Expression of the pNR-2/pS2 protein in diverse human epithelial tumours

J.A. Henry1, M.K. Bennett1, N.H. Piggott1, D.L. Levet1, F.E.B. May1 & B.R. Westley1

1Department of Pathology, University of Newcastle upon Tyne, Royal Victoria Infirmary, Newcastle upon Tyne; 2Department of Pathology, Freeman Hospital, Newcastle upon Tyne, UK.

Summary The pNR-2/pS2 protein is regulated by oestrogens in breast cancer cell lines. This report describes a systematic survey of pNR-2/pS2 expression in a number of common epithelial tumours. Expression was evaluated immunohistochemically in an archival series using antisera raised against the C-terminus of the pNR-2/pS2 protein. Expression of pNR-2/pS2 by malignant epithelial tumours was widespread. Intense immunohistochemical staining was found in tumour cells in a proportion of pancreatic (6/8), large intestinal (7/12), gastric (9/16) and endometrial (4/12) carcinomas. Positive staining for the pNR-2/pS2 protein was also found in both benign and malignant ovarian epithelial tumours and was very significantly associated with mucinous differentiation (P < 0.0001). Small numbers of carcinomas of bladder (2/10) and prostate (2/7) showed less intense staining and single examples of cervical carcinoma (1/7) and lung carcinoma (1/19) stained positively. None of the renal carcinomas (0/16) examined stained positively. Positive staining showed no correlation with gender. Although there are reports of oestrogen receptor expression in most of the tumour types considered, the possibility of other regulatory influences must also be considered. The pNR-2/pS2 protein may well have a more general role in human epithelial neoplasia than hitherto realised.

The pNR-2/pS2 messenger RNA was originally detected in oestrogen-responsive breast cancer cell lines by virtue of its regulation by oestrogen (Masiakowski et al., 1982; Prud'homme et al., 1985; May & Westley, 1986). The pNR-2/pS2 mRNA encodes a small, cysteine-rich protein of 84 amino acids (Jakowlew et al., 1984) which is secreted from breast cancer cells (Nunez et al., 1987) as a mature 60 amino acid protein (Rio et al., 1988a). Oestrogen regulation of the pNR-2/pS2 gene is conferred by a short enhancer region in the 5' flanking region of the gene (Berry et al., 1989). The function of the pNR-2/pS2 protein is unknown but it shows some structural similarity to small peptide growth factors such as insulin-like growth factor I and a high degree of homology to porcine pancreatic spasmolytic polypeptide (Rio et al., 1988b). In breast cancer the pNR-2/pS2 mRNA and protein are expressed predominantly in oestrogen receptor positive tumours (Rio et al., 1987; Henry et al., 1990; Henry et al., 1991) and high levels of the protein are predictive of favourable prognosis (Foekens et al., 1990). Expression of pNR-2/pS2 is also predictive of a favourable response to endocrine therapy in advanced breast cancer (Henry et al., 1991).

In normal tissue, the pNR-2/pS2 protein is expressed in gastric mucosa (Rio et al., 1988a), small intestinal mucosa and in normal breast epithelium (Piggott et al., 1991). Amongst malignant epithelial tumours, the pNR-2/pS2 protein has been detected in gastric carcinomas (Luqmani et al., 1989) and gynaecological cancers (Wysoczki et al., 1990).

To date there has been no systematic evaluation of the expression of this intriguing protein in primary epithelial tumours arising in different organs. We have recently raised a rabbit polyclonal antiserum to a synthetic peptide derived from the C-terminal region of the pNR-2/pS2 protein (Piggott et al., 1991). This antiserum is effective on conventionally fixed, paraffin embedded, histological material. We now report an immunohistochemical survey of pNR-2/pS2 expression in common epithelial tumours.

Materials and methods

Surgically resected tumours and tumour biopsies were fixed overnight in phosphate-buffered 4% formalin and representa-

Correspondence: J.A. Henry, Department of Pathology, University of Newcastle upon Tyne, Royal Victoria Infirmary, Newcastle upon Tyne NE1 4LP, UK.

Received 4 March 1991; and in revised form 28 May 1991.

© Macmillan Press Ltd., 1991
The specific detection of pNR-2/pS2 expression was confirmed by the absence of staining when the antisera was preabsorbed with the synthetic peptide that had been used as the immunogen. There was considerable variation in the proportion of tumour cells that expressed pNR-2/pS2 and in the intensity of staining.

The highest incidence of positive staining was found in pancreatic carcinomas (75%; Table I). Positive pNR-2/pS2 immunohistochemical staining was cytoplasmic and showed a tendency to perinuclear accentuation (Figure 1). Pancreatic tumours also contained some of most intensely stained cells and some of them contained high proportions of positively stained cells (Table I). Positive cytoplasmic staining was found in tumour cells scattered throughout the neoplastic glandular elements: background staining was minimal (Figure 1).

Intense cytoplasmic staining of tumour cells was also found in appreciable numbers of carcinomas of large bowel (58%), stomach (56%) and endometrium (33%; Table I). In all of the positive examples, tumour cell staining was again cytoplasmic with a tendency to perinuclear accentuation. In positively stained large bowel carcinomas both diffuse and focal patterns of staining were observed (Figure 2). In general, if large proportions of tumour cells stained, positively stained cells were present scattered diffusely throughout the tumour whereas staining tended to be more focal in large bowel carcinomas containing smaller proportions of positive cells. Focal positive staining was also present in apparently normal large bowel epithelium adjacent to one tumour. Both diffuse and focal patterns of staining were observed in positively stained gastric carcinomas. Positive staining was observed in tumours exhibiting both glandular and diffuse histological patterns (Figure 3a,b). Epithelial cells in adjacent uninvolved gastric mucosa showed the pattern of staining previously observed in normal gastric mucosa (Piggott et al., 1991). In all four positively stained endometrial carcinomas, the positive tumour cells were present scattered widely throughout the tumour: this included one tumour where only approximately 1% of tumour cells stained positively. Positive staining was confined to malignant epithelial cells; the stromal component only demonstrated weak background staining (Figure 4).

Ovarian tumours formed a particularly interesting group.

![Figure 1](image1.png)  **Figure 1** Positive immunohistochemical staining for pNR-2/pS2 in a proportion of cells in a pancreatic adenocarcinoma. Staining is cytoplasmic with a tendency to perinuclear accentuation (arrow). Bar = 50 μm.

![Figure 2](image2.png)  **Figure 2** A colorectal carcinoma with positive pNR-2/pS2 immunohistochemical staining of varying intensity in a proportion of tumour cells lining glandular structures (arrow). Bar = 50 μm.

| Primary site | Total no. cases | No. pNR-2/pS2 positive cases (%) | Proportion of male pNR-2/pS2 positive cases (%) | Proportion of female pNR-2/pS2 positive cases (%) | Positive tumours, mean proportion positively-stained cells (%) | Range |
|--------------|----------------|----------------------------------|-----------------------------------------------|-----------------------------------------------|-------------------------------------------------|-------|
| Pancreas     | 8              | 6 (75%)                          | 2/3 (67%)                                     | 4/5 (80%)                                     | 42%                                             | 7–57% |
| Large bowel  | 12             | 7 (58%)                          | 5/8 (63%)                                     | 2/4 (50%)                                     | 20%                                             | 4–60% |
| Stomach      | 16             | 9 (56%)                          | 4/7 (57%)                                     | 5/9 (55%)                                     | 27%                                             | 0.5–77%|
| Ovary        | 25             | 9 (36%)                          | –                                             | 9/25 (36%)                                    | 21%                                             | 1–61% |
| Endometrium  | 12             | 4 (33%)                          | –                                             | 4/12 (33%)                                    | 26%                                             | 1–56% |
| Prostate     | 7              | 2 (29%)                          | 2/7 (29%)                                     | –                                             | 2.5%                                            | 2–3%  |
| Bladder      | 10             | 2 (20%)                          | 2/7 (29%)                                     | 0/3 (0%)                                      | 30%                                             | 7–53% |
| Cervix       | 7              | 1 (14%)                          | –                                             | 1/7 (14%)                                     | 1%                                              | –     |
| Lung         | 19             | 1 (5%)                           | 0/14 (0%)                                     | 1/5 (20%)                                     | 62%                                             | –     |
| Kidney       | 16             | 0 (0%)                           | 0/5 (0%)                                      | 0/11 (0%)                                     | 0%                                              | –     |
| Breast       | 171            | 117 (67%)                        | –                                             | 117/171 (67%)                                 | 14.9%                                           | 1–81% |
In total 25 malignant tumours were stained, comprising 14 serous cystadenocarcinomas, eight mucinous cystadenocarcinomas and three unclassifiable, poorly differentiated carcinomas. In the nine positively stained tumours, strongly stained tumour cells were typically present scattered diffusely throughout the tumour, with only minimal stromal staining (Figure 5a). Further analysis of the pattern of staining in the ovarian tumours showed that there was a significant association between pNR-2/pS2 positivity and mucinous subtype (Table II; Fisher's exact probability = 0.0083). A group of benign serous and mucinous cystadenomas were stained immunohistochemically to investigate whether mucinous differentiation was associated with pNR-2/pS2 expression in benign tumours. In these benign tumours, positive staining was confined to epithelial cells lining cystic spaces and although more prevalent was often less intense than that observed in their malignant counterparts (Figure 5b). In general, positively stained cells were found scattered around the circumference of the cystic spaces in these benign tumours, but in some instances the pattern was more focal. The association between positive pNR-2/pS2 immunohistochemical staining and mucinous differentiation was significant in the group of benign tumours (Table II; Fisher's exact probability = 0.001). The association between pNR-2/pS2 expression and mucinous differentiation became even more highly significant when the groups of benign and malignant ovarian tumours were combined (Fisher's exact probability <0.00001).

Strong positive immunohistochemical staining was present in a large proportion of cells present diffusely throughout a single adenocarcinoma of lung. The remaining lung cancers (comprising eight adenocarcinomas and ten undifferentiated large cell carcinomas) did not appear to express pNR-2/pS2 (Table I).

Less intense immunohistochemical staining for pNR-2/pS2 was present in cells scattered throughout two transitional cell carcinomas of bladder and in small foci of cells present in two prostatic adenocarcinomas. A single cervical carcinoma contained a small focus of positively stained tumour cells. This was the only adenocarcinoma analysed in a group which otherwise comprised squamous carcinomas of cervix. None of the 16 renal adenocarcinomas examined showed any evidence of pNR-2/pS2 expression (Table I).
The pNR-2/pS2 protein was discovered in human breast cancer cells (Masiakowski et al., 1982; Prud'homme et al., 1985; May & Westley, 1986; Skilton et al., 1989) but its expression has since been reported in gastric (Luqmani et al., 1989) and gynaecological cancers (Wysocki et al., 1990). This report is the first survey of the extent of the pNR-2/pS2 expression in common human epithelial tumours. We have found that pNR-2/pS2 expression is a widespread phenomenon: of the tumours examined only renal adenocarcinomas did not show any expression at all (Table I). Convincing pNR-2/pS2 immunohistochemical staining was found in appreciable numbers of carcinomas of pancreas, large bowel, stomach, ovary and endometrium: weaker staining was found in a proportion of prostatic and bladder carcinomas. Single examples of both cervical and lung carcinomas stained positively. We detected a lower proportion of pNR-2/pS2 positive gastric carcinomas than Luqmani et al. (1989) but as both studies included only relatively small numbers of gastric carcinomas the proportions may be considered broadly comparable. The proportion of ovarian and endometrial carcinomas in which pNR-2/pS2 was detectable immunohistochemically is however considerably higher than that reported by Wysocki et al. (1990) who studied expression of pNR-2/pS2 mRNA in Northern blots of total cellular RNA extracts: it is possible that the significant proportion of stromal cells in these tumours militates against detection of a mRNA expressed solely in a proportion of the malignant epithelial cells.

For the purposes of comparison, the results obtained with a large series (171) of primary breast tumours (Henry et al., 1991) are shown in Table I. While the numbers of tumours in each group in the present series are smaller, it is interesting that pNR-2/pS2 is expressed in a similar proportion of breast, pancreatic, large bowel and gastric tumours. In addition, the mean proportion of cells in which pNR-2/pS2 expression was detected in the breast tumours is lower than in some of the other tumour types. Thus pNR-2/pS2 expression is at least as prevalent in some other epithelial tumours as it is in breast cancer.

Malignant ovarian tumours of surface epithelium form a heterogeneous group and the current series was chosen to represent the more common tumours which show either serous or mucinous differentiation. Immunohistochemical staining for the pNR-2/pS2 protein was found to associate very significantly with mucinous differentiation (Table II): a similar association with mucinous differentiation has been observed by Wysocki et al. (1990) but they did not have sufficient numbers of pNR-2/pS2 positive tumours for statistical analysis. We have investigated this association further in a series of benign serous and mucinous cystadenomas of ovary and found a similar and significant association. Although the association of pNR-2/pS2 expression with mucinous differentiation is not absolute, antibodies to pNR-2/pS2 may be useful reagents for determining differentiation in diagnostically problematic ovarian tumours. The divergent differentiation found in ovarian epithelial tumours is thought to result from the capacity of ovarian surface epithelium to differentiate into each of the types of Mullerian epithelium, with mucinous tumours differentiating along the line of endocervical epithelium. A series of normal endocervices in ten hysterectomy specimens was also stained immunohistochemically with the pNR-2/pS2 antisera but there was no evidence of pNR-2/pS2 expression (data not shown). It is however interesting to note that mucinous tumours positively were the only adenocarcinoma of endocervix in a group otherwise composed to squamous carcinomas. Expression of pNR-2/pS2 in epithelial tumours of Mullerian origin may occur as part of neoplastic progression.

We have detected pNR-2/pS2 expression in normal breast, stomach, small intestine and prostate in a previous study (Piggott et al., 1991) and in apparently normal large bowel epithelium adjacent to a colonic tumour in the present study. Wright et al. (1991) have described pNR-2/pS2 expression in intestinal mucosa adjacent to areas of mucosal damage and it is possible that a similar phenomenon occurs in normal colorectal epithelium adjacent to some tumours. Clearly pNR-2/pS2 expression by human malignant epithelial tumours is not restricted to those tissues in which the protein...
is normally expressed. The factors that control the ectopic expression of this protein are currently unknown.

In human breast cancer at least, expression of the pNR-2/pS2 mRNA or protein is almost entirely confined to a proportion of breast cancers that express oestrogen receptor (Río et al., 1987; Skilton et al., 1989; Henry et al., 1990; Foekens et al., 1990). This implies that oestrogens regulate the expression of the pNR-2/pS2 gene in breast cancer cells. The question then arises, is this gene regulated by oestrogens acting through the oestrogen receptor in the other malignant tumours in which it is expressed? Oestrogen receptor has been detected in a proportion of ovarian epithelial tumours (Schwartz et al., 1982; Bizi et al., 1988) and endometrial carcinomas (Palmer et al., 1988). More surprisingly, oestrogen receptor has also been detected in gastric carcinomas (Sica et al., 1984; Tokunaga et al., 1986), colorectal carcinomas (McClenndon et al., 1977; Sica et al., 1984) and pancreatic carcinomas (Greenway et al., 1981). Furthermore, a favourable response to antioestrogen therapy and other endocrine therapies has been recorded in pancreatic carcinoma (reviewed by Greenway, 1987). Expression of pNR-2/pS2 in normal gastric mucosa and gastric carcinomas has however been reported to be independent of oestrogen receptor expression (Río et al., 1988a; Luqmani et al., 1989) and Wysocki et al. (1990) only observed a weak correlation between oestrogen receptor mRNA and pNR-2/pS2 mRNA expression in ovarian epithelial tumours. Unfortunately, as the tumours in this study are an archival series from which only formalin fixed, paraffin embedded material is available and as receptor assays were not performed at the time of resection we are unable to correlate oestrogen receptor and pNR-2/pS2 expression. However in the current series, gender (and consequent gender dependent differences in sex steroid levels) did not show any relationship to the proportion of tumours expressing pNR-2/pS2 (Table I). As the majority of tumours from women were from elderly, postmenopausal women analysis of the effect of menopausal status was not possible.

Other factors may regulate pNR-2/pS2 gene expression in these tumours. The pNR-2/pS2 gene has a complex upstream promoter region that contains enhancer elements responsive to epidermal growth factor, the c-Ha-ras oncoprotein, the c-jun protein and a tumour promoter (Nunez et al., 1989). Co-expression of epidermal growth factor and pNR-2/pS2 has been described in intact mucosa at the edge of intestinal ulcers and it has been suggested that in this instance pNR-2/pS2 expression is controlled by epidermal growth factor (Wright et al., 1991). It is also possible that pNR-2/pS2 is expressed constitutively in a proportion of tumours.

The function of the pNR-2/pS2 protein however remains enigmatic. If the pNR-2/pS2 protein is indeed a growth factor it is possible that it may stimulate tumour growth by autocrine means. The biological and prognostic significance of pNR-2/pS2 expression, particularly in non-mammary tumours, remains to be determined. The frequent expression of this protein in diverse human malignancies suggests that pNR-2/pS2 could have a more general role in human neoplasia than hitherto realised.

This work was supported by the Gunnar Nilsson Cancer Research Trust, the North of England Cancer Research Campaign, the Breast Cancer Research Trust, and the Medical Research Council. We thank Mrs R. Brown for technical assistance, Mrs E. Tweedy for secretarial assistance and Mr S. Brabazon for photography. We are indebted to Ms S. Cousen for peptide synthesis. F.E.B. May thanks the Royal Society for a University Research Fellowship.

### Table 11 pNR-2 immunohistochemical staining in benign and malignant primary ovarian tumours

| Tumour type                  | Total no. cases | pNR-2/pS2 positive cases | Positive tumours, mean proportion positively-stained cells (%) | Range |
|------------------------------|-----------------|--------------------------|----------------------------------------------------------------|-------|
| Serous cystadenocarcinoma     | 14              | 2 (14%)                  | 3%                                                              | 1–5%  |
| Mucinous cystadenocarcinoma   | 8               | 6 (75%)                  | 20%                                                             | 14–29%|
| Serous cystadenoma            | 14              | 5 (36%)                  | 18%                                                             | 15–24%|
| Mucinous cystadenoma          | 11              | 11 (100%)                | 37%                                                             | 10–71%|

**References**

BERRY, M., NUNEZ, A.-M. & CHAMON, P. (1989). Estrogen-responsive element of the human pS2 gene is an imperfectly palindromic sequence. *Proc. Natl Acad. Sci. USA*, 86, 1218.

BIZZI, A., CODEGONI, A.M., LANDONI, F. & 5 others (1988). Steroid receptors in epithelial ovarian carcinoma: relation to clinical parameters and survival. *Cancer Res.*, 48, 6222.

FOEKENS, J.A., RIO, M.-C., SEGUN, P. & 5 others (1990). Prediction of relapse and survival in breast cancer patients by pS2 protein status. *Cancer Res.*, 50, 3832.

GREENWAY, B., IQBAL, M.J., JOHNSON, P.J. & WILLIAMS, R. (1981). Oestrogen receptor proteins in malignant and fetal pancreas. *Br. Med. J.*, 283, 751.

GREENWAY, B.A. (1987). Carcinoma of the exocrine pancreas: a sex hormone responsive tumour? *Br. J. Surg.*, 74, 441.

HENRY, J.A., NICHOLSON, S., HENNESSY, C., LENNARD, T.W.J., MAY, F.E.B. & WESTLEY, B.R. (1990). Expression of the oestrogen regulated pNR-2 mRNA in human breast cancer: relation to oestrogen receptor mRNA levels and response to tamoxifen therapy. *Br. J. Cancer*, 61, 32.

HENRY, J.A., PIGGOTT, N.H., MALLICK, U.K. & 4 others (1991). pNR-2/pS2 immunohistochemical staining in breast cancer: correlation with prognostic factors and endocrine response. *Br. J. Cancer*, 63, 615.

JAKOWLEW, S.B., BREATHNACH, R., JELTSCH, J.-M., MASIAKOWSKI, P. & CHAMON, P. (1984). Sequence of the pS2 mRNA induced by estrogen in the human breast cancer cell line MCF-7. *Nucleic Acids Res.*, 12, 2861.

LUQMANI, Y., BENNETT, C., PATERSON, I. & 4 others (1989). Expression of the pS2 gene in normal, benign and neoplastic human stomach. *Int. J. Cancer*, 44, 806.

MCCLENNDON, J.E.,APPLEBY, D., CLAUDON, D.B., DONEGAN, W.L. & DECOSSE, J.J. (1977). Colonic neoplasms: tissue estrogen receptor and carcinoembryonic antigen. *Arch. Surg.*, 112, 240.

MASIAKOWSKI, P., BREATHNACH, R., BLOCH, J., GANNON, F., KRUST, A. & CHAMON, P. (1982). Cloning of cDNA sequences of hormone regulated genes from the MCF-7 human breast cancer cell line. *Nucleic Acids Res.*, 10, 7895.

MAY, F.E.B. & WESTLEY, B.R. (1986). Cloning of estrogen regulated messenger RNA sequences from human breast cancer cells. *Cancer Res.*, 46, 6034.

NUNEZ, A.-M., JAKOWLEW, S., BRIAND, J.-P., GAIRE, M., KRUST, A., RIO, M.-C. & CHAMON, P. (1987). Characterization of the estrogen-induced pS2 protein secreted by the human breast cancer cell line MCF-7. *Endocrinology*, 121, 1759.
The 5' flanking region of the pS2 gene contains a complex enhancer region responsive to oestrogens, epidermal growth factor, a tumour promoter (TPA), the c-Ha-ras oncoprotein and the c-jun protein. *EMBO J.*, 8, 823.

PALMER, D.C., MUIR, I.M., ALEXANDER, A.I., CAUCHI, M., BENNETT, R.S. & QUINN, M.A. (1988). The prognostic importance of steroid receptors in endometrial carcinoma. *Obstet. Gynecol.*, 72, 388.

PIGGOTT, N.H., HENRY, J.A., MAY, F.E.B. & WESTLEY, B.R. (1991). Antipeptide antibodies against the pNR-2 oestrogen-regulated protein of human breast cancer cells and detection of pNR-2 expression in normal tissues by immunohistochemistry. *J. Pathol.*, 163, 95.

PRUD'HOMME, J.-F., FRIDLANSKY, F., LE CUNFF, M. & 4 others (1985). Cloning of a gene expressed in human breast cancer and regulated by estrogen in MCF-7 cells. *DNA*, 4, 11.

RIO, M.C., BELLOCQ, J.P., GAIRARD, B. & 7 others (1987). Specific expression of the pS2 gene in subclasses of breast cancers in comparison with expression of the estrogen and progesterone receptors and the oncogene ERBB2. *Proc. Natl Acad. Sci. USA*, 84, 9243.

RIO, M.-C., BELLOCQ, J.-P., GAIRARD, B., KOEHL, C., RENAUD, R. & CHAMBERON P. (1988a). Expression spécifique de gene humain pS2 dans les cancers du sein. *Biochimie*, 70, 961.

RIO, M.C., BELLOCQ, J.P., DANIEL, J.Y. & 5 others (1988b). Breast cancer-associated pS2 protein: synthesis and secretion by normal stomach mucosa. *Science*, 241, 705.

SCHWARTZ, P.E., LIVOLSI, V.A., HILDRETH, N., MACLUSKY, N.J., NAFTOLIN, F.N. & EISENFIELD, A.J. (1982). Estrogen receptors in ovarian epithelial carcinoma. *Obstet. Gynecol.*, 59, 229.

SICA, V., NOLA, E., CONTIERI, E. & 7 others (1984). Estradiol and progesterone receptors in malignant gastrointestinal tumours. *Cancer Res.*, 44, 4670.

SKILTON, R.A., LUQMANI, Y.A., MCCLELLAND, R.A. & COOMBES, R.C. (1989). Characterisation of a messenger RNA selectively expressed in human breast cancer. *Br. J. Cancer*, 60, 168.

STERNBERGER, L.A., HARDY, P.H., CUCULIS, J.J. & MEYER, H.G. (1970). The unlabelled antibody method of immunohistochemistry: preparation and properties of soluble antigen-antibody complex (horseradish peroxidase: antiperoxidase) and its use in the identification of spirochaetes. *J. Histochem. Cytochem.*, 18, 315.

TOKUNGA, A., NISHI, K., MATSUKURA, N. & 5 others (1986). Estrogen and progesterone receptors in gastric cancer. *Cancer*, 57, 1376.

WRIGHT, N.A., POULSON, R., STAMP, G.W.H. & 6 others (1991). Epidermal growth factor (EGF/URO) induces expression of genes encoding regulatory peptides in damaged human gastrointestinal tissues. *J. Pathol.*, 162, 279.

WOOD, G.S. & WARNKE, R. (1981). Suppression of endogenous avidin-binding in tissues and its relevance to biotin-avidin detection systems. *J. Histochem. Cytochem.*, 29, 1196.

WYSOCKI, S.J., HAHNEL, E., MASTERS, A., SMITH, V., MCCARTNEY, A.J. & HAHNEL, R. (1990). Detection of pS2 messenger RNA in gynaecological cancers. *Cancer Res.*, 50, 1800.