International Food Research Journal 26(1): 1 - 10 (February 2019)

Journal homepage: *http://www.ifrj.upm.edu.my*

Review

Loop mediated isothermal amplification; a review on its application and strategy in animal species authentication of meat based food products

[1]Tasrip, N. A., [1]Khairil Mokhtar, N. F., 1Hanapi, U. K., [1]Abdul Manaf, Y. N., [2]Ali, M. E., [3]Cheah, Y. K., [4]Mustafa, S. and [1,3*]Mohd Desa, M. N..

[1]Halal Products Research Institute, Universiti Putra Malaysia, 43400, Serdang, Selangor, Malaysia
[2]Nanotechnology and Catalysis Research Centre, University of Malaya, 50603, Kuala Lumpur, Malaysia
[3]Department of Biomedical Science, Faculty of Medicine & Health Sciences, Universiti Putra Malaysia, 43400, Serdang, Selangor, Malaysia
[4]Department of Microbiology, Faculty of Biotechnology and Biomolecular Sciences, Universiti Putra Malaysia, 43400, Serdang, Selangor, Malaysia

Article history

Received: 15 January, 2018
Received in revised form: 28 June 2018
Accepted: 19 July 2018

Keywords

LAMP
Meat
Meat Speciation
Authentication
Adulteration

Abstract

Isothermal amplification is a technique that can amplify target DNA sequences at a single incubation temperature. Loop-mediated isothermal amplification (LAMP) is an extension of the isothermal DNA amplification technique that combines rapidity, simplicity, high specificity and sensitivity. Due to its overwhelming characteristics, LAMP has been explored for its feasibility in detecting various subjects, and recently in meat-based food products for DNA-based meat species authentication. It has been developed to target various meat species such as porcine, chicken, horse, and ostrich with sensitivity as low as 0.1 pg/µL. Further improvement with the use of magnetic beads, electrochemiluminescence and special dye such as calcein and crystal violet had increased the sensitivity of the LAMP assay. Other important characteristics were specific target gene primers as well as a shorter incubation time, warranting a good prospect for rapid testing authentication.

© All Rights Reserved

Introduction

Utilisation of animal and its by-products such as meat, gelatine and fat in various foods and non-food products has been increasing tremendously. In addition, the admixture of meats with that of a lower value without proper declaration has also been reported (Ayaz *et al*., 2006; Fajardo *et al*., 2008; Özpinar *et al*., 2013; Soares *et al*., 2013; Amin *et al*., 2015). This has triggered a great concern among consumers particularly the religious devotees who are subjected to certain regulation in meat consumption.

From a religious perspective, porcine is forbidden for consumption in Islam and Judaism (Soares *et al*., 2010; Nakyinsige *et al*., 2012), and yet porcine and its derivative have been largely utilised either as food or derivative of other form of products. Moreover, although not widely reported, porcine serum albumin (PSA) has been acknowledged as a significant allergen found in porcine meats (Kim *et al*., 2011). Meanwhile, dog, which is also forbidden in Islam, is not allowed for consumption in Buddhism (Soares *et al*., 2010; Rahman *et al*., 2014). Consuming dog meats might put individuals at risk of infection from deadly pathogens such as *Escherichia coli* serotype 107 and *Salmonella sp*., as well as at risk of contracting other harmful microbes such as anthrax, brucellosis, hepatitis and leptospirosis (Sirius Global Animal Charitable Trust, 2007).

Another example of food fraud involves beef pertaining to its origin and gender. Origin fraud occurred when beef from different countries might cost higher than from some others. For example based on Peter Shears report (2010), British sirloin beef costs £8.50/kg meanwhile Brazilian beef costs about £4.50/kg. It is difficult to differentiate the country of origins once the meat is unbadged and unbagged. As for the gender issue, beef from male animals is more valuable as compared to beef from female animals. Female cows are used for reproduction and only slaughtered when they get older, resulting in a lower quality meat as the meat has turned into

*Corresponding author.
Email: mnasir@upm.edu.my

tougher texture due to the rigidity of muscle fibres. Unscrupulous sellers sell the meats with the price of that of male cows.

In early 2013, a two-month long investigation on the authenticity of beef products revealed the presence of undeclared horse meat in a frozen burger produced in Ireland (O'Mahony, 2013). Out of 27 burgers tested, 37% of them had been found containing traces of equine DNA. Similar results have been reported when further samples of the same burger brand were purchased at the same retail outlets and tested (O'Mahony, 2013). Since labelling requirements were violated, the fraudulent nature of this matter could be assumed. O'Mahony (2013) also found salami products containing bovine and/ or porcine DNA, but not equine DNA. Low levels of porcine DNA might suggest an unintentional presence reflecting poor operation system.

Many studies have thus focused on the authentication approach to verify the originality of meat origin in related products available in the market. PCR has been widely used for authentication purposes but isothermal approach particularly the LAMP is also promising, and has recently gained much interest. The present review will therefore present the principle of LAMP, its application and strategies in meat species identification.

PCR vs Isothermal approaches in DNA detection

Polymerase chain reaction (PCR) based methods targeting specific DNA marker have been proven to be effective in identifying and revealing even in minute levels of adulteration in highly processed meat products (Fajardo, Gonza and Garcı, 2010; Ali *et al*., 2012; Doosti and Dehkordi 2014). Further enhancements involve PCR-based amplified fragment length polymorphism (PCR-AFLP) (Sasazaki *et al*., 2004), random amplification of polymorphic DNA (RAPD) (Calvo, Zaragoza and Osta, 2001), quantitative competitive (QC) PCR (Wolfand Lüthy, 2001), multiplex-PCR (Asensio *et al*., 2008), nested primer PCR (Pardo and Pérez-Villareal, 2004), microsatellite (Tajima *et al*., 2002; Rikimaru and Takahashi, 2007) and real-time PCR using probes (Fajardo *et al*., 2008; Ahmed *et al*., 2010).

Nevertheless, these methods should be performed in a well-equipped lab with a thermal cycler machine and skilled personnel (Dhama *et al*., 2014). Alternatively, approaches of DNA amplification without the use of thermal cycler have also been developed. These methods are referred to as isothermal amplification as the reaction works at a constant temperature. An important advantage of these assays is that there is no need for a high temperature for initial heat denaturation and lower temperature in the amplification steps as in PCR assays. The methods include Nucleic Acid Sequence-Based Amplification (NASBA), Transcription Mediated Amplification (TMA), Strand Displacement Amplification (SDA), Rolling Circle Amplification (RCA), Signal Mediated Amplification of RNA Technology (SMART), Loop-Mediated Isothermal Amplification (LAMP), Helicase-Dependent Amplification (HDA), Single Primer Isothermal Amplification (SPIA) and Circular Helicase Dependent Amplification (cHDA) (Walker *et al*., 1992; Lizardi *et al*., 1998; Notomi *et al*., 2000; Gill and Ghaemi 2008; Mugasa *et al*., 2009). Each of the amplification systems has different processes that result in different outcome of detections.

Some of these isothermal amplification methods are related to the complexity in primer design and enzyme regiments used in the assay. Based on primer design, SDA, SMART and LAMP are considered complicated for a new user. SDA uses four primers to generate initial amplicons; SMART assay consists of two single stranded oligonucleotide probes ("extension" and "template") that anneal to template DNA forming a three-way-junction (Tröger *et al*., 2015); meanwhile, LAMP requires four to six specific designed primers for each target region. Compared to PCR which only use one enzyme (DNA polymerase), NASBA, SMART, SDA, HDA and SPIA need more than one enzyme to allow amplification of DNA or RNA. SPIA assay needs DNA polymerase with strand displacement activity and RNaseH (Karami *et al*., 2011), meanwhile NASBA needs three different enzymatic steps (transcription/ cDNA synthesis/ RNA degradation) to accomplish an isothermal RNA amplification assays (Cho *et al*., 2005).

Among these approaches, LAMP provides a better efficiency in the amplification of large DNA sequences with better sensitivity and specificity (Mori and Notomi, 2009). In LAMP assay, pyrophosphate ions are produced in a large amount which results in the formation of white precipitate magnesium pyrophosphate. The appearance of this precipitate provides an indication that the target DNA has been amplified (Mori *et al*., 2001). In addition, an important advantage of this technique is related to how it can tolerate to some inhibitory materials that might be present and usually affect the PCR efficiency (Enomoto *et al*., 2005; Kaneko *et al*., 2005).

In a study done by Kaneko *et al*. (2007), LAMP managed to detect DNA targeting Herpes Simplex Virus-1 that was suspended in a culture medium with various biological substances, in which potential inhibitors such as amino acids or salts might be present. They concluded that the tolerance of LAMP

to biological substances was superior. Although the extent of the inhibitors and their effects in the process were not known, successful detection could indicate the ability of LAMP to overcome this problem. In addition, Kaneko *et al*. (2007) suggested that DNA extraction step can be omitted in LAMP assay, and this was demonstrated in Abdulmawjood *et al*. (2014)'s study. However the later study reported that the amplification times were longer than those using extracted DNA.

Meanwhile, Yang *et al*. (2014)'s study used a similar target region (cytochrome b gene) to be amplified by using both LAMP and PCR assays. This was done using the outer primers (F3 and B3) of the LAMP assay. The results showed that LAMP assay could detect DNA as low as 1 pg/µL in 45 min as compared to PCR assay which was only at 100 pg/µL in more than 65 min. This showed that LAMP assay could be more sensitive and rapid as compared to PCR assay. Therefore, the application of LAMP in authentication of meat fraud or adulterated meat products has come into attention as a promising tool for identifying the meat species in meat-based products.

Application of LAMP and strategies in meat spp. authentication

LAMP has been demonstrated to be sufficiently sensitive such that it allows amplification of a few copies of DNA to 109 in less than an hour under isothermal conditions which eliminates the need for thermal cycling. It employs four to six primers and specific DNA polymerases such as Bst DNA polymerase (New England Biolabs, Ipswich, MA) (Yang *et al*., 2014) or GspSSD DNA polymerase (Optigene Ltd., UK) (Cho, Dong and Cho, 2014) which are capable of inducing auto-cycling strand displacement with enhanced specificity (Cho *et al*., 2014).

The performance of LAMP mainly relies on the primers' design, successful DNA extraction and optimal incubation temperature for primers to bind at the target region. A minimum of four primers labelled as forward outer (F3), backward outer (B3), forward inner (FIP) and backward inner (BIP) primers are required, while additional two primers labelled as loop forward (LF) and loop Backward (LB) are added to accelerate the reaction (Nagamine, Hase and Notomi 2002). F3 and B3 take place during DNA strand displacement, thus referred to as strand displacing primers, while FIP and BIP are involved in loop formation (Parida *et al*., 2008). An online tool such as Primer Explorer could assist in designing LAMP primers (Dhama *et al*., 2014). The main challenge in LAMP primers' design is to ensure a low possibility of dimers formation among the primers. As of PCR, dimers formation will completely halt the amplification of target sequence. Thus, the potential of homo- and heterodimer formation among the primers must be assessed in details prior to DNA amplification process.

The principle underlying the amplification of specific target sequences in LAMP assay has been discussed in details by Notomi *et al*. (2000). Utilisation of LAMP has been initially applied in the diagnosis of pathogenic bacteria, parasites, viruses (Notomi *et al*., 2000; Tlili *et al*., 2013; Safavieh *et al*., 2014a; Safavieh *et al*., 2014b; Fernandez-Soto *et al*., 2014; Zeng *et al*., 2014), and genetically-modified organisms (Ahmed *et al*., 2009; Kiddle *et al*., 2012; Zhang *et al*., 2013; Huang *et al*., 2014; Wang *et al*., 2015), as well as in authenticating medicinal materials (Mahony *et al*., 2013). Due to the simplicity of this technique, LAMP has been extended to food analysis. For instance, some studies have recently utilised this approach in the identification of meat species of meat products (Ahmed *et al*., 2010; Abdulmawjood *et al*., 2014; Cho *et al*., 2014; Kanchanaphum, Maneenin and Chaiyana, 2014; Yang *et al*., 2014; Zahradnik *et al*., 2015; Lee *et al*., 2016; Ran *et al*., 2016; Roy *et al*., 2016a; Roy *et al*., 2016b). Table 1 shows the summary of meat identification studies using LAMP assays, while Figure 1 shows the summary of approaches in the LAMP assays that have been developed thus far.

Simplex LAMP

Single animal species detection using simplex-LAMP has been successfully developed for the detection of porcine (Ahmed *et al*., 2010; Kanchanaphum *et al*., 2014; Yang *et al*., 2014; Lee *et al*., 2016; Ran *et al*., 2016), horse (Zahradnik *et al*., 2015) and ostrich meats (Abdulmawjood *et al*., 2014). In these studies, two modes of LAMP assay were employed, which were direct-real time LAMP and real time LAMP. Direct-real time PCR LAMP allows for direct detection of DNA without prior DNA extraction. On the other hand, real time LAMP requires the DNA to be extracted and purified prior to LAMP analysis. DNA extraction can be various utilising the common processes such as kits or conventional approach as long as the concentration and purity of DNA is sufficient as of PCR standard.

Lee *et al*. (2016), Ran *et al*. (2016) and Yang *et al*. (2014) showed a high potential of LAMP for rapidity and good sensitivity in porcine identification. The assays developed by Yang *et al*. (2014) and Lee *et al*. (2016) had a similar sensitivity of 1 pg porcine DNA,

Table 1 Summary of meat identification studies using LAMP assays.

Target species	Modes (S/M)	Target gene	Length (bp)	Sensitivity (pg/µL)	Specificity Admixture (%)	Duration (m)	Reference
Porcine	Indirect S	12S rRNA	229	~20330	-	65	Ahmed *et al*. (2010)
	Indirect M	COI and II	378	1	-	30	Cho *et al*. (2014)
	Indirect S	Cytochrome b	347	1	0.01	45	Yang *et al*. (2014)
	Indirect S	D-loop	185	10000	-	50	Kanchanaphum *et al*. (2014)
	Indirect S	ND1	274	0.5	0.0001	65	Ran *et al*. (2016)
	Direct & Indirect S	D-loop	221	1	0.001	30	Lee *et al*. (2016)
	Indirect S	Cytochrome b	207	100	-	20	Roy *et al*. (2016a)
	Indirect S	Cytochrome b	252	1	-	120	Roy *et al*. (2016b)
	Indirect S	Cytochrome b	252	1	-	120	Roy *et al*. (2017)
	Indirect S	Cytochrome b	252	0.1	-	120	Azam *et al*. (2018)
Chicken	Indirect S	12S rRNA	222	~78.68	-	65	Ahmed *et al*. (2010)
	Indirect M	ATP synthase	367	0.1	0.0001	30	Cho *et al*. (2014)
	Indirect S	Cytochrome b	190	1	-	20	Roy *et al*. (2016a)
Beef	Indirect S	12S rRNA	203	~23.63	-	65	Ahmed *et al*. (2010)
	Indirect M	D-loop	331	10	-	30	Cho *et al*. (2014)
Horse	Indirect M	ATP synthase	369	1	0.001	30	Cho *et al*. (2014)
	Indirect S	NADH dehydrogenase	213	100	0.001	90	Zahradnik *et al*. (2015)
Ostrich	Direct & Indirect S	Cytochrome b	213	0.35	0.01	>20	Abdulmawjood *et al*. (2014)
Goat	Indirect M	D-loop	247	1	-	30	Cho *et al*. (2014)
Sheep	Indirect M	Cytochrome b	320	1	0.001	30	Cho *et al*. (2014)
Duck	Indirect M	COI and II	363	1	-	30	Cho *et al*. (2014)
Turkey	Indirect M	D-loop	309	1	-	30	Cho *et al*. (2014)

S = Simplex, M = Multiplex, bp = Base pair, pg = picogram, m = Minute, - = Not mentioned

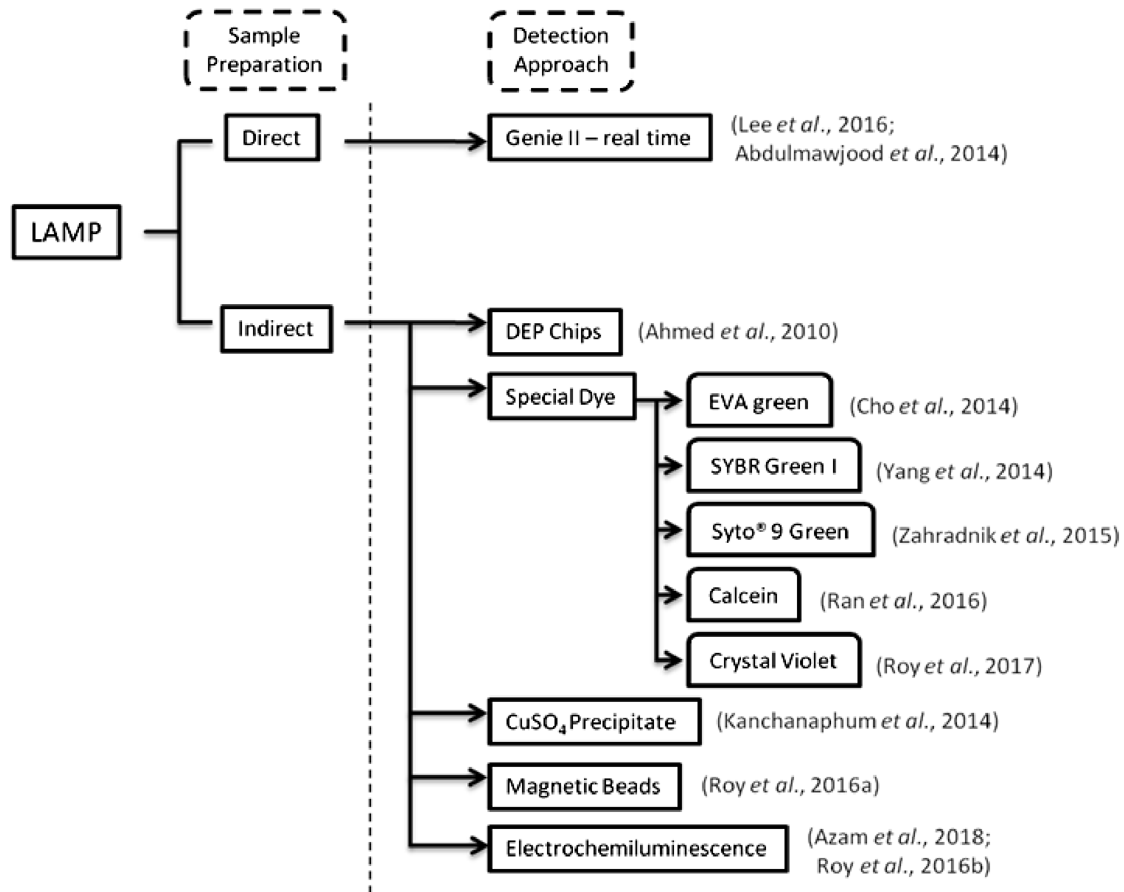

Figure 1 Summary of approaches in the LAMP assays that have been developed.

but Lee *et al*. (2016) employed a direct real-time LAMP assay which required only 30 min to amplify the porcine DNA. However, the amplification time for direct real-time LAMP in Lee *et al*. (2016) showed a slight delay as compared to that of real-time LAMP. Meanwhile, the lowest sensitivity of porcine DNA was achieved by LAMP assay developed by Ran *et al*. (2016) with a detection limit of 0.5 pg, but it took more than 60 min for the amplification. This might be due to the different buffer and target region utilised in these studies which could result in variable detection sensitivities and amplification times.

Other than porcine, Zahradnik *et al*. (2015) designed a specific LAMP primers targeting the NADH dehydrogenase gene of horse mitochondrial DNA, for detecting horse meat in processed meat products. The outcome showed a limit of detection as low as 0.1 ng of extracted horse DNA with zero cross reactions against beef, porcine and chicken. For visual detection, following the LAMP assay 1 μL×1 SYBR Green® I was added to each reaction tube. The study also tested the LAMP assay on a few food products containing various horse meat recipes (two differently spiced, hard cured sausages; meat loaf; blood pudding and knackwurst), and the results showed a reliable detection in all products except for blood pudding. The later might be associated with the fact that blood pudding consists of mainly coagulated blood with high erythrocytes content (>90%), and less nuclei or mitochondria. Hence, the extracted DNA originated from thrombocytes and leucocytes will be lower (Bowen, 1963).

Further, Abdulmawjood *et al*. (2014) successfully identified ostrich meat based on the mitochondrial DNA-encoded cytochrome b gene of the ostrich using both indirect and direct real-time LAMP assay. For the former, the LAMP assay was used in combination with a real-time fluorometer, Genie II. The system was able to detect as low as 0.01% of ostrich species in a mixture of bovine meat. At the same time, a direct swab method without nucleic acid extraction using the HYPLEX LPTV buffer was also tested. However, the amplification time was longer than that using extracted DNA. Nonetheless, the fluorescent signals from all samples were positive after approximately 12 min. In spite of the slightly longer amplification times used in the swab and HYPLEX LPTV buffer processing method, this approach still provided an accurate detection in less than 20 min using the portable equipment. This rapid processing method

also allowed the detection of ostrich meat without the need for incubation steps. This rapid lysis procedure also worked well with both minced meat and steak, giving positive results by LAMP assay without the need for DNA extraction.

Multiplex-LAMP

In order to increase the applicability of LAMP in animal species speciation, multiplex LAMP has been developed. Cho *et al*. (2014) designed a LAMP assay targeting a mitochondrial DNA which was respectively designed in combination to be specific for identifying and discriminating eight animal species simultaneously namely cattle, pig, horse, goat, sheep, chicken, duck and turkey. Animal species discrimination was possible in this process by taking advantage on the unique annealing temperature generated by the respective amplicons with no cross-reactivity observed in each set. The annealing temperature was effective for species discrimination because it differed proportionately with the nucleotide composition and amplicons size of each target sequence (Mouillesseaux *et al*., 2003). In comparison with probe-based real-time DNA amplification assay, intercalating dye-based is more cost-effective and robust to be used (Cho *et al*., 2014). The limits of detection of the LAMP assays in raw and cooked meat were 10 pg/µL to 100 fg/µL levels in 30 min reaction time. This shows the ability of multiplex LAMP assays for discriminating multiple meat species simultaneously. Therefore, while providing the simplicity, rapidity, accuracy and good sensitivity, it looks promising for this approach to be applied in the routine services for multi-meat species identification to detect potential meat source fraud and adulteration involving more than one species.

Further improvement of LAMP based approaches

To assimilate LAMP application as a point-of-care device, it has been further modified to increase the sensitivity of detection. Azam *et al*. (2018) developed a novel, simple, sensitive and rapid luminol-based electrochemiluminescence (ECL) technique for wild boar DNA detection. Luminol or 5-amino-2,3-dihydro-1,4-phthalazinedione acts as luminophore dye in this study. DNAs of LAMP products from this assay were added into luminol solution for further ECL analysis for quantification. The DNA-luminol complexes caused the diffusion of luminol towards the electrode surface to be slowed down and hindered that resulted in low luminol intensity. Hence, by employing this assay to carbon screen-printed electrode (SPE), this technique has a potential to be developed further into a compact biosensor for verifying food authenticity. This LAMP-ECL sensor could detect DNA as low as 0.1 pg/µL in ~5 min.

On the other hand, Roy *et al*. (2017) presented a paper microchip to detect and quantify utilising colour changes in presence of targeted nucleic acid. After DNA extraction and amplification of DNA using LAMP assay, LAMP products was then detected and quantified on a paper microchip fabricated in a cellulose paper and a small wax chamber utilising crystal violet (CV) dye. Crystal violet is a triphenylmethane dye containing a p-quinoid group that acts as a chromophore. It is widely being used in Gram staining for bacterial classification, and also DNA staining in agarose gels, as CV has been shown to have a high affinity for dsDNA compared to ssDNA or RNA (Norden *et al*., 1978; Miyamoto *et al*., 2015) to form a CV-dsDNA stable complex. The colour change from colourless to purple indicates a positive signal for the assay. This assay was able to detect as low as 1 pg/µL for wild boar DNA and 10 pg/µL for Bacillus subtilis DNA.

Roy *et al*. (2016b) integrated LAMP with electrochemiluminescence (ECL) technique for both DNA detection and quantification. The amplified DNA from LAMP reaction bound electrostatically with $[Ru(bpy)_3]^{+2}$ on a carbon electrode surface, which was later triggered by tripropylamine (TPrA) to yield luminescence. In this process, target DNA of wild boar meat was detected as low as 1pg/mL (3.43 $\times 10^{-1}$ copies/mL) within 60 min.

Meanwhile, Roy *et al*. (2016a), developed a sensitive and specific paper-based method for detection of chicken and porcine DNA with a detection limit of 1 pg/µL and 100 pg/µL, respectively by utilising magnetic beads. The principle lies on the formation of dark red aggregates on filter paper upon addition of magnetic bead to LAMP products in magnetic field. The detection method was also shown to be time-effective as a positive result could be obtained after 20 min of LAMP reaction.

Previously, Ahmed *et al*. (2010) reported the use of disposable electrochemical printed (DEP) chip-based detection of loop amplicons using a combination of primers targeting porcine, chicken and bovine simultaneously. Mitochondrial 12S rRNA gene was used in the system due to the characteristics of the gene that provide suitable length and ample grade of interspecies variation. The interaction between target loop amplicons and DNA binder H33258 was measured based on the changes in the anodic peak current (mA) due to oxidation of H33258 molecule using Linear Sweep Voltammetry (LSV). Limit of detection of loop amplified target DNA for porcine

was ~20.33 ng/µL, meanwhile for chicken and bovine were ~78.68 pg/µL and 23.63 pg/µL, respectively. In addition, the sensitivity of the detection in the study was further validated by comparing the results with those generated by multiplex-PCR. It was shown that the LAMP based electrochemical sensor had a higher specificity with no non-specific amplicons. Another advantage of this method was its rapidity as it took less time 65 min while multiplex based electrophoretic analysis took more than 90 min to complete.

Conclusion

Accurate species identification of meat products, detection of adulteration or fraudulent substitution and Halal authentication might gain benefits from this LAMP technique (Premanandh and Salem 2017). This method can be conducted quickly and without complex instrumentation with good sensitivity for meat species detection. The sensitivity limit can be as low as 0.1 pg/µL in 20 min, which is better than PCR that needs more than 1 h to complete. LAMP could also be incorporated with other tools such as the use of magnetic beads and special dye such as calcein and crystal violet that helps in improving detection and also reducing contamination. These make LAMP suitable for use in the field, where the application of gel electrophoresis apparatus and PCR instrument is not practical.

Acknowledgement

The authors would like to acknowledge the financial support provided by Universiti Putra Malaysia grant (GP-IPS/2018/9625800) and Halal Products Research Institute for cordial support in completing this review. Nor Asmara Tasrip wishes to acknowledge the financial support from MyBrain15 under the Ministry of Education, Malaysia for her PhD fellowship.

References

Abdulmawjood, A., Grabowski, N., Fohler, S., Kittler, S., Nagengast, H. and Klein, G. 2014. Development of Loop-Mediated Isothermal Amplification (LAMP) assay for rapid and sensitive identification of ostrich meat. PLoS One 9(6): e100717.

Ahmed, M. U., Saito, M., Hossain, M. M., Rao, S. R., Furui, S., Hino, A., Takamura, Y., Takagi, M. and Tamiya, E. 2009. Electrochemical genosensor for the rapid detection of GMO using loop-mediated isothermal amplification. Analyst 134(5): 966-72.

Ahmed, M. U., Hasan, Q., Hossain, M. M., Saito, M. and Tamiya, E. 2010. Meat species identification based on the loop mediated isothermal amplification and electrochemical DNA sensor. Food Control 21(5): 599-605.

Ali, M. E., Hashim, U., Dhahi, T. S., Mustafa, S., Man, Y. B. C. and Latif, M. A. 2012. Analysis of pork adulteration in commercial burgers targeting porcine-specific mitochondrial cytochrome b gene by taqman probe real-time polymerase chain reaction. Food Analytical Methods 5(4): 784-794.

Amin, A., Bee, S., Hamid, A. and Ali, E. 2015. A method for the detection of potential fraud of bringing feline meat in food chain. International Journal of Food Properties 2912(December): 1532–2386.

Asensio, L., Gonzalez, I., Pavon, M. A. and Garcia, T. R. M. 2008. An indirect ELISA and a PCR technique for the detection of grouper (Epinephelus marginatus) mislabeling. Food Additives and Contaminants 6: 677-683.

Ayaz, Y., Ayaz, N. D. and Erol, I. 2006. Detection of species in meat and meat products using enzyme-linked immunosorbent assay. Journal of Muscle Foods 17: 214-220.

Azam, N. F. N., Roy, S., Lim, S. A. and Ahmed, M. U. 2018. Meat species identification using DNA-luminol interaction and their slow diffusion onto the biochip surface. Food Chemistry 248: 29-36.

Bowen, H. J. M. 1963. The Elementary Composition of Mammalian Blood. United Kingdom: United Kingdom Atomic Energy Authority.

Calvo, J. H., Zaragoza, P. and Osta, R. 2001. Random amplified polymorphic DNA fingerprints for identification of species in poultry pâté. Poultry Science 80(4): 522-524.

Cho, A., Dong, H. and Cho, S. 2014. Meat species identification using loop-mediated isothermal amplification assay targeting species-specific mitochondrial DNA. Korean Journal for Food Science of Animal Resources 34(6): 799-807.

Cho, E. J., Yang, L., Levy, M. and Ellington, A. D. 2005. Using a deoxyribozyme ligase and rolling circle amplification to detect a non-nucleic acid analyte, ATP. Journal of the American Chemical Society 23: 2022-2023.

Dhama, K., Karthik, K., Chakraborty, S., Tiwari, R., Kapoor, S., Kumar, A. and Thomas, P. 2014. Loop-mediated isothermal amplification of DNA (LAMP): a new diagnostic tool lights the world of diagnosis of animal and human pathogens: A review. Pakistan Journal of Biological Sciences 17(2): 151-166.

Doosti, A. and Dehkordi, P. G. 2014. Molecular assay to fraud identification of meat products. Journal of Food Science and Technology 51(January): 148-152.

Enomoto, Y., Yoshikawa, T., Ihira, M., Akimoto, S., Miyake, F., Usui, C., Suga, S., ... and Asano, Y. 2005. Rapid diagnosis of herpes simplex virus infection by a loop-mediated isothermal amplification method. Journal of Clinical Microbiology 43(2): 951-955.

Fajardo, V., Gonza, I. and Garcı, T. 2010. A review of current methodologies for the authentication of meats from game animal species. Trends in Food Science and Technology 21: 408-421.

Fajardo, V., González, I., Martín, I., Rojas, M., Hernández, P. E., García, T. and Martín, R. 2008. Real-time PCR for detection and quantification of red deer (Cervus elaphus), fallow deer (Dama dama), and roe deer (Capreolus capreolus) in meat mixtures. Meat Science 79(2): 289-298.

Fernandez-Soto, P., Arahuetes, J. G., Hernandez, A. S., Aban, J. L., Santiago, B. V. and Muro, A. 2014. A Loop-Mediated Isothermal Amplification (LAMP) assay for early detection of Schistosoma mansoni in stool samples : a diagnostic approach in a murine model. PLOS Neglected Tropical Diseases 8(9): e3126.

Gamble, H. R. 1997. Parasites associated with pork and pork products. Scientific and Technical Review of the Office International des Epizooties 16(2): 496-506.

Gill, P. and Ghaemi, A. 2008. Nucleic acid isothermal amplification technologies: A review. Nucleosides, Nucleotides and Nucleic Acids 27(3): 224-243.

Huang, X., Chen, L., Xu, J., Ji, H.-F., Zhu, S. and Chen, H. 2014. Rapid visual detection of phytase gene in genetically modified maize using loop-mediated isothermal amplification method. Food Chemistry 156 (Supplement C): 184-189.

Kanchanaphum, P., Maneenin, S. and Chaiyana, W. 2014. Analysis of pork meat using loop mediated isothermal amplification (LAMP) to confirm halal status. International Journal of Biosciences 4(9): 62-68.

Kaneko, H., Iida, T., Aoki, K., Ohno, S. and Suzutani, T. 2005. Sensitive and rapid detection of herpes simplex virus and varicella-zoster virus DNA by loop-mediated isothermal amplification. International Journal of Biosciences 43(7):3290-3296.

Kaneko, H., Kawana, T., Fukushima, E. and Suzutani, T. 2007. Tolerance of loop-mediated isothermal amplification to a culture medium and biological substances. Journal of Biochemical and Biophysical Methods 70: 499-501.

Karami, A., Gill, P., Motamedi, M. and Saghafinia, M. 2011. A review of the current isothermal amplification techniques: applications, advantages and disadvantages. Journal of Global Infectious Diseases 3(3): 293-302.

Kiddle, G., Hardinge, P., Buttigieg, N., Gandelman, O., Pereira, C., Mcelgunn, C.J., Rizzoli, M., … and Murray, J.A.H. 2012. GMO detection using a bioluminescent real time reporter (BART) of loop mediated isothermal amplification (LAMP) suitable for field use. BMC Biotechnology 12(15): 1-13.

Kim, K. B. W. R., Song, E. J., Lee, S. Y., Park, J. G., Lee, J. W., Byun, M. W. and Ahn, D. H. 2011. Changes in antigenicity of porcine serum albumin in gamma-irradiated sausage extract by treatment with pepsin and trypsin. Radiation Physics and Chemistry 80(11): 1258-1262.

Lee, S., Kim, M., Hong, Y. and Kim, H. 2016. Development of a rapid on-site detection method for pork in processed meat products using real-time loop-mediated isothermal amplification. Food Control 66: 53-61.

Lizardi, P. M., Huang, X., Zhu, Z., Bray-Ward, P., Thomas, D. C. and Ward, D. C. 1998. Mutation detection and single-molecule counting using isothermal rolling-circle amplification. Nature Genetics 19: 225-232.

Mahony, J., Chong, S., Bulir, D., Ruyter, A., and Mwawasi, K. 2013. Development of a sensitive loop-mediated isothermal amplification assay that provides specimen-to-result diagnosis of respiratory syncytial virus infection in 30 minutes. Journal of Clinical Microbiology 51(8): 2696-2701.

Miyamoto, S., Sano, S., Takahashi, K. and Jikihara, T. 2015. Method for colorimetric detection of double-stranded nucleic acid using leuco triphenylmethane dyes. Analytical Biochemistry 15(473): 28-33.

Mori, Y., Nagamine, K., Tomita, N. and Notomi, T. 2001. Detection of loop-mediated isothermal amplification reaction by turbidity derived from magnesium pyrophosphate formation. Biochemical and Biophysical Research Communications 289(1): 150-154.

Mouillesseaux, K. P., Klimpel, K. R. and Dhar, A. K. 2003. Improvement in the specificity and sensitivity of detection for the Taura syndrome virus and yellow head virus of penaeid shrimp by increasing the amplicon size in SYBR Green real-time RT-PCR. Journal of Virological Methods 111(2): 121-127.

Mugasa, C. M., Laurent, T., Schoone, G. J., Kager, P. A., Lubega, G. W. and Schallig, H. D. F. H. 2009. Nucleic acid sequence-based amplification with oligochromatography for detection of Trypanosoma brucei in clinical samples. Journal of Clinical Microbiology 47(3): 630-635.

Nagamine, K., Hase, T. and Notomi, T. 2002. Accelerated reaction by loop-mediated isothermal amplification using loop primers. Molecular and Cellular Probes 16 (3): 223-229.

Nakyinsige, K., Bin, Y., Man, C. and Qurni, A. 2012. Halal authenticity issues in meat and meat products. Meat Science 91(3): 207-214.

Nordén, B., Tjerneld, F. and Palm, E. 1978. Linear dichroism studies of binding site structures in solution. Complexes between DNA and basic arylmethane dyes. Biophysical Chemistry 8(1): 1-15.

Notomi, T., Okayama, H., Masubuchi, H., Yonekawa, T., Watanabe, K., Amino, N. and Hase, T. 2000. Loop-mediated isothermal amplification of DNA. Nucleic Acids Research 28(12): E63.

O'Mahony, P. J. 2013. Finding horse meat in beef products-a global problem. Quarterly Journal of Medicine 106(6): 595-597.

Özpinar, H., Tezmen, G., Gökçe, İ. and Tekiner, İ. H. 2013. Detection of animal species in some meat and meat products by comparatively using DNA microarray and real time PCR methods detection of animal species in some meat and meat products by comparatively using DNA microarray and real time PCR methods. Kafkas

Üniversitesi Veteriner Fakültesi Dergisi 19(February): 245-252.

Pardo, M. A. and Pérez-Villareal, B. 2004. Identification of commercial canned tuna species by restriction site analysis of mitochondrial DNA products obtained by nested primer PCR. Food Chemistry 86(1): 143-150.

Parida, M., Sannarangaiah, S., Dash, P. K., Rao, P. V. L. and Morita, K. 2008. Loop mediated isothermal amplification (LAMP): a new generation of innovative gene amplification technique ; perspectives in clinical diagnosis of infectious diseases. Reviews in Medical Virology 18(August): 407-421.

Peter, S. 2010. Food fraud – a current issue but an old problem. British Food Journal 112(2): 198-213.

Premanandh, J. and Bin, S. S. 2017. Progress and challenges associated with halal authentication of consumer packaged goods. Journal of the Science of Food and Agriculture 97(14): 4672-4678.

Rahman, M., Ali, E., Bee, S., Hamid, A., Mustafa, S., Hashim, U. and Kalthum, U. 2014. Polymerase chain reaction assay targeting cytochrome b gene for the detection of dog meat adulteration in meatball formulation. Meat Science 97(4): 404-409.

Ran, G., Ren, L., Han, X., Liu, X. and Li, Z. 2016. Development of a rapid method for the visible detection of pork DNA in halal products by loop-mediated isothermal amplification. Food Analytical Methods 9: 565-570.

Rikimaru, K. and Takahashi, H. 2007. A method for discriminating a japanese brand of chicken, the hinai-jidori, using microsatellite markers. Poultry Science 86(9): 1881-1886.

Roy, S., Mohd-Naim, N. F., Safavieh, M. and Ahmed, M. U. 2017. Colorimetric nucleic acid detection on paper microchip using loop mediated isothermal amplification and crystal violet dye. ACS Sensors 2(11): 1713-1720.

Roy, S., Rahman, I. A. and Ahmed, M. U. 2016a. Paper-based rapid detection of pork and chicken using LAMP – magnetic bead aggregates. Analytical Methods 8: 2391-2399.

Roy, S., Xiao, S., Liew, J., Ying, Z., Safavieh, M. and Ahmed, M. U. 2016b. A novel, sensitive and label-free loop-mediated isothermal amplification detection method for nucleic acids using luminophore dyes. Biosensors and Bioelectronic 86: 346-352.

Safavieh, M., Ahmed, M. U., Sokolullu, E., Ng, A., Braescuac, L. and Zourob, M. 2014a. A simple cassette as point-of-care diagnostic device for naked-eye colorimetric bacteria detection. Analyst 139(2): 482-487.

Safavieh, M., Ahmed, M. U., Ng, A. and Zourob, M. 2014b. High-throughput real-time electrochemical monitoring of LAMP for pathogenic bacteria detection. Biosensors and Bioelectronics Aug 15(58): 101-106.

Sasazaki, S., Itoh, K., Arimitsu, S., Imada, T., Takasuga, A., Nagaishi, H., Takano, S., Mannen, S. and Tsuji, S. 2004. Development of breed identification markers derived from AFLP in beef cattle. Meat Science 67(2): 275-280.

Sirius Global Animal Charitable Trust. 2007. International ban on dog meat products [report submitted to the World Health Organization]. Retrieved from website: http://www.siriusgao.org/submission/Submission%20-%20International%20Ban%20on%20Dog%20Meat%20Products%204.pdf

Soares, S., Amaral, J. S., Mafra, I. and Oliveira, M. B. P. P. 2010. Quantitative detection of poultry meat adulteration with pork by a duplex PCR assay. Meat Science 85(3): 531-536.

Soares, S., Amaral, J. S., Oliveira, M. B. P. P. and Mafra, I. 2013. A SYBR Green real-time PCR assay to detect and quantify pork meat in processed poultry meat products. Meat Science 94(1): 115-120.

Tajima, K., Enishi, O., Amari, M., Mitsumori, M., Kajikawa, H., Kurihara, M., Yanai, S. … and Matsumoto, M. 2002. PCR detection of DNAs of animal origin in feed by primers based on sequences of short and long interspersed repetitive elements. Bioscience, Biotechnology, and Biochemistry 66(10): 2247-2250.

Tlili, C., Sokullu, E., Safavieh, M., Tolba, M., Ahmed, M. U. and Zourob, M. 2013. Bacteria screening, viability, and confirmation assays using Bacteriophage-Impedimetric/Loop-Mediated Isothermal Amplification Dual-Response Biosensors. Analytical Chemistry 85(10): 4893-4901.

Tröger, V., Niemann, K., Gärtig, C. and Kuhlmeier, D. 2015. Isothermal amplification and quantification of nucleic acids and its use in microsystems. Journal of Nanomedicine and Nanotechnology 6(3): article number 1000282.

Walker, G. T., Little, M. C., Nadeau, J. G. and Shank, D. D. 1992. Isothermal in vitro amplification of DNA by a restriction enzyme / DNA polymerase system. Proceedings of the National Academy of Sciences of the United States of America 89(January): 392-396.

Wang, C., Li, R., Quan, S., Shen, P., Zhang, D. and Shi, J. 2015. GMO detection in food and feed through screening by visual loop-mediated isothermal amplification assays. Analytical and Bioanalytical Chemistry 407: 4829-4834.

Wolf, C. and Lüthy, J. 2001. Quantitative competitive (QC) PCR for quantification of porcine DNA. Meat Science 57(2): 161-168.

Yang, L., Fu, S., Peng, X., Li, L., Song, T. and Li, L. 2014. Identification of pork in meat products using real- time loop-mediated isothermal amplification. Biotechnology and Biotechnological Equipment 28(May): 882-888.

Zahradnik, C., Martzy, R., Mach, R. L., Krska, R., Farnleitner, A. H. and Brunner, K. 2015. Loop-mediated isothermal amplification (LAMP) for the detection of horse meat in meat and processed meat products. Food Analytical Methods 8: 1576-1581.

Zeng, J., Wei, H., Zhang, L., Liu, X., Zhang, H., Cheng, J., Ma, D., … and Liu, L. 2014. Rapid detection of Vibrio parahaemolyticus in raw oysters using immunomagnetic separation combined with loop-

mediated isothermal amplification. International Journal of Food Microbiology 174(Supplement C): 123-128.

Zhang, M., Liu, Y., Chen, L., Quan, S., Jiang, S., Zhang, D. and Yang, L. 2013. One simple DNA extraction device and its combination with modified visual loop-mediated isothermal amplification for rapid on-field detection of genetically modified organisms. Analytical Chemistry 85(1): 75-82.