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Detection of melamine by voltammetry at a carbon electrode decorated with metal nanoparticles and assessment in commercial sample

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Abstract
In this work, the voltammetric behavior of melamine was investigated on a nanoparticles modified electrode. The Fe_2O_3 nanoperticles were prepared and characterized. The melamine showed well defined anodic peak current on the modified electrode. The electro-oxidation was studied in phosphate buffer solution of pH 6.0. The effect of scan rate, volume of suspension and pH were studied for the title reaction. Further, linear sweep voltammetry was used to develop a method to detect melamine. Under optimized conditions, the peak current was linear with the concentration in the range 50 to 160 µM. The limit of detection of 10.5 µM and limit of quantification of 35.5 µM was obtained. Using this optimized method, melamine was detected in commercial milk powder successfully.

Keywords: Melamine, voltammetry, modified electrode, nanoparticles, analysis

Introduction

Adulteration of food is of global concern and especially people in the developing countries have been affected severely because of lack of monitoring. Milk adulterants can affect health hazards severely which may lead to deadly diseases. Milk has been considered as a complete food, because it contains nutrients in abundance which is needed for both infants and adults. There are some major compounds which are used to adulter the milk are urea, formalin, detergents, benzoic acid, caustic soda, hydrogen peroxide, sugars, salicylic acid and melamine. Some of these adulterants can cause serious adverse health issues, sometime when used for a long time. To increase the protein content in milk wrongly, melamine is being added. If the added melamine level is well above the limit set by the policies, can cause renal failure and death in infants. Melamine is a little poisonous by its nature. But when it combines with cyanuric acid, it yields a compound called melamine cyanurate. This compound so formed may become basis for formation of kidney stones, damage of tissue, bladder cancer, and finally to death [1, 2].

Therefore, a simpler, solvent free, robust, quick, reliable, sensitive and greener method is very much essential for the assessment of melamine. The electrochemical methods are very simple, sensitive, reliable, solvent free and selective methods which have the capacity of giving results in real time. These electrochemical, in particular, voltammetric methods once developed for the assessment, that can be used to create awareness among local public and also will motivate them not to use adultered milk products. It may also motivate the manufacturer of milk products, not to add this lethal melamine to the dairy products.

Various methods of analysis of melamine were available. The classical methods like infrared spectroscopy [3], Raman spectroscopy [4], gas chromatography-mass spectrometry [5], mass spectroscopy [6] and HPLC [7] were developed to detect melamine. But these methods have some limitations like expensive equipment, extraction/pretreatments are time consuming and usage of organic solvents, out of which some solvents are carcinogenic. The electrochemical methods have many advantages like, ultra sensitivity, simple procedures, fast response and reliability. There are many reports on the detection of melamine using electrochemical methods. A molecuraly imprinted sensors were developed and used successfully for the detection of melamine [8]. There are few attempts to develop sensor based on molecularly imprinted polymer based electrodes [9, 15]. Bakas *et al* detected melamine in picomolar range using UV-graft polymerization [9].

Ji *et al* modified pre-polymerized electrode with graphene to get better sensitivity. Regas *et al* constructed a conducting polymer based imprinted electrode so that enhanced conductivity will increase the sensitivity of the method [12]. There was an attempt to decorate the imprinted electrode with carbon nanotubes to achieve better sensitivity [13]. A molecular receptor based method was developed so that melamine can react with the receptor, which in turn becomes a very specific detection of analyte [15]. Some sensing was done using a recognition element 3,4-dihydroxyphenyl acetic acid [16]. Carbon based electrodes were modified with various carbon based nanoparticles for the assessment of melamine. The reduced graphene oxide was used as modifier along with copper nano flowers [17], and with gold nano particles [18], respectively for the sensing. Peng at al developed a gold nanoparticles modified carbon paste electrode, since these carbon paste electrodes are very easy to modify and renew [19]. DNA based sensor platforms were also developed [20, 21]. Li *et al* constructed a DNA based biosensor for real time detection of melamine with superior sensitivity and selectivity [20]. An electrochemical biosensor was constructed for the analysis of melamine together with urea [22].

Indian Institute of Science researchers have developed a device to detect melamine in adultered milk samples based

on its reaction with silver nano particles. But this is purely qualitative and not possible to quantitate the melamine amount. There were few colorimetric methods [23] where gold nanoparticles got aggregated so that the colour of nanoparticles changes and this was measured by a colorimeter. In another report, authors have used [24] different sized gold nanoparticles with good sensitivity. One more method reported uses unmodified silver nanoparticles [25]. Melamine was detected in raw milk sample using biofunctionalized silver nanoparticles [26].

There were some reports using reverse phase Liquid Chromatography to determine melamine [27]. There were very few reports on detection of melamine using electrochemical methods. There was an attempt from BARC researchers on a carbon paste electrode modified with grapheme oxide-gold nanoparticles [28]. Another report was from SASTRA University, where they used Platinum/ZnO modified electrode for the assessment of melamine [29]. There was report recently [30] which uses printed circuit board electrode for the detection of melamine.

Materials and Methods

Materials: Melamine, Methanol, Sodium dihydrogen phosphate, Disodium hydrogen phosphate, acetonitrile and all other chemicals were received from Rankem and all are of analytical grade. Deionised water was used to prepare phosphate buffer solution (PBS). The pH of PBS was varied using different volumes of stock solution of monobasic and dibasic sodium phosphate. Stock solution of melamine was made in methanol. Amul milk was used to analyse melamine which was obtained from a local medical store. Small portion was weighed and dissolved in methanol to prepare stock solution. This stock solution was diluted with PBS to get a solution with adequate concentration for analysis.

Instrumentation: Electrochemical measurements were carried out on a CH Instruments, CHI660E Electrochemical Analyser (USA) coupled with a conventional three-electrode cell. A three-electrode cell was used with a Ag/AgCl as reference electrode, a Pt wire as counter electrode and a bare glassy carbon electrode with a diameter of 2 mm (modified and unmodified) were used as working electrodes, respectively. All the potentials in this paper are given against the Ag/AgCl (3 M KCl). Solution pH was measured with an Elico LI120 pH meter (Elico Ltd). Scanning electron microscope (SEM XL 30 ESEM with EDAX) was used to characterize the Fe_2O_3 nanoparticles.

Preparation and Characterization of Nanoparticles: The Fe_2O_3 nanoparticles were prepared as described in literature. $FeCl_3$ and $FeCl_2.4H_2O$ were dissolved in a 2 N hydrochloric acid to form a solution with the concentration of 1 M for $FeCl_3$ and 2 M for $FeCl_2.4H_2O$. The $NH_3.H_2O$ solution (2 M) was dropped to this solution with vigorous stirring at room temperature for 2 hours. The final pH was 9.70. The brown precipitate was then collected by filtration and rinsed three times with deionized water and ethanol. Finally, the washed precipitate was dried at 80 °C overnight. The SEM image of nanoparticles obtained after calcinations was recorded and is as shown in below Figure 1. The Figure 2 shows the FT-infrared (FTIR) spectrum of the sample, the absorption peak at 460, 536 and 3419 cm^{-1} identified Vibration of Fe–O.

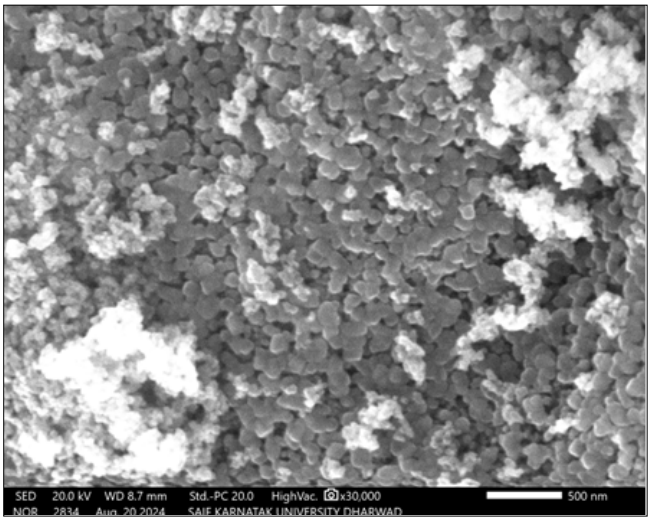

Fig 1: SEM image of Fe_2O_3 nanoparticles

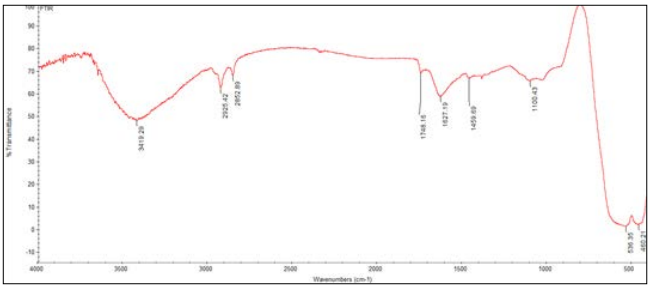

Fig 2: FTIR spectrum of Fe_2O_3 nanoparticles

Method of Analysis: The Fe_2O_3 nanoparticles suspension was prepared by dispersing 2 mg of nanoparticle in 10 mL acetonitrile separately using ultrasonic agitation to obtain a relative stable suspension. The GCE was carefully polished with 0.30 and 0.05 α-alumina slurry on a polishing cloth, and then washed in an ultrasonic bath of methanol and water, respectively. The cleaned GCE was initially coated by casting 2 μL of the black suspension of Fe_2O_3 at a time and dried in air.

The modified GCE was first activated in PBS (pH 6.0) by cyclic voltammetric sweeps between -0.1 and 0.7 V until stable cyclic voltammograms were obtained. Then electrodes were transferred into another cell of PBS (pH 6.0) containing proper amount of melamine. After accumulating for 150 s at open circuit under stirring and following quiet for 20 s, potential scan was initiated and cyclic voltammograms were recorded between -0.1 and 0.7 V, with a scan rate of 50 mV s^{-1}. All measurements were carried out at room temperature and without removing dissolved oxygen.

Results and Discussion

Voltammetric Response of Melamine: The cyclic voltammograms of melamine at a bare GCE and at modified GCE were shown in Figure 3. It can be seen that the melamine oxidation peak at the bare GCE was weak and broad due to slow electron transfer, while the response was considerably improved at the modified GCE. At the bare GCE, the peak was at about 0.5 V (Figure 3, curve *a*), but

on the modified GCE, the peak appeared at about 0.28 V (Figure 3, curve b), This was attributed to the electro catalytic effect caused by Fe_2O_3 nanoparticles. The reason for the better performance of the modified GCE may be due to the nanometer dimensions of the nanoparticles. Meanwhile the nanoparticles increase the effective area of the electrode. The modified electrode has no electrochemical activity in phosphate buffer solution.

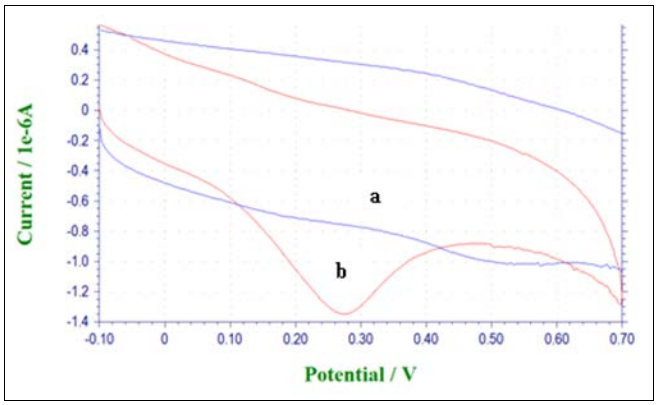

Fig 3: Cyclic voltammograms of melamine at (a) bare GCE and (b) at modified GCE

Optimization of Parameters: The influence of the amount of nanoparticles on the peak current was studied. At 6 µL of Fe_2O_3 suspension, the peak current was highest. After that amount, it decreases. This is related to the thickness of the film. If the film was too thin, the melamine amount adsorbed was small, resulting in the small peak current. When it was too thick, the film conductivity reduced and the film became not as stable as nanoparticles film could leave off the electrode surface. Thus it blocks the electrode surface and hence the peak current decreases. Therefore, 6 µL of Fe_2O_3 suspension solution was used in the remaining studies.

The electrode reaction might be affected by the buffer solution and pH of the medium. The effect of pH on the current response was investigated by CV in PBS a shown in Figure 4. Within the range of pH 1.0 – 7.0, the peak potential shifted considerably as pH of solution increased and highest peak current was observed for pH 6.0. Therefore the pH of 6.0 was used for further studies.

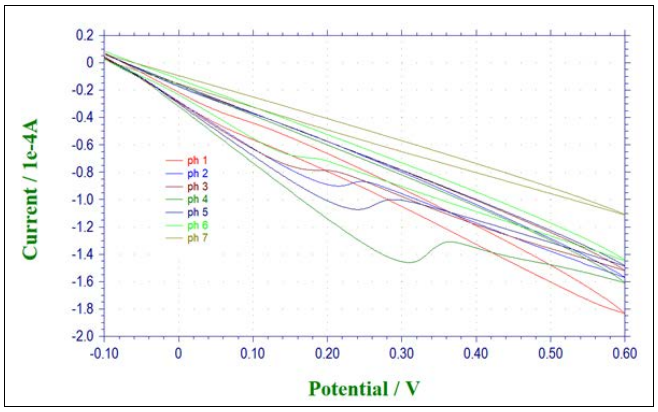

Fig 4: Influence of pH on electro-oxidation of melamine

Useful information involving electrochemical mechanism usually can be acquired from the relationship between peak current and scan rate. Therefore, the voltammetric behavior of melamine at different scan rates from 10 to 100 mV s^{-1} was studied (Figure 5.). There is a good linear relationship between peak current and square root of scan rate. The equation representing this was $Ip = 11.04\ (v)^{1/2} - 1.132$; $R^2 = 0.994$. This indicates that the electrode process was controlled by diffusion rather than adsorption. In addition, there was a linear relation between log*I*p and log*v*, corresponding to the following equation: log$Ip = 0.513\log v + 0.939$; $R^2 = 0.987$, which has the slope values of 0.513, which is very close to the theoretical value of 0.5 for diffusion controlled process. The peak potential not altered values with increasing the scan rates.

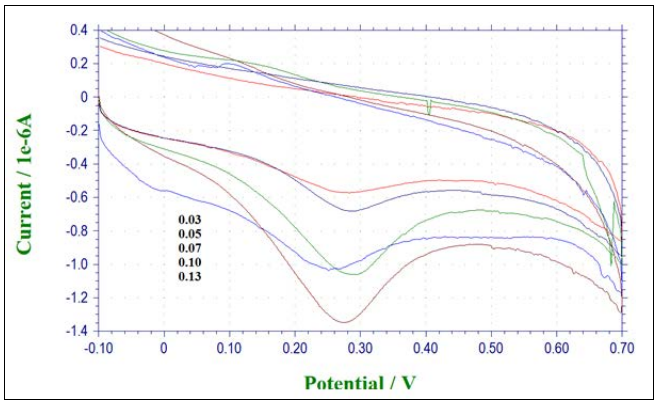

Fig 5: Effect of scan rate on the electro-oxidation of melamine in PBS of pH 6.0.

Application

In order to develop a voltammetric method for determining the analyte, linear sweep voltammetry was used. According to the obtained results, it was possible to apply this technique to the quantitative analysis of melamine. The PBS solution of pH 6.0 was selected as the supporting electrolyte for the quantification. Linear sweep voltammograms obtained with increasing amounts of melamine showed that the peak current increased linearly with increasing concentration, as shown in Fig. 6. Using the optimum conditions described above, linear calibration curves were obtained for melamine in the range of 50 to 160 µM (Figure 7). The linear equation was *I*p (µA) = 0.231 + 0.018*C* (R^2 = 0.998*C* is in µM). Deviation from linearity was observed for more concentrated solutions, due to the adsorption of melamine or its oxidation product on the electrode surface. Related statistical data of the calibration curves were obtained from five different calibration curves. The limit of detection (LOD) and quantification (LOQ) were 10.5 and 35.5 µM, respectively.

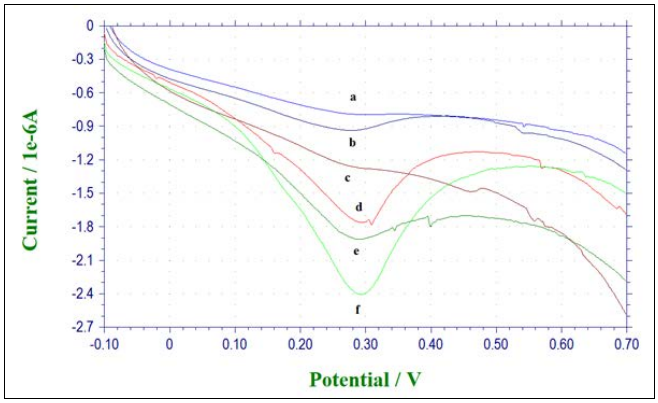

Fig 6: Variation of concentration of melamine at pH 6.0 in PBS solution

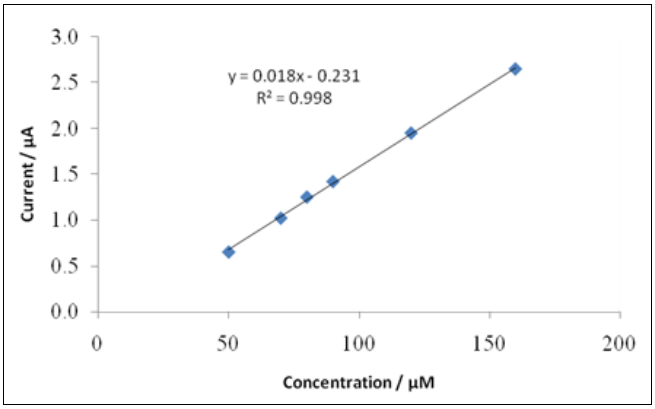

Fig 7: Calibration plot

Analysis in Commercial Sample: In order to evaluate the applicability of the proposed method in the commercial sample analysis, milk sample containing melamine was studied. The results showed that there was some amount of melamine present. The standard addition method was used to detect melamine because of non-availability of the standard reference method. Under the optimized conditions, the current at about 0.30 V was measured at various addition of known amount of melamine and results were given in below Table 1.

Table 1: Analysis data for melamine detection in commercial sample

Sample	Spiked (µM)	Found (µM)	Recovery (%)	RSD (%)
Commercial milk	0	----	----	----
	70	69.23 ± 0.13	98.90	3.52
	110	120.56 ± 0.87	100.47	4.63
	140	139.17 ± 0.42	99.41	2.76

Conclusion

In the present work, a Fe_2O_3 nanoparticles modified glassy carbon electrode has been successfully developed for voltammetric oxidation and assessment of melamine in PBS. The modified electrode showed electrocatalytic action for the oxidation of melamine, characterizing by the enhancement of the peak current, which was probably due to the larger surface area of nanoparticles. The peak at about 0.28 V was suitable for analysis and the peak current was linear to melamine concentrations over a certain range under the selected conditions. This sensor can be used for voltammetric determination of melamine with more sensitivity and good reproducibility. The modified electrode has been used to determine melamine in commercial samples. The proposed method offered the advantages of accuracy and time saving as well as simplicity of reagents and apparatus. This work adds a new modified electrode to voltammetric field.

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Conflicts of interest- The author declare that they have no conflict of interest.

References

1. Langman CB, Alon U, Ingelfinger J. A position statement on kidney disease from powdered infant formula-based melamine exposure in Chinese infants. Pediatric Nephrology,2009:24:1263-1266. https://doi.org/10.1007/s00467-009-1129-6.
2. Sun P, Wang J, Shen J, Wei H. Residues of melamine and cyanuric acid in milk and tissues of dairy cows fed different doses of melamine. Journal of Dairy Science,2011:94:3575-3582. https://doi.org/10.3168/jds.2010-4018.
3. Mauer LJ, Chernyshova AA, Hiatt A, Deering A, Davis R. Melamine Detection in Infant Formula Powder Using Near- and Mid-Infrared Spectroscopy. Journal of Agricultural and Food Chemistry,2009:57:3974-3980. https://doi.org/10.1021/jf900587m.
4. Mecker LC, Tyner KM, Kauffman JF, Arzhantsev S, Mans DJ, Gryniewicz Ruzicka CM. Selective melamine detection in multiple sample matrices with a portable Raman instrument using surface enhanced Raman spectroscopy-active gold nanoparticles. Analytica Chimica Acta,2012:733:48-55. https://doi.org/10.1016/j.aca.2012.05.001.
5. Xu XM, Ren YP, Zhu Y. Direct determination of melamine in dairy products by gas chromatography/mass spectrometry with coupled column separation. Analytica Chimica Acta,2009:650:39-43. https://doi.org/10.1016/j.aca.2009.04.026.
6. Yang SP, Ding JH, Zheng J. Detection of Melamine in Milk Products by Surface Desorption Atmospheric Pressure Chemical Ionization Mass Spectrometry. Analytical Chemistry,2009:81:2426-2436. https://doi.org/10.1021/ac900063u.
7. Lin ML, Awika J, Yang L. Detection of Melamine in Gluten, Chicken Feed, and Processed Foods Using Surface Enhanced Raman Spectroscopy and HPLC. Journal of Food Science,2008:73:129-134. https://doi.org/10.1111/j.1750-3841.2008.00901.x.
8. An J, Li L, Ding Y. A novel molecularly imprinted electrochemical sensor based on Prussian blue analogue generated by iron metal organic frameworks for highly sensitive detection of melamine. Electrochimica Acta,2019:326:134946. https://doi.org/10.1016/j.electacta.2019.134946.
9. Bakas I, Salmi Z, Jouini M. Picomolar Detection of Melamine Using Molecularly Imprinted Polymer-Based Electrochemical Sensors Prepared by UV-Graft Photopolymerization. Electroanalysis,2015:27:429-439. https://doi.org/10.1002/elan.201400382.
10. Ji Z, Chen W, Wang E, Deng R. Electropolymerized Molecular Imprinting & Graphene Modified Electrode for Detection of Melamine. International Journal of Electrochemical Science,2017:12:11942-11954. https://doi.org/10.20964/2017.12.34.
11. Li J, Chen Z, Li Y. A strategy for constructing sensitive and renewable molecularly imprinted electrochemical sensors for melamine detection. Analytica Chimica Acta,2011:706:255-260. https://doi.org/10.1016/j.aca.2011.08.048.
12. Regasa MB, Soreta TR, Femi OE, Ramamurthy PC. Development of Molecularly Imprinted Conducting Polymer Composite Film-Based Electrochemical Sensor for Melamine Detection in Infant Formula. ACS

Omega,2020:5:4090-4099. https://doi.org/10.1021/acsomega.9b03747.
13. Xu S, Lin G, Zhao W. Necklace-like Molecularly Imprinted Nanohybrids Based on Polymeric Nanoparticles Decorated Multiwalled Carbon Nanotubes for Highly Sensitive and Selective Melamine Detection. ACS Applied Materials and Interfaces,2018:10:24850-24859. https://doi.org/10.1021/acsami.8b08558.
14. Heydarian-Dehkordi N, Saei-Dehkordi SS, Izadi Z, Ghasemi-Varnamkhasti M. Development of an ultrasensitive molecularly imprinted poly-(ortho-phenylenediamine) based sensor for the determination of melamine adulteration in milk and infant formula. Food Science and Nutrition,2022:10:3154-3164. https://doi.org/10.1002/fsn3.2914.
15. Regasa MB, Soreta TR, Femi OE, Ramamurthy PC, Kumar S. Molecularly imprinted polyaniline molecular receptor–based chemical sensor for the electrochemical determination of melamine. Journal of Molecular Recognition,2020:33:e2836. https://doi.org/10.1002/jmr.2836.
16. Cao Q, Zhao H, He Y, Ding N, Wang J. Electrochemical sensing of melamine with 3,4-dihydroxyphenylacetic acid as recognition element. Analytica Chimica Acta,2010:675:24-28. https://doi.org/10.1016/j.aca.2010.07.002.
17. Daizy M, Tarafder C, Al-Mamun MR, Liu X. Electrochemical Detection of Melamine by Using Reduced Graphene Oxide–Copper Nanoflowers Modified Glassy Carbon Electrode. ACS Omega,2019:4:20324-20329. https://doi.org/10.1021/acsomega.9b02827.
18. Ren QX, Shen XY, Sun YY, Fan RH, Zhang J. A highly sensitive competitive immunosensor based on branched polyethyleneimine functionalized reduced graphene oxide and gold nanoparticles modified electrode for detection of melamine. Food Chemistry,2020:304:125397. https://doi.org/10.1016/j.foodchem.2019.125397.
19. Peng J, Feng Y, Han XX, Gao ZN. Sensitive electrochemical detection of melamine based on gold nanoparticles deposited on a graphene doped carbon paste electrode. Analytical Methods,2016:8:2526-2532. https://doi.org/10.1039/C6AY00287K.
20. Li H, Somerson J, Xia F, Plaxco KW. Electrochemical DNA-Based Sensors for Molecular Quality Control: Continuous, Real-Time Melamine Detection in Flowing Whole Milk. Analytical Chemistry,2018:90:10641-10645. https://doi.org/10.1021/acs.analchem.8b01993.
21. Liu XJ, Song MM, Li F. Triplex DNA-based Bioanalytical Platform for Highly Sensitive Homogeneous Electrochemical Detection of Melamine. Scientific Reports,2017:7:4490. https://doi.org/10.1038/s41598-017-04812-4.
22. Ezhilan M, Gumpu MB, Ramachandra BL. Design and development of electrochemical biosensor for the simultaneous detection of melamine and urea in adulterated milk samples. Sensors & Actuators, B: Chemical,2017:238:1283-1292. https://doi.org/10.1016/j.snb.2016.09.100.
23. Kumar N, Seth R, Kumar H. Colorimetric detection of melamine in milk by citrate-stabilized gold nanoparticles. Analytical Biochemistry,2014:456:43-49. https://doi.org/10.1016/j.ab.2014.04.002.
24. Paul IE, Rajeshwari A, Prathna TC, Raichur AM, Chandrasekaran N, Mukherjee A. Colorimetric detection of melamine based on the size effect of AuNPs. Analytical Methods,2015:7:1453-1462. https://doi.org/10.1039/C4AY02622E.
25. Kumar N, Kumar H, Mann B, Seth R. Colorimetric determination of melamine in milk using unmodified silver nanoparticles. Spectrochimica Acta, Part A: Molecular and Biomolecular Spectroscopy,2016:156:89-97. https://doi.org/10.1016/j.saa.2015.11.028.
26. Borase HP, Patil CD, Salunkhe RB, Suryawanshi RK, Salunke BK, Patil SV. Biofunctionalized silver nanoparticles as a novel colorimetric probe for melamine detection in raw milk. Biotechnology and Applied Biochemistry,2015:62:652-662. https://doi.org/10.1002/bab.1306.
27. Srimathi R, Priya D, Kumar BS. Determination of Melamine in Dry Milk by New Infrared Spectroscopy Method. Research Journal of Pharmacy and Technology,2017:10(12):6520660. https://doi:10.5958/0974-360X.2017.00782.X.
28. Dey MK, Reddy AVR, Satpati K. Determination of Melamine by a Reduced Graphene Oxide–Gold Nanoparticle Composite Carbon Paste Electrode. Analytical Letters,2016:49(17):2703-2715. https://doi:10.1080/00032719.2016.1154065.
29. Ezhilan M, Gumpu MB, Ramachandra BL. Design and development of electrochemical biosensor for the simultaneous detection of melamine and urea in adulterated milk samples. Sensors & Actuators, B: Chemical,2017:238:1283-1292. https://doi.org/10.1016/j.snb.2016.09.100.
30. Nandeshwar R, Malik M, D'Costa M. Molecular imprinting with polyaniline on ENIG finish PCB electrodes for electrochemical detection of melamine. IEEE Sensors,2022:22(3):1898-1904. https://doi:10.1109/JSEN.2021.3137515.