Electronic Supplementary Material (ESI) for Analytical Methods.

A Self-referenced Method for Determination of Patulin by Surface-enhanced Raman Scattering Using Gold Nanobipyramids as substrate

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Preparation of gold nanobipyramids, gold nanorode and gold nanosphere

The Au nanobipyramids were prepared using the seed-mediated growth method, as described in previous works [1-3]. Briefly, a freshly prepared, ice-cold $NaBH_4$ solution (0.01 M, 0.15 mL)was added under vigorous stirring into an aqueous solution that was pre-made by mixing together $HAuCl_4$ (0.01 M, 0.125 mL), trisodium citrate (0.01 M, 0.25 mL), and water (9.625 mL). The resultant seed solution was kept at room temperature for 2 h before use. The seed solution (0.2 mL) was injected into the growth solution that was made in advance by mixing together CTAB solution (0.1 M, 40 mL), $HAuCl_4$ (0.01 M, 2 mL), $AgNO_3$ (0.01 M, 0.4 mL), HCl (1 M, 0.8 mL), and ascorbic acid (0.1 M, 0.32 mL), followed by gentle inversion mixing for 10 s. The reaction solution was left undisturbed overnight at room temperature. The purification of the as-prepared Au nanobipyramids was conducted by using a depletion-induced separation method, as reported in a previous work [4].

Au nanorods were prepared by employing a seed-mediated, surfactant assisted method [5]. Firstly, Au seed solution was prepared by adding a freshly ice-cold $NaBH_4$ solution (0.60 mL, 0.01 M) into a mixed aqueous solution composed of CTAB (7.50 mL, 0.1 M) and $HAuCl_4$ (0.25 mL, 0.01 M), which was maintained under gently stirring for 2 min, and then kept at room temperature (25 ^oC) for 2 h. Secondly, a growth solution was prepared by mixing $HAuCl_4$ (5 mL, 0.01 M), $AgNO_3$ (0.35 mL, 10 mM) and CTAB (95 mL, 0.1 M), followed by adding a freshly prepared ascorbic acid solution (0.55 mL, 0.1 M). Thirdly, 120 mL of Au seed solution was dropped into the Au nanorods growth solution with rapid stirring for 2 min. The Au nanorods were left to grow during 4 h under stirring at 25 °C. Finally, the resulting Au nanorods were separated and purified by centrifuging at 10000 rpm for 30 min.

Gold colloid was synthesized by the G. Frens method of reducing chloroauric acid tetrahydrate with sodium citrate as reductant [6, 7]. 50 mL of 1 mM chloroauric acid tetrahydrate was boiled in a 250 mL round-bottom flask. Upon boiling, 1.85 mL 1% (w/v) sodium citrate aqueous solution was added into solution and the solution was then left to boil for an additional 15 min with mild stirring. When the solution turned wine red, the reduction was completed and the gold colloid was stored in a reagent bottle.

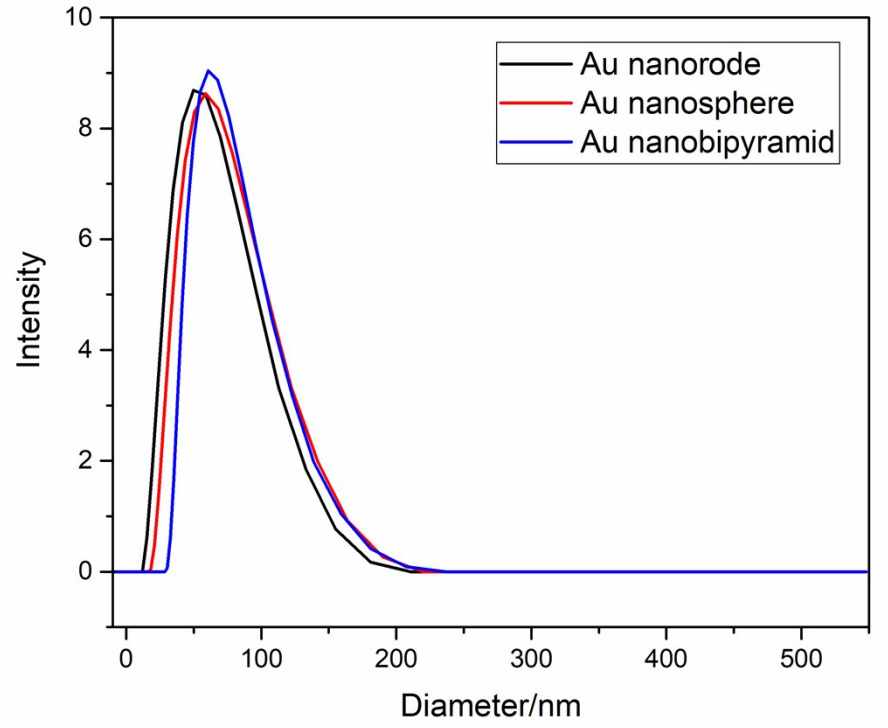

Fig. S1 The DLS spectra of Au nanobipyramid, Au nanorod and Au nanosphere

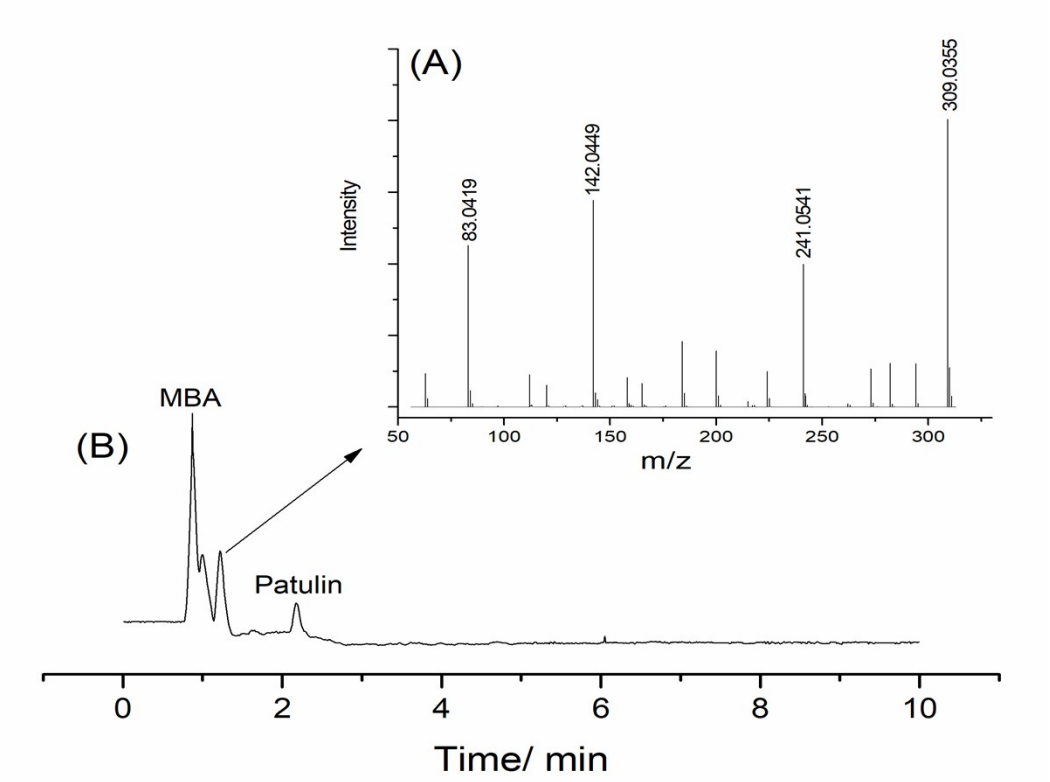

Fig. S2 (A) The MS spectrum of product A detected by HPLC-ESI(+)-MS/MS. (B) The HPLC spectrum of patulin with MBA adducts after incubation time of 2 min analyzed by HPLC-ESI(+)-MS/MS.

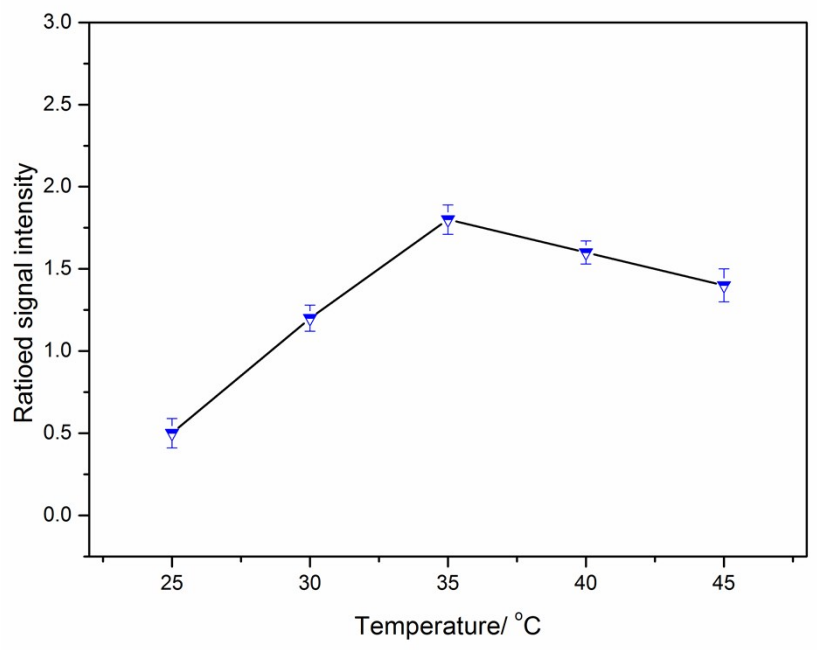

Fig. S3 The effect of reaction temperature on ratioed signal intensity of I_{1641}/I_{1586}.

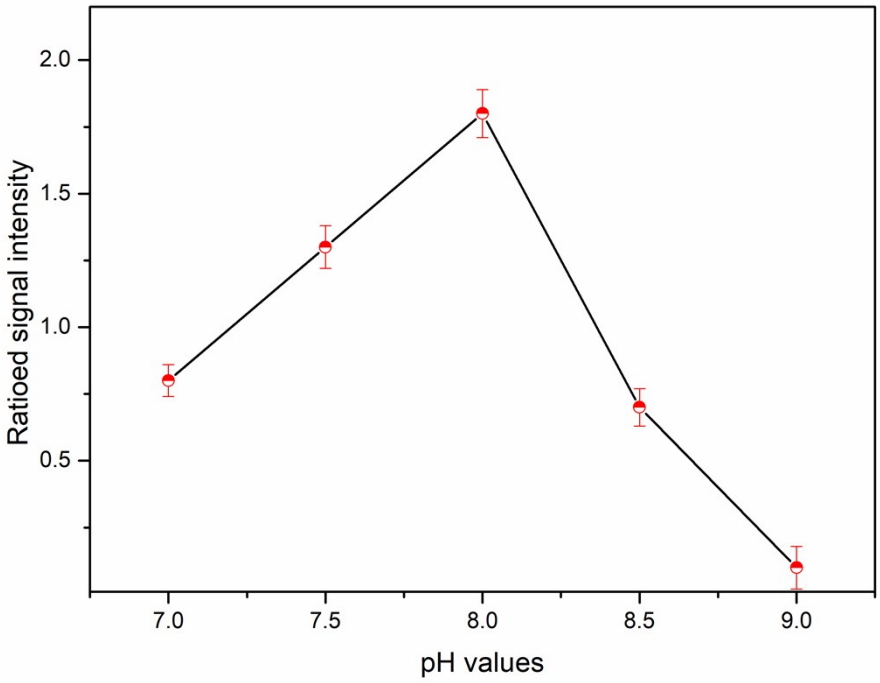

Fig. S4 The effect of pH on ratioed signal intensity of I_{1641}/I_{1586}

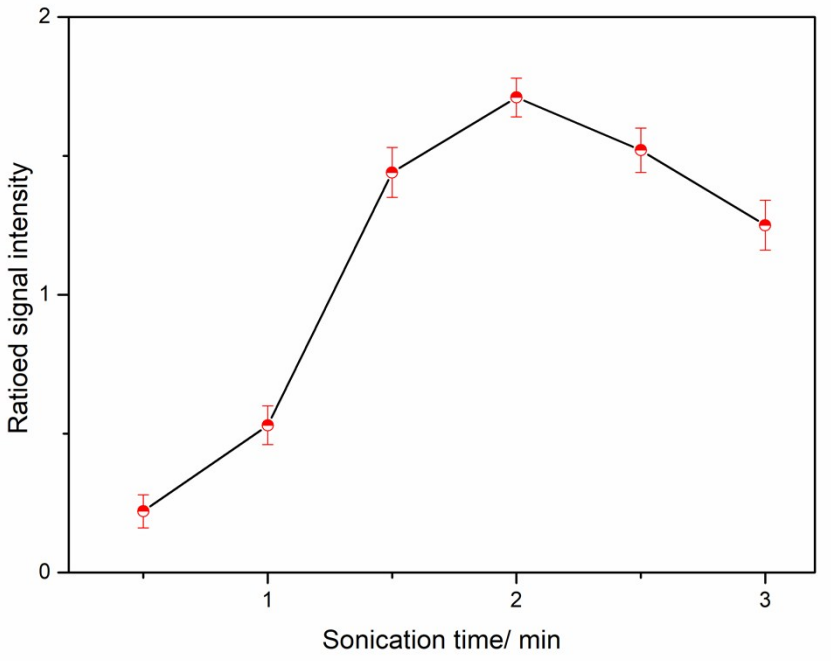

Fig. S5 The relationship between the ratioed signal intensity (I_{1641}/I_{1586}) and incubation time.

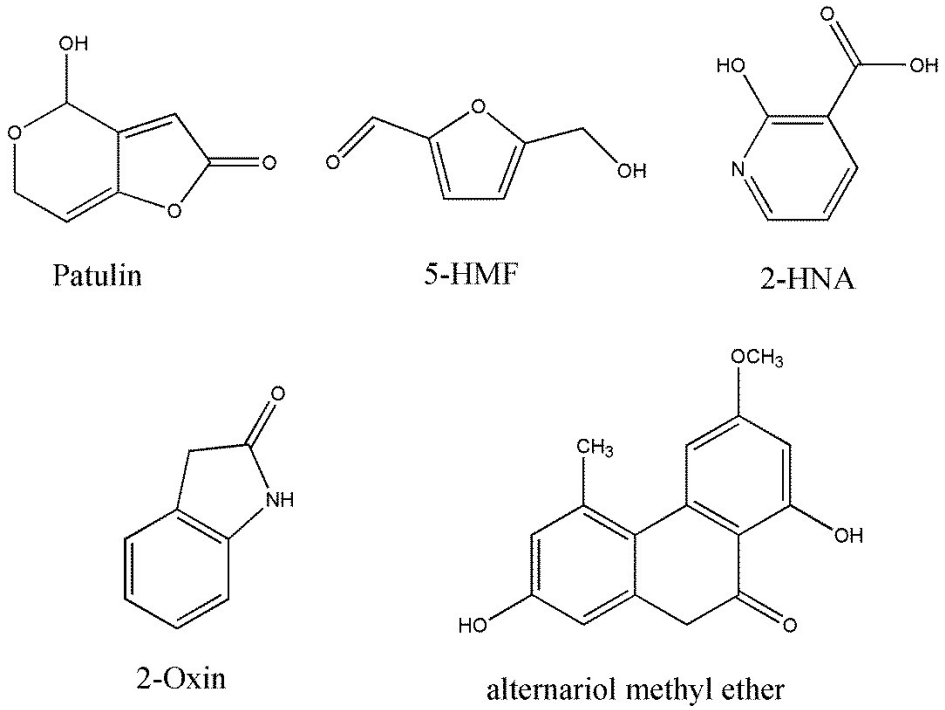

Fig. S6 Chemical structure of patulin and its analogue molecules; (Patulin, 5-HMF, 2-HNA, 2-Oxin and alternariol methyl ether.

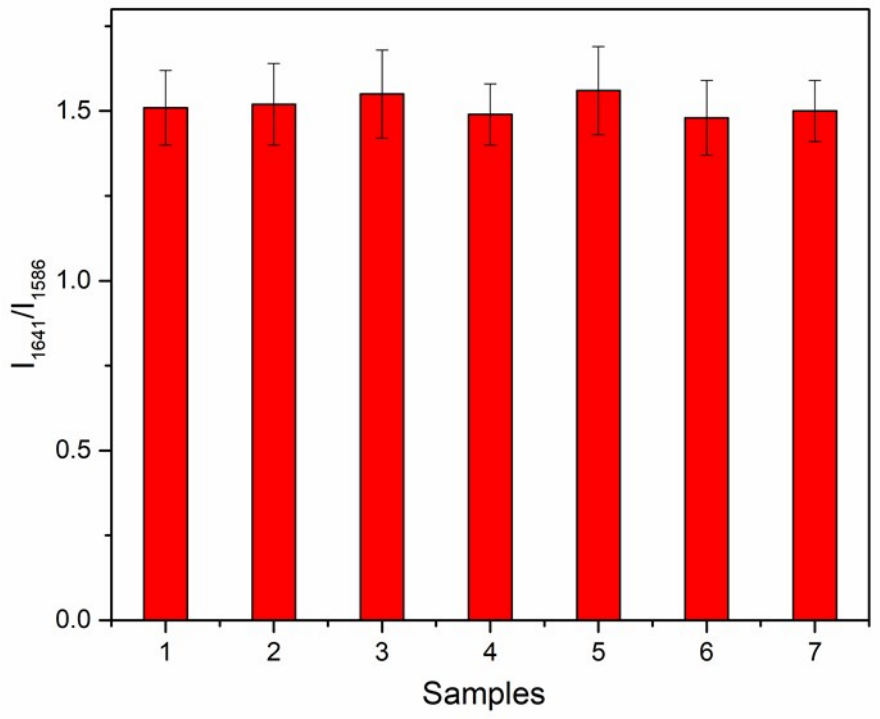

Fig. S7 The ratioed signal intensity of seven samples of 50μM 4-MBA with 100 μg/L patulin.

Table S1 Application of Au nanobipyramids for determination of patulin in real sample using this developed method (n=3) and HPLC (n=3).

Samples	SERS method (μg/L)	HPLC (μg/L)
Healthy apple juice	<LOD	<LOD
Rotten apple juice	126± 3	131± 1
Healthy pear juice	<LOD	<LOD
Rotten apple juice	78±6	72±3

Table S2. Comparison of previously reported method with the present method for detection of patulin.

Method	LOD	Analytical range	Comments	Ref.
SERS	6 μg/L	10-1000 μg/L	self-referenced; simple, cost-effective; high specificity; wide linear range.	This work
HPLC	3.99 μg/kg	2.5–250 μg/kg	Using solid-phase extraction as a pretreatment method; much organic solvent consumption	[8]
Electrochemistry	7.57×10^{-13} M	10^{-9} to 10^{-12} M	Using molecular imprinting technique as extracting reagent.	[9]
Fluorescence	10 μg/L	-	High selectivity; based on an antigen-antibody interactions; complicated synthesis of antibody	[10]
colorimetric	48 pg/mL	50-2500 pg/mL	High selectivity; aptamer-based method; complicated operation.	[11]
LC-MS	0.5 μg/L	2-2000 μg/L	Using a liquid-liquid-liquid microextraction as a pretreatment method; wide linear range.	[12]
Chemiluminescen-ce	0.01 ng/mL	0.05 to 80 ng/mL	High sensitivity; wide linear range; rapid.	[13]
HPLC	0.6 μg/kg	2–100 μg/kg	Selective extraction by molecularly imprinted method; complex operation.	[14]
Quartz-crystal microbalance	140 nM	-	High cost; good specificity; needing surface functionalization of antibody; complicated sample preparation processes;	[15]
MEKC*	0.6 μg/L	-	Using a microextraction process; much organic solvent consumption.	[16]

* Micellar electrokinetic capillary chromatography

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