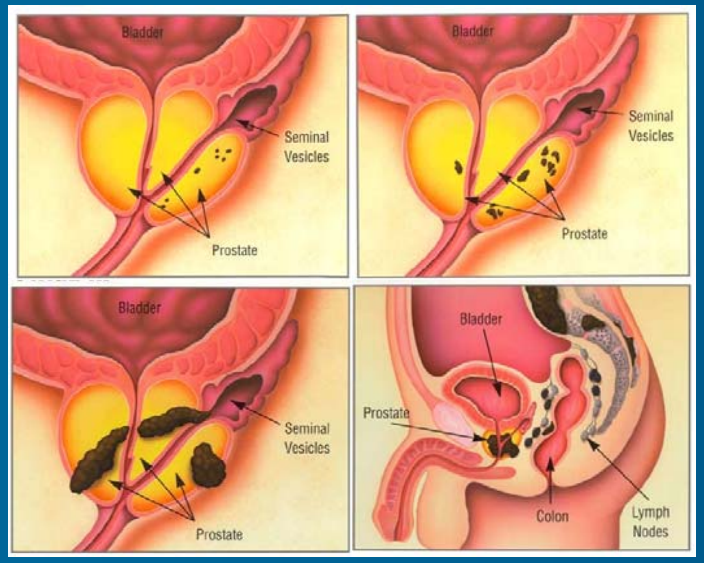
Departamento de Química Física y Analítica

UNIVERSIDAD DE OVIEDO

Dual sensor based on gold nanostructured screen-printed carbon electrodes for the detection of prostate specific antigen (PSA)

DUAL SENSOR BASED ON GOLD NANOSTRUCTURED SCREEN-PRINTED CARBON ELECTRODES FOR THE DETECTION OF PROSTATE SPECIFIC ANTIGEN (PSA)

INTRODUCTION



Prostate specific antigen (PSA) is a glycoprotein (93% peptide, 7% sugar) produced by prostate to liquefy the seminal fluid, and is the most reliable tool for diagnosing prostate cancer. PSA in serum circulates non-complexed (free PSA, fPSA, , MW 33 KDa) and complexed in several forms, being the predominant one the complex with α1-antichymotripsin (PSA-ACT, MW 90 KDa)

Free PSA (fPSA)

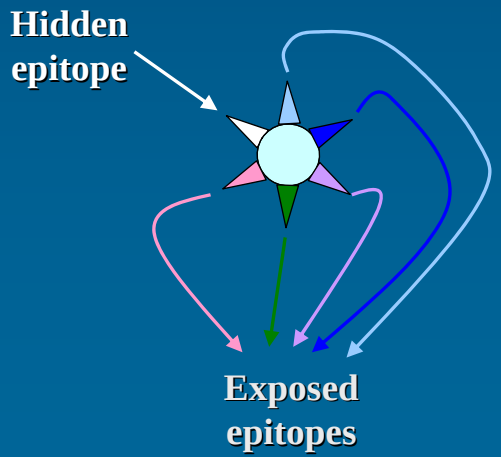
Total PSA (tPSA)

[tPSA]

< 2.5 ng/mL	2.5 ng/mL - 10 ng/mL	> 10 ng/mL
Low cancer risk	Grey zone	Cancer risk

[fPSA]/[tPSA]

> 0.25	< 0.25
Low cancer risk	Cancer risk



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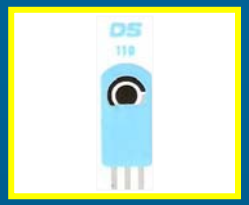
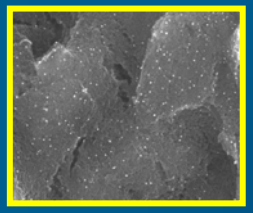
FUNDAMENTALS

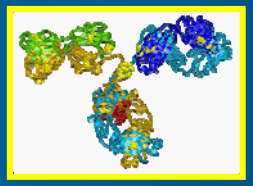
Screen-printed Electrodes



Nanostructuration

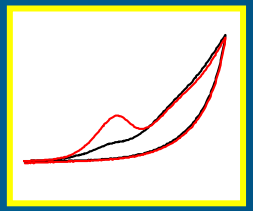


Immunological reaction



Enzymatic catalysis

Voltammetric detection



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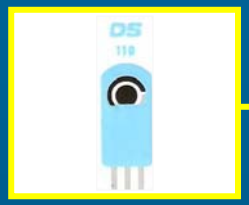

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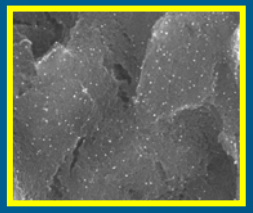

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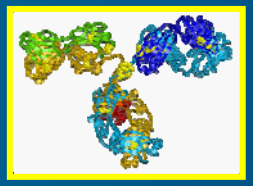
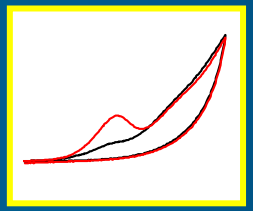
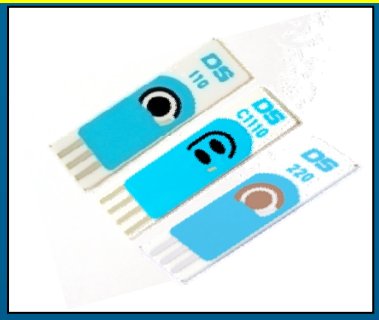


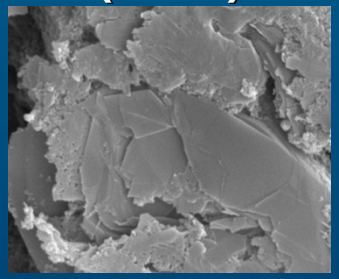
Enzymatic catalysis

Voltammetric detection



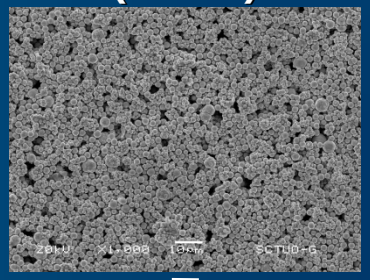


Screen-printed Carbon Electrode (SPCE)



5 µm

Screen-printed Gold Electrode (SPGE)



10 µm

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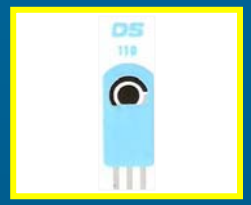
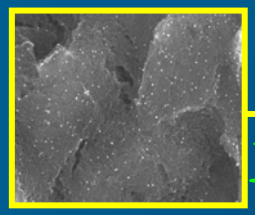
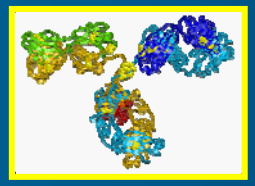


Nanostructuration

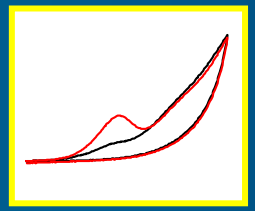


Immunological reaction



Enzymatic catalysis

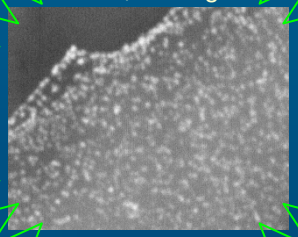
Voltammetric detection



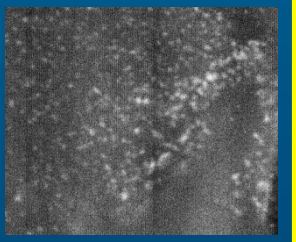
Nanostructuration by synthesis *in situ* of gold nanoparticles (gold electrodeposition)

Screen-printed Carbon – nanoGold Electrode (SPC-nAu-E)

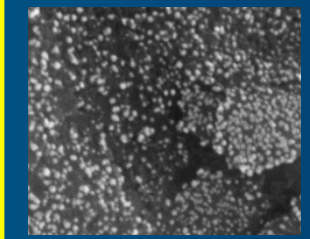
S1
70 nm, 100 ng Au



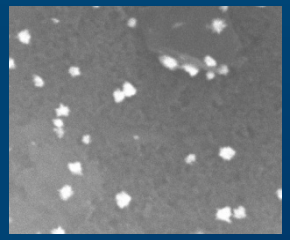
S2
70 nm, 100 ng Au



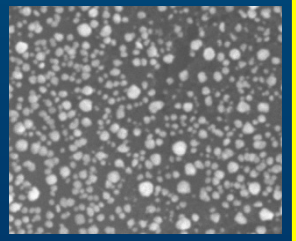
S3
70 nm, 700 ng Au



S4
160 nm, 700 ng Au



S5
160 nm, 1850 ng Au



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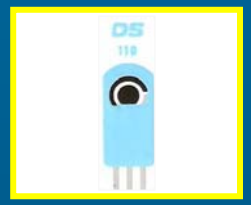
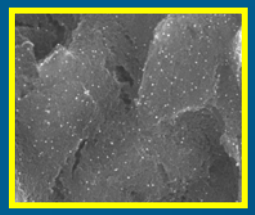
FUNDAMENTALS

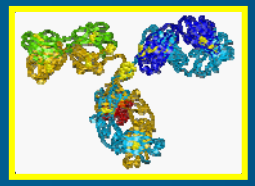
Screen-printed Electrodes



Nanostructuration

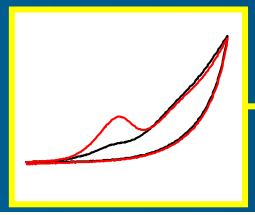


Immunological reaction



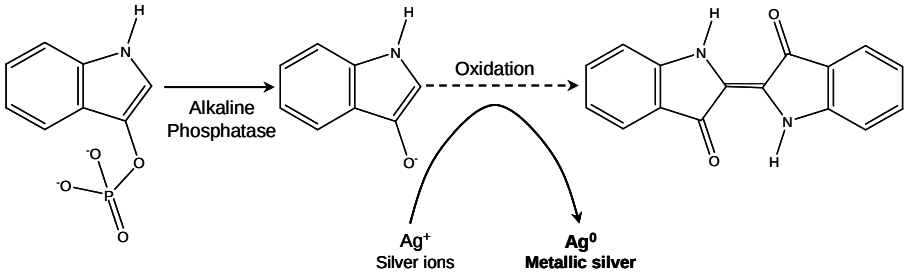
Enzymatic catalysis

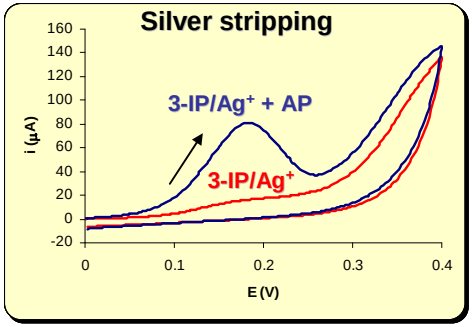
Voltammetric detection



P. Fanjul-Bolado, D. Hernández-Santos, M.B. González-García, A. Costa-García, ***Anal. Chem.***, 2007 (*79*), 5272

Metalloenzymatic detection





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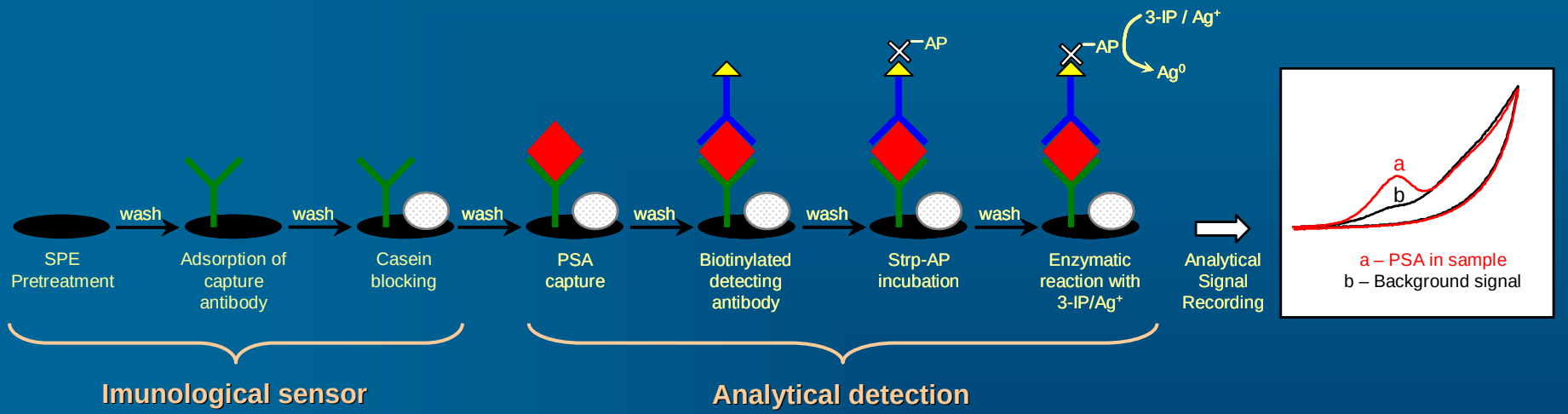
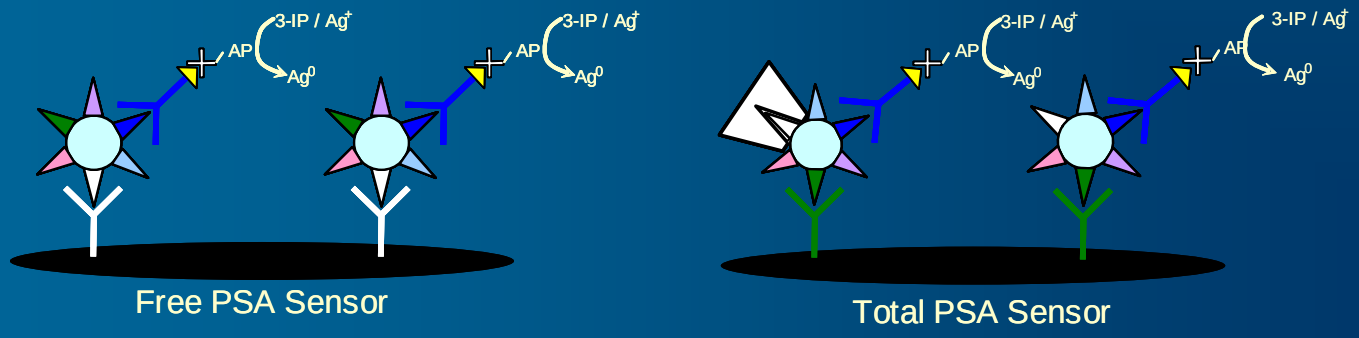
IMMUNOSENSOR DEVELOPMENT

ANALYTICAL SCHEME



Immunosensor formats



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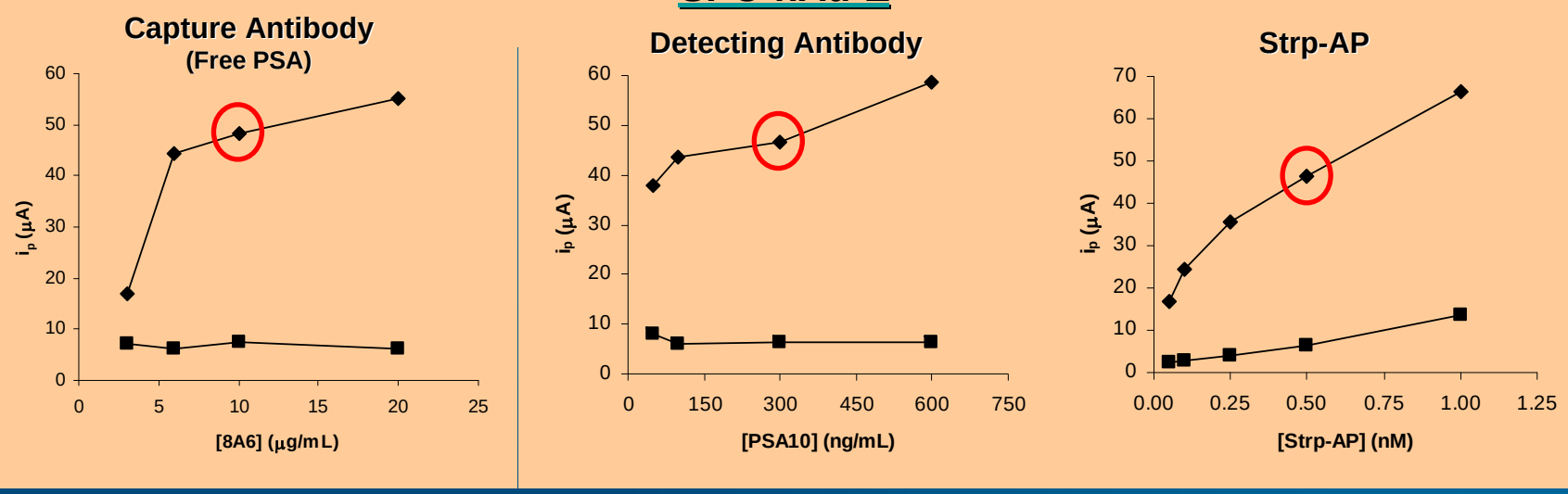
IMMUNOSENSOR DEVELOPMENT

OPTMIZATION OF PAREMETERS AND CALIBRATIONS

SPC-nAu-E



PARAMETERS	SPCEs	SPGEs	SPC-nAu-Es
Capture antibody [8A6] (f) , [5G6] (t) (µg/mL)	6	10	10
[PSA10] (ng/mL)	300	100	300
[Strp-AP] (nM)	0.5	1	0.5

CALIBRATIONS		Linear range (ng/mL)	Slope µA/(ng/mL)	r
SPCE	5G6 (t)	10 - 100	0.587	0.990
	8A6 (f)	10 - 100	0.377	0.999
SPGE	5G6 (t)	10 - 100	0.496	0.990
	8A6 (f)	10 - 100	0.277	0.990
SPC-nAu-E	5G6 (t)	**1 - 10**	**12.300**	0.994
	8A6 (f)	**1 - 10**	**6.081**	0.992

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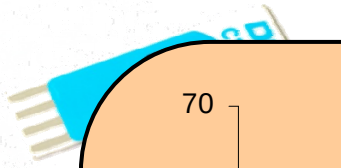
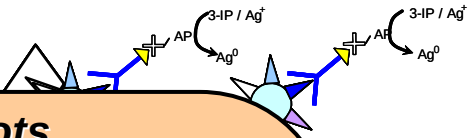
IMMUNOSENSOR DEVELOPMENT

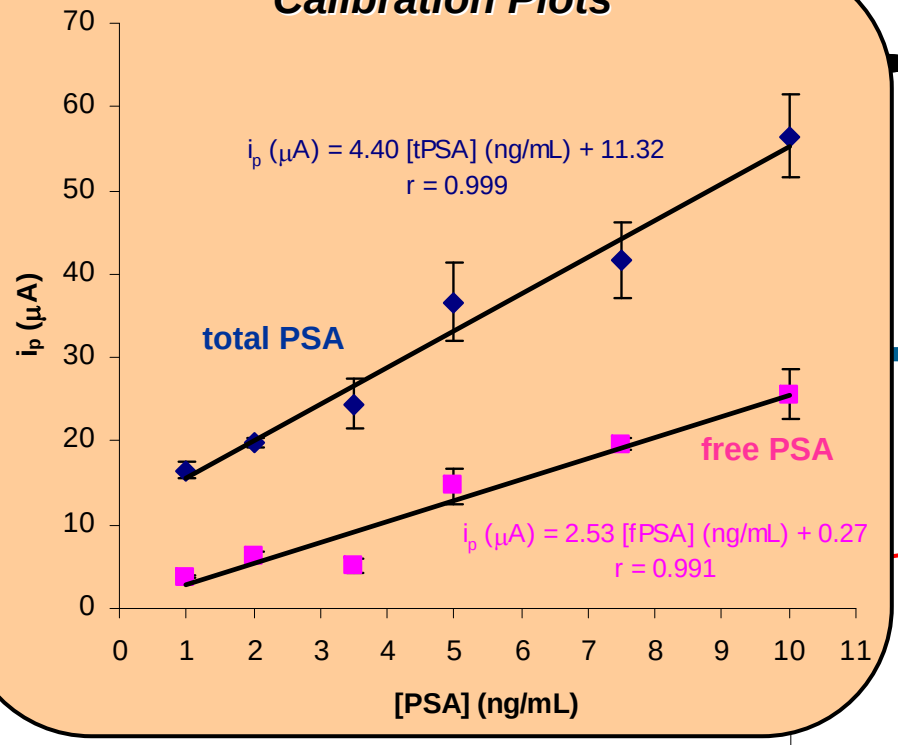
SIMULTANEOUS DETECTION OF fPSA AND tPSA

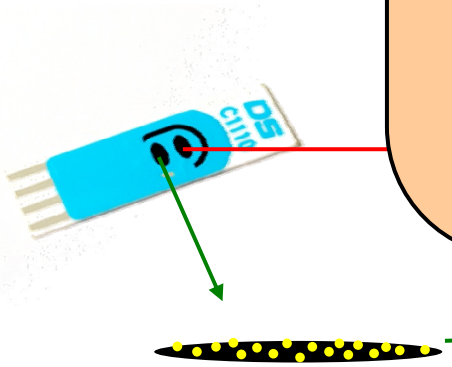
Different coating on each electrode

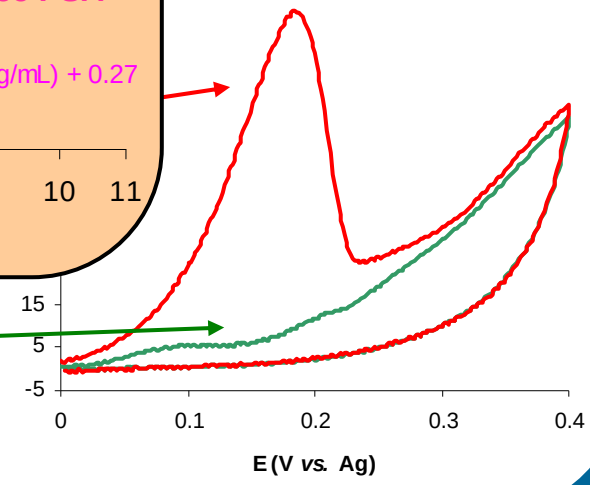




Calibration Plots







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IMMUNOSENSOR DEVELOPMENT

SIMULTANEOUS DETECTION OF fPSA AND tPSA

Serum samples

Monitor PSA production from three different cultures of human androgen-sensitive LNCaP prostate tumor cells

CON

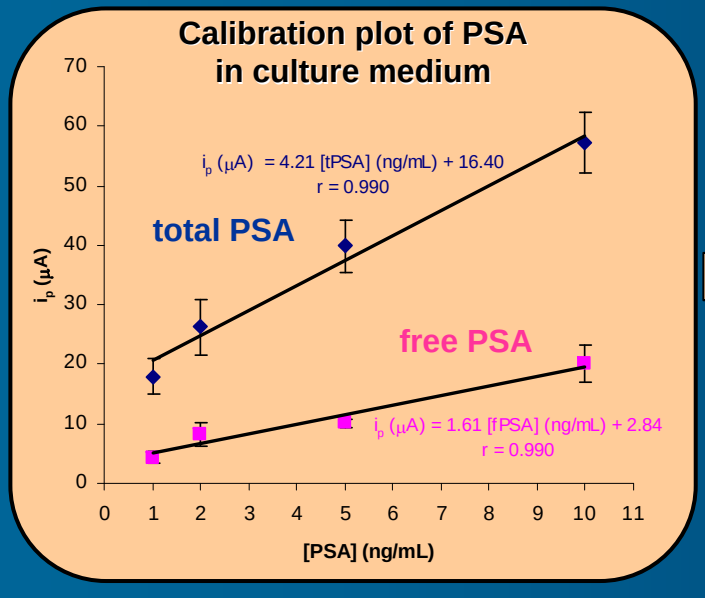
LNCaP cells maintained in RPMI 1640 culture medium supplemented with 10% Fetal Bovine Serum (FBS)
(culture medium with normal content of androgens)

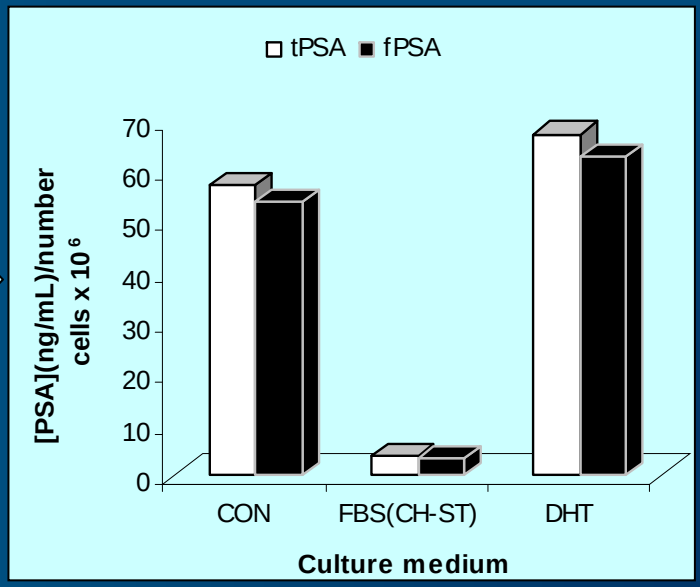
$FBS_{(CHT-ST)}$

Charcoal-stripped FBS was used instead of normal complete FBS
(culture medium without androgens)

DHT

Dihidrotestosterone, the active form of testosterone, was used in the culture
(culture medium with enhanced content of androgens)

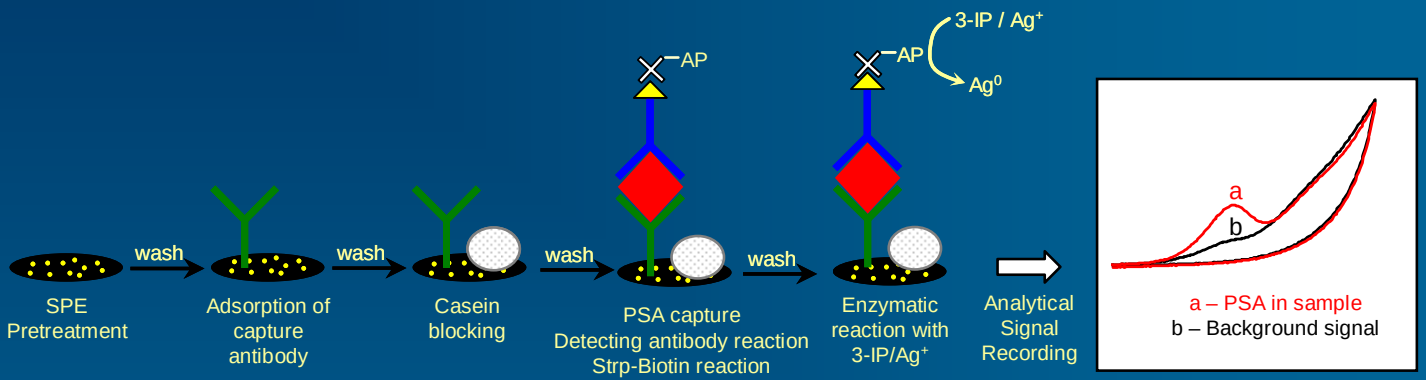


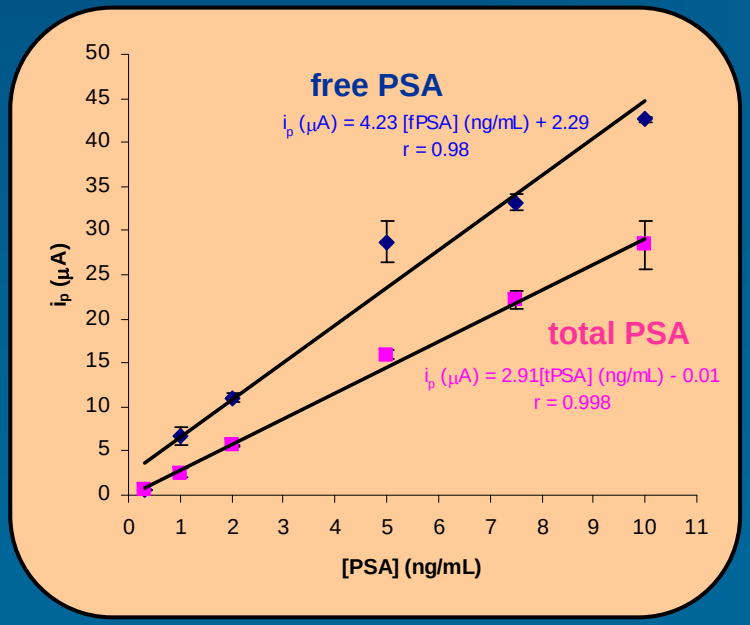


IMMUNOSENSOR DEVELOPMENT

SIMULTANEOUS DETECTION OF fPSA AND tPSA

ONE-STEP FORMAT





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AUTHORS

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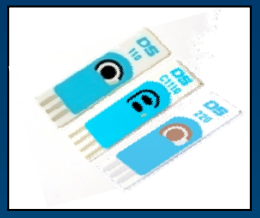
FINANCIAL SUPPORT

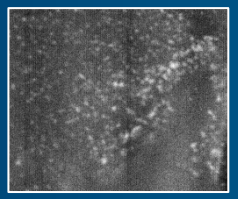
Gobierno del Principado de Asturias
Consejería de Educación y Ciencia

Project PC06-004

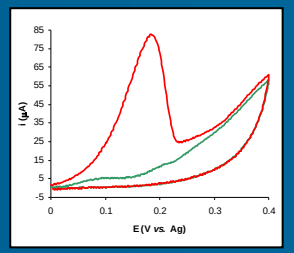
PCTI Asturias
Plan de Ciencia, Tecnología e Innovación de Asturias 2006 - 2009

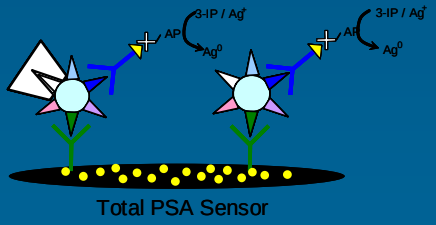
Immunobisensor for free and total PSA detection in serum

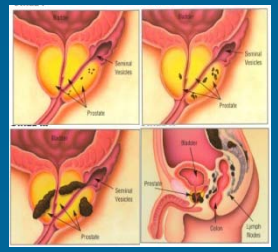




Thank you for your kind attention !







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