DCE-MRI Assessment of Responses to Hormonotherapy in Preclinical Breast and Prostate Cancer Models in Rats

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Abstract#1941

INTRODUCTION - OBJECTIVES

Angiogenesis has been shown to be an essential factor for tumour growth and development. Recent studies have reported that estrogen regulates angiogenesis in breast cancer, although the mechanism is not yet clearly defined. Several hormonodependent tumour models which have been developed in rodents and characterized, could be particularly helpful to study this phenomenon *in vivo*.

Among these models, the DMBA chemoinduced mammary cancer in Sprague Dawley rats and R3327H prostatic adenocarcinoma in Copenhagen rats (Dunning model) have been successfully used for the evaluation of new therapeutic treatments.

In the present study, we have evaluated and compared the effects of hormone deprivation on angiogenesis related parameters on breast and prostate cancer models using the Dynamic Contrast Enhanced (DCE)-MRI technique. The objective was to investigate whether tumour regression following hormone deprivation could be associated with a change in parameters of tumour blood perfusion by quantitative kinetic model analysis of DCE-MRI using Gadomer (Supplied by Schering AG, Berlin, Germany) as contrast agent.

METHODOLOGY

◆ *Animals :*

Female Sprague Dawley rats were used for the DMBA-induced breast cancer model (induction with 20 mg/rat of DMBA given PO at D0)

Male Copenhague rats were used for the R3327H prostate cancer model

Male and female athymic *Nude* rats were used for the human breast or prostate tumour xenograft models

◆ *Hormonotherapy procedure :*

For DMBA-induced tumour model, hormonal manipulation was performed by ovariectomy or Tamoxifen treatment (10 mg/kg, PO, Q1Dx28), starting at D66 after DMBA induction when 50% of rats had development a tumour or at D86 when each rat had developed at least one tumour.

For the R3327H prostate cancer model, hormone deprivation was performed by castration at D97 when the mean tumour volume reached 350 mm^3

Plasma concentrations of estradiol or testosterone levels were measured by ELISA.

◆ *Histological profil characterization of DMBA-induced tumours*

Tissue Arrays were constituted with different tumour samples excised from control or ovariectomized animals

◆ *DCE-MRI procedures :*

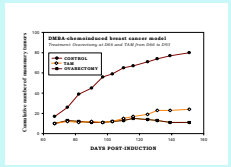
MRI experiments were carried out on a Siemens 1.5 T Magnetom Vision. A flexible surface coil of dimensions 16x34 cm was used. During MRI, the rat was anaesthetized with a ketamine/xylazine mixture administered by intramuscular injection. The rat tail vein had been cannulated for contrast agent bolus administration before placing the animal in the magnet. Rats were positioned in the supine position within the coil. Volume measurement and anatomical description of the tumours were carried out using a multi-slice T2-weighted sequence (TR 4500 ms/TE 54 ms/NEX 2/Slice 2 mm) with an inplane resolution of 400 µm. A FLASH2D gradient recalled echo imaging sequence (TR 200 ms/TE 6 ms/NEX 1/Slice 3 mm) with an inplane resolution of 600 µm was used to evaluate the tumour blood vessel permeability.

The macromolecular contrast agent Gadomer was injected at 0.06 mmol/kg as this type of agent diffuses very slowly, if at all, through normal endothelial barriers and is well suited to evaluate the hypervascularity and hyperpermeability inherent in tumour microvasculature (Brasch et al.,JMRI, 1997).

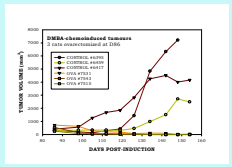
A bi-compartmental and bi-directional kinetic model for the measurement of vascular permeability (K_{tofts}) and tracer leakage space (υ_e) has been previously described by Tofts *et al.*(JMRI, 1997). The tracer uptake curves derived from the signal enhancement in the selected regions of interest (ROI) (eg. on the periphery of the tumour) were analysed using a program developed under PV-Wave (VNI, Boulder, Colorado, US).

Results of *in vivo* antitumour efficacy of hormone deprivation on DMBA-induced and R3327H tumour models

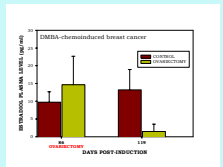
Antitumour activity of Tamoxifen and ovariectomy on DMBA-induced tumour



Antitumour effect of ovariectomy on DMBA-induced tumour

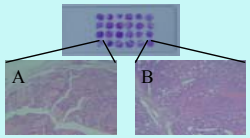


Plasma Estradiol levels



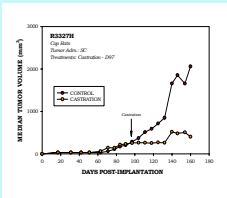
Hormonal manipulation (ovariectomy, Tamoxifen treatment) inhibited the development of new mammary tumours and induced the regression of established ones in DMBA-chemoinduced breast cancer model.

Histology analysis of DMBA-induced tumours by TISSUE-ARRAY

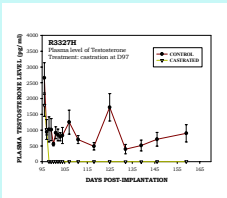


In a panel of 48 DMBA-induced tumours excised from control rats, 85% of the tumours were analysed as invasive carcinomas (B) and 15% were considered as benign tissues (A: a cystadenofibroma)

Antitumour effect of castration on R3327H tumour

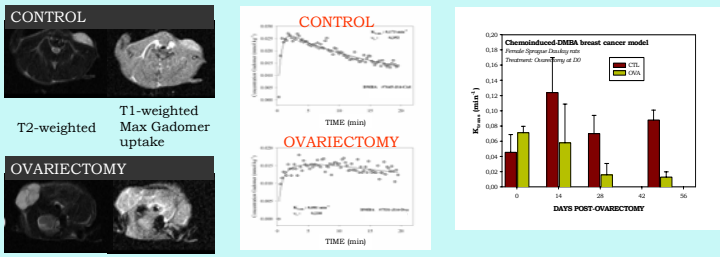


Plasma Testosterone levels

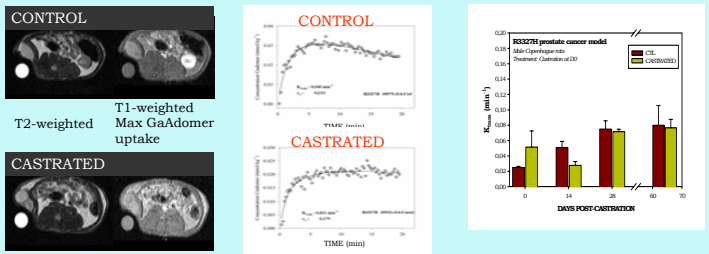


Hormone deprivation caused by castration of R3327H-tumour bearing rats was associated with significant tumour growth inhibition.

Results of MRI analysis for tumour vascular hyperpermeability quantification



T2-weighted anatomical and T1-weighted perfusion images showing localisation and contrast medium uptake of tumors in Control and Ovariectomized rats at D14. Perfusion curves generated from ROIs on the periphery of the tumor show lower microvascular permeability in the ovariectomized rats and the bar chart illustrates the progressive diminution in K_{TOFTS} in the ovariectomy group over time.



T2-weighted anatomical and T1-weighted perfusion images showing localisation and contrast medium uptake of tumors in Control and castrated rats. Perfusion curves generated from ROIs on the periphery of the tumor at D14 after castration illustrate lower microvascular permeability in the castrated rats. The bar chart suggests that the reduction in K_{Tofts} is transitory.

Values of tumour vascular hyperpermeability in a panel of HD and HID cancer models

TUMOUR MODEL	Hormone dependency	Treatment group	K_{TOFTS} (min^{-1}) D0	K_{TOFTS} (min^{-1}) D14	K_{TOFTS} (min^{-1}) D28	υ_e D0	υ_e D14	υ_e D28
PROSTATE CANCER MODELS								
R3327H	HD	Control	0.041 ± 0.021	0.046 ± 0.016	0.091 ± 0.025	0.112 ± 0.049	0.255 ± 0.038	0.694 ± 0.142
		Castrated		0.023 ± 0.011	0.069 ± 0.006		0.204 ± 0.042	0.834 ± 0.041
PAC120	HD	Control	0.043 ± 0.004	0.069 ± 0.005	0.035 ± 0.004	0.280 ± 0.025	0.326 ± 0.006	0.182 ± 0.008
		Castrated		0.039 ± 0.012	0.031 ± 0.002		0.297 ± 0.081	0.168 ± 0.044
PC3	HID	Control	0.083 ± 0.009	0.096 ± 0.018	ND	0.576 ± 0.073	0.440 ± 0.090	ND
		Castrated		0.108 ± 0.013	ND		0.528 ± 0.017	ND
BREAST CANCER MODELS								
DMBA	HD	Control	0.061 ± 0.001	0.173 ± 0.035	0.073 ± 0.022	0.265 ± 0.070	0.381 ± 0.063	0.283 ± 0.070
		Ovariectomy		0.082 ± 0.008	0.023 ± 0.004		0.256 ± 0.022	0.115 ± 0.011
MDA-MB231	HID	Control	0.044 ± 0.013	0.056 ± 0.021	0.051 ± 0.017	0.270 ± 0.079	0.229 ± 0.059	0.236 ± 0.061
		Ovariectomy		0.057 ± 0.005	0.050 ± 0.010		0.323 ± 0.071	0.250 ± 0.059

K_{TOFTS} and υ_e values measured just before hormone withdrawal (D0) were well correlated to the tumour doubling time (data not shown). Hormone deprivation had no effects on K_{TOFTS} and υ_e values in HID models. Hormone deprivation had similar effects on K_{TOFTS} and υ_e values in PAC120 and R3327H prostate models.

CONCLUSIONS

◆ **Gadomer contrast agent was shown to be well suited to quantify small variations in tumour vascular permeability parameters**

◆ **Hormone deprivation was highly effective to inhibit tumour growth in breast DMBA-chemoinduced and prostatic R3327H cancer models in rats.**

◆ **Estradiol deprivation induced significant and prolonged decrease in K_{TOFTS} and υ_e vascular permeability parameters in DMBA-chemoinduced breast cancer model.**

◆ **Testosterone deprivation induced a significant but transient decrease (D14 after castration) in K_{TOFTS} and υ_e parameters both in syngeneic (R3327H) and xenogeneic (PAC120) hormonodependent (HD) prostate cancer models. After D14, K_{TOFTS} and υ_e values of the castrated rats returned to the initial values that was not correlated with a regrowth of the tumours.**

◆ **Hormone withdrawal had no effects on K_{TOFTS} and υ_e parameters in hormonoindependent (HID) breast and prostate cancer models**

◆ **Based on these results, experiments are in progress to quantify hormonotherapy effects on hormonodependent breast tumour vascular permeability.with our MRI system.**

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