



Cytokeratins as Tumor Markers

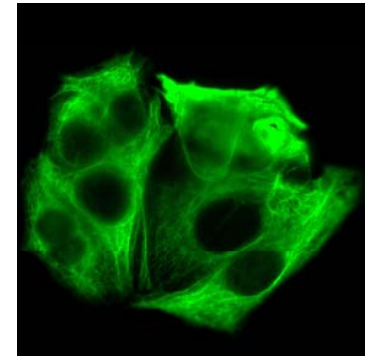


Roland Einarsson

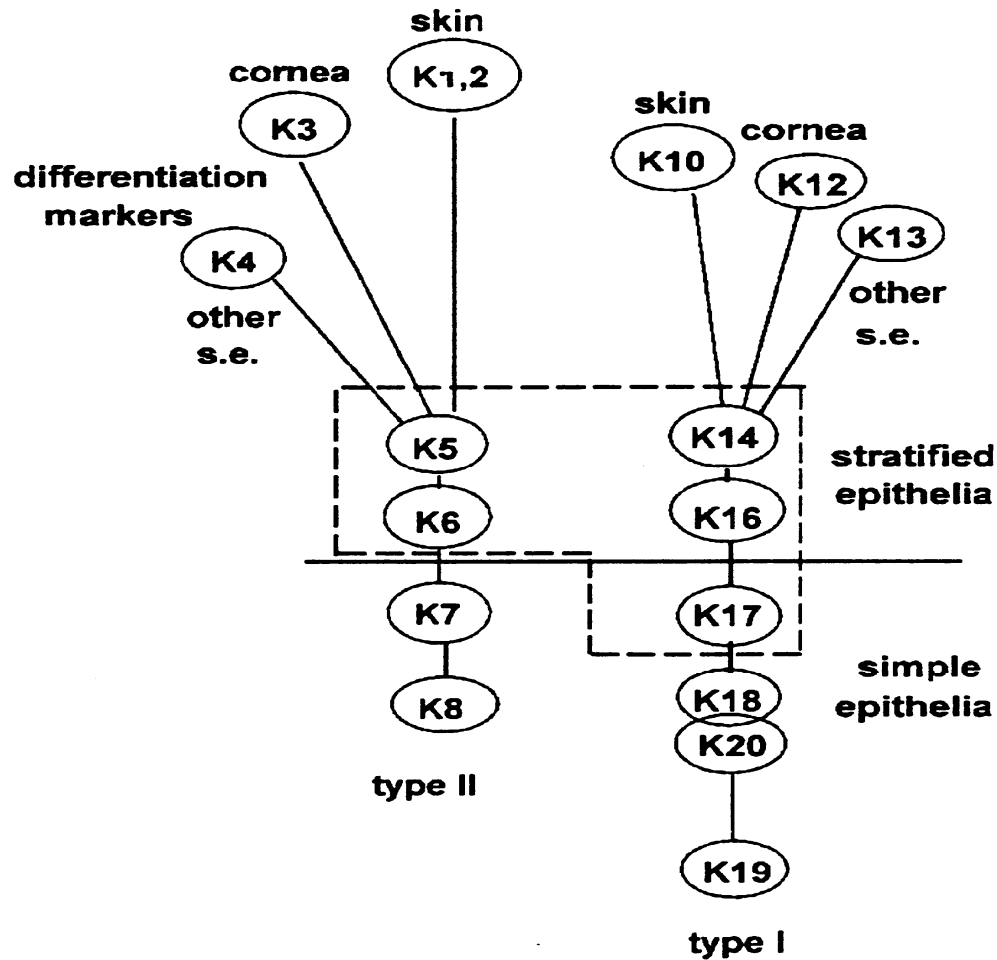
What are cytokeratins?



- Cytoskeletal proteins
 - Intermediate filament proteins
 - Family of >20 different proteins
 - CKs 8, 18 and 19 most abundant
- Epithelial cell specific expression
 - Overexpressed in transformed cells
- Cell death mode - necrosis
and/or apoptosis

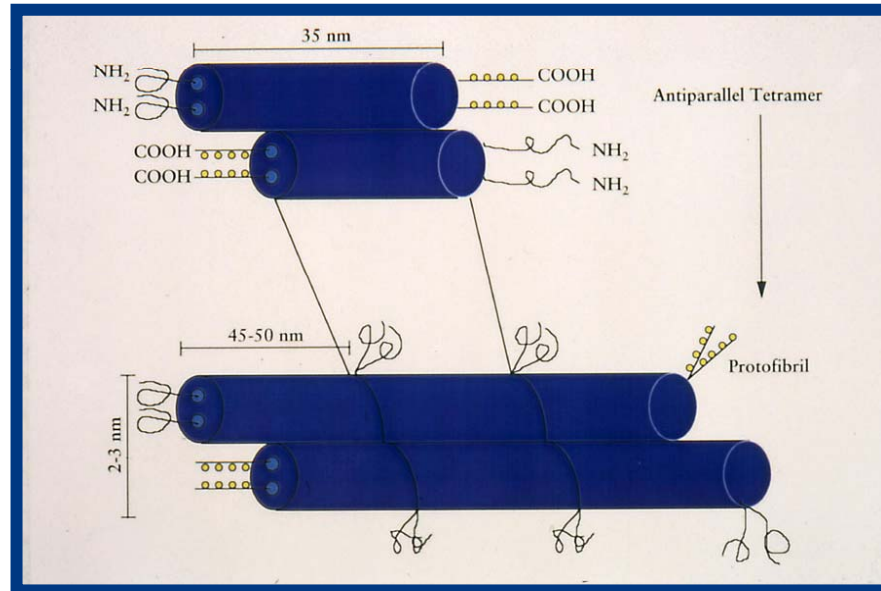


Expression of cytokeratins in various epithelia.



B. Sundström, T. Stigbrand: *Int J Biol Markers* 1993; 9, 102-108.

Cytokeratin protein structures

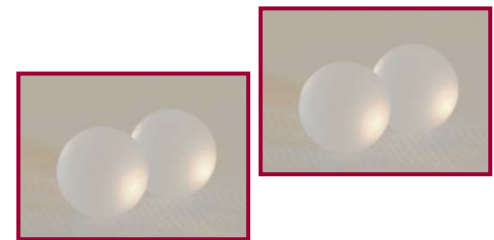


- Acidic (type I, CK 9-20, 40-56 kDa); basic proteins (type II, CK 1-8, 53-68 kDa)
- Obligate heterodimer formations
- Detection of CK fragments in circulation

Cytokeratin tumor markers



- Tumor cell activity markers
 - Early and distinct signals
- Management of patients with carcinomas
 - Prognosis, monitoring and follow-up
- Complementary to volume markers
 - Defined panels give increased sensitivity and prolonged lead times
- Not organ specific



Cytokeratin panel - Indication



TPS®

Breast, Prostate, GI &
Gynecological cancers

UBC®

Urinary Bladder cancer

TPAcyk™

General epithelial cancers

MonoTotal®

Non-Small Cell Lung cancer
Available in IRMA format only

Cytokeratin panel - Details



TPS[®]

Tissue Polypeptide Specific ag
Cytokeratin 18

UBC[®]

Urinary Bladder Cancer ag
Cytokeratins 8/18

TPAcyk[™]

Tissue Polypeptide Antigen cyk
Cytokeratins 8/18

MonoTotal[®]

Cytokeratins 8/18/19

Clinical documentation



TPS[®]

~ 400 scientific studies

UBC[®]

~ 45 scientific studies

TPAcyk[™]

~ 25 scientific studies / excl TPA

MonoTotal[®]

~ 10 scientific studies

Commercially available cytokeratin markers



- TPA (Tissue Polypeptide Antigen)
 - cytokeratins 8, 18 and 19
 - general marker for epithelial cancer
- CYFRA 21-1 (Cytokeratin Fragment)
 - cytokeratin 19
 - mainly small cell lung cancer (NSCLC)

Non-malignant conditions

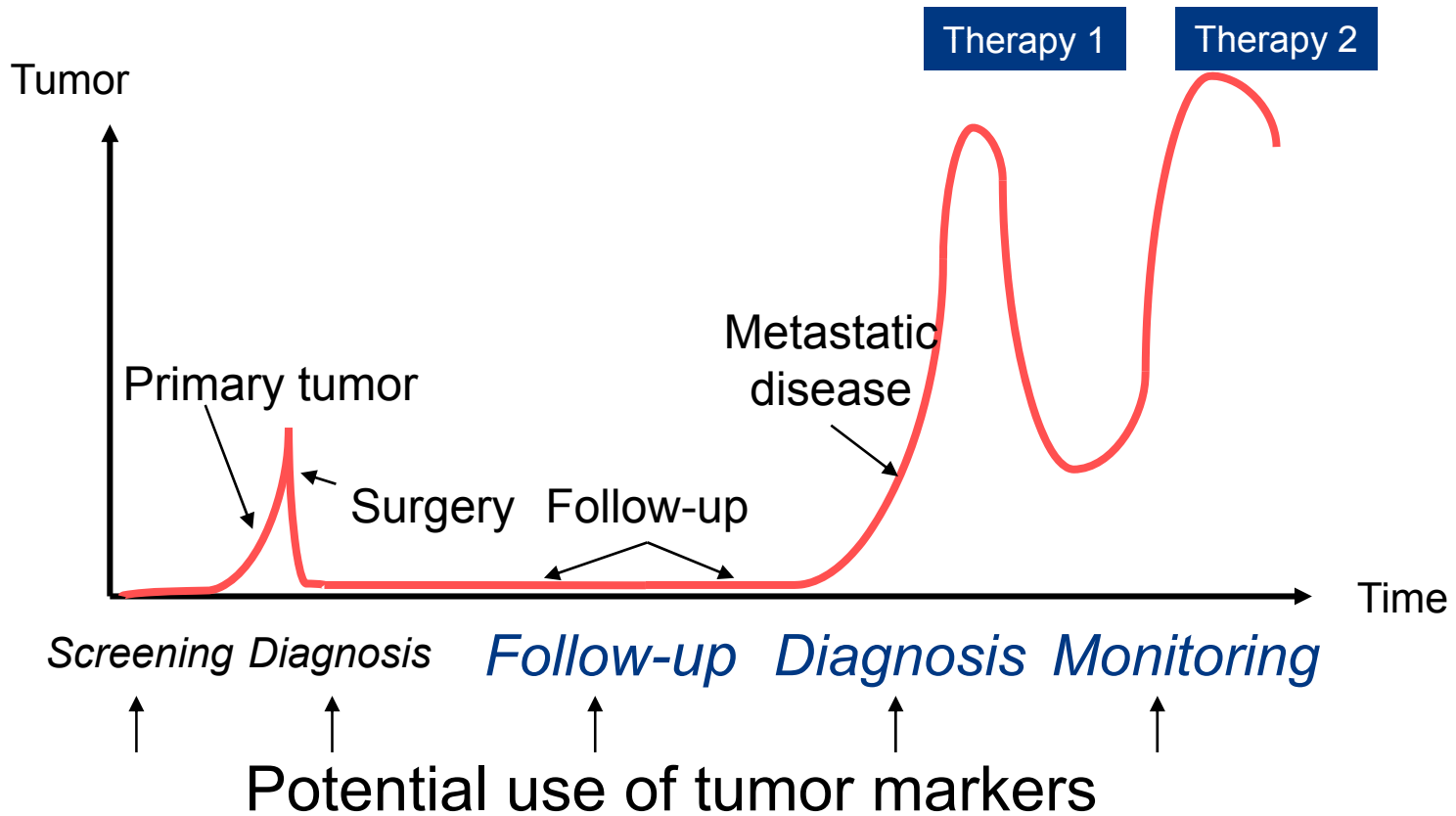


Increased cytokeratin serum levels can be found in case of

- pregnancy
- liver disease
- renal failure
- general infections
- diabetes



TMs - Course of events



Added clinical values



PROGNOSIS

Prediction of possible therapeutic response

Prediction of rapid clinical progression

Optimized therapy based on early response

Motivation to complete therapy courses

Termination of ineffective therapy

– reduced toxicity and society costs

MONITORING

Simplified & sensitive follow up – long lead times

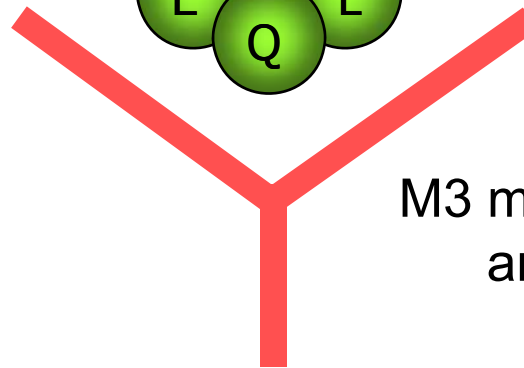
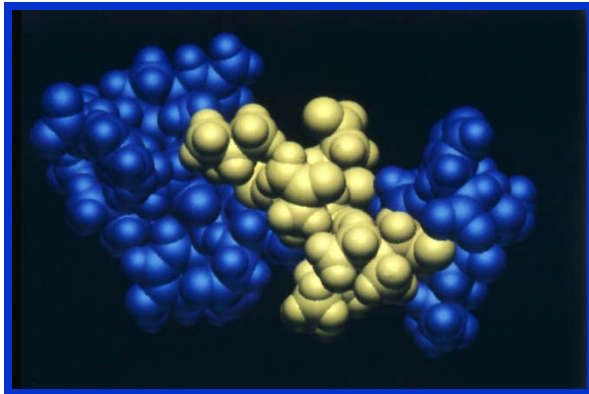
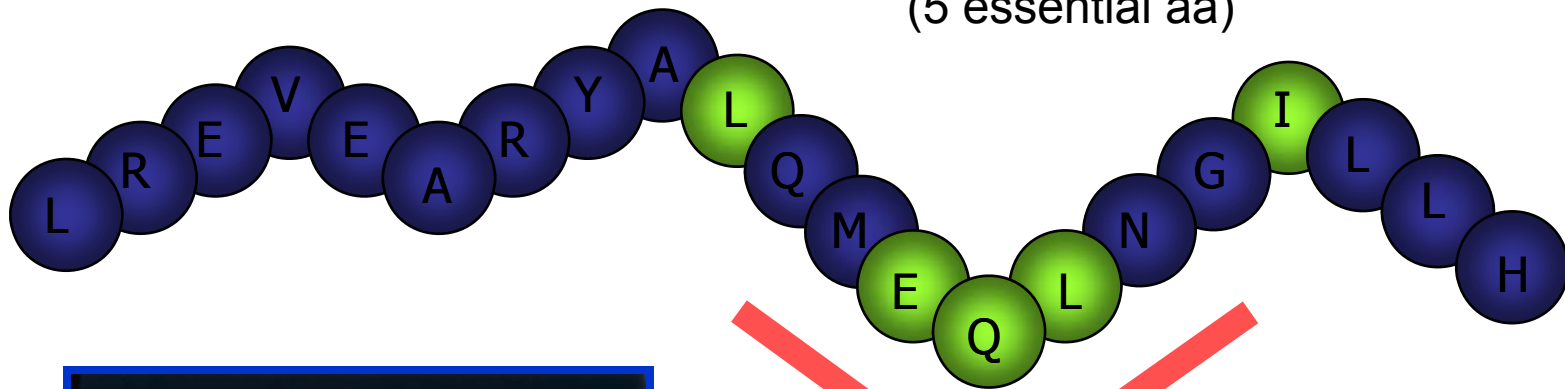
Reduced use of more expensive examinations

FOLLOW UP

TPS epitope M3



21 amino acid residues
(5 essential aa)



M3 monoclonal
antibody



TPS[®]

Advanced Breast Cancer

Breast cancer management

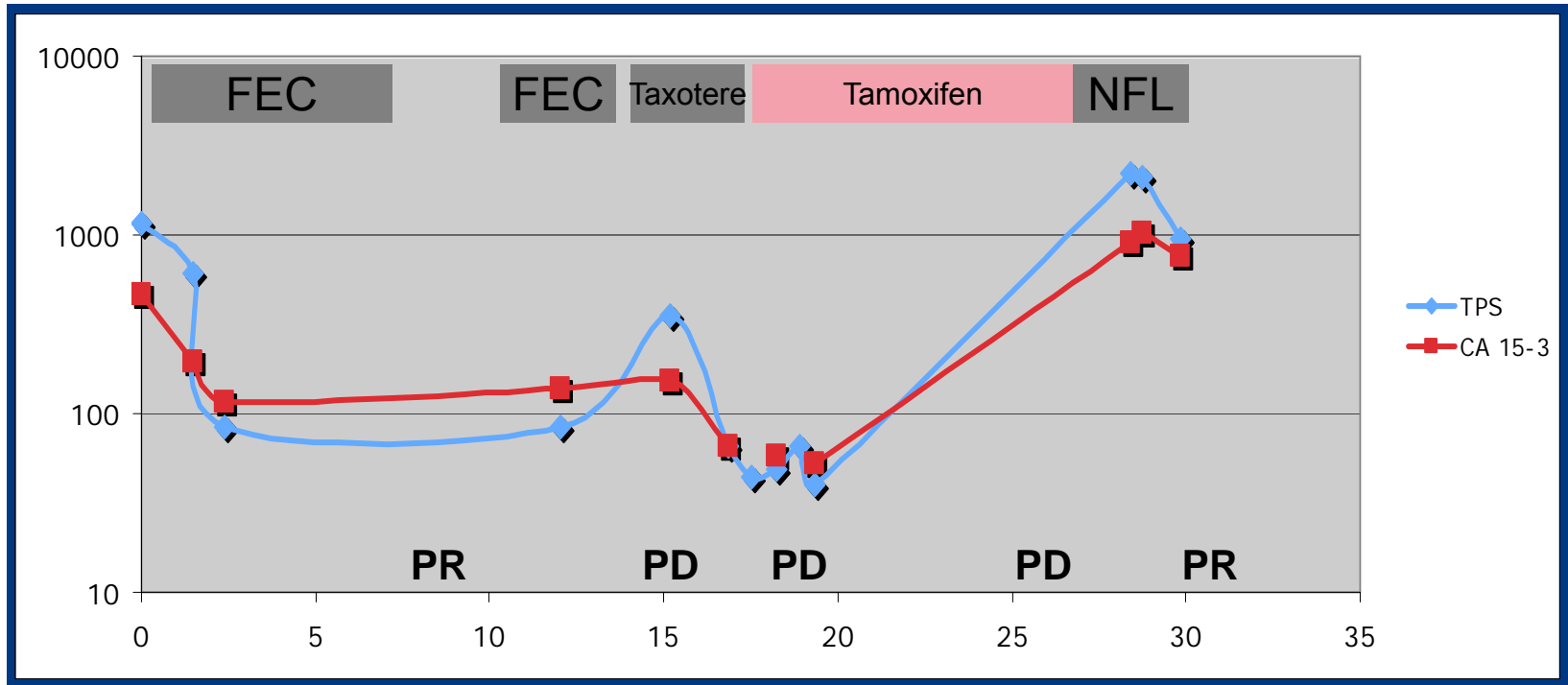


Regular analysis using the combination CA15-3 and TPS in patients with advanced breast cancer for

- follow-up
- diagnosis of suspicious metastatic disease
- differential diagnosis
- therapy monitoring



TMs & therapy response



PR partial response
PD progressive disease

Cut points applied: TPS 80 U/l, CA15-3 30 U/ml

Lindman J Tumor Marker Oncol 2000
Einarsson Anticancer Res 2000

Pretreatment TM levels



“Diagnostic sensitivity” baseline

TPA	81%	TPA+TPS	87%
TPS	81%	TPA+CA 15-3	89%
CA 15-3	79%	TPS+CA 15-3	92%

Sensitivity - dominant site of disease

	visceral/bone	soft tissue/loc.adv.
TPA	91%	50%
TPS	94%	44%
CA 15-3	89%	50%
TPS + CA 15-3	98%	75%

*Lindman J Tumor Marker Oncol 2000
Einarsson Anticancer Res 2000*

Breast cancer - summary



- Pretreatment TPS correlated with prognosis
- High sensitivity visceral/bone metastases
- Therapy response vs. decreasing TPS
TPS 69 - 84% CA 15-3 46 - 68%
- Disease progression vs. increasing TPS
TPS 82 - 85% CA 15-3 30 - 62%

Combination TPS & CA 15-3 >90%



TPS[®]

Advanced Prostate Cancer

Prostate cancer management

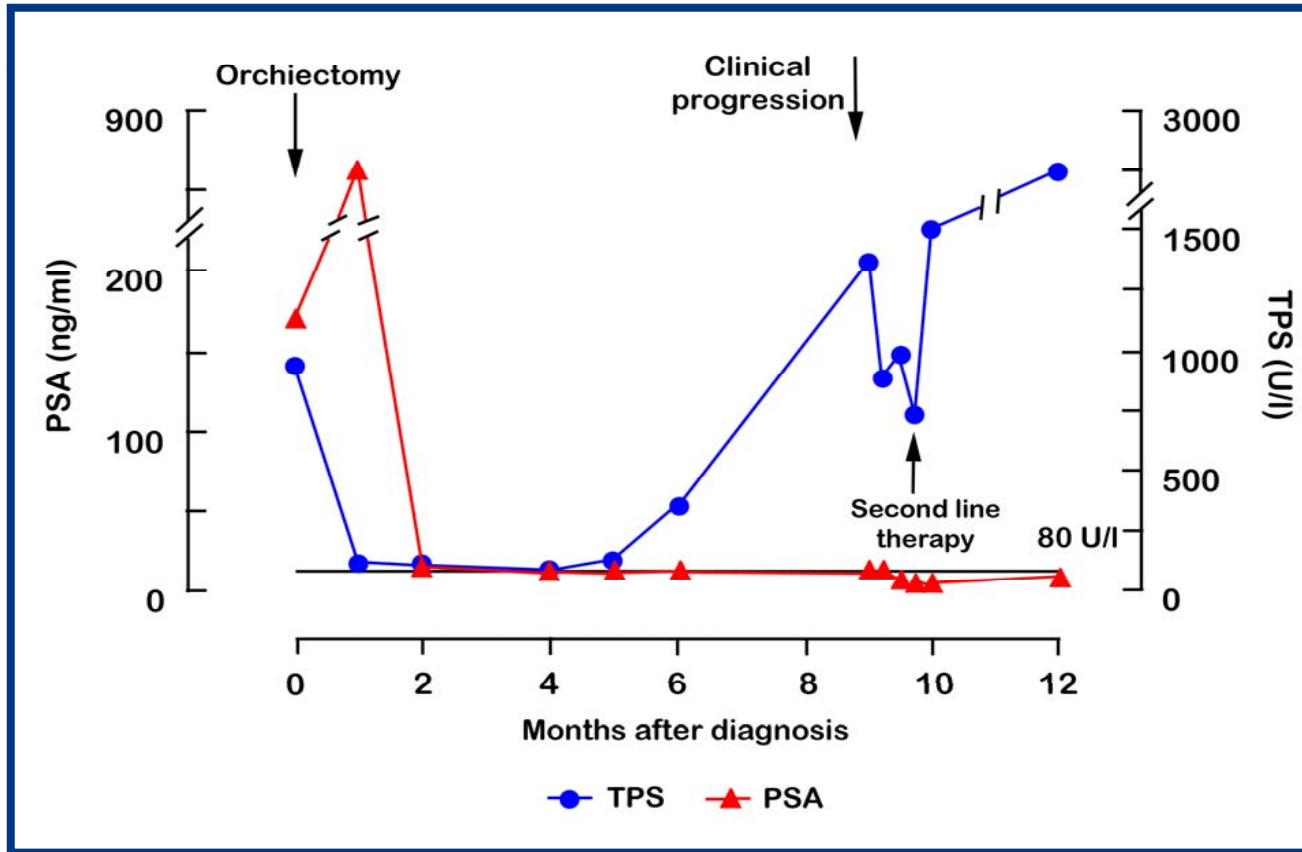


Regular analysis using the combination PSA and TPS in patients with advanced prostate cancer for

- follow up
- differentiation of stable & progressive disease
- therapy monitoring of hormone refractory metastasized disease
 - therapy effect
 - identification of patients with high progression risk (pretreatment prognosis)

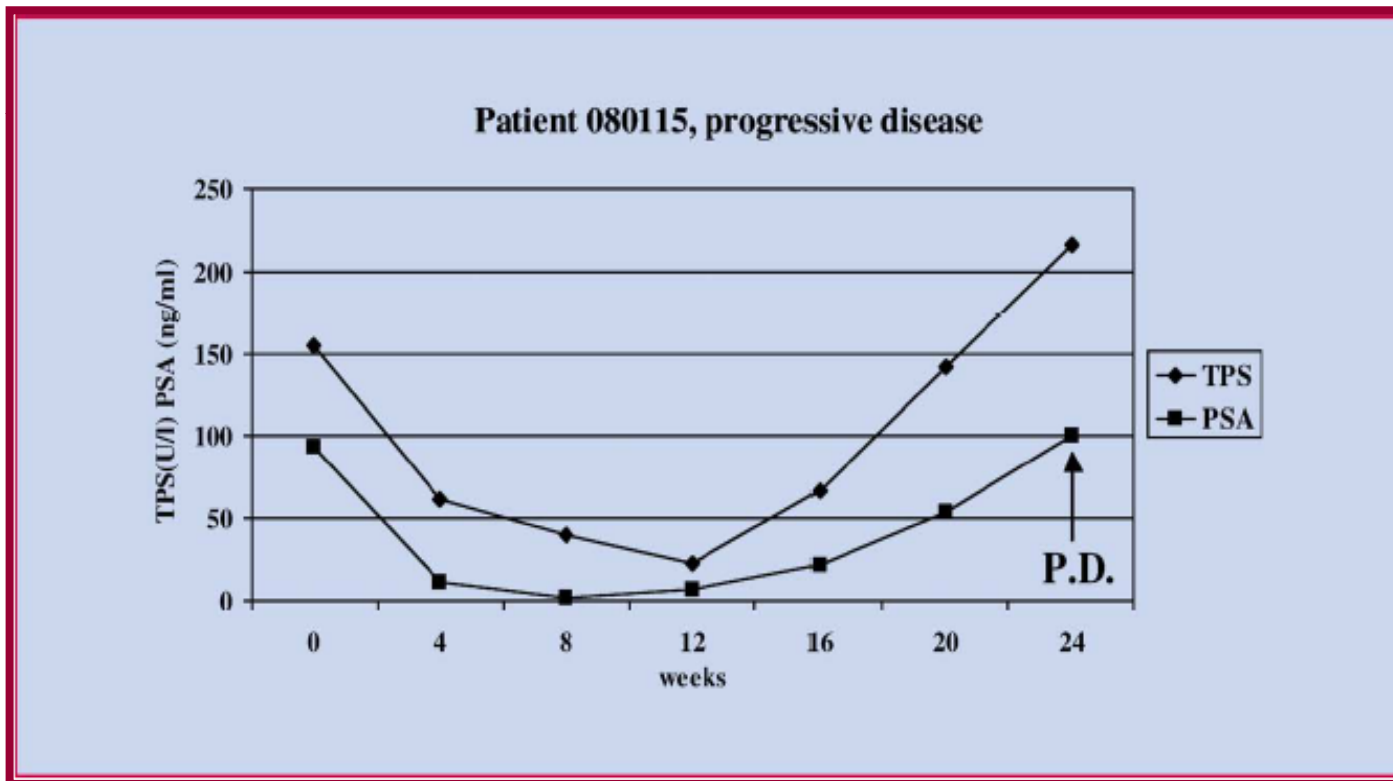


Therapy response TPS & PSA



Kramer J Urol 1997

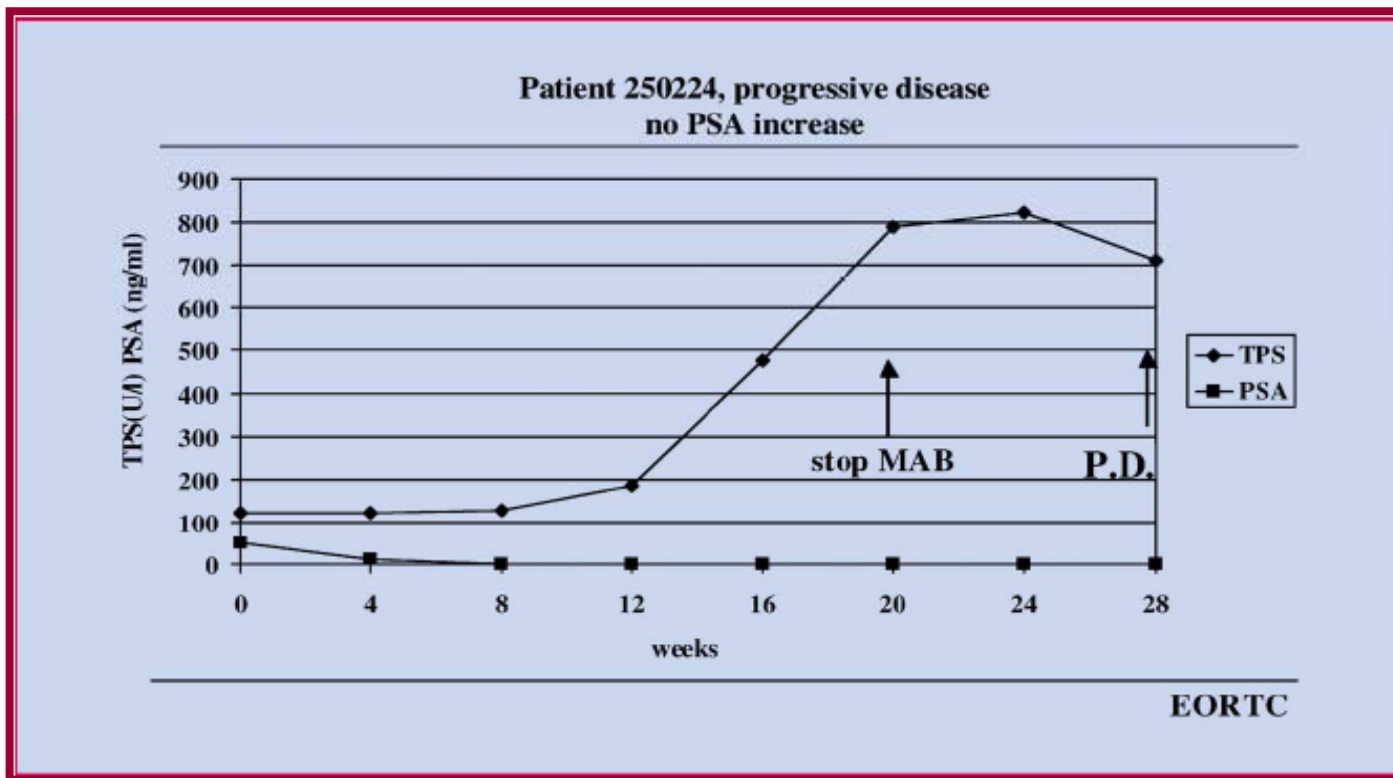
EORTC - Therapy response



Patient during MAB. Simultaneous decrease and increase in TPS and PSA. Clinical progression (24 weeks). *EORTC protocol 30954*

Kil Eur Urol 2003

EORTC - Therapy response



Patient during MAB. Treatment stopped at PSA nadir according to protocol (w. 20). TPS remains increased. Clinical progression (28 weeks), PSA shows normal levels. *EORTC protocol 30954*

Kil Eur Urol 2003

Prostate cancer - summary



- Pretreatment TPS significantly associated with prognosis
 - elevated TPS levels = 3.8 increased risk for disease progression
- Therapy monitoring and follow up
 - response vs. decreasing levels; TPS 90% PSA 64%
 - progression in high grade PSA-negative disease; TPS increase in 28% of the patients
- EORTC recommends combined use of TPS and PSA in patients with metastasized prostate cancer

Kramer J Urol 1997

Kil Eur Urol 2003



TPS[®]

Advanced Gastrointestinal Cancer

GI cancer management

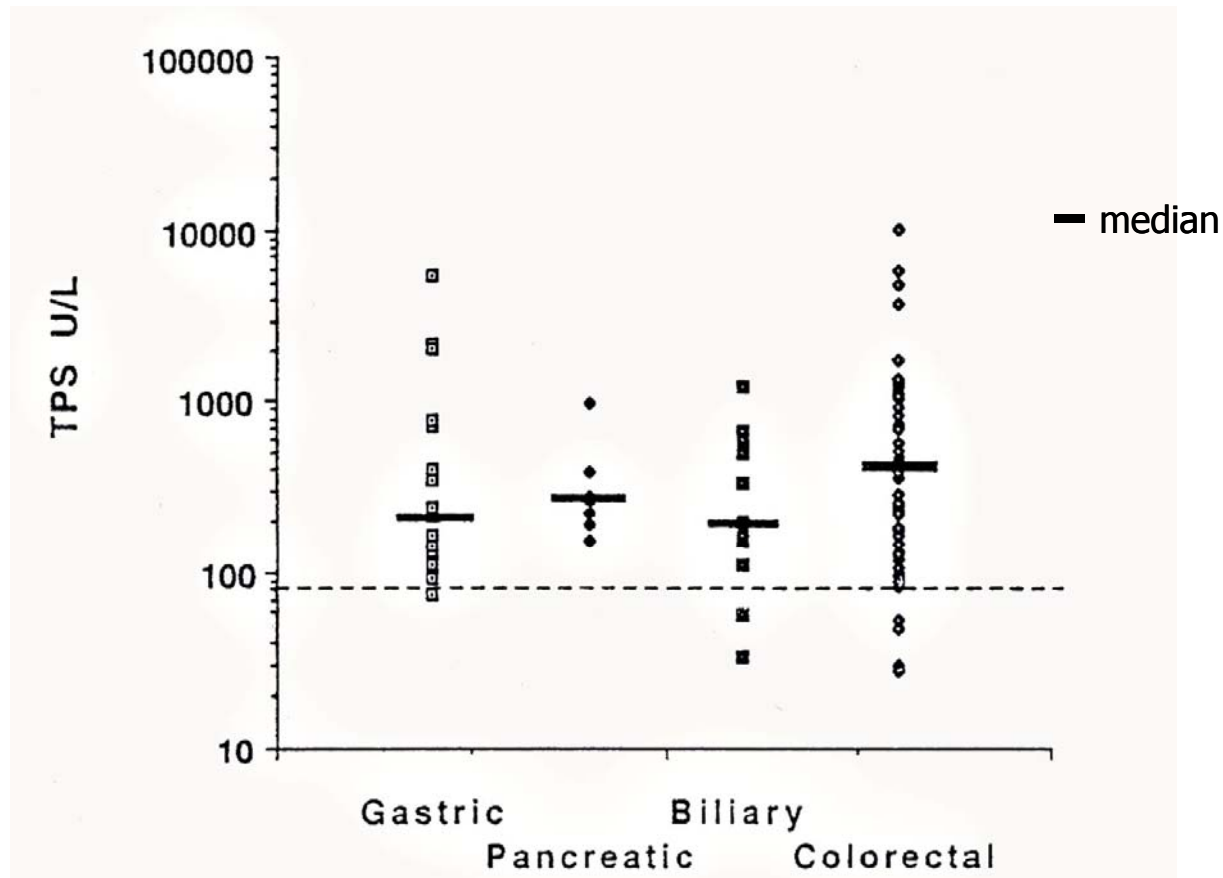


Regular analysis using TPS in patients with advanced gastrointestinal cancer

- therapy monitoring
 - early predictor of palliative therapy response
 - replace/reduce number of radiological assessments
- pretreatment prognosis
 - baseline value shows correlation to survival

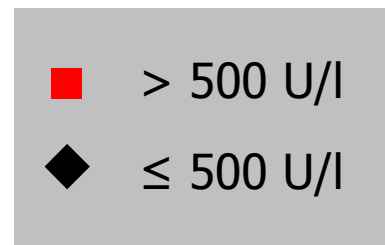
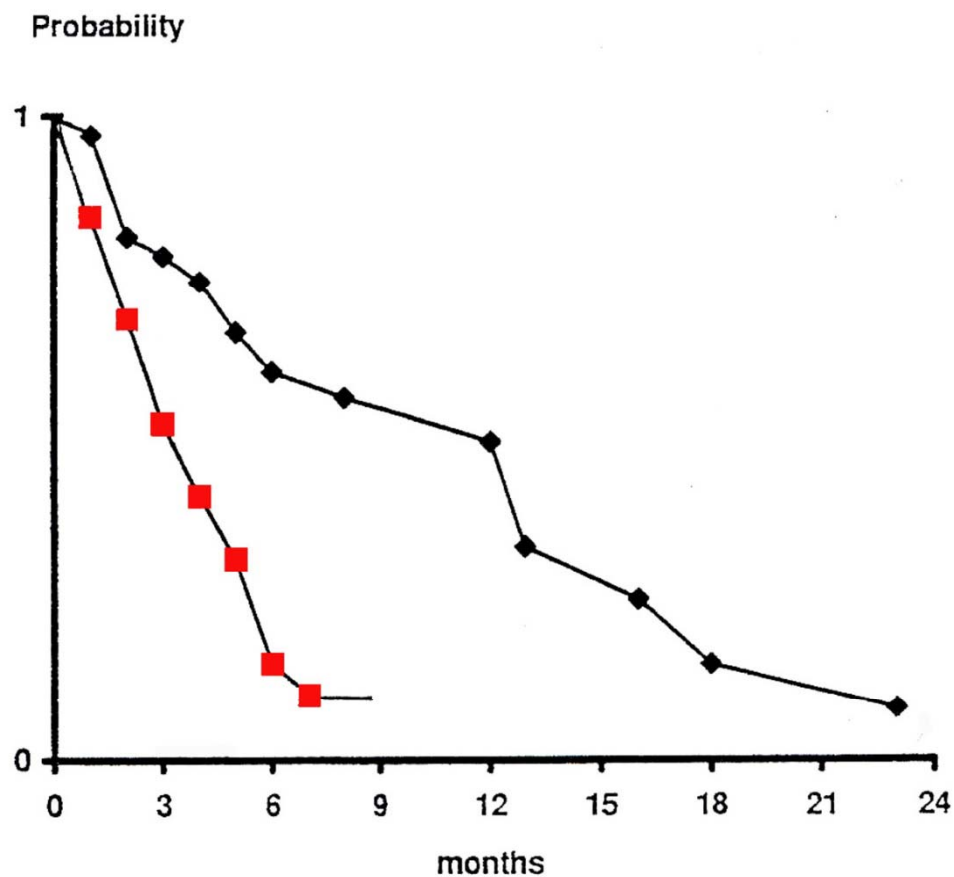
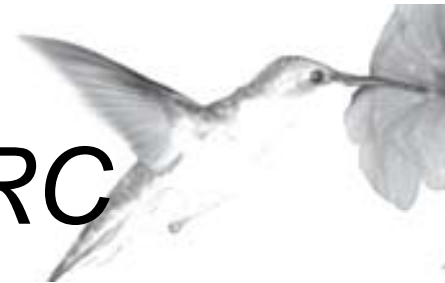


Diagnosis vs. baseline TPS



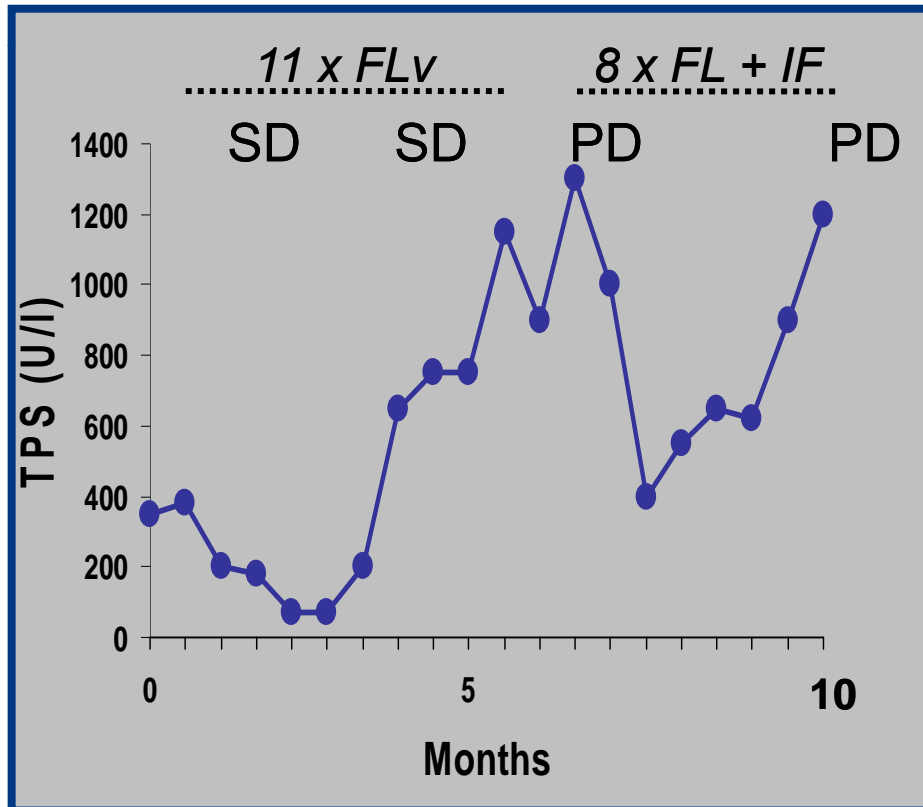
Glimelius, Einarsson Acta Oncol 1996
Berglund, Einarsson Ann Oncol 2002

TPS - Survival analysis in CRC



Glimelius, et al. Acta Oncol 1996
Berglund, et al. Ann Oncol 2002

Therapy response by TPS



Colorectal cancer patient
during chemotherapy

SD stable disease
PD progressive disease

Glimelius, et al. Acta Oncol 1996
Berglund, et al. Ann Oncol 2002

GI cancer - summary



- Elevated TPS levels is found in $>90\%$ of GI patients prior to treatment (baseline)
- Normal baseline levels correlates to a favorable outcome
 - prolonged survival
 - higher rate objective response
- TPS decrease $>50\%$ after two therapy courses highly correlates to a favorable treatment outcome (90% PR/SD) or a subjective response (100%)



GI cancer – summary *cont.*



- Serial TPS measurements can with high accuracy early identify patients with advanced gastrointestinal cancer who will not benefit from the treatment (palliative chemotherapy)
 - Indications already after 2 – 4 weeks





TPS®

Ovarian Cancer

Ovarian cancer management

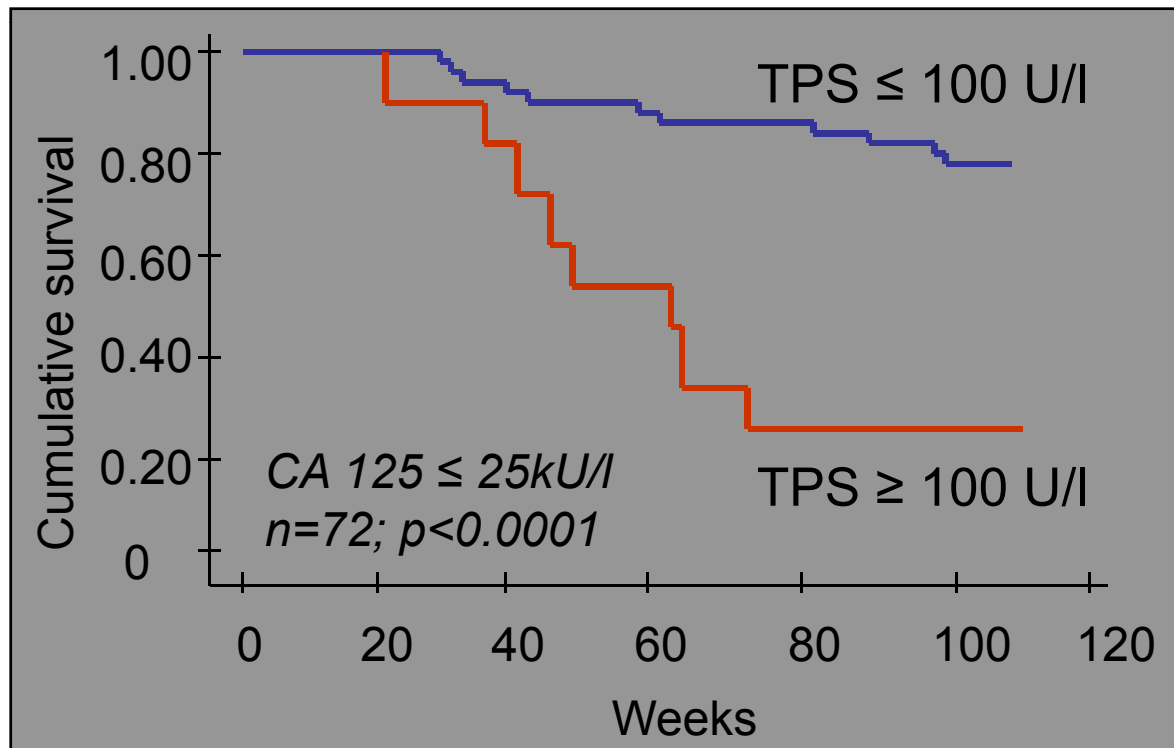


Regular analysis using the combination CA125 and TPS in patients with advanced ovarian cancer to

- predict clinical outcome
 - added prognostic value after chemotherapy
- establish response to treatment
 - increased sensitivity during therapy monitoring



Cumulative survival by TPS



*CA125 ≤ 25 kU/l
FIGO stage III + IV*

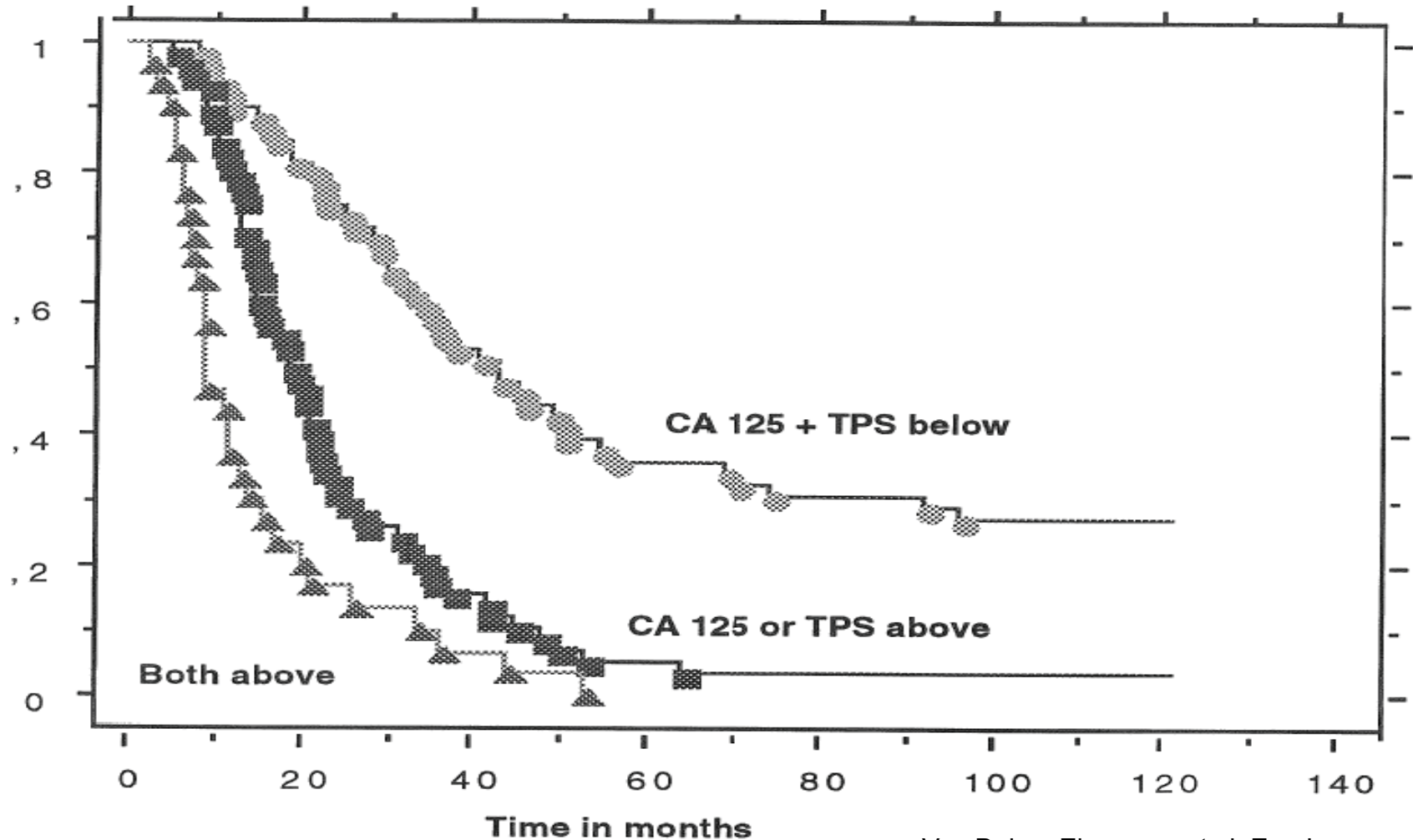
van Dalen, Einarsson et al.
Gynecol Oncol 2000

Two-year overall survival in ovarian cancer patients using TPS as discriminator (100 U/l)

CA125 and TPS in OvCa - clinical follow up after 6 cycles (carboplatin/Taxol)



Cum. Survival



Ovarian cancer - summary

- Advanced ovarian cancer patients with low or elevated CA125 and TPS after 3 chemotherapy courses demonstrates prognostic significance - clinical follow-up 1, 2 and 10-years
 - Both TPS and CA125 are independent factors of 2-years overall survival
 - TPS alone has additional prognostic value
 - TPS is a single independent factor predictive of 1-year overall survival
 - FIGO III/IV patients with TPS and CA125 below discrimination level showed good prognosis and elevated levels bad prognosis after 10-years clinical follow-up



Combination of cancer markers in different carcinomas

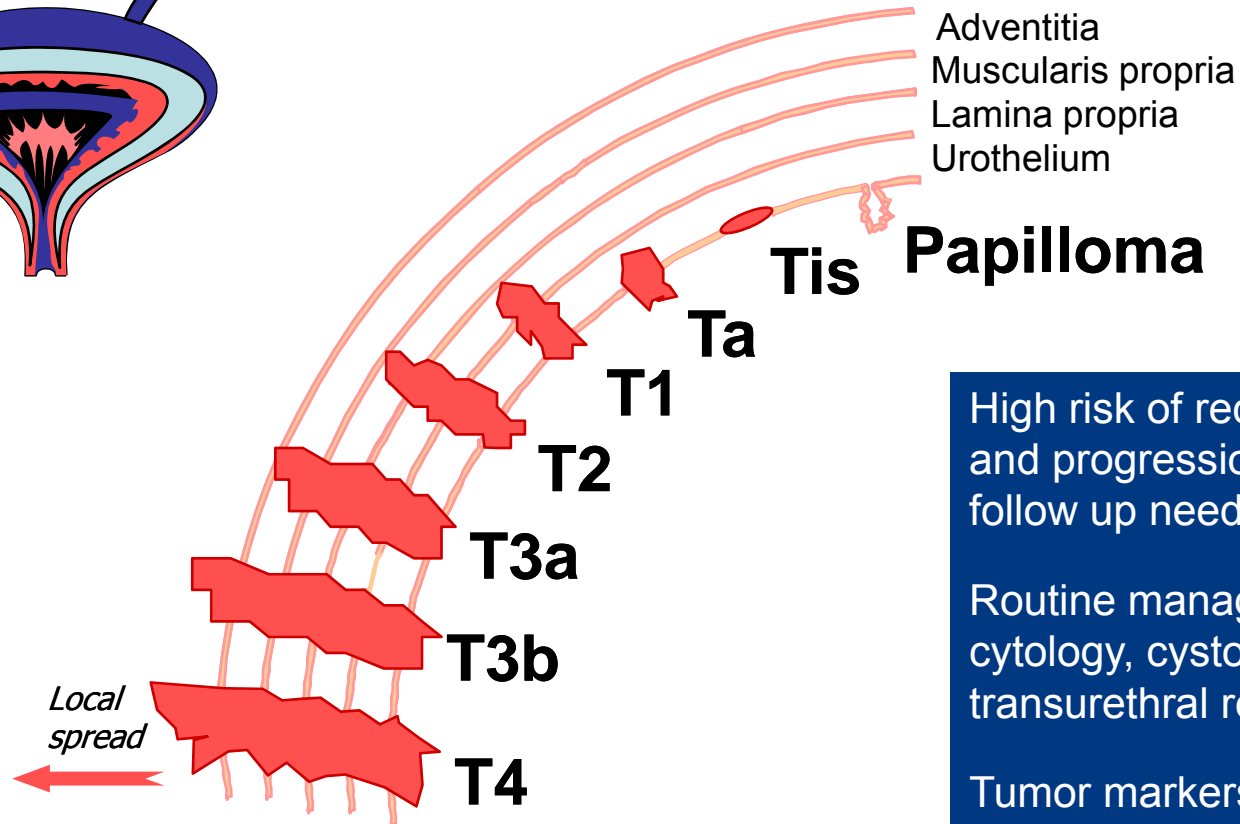
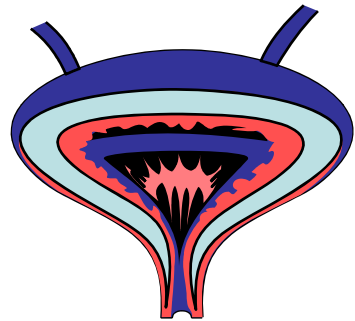


Indication	TM Combination
Breast cancer	TPS & CA 15-3
Prostate cancer	TPS & PSA
Colorectal cancer	TPS & CEA
Pancreatic cancer	TPS & CA 19-9
Ovarian cancer	TPS & CA 125



UBC[®] Urinary Bladder Cancer

Bladder Cancer - Clinic



High risk of recurrence and progression – long follow up needed

Routine management; cytology, cystoscopy, and transurethral resection

Tumor markers?

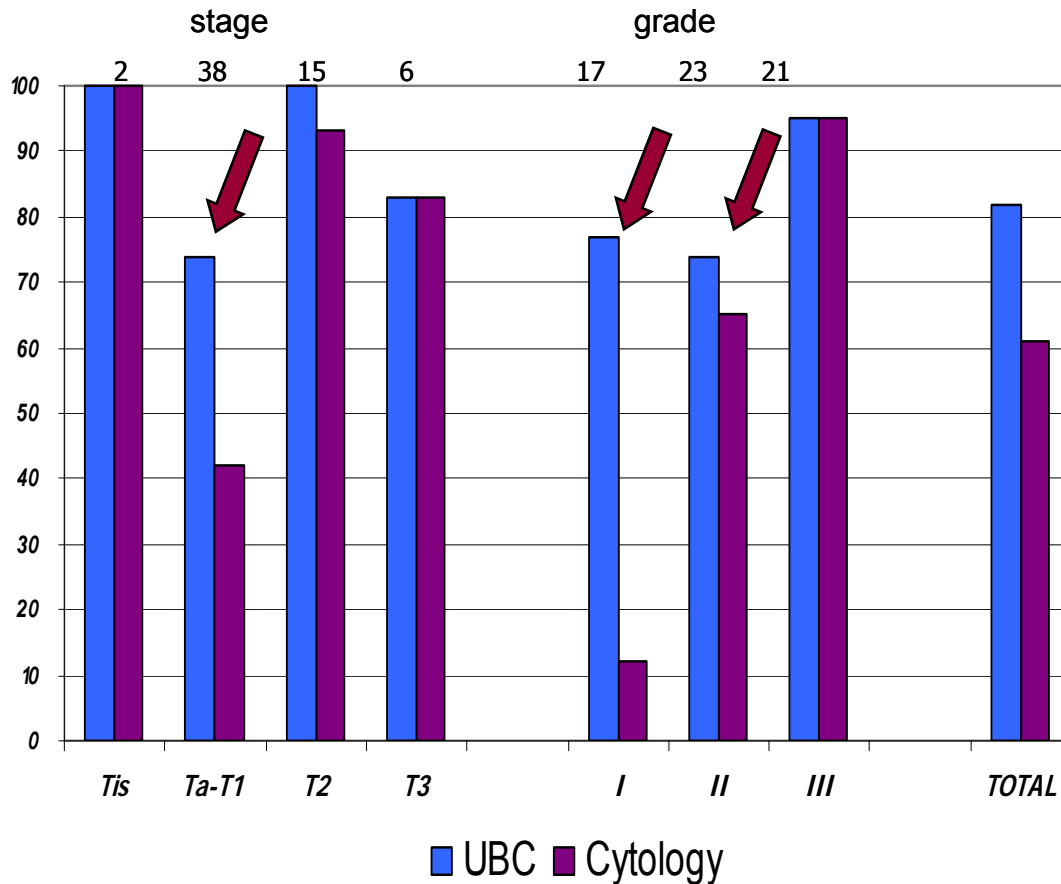
TMs in Bladder Cancer



- Sensitive and non-invasive adjunctive tool in primary disease
- Possibility to postpone and reduce the number of cystoscopies during follow-up
 - Individualized surveillance schedules



UBC & Voided Urine Cytology

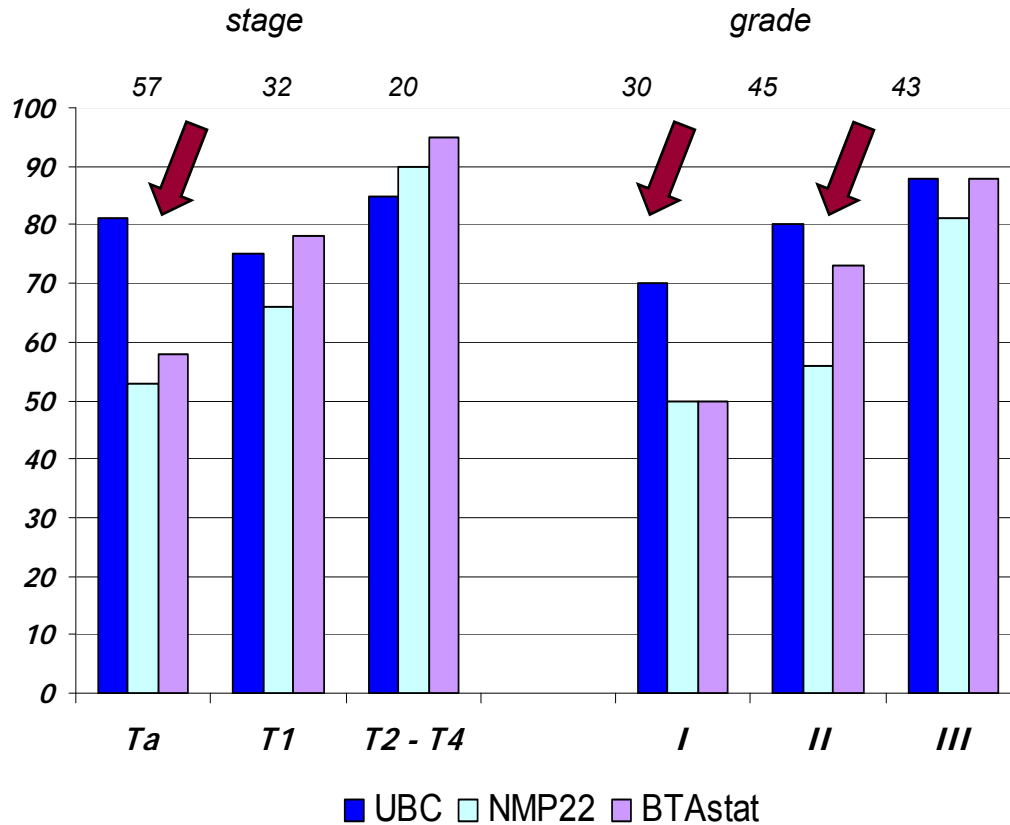


	Sensitivity
VUC	61%
UBC	82%

Sumi, Clin Chim Acta 2000



UBC vs. BTAstata & NMP22



	Sensitivity
BTAstata	73%
NMP22	64%
UBC	81%
UBC & BTAstata	92%

Giannopoulos J Urol 2001

Comparative evaluations

UBC 82% vs. Cytology 61%

UBC 81% vs. BTAstata 73%

NMP22 64%

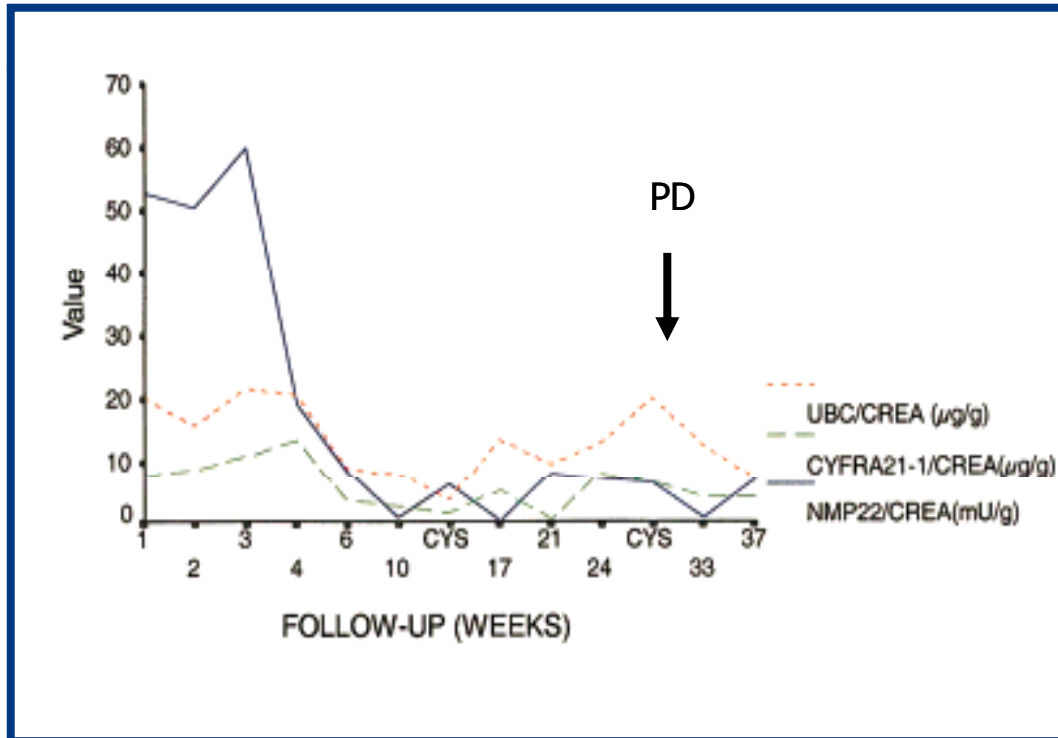
Excluding patients with e.g. urinary tract infections increased UBC sensitivity to 87%

UBC shows significantly better sensitivity both for lower stage and lower grade urinary bladder tumors

Combination UBC & BTAstata 92%



TMs vs. Scheduled cystoscopies



Serial UBC could detect recurrent disease earlier than scheduled cystoscopies in;
87% follow-up
67% monitoring

Serial negative results indicated disease free follow-up status in 87%

Patient with resected superficial bladder tumor monitored during therapy. Initial TM decrease after surgery is followed by peaking TM increases during follow-up, reflecting progressive disease as detected at scheduled cystoscopic examination week 25.

Sanchez-Carbayo Cancer 2001

UBC[®] Added clinical value



- Serial tumor markers can detect recurrence earlier than scheduled cystoscopies
 - Persistent negative tumor marker results strongly indicates a disease free status
- Methodology comparisons
 - UBC generally superior to cytology
 - UBC show better overall performance than BTAstat and NMP22; including higher sensitivity for lower stage/grade tumors
 - Combination UBC & BTAstat yields >90% sensitivity

Cytokeratin panel - Focus



TPS®

Breast cancer

Prostate cancer

Gastrointestinal cancer

UBC®

Urinary bladder cancer

TPAcyk™

General epithelial cancer



TPAcyk[®] Epithelial Cancer

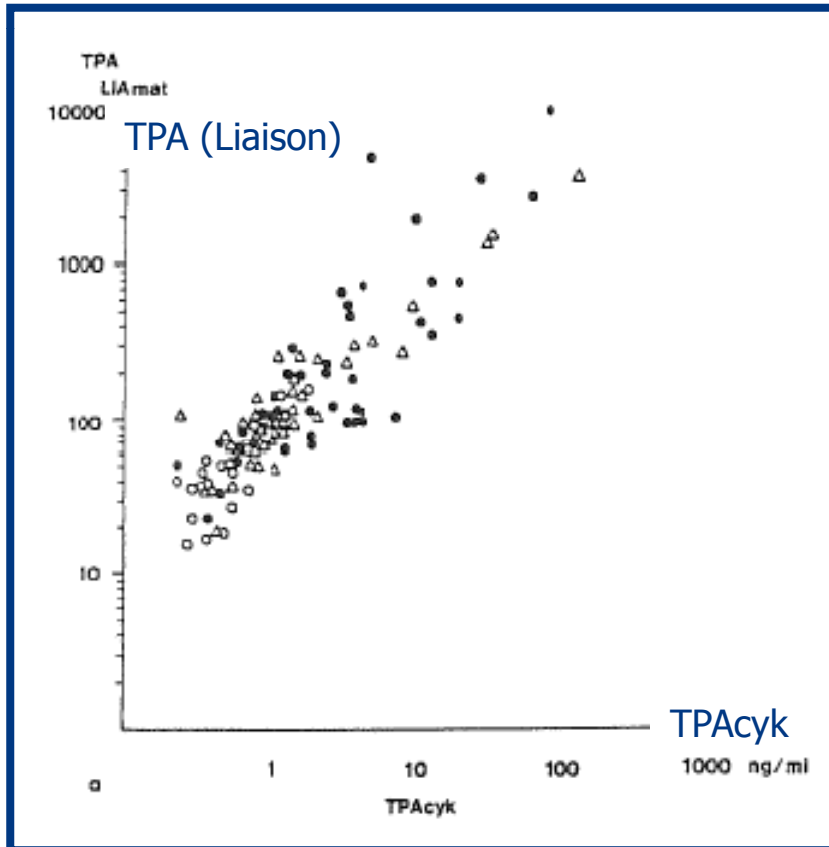
TPAcyk carcinoma marker



- TPAcyk is a general cytokeratin marker
 - particularly for treatment monitoring and follow-up of epithelial carcinomas
- TPAcyk is a tumor cell activity marker
 - cytokeratins 8 and 18
- TPAcyk is a TPA tumor marker
 - supported by the clinical background of the TPA concept
 - in clinical routine for > 20 years



TPAcyk Comparative data

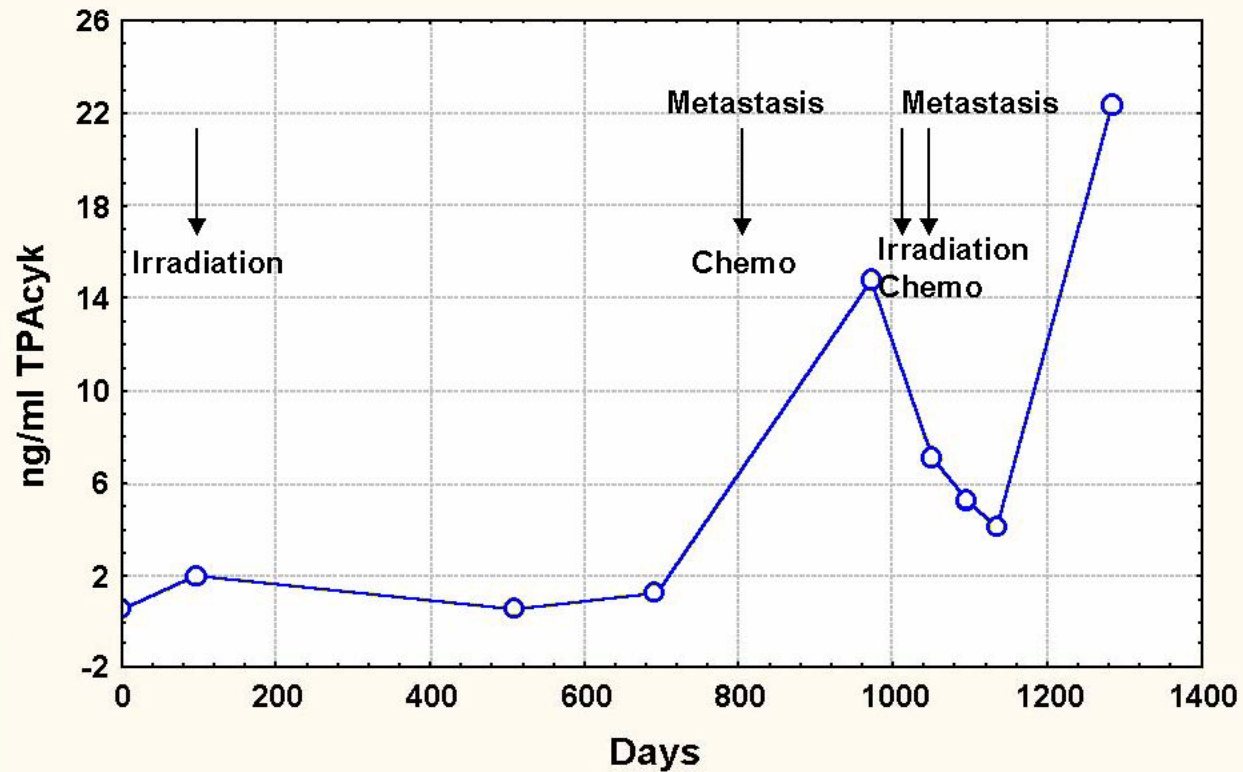


Correlation analyses TPAcyk and TPA

	Controls	Benign	Malign
TPA Lia	0.879	0.986	0.955
TPA Irma	0.880	0.958	0.952

Bahlo GIT Labor-Medizin 1993

Patient management by TPACyk



Wiklund, unpublished data

TPAcyk™ - Short Summary



- TPAcyk is a Tissue Polypeptide Antigen marker
 - TPAcyk and TPA show >0.96 correlation
- TPAcyk indicates ongoing tumor cell activity
 - particularly useful in treatment monitoring and patient follow-up
- Elevated TPAcyk often precedes clinical confirmation of tumor progression with long lead times



MonoTotal™ in Non Small Cell Lung Cancer

Potential marker for diagnosis and management of patients with NSCLC

Significantly higher values in sera in comparison with other cytokeratins

More sensitive than other conventionally used cytokeratin tumor markers

Cytokeratin Tumor Markers



Valuable in

- disease prognosis
- diagnosis advanced disease
- therapy monitoring
- patient follow-up



Early and distinct response upon therapy failure or success, and tumor recurrence

- long lead times to clinical manifestation compared to conventional methods



Thank you for your attention!