## **Electronic supplementary information (ESI)**

## A novel third generation xanthine biosensor with enzyme modified glassy carbon electrode

## using electrodeposited MWCNT and nano-gold polymer composite film

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Measurements and Instrumentation

The electrochemical measurements were conducted using IVIUMStat electrochemical analyzer (Model: A09050, Iviumstat Technologies, USA) with IviumSoft software. Electrochemical impendence spectroscopy (EIS) was performed at each step of electrode modification using frequency response analyser (Eco Chemie B.V, Utrecht, Netherlands) attached with Autolab Serial nr. AUT72660 and controlled by FRA 4.9.006 software. EIS study of the modified electrodes was carried out in 5mM of  $[Fe(CN)6]_3^-$  and  $[Fe(CN)6]_4^-$  with 0.1M KCl within frequency range 1MHz to 0.01 Hz, amplitude 10mV at fixed potential of 0.28V.

Purified deionized water with conductivity of 18.2 M $\Omega$  cm (Model no: arium®611UF, Sartorius stedim biotech GmbH, Germany) was used for preparation of all solutions and buffers. Scanning electron microscopy (SEM) of the PPD film was conducted using JEOLEVO® 18 special edition

(Model: ZEISS EVO-MA 10), at an acceleration voltage 15kV. Field emission scanning electron microscopic (FESEM) image of electrodeposited fMWCNT onto the PPD/Au film was obtained by JEOL JSM-7600F. The sample for SEM and FESEM was mounted in the stub using carbon tap and coated with platinum (5-7nm thickness) by a Quorum Q150TES Ion Sputter.

The characterization of gold nanoparticles in PPD film was monitored using transmission electron microscopy (TEM model: JEOL JEM 2100 HR with EELS and EDS attachment of INCAx-sight Energy TEM 200, Oxford Instruments, UK) at an acceleration voltage of 200 kV. The presence of specific atom was confirmed by energy diffraction atomic spectroscopic (EDAS) attachment of INCA Penta FETX3, OXFORD Instruments, UK with TEM.

Topographical characterization of the modified electrode was performed using atomic force microscopic (AFM) in tapping mode with large area scanner probe (RTESPA) of Model Innova, Bruker AXS Pte Ltd. AFM images were taken with a resonance frequency of 285 kHz and scan rate of 1.0 Hz.

The X-ray diffraction analysis was performed in the range of 10-90° 2 $\Theta$  at a step-scan of 0.0332 $\Theta$ /s on Phillips X'pert wide angle X-ray diffractometer (USA) in reflection mode using Ni filtered Cu-ka (30 mA, 40 kV) radiation ( $\lambda = 1.5406$  Å).

The Fourier transform infrared analysis was performed by ThermoScientific FTIR Instrument (model no: Nicolet iS10) in the range of 400-4000 cm<sup>-1</sup> with a scan rate of 4cm<sup>-1</sup>.



S-Fig 1.XRDof Au-PPD film.



**S-Fig 2.**XRD spectra of before and after functionalization of MWCNT(a) and EDAX of fMWCNT(b) showing presence of oxygen molecules on the carboxylatedMWCNT.



S-Fig 3.AFM topography images intapping phase mode of PPD(a), Au-PPD(b), fMWCNT/Au-

PPD(c);



S-Fig 4.Relative stability of the proposed XO/fMWCNT/Au-PPD/GCEsensor with time.

**Table** Within and between batch assay coefficients of variation (CV) for determination of xanthine in fish samples by XO/fMWCNT/Au-PPD/GCE

Ν	Xanthine (µM)	CV (%)
Within assays (6)		
26.55		
26.48		
25.95	26.47±0.51	1.93
27.24		
26.74		
25.86		
between assays (6)		
27.92		
25.78		
26.16	26.61±0.82	3.08
26.65		
27.19		
25.98		