Unitat de Recerca Biomèdica i d'Oncologia Translacional i Pediàtrica

Institut de Recerca i Hospital Universitari Vall Hebron

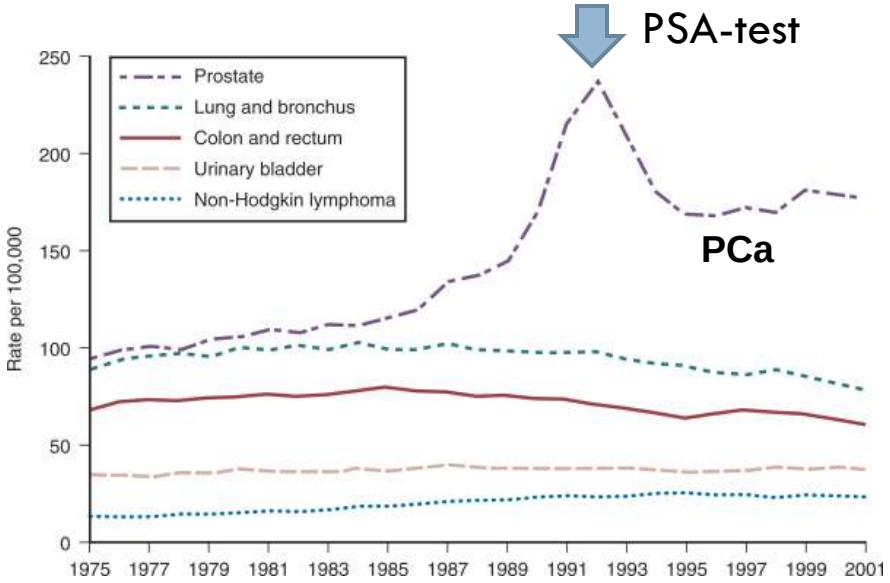
"Nuevos marcadores de diagnóstico en el cáncer de próstata"

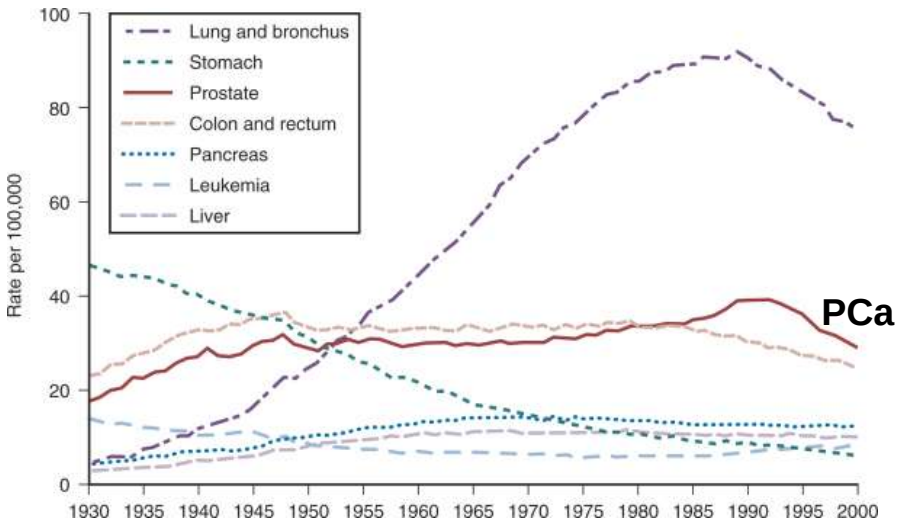
Jaume Reventós M.D.,Ph.D.

Congreso Nacional de Patología, Zaragoza, 18.05.2011

Incidence and Mortality in Prostate Cancer

- Most frequent cancers in men
- Most mortal cancers in men





Prostate cancer is the **most common malignancy** and **the second cause of death** after lung cancer in **men in the western world**

Since the introduction of the PSA-test the incidence of localized prostate cancer has increased but the incidence of metastatic cancer has decreased

Actual Diagnostic Tripod of Pca

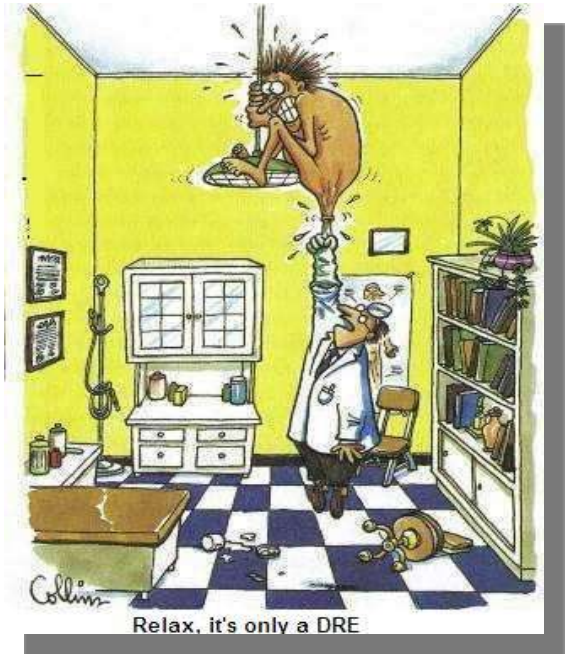
Serum PSA level (≥4.0ng/ml) or annual tendency to increase ≥ 2ng/ml

Serum PSA [ng/mL]	DRE- % PCa	DRE+ % PCa	Bx
0-2	1	5	no
2-4	15	20	no
4-10	25	45	yes
>10	>50	>75	yes
>20	95		yes

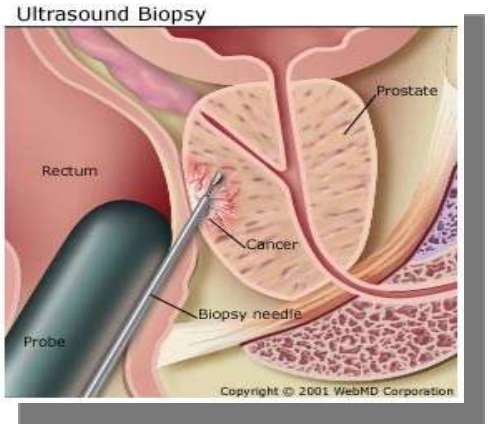
PSA 4-10 ng/ml → HUVH >**66%** of men will not have PCa (**false positive**).

Poor PSA **specificity (33 %)**

Digital rectal examination (DRE)



Biopsy



Prostate biopsy is the "**gold standard**" for the definitive diagnostic of PCa

Up to **12-30% false negative** results in the first biopsy

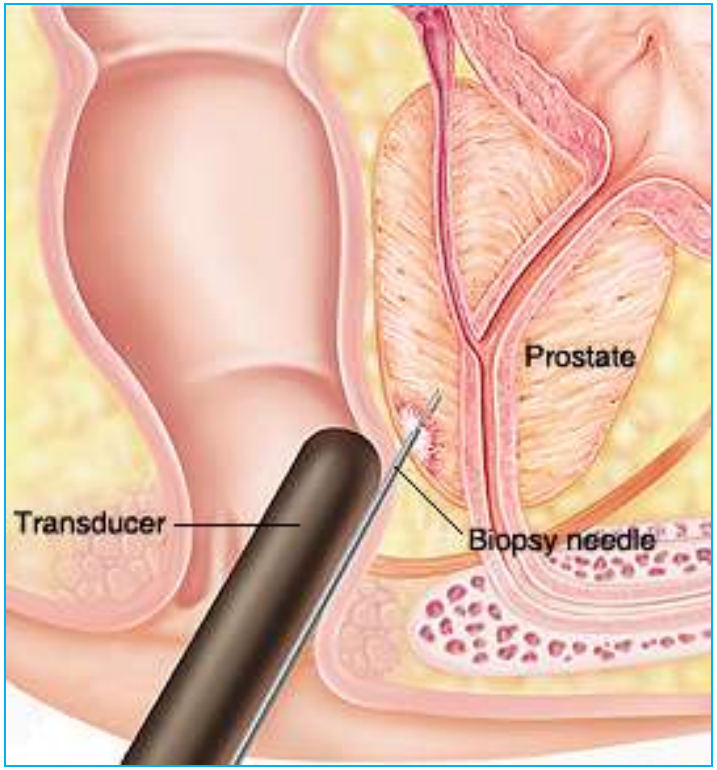
There is a diagnostic dillema and more PCa specific biomarkers are needed

The Digital Rectal Exam (DRE)

- Can detect a tumors since most prostate cancers arise within the peripheral zone.
- **The formation of a prostatic tumor can be detected as a hard or abnormal mass, discrete nodule, asymetric shape, …**
- But…it is subjected to some human variability.
- If PSA< 4 ng/ml detection rate is max 10-15%
- 40-70% already no more organ confined

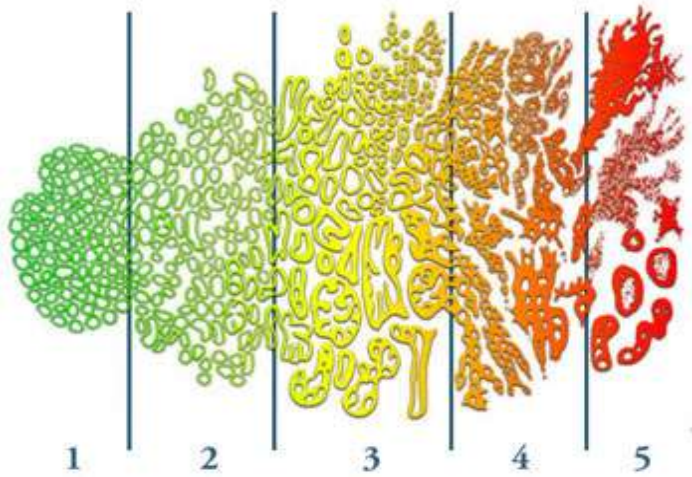
Prostate Biopsy

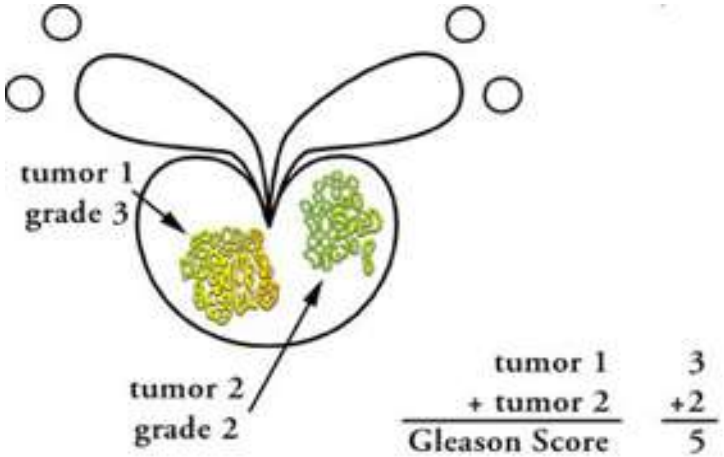
- **Gold standard to confirm the presence of prostate cancer**
- Performed after rising PSA and/or suspicious DRE
- Purpose: take prostate tissue and examine under a microscope (histology)



Gleason Grading

Sum of primary and secondary **Gleason Grades** within a prostate cancer tissue specimen. (Lowest 1+1=2; Highest 5+5=10)





Gleason Score 2-4

Low-grade Tumor

Lowest Chance of Cancer Invading and Metastasizing

Gleason Score 5-7

Mid-Grade Tumor

Gleason Score 8-10

High-Grade Tumor

Highest Chance of Cancer Invading and Metastasizing

Diagnostic dilemma

Men with at least one **negative prostatic biopsy** sometimes have an **increased PSA value** caused by **BPH, prostatitis, prostatic infarct, urine retention** etc.

FALSE POSITIVES (aprox. 66%) = not necessary biopsy

A significant portion of men with only **moderate increased PSA** values oscillating around (2.5 – 4.0 ng/mL) harbor a **not diagnosed prostate cancer** because the PSA level is to low to justify a biopsy

FALSE NEGATIVES (aprox. 20%) = cancer present but not detected

Only a fraction of men with increased PSA represent the group of detectable cancers

The Prostatectomy – The 2nd Dilemma

apparently organ-confined

evidently metastatic

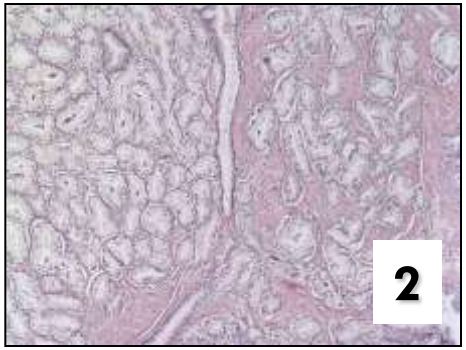
Gleason grading score

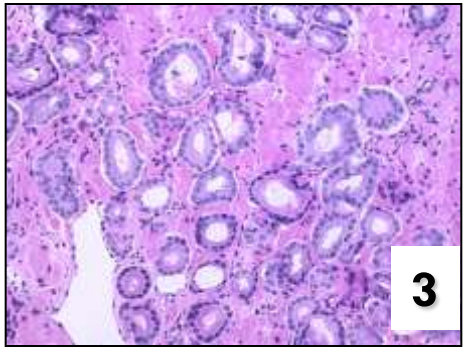
Cancer aggresiveness

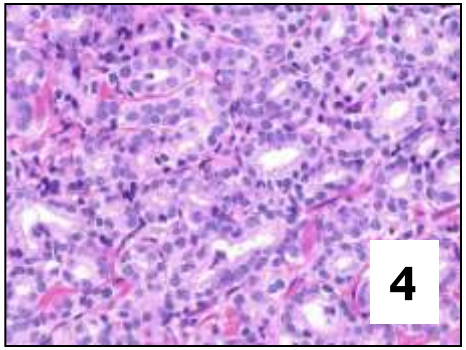
unnecessary

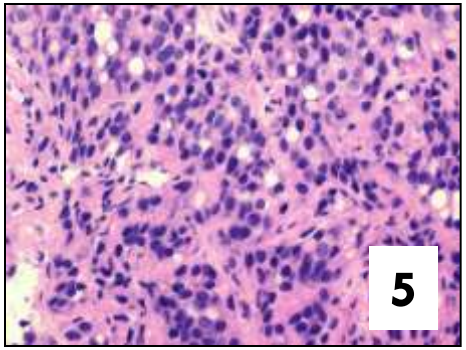
curative

useless









The Need for Biomarkers - Cancer

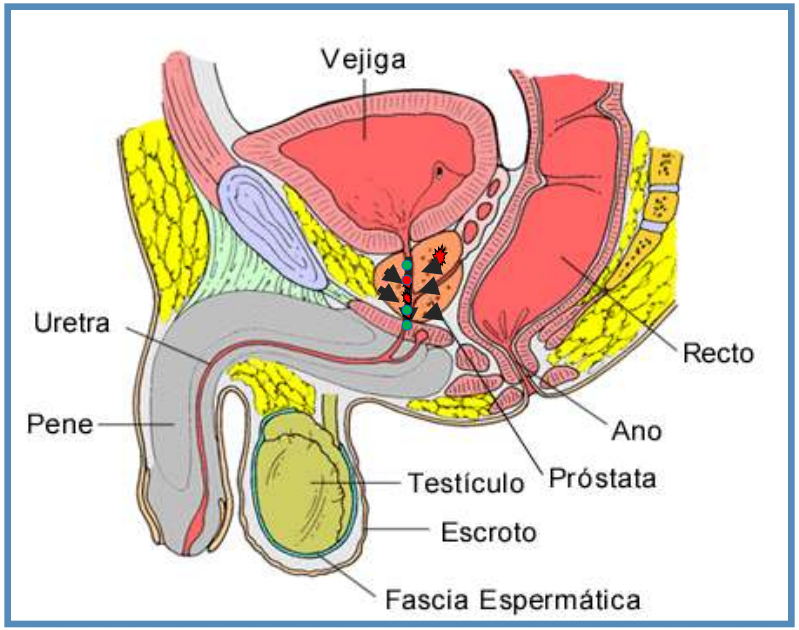
- **Cancer diagnosis** is typically based on assessment of morphological alterations of cells and tissues
  - Significant number of cases are ambiguous
  - Cannot predict patient responses to treatment

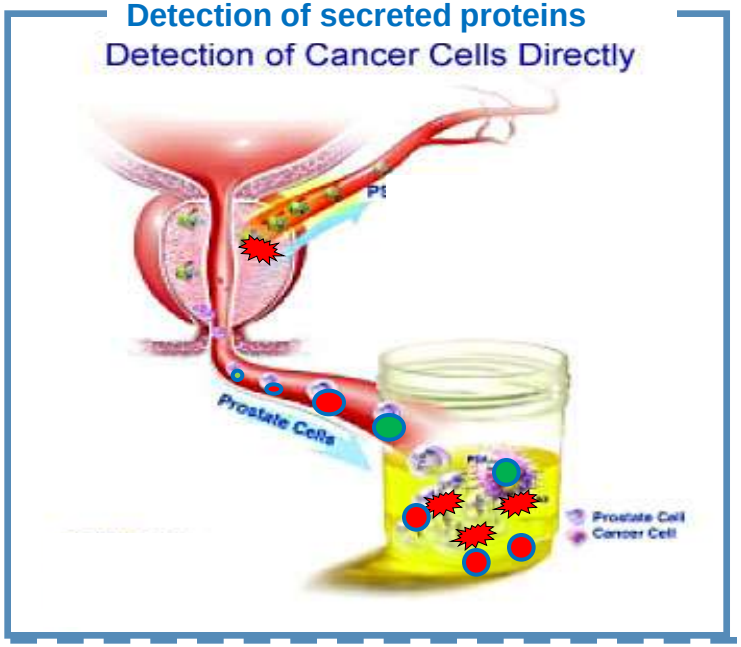
Biomarkers are needed:

- **Diagnostic markers** - aid in tumor classification - what type of tumor?
- **Prognostic markers** - provide info on malignant potential of tumor
- **Predictive markers** - aid in the choice of treatment modalities
  - i.e. breast cancer patients with estrogen receptor positive tumors get treated with anti-estrogen drugs

How to find prostate cancer biomarkers?
Working Hypothesis

- A **small portion** of normal **prostate cells** and cancer cells and their products **continuously disseminate** from the epithelium and can be found in urine
- **Prostate cancer cells** are **secreting different products** which can be detected in the urine
- A **prostatic massage** leads to an **enrichment** of prostatic fluid and prostatic cells in the first urine catch after massage

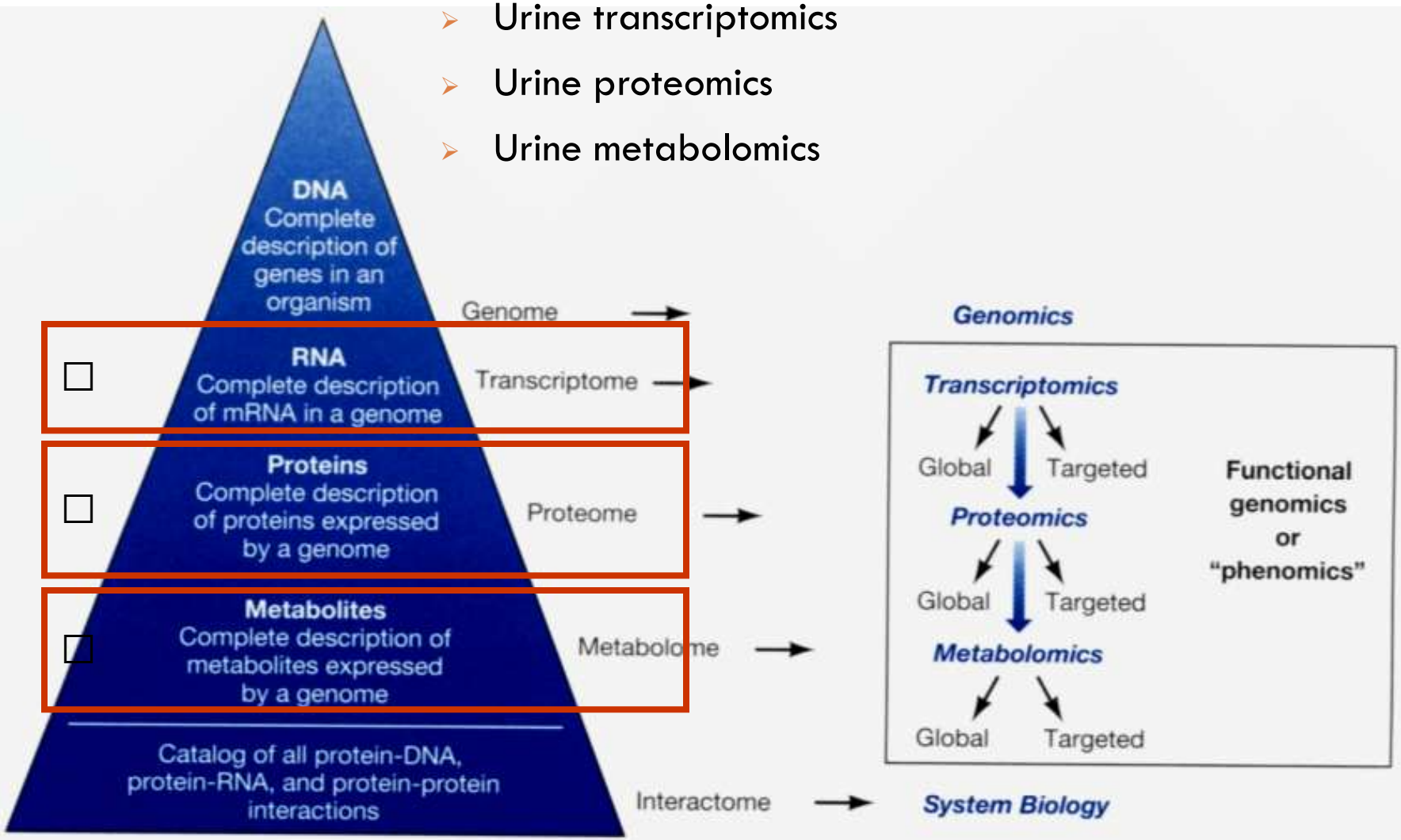


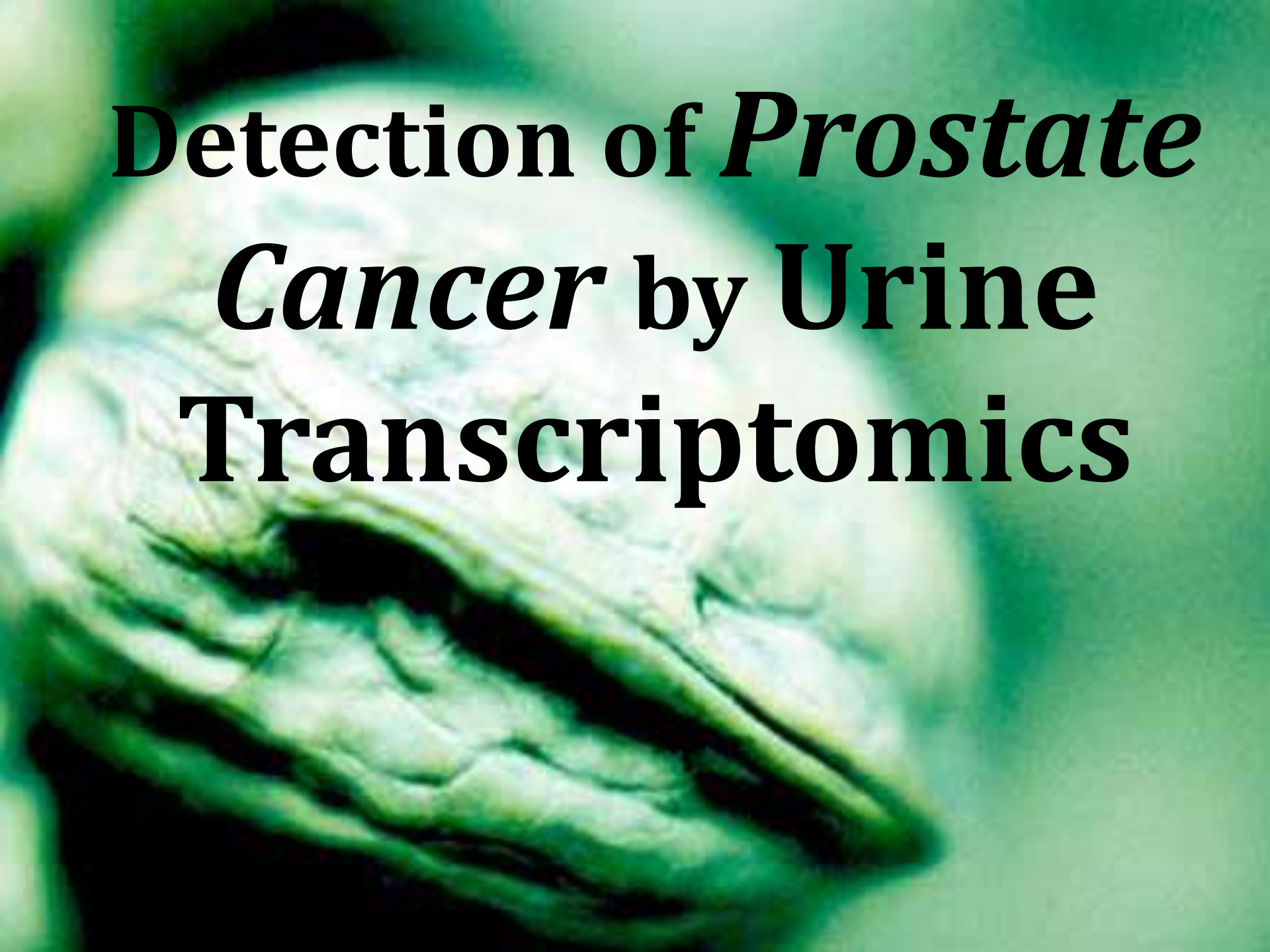


Urine after a (firm) massage → Non invasive method for the detection of PCa

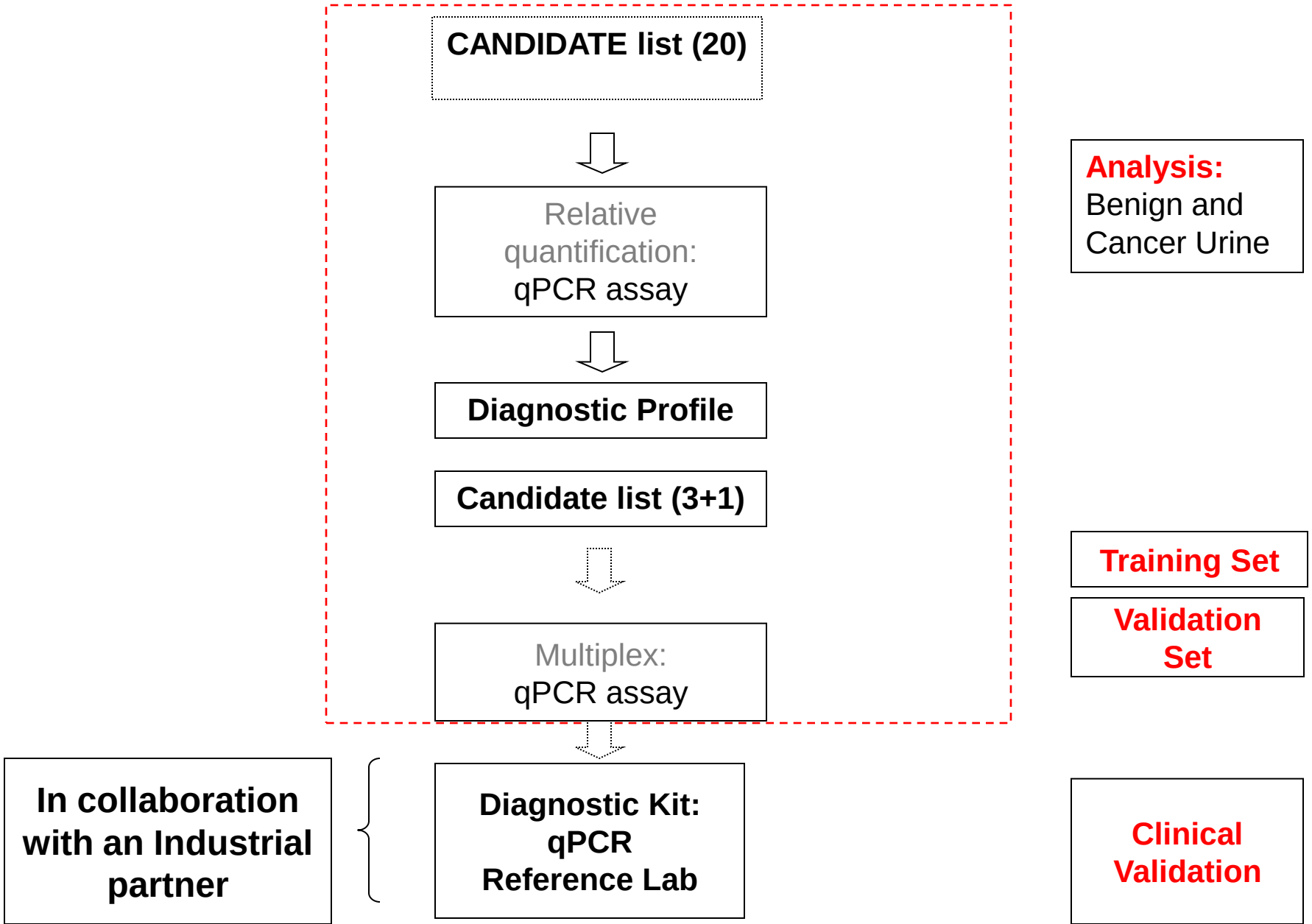
Current -omics

- Urine transcriptomics
- Urine proteomics
- Urine metabolomics

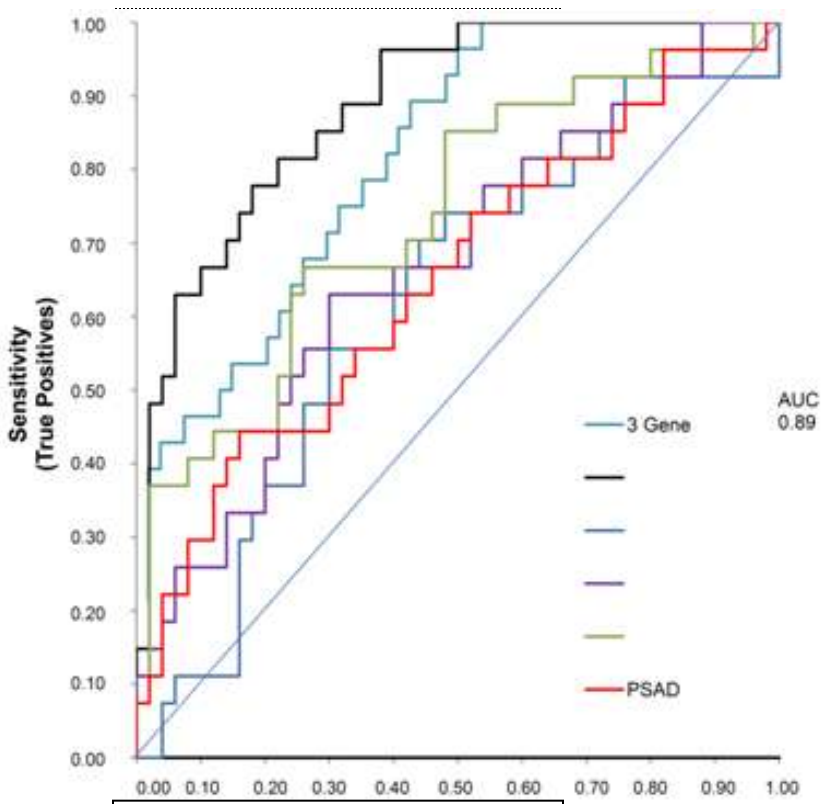



Detection of *Prostate Cancer* by Urine Transcriptomics

IDENTIFY NEW BIOMARKERS BY GENOMICS



IDENTIFY NEW BIOMARKERS BY GENOMICS



Multiplex:
qPCR assay

Analysis:
Benign and
Cancer Urine

Training Set

Validation Set

In collaboration with an Industrial partner

Diagnostic Kit:
qPCR
Reference Lab

Clinical Validation

IDENTIFY NEW BIOMARKERS BY GENOMICS

- Our multiplex qPCR marker panel represents a sensitive method to suspect prostate cancer in urine which can be used to increase the specificity of PSA avoiding a significant number of unnecessary biopsies.
- This novel method increases the efficiency of PSA and it is especially useful in the gray zone of PSA.
- Having a sensitivity of 96% the specificity was 62% would allow us to save 42% of unnecessary performed biopsies.

Analysis:
Benign and Cancer Urine

Training Set

Validation Set

In collaboration with an Industrial partner

Diagnostic Kit:
qPCR
Reference Lab

Clinical Validation

Transcriptomic identified biomarkers for the diagnosis of prostate cancer in urine:

Prostate specific membrane antigen (PSMA);
Prostate-specific G-protein coupled receptor (PSGR);
Prostate cancer antigen 3 (PCA3).

PSGR and PCA3 as Biomarkers for the Detection of Prostate Cancer in Urine

Marina Rigau,[1] Juan Morote,[2,3] Maria Carmen Mir,[2] Carlos Ballesteros,[2] Israel Ortega,[4] Alex Sanchez,[4,5] Eva Colás,[1] Marta Garcia,[1] Anna Ruiz,[1] Miguel Abal,[6] Jacques Planas,[2] Jaume Reventós,[1,3] and Andreas Doll[1]*

[1]*Biomedical Research Unit, Research Institute, Vall d'Hebron University Hospital, Barcelona, Spain*
[2]*Department of Urology, Vall d'Hebron University Hospital, Barcelona, Spain*
[3]*School of Medicine, Autonomous University of Barcelona, Spain*
[4]*Statistics and Bioinformatics Unit, Vall d'Hebron University Hospital, Barcelona, Spain*
[5]*Statistics Department, University of Barcelona, Barcelona, Spain*
[6]*Translational Oncology Laboratory, Instituto de Investigaciones Sanitarias, Complejo Hospitalario Universitario de Santiago (CHUS), Santiago de Compostela, Spain*

BACKGROUND. Several studies have demonstrated the usefulness of monitoring an RNA transcript in urine, such as PCA3, for prostate cancer (PCa) diagnosis. PCa screening would benefit from additional biomarkers of higher specificity and could be used in conjunction with

METHODS AND KITS FOR THE DIAGNOSIS OF PROSTATE CANCER

FIELD OF THE INVENTION

The invention fall within the field of diagnosis and, more specifically, in the field of diagnosis of prostate cancer by means of using a marker gene the expression of which is over-expressed in urine samples from prostate cancer patients.

BACKGROUND OF THE INVENTION

Prostate cancer (PCa) has transformed into the most common type of cancer in the Western male population, where it is responsible for more male deaths than any other cancer, except lung cancer. Even though the introduction of the prostate specific antigen (PSA) test in the late 80s of the past century has led to a dramatic increase its detection (Jemal A. et al. Cancer statistics, 2007,57:43-66), the risk of developing this type of cancer during a lifetime is estimated at one in six men, and the risk of death due to metastatic PCa is 1 in 30.

The diagnostic tools for detecting PCa can be separated into those that screen for the disease, such as PSA and digital rectal exams (DRE), and the decisive diagnosis set of transrectal ultrasound guided prostate biopsies (TRUS).

One of the limitations of serum PSA as a tumor marker is its lack of specificity, which results in a high rate of negative biopsies. There is a substantial overlap in the serum PSA values of men with benign prostatic hyperplasia (BPH) and those with PCa. Elevated PSA levels can also be attributed to other factors such as prostatitis, prostate irritation, and recent ejaculation (Pannek J y Partin AW, Oncology, 11:1273-8 y Loeb, S. 2009, Urol. Oncol.27:64-6). As a consequence of the current screening parameters, around 2/3 of the approximately 1,300,000 biopsies made yearly in the United States (390,000 in Europe) are unnecessary (Catalona WJ. et al., 1991, N. Engl. J. Med. 324:1156-61 y Makinen, T. et al. 2004, Clin Cancer Res 10:2231-6). In contrast, the false positive rate of a biopsy is about zero, although the false negative rate in the first

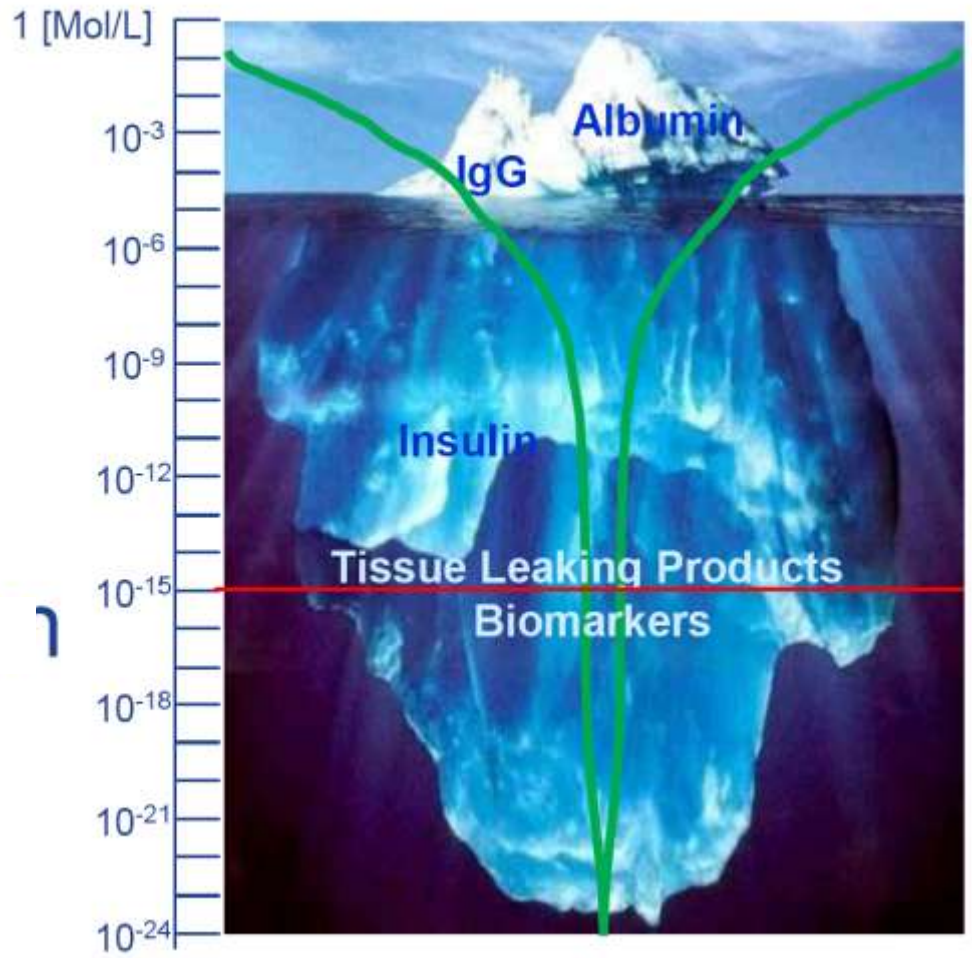
Detection of PROSTATE CANCER by Urine Proteomics

Vall d'Hebron Hospital

RESEARCH UNIT IN BIOMEDICINE AND TRANSLATIONAL AND PEDIATRIC ONCOLOGY

VALL D'HEBRON RESEARCH INSTITUTE AND HOSPITAL - BARCELONA

Why Urinary Proteomics?



DISADVANTAGES

- **Low protein concentration** (150mg in 24h urine)
- High levels of **salts** and/or other **interfering compounds**
- **High degree of variation** (intra-individual and inter-individual)
- **High complexity** of the sample
- High dynamic range
- High levels of **abundant protein** (albumin and IgG) → **DEPLETION/ENRICHMENT**

Urinary proteomics (DIGE-MS) a technical challenge

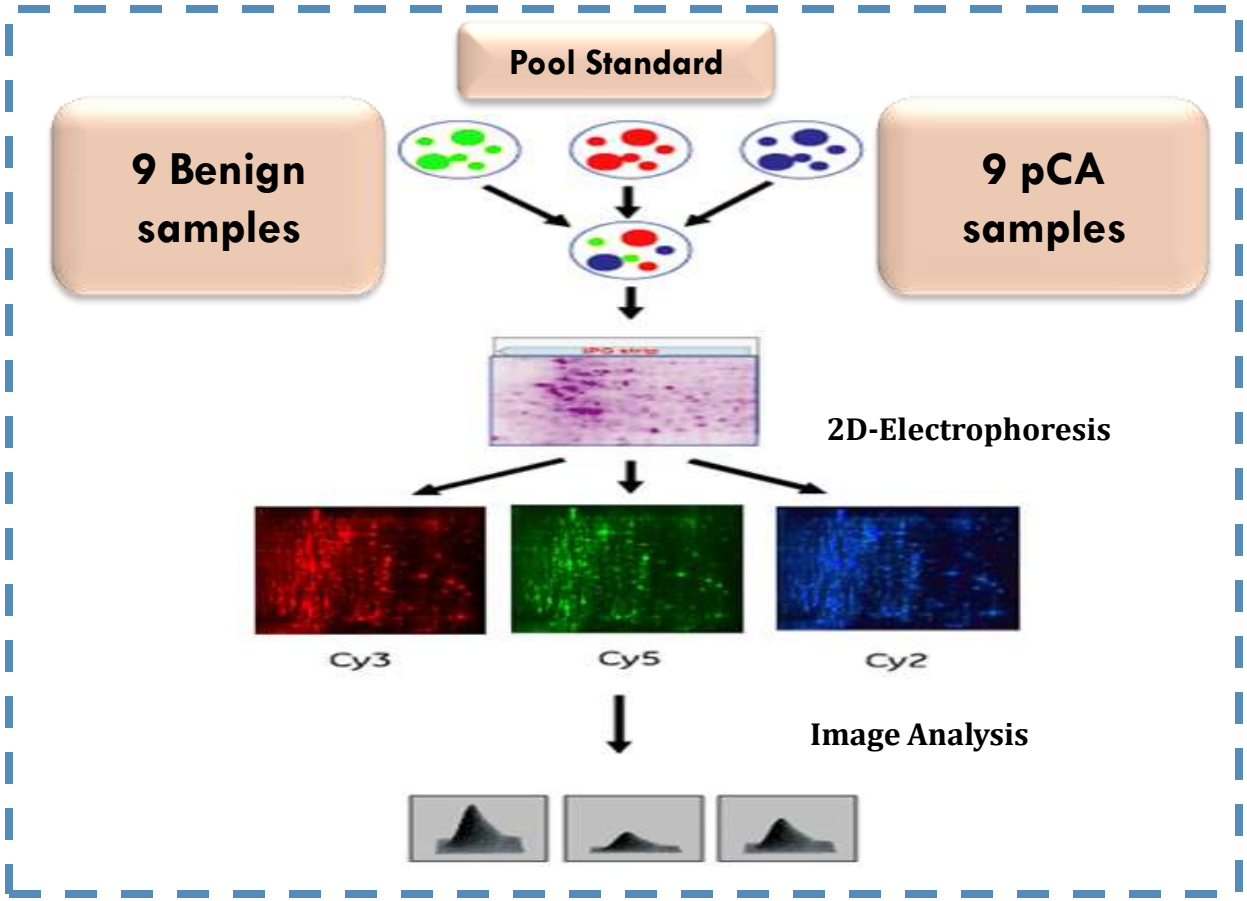
Comparative Proteomic Analysis

Proteins from urine supernatants

Differential In Gel Electrophoresis **(DIGE)**

- **1500 – 2000 proteins/gel**
- **Compare / quantify**
- **Compatible to MS**

MS/MS Mass Spectroscopy **(MALDI-TOF)**

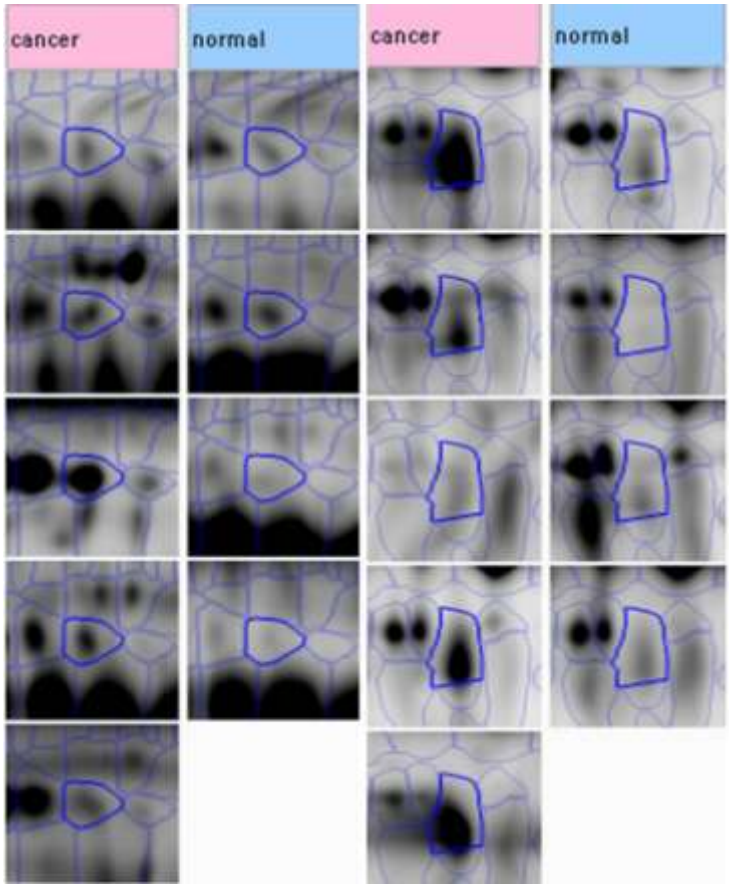


Protein Identification

Protein Validation SRM LC-MS (Ongoing)

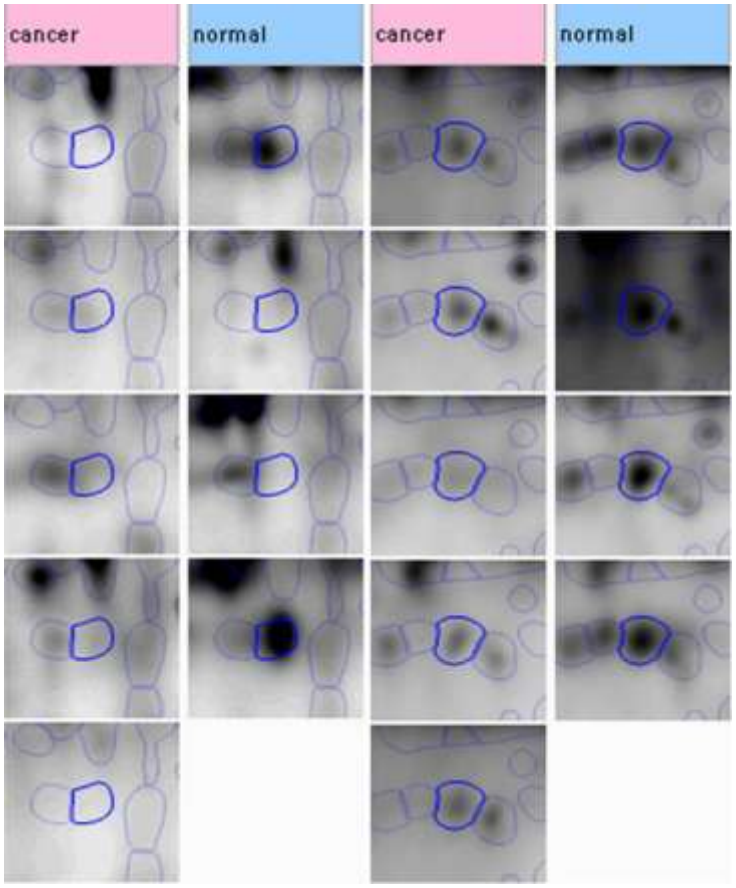
ELISA

Image Analysis



Overexpressed in cancer

(ANOVA < 0,05 and FOLD > 1,3)

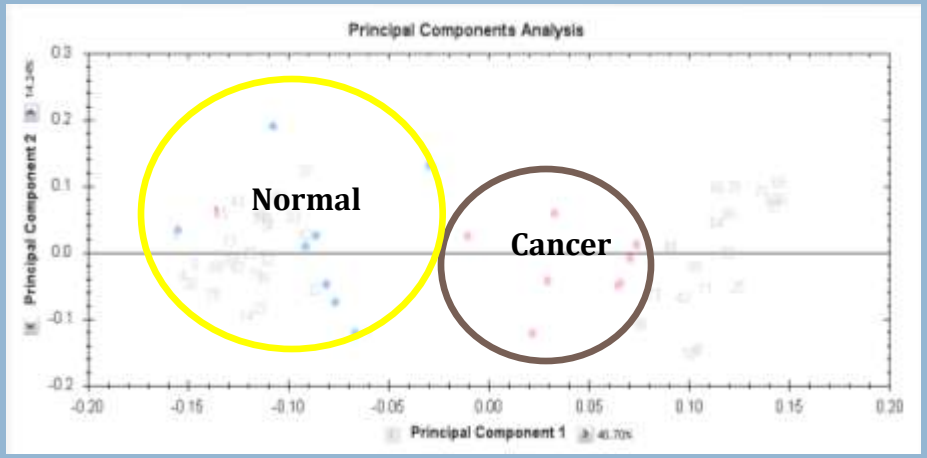


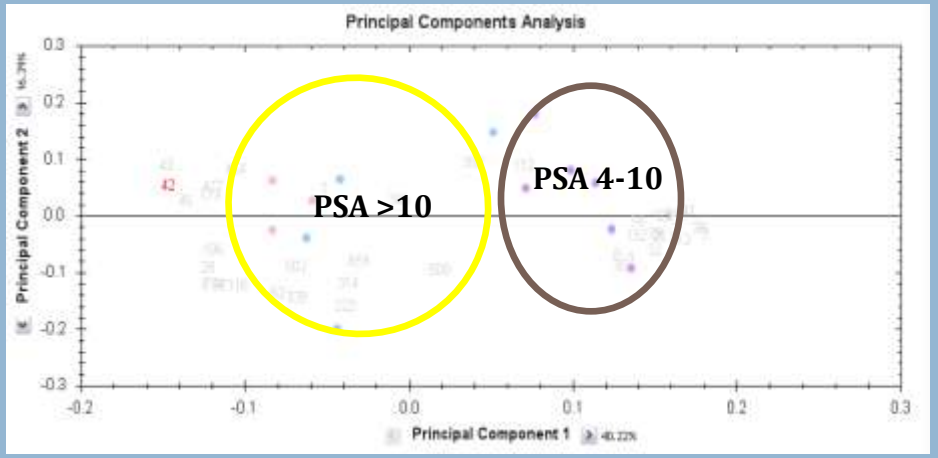
Underexpressed in cancer

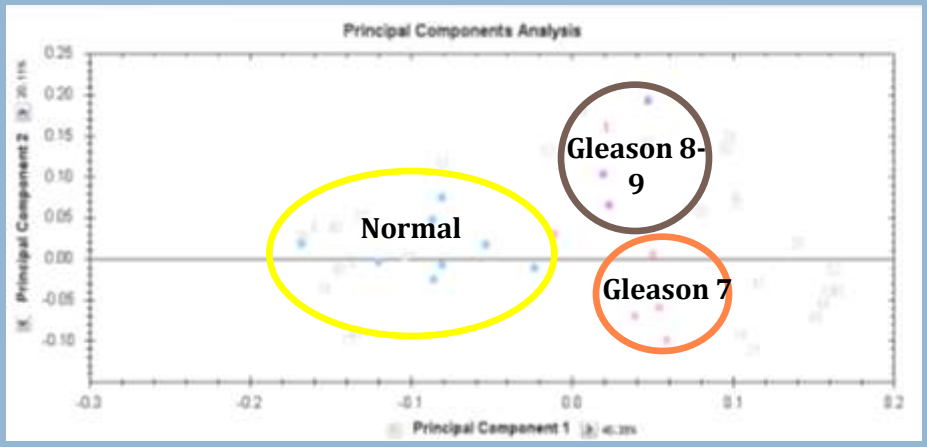
(ANOVA < 0,05 and FOLD > 1,3)

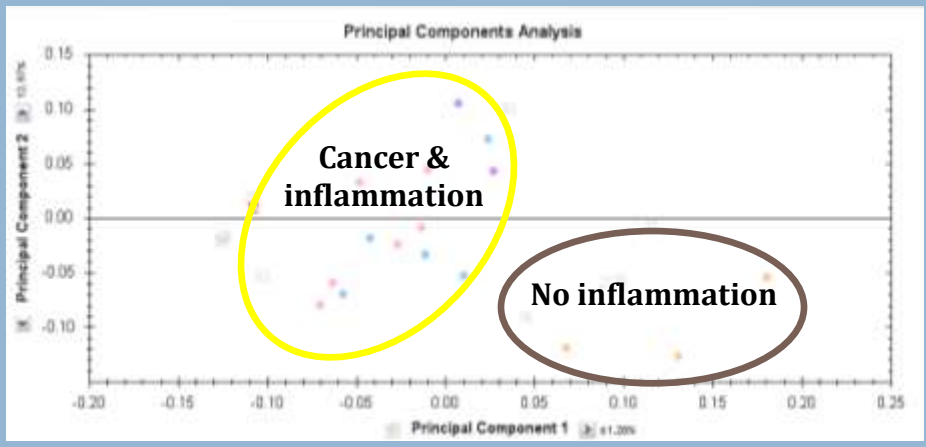
Statistical Data Analysis

Principal Component Analysis



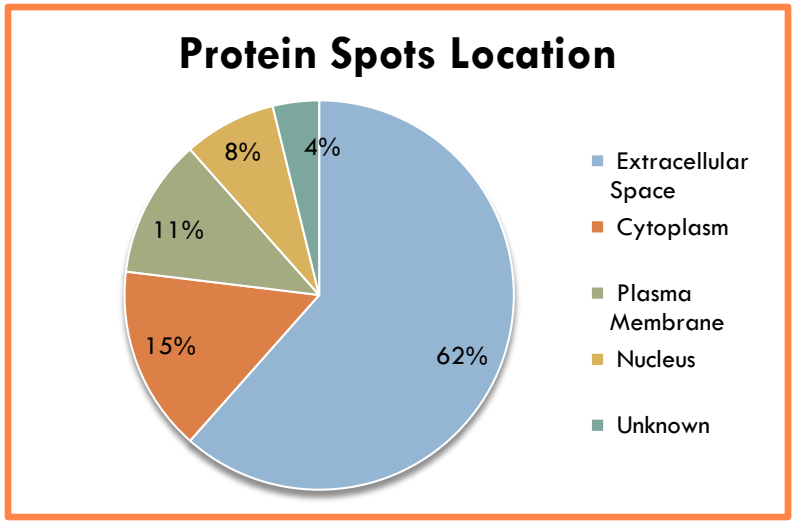


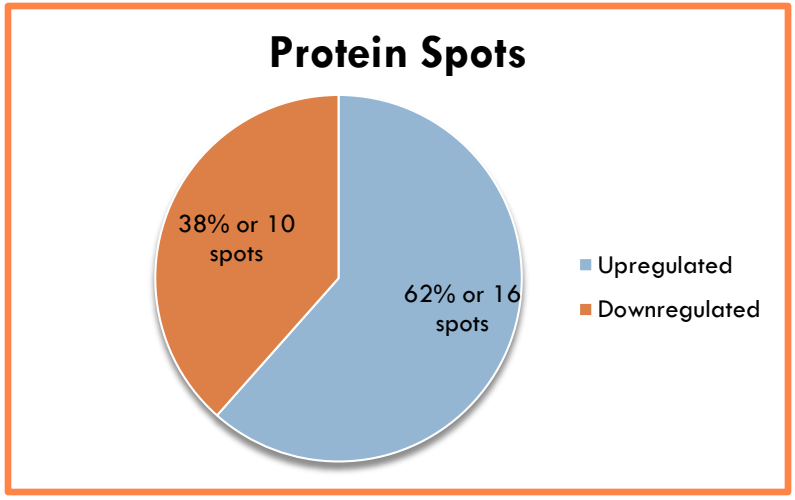




Identified Proteins

Fold	ID	Location	Type
-8,217	1	Extracellular Space	phosphatase
-6,130	PSA	Extracellular Space	peptidase
-5,758	3	Unknown	other
-3,351	4	Plasma Membrane	transporter
-3,351	5	Cytoplasm	other
-2,775	6	Extracellular Space	other
-2,686	7	Cytoplasm	enzyme
-2,683	8	Extracellular Space	other
-2,500	9	Extracellular Space	transporter
-2,493	10	Cytoplasm	transporter
-2,395	11	Unknown	other
-2,282	12	Plasma Membrane	other
-2,282	13	Extracellular Space	peptidase
-2,109	14	Extracellular Space	other
-1,804	15	Plasma Membrane	peptidase
-1,725	16	Nucleus	other
1,439	17	Extracellular Space	peptidase
1,479	18	Extracellular Space	enzyme
1,746	19	Extracellular Space	transporter
1,835	20	Extracellular Space	peptidase
1,850	21	Extracellular Space	transporter
1,990	22	Extracellular Space	other
2,289	23	Extracellular Space	other
2,845	24	Cytoplasm	transporter
2,845	25	Nucleus	other
3,037	26	Extracellular Space	enzyme



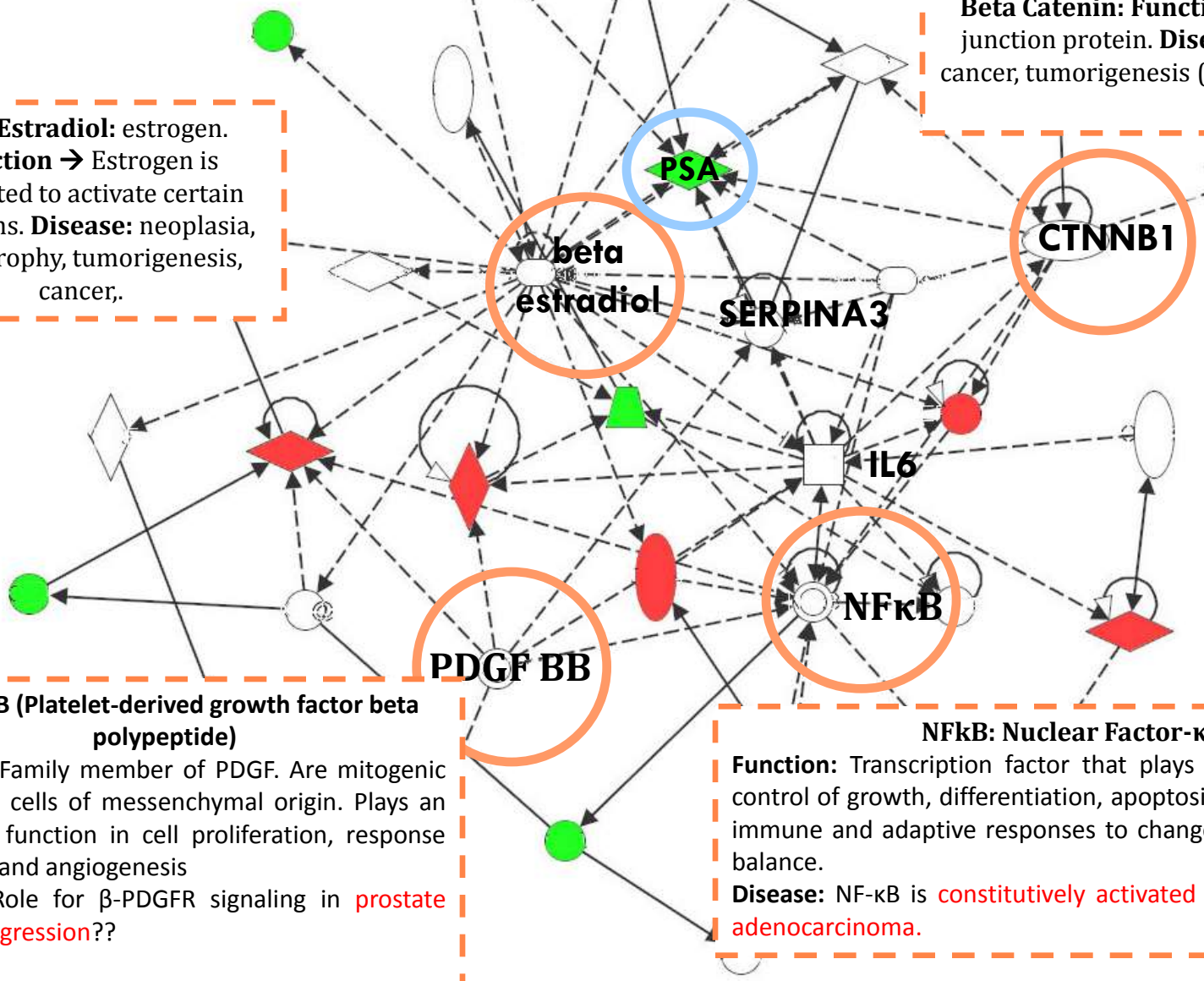


IPA-Network

PSA/KLK3 → peptidase. **Function**: proteolysis; negative regulation of angiogenesis. **Disease**: prostate cancer, breast cancer, breast carcinoma, neoplasia, cancer

Beta Catenin: Function → adherens junction protein. **Disease:** neoplasia, cancer, tumorigenesis (Prostate Cancer).

Beta Estradiol: estrogen. **Function →** Estrogen is suspected to activate certain Oncogens. **Disease:** neoplasia, hypertrophy, tumorigenesis, cancer,.



PDGF-BB (Platelet-derived growth factor beta polypeptide)

Function: Family member of PDGF. Are mitogenic factors for cells of messenchymal origin. Plays an important function in cell proliferation, response wounding and angiogenesis

Disease: Role for β-PDGFR signaling in prostate cancer progression??

NFkB: Nuclear Factor-κB

Function: Transcription factor that plays important roles in control of growth, differentiation, apoptosis and is involved in immune and adaptive responses to changes in cellular redox balance.

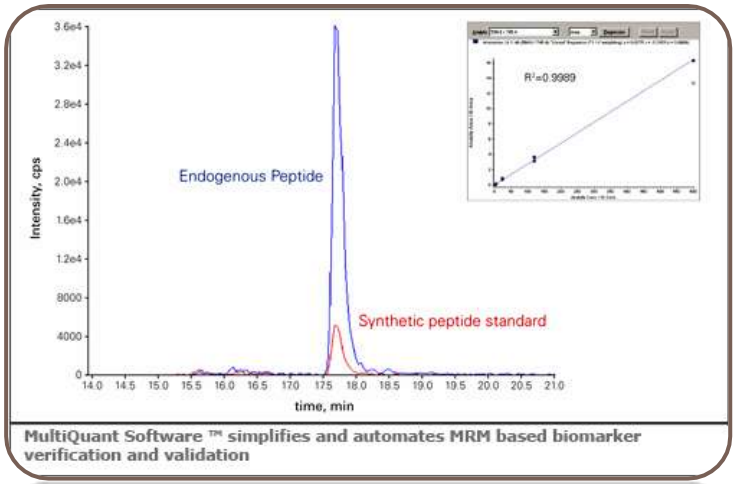
Disease: NF-κB is constitutively activated in human prostate adenocarcinoma.

Protein Validation by SRM (ongoing work)

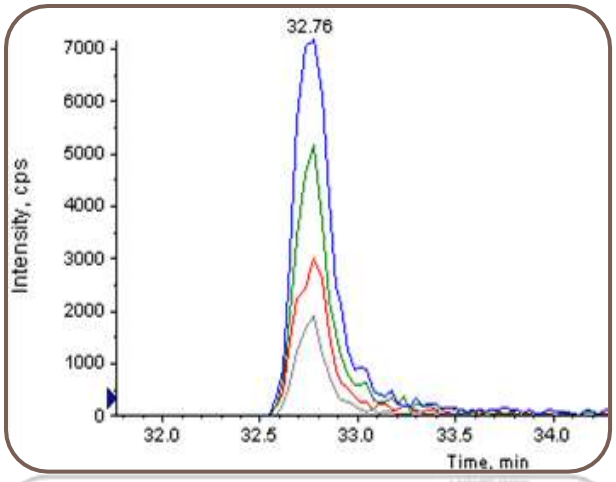
CANDIDATE BIOMARKER

Define "SIGNATURE PEPTIDES"

ABSOLUTE QUANTITATION



MultiQuant Software ™ simplifies and automates MRM based biomarker verification and validation



Selected Reaction Monitoring (SRM): Conducted by Liquid Cromatography mass spectometry (LC-MS)

- Enables direct quantification of proteins in complex samples
- Up to tens of candidates can be nearly simultaneously targeted and qunatified by detecting "Signature peptides"

- We identified **26 proteins** which are 16 under- and 10 overexpressed when comparing **urines** from **prostate cancer** patients against **healthy** age-matched individuals
- Urine proteomics can further distinguish between **PSA 4-10, and PSA >10**
- Urine proteomics can distinguish between **Gleason 8-9, Gleason 7 and Normal** age matched individuals.
- Most of the identified proteins are involved in **pathways associated with cancer or inflammation**
- **Biomarkers validation** by **SRM -** based assay

These data demonstrate the ability of **proteomic analysis** to reveal **potential biomarkers for PCa in urine.**

CONCLUSIONS

Detection of PROSTATE CANCER by building an odorant nanosensor ("intelligent nose")

Three different strategies are being followed by our research teams:

- Urine metabolomics: Identification of volatile products in the urine of Pca patients, and establishment of a odorant nano-sensor (intelligent nose).

THESE DOGS MIGHT SAVE YOUR LIFE

Can sensitive canine noses detect cancer even before your doctor can? Researchers working with these four-legged lab partners think it's possible

BY JOHNNY DODD

"They all have different working styles," McCulloch says of his team (which includes Majestic, Freeman and Divine).

On the floor in Michael McCulloch's research lab is a row of five identical plastic specimen boxes: One contains a breath sample from a woman with ovarian cancer; four are controls from healthy women. Each box is weighted with concrete. Why the concrete? "Some of the dogs come bursting in and make

For the past 10 years McCulloch—the director of research at San Anselmo, Calif.'s Pine Street Foundation, which studies integrative medicine—has been exploring whether dogs can reliably detect the disease. In 2003 "we trained dogs to smell lung cancer 99 percent of the time and breast cancer 88 percent of the time." Now partnered with the Uni-

has a high 5-year survival rate when detected and treated at a localized stage, but is not often diagnosed that early.

To make the team, each dog had to correctly identify a sample 30 consecutive times. Freeman, a 6-year-old black Lab, is the standout. "He takes his time, walking the entire line before he shows us where it is. He's very con-

Tessy analyzes breath samples in the lab.

> "We're starting to learn that your dog probably knows more about your health than you know about theirs" —MICHAEL McCULLOCH

But these aren't wonder dogs bred to work in labs; they are family pets whose owners volunteer them one or two days a week. Anecdotally, it seems most dogs can sense illness, says retired Tallahassee police-dog trainer Duane Pickle. Anything that "smells different

puncturist, first got the idea when he was studying Chinese medicine. "A text from the third century B.C. mentions how liver disease causes a change in body odor." Then he saw a 1989 report in the medical journal *Lancet* that told of a young woman whose

he could re-create that phenomenon in a lab. To teach the Pine Street dogs their task, he initially combines the known cancer samples with dog food, while leaving healthy samples untouched. Once the dogs understood what they were looking for, they could pick out the cancer scent without a food hint.

So does cancer have a smell? What McCulloch thinks the dogs are detecting is metabolic waste "from the tumor cells, which is chemically different from normal cells. The waste travels through the bloodstream and is exhaled out through the lungs."

Since news of his study broke, McCulloch has received dozens of letters from people who say their pets saved their lives by drawing attention to undiagnosed cancer (see box). Even mainstream medicine is taking note. "An enormous amount of research is being done to find those proteins present in small quantities in the bloodstream that may signal cancer," says Dr. J. Leonard Lichtenfeld, the American Cancer Society's deputy chief medical officer. "That a dog could smell these is definitely within the realm of possibility."

The dogs' part will end in December, and McCulloch's data will begin to be analyzed. Eventually, it is possible dogs may be used to find cancer in its early stages. For now, McCulloch says his findings offer some insight into canine behavior: "When you see dogs on the

THE DIACPROL PROJECT: Building a nanosensor ("electronic nose") for prostate cancer diagnosis

Non-invasive olfactory nanobioplatform for human disease diagnosis and monitoring.

Odors emitted by the human body, by its secretions and exhalations depend both on its genetic signature and on its physio-pathological status. As a matter of fact, some medical practices used or are still using sensorial diagnosis (mainly olfactory) to check the good health or to determine various pathologies (phenylcetonuria, typhoïd, scorbut, etc), and the potential interest of dogs for the diagnosis of some melanoma was recently pointed out

Pickel, D., Manucy, G. P., Walker, D. B., Hall, S. B. & Walker, J. C. (2004) Evidence for canine olfactory detection of melanoma, Applied Animal Behaviour Science.89, 107.

Willis, C. M., Church, S. M., Guest, C. M., Cook, W. A., McCarthy, N., Bransbury, A. J., Church, M. R. T. & Church, J. C. T. (2004) Olfactory detection of human bladder cancer by dogs : proof of principle study, British Medical Journal. 329, 712-717

Another sniffer dog for the clinic?, The Lancet (2001) . 358, 930.

In vitro Diagnosis

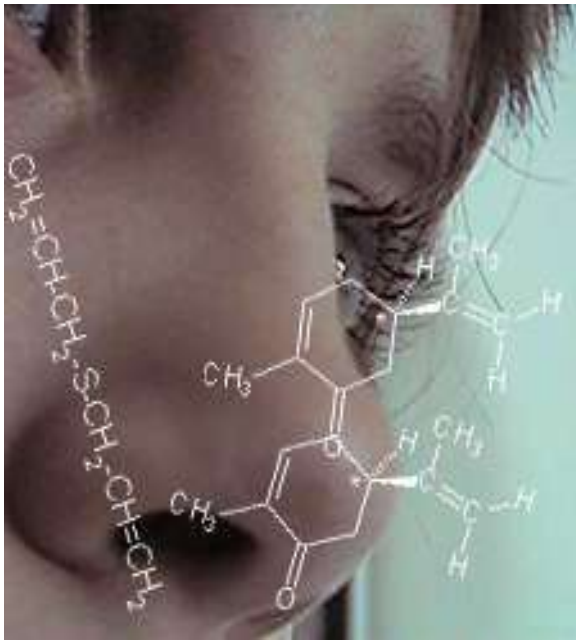
An in-vitro diagnostic tool can be a single biosensor, or an integrated device containing many biosensors. A biosensor is a sensor that contains a biological element, such as an enzyme, capable of recognising and 'signalling' (through some biochemical change) the presence, activity or concentration of a specific biological molecule in solution. A transducer is used to convert the biochemical signal into a quantifiable signal. Key attributes of biosensors are their specificity and sensitivity. Nanoanalytical tools like scanning probe microscopy or imaging mass spectrometry offer new opportunities for in-vivo diagnostics, like molecular pathology or reading out highly integrated ultra-sensitive biochips.

Table 1 | **Summary of key volatiles associated with different disease types analysed by GC–MS**

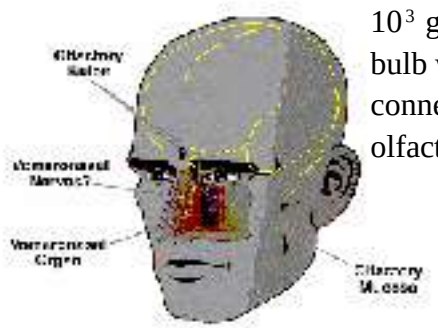
Sample	Disorder/Infection	Volatile compounds	References
Microorganism-associated disorders			
Urine	Urinary tract infection	Isovaleric acid, alkanes	28
Intraperitoneal fluid	Aerobic Gram-negative bacteria	Terpenes, ketones	29
Intraperitoneal fluid	Anaerobic bacterial infections	Acetic, butyric acids	30
Human pus	–	Isobutyric, isovaleric, isocaproic acids	31
Other disorders			
Human breath	Breast cancer	Alkanes, monomethylated alkanes	32
Human breath	Lung cancer	Alkanes, monome anes	33
Human breath	Acute asthma	Pentane	34
Urine	Metabolic disorders	Isovaleric acid	35
Alveolar air	Hepatic coma	Methyl-mercaptan	36
Alveolar air	Rheumatoid arthritis	Pentane	37
Alveolar air	Schizophrenia	Pentane, carbon disulphide	38
Alveolar air	Ketosis	Acetone	39
Alveolar air	Cardiopulmonary disease	Acetone, ethanol	40
Blood plasma, cerebrospinal fluid	Hepatic encephalopathy	3-methylbutanol	41

Overall Aim

- **Develop a bio-electronic olfactory sensor based on the electrical properties of single olfactory receptors**

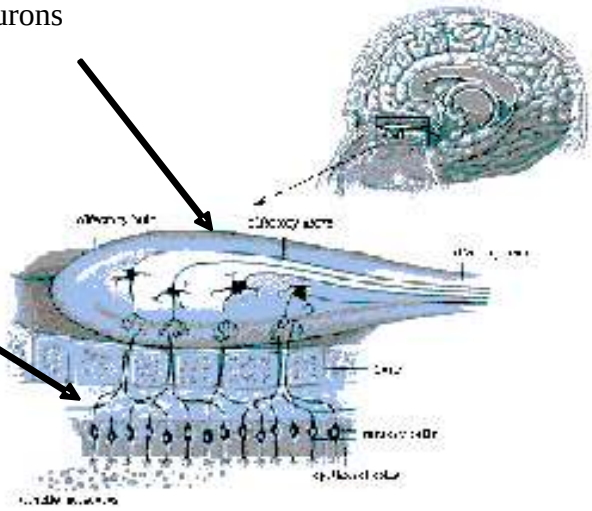


Biological Nose



10^3 glomeruli in the bulb which receive the connections of 25000 olfactory neurons

10^8 neurons in the piriform cortex



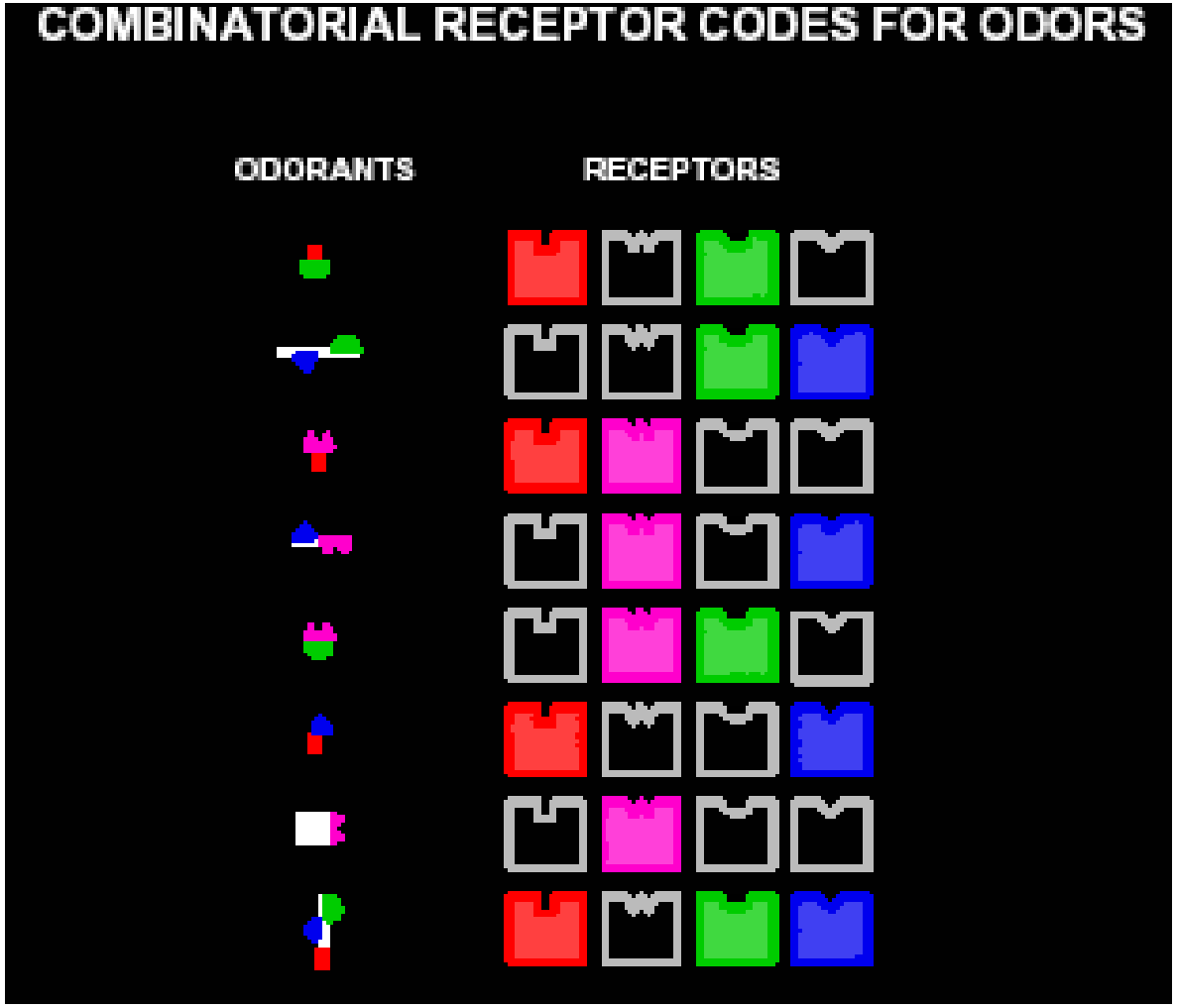
10^7 receptor cells

10^3 different types

Each type display a unique profile of sensitivity to a battery of odorants

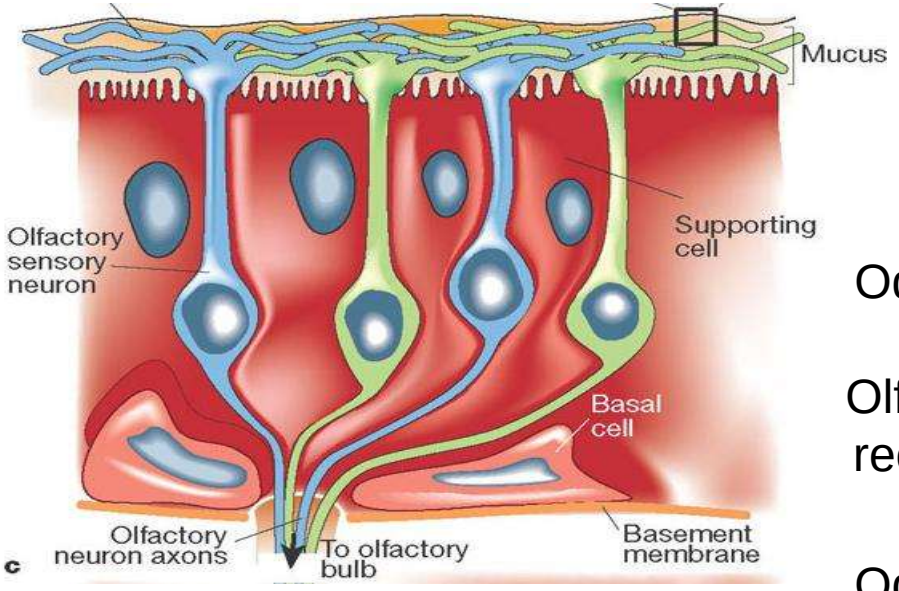
Nobel Prize 2004

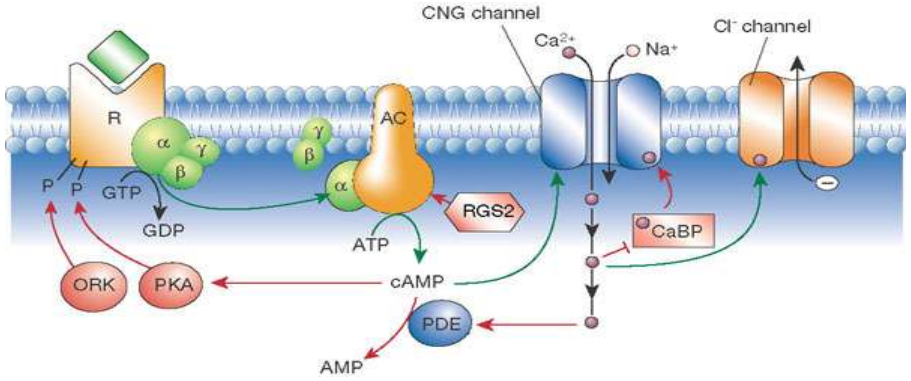
Linda Buck



Richard Axel

OLFACTORY RECEPTORS



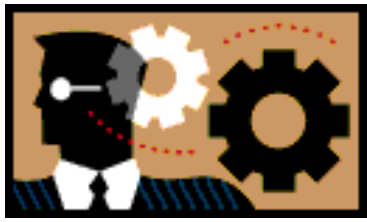


Natural odorant identification

Odorant + Olfactory receptor + Many chemical processes → Electric signal + Brain → Odorant identification

Artificial odorant indentification

Odorant + Olfactory receptor → Electric signal + Processing → Odorant identification



olfactory receptor

odorant

physiological medium

electrode

R

I

Amp

vo

output

vi

input

pre-amplifier

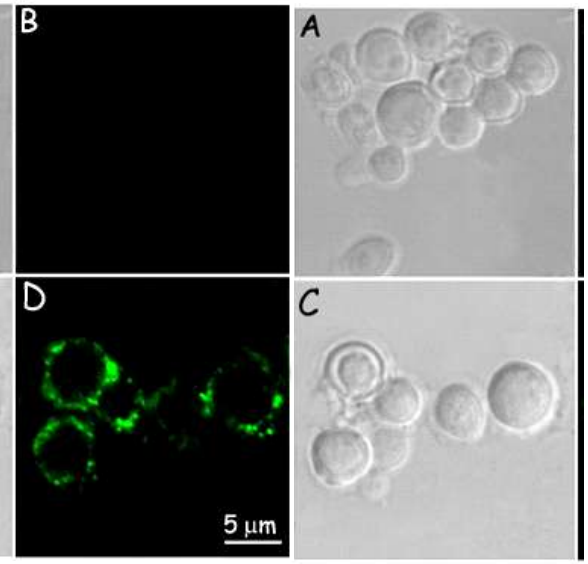
output

with odorant

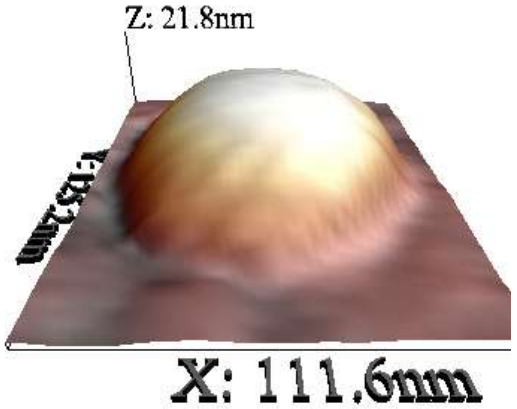
whitout odorant

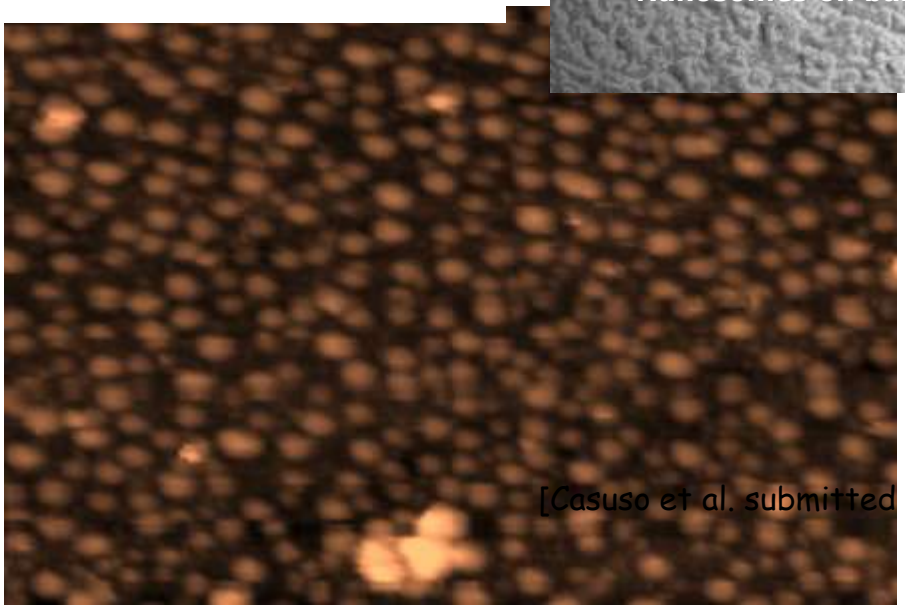
input

Bioelectronic Sensor



The Olfactory Receptors





[Casuso et al. submitted to Biosens. Bioelectron.]

Strategies of analysis

1. Volatile compounds

Table 3. Peak areas of hexanal and 1-octen-3-ol in 19 liver cancer patients and 18 normal ones

Patient No.	Peak area		Normal No.	Peak area	
	Hexanal	1-Octen-3-ol		Hexanal	1-Octen-3-ol
1	2760059	418158	1	0	0
2	2806264	129048	2	0	0
3	0	1031933	3	0	0
4	3495954	449983	4	0	0
5	2210171	170786	5	0	0
6	2854045	398115	6	0	0
7	2368412	291152	7	0	0
8	2985494	408808	8	1173333	57998
9	5165802	232646	9	194947	0
10	4813377	1184808	10	0	0
11	1651183	0	11	0	0
12	2982424	370943	12	816748	0
13	9068700	861267	13	0	0
14	0	2250239	14	0	0
15	4649721	782300	15	716832	0
16	3291170	2338376	16	0	0
17	4230053	636561	17	0	0
18	7135117	776768	18	265436	0
19	14033917	801794			

RAPID COMMUNICATIONS IN MASS SPECTROMETRY
Rapid Commun. Mass Spectrom. 2008; 22: 1301–1306
Published online in Wiley InterScience (www.interscience.wiley.com) DOI: 10.1002/rcm.3484

RCM

Investigation of volatile biomarkers in liver cancer blood using solid-phase microextraction and gas chromatography/mass spectrometry

Ruyi Xue[1†], Ling Dong[1†], Si Zhang[2], Chunhui Deng[3*], Taotao Liu[1], Jiyao Wang[1] and Xizhong Shen[1**]

[1]Zhongshan Hospital, Shanghai Medical College, Fudan University, Shanghai 200032, China
[2]Gene Research Center, Shanghai Medical College, Fudan University, Shanghai 200032, China
[3]Department of Chemistry, Fudan University, Shanghai 200433, China

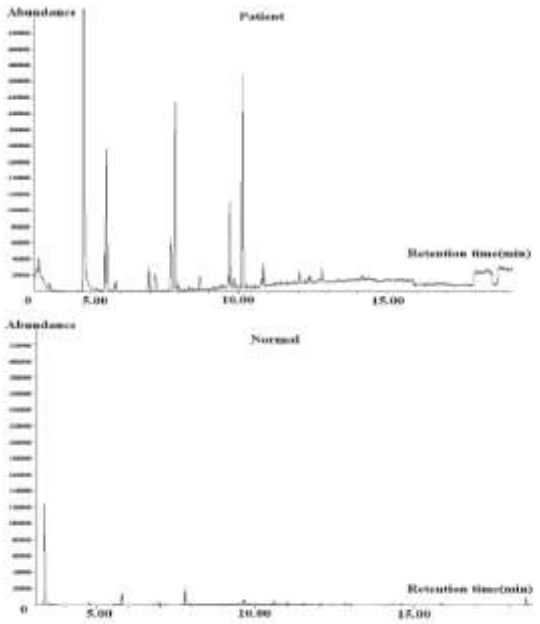
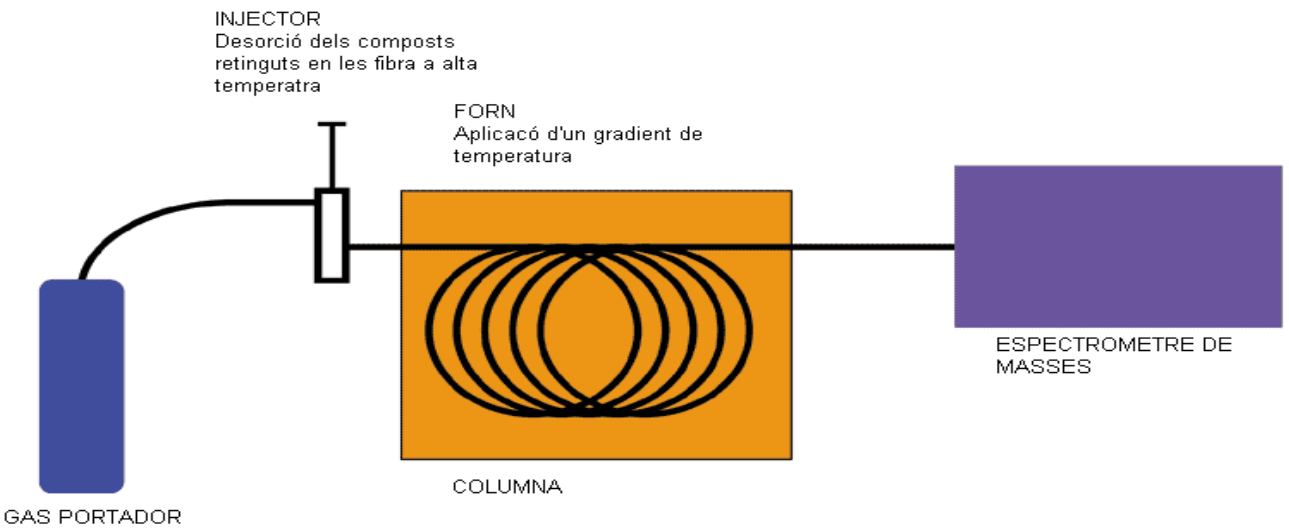


Figure 2. SPME-GC/MS TICs of volatile compounds in a liver cancer blood (52, male) (patient) and a normal blood (53, male) (normal). Extraction was performed at 60°C for 15 min using a CAR–PDMS fiber; a desorption temperature of 250°C and a time of 30 s were used.

Measurement method



sample treatment

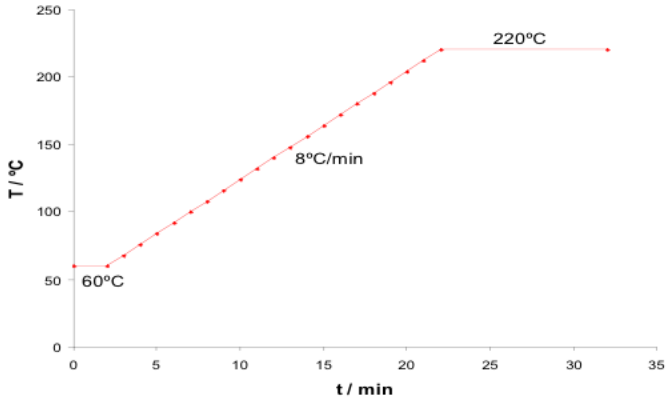
_ Neutral sample pretreatment

3ml urine + 2g NaCl

_ Basic sample pretreatment

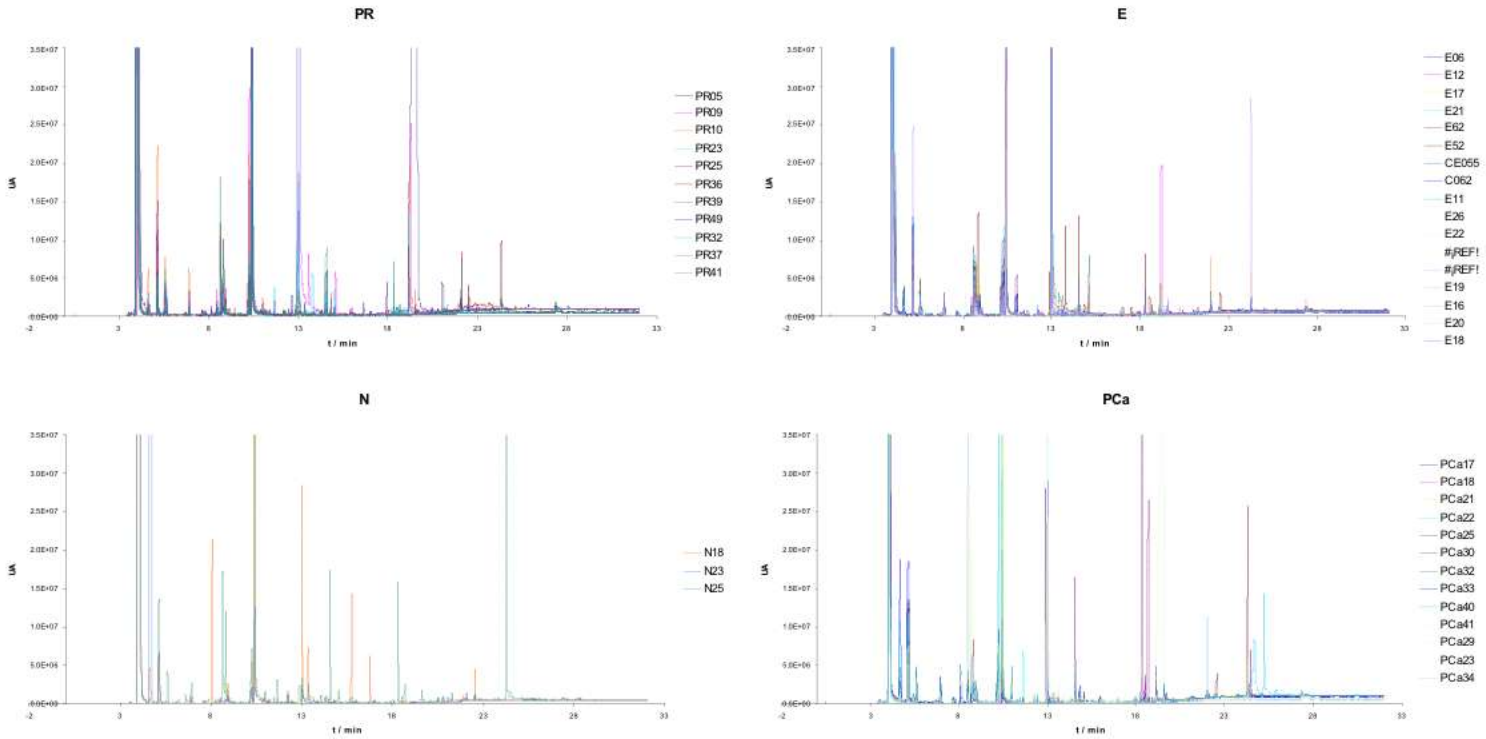
3ml urine + 2.25g K_2CO_3 + 1 KCl lentil

_ GC/MS conditions



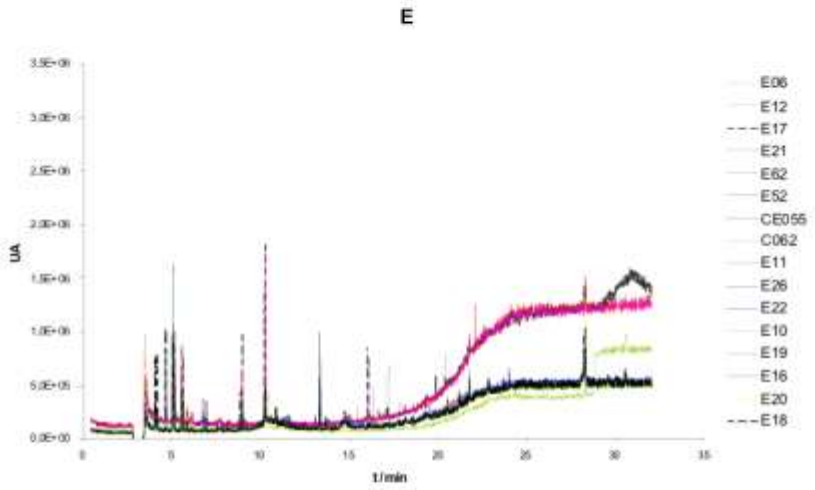
results

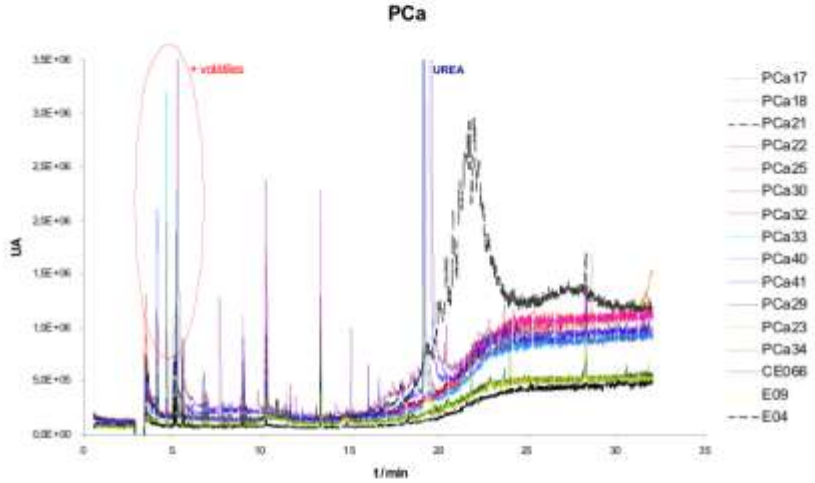
Gas chromatograms at basic pH



results

_ Gas chromatograms at neutral pH

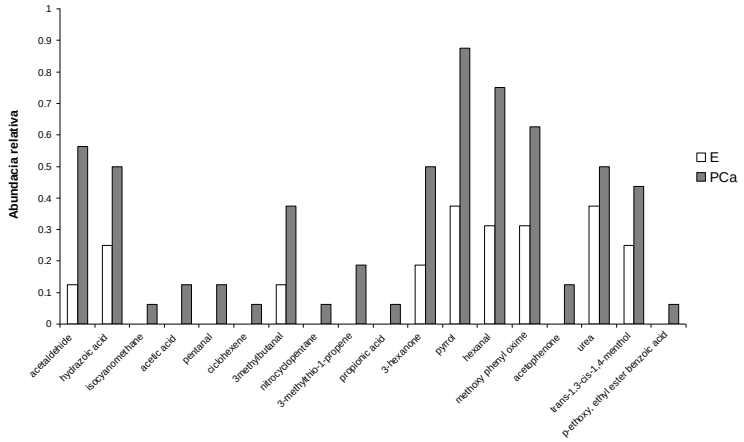


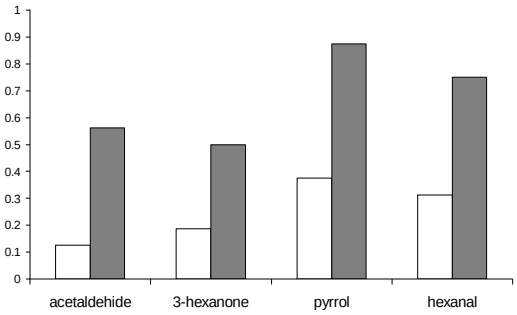


t / min	compuesto	Promedio Área Pca	Promedio Área E	Incremento / %
3.9	acetaldehide	1.48E+06		
4.11	methanethiol	2.71E+06	7.31E+05	271
4.64	ethanol	5.63E+06	2.54E+06	122
5.12	acetone	3.70E+06	2.47E+06	49.8
5.57	2-methyl-2-propanol	1.64E+06	1.43E+06	14.7
6.7	2-ethoxy-2methylpropane	7.34E+05	4.45E+05	64.9
6.91	2-butane	3.96E+05	3.84E+05	3.13
19.46	urea	2.47E+08		

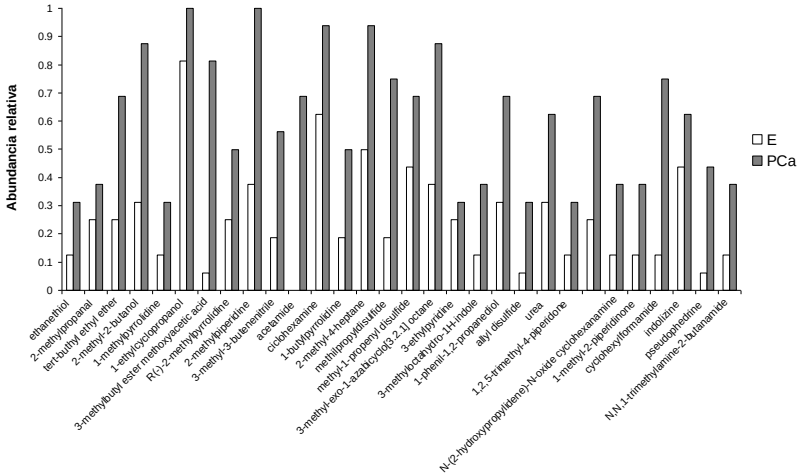
conclusions

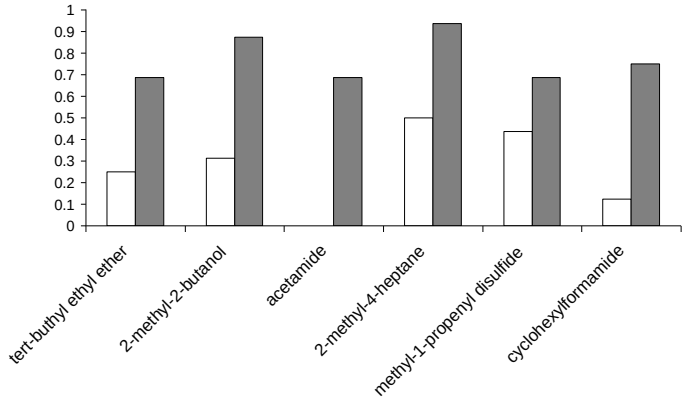
pH neutral





pH basic





conclusions

· **Fingerprint compounds**

pH neutre	pH basic
	tert butylethyl ether
acetaldehide	2-methyl 2-butanol
3-hexanone	2-methyl 4-heptane
pyrrol	acetamide
hexanal	methylpropyldidulfide
	cyclohexylformamide

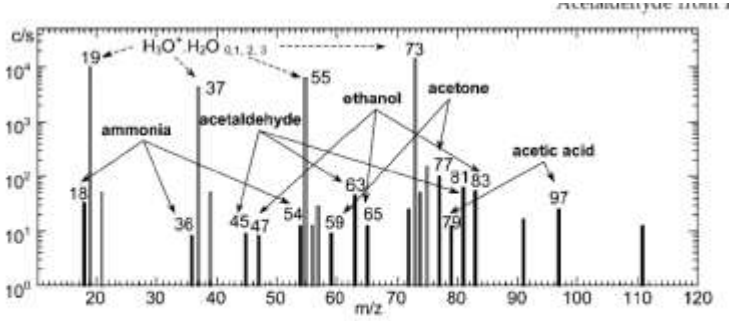


Figure 2. SIFT-MS full-scan mode (FSM) spectra obtained from the headpace above a CALU-1 cell culture (80 million cells, 4.5 g/L glucose in the medium).

RAPID COMMUNICATIONS IN MASS SPECTROMETRY
Rapid Commun. Mass Spectrom. 2003; **17**: 845–850

Smith et al.

· ↑ **VOCs at neutral pH**

Compound	Area increase / %
acetaldehide	appears
methanethiol	271
ethanol	122
acetone	49.8
2-methyl-2-propanol	14.7
2-ethoxy-2methylpropane	64.9
2-butane	3.13
urea	appears

Papers

Olfactory detection of human bladder cancer by dogs: proof of principle study

Carolyn M Willis, Susannah M Church, Claire M Guest, W Andrew Cook, Noel McCarthy, Anthea J Bransbury, Martin R T Church, John C T Church

Abstract

Objective To determine whether dogs can be trained to identify people with bladder cancer on the basis of urine odour more successfully than would be expected by chance alone.
Design Experimental, "proof of principle" study in which six dogs were trained to discriminate between urine from patients with bladder cancer and urine from diseased and healthy controls and then evaluated in tests requiring the selection of

Although these anecdotal events remain unsupported by experimental evidence, the concept that dogs can "smell" cancer is not unreasonable. Tumours produce volatile organic compounds, which are released into the atmosphere through, for example, breath and sweat.[5-8] Some of these volatile organic compounds are likely to have distinctive odours; even when present in minute quantities, they could be detectable by dogs, with their exceptional olfactory acuity.[9-11]

Integrative Cancer Therapies
Volume 7 Number 2
June 2008 76-80

10.1177/1534735408319058
http://ict.sagepub.com
hosted at
http://online.sagepub.com

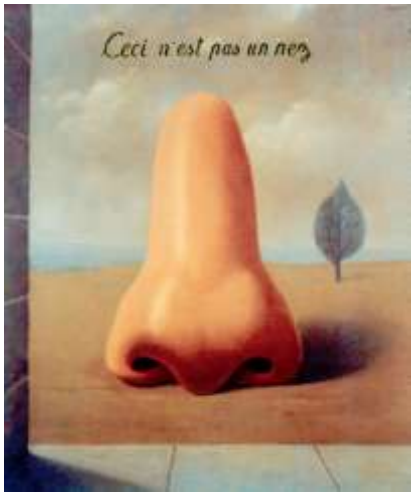
Human Ovarian Carcinomas Detected by Specific Odor

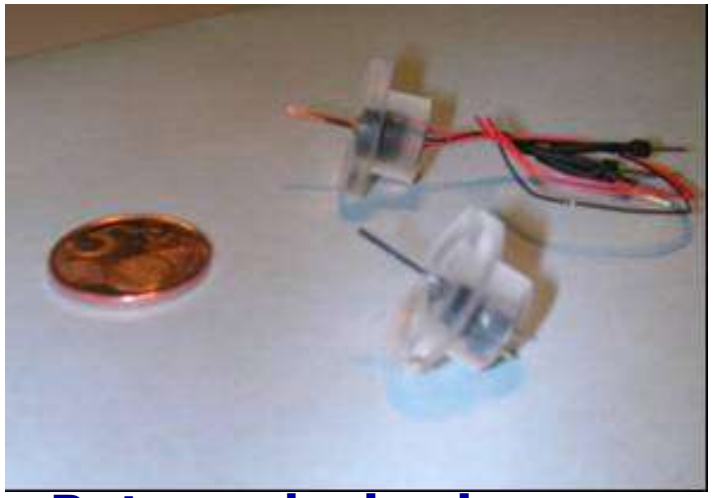
György Horvath, Gunvor af Klinteberg Järverud, Sven Järverud, István Horváth

The high mortality rate associated with ovarian carcinoma is mainly owing to late diagnosis. It is thus essential to develop inexpensive and simple methods for early diagnosis. Papers on canine scent detection of malignancies such as melanoma and bladder, lung, and breast cancer have recently been published in peer-reviewed journals, indicating a new diagnostic tool for

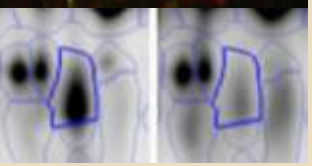
Using our training method, we taught a dog to distinguish different histopathological types and grades of ovarian carcinomas, including borderline tumors, from healthy control samples. Double-blind tests showed 100% sensitivity and 97.5% specificity. Moreover, the odor of ovarian carcinomas seems to differ from those of other gynecolog-

Josep Samitier, Elena Martínez, Marta Poch, Eva Alvárez de Eulate.
Institut de Bioenginyeria de Catalunya, Universitat de Barcelona.





But our device is near…



UNITAT DE RECERCA
BIOMÈDICA i D'ONCOLOGIA
TRANSLACIONAL I
PEDIÀTRICA

ACKNOWLEDGMENTS

Marina Rigau
Marta Garcia
Eva Colás
Marta Llauradó
Núria Pedrola
Hafid Alazzouzi
Anna Ruiz
Andreas Doll
Jaume Reventós

SERVEI DE PROTEÒMICA
Francesc Canals
Núria Colomé
Marta Monge

SERVEI DE UROLOGIA
Joan Morote Robles
Mª Carme Mir
Carlos Ballesteros

SERVEI D'ANATOMIA PATOLÒGICA
Inés De Torres
Estefania Landolfi
Santiago Ramón y Cajal

UCTS
Mariangeles Artaza

PERSONAL QUIRÒFAN URO
Mª Dolors Argás
Luis Ortega

CRP-SANTÉ-UNIVERSITÉ DE LUXEMBOURG
Bruno Domon
Elodie Duriez

GRANT SUPPORT

Vall d'Hebron Institut de Recerca
VHIR

Agència de Gestió d'Ajuts Universitaris i de Recerca

Generalitat de Catalunya
Departament d'Innovació, Universitats i Empresa

ASOCIACIÓN ESPAÑOLA DE UROLOGÍA

MARIE CURIE ACTIONS

PEOPLE

EMBO

FFC

Instituto de Salud Carlos III

MINISTERIO DE CIENCIA E INNOVACIÓN

Fonds National de la Recherche Luxembourg

Transbiomed

RTICC