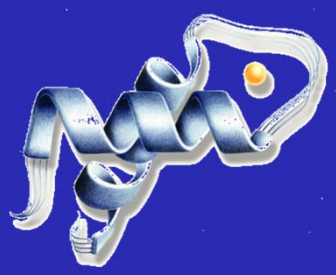


Tumor markers: principles and their clinical use

Newly diagnosed cases of cancer in Hungary, 2011

Total number	77421
Males	38998 (50.4%)
Females	38440 (49.6%)

Factors predicting chances of survival

- Early recognition
- Biochemical features: proliferation, invasion, metastasis

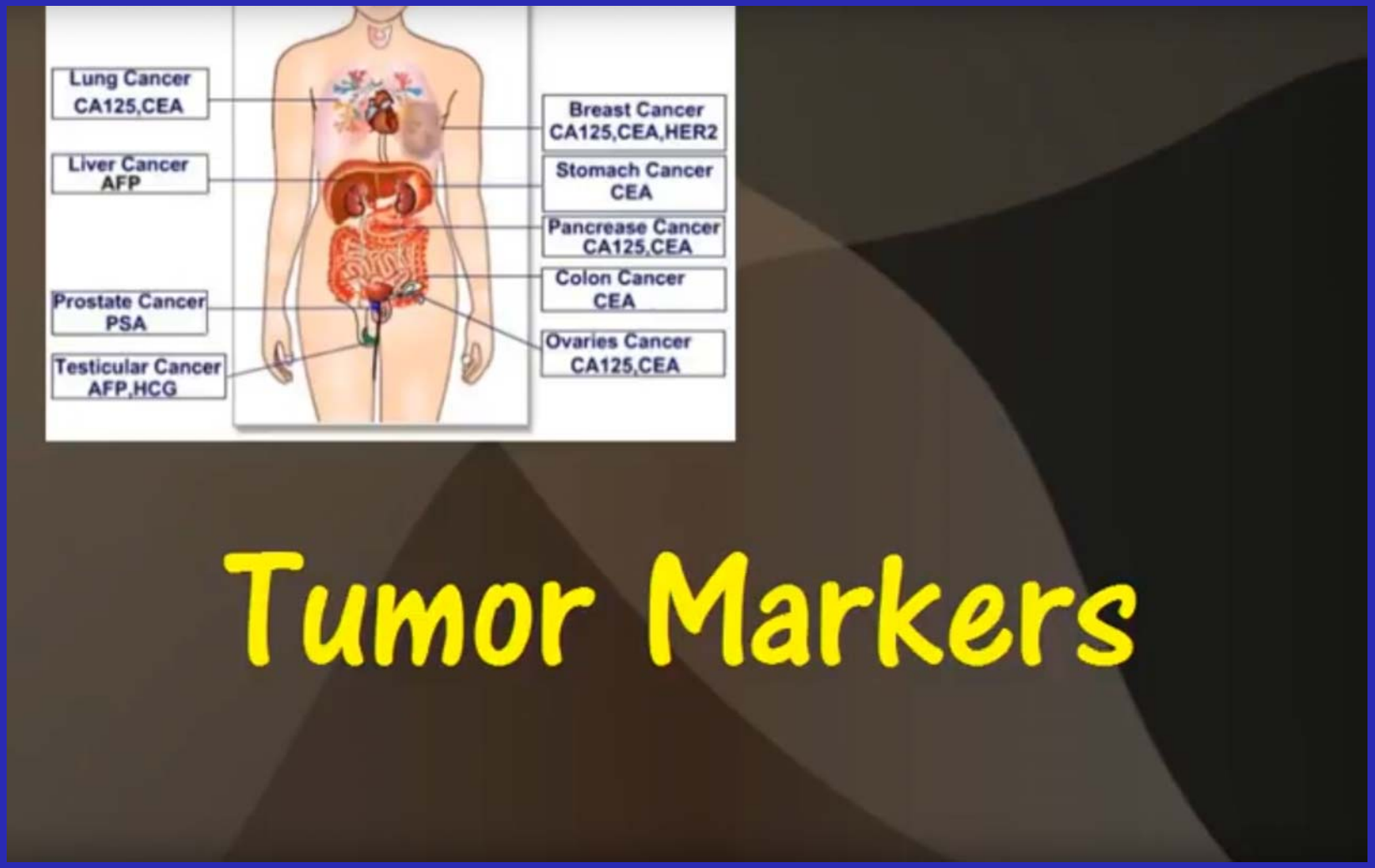
↓

Pathology, laboratory

↓

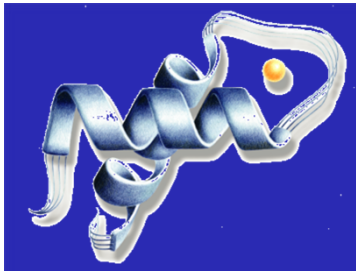
Efficient individual therapy

https://www.youtube.com/watch?v=SyhXLaPHxXk



https://www.youtube.com/watch?v=Nz2YGL4NWEE

https://www.youtube.com/watch?v=46Xh7OFkkCE



Clinical tasks to be fulfilled by biomarkers in oncology

1. Diagnosis, early detection
2. Prognosis (survival)
3. Prediction of response to adjuvant / palliative therapy
4. Monitoring of therapeutic response

An ideal Tumor marker

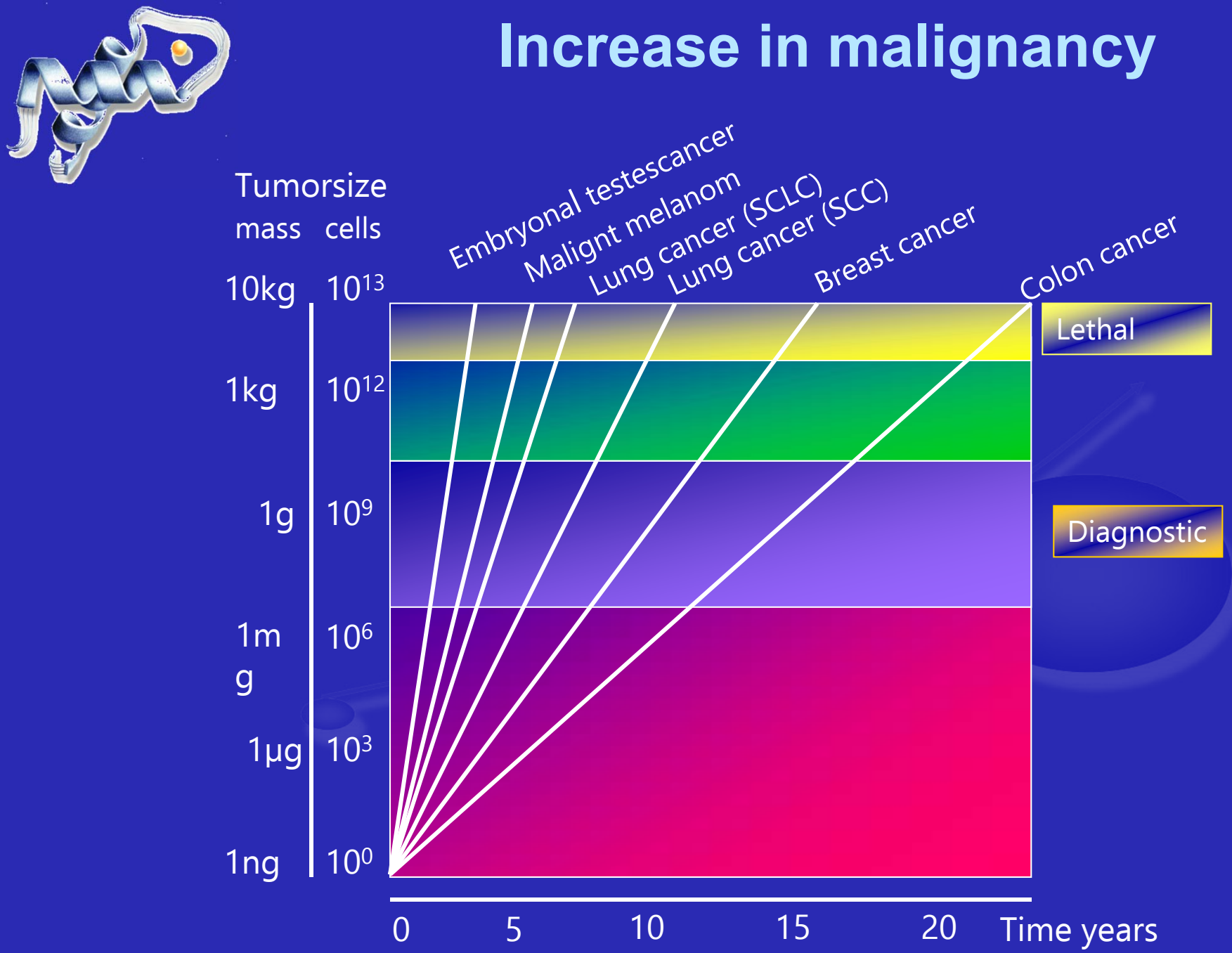
- Indicates early phase
- Specific for the tumor
- No interference with other disorders
- Easy to measure
- Prognostic & predictive
- May be caused by non-malignant disorders

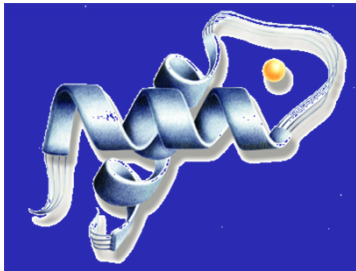
- …and does not exist (still)

Early recognition

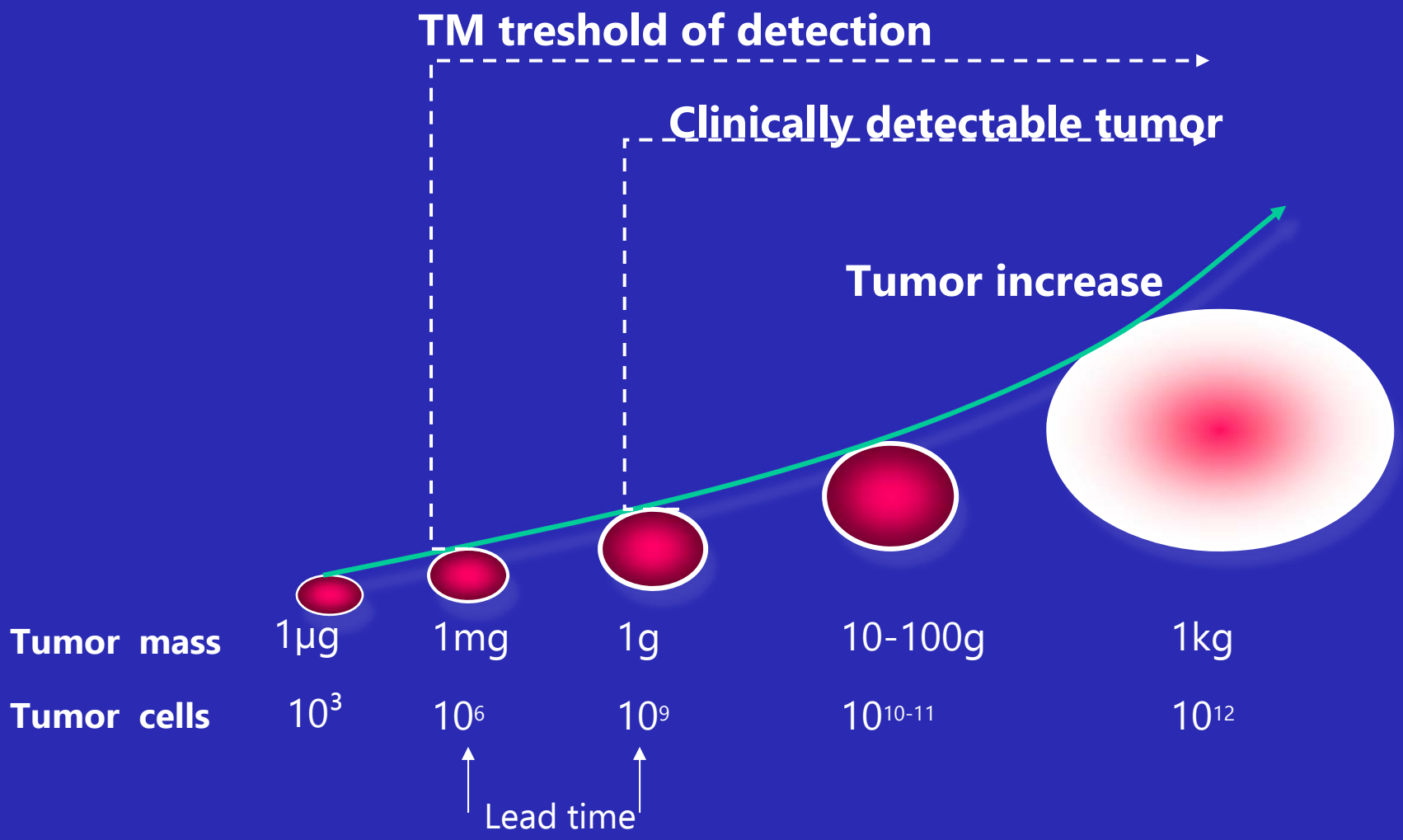
- Lab does not help (usually)
- When tumor enters into the blood it is not an early stage
- General signs and symptoms: anaemia, low iron stores, blood in excretions
- Fecal blood test; urinary blood test – early warning signs
- May be caused by non-malignant disorders

Increase in malignancy



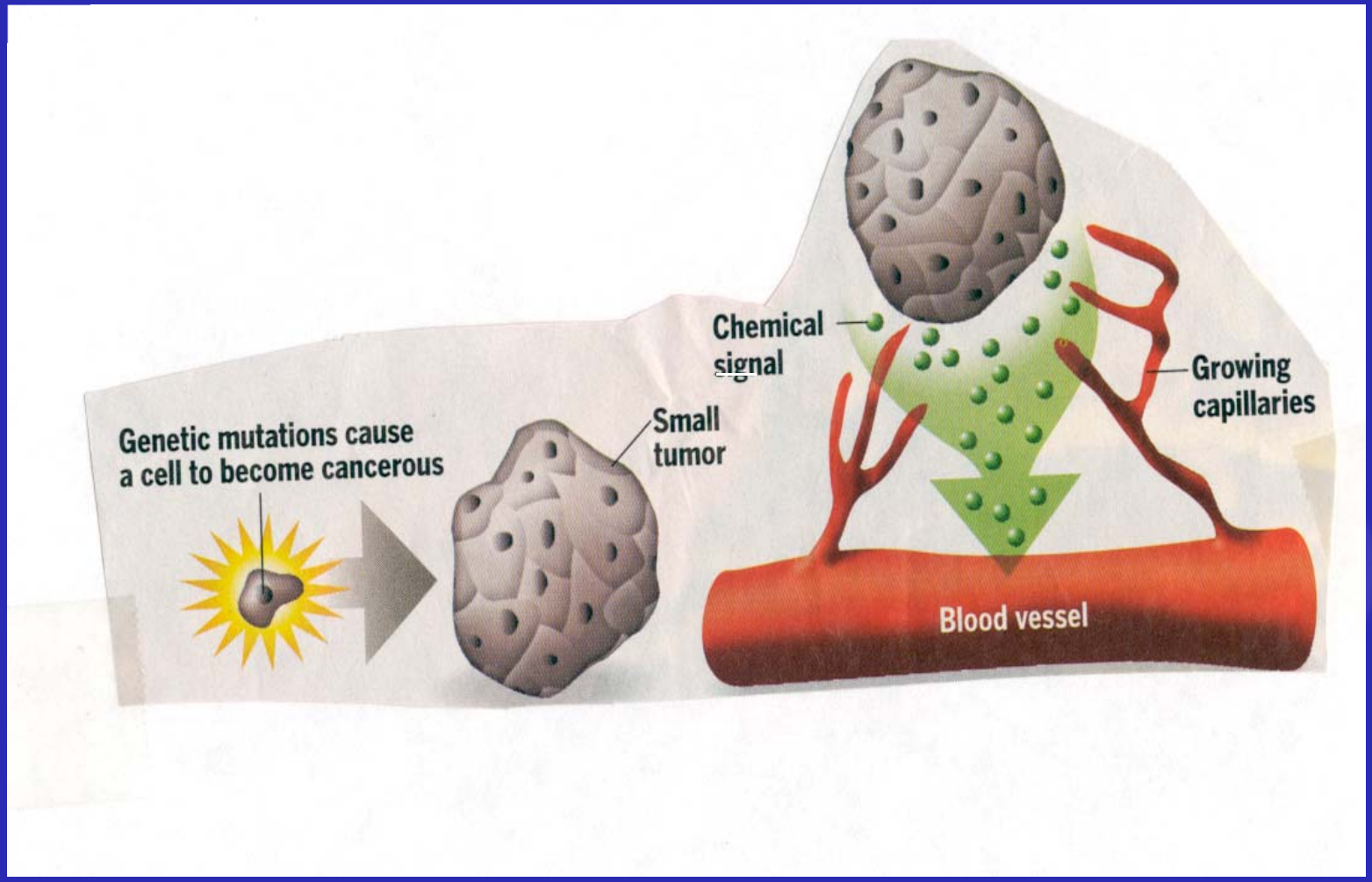


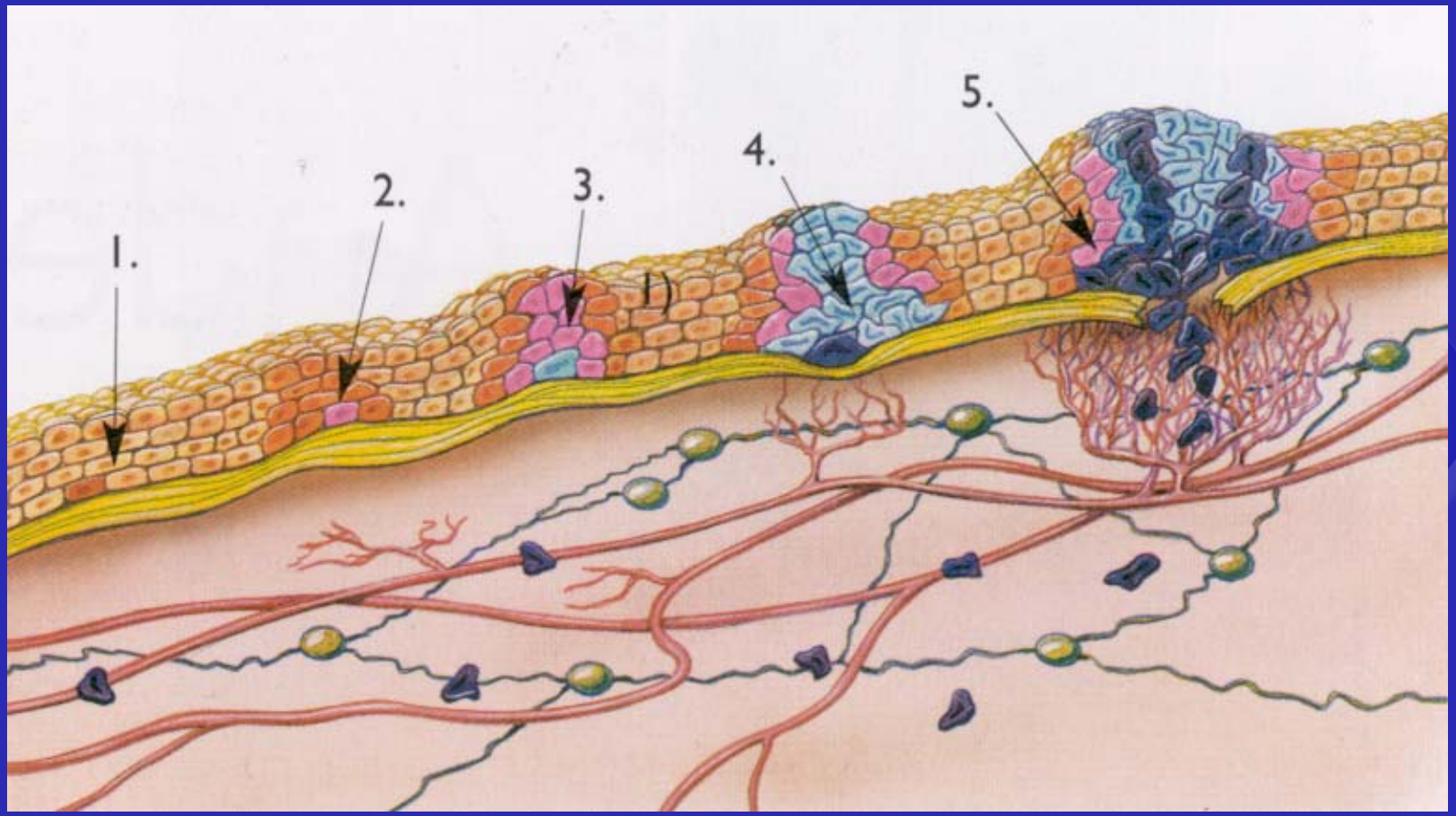
Detection of malignancy



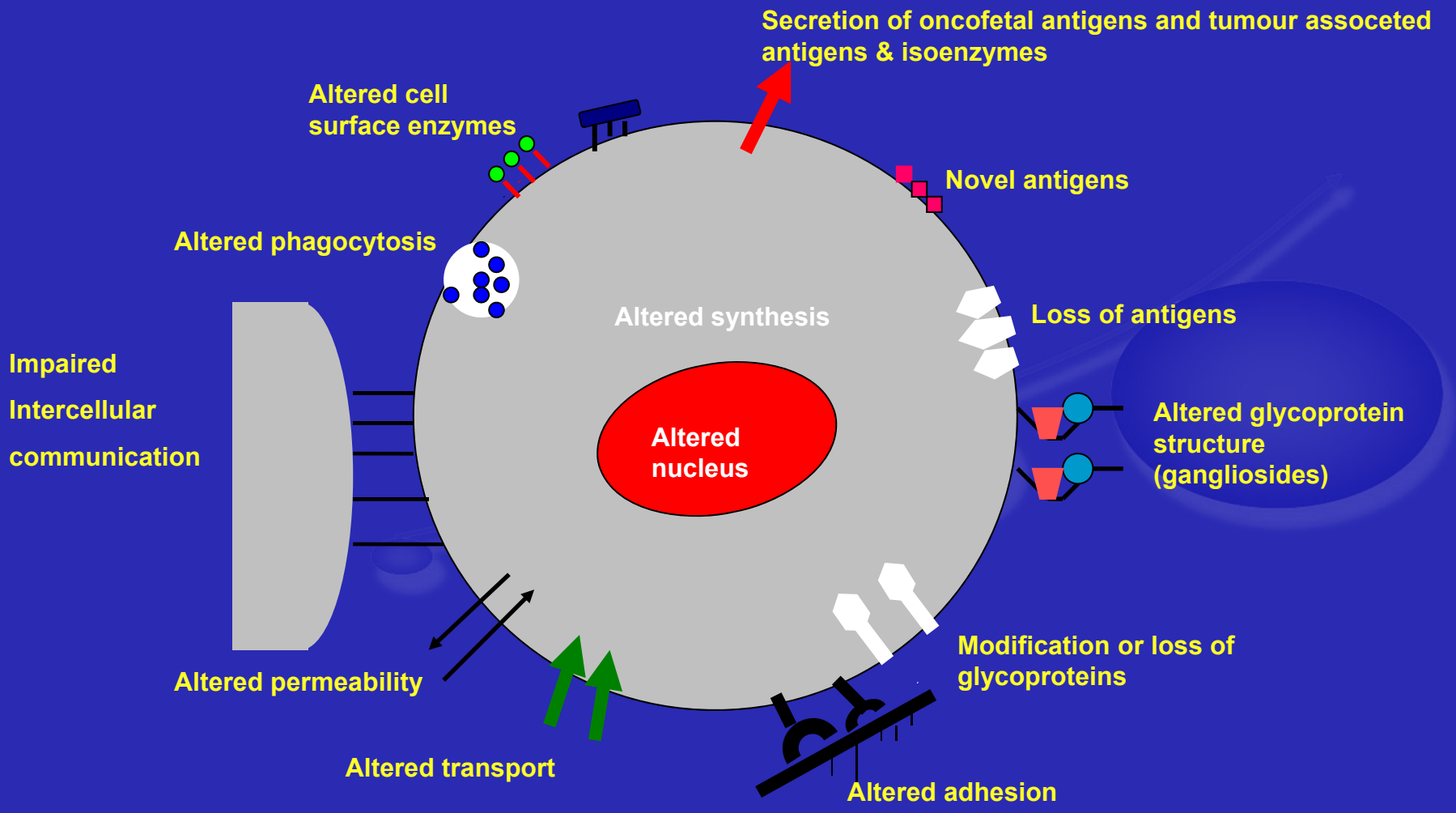
Factors determining tumor marker serum levels

- Size and extention of tumors
- TM synthesis
- TM secretion
- Assocetion between tumor & circulation
- TM catabolism (kidney, liver function, etc.)


Genetic mutations cause
a cell to become cancerous
Small
tumor
Chemical
signal
Growing
capillaries
Blood vessel

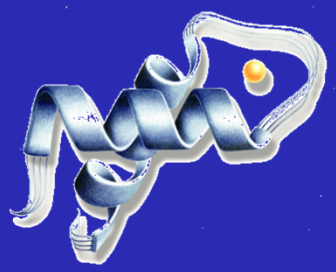

1.
2.
3.
4.
5.

Possible sources of TM in tumor cells



Circulating tumor markers

- Tumor associated antigens
- Hormones and subunits
- Enzymes and isoenzymes
- Specific serum proteins
- Metabolites



TUMOR ASSOCIATED ANTIGENS

CEA	gastrointestine
CA125	ovaries
CA19-9	pancreas
CA15-3	breast
CA72-4	stomach
PSA	Prostate
fPSA	
TPA	Urinary Bladder, proliferation marker

Tissue polypeptide antigen (TPA)

TPA is a polypeptide of 180 kD mol weight and is the mixture of cytokeratines:

Cytokeratin 19	**44%**
Cytokeratin 18	**36%**
Cytokeratin 8	**30%**

Physiologic presence:

- **Fetal tissues, placenta, epithelium**

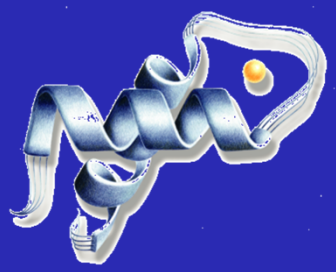
Normal range:

- serum: **0 – 90 U/l**
- urine: **<500 U/l**

Half life: **1 day**

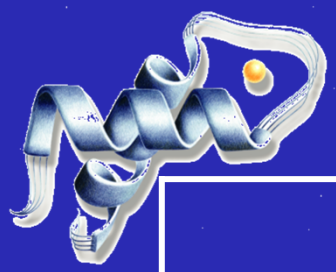
A TPA is produced in cancer cell membranes, secreted actively into biological fluids.

Universal tumor-assoceted antigen as its levels increase with proliferation.



HORMONES

- EUTROPIC hormone synthesis — Tumor of endocrine tissues
- ECTOPIC hormone synthesis — Hormone production of non-endocrine tissues

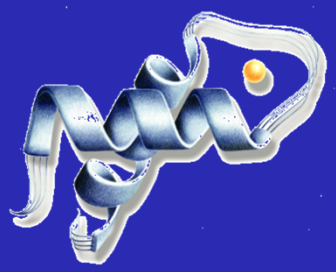


EUTROPIC HORMONE production

TUMOR	HORMONE
Adrenomedullar tumors Pheochromocytoma Neurobastoma	Epinephrine
Adrenocortical tumors	Aldosterone Cortisol
Pituitary tumors	Prolactin GH
Iinzulinoma	Insulin
Pancreas non-beta insulinoma (gastrinoma)	Gastrin
Parathyroid cancer	Parathormone
Medullar thyroid cancer	Calcitonin
Chorioncancer	hCG

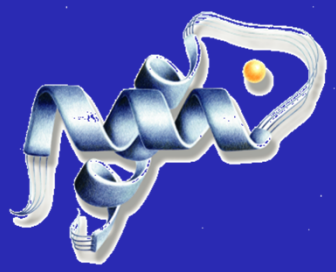
ECTOPIC HORMON productions

TUMOR	HORMONE
1. APUDOMAS (**A**mine **P**recursor **U**ptake - for high uptake of amine precursors.(HISTAMINE) **D**ecarboxylase - for high content of the enzymeamino acid decarboxylase (for conversion of precursors to amines)	
Small cancer of the lung	ACTH
Carcinoid tumors	Lipotrophin (LPH)
Pancreas islet cell tumors	Vasopressin (AVP
Malignant epithelial thymomas	Calcitonin, parathormone, gastrin, (insulin, glukagon)
2. Non-APUDOMAS	
Lung epidermoid and adeno cancers	Calcitonin, parathormone, GH, prolactin, insulin, glukagon
Breast cancer	Parathormone, ACTH



SPECIFIC SERUM PROTEINS

TG (Thyreoglobulin)	Thyroid cancer
Beta-2-microglobulin	
S100 protein	Melanoma
AFP (alfa-fetoprotein)	- liver cancer & metastasis



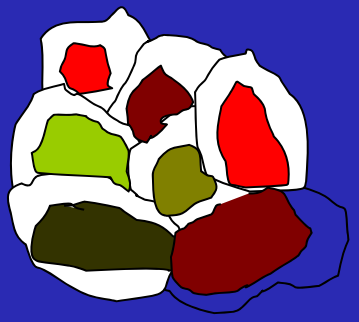
ENZYMES

- NSE (Neuron specific enolase)
  - Neuroblastoma
  - Small cell lung cancers
  - Apudoma

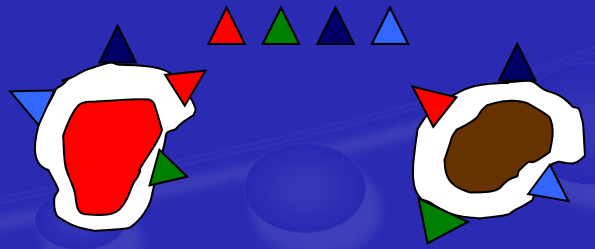
- TK1 (Timidine kinase)
  - Lymphoproliferation malignancies (leukemia, lymphomas, myeloma, etc.)
  - Activity marker in solid tumors

Impact of tumor heterogeneity on TM markers determined with poly- and monoclonal Abs

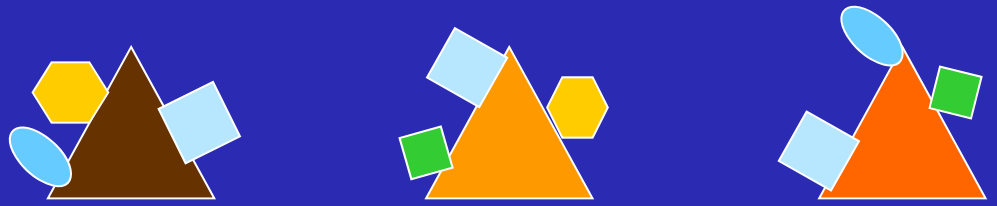
Heterogeneous tumor cells



Tumor cells may vary in term of TMs produced



TMs may contain different epitopes with different patterns



TMs in suspected cancer

Tumor	TM	Tumor	TM
Liver (primer)	AFP; CEA	Prostate	PSA; fPSA
Testis	AFP; hCG	Urinary bladder	TPA; CEA; CA 19-9
Head/Neck	SCC; CEA	breast	CA 15-3;CEA; TPA
Lung	NSE; SCC; CEA; TPA	Cervix	SCC; CEA; CA 125; TPA
Biliary	CA 19-9; CEA; TPA	Ovaries	CA 125; CA19-9; CA 72-4; TPA
Pancreas	CA 19-9; CEA	Neuroblastoma	NSE; VMA; HVA
Gastric	CA 72-4; CA 19-9; CEA; TPA	Limphoma, leukemia	TK1; ß-2-mikrogl.; Bence-Jones fehérje; monoklon. Immunglob.
Colon cancer	CEA; CA 19-9; TPA		
Thyroid	Tg (papill.); Calcitonin (Medul.)	Melanoma	S-100

Multiparametric TM test

- Tumors are mixture of different tumor cells
- Simultaneous test of several TMs increases the sensitivity
- TMs are specific for tissues, not for organs.

Epithelial cell express:

CA 125 (ovarium)

CA 15-3 (breast)

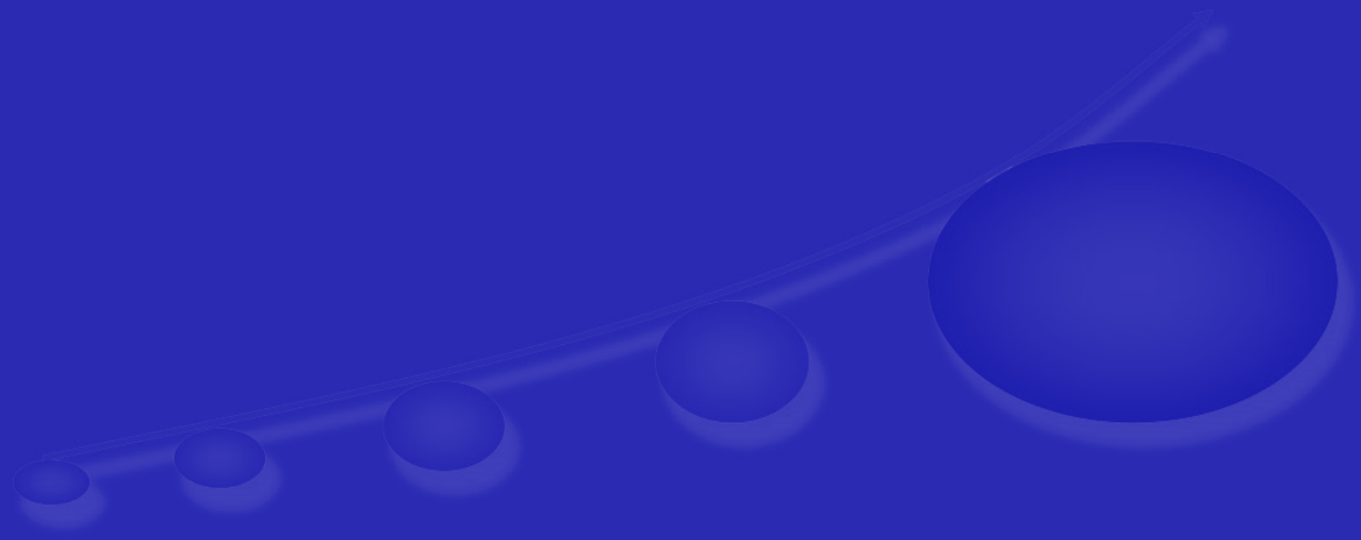
CA 19-9 (pancreas)

CEA (vastagbél)

antigens with specific patterns for one localization.

Screening

- There is no generally recommended TM for screening of any cancer



Screening of populations at risk: some guidelines

- Prostate cancer
  - Above 50 years in males (in general)
  - Above 40 years in high risk males
  - PSA and RDE test yearly; if PSA is 4-10 ng/ml & RDE negative, fPSA (warning: fPSA is decreased in vitro after sampling and may indicate cancer falsely)

- Ovarian cancer
  - Yearly transvaginal US and CA 125 in women with family history of breast- and ovary cancer.
  - Postmenopausal women with pelvic lesions: CA 125 & HE4 tests.

Diagnosis & prognosis: the position of TMs

General exam

- Physical exam
- Endoscopy
- Imaging
- Surgery

Specific workup

- Histology, cytology
- Clinical chemistry
- Tumor markers
- molecular biology

Tumor classification

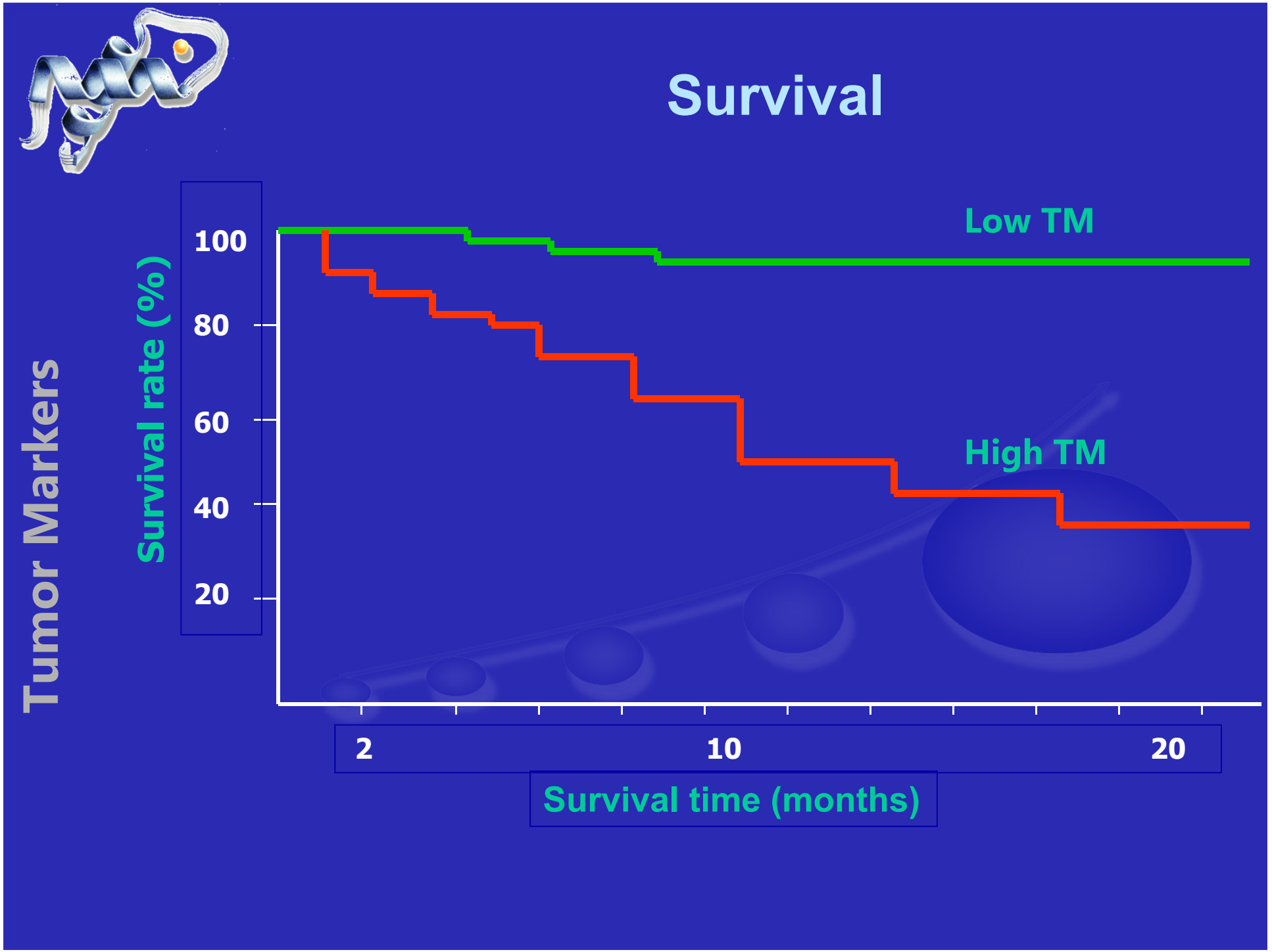
- Staging
- Localization
- Grading (low/ high risk)

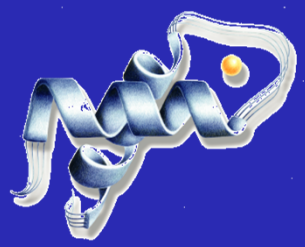
Therapy

Example for breast cancer

Preoperative TM values

		TPA		CA 15-3		CEA		TK	
	N	mean	Std.Dev.	mean	Std.Dev.	mean	Std.Dev.	mean	Std.Dev.
Benign	150	43,8	27,1	19,8	15,8	1,7	1,6	3,9	1,7
Primary breast cc	450	98,9	235,4	32,6	111,7	3,5	11,1	4,8	4,9
Recurrence	80	139,1	224,5	113,9	550,5	8,4	47,1	6,7	9,2
Metastasis	**24**	**93,2**	**58,4**	**22,6**	**11,5**	**2,2**	**1,7**	**5,6**	**3,5**

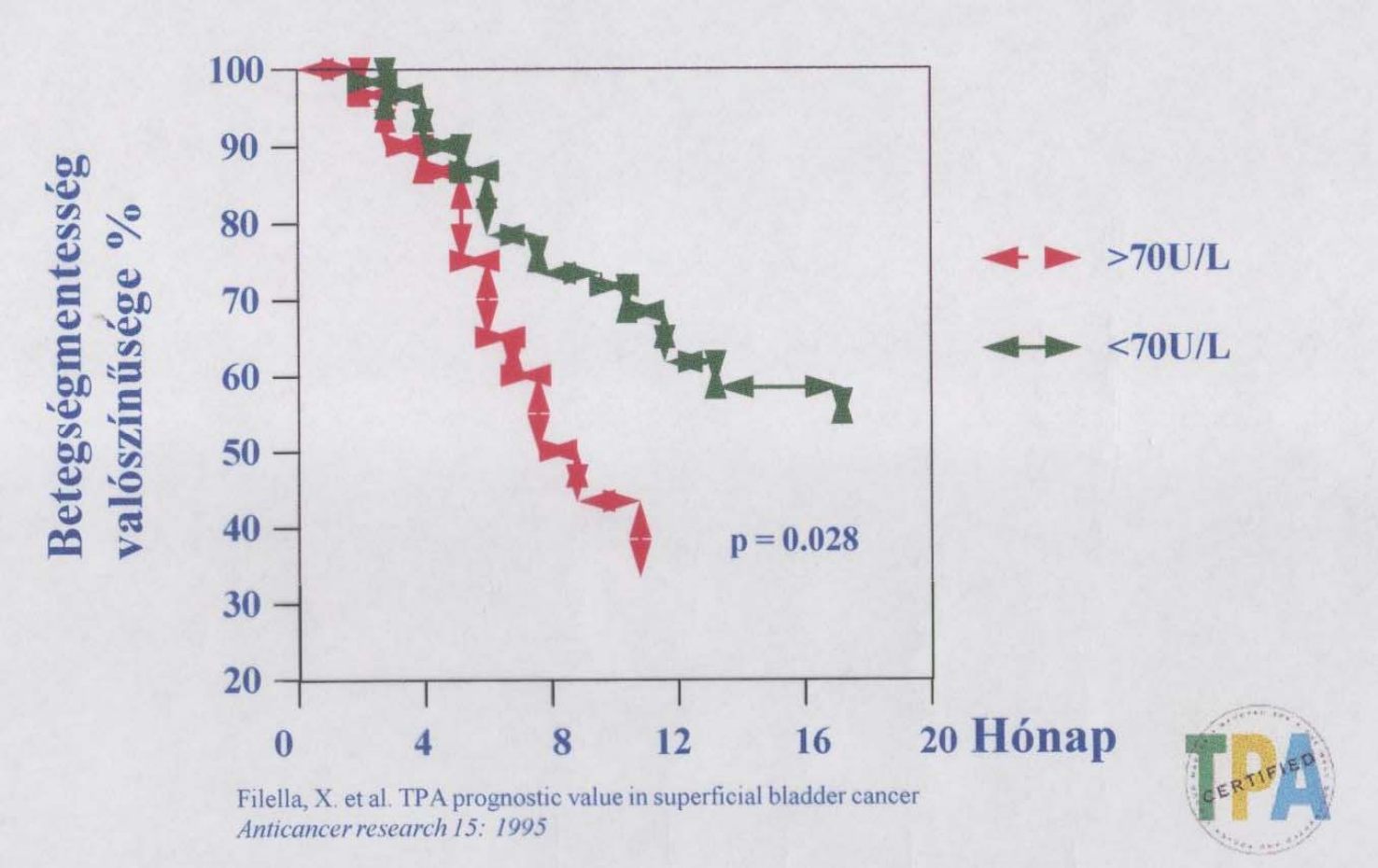

Survival
Tumor Markers
Survival rate (%)
100
80
60
40
20
Low TM
High TM
2
10
20
Survival time (months)



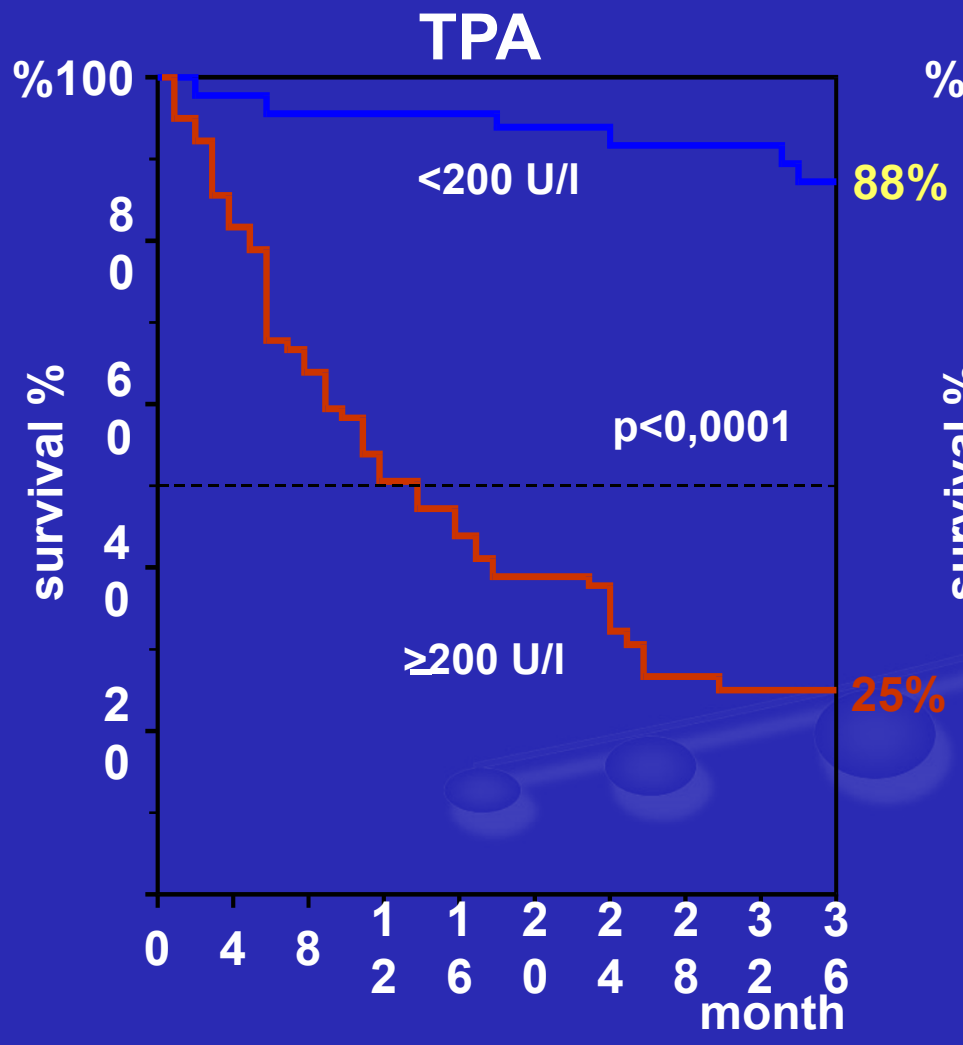
Tumor markers and prognosis in liver tumors

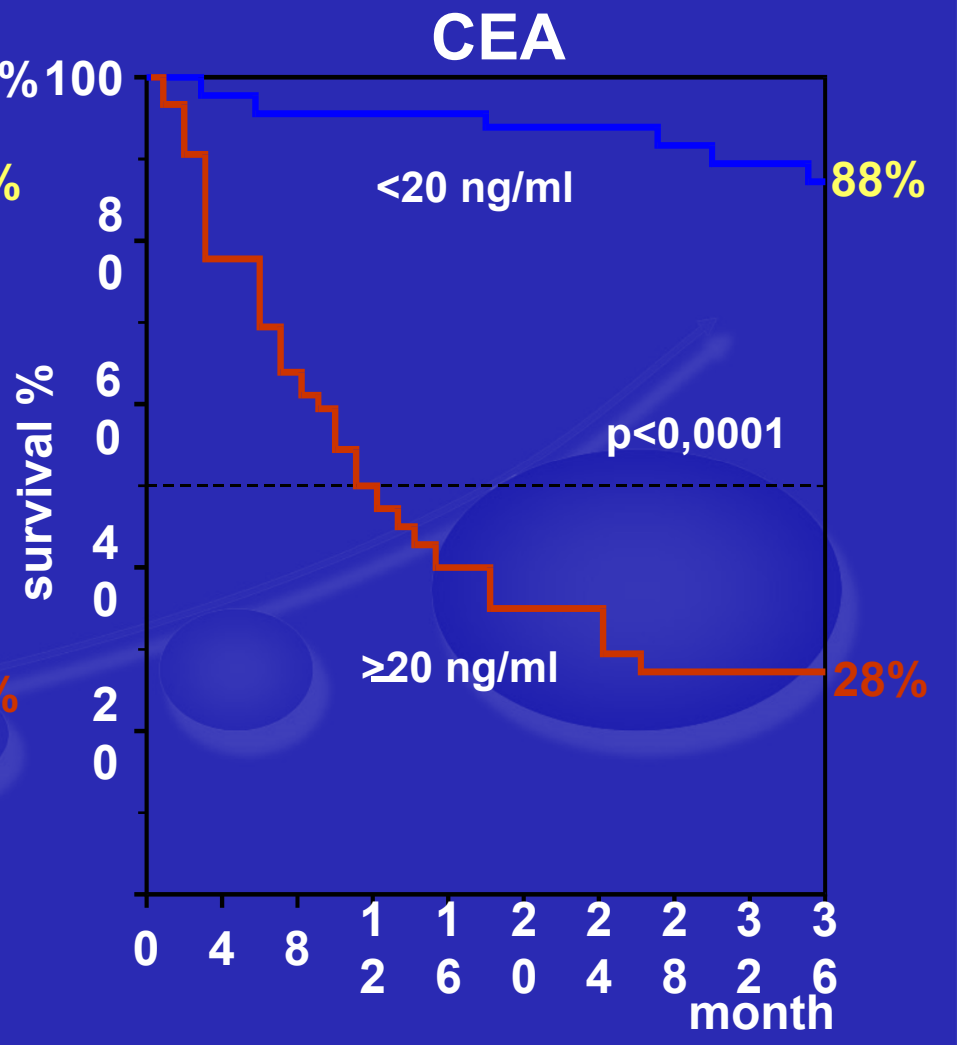
Prognosis	AFP (ng/mL)	hCG (U/L)
Good	< 1.000	< 5.000
Intermediate	1.000 – 10.000	5.000 – 50.000
Poor	> 10.000	> 50.000

Serum TPA – prediction of recidiva in bladder cancer

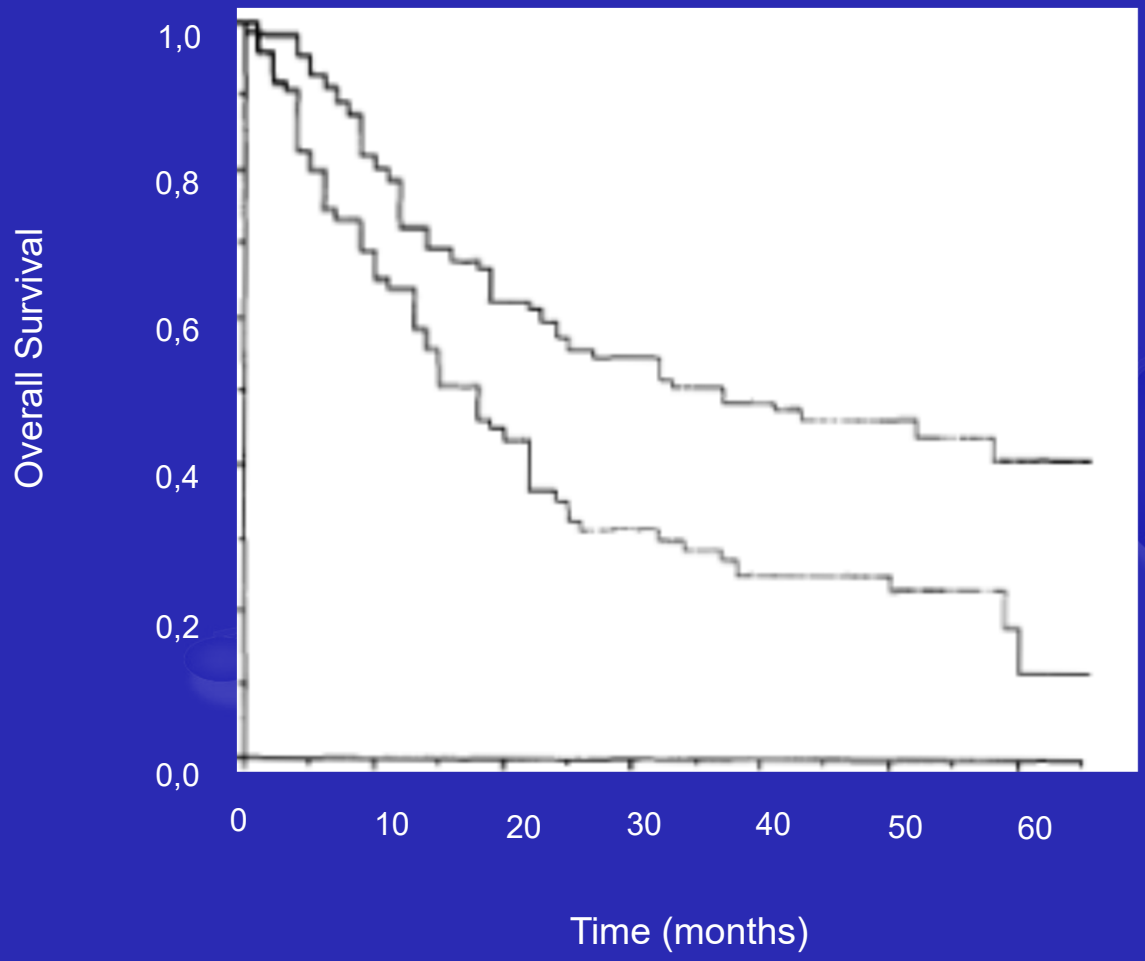


Preoperative levels & prognosis





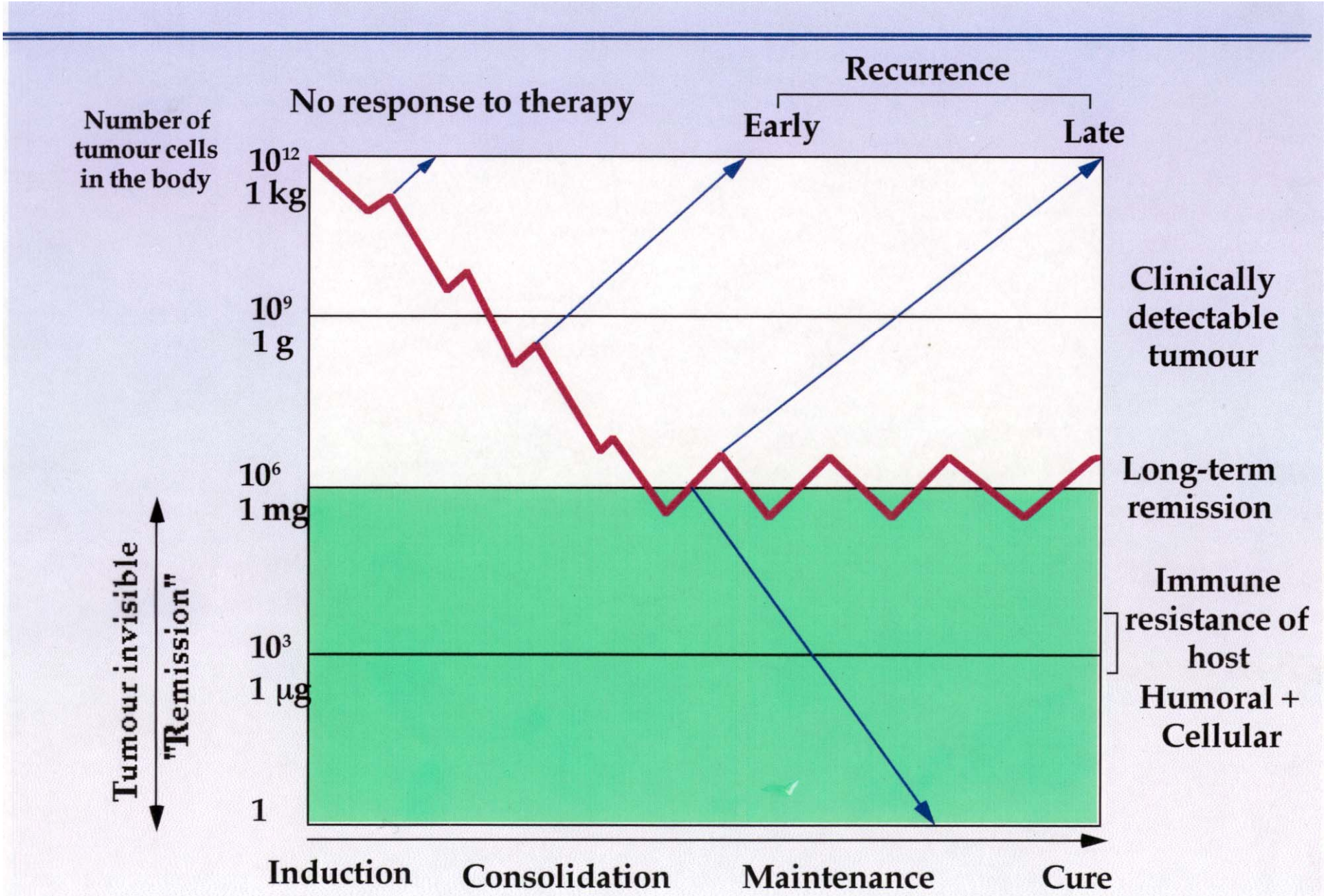
TK1: prognosis in head-neck cancers



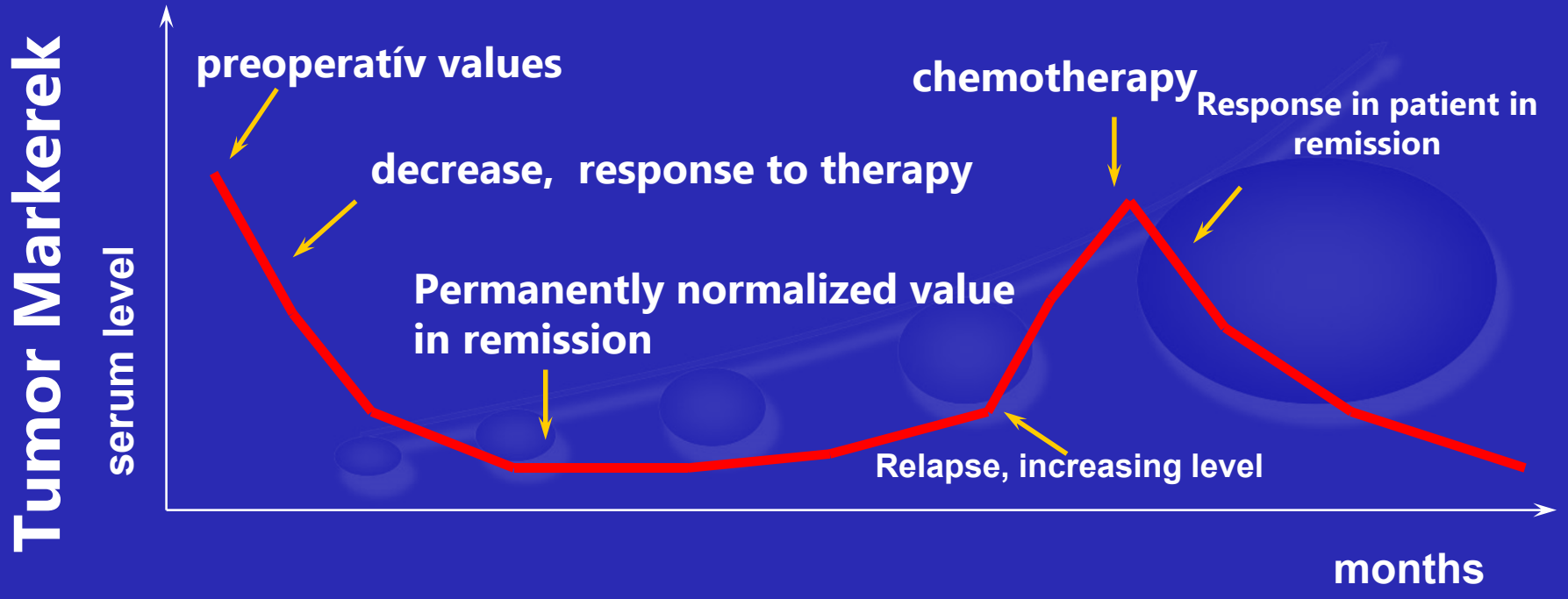
Fontana et at Head&Neck 1993;15; 425-432

Follow-up

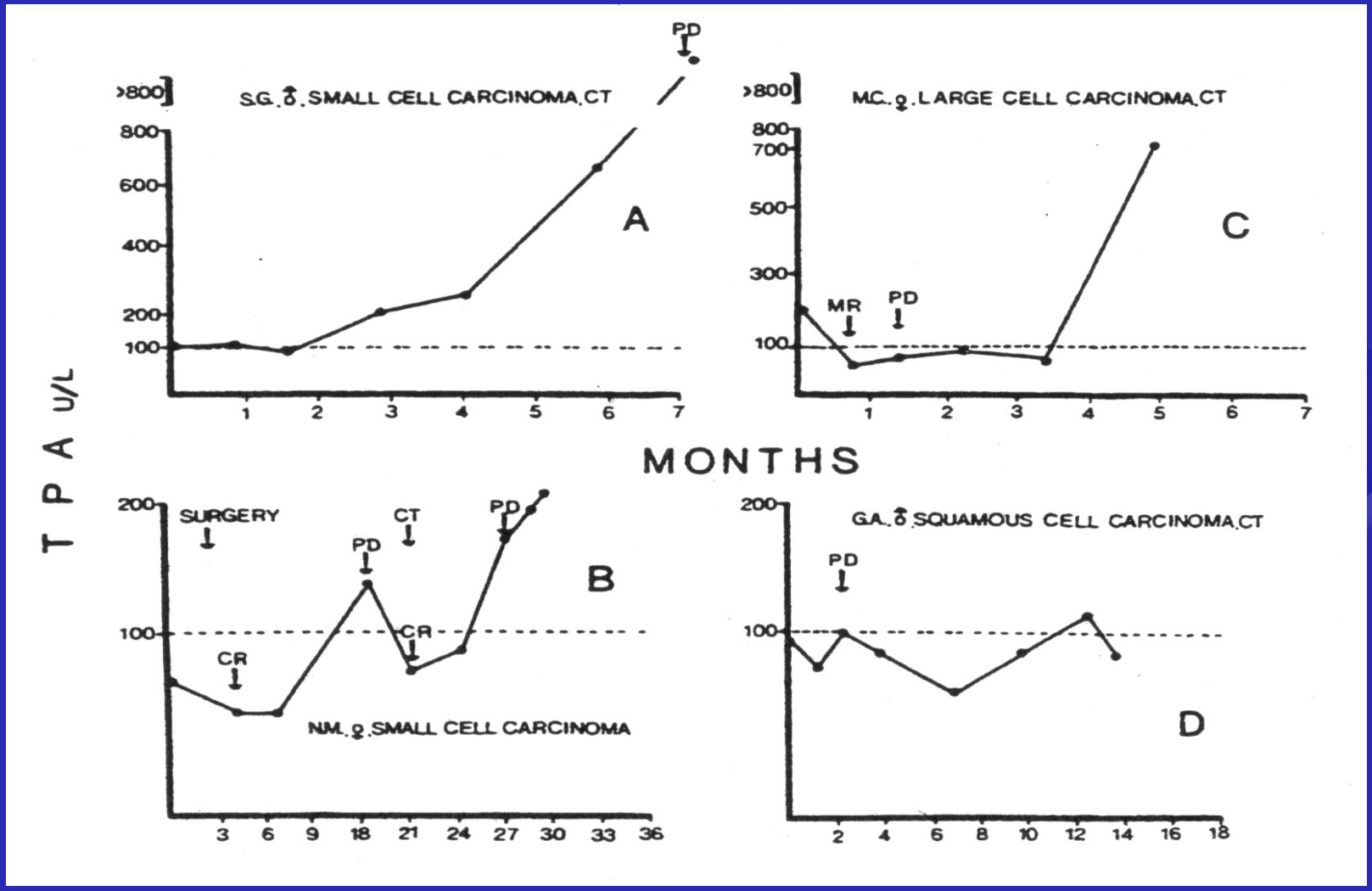
- Establishing the individual change in status
  - Tumor free
  - progression
  - response
- Analytical change = ?biological change
- Sequential tests
  - Extent of change
  - kinetics
- Normal value is inappropriate for this purpose
- Factors having an influence on analytical results
  - C_A – imprecision
  - C_G – intrapersonal variability
  - C_P – interpersonal variability
- C_p – in healthy female patients
    - CA 15-3 - 6.2%
    - CEA - 3.3%
    - TPA - 28.3%


Recurrence
No response to therapy
Early
Late
Number of tumour cells in the body
10^{12}
1 kg
10^{9}
1 g
10^{6}
1 mg
10^{3}
1 µg
1
Clinically detectable tumour
Long-term remission
Immune resistance of host
Humoral + Cellular
Tumour invisible
"Remission"
Induction
Consolidation
Maintenance
Cure

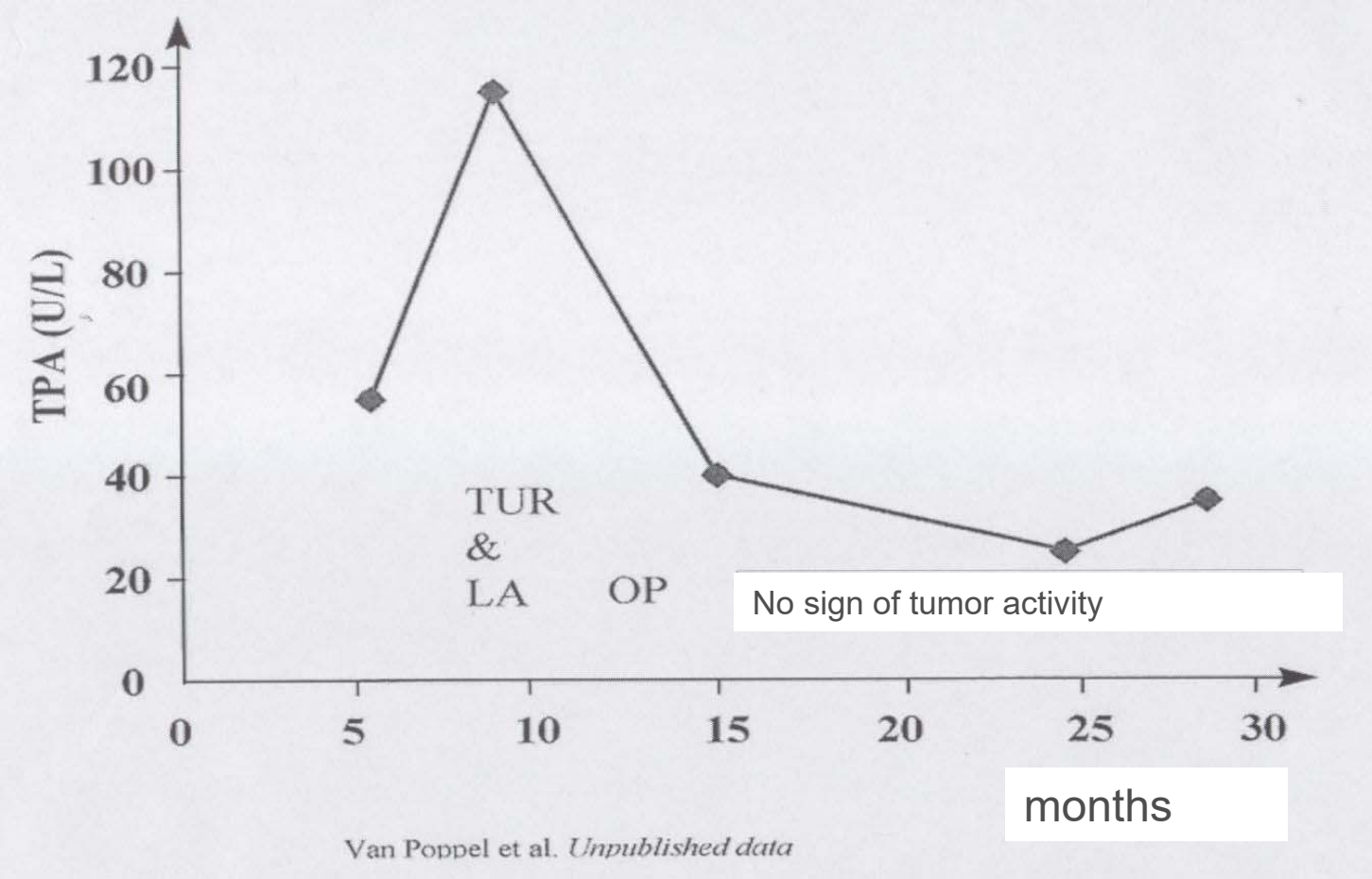
Characteristic alteration of TM levels during monitoring



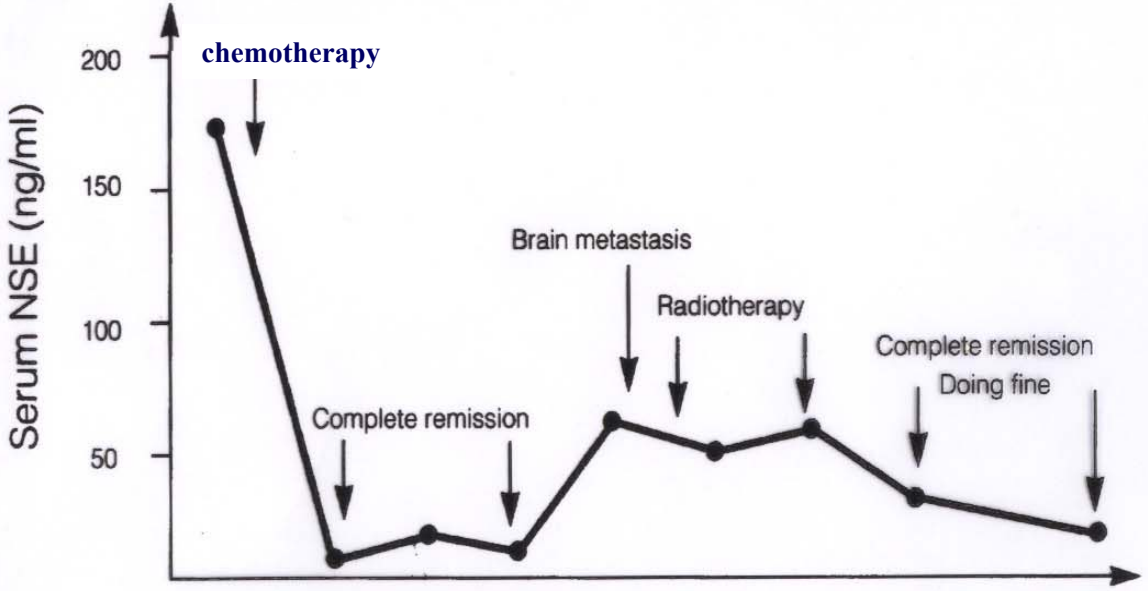
Use of TPA in lung cancer monitoring



serum TPA values in patients with urinary bladder cc



Serum NSE levels, in SCLC patient with high (170 ng/ml) initial serum conc., followed during treatment for 22 months

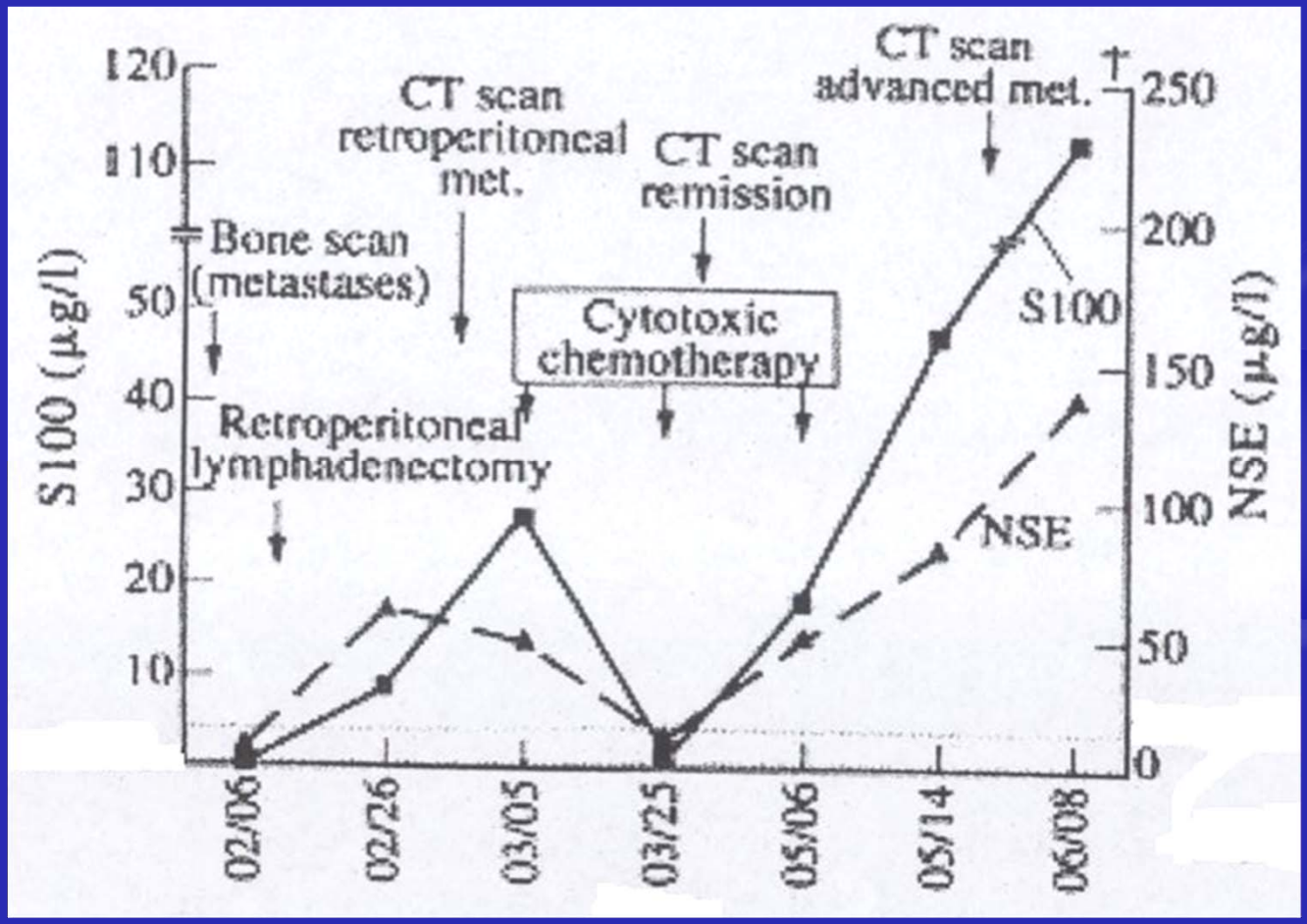


idő

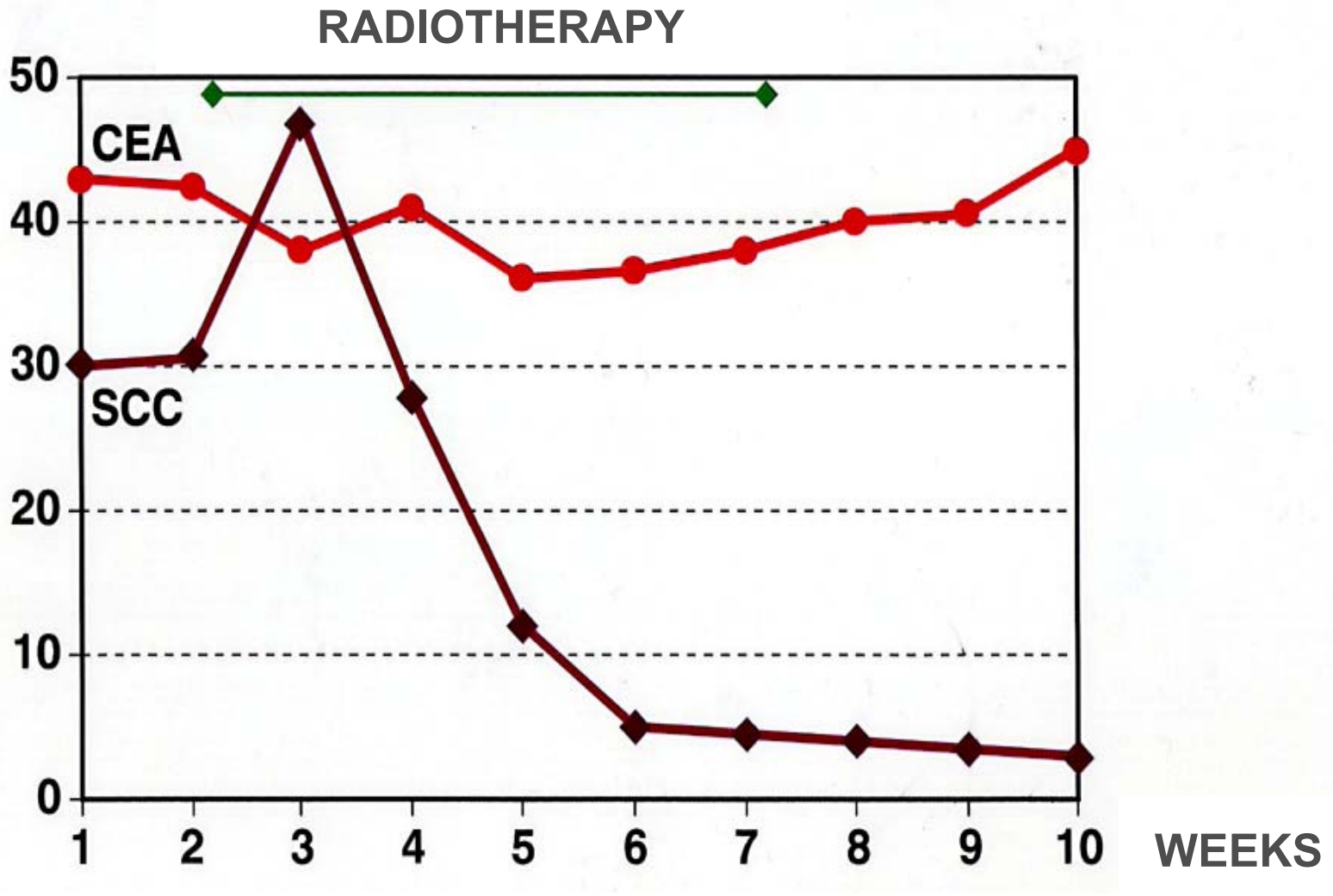
Neuron-Specific Enolase as a Marker for Neuroblastoma and Small-Cell Carcinoma of the Lung.
S.Påhlman, T.Esscher, J Bergh, L. Steinholtz, E.Nöu, K. Nilsson.
Tumour Biology, 5 (1984) 119-126

luresnse.2

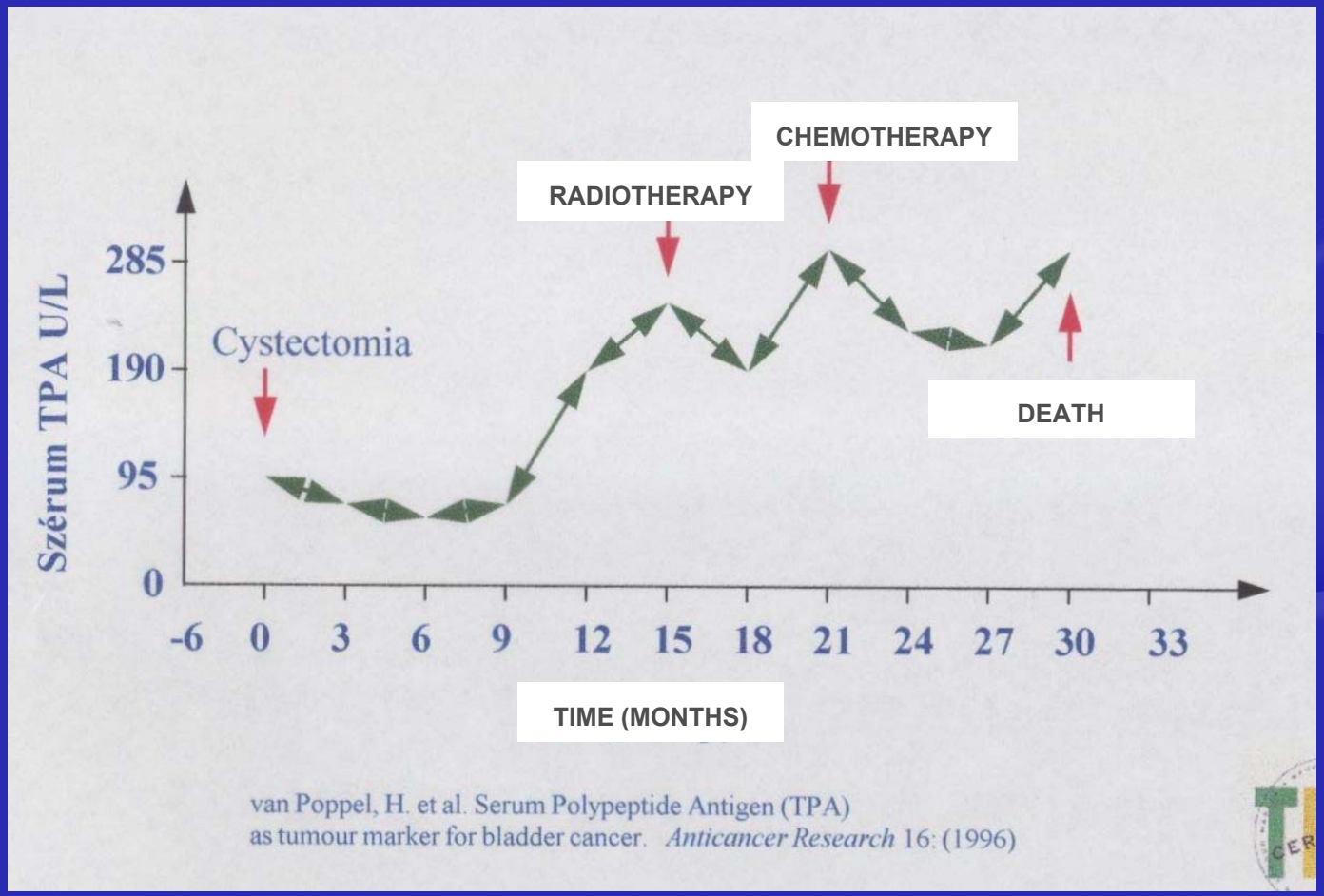
S-100 & NSE values in glioblastoma



MONITORING THE EFFECT OF RADIOTHERAPY IN A PATIENT WITH ORAL EPITHELIAL CANCER AND BREAST METASTASIS

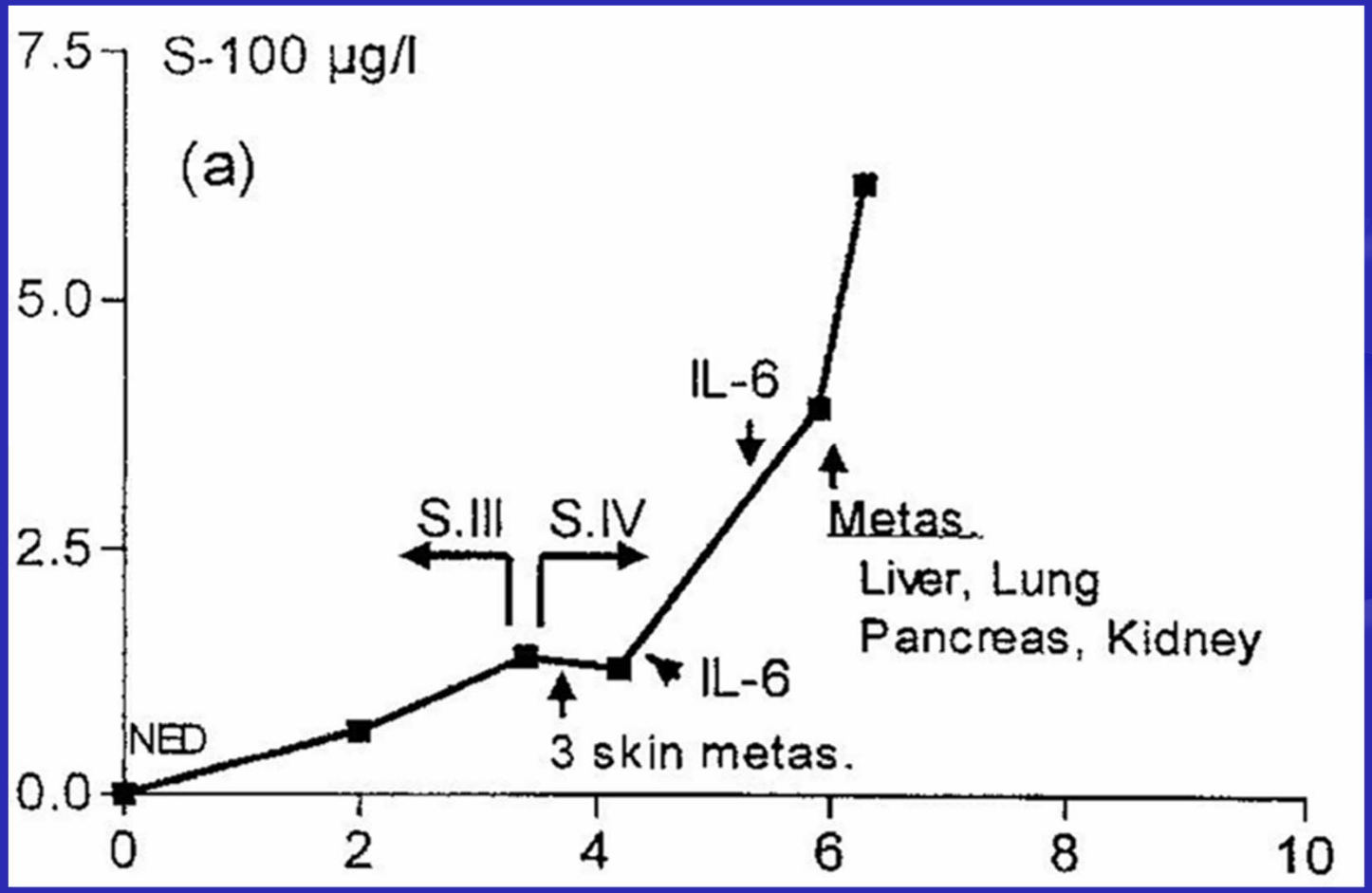


TPA for the assessment of therapeutic response



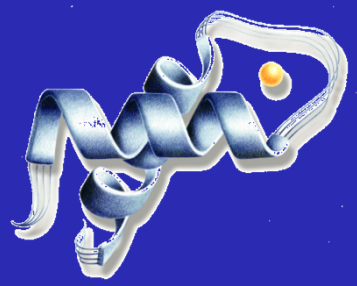
van Poppel, H. et al. Serum Polypeptide Antigen (TPA) as tumour marker for bladder cancer. *Anticancer Research* 16: (1996)

S-100 levels indicate the progression of melanoma during interleukin therapy



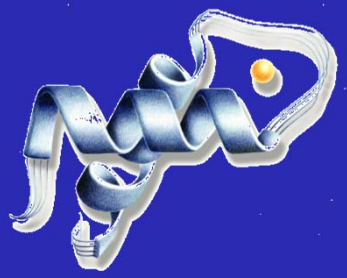
The use of some TMs to early detection of recurring and metastatic breast cancer

Marker	Sensitivity (%)	Specificity (%)
TPA	63	98
CA 15-3	46	98
CEA	7	99
CA 15-3+TPA	83	96
CEA +TPA	70	98



Recommended frequency of TM tests during monitoring

- Postoperative phase / chemotherapy or irradiation
  - Before therapy, then 2 – 10 days after therapy (depending on half life)
  - At least quarterly within 2 years after the surgery
  - At least twice a year 3 – 5 years after the surgery
- Before therapy switch
- When relapse or metastases are suspected
- New staging
- 14 – 30 days after measuring an abnormal TM

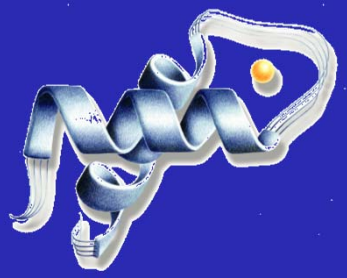


Factors influencing TM results & interpretation

- Preanalytical consideration
  - Analytical variables
  - Postanalytical issues

Factors – in detail

Preanalytical phase	Analytical phase	Postanalytical phase
● Inappropriate sampling	● failure of the analytical system	● Misinterpretation
● Patient ID errors	● insufficient analytical specificity	● Data management failure
● inappropriate order	● performance of analytical system is suboptimal	● long Turn-around-time
● Data management failure		● etc.
● **etc.**		
48%	**13%**	**39%**



Preanalytical phase

- Sampling, treatment of samples, storage

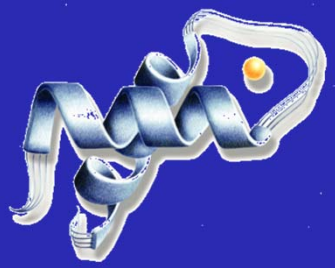
- Clinical data
  - Patient's actual status (e.g. timing of sampling)
  - in vivo interfering factors (pl. co-morbidity, drugs)
  - Prior TM that was performed elsewhere (result, date, method)

Analytics

- Modern, quick and automated immunanalyzers
- Intra-assay variability < 5%
- Inter-assay variability < 10%
- Internal and external QC
- Standardisation, use of reference methods (AFP, hCG, CEA, PSA international standard materials); in some cases there is no reference material

Postanalytical phase

- Factors to be considered when TMs are interpreted
  - Analytical result
  - Reference range
  - Method used
  - Analitical sensitivity
  - Prior TM values
  - Kinetics (biologically significant: > ± 25%)
  - Recommended confirmatory tests (repeated dampling)
  - Patient information (TM tests alone have no diagnostic value)
- Laboratory consultants are highly recommended



Abnormal TM value without clinical basis

Possibility	Remedy
• Analytical failure	Repeated testing
• Non malignant illness	Further tests repeated TM test
• Malignancy (subclinical stage)	Multiparametric TM close monitoring Oncological exam
• Other factors (heterophyl antibody)	History to identify interfering factors

Factors influencing the use of TM tests

- Unrealistic expectations regarding the information provided by TM (e.g. cancer test)
- Lack of information how to use TM & its combinations
- Inaccuracies regarding clinical interpretation os results
- No clinical and laboratory protocols
- Problems with labs

Take home message

- TMs are suitable for MONITORING /FOLLOW-UP
- Multiple use of TMs has additional value to therapeutic decision making
- TMs are inappropriate for diagnosis or general screening

BASELINE TUMOR MARKER VALUES ARE REQUIRED TO ASSESS FLUCTUATION