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Thema Tumormarkers*

Practice guidelines for tumour marker use in the clinic

C.M. STURGEON

Increasing pressure to provide health care based on "best practice" has stimulated local, national and international groups to develop guidelines in a number of clinical areas, particularly in cancer medicine, where diagnostic procedures are often invasive and therapy expensive (1). The broad recommendations of the American Society of Clinical Oncology (ASCO) (2) have recently been complemented by more detailed guidelines from the National Academy of Clinical Biochemistry (NACB) (3) in the United States and the European Group for Tumour Markers (EGTM) (4) in Europe. Based on expert opinion and published reports, these guidelines include recommendations about which tumour markers are likely to be most helpful in given clinical circumstances. The NACB and EGTM guidelines also highlight requirements and pitfalls of tumour marker measurements in the pre-analytical, analytical and post-analytical phases.

Recommendations relating to the pre-analytical phase

The pre-analytical phase is in many respects the most important phase of laboratory analysis. Errors in the pre-analytical phase reportedly occur up to ten times as often as in the analytical phase, are often difficult to identify and if unrecognised can seriously compromise patient care. Care and attention to detail when requesting a tumour marker measurement is therefore essential.

Selecting the most appropriate tumour marker

Tumour markers may be regarded as surrogate indicators that can increase or decrease a clinician's suspicion that a future clinically important event will or will not occur, and/or that a specific treatment will reduce its likelihood (3). Their value is in permitting an assessment of risk that should enable therapy to be offered to those patients most likely to benefit, while reducing exposure to toxicities for those who would not benefit. Selection of the most appropriate marker should therefore take heed both of the clinical question – whether risk assessment, screening, diagnosis, prognosis, prediction or monitoring – and of the reliability of the separation in outcomes for marker positive and marker negative patients.

Unfortunately there are as yet relatively few well-designed and validated prospective studies for individual tumour markers. Many of the studies reported in the literature have not been validated thoroughly (i.e. in the same assay, using the same cut-off limits and types of patients), while in others statistical significance ($p<0.05$) in outcomes of two groups separated by marker results has incorrectly been regarded as evidence of clinical utility, which is not always the case. With these caveats in mind, it is nevertheless possible to make some recommendations about the most appropriate markers in given clinical situations, as summarised in Table 1 for some major cancer sites (3). [It should be noted that these recommendations apply to requests for tumour marker measurement in routine clinical practice and not necessarily to their use in clinical trials where other considerations may apply.]

Optimising specimen collection

There is no strong evidence of diurnal variation for most markers, so specimens can be taken at any time of day. While the advent of the electronic health record should make it possible to link the ordering process with advice about relevant pre-analytical concerns, pending such developments it is important for both clinical and laboratory staff to be aware of clinical interventions and conditions which may transiently increase tumour markers concentrations. For example, blood for PSA should be taken before any manipulation of the prostate, and blood for CA125 should not be taken during menstruation, which may increase the serum concentration two to three-fold. PSA may also be increased markedly in men with urinary tract infections and prostatitis while CA125 may be mildly elevated in endometriosis and the first two trimesters of pregnancy and markedly raised in any patient with benign ascites. CA19.9 may be significantly increased in patients with cholestasis and patients in this category should be noted on the clinical report. The effect of medication and other treatment should also be considered: 5α-reductase inhibitors cause a median decrease in *f*PSA concentration of approximately 50%. Transient increases in tumour marker concentrations can also occur following

Correspondence: dr Catherine M. Sturgeon, clinical researcher. Dept. of Clinical Biochemistry, Royal Infirmary of Edinburgh, 51 Little France Crescent, Edinburgh EH16 4SA, UK
E-mail: cs@csturgeon.net

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chemotherapy. Cannabis may increase serum levels of hCG while smoking may slightly increase apparent CEA levels in some immunoassays.
The laboratory should provide clear and readily available advice about the appropriate tube type for each test, so as to ensure that manufacturers' instructions are always followed. Standardised conditions of specimen collection and fixation are crucial for immunohistochemical analyses. Although most tumour markers are reasonably stable, serum or plasma should be separated and stored appropriately as soon as possible. At high ambient temperature the potential influence of transit time on analyte results should be considered. As for other analytes, the majority of pre-analytical errors for tumour markers will be simple specimen handling errors – hemolyzed specimens, insufficient specimens, and incorrect specimens – and their occurrence should be minimized by adherence to good laboratory practice. Extra vigilance is required in ensuring that correct tumour marker results are reported, since reporting erroneous results is more likely to cause patients undue alarm than is the case for many other laboratory tests.

Recommendations relating to the analytical phase
In the analytical phase it is essential for satisfactory measurement of any analyte that laboratories ensure that methods used, whether immunoassay or immunohistochemical, are well validated and that their performance is carefully monitored. Implementation of rigorous Internal Quality Control (IQC) procedures and participation in well-designed Proficiency Testing [External Quality Assessment (EQA)] programs should help to ensure that methods are performing according to specification. The NACB has made recommendations for IQC and EQA provision

Table 1. Summary or current NACB recommendation for the use of tumour markers in specific malignancies

	Screening / early detection	Diagnosis / case-finding Staging / prognosis	Detecting	Monitoring recurrence	therapy
Testicular tumours	No tumour markers recommended	AFP, hCG, LDH	AFP, hCG, LDH	AFP, hCG, LDH	AFP, hCG, LDH
Prostate cancer	PSA, cPSA, %fPSA [with DRE]	PSA, cPSA, %fPSA [with DRE]	PSA, cPSA [with DRE & biopsy Gleason Grade]	PSA, cPSA	PSA, cPSA
Colorectal cancer	FOB [in subjects >50 years old; genetic testing in high risk subjects]	No tumour markers recommended	CEA	CEA	CEA
Liver cancer	AFP [in high risk subjects]	AFP	AFP	AFP	AFP
Ovarian cancer	CA125 [only in combination with TVUS for early detection in hereditary syndromes]	CA125 [post-menopausal women only]	CA125	CA125	CA125
Breast cancer	No tumour markers recommended	No tumour markers recommended	ER, PR, HER-2, uPA, PAI-1	No tumour markers recommended	CA 15-3, CEA [monitoring advanced disease]
Gastric cancer	No tumour markers recommended	No tumour markers recommended	No tumour markers recommended	No tumour markers recommended	No tumour markers recommended
Pancreatic cancer	No tumour markers recommended	CA19-9 [if used, **only** with CT or EUS and in an appropriate clinical context]	CA19-9	No tumour markers	CA19-9 [during palliative therapy with imaging tests or after potentially curative surgery]
Cervical cancer	No tumour markers recommended	SCC [possibly in squamous cell cervical carcinoma]	SCC [possibly in squamous cell cervical carcinoma]	SCC [possibly in squamous cell cervical carcinoma]	SCC [possibly in squamous cell cervical carcinoma]

AFP, α-fetoprotein; CEA, carcinoembryonic antigen; CT, computed tomography; DRE, digital rectal examination; ER, estrogen receptor; EUS, examination under ultrasound; FOB, faecal occult blood; hCG, human chorionic gonadotropin; LDH, lactate dehydrogenase; MIA, melanoma inhibiting activity; PAI-1, plasminogen activator inhibitor-1; PR, progesterone receptor; PSA, prostate specific antigen; cPSA, complexed PSA; %fPSA, % of free (uncomplexed) PSA to total (complexed + free) PSA; SCC, squamous cell carcinoma antigen; TVUS, transvaginal ultrasound; uPA, urokinase plasminogen activator.

that are generally applicable to all analytes, several factors being especially relevant for tumour marker measurement (3, 4).
Excellent precision and reproducibility (intra-assay variability <5%; inter-assay variability <10%) is essential particularly near critical clinical decision points, for example in screening programs (e.g. when using PSA to select asymptomatic patients for biopsy) or where chemotherapy may be instituted on the basis of a rising tumour marker level in the absence of other scan evidence (e.g. when monitoring testicular cancer patients with AFP and hCG). Long-term assay stability, which can be readily assessed by proficiency testing schemes, should also be demonstrated since tumour markers are often measured for cancer patients over months or years.
Such long-term monitoring presents major analytical challenges as patients may be treated in different hospitals using different methods and laboratories may change tumour marker methods during the relevant time period. Data from proficiency testing schemes confirm that there are still significant between-method differences in results, with coefficients of variation in excess of 20% observed for some tumour markers. Poor calibration and differences in the specificity of antibodies used as well as method design all contribute to this variation. A number of international initiatives to address these issues are currently in progress. For the present, considerable care must be taken when changing tumour marker methods or when interpreting cumulated results for individual patients that have been obtained in different methods.

Differences in method design contribute not only to the numerical differences in results observed, but also influence method robustness to clinically relevant interferences. These include cross-reactions with closely related molecules, the high dose hook effect and interference from heterophilic or human anti-mouse antibodies. Maintaining vigilant awareness of the potential for such interference is very important and requires good understanding of the characteristics of the assays being used. Regular dialogue between laboratory and clinical staff should be actively promoted, since early discussion and investigation of any results that are not in accord with the clinical picture is likely to facilitate early identification of erroneous results caused by interference.

Recommendations relating to the post-analytical phase
In the post-analytical phase, dialogue can be encouraged by provision of brief clinical information by clinicians when requesting tumour marker measurements. It is recommended that laboratory reports for tumour markers include fully cumulated results, an appropriate reference interval and the name of the assay method used, together with an indication of whether any change in marker level is significant and, if appropriate, whether any change of method is likely to have affected interpretation of the trend in marker level (3, 4).
Laboratories should also be actively involved in ongoing audit of the clinical utility of the tumour marker results they provide and of their influence on clinical outcome. Proficiency testing schemes can also contribute by undertaking occasional surveys to compare practice in participating laboratories, highlighting differences in reference intervals, reporting practice and interpretation of clinical results. Such surveys can also provide an indication of how effectively guidelines such as those outlined here are being implemented in routine practice.

References

1. Sturgeon CM. Practice guidelines for tumour marker use in the clinic. Clin Chem 2002; 48: 1151-9.
2. Bast RC, Radvin P, Hayes DF, Bates S, Fritsche H, Jessup JM, et al. 2000 update of recommendations for the use of tumour markers in breast and colorectal cancer: clinical practice guidelines of the American Society of Clinical Oncology. J Clin Oncol 2001; 19: 1865-78.
3. NACB Practice Guidelines and Recommendations for use of tumour markers in the clinic. Available in electronic form at http://www.aacc.org/NR/rdonlyres.
4. European Group on Tumour Markers (EGTM) Consensus Recommendations. Available in electronic form at http://egtm.web.med.uni-muenchen.de/index2.html.