence designed in NS2A region conserved among all Zika virus lineages | Real-time fluorescent detection | 21 RNA copies | 25 positive and 9 negative urine samples collected during the Zika virus epidemic in Tuparetama, Brazil | 92% | 100% | Real-time RT[a]-PCR | — | Higher | The same | 138 |

**Table 5** *(Contd.)*

| Analyte(s) | Detection method | Limit of detection | Clinical/field sample(s) | Clinical sensitivity | Clinical specificity | Benchmark method | Limit of detection of benchmark method | Clinical sensitivity of benchmark method compared to RPA | Clinical specificity of benchmark method compared to RPA | Ref. |
|---|---|---|---|---|---|---|---|---|---|---|
| G-protein-coupled chemokine receptor (GPCR) gene of lumpy skin disease virus (LSDV) | Real-time fluorescent detection | 100 DNA copies (179 DNA copies by probit analysis) | 12 negative skin samples and 22 skin nodules of suspected LSDV-infected cattle collected during the summer of 2012 in Dakahlia Governorate, Egypt | 100% | 100% | Real-time PCR | 37 DNA copies | The same | The same | 139 |
| *IS900* gene of *Mycobacterium avium* subsp. *paratuberculosis* (MAP) | Real-time fluorescent detection | 16 plasmid copies per μL; 500 fg genomic DNA/ reaction | Archived DNA of MAP positive blood ($n$ = 14), sperm ($n$ = 18), faecal ($n$ = 12) and tissue ($n$ = 4) samples and 20 MAP-negative faecal samples | 89.5% | — | Real-time PCR | 1 plasmid copies per μL; 50 fg genomic DNA/ reaction | Higher | — | 140 |
| T1E4 gene of prostate cancer | Real-time fluorescent detection | 1000 RNA copies | 9 urine samples obtained from prostate cancer and 2 urine samples from healthy individuals | 90% | 100% | Real-time RT[a]-PCR | — | The same | The same | 141 |
| NS1 gene of porcine parvovirus (PPV) | Real-time fluorescent detection | 300 DNA copies | 101 clinical tissue samples (serum, liver, kidney, lymph node, spleen and duodenum) collected from pig farms with suspected cases of PPV in Gansu province, China, and 27 clinical samples (serum, kidney and duodenum) collected from healthy pigs | 94.4% | 100% | Real-time PCR | — | Higher | The same | 54 |
| Nucleocapsid gene of type 2 porcine reproductive and respiratory syndrome virua (PRRSV) | Real-time fluorescent detection | 100 RNA copies (690 RNA copies by probit analysis) | 60 clinical samples (lymph node, lung, spleen and liver) collected from diseased pigs suspected of having PRRS from 5 pigs farms in Hebei province, China from 2015–2016 | — | — | Real-time RT[a]-PCR | 100 RNA copies | — | — | 142 |
| Cytochrome b gene of *Theileria annulata* | Lateral flow strip detection | 2 pg genomic DNA | 17 anticoagulated blood samples collected from tropical theileriosis endemic areas in Gansu province, China | — | — | Real-time PCR | — | — | — | 67 |
| pirA-like gene of *Vibrio owensii* | Real-time fluorescent detection | 2 plasmid copies (2.84 plasmid copies by probit analysis) | 138 clinical shrimp obtained from immersion bioassay, including 70 shrimp acute hepatopancreatic necrosis disease (AHPND) infected shrimp and 68 non-AHPND infected shrimp | 100% | 100% | Real-time PCR | — | Lower | Lower | 143 |

Table 5 *(Contd.)*

| Analyte(s) | Detection method | Limit of detection | Clinical/field sample(s) | Clinical sensitivity | Clinical specificity | Benchmark method | Limit of detection of benchmark method | Clinical sensitivity of benchmark method compared to RPA | Clinical specificity of benchmark method compared to RPA | Ref. |
|---|---|---|---|---|---|---|---|---|---|---|
| rRNA gene of *Fasciola hepatica* | Gel electrophoresis detection | 1.6 pg $\mu L^{-1}$ DNA copies | 102 human stool samples selected from banked specimens | 87.8% | 100% | Real-time PCR | 1.6 pg $\mu L^{-1}$ DNA copies | Lower | The same | 144 |
| | Lateral flow strip detection | 1.0 pg $\mu L^{-1}$ DNA copies | | 95.2% | 90.4% | | | Lower | Higher | |
| N gene of pest des petits ruminants virus (PPRV) | Real-time fluorescent detection | 100 plasmid copies | 32 clinical samples collected from suspected cases of PPRV in Gansu province, China and 5 samples obtained from healthy sheep | 90% | 100% | Real-time RT[a]-PCR | 10 plasmid copies | Higher | The same | 145 |
| | Lateral flow strip detection | 150 plasmid copies | | 90% | 100% | | | Higher | The same | |
| ITS2 gene of *Phytophthora infestans* | Real-time fluorescent detection | 50 fg $\mu L^{-1}$ of genomic DNA | 24 potato leaf samples collected from fields with and without visible symptoms of late blight infections in New Brunswick and Quebec provinces, Canada, respectively | 33.3% | 100% | LAMP | 50 fg $\mu L^{-1}$ of genomic DNA | Higher | Lower | 146 |
| ORF2 gene of porcine circovirus type 2 (PCV2) | Real-time fluorescent detection | 100 plasmid copies | 65 clinical samples (spleen, inguinal lymph node, tonsil, lung and serum) collected from suspected PCV2 infection pigs from 8 pig farms in Shandong province, China; 37 clinical samples (inguinal lymph node, tonsil, lung and serum) collected from Gansu Province, China, and 10 PCV1 positive samples conserved in the laboratory | 100% | 100% | Real-time PCR | 80 plasmid copies | The same | The same | 69 |
| | Lateral flow strip detection | 100 plasmid copies | | 100% | 100% | | | The same | The same | |
| gD gene of pseudorabies virus | Real-time fluorescent detection | 100 DNA copies | 76 clinical samples (tonsil, heart, spleen, lymph nodes, lung and serum) collected from pig farms in Shandong province, China, and 26 clinical samples (lymph nodes, tonsil and serum) collected from healthy pigs | 93.3% | 100% | Real-time PCR | — | Higher | The same | 70 |
| | Lateral flow strip detection | 160 DNA copies | | 93.3% | 100% | | | Higher | The same | |
| B1 gene of *Toxoplasma gondii* | Lateral flow strip detection | 0.1 oocysts/reaction | 35 soil samples and 15 water samples collected from parks, residential areas, schools and gutterways in Lanzhou city, Gansu rovince, China, during August 2016 | 100% | 100% | Nested PCR | 1 oocyst/reaction | The same | The same | 71 |

**Table 5** *(Contd.)*

| Analyte(s) | Detection method | Limit of detection | Clinical/field sample(s) | Clinical sensitivity | Clinical specificity | Benchmark method | Limit of detection of benchmark method | Clinical sensitivity of benchmark method compared to RPA | Clinical specificity of benchmark method compared to RPA | Ref. |
|---|---|---|---|---|---|---|---|---|---|---|
| RNA transcript of TMPRSS2:ERG (a fusion gene for prostate cancer) | RPA fluocculation assay | $10^5$ RNA copies | Clinical urine specimens from 10 metastatic castration-resistant promising prostate cancer patients and 5 healthy control patients | 70% | 100% | Conventional RT[a]-PCR | — | The same | The same | 101 |
| VP2 gene of porcine parvovirus | Real-time fluorescent detection | 100 DNA copies (103 DNA copies by probit analysis) | 115 clinical samples (lymph node, lung, spleen, kidney and duodenum collected from pigs with reproductive disorders, diarrhea or respiratory disease in Hebei province, China from 2014 to 2016 | 100% | 100% | Real-time PCR | 100 DNA copies | The same | The same | 147 |
| G-protein-coupled chemokine receptor (GPCR) gene of *Capripoxvirus* | Real-time fluorescent detection | 300 plasmid copies | 107 clinical samples (liver, lung, kidney, spleen, skin and blood) collected from 14 suspected sheep and 6 suspected goats in Gansu province which were characterised by pyrexia, excessive salivation and generalised pock lesions in the skin during the period of October 2014 to August 2015 | 97% | 100% | Real-time PCR | — | Higher | The same | 148 |
| | Lateral flow strip detection | 300 plasmid copies | | 97% | 100% | | | Higher | The same | |
| Nucleocapsid protein gene of canine distemper virus | Real-time fluorescent detection | 9.4 RNA copies (31.8 RNA copies by probit analysis) | 32 nasal/oropharyngeal swabs collected from 20 dogs of both sexes (various breeds and ages) from the animal hospital of Agricultural University of Hebei and 12 raccoon dogs from the farms in Hebei Province, China from 2014 to 2016 | 100% | 100% | Real-time RT[a]-PCR | 94 RNA copies | The same | The same | 149 |
| *imp* gene of *Candidatus* Phytoplasma oryzae | Real-time fluorescent detection | 1–10 plasmid copies | 66 Napier grass samples from various geographical locations in western Kenya | 100% | 57.1% | Real-time PCR | — | Lower | The same | 79 |
| | Lateral flow strip detection | 10–100 plasmid copies | | — | — | | | | | |

**Table 5** *(Contd.)*

| Analyte(s) | Detection method | Limit of detection | Clinical/field sample(s) | Clinical sensitivity | Clinical specificity | Benchmark method | Limit of detection of benchmark method | Clinical sensitivity of benchmark method compared to RPA | Clinical specificity of benchmark method compared to RPA | Ref. |
|---|---|---|---|---|---|---|---|---|---|---|
| *imp* gene of *Candidatus* Phytoplasma mali | Real-time fluorescent detection | 10 copies of cloned plasmid | 38 roots of field samples from apple (*Malus domestica*) trees collected in autumn | 100% | 100% | Real-time PCR | — | The same | The same | 72 |
| | Lateral flow strip detection | 10 copies of cloned plasmid | 2014, in spring 2015 and in June 2016 in private orchards or in the experimental field of the Institute for fruit growing in Samochvalovichi, Belarus | 100% | 100% | | | The same | The same | |
| N gene of rabies | Real-time fluorescent detection | 1000 RNA copies per μL of strains SAD B19, Bobcat USA and Kelev | A panel of RNA from 33 field samples | 97% | — | Real-time PCR | 1 RNA copies per μL of strains SAD B19, Bobcat USA and Kelev | Higher | — | 150 |
| *KRAS* oncogenic mutation gene G12D on Exon 12 | Real-time silicon photonic microring-based detection | 1% to 100% of the mutant cells | 70 frozen tissues samples from colorectal cancer patients in Bio-Resource Center of Asian Medical Center, including 24 samples with the G12D mutation (34.3%), 26 samples with G13D mutation (37.1%) and 20 samples with no mutation (28.6%) | 100% | 100% | Conventional PCR | 30% to 100% of the mutant cells | Lower | The same | 151 |
| *KRAS* oncogenic mutation genes G13D on Exon 13 | | | | 100% | 100% | | | Lower | The same | |
| A consensus region that covers all 7 S-segment clades of Crimean-Congo Hemorrhagic fever virus (CCHFV) | Real-time fluorescent detection | 500 RNA copies (251 RNA copies by probit analysis) | 21 extracted patient sera samples obtained in relation to outbreaks of CCHFV in 2013–2015 in Tajikistan | 88% | 100% | Real-time PCR | — | Higher | The same | 152 |
| Canine parvovirus 2 (CPV-2) nucleocapsid protein gene | Real-time fluorescent detection | 10 copies of recombinant plasmid | 91 fecal swab samples collected from the dogs from 2012 to 2016 | 100% | 100% | Real-time PCR | 10 copies of recombinant plasmid | The same | The same | 153 |
| G gene of bovine ephemeral fever virus (BEFV) | Lateral flow strip detection | 8 plasmid copies/ reaction (corresponding to 24 RNA copies) | 104 clinical blood specimens and 24 tissue samples including 16 lung tissue specimens, 8 lymph gland specimens collected from suspected dairy cattle cases of BEFV infections in eastern China | 97.89% | 90.91% | Real-time PCR | — | Higher | Higher | 74 |

**Table 5** *(Contd.)*

| Analyte(s) | Detection method | Limit of detection | Clinical/field sample(s) | Clinical sensitivity | Clinical specificity | Benchmark method | Limit of detection of benchmark method | Clinical sensitivity of benchmark method compared to RPA | Clinical specificity of benchmark method compared to RPA | Ref. |
|---|---|---|---|---|---|---|---|---|---|---|
| *IS900* gene of *Mycobacterium avium* subsp. *paratuberculosis* | Lateral flow strip detection | 8 plasmid copies/ reaction | 320 individual fecal samples collected between September 2016 and September 2017 from 10 different dairy farms located in 10 distinct geographic regions of Shandong province, China | 100% | 97.63% | Real-time PCR | 8 plasmid copies/ reaction | The same | Higher | 77 |
| *Fno* FSC771 hypothetical protein gene of *Francisella noatunensis* subsp. *Orientalis* | Real-time fluorescent detection | 10 plasmid copies (15 plasmid copies by probit analysis) | Samples of spleen ($n$ = 78), head kidney ($n$ = 78) and water ($n$ = 5) | 100% | 84.89% | Real-time PCR | 10 plasmid copies (11 plasmid copies by probit analysis) | The same | Higher | 154 |
| VP1 gene of Enterovirus 71 subgenotype C4 (EV71-C4) | Real-time fluorescent detection | 3.767 log 10 genomic copies (LGC) | Stool samples ($n$ = 44) collected in 2017 by Shenzhen Center for Disease Control and Prevention | 100% | 100% | Real-time PCR | 2.026 log 10 genomic copies (LGC) | The same | The same | 155 |
| | | | Stool samples ($n$ = 134) collected from patients with suspected hand–foot–mouth disease at the pediatrics department of Zhujiang Hospital (Southern Medical University, Guangzhou, China) in 2009 | 89.5% | 100% | | | Lower | The same | |
| 56 kDa gene of a Karp-like strain of *Orientia tsutsugamushi* | Lateral flow strip detection | 10 copies (recombinant plasmid); 12 copies of genomic DNA | 62 animal (including *Apodemus agrarius*, *Rattus norvegicus*, *Microtus fortis* and *Neomys fodiens*) organ samples including 5 infected animals trapped in the wild, 2 infected in the laboratory and 55 uninfected animals trapped in the wild | 100% | 100% | Real-time PCR | 12 copies of genomic DNA | The same | The same | 156 |
| 23S rRNA gene of *Coxiella burnetii* | Lateral flow strip detection | 10 copies (recombinant plasmid); 7 copies of genomic DNA | DNA of spleens from 5-week old C57BL/6 female *Coxiella burnetii*-infected mice and 9 control PBS-infected mice | 100% | 100% | Real-time PCR | 7 copies of genomic DNA | The same | The same | 157 |

[a] RT: reverse transcription. [b] Clinical sensitivity and specificity were recalculated using the ESI.

The Royal Society of Chemistry apologises for these errors and any consequent inconvenience to authors and readers.

## Acknowledgements

The authors thank Valentina Paz Gacitúa Miranda, Hahn-Schickard, for thoroughly studying all of the referenced literature and for spotting gaps and mistakes.

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