Characterisation of a new brain metastases model in the nude rat

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Introduction

Animal models are of vital importance in the study of cancer development and progression. The relevance of each model depends on how closely it mimics the histology, physiological parameters, biochemical pathways and metastatic spread. Orthotopic xenografts of human tumours into immunodeficient animals have previously been shown to reproduce the histology and metastatic patterns of such tumours at advanced stages in mice (1). Here we present for the first time a new rat/tumour model, where brain metastases directly harvested from cancer patients are xenografted into the brains of immunocompromised rats.

Materials and Methods

Brain metastases were collected during operation, and put in culture (2). Then the biopsy materials were minced into small fragments, and placed in nutrient medium in culture flasks coated agar. After 10-20 days, they rounded up into spheroids. 10 such spheroids were stereotactically implanted into each brain of immunodeficient (Rowett rnu/rnu) rats. The animals where followed by MR imaging for up to 30 weeks, and T1 weighted (T1W) images (with and without Gadolinium contrast) as well as T2 weighted (T2W) MR images was acquired with a Bruker Pharmascan 70/16 7T small animal MR scanner. As tumours developed, diffusion weighted images were also acquired. The rats where sacrified upon sign of illness, and histological examinations of the tumours were performed.

Results and Discussion

Brain metastases from five different patients were implanted (primary: 2 colon, 2 ovarian, 1 lung), and all developed tumours in the rat brain after 6-32 weeks (Figure 1). T1W images with contrast enhancement of colon metastases (Fig 1B) and ovarian metastases (Fig 1E) in the rat brains showed similar contrast enhancement to clinical pictures (Figs 1A, 1D). T2W images displayed extensive edema inside the tumour area (Figs 1C and 1F). Diffusion weighted imaging of the colon metastases in the rat brains further verified the increased intratumoral edema, and the apparent diffusion constant (ADC) increased from $0.682*10^{-3}$ mm^2/sec (control animals) to $0.947*10^{-3}$ mm^2/sec (n=4 rats in each group, $p<0.04$). The histological evaluations demonstrated a demarcated border between the tumours and the normal brain parenchyma (Fig 2A). The tumours exhibited areas with high tumour cell proliferation (Fig 2B), in between regions of apoptosis/necrosis (Fig 2C). Immunostaining for nestin (a stem-like cell marker), showed nestin positivity in the primary biopsies from patients (Fig 2D), which was maintained in tumours developing in the rat brain (Fig 2E).

Conclusions

A novel brain metastases model has been developed in immunodeficient rats, which reflects radiological characteristics, as well as histological and metastatic patterns seen in patients. The model thus represents a promising tool for investigating molecular mechanisms behind metastatic tumour spread, as well as studying the effects of different treatment strategies, such as for instance anti-angiogenic therapy.

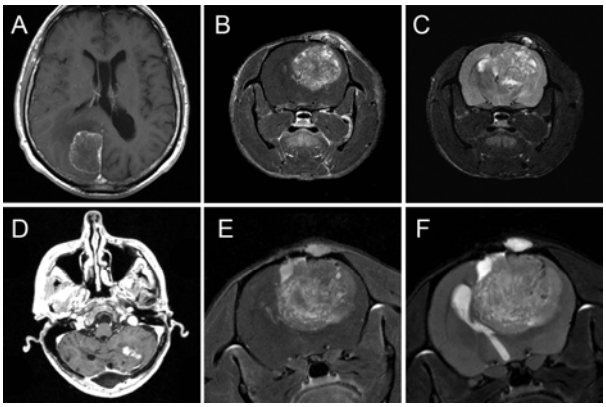

Figure 1. MR images of a colon metastases (A, B and C) and an ovarian metastases (D, E and F). A. T1W MR-image with contrast of patient with colon metastases. B. T1W image with contrast of same tumour four weeks after implantation into the nude rat brain. C. T2W MR image of same tumour shown in B. D. CT image with contrast of a patient with an ovarian metastases. E. T1W MR image with contrast in tumours after implantation into the rat brain. of same tumour 21 weeks after implantation into the nude rat brain. F. T2W MR image of same tumour shown in E.

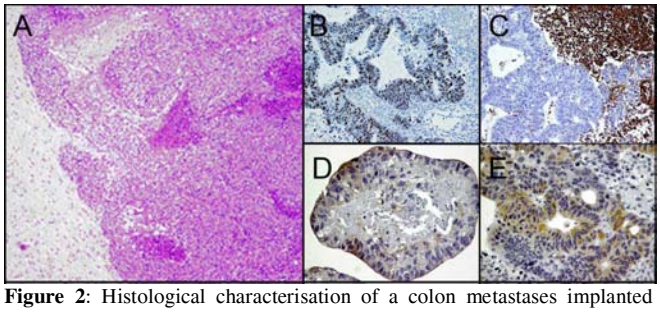

Figure 2: Histological characterisation of a colon metastases implanted into the nude rat brain. A. HE staining of the tumour. B. Ki67 staining showing extensive tumour cell proliferation. C. Necrotic and apoptotic areas were seen after TUNEL staining of the tumours. D. The primary biopsy were positive for nestin. E. The nestin positivity was maintained in tumours developing in the rat brain. All images X20.

References

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