Virchows Archiv A Pathol Anat (1990) 416:491–496

Virchows Archiv A
Pathological Anatomy and Histopathology

Transferrin receptor expression in tumours of the human nervous system: relation to tumour type, grading and tumour growth fraction

Reinhard Prior, Guido Reifenberger, and Wolfgang Wechsler

Department of Neuropathology, Heinrich-Heine-University, Moorenstrasse 5, D-4000 Düsseldorf 1, Federal Republic of Germany

Received August 16, 1989 / Received after revision December 11, 1989 / Accepted December 12, 1989

Summary. The expression of transferrin receptor (Tr) was investigated by means of immunohistochemistry in 101 tumours of the human central and peripheral nervous system. The results were compared with the proliferative activity of the tumours, determined by immunostaining for the proliferation-associated antigen Ki-67. In addition to immunostaining of normal and proliferated blood vessel endothelium and of a fraction of tumour infiltrating lymphocytes, we observed staining for Tr in a variable fraction of neoplastic cells of all histological tumour types. Immunoreactivity in the majority of tumour cells was found only in anaplastic tumours such as glioblastomas. Furthermore, a positive correlation between Tr expression and the Ki-67 growth fraction was established for gliomas. Non-glial tumours strongly expressing Tr included one metastatic rhabdomyosarcoma, one intracerebral malignant lymphoma, two of four plasmocytomas and seven of nine metastatic carcinomas. Our results indicate that immunohistochemistry for Tr and Ki-67 can provide additional information about the biological behaviour of nervous system tumours, thus complementing conventional histopathological criteria for anaplasia.

Key words: Brain neoplasm – Transferrin receptor – Tumour growth fraction – Ki-67

Introduction

The transferrin receptor (Tr) has been characterized as a membrane-spanning glycoprotein composed of two identical disulphide-bonded 95000 kDa subunits containing 760 amino acids (Trowbridge et al. 1984; Schneider et al. 1982, 1984). Cellular iron uptake, mediated by the binding of transferrin to its receptor and subsequent endocytosis of the receptor-ligand complex (Dautry-Varsat et al. 1983; Klausner et al. 1983a, b) is an essential prerequisite for DNA synthesis and cell replication (Hamilton 1982; Larrick and Cresswell 1979; Neckers et al. 1984).

Tr has been demonstrated immunohistochemically in epithelial cells of normal human epidermis, gastrointestinal tract, liver, kidney, testis, anterior pituitary, endocrine pancreas (Gatter et al. 1983) and in the endothelial cells of the brain capillaries (Jefferies et al. 1984). Tr is further expressed in various malignant human tumours including carcinoma of breast (Wrba et al. 1986), lung (Doria et al. 1988), gastrointestinal tract (Niitsu et al. 1987), hepatocellular (Sciot et al. 1988) and bladder transitional cell carcinoma (Seymour et al. 1987), malignant melanoma (Soyer et al. 1987), non-Hodgkin's lymphoma (Medeiros et al. 1988) and leukaemia (Barnett et al. 1987). For some of these tumour types a correlation has been established between the amount of Tr expression and certain tumour growth variables including rate of [^{3}H]thymidine incorporation (Kvaloy et al. 1984), DNA flow cytometry (Walker and Camplejohn 1986; Wain et al. 1987) and immunostaining for the Ki-67 antigen (Wrba et al. 1988) or for incorporated bromodeoxyuridine (Schrape et al. 1987). Knowledge of the in situ expression of Tr in tumours of the human nervous system, is still lacking however.

We investigated the expression of Tr in a large series of benign and malignant nervous system tumours by means of immunohistochemistry, using a specific monoclonal antibody against Tr. We further determined the tumour growth fractions by using the monoclonal antibody Ki-67. The purpose of the present study was to find out whether Tr expression in nervous system tumours is correlated with specific tumour types or with the degree of malignancy assessed by conventional histopathological criteria for anaplasia as well as by Ki-67 immunostaining.

Materials and methods

A total of 101 tumours of the human nervous system were examined on acetone-fixed cryostat sections (Table 1). All tumours were

Offprint requests to: W. Wechsler

Table 1. Transferrin receptor (Tr) expression and Ki-67 labelling index (LI) in tumours of the human nervous system

Diagnosis and grading	No.	Tr expression				Ki-67-LI (%)		
		0	1	2	3	$\bar{x}$	SD	range
Astrocytoma, pilocytic (I)	2	2	0	0	0	<1	<1	–
Astrocytoma (II)	12	4	7	1	0	1.8	2.1	<1–7.0
Astrocytoma, anaplastic (III)	5	0	2	3	0	8.4	7.8	1.3–21.2
Astrocytoma, anaplastic (IV)[a]	2	0	0	0	2	48.6	6.2	44.2–52.9
Pleomorphic Xanthoastrocytoma	1	0	1	0	0	<1	–	–
Oligodendroglioma (II)	3	1	1	1	0	3.3	4.5	<1–8.5
Oligodendroglioma, anaplastic (III)	4	0	4	0	0	12.8	3.5	10.4–18.0
Mixed glioma (II)	2	0	2	0	0	4.3	5.3	<1–8.0
Mixed glioma, anaplastic (III)	6	0	3	2	1	14.1	10.4	3.0–29.0
Ependymoma (II)	3	2	1	0	0	1.5	1.5	<1–3.2
Ependymoma, anaplastic (III)	1	0	0	0	1	21.0	–	–
Subependymoma (I)	2	0	2	0	0	<1	<1	–
Glioblastoma (IV)	16	0	1	5	10	12.3	7.3	3.7–28.0
Medulloblastoma (IV)	5	1	1	2	1	24.7	14.5	6.0–42.0
Primitive neuroectodermal tumour (IV)	1	0	0	1	0	13.4	–	–
Meningioma (I)	12	2	2	7	1	1.2	1.0	<1–3.8
Meningioma, anaplastic (III)	1	0	0	0	1	10.0	–	–
Neurinoma (I)	5	5	0	0	0	<1	–	–
Neurinoma, anaplastic (III)	1	0	0	1	0	22.0	–	–
Ganglioneuroma (I)	1	1	0	0	0	<1	–	–
Ganglioneuroblastoma (III)	1	0	1	0	0	7.7	–	–
Metastatic carcinoma	9	2	0	1	6	10.8	12.4	<1–34.2
Metastatic intracerebral rhabdomyosarcoma	1	0	0	0	1	8.0	–	–
Malignant lymphoma	1	0	0	0	1	27.0	–	–
Plasmocytoma[b]	4	2	0	0	2	10.7	9.6	<1–20.0

[a] Highly anaplastic recurrencies of primary WHO grade II astrocytomas (see Winkler et al. 1988)
[b] Vertebral

The fraction of Tr-positive tumour cells in each tumour was estimated on a rating scale where 0=no positive cell, 1=single positive cells (<10%), 2=moderate fraction of positive cells (10–50%), 3=large fraction of positive cells (>50%). Ki-67 staining is expressed as the percentage of positive cell nuclei given as mean value ($\bar{x}$) with standard deviation (SD) and range (minimal – maximal value). Grade I–IV refers to WHO grade of malignancy

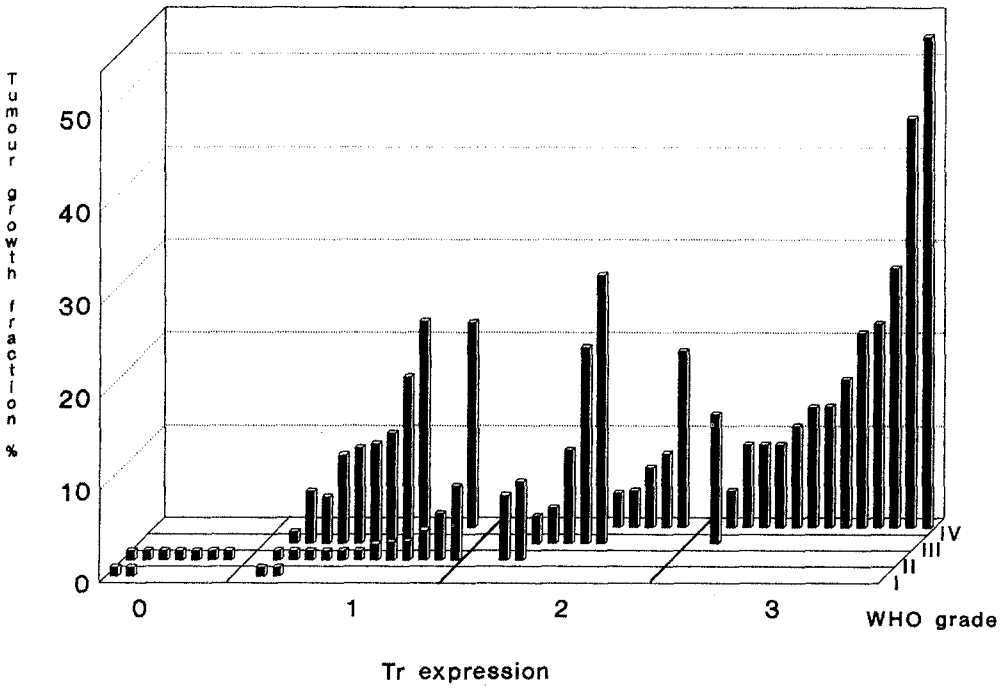

Fig. 1. Schematic representation of transferrin receptor (*Tr*) expression, tumour growth fraction and WHO grade in human gliomas. The fraction of Tr-positive tumour cells in each tumour was estimated on a rating scale where 0=no positive cell, 1=single positive cells (<10%), 2=moderate fraction of positive cells (10–50%), 3=large fraction of positive cells (>50%). The tumour growth fraction is given as a percentage of Ki-67 positive nuclei. Each *bar* represents one tumour. The mean Ki-67 indices [mean ($\bar{x}$), standard deviation (SD)] increased with the degree of Tr immunoreactivity: Tr=0, $\bar{x}$<1%; Tr=1: $\bar{x}$=6.1%, SD=7.0; Tr=2: $\bar{x}$=10.3%, SD=8.3; Tr=3: $\bar{x}$=19.0%, SD=14.1). Statistical significances according to the Mann-Whitney U test were Tr=0 vs Tr=1: P=0.0006; Tr=0 vs Tr=2: P=0.00004; Tr=0 vs Tr=3: P=0.00002; Tr=1 vs Tr=2: P=0.025; Tr=1 vs Tr=3: P=0.0002; Tr=2 vs Tr=3: P=0.011

classified according to the WHO classification (Zülch 1979). Tissue samples from surgically removed tumours were divided, one part being fixed in 4% buffered formalin for routine tumour diagnosis, the other being shock-frozen in isopentane at −150° C for immunohistochemistry, which was performed using the avidin-biotin-peroxidase method (Hsu et al. 1981; for details of the protocol, see Reifenberger et al. 1989). As primary antibody we used a commercially available mouse monoclonal anti-Tr antibody (clone 2EB, Amersham-Buchler, Braunschweig, FRG) which was applied to the sections at a concentration of 2 µg/ml for 12 h at room temperature. The mouse monoclonal IgG1 antibody Ki-67 was obtained by immunization against a crude nuclear fraction of L428 cells (Gerdes et al. 1983) and recognizes a nuclear antigen present in proliferating but absent in resting (Go) cells (Gerdes et al. 1984). We used the Ki-67 antibody (obtained from Dakopatts, Hamburg, FRG) diluted 1:20 for 12 h at room temperature. Negative controls were performed by omitting the primary antibody and applying non-specific mouse immunoglobulins instead. Tr expression was evaluated as indicated in Table 1 and Fig. 1 by using a modified version of the semi-quantitative rating scale already employed in previous publications (Reifenberger et al. 1989; Prior et al. 1989). This semiquantitative evaluation was choosen because accurate counting of Tr-positive cells was impossible in many cases due to the fact that immunoreactivity was not nuclear (as in Ki-67 staining) but rather cytoplasmic and cell membrane associated. This staining pattern makes a clearcut distinction of positive and negative tumour cells very difficult on cryostat sections. In contrast, Ki-67 positive or negative cell nuclei are easily recognizable, thus allowing a quantitative assessment of proliferative activity by counting (Deckert et al. 1989). For statistical analysis we used the Mann-Whitney U test (see Fig. 1).

Results

Immunostaining for Tr was observed as a fine granular reaction product located both on the cell membrane and in the cytoplasm of tumour cells. In addition, we found normal and proliferated intra- and extratumoral blood

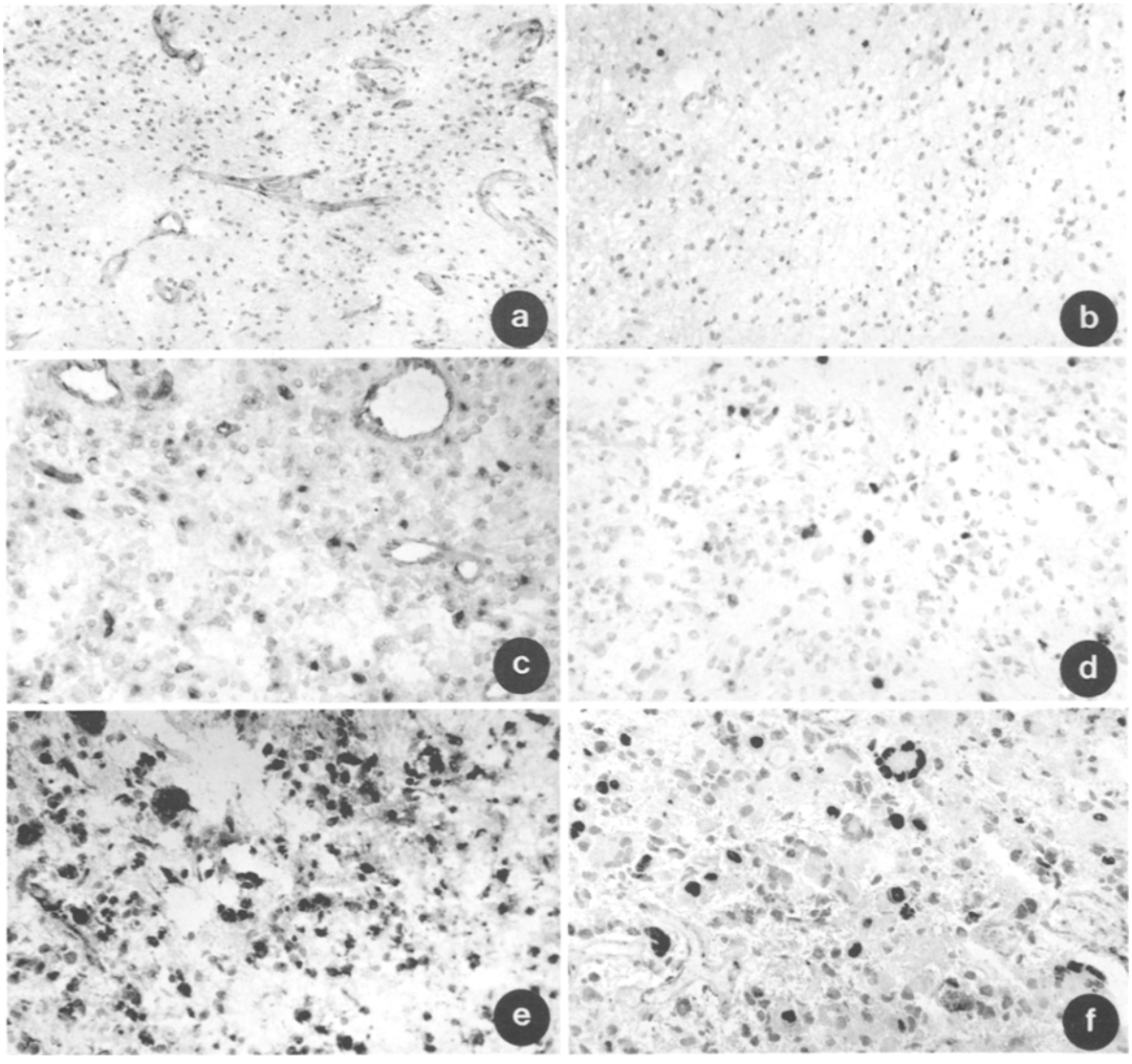

Fig. 2. a, b Astrocytoma (WHO grade II). **a** Tr expression is mainly restricted to intratumour blood vessels. **b** The Ki-67 index is below 1%. NP 678/88, ×120. **c, d** Oligodendroglioma (WHO grade II). **c** Tr expression is present on blood vessels and a minor fraction of tumour cells. **d** The Ki-67 index is elevated to 8.5%. NP 699/88, ×150. **e, f** Giant cell glioblastoma (WHO grade IV). **e** Tr is strongly expressed on the majority of tumour cells. **f** The Ki-67 index is 13%. NP 602/88, ×150

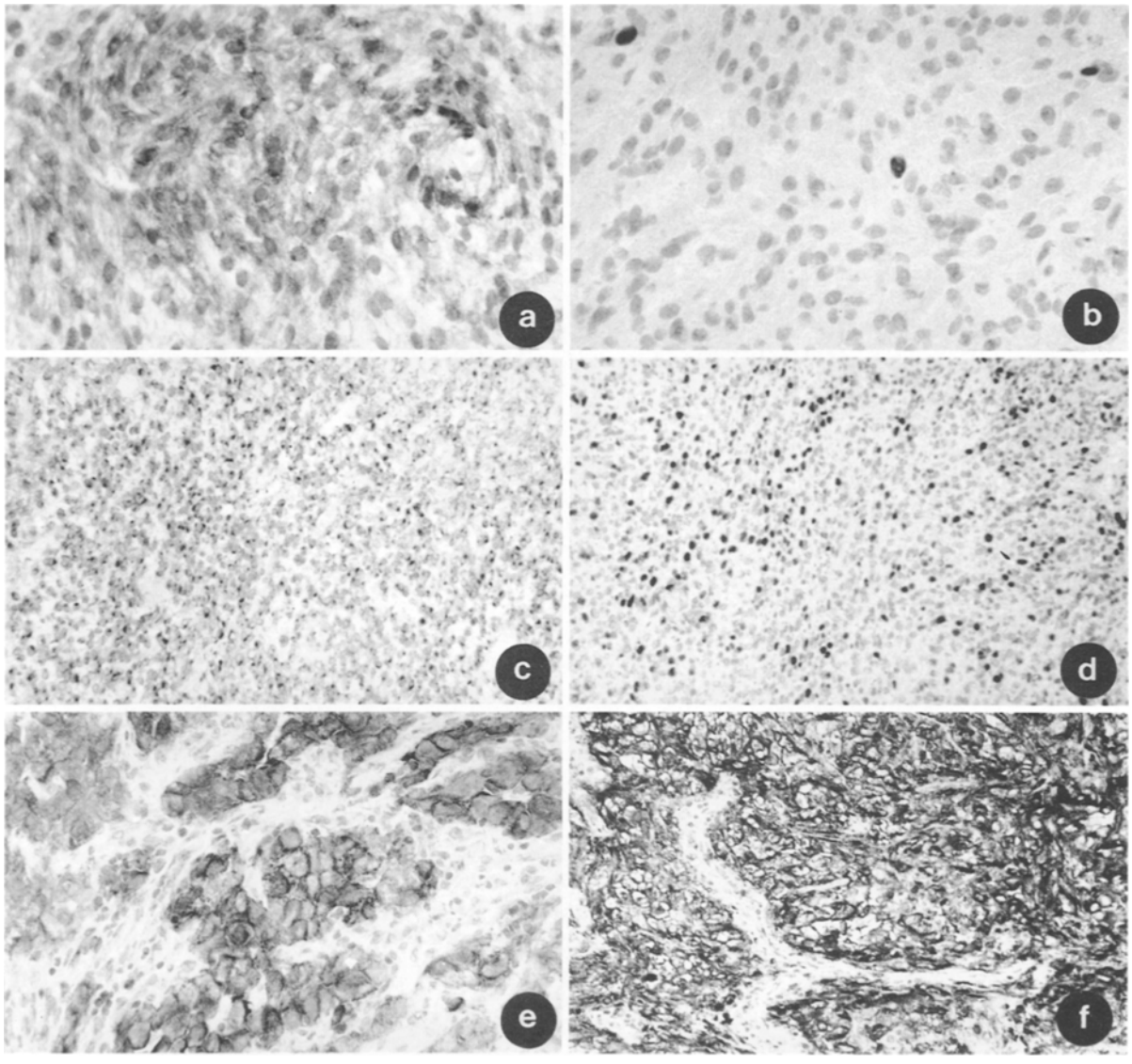

Fig. 3. a, b Endotheliomatous meningioma (WHO grade I). **a** A cluster of Tr-positive meningioma cells. **b** The Ki-67 index is less than 1%. NP 1103/88, ×400. **c, d** Plasmocytoma. **c** Many tumour cells demonstrate Tr immunoreactivity. **d** The Ki-67 index is 18%. NP 493/88, ×120 **e** Metastatic carcinoma. Note strong immunoreactivity for Tr in the carcinoma cells, while the mesenchymal stroma is unstained. The Ki-67 index of this tumour was 17%. NP 313/88, ×240. **f** Metastatic hypernephroma. Note generalized strong Tr expression. The Ki-67 index of this tumour was 9%. NP 675/88, ×150

vessel endothelium positivity and in some cases a fraction of the infiltrating lymphocytes were also Tr positive. With the exception of this blood vessel and lymphocyte associated staining, no immunoreactivity was seen in non-neoplastic nervous tissue adjacent to the tumours.

Immunoreactivity for Tr and Ki-67 growth fractions in the different tumour types is summarized in Table 1. Fig. 1 represents the relationship between Tr expression, tumour growth fraction and WHO grade in gliomas graphically. Among the gliomas, 25 low-grade (WHO grade I and II) tumours comprising 2 pilocytic astrocytomas (grade I), 12 astrocytomas (grade II), 1 pleomorphic xanthoastrocytoma, 3 oligodendrogliomas (grade II), 2 mixed gliomas (grade II), 2 ependymomas (grade II), and 2 subependymomas (grade I) were either completely negative or showed Tr expression restricted to a minor fraction of neoplastic cells (Fig. 2a–d). However in all of the 34 anaplastic (WHO grade III and IV) gliomas investigated, including 7 astrocytomas (grade III and IV), 4 oligodendrogliomas (grade III), 6 mixed gliomas (grade III) and 16 glioblastomas (grade IV), Tr immunoreactive tumour cells were detected. In many grade III gliomas, particularly in anaplastic oligodendrogliomas and mixed gliomas, however, the percentage of Tr positive cells was low or moderate, whereas in most glioblastomas (10/16) the majority of tumour cells were strongly labelled (Fig. 2e, f). All Tr negative gliomas had proliferation indices below 1% and showed no histological signs of anaplasia. In contrast, gliomas with Tr labelling in a large fraction of tumour cells were almost exclusively grade IV anaplastic gliomas with elevated growth fractions. In spite of the wide ranges of

Ki-67 proliferation indices associated with each degree of Tr expression, statistical evaluation demonstrated an overall positive correlation between Tr expression and growth fractions in gliomas (see legend to Fig. 1).

Other tumours of the central nervous system, including 4 of 5 medulloblastomas and 1 primitive neuroectodermal tumour showed variable, but mostly moderate degrees of Tr immunoreactivity. In tumours of the peripheral nervous system a moderate degree of Tr expression was observed in 1 anaplastic neurinoma (grade III) and 1 anaplastic ganglioneuroblastoma (grade III), while all benign neurinomas and 1 ganglioneuroma were Tr negative. Ten of 12 benign (grade I) meningiomas showed weak immunostaining for Tr in a moderate fraction of tumour cells (Fig. 3a, b). There was no relation to particular histological subtypes. In many cases, however, the Tr-positive meningioma cells were arranged in small islands. A more widespread staining was observed in one case of recurrent anaplastic (grade III) meningioma. The majority (6/9) of intracerebral and spinal metastatic carcinomas expressed high amounts of Tr (Fig. 3e, f). This group was composed of 2 metastases derived from bronchial adenocarcinomas, 1 metastatic bronchial squamous epithelial carcinoma, 2 metastases from renal cellcarcinoma, 1 metastatic follicular thyroid carcinoma, 2 adenocarcinomas of unknown origin and 1 paravertebral undifferentiated metastatic carcinoma of unknown origin. The metastasis of the thyroid carcinoma and 1 of the metastatic adenocarcinomas of unknown origin were Tr negative. Strong immunoreactivity for Tr was present in 1 intracerebral malignant lymphoma (centrocytic-centroblastic subtype), in 1 intracerebral metastatic pleomorphic rhabdomyosarcoma and in 2 of 4 vertebral plasmocytomas. The Tr-negative plasmocytomas had low Ki-67 indices (0.9 and 4.2%), while the Tr-positive cases showed elevated values of 18% and 20% respectively, thus indicating a positive correlation between Tr immunoreactivity and tumour growth fraction (Fig. 3c, d).

Discussion

The present study describes the in situ distribution of Tr in tumours of the human nervous system for the first time. Using immunohistochemistry with a monoclonal antibody on cryostat sections, we detected Tr on the endothelium of intracerebral and intratumoral normal and proliferating blood vessels, corroborating previous data reported by Jefferies et al. (1984). Tr was further present in a fraction of tumour-infiltrating lymphocytes. In addition, Tr expression was observed in neoplastic cells in the majority of benign (WHO grade I and II) and anaplastic (WHO grade III and IV) glial tumours. Gliomas with more than 50% of Tr-positive tumour cells all belonged to the high grade group, particularly to the glioblastomas. This finding parallels the association of high Tr expression with anaplastic tumour growth described for various malignant tumour types of non-neuroectodermal origin (Barnett et al. 1987; Doria et al. 1988; Medeiros et al. 1988; Niitsu et al. 1987; Seymour et al 1987; Sciot et al. 1988; Wrba et al. 1986).

In comparison with the expression of epidermal growth factor receptor (Reifenberger et al. 1989), Tr expression demonstrated a much clearer correlation with the tumour growth fraction determined by Ki-67 in the glioma group. Tumour growth fractions were constantly below 1% for Tr-negative gliomas, whereas gliomas with moderate or high Tr expression showed increased proliferative activity. The Ki-67 indices, however, covered a relatively broad range of values for each degree of Tr expression. This may partly be a consequence of the relatively crude semi-quantitative score we applied to classify Tr immunoreactivity. It has, however, been demonstrated that the correlation of Ki-67 growth fractions with the conventional histopathological grading according to the WHO classification does result in considerable ranges for each grade of malignancy (Deckert et al. 1989; Kleihues et al. 1988).

Peripheral nervous system tumours showed a similar tendency to a more pronounced Tr expression in tumours with histological signs of anaplasia. The association between increased Tr expression and high grade of malignancy appears not to exist for medulloblastomas, since only one of five medulloblastomas showed Tr immunoreactivity in a majority of tumour cells, and one medulloblastoma was even negative for Tr. Meningiomas represented the only tumour type among the neoplasms investigated where Tr expression was constantly associated with a low grade of malignancy and a low proliferative potential. The intensity of Tr staining in meningiomas, however, was considerably lower than in malignant tumours of glial origin. The strong immunoreactivity we observed in most metastatic carcinomas is in keeping with previous results demonstrating Tr in malignant tumours of epithelial origin (Doria et al. 1988; Gatter et al. 1983; Wrba et al. 1986).

Tr-directed immunotoxins have been shown to be highly effective against cell lines derived from human malignant gliomas and medulloblastomas (Colombatti et al. 1988; Zovickian et al. 1987). Our results on the expression of large amounts of Tr in many malignant brain tumours in situ indicate that Tr-directed immunotoxins may be effective additional therapeutics in vivo (for example, in cases of leptomeningeal blastomatosis). Johnson et al. (1989) have recently proposed the first clinical trials using such an approach.

In conclusion, Tr is expressed in various tumours of the human nervous system. In gliomas a close relationship between Tr expression, WHO grade of malignancy and tumour growth fraction has been established. Immunohistochemistry for Tr can therefore reveal additional information on the biological behaviour of gliomas and may thus supplement conventional criteria for anaplasia in the histopathological grading of these neoplasms.

Acknowledgements. The authors would like to thank E. Gross, C. Mähler, U. Tuma and L. Willer for excellent technical assistance and S. Wist for stylistic revision of the manuscript. The work was supported by the Deutsche Forschungsgemeinschaft, SFB 200.

References

Barnett D, Wilson GA, Lawrence AC, Buckley GA (1987) Transferrin receptor expression in the leukaemias and lymphoproliferative disorders. Clin Lab Haematol 9:361–370

Colombatti M, Bisconti M, Dell'Arciprete L, Gerosa MA, Tridente G (1988) Sensitivity of human glioma cells to cytotoxic heteroconjugates. Int J Cancer 42:441–448

Dautry-Varsat A, Ciechanover A, Lodish HG (1983) pH and the recycling of transferrin during receptor mediated endocytosis. Proc Natl Acad Sci USA 80:2258–2262

Deckert M, Reifenberger G, Wechsler W (1989) Determination of the proliferative potential of human brain tumors using the monoclonal antibody Ki-67. J Cancer Res Clin Oncol 115:179–188

Doria MI Jr, Montag AG, Franklin WA (1988) Immunophenotype of small cell lung carcinoma. Expression of NKH-1 and transferrin receptor and absence of most myeloid antigens. Cancer 62:1939–1945

Gatter KC, Brown G, Trowbridge IS, Woolston R-E, Mason DY (1983) Transferrin receptors in human tissues: their distribution and possible clinical relevance. J Clin Pathol 36:539–535

Gerdes J, Schwab U, Lemke H, Stein H (1983) Production of a mouse monoclonal antibody reactive with a human nuclear antigen associated with cell proliferation. Int J Cancer 31:13–20

Gerdes J, Lemke H, Baisch H, Wacker HH, Schwab U, Stein H (1984) Cell cycle analysis of a cell proliferation-associated human nuclear antigen defined by the monoclonal antibody Ki-67. J Immunol 133:1710–1715

Hamilton TA (1982) Regulation of transferrin receptor expression in concanavalian A stimulated and Gross virus transformed rat lymphoblasts. J Cell Physiol 113:40–46

Hsu SM, Raine L, Fanger H (1981) The use of anti-avidin antibody and avidin-biotin-peroxidase-complex in immunoperoxidase techniques. Am J Clin Pathol 75:816–821

Jefferies WA, Brandon MR, Hunt SV, Williams AF, Gatter KC, Mason DY (1984) Transferrin receptor on endothelial cells of brain capillaries. Nature 312:162–163

Johnson VG, Wrobel C, Wilson D, Zovickian J, Greenfield L, Oldfield EH, Youle R (1989) Improved tumor-specific immunotoxins in the treatment of CNS and leptomeningeal neoplasia. J Neurosurg 70:240–248

Klausner RD, Ashwell G, Renswoude J van, Harford JB, Bridges KR (1983a) Binding of apotransferrin to K562 cells: explanation of the transferrin cycle. Proc Natl Acad Sci USA 80:2263–2266

Klausner RD, Renswoude J van, Ashwell G, Kempf C, Schechter AN, Dean A, Bridges KR (1983b) Receptor mediated endocytosis of transferrin in K562 cells. J Biol Chem 258:4715–4724

Kleihues P, Meer L, Shibata T, Bamberg M (1988) DNA-Reparatur und DNA-Replikation als Kriterien für therapeutische Effizienz. In: Bamberg S, Sack H (eds) Therapie primärer Hirntumoren, Zuckschwerdt, Munich, pp 1–8

Kvaloy S, Langholm R, Kaalhus O, Michaelsen T, Funderud S, Foss Abrahamsen A, Godal T (1984) Transferrin receptor and B-lymphoblast antigen: their relationship to DNA synthesis, histology and survival in B-cell lymphomas. Int J Cancer 33:173–177

Larrick JW, Creswell P (1979) Modulation of cell surface iron transferrin receptors by cellular density and state of activation. J Supramol Struct 11:579–586

Neckers LM, Yenokida G, James SP (1984) The role of transferrin receptor in human B lymphocyte activation. J Immunol 133:2437–2441

Niitsu Y, Kohgo Y, Nishisato T, Kondo H, Kato J, Urushizaki Y, Urushizaki I (1987) Transferrin receptors in human cancerous tissues. Tohoku J Exp Med 153:239–243

Medeiros LJ, Picker LJ, Horning SJ, Warnke RA (1988) Transferrin receptor expression by non-Hodgkin's lymphomas. Correlation with morphologic grade and survival. Cancer 61:1844–1851

Prior R, Reifenberger G, Wechsler W (1989) Nerve growth factor receptor in tumours of the human nervous system. Immunohistochemical analysis of receptor expression and tumour growth fraction. Pathol Res Pract 185:332–338

Reifenberger G, Prior R, Deckert M, Wechsler W (1989) Epidermal growth factor receptor expression and growth fraction in human tumours of the nervous system. Virchows Arch [A] 414:147–155

Schneider C, Sutherland R, Newman R, Greaves M (1982) Structural features of the cell surface receptor for transferrin that is recognised by the monoclonal antibody OKT9. J Biol Chem 257:8516–8522

Schneider C, Owen M, Banville D, Williams JG (1984) Primary structure of human transferrin receptor deduced from the mRNA sequence Nature 311:675–678

Schrape S, Jones DB, Wright DH (1987) A comparison of three methods for the determination of the growth fraction in non-Hodgkin's lymphoma. Br J Cancer 55:283–286

Sciot R, Paterson AC, Eyken P vyn, Callea F, Kew MC, Desmet VJ (1988) Transferrin receptor expression in human hepatocellular carcinoma: an immunohistochemical study of 34 cases. Histopathology 12:53–63

Seymour GJ, Walsh MD, Lavin MF, Strutton G, Gardiner RA (1987) Transferrin receptor expression by human bladder transitional cell carcinomas. Urol Res 15:341–344

Soyer HP, Smolle J, Torne R, Kerl H (1987) Transferrin receptor expression in normal skin and in various cutaneous tumors. J Cutan Pathol 14:1–5

Trowbridge IS, Newman RA, Domingo DL, Sauvage C (1984) Transferrin receptors: structure and function. Biochem Pharmacol 33:925–932

Wain SL, Braylan RC, Borowitz MJ (1987) Correlation of monoclonal antibody phenotyping and cellular DNA content in non-Hodgkin's lymphoma. The Southeastern Cancer Study Group experience. Cancer 60:2403–24

Walker RA, Camplejohn RS (1986) DNA flow cytometry of human breast carcinomas and its relationship to transferrin and epidermal growth factor receptors. J Pathol 150:37–42

Winkler J, Roosen N, Reifenberger G, Wechsler W (1988) Dedifferentiation in recurrent astrocytoma: clinicopathological study of two cases (abstract). Clin Neuropathol 7:222

Wrba F, Ritzinger E, Reiner A, Holzner JH (1986) Transferrin receptor (TrfR) expression in breast carcinoma and its possible relationship to prognosis. An immunohistochemical study. Virchows Arch [A] 410:69–73

Wrba F, Reiner A, Markis-Ritzinger E, Holzner JH, Reiner G, Spona J (1988) Prognostic significance of immunohistochemical parameters in breast carcinomas. Pathol Res Pract 183:277–283

Zovickian J, Johnson VG, Youle RJ (1987) Potent and specific killing of human malignant brain tumor cells by an antitransferrin receptor antibody-ricin immunotoxin. J Neurosurg 66:850–861

Zülch KJ (1979) Histological typing of tumours of the central nervous system. International histological classification of tumours. WHO, Geneva