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Quantitative Studies of 99 m-Technitium Glucoheptonate Specific Uptake in Human Brain Tumours

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Abstract

Specific uptakes of 99m Technitium glucoheptonate were measured in human cerebral gliomas using gamma camera / computer system. Build up factors measured for 99m-Tc using a water phantom were used to correct for attenuation. The measurements carried out on 19 patients show that the specific uptake of 99m -TcGH in malignant tumor is higher, at least by a factor of 2, than in benign Tumours.

Key words -
Technitium glucoheptonate,
Specific uptake,
Malignant gliomas,
Gamma camera

The role of radionuclide imaging as a primary screening modality for the detection and localisation of brain Tumours has diminished with the advent of x-ray computerized tomography (CT). However, the radioisotopic investigations can better characterise the functional and metabolic activity of different tissues. Cancer cells and tumours are known to manifest higher rates of glucose uptake and glycolysis [1], [2]. Measurements of glucose utilization rates in cerebral neoplasms, using positron emission tomography (PET) with 18F-fluorodeoxyglucose (18F-DG) have been observed to correlate well with the degree of malignancy [3] and could be helpful in differentiating between radiation necrosis and tumor recurrence [4] and predicting the survival in glioma patients [5].

Our earlier work [6], [7] has shown that radiation damage in tumours could be differentially enhanced by the presence of 2-deoxy-D-glucose (2-DG), since (2-DG) inhibits DNA and cellular repair process in cells with high glycolytic activity presumably by reducing the energy supply below a critical threshold level [8], [9], [10], [11]. Combination of 2-DG with ionizing radiations is expected, therefore, to improve the therpeutic efficacy. Malignant cerebral gliomas would be appropriate systems to test this approach since these tumours usually do not metastasize and are known to have high rates of glycolysis [2], [12], [13]. In order to carry out satisfactory clinical trials, it would be desirable to select tumours with high rates of glucose uptake and to monitor the 2-DG concentrations in the tumor. Tumor imaging with PET using FDG

would be the most appropriate method for this purpose. However, PET scanning is very expensive and facilities for PET are at present not available in India. Therefore, we have undertaken research work to develop alternative techniques for this purpose, using the conventional gamma cameras which are commonly used for radioactive isotope scanning.

99m- Technitium labelled glucoheptonate (Tc-GH), another glucose analogue, has been shown to concentrate in majority of the brain tumours [14], [15]. It has been suggested that the increased accumulation of TcGH in the tumor could be related to its being an analogue of glucose and a substrate for energy [16], [17]. In the present work, we have quantitatively investigated the specific uptake of TcGH in different types of cerebral gliomas from the image acquired by a conventional gamma camera / computer system and examined the correlation between the degree of malignancy and the specific TcGH uptake by the tumor.

The difficulties in accurately quantitating absolute radioactivity from projected image of a lesion arise due to the surrounding background radioactivity and irregular intervening structures of variable attenuation characteristics. Further, the variations in countrates across the region of hyper concentration could also be due to thickness variations of volume source, masking the actual distribution of radioactivity.

To overcome these difficulties, different methods for quantitation of organ radioactivity have been proposed [18], [19], [20], [21], [22]. These methods employ geometric means of countrates with 180° separated views [19], [20] or use measured build up factors [22] or transmission with external radioactivity sources to correct for attenuation [21], [23] along with projected countrates. In the present work, we have applied the method based on the use of a set of measured build up factors [22].

The build up factor B(d) is defined as

$$B(d)=c / co . \exp (+\mu d) \quad (1)$$

where c is the countrate measured for a radiation source situated at depth 'd' in the tissue equivalent phantom, co the countrate measured in air at the same source to collimator distance, μ is the linear attenuation co-efficient for soft tissue ($0.15\ cm^{-1}$ for 99m-Tc 140 KeV gamma rays).

For absolute activity measurements using the build up factors, anterior and posterior countrates CA and Cp respectively are acquired with parallel hole collimator. If T is the total phantom thickness and x is the thickness of the volume source, the simultaneous equations for Co and d are solved using CA and Cp.

$$CA=Co .B(d). \exp (-0.15d) .[\mathrm{Sinh} (0.15 x /2) / (0.15 x/2)] \quad (2)$$

$$Cp =Co. B(T-d).\exp[(-0.15) (T-d)].[\mathrm{Sinh}(0.15x/2)/ (0.15x/2)] \quad (3)$$

where 'd' is the depth from which countrate CA is available. Co is the corrected countrate in counts / sec for the region of interest. If E is the calibration factor of the gamma camera system for the measured isotope in counts / MBq, the absolute radioactive tracer uptake A, in the region of interest can be calculated as

$$A= Co/E \quad (4)$$

Material and Methods

A gamma camera with 37 photomultiplier tubes (Phillips, Gamma Diagnost A) with PDP 11/34 computer (DEC) has been used along with a low energy general purpose parallel hole collimator, at 20 % window selection for a single isotope study. 99m-Technitium isotope in the form of pertecnetate is supplied by the Regional Centre for Radiopharmaceuticals, Bhabha Atomic Research Centre, Bangalore.

Measurement of build up factors and calibration of the camera

A precalibrated 99m-Tc pertechnetate standard source is mounted in a vial at known distances from the face of the collimator. A water phantom made of plexiglass of approximate size of a normal skull is used to measure build up factors B(d) as defined in equation (1). Both the build up factors and the calibration factor of the gamma camera for this geometry in counts / MBq are determined. The variation of B(d) for different depths of water is shown in Fig. 1.

Variation of build up factor B(d) as a function of depth in water

Brain tumor scanning with 99m-TcGH

The brain tumor patients were referred either from the Department of Neurosurgery, National Institute of Mental Health & Neuro Sciences or from the Department of Radiotherapy, Kidwai Memorial Institute of Oncology, Bangalore . The diagnosis of the tumor is according to the criteria laid down by Zulch [24]. 555 MBq-925 MBq (15m Ci-25m Ci) of 99m-Technitium glucoheptonate is injected intravenously into the patient, positioned under the gamma camera. The syringe activity before and after injection has been measured with a digital electrometer and pressurized re-entrant ionization chamber (ECIL India). The radio-chemical purity and tagging efficiency are routinely checked by the suppliers of 99m-Tc radiopharmaceuticals. First pass perfusion images and 3 hour static images are acquired by computer and multi format film.

Brain blood pool scanning

The brain blood pool scanning was carried out by the in vivo labelling of red blood cells using the method described by Pavel et al. [25.] 1.2 ml of DTPA kit (BARC) containing stannous chloride (2 mg of stannous chloride, 20 mg DTPA) was injected intravenously in normal saline initially followed by an i.v injection of about 500 MBq 99m-Tcpertechnetate after 30 min. Blood pool scanning was performed after 3 hours as in the case of TcGH brain imaging.

Calculation of the specific uptake

The countrates CA and Cp are available from the computer display for regular regions of interest. Using equations (2) - (4) the TcGH uptake is quantitated in the regions of interest containing the tumor and also in the normal brain by means of an iterative procedure. For the first iteration a value of build up factor B(T/2) is used in both equations (2) and (3) assuming the presence of the source in the midline and a 'semi-correct' value of 'd' is obtained. This value of 'd' is used for referring the numerical values of B(d) and B(T-d) from the graph (fig. 1). Once again the equations are solved for depth 'd' and countrate Co. This process is repeated to get converging values of calculated Co. Depending on the appearance of tumor outline, either A-P (or) R-L pairs of views are used for quantitation. Using the corrected countrate Co, the TcGH uptake is obtained from equation (4). The calculated radioactivity is

expressed as specific uptake by dividing by the volume. The normal brain specific uptake (in Bq/ml) is used to subtract the contributed radioactivity from the normal brain volume overlapping the tumor and to quantify the TcGH uptake in the tumor. The volume of the tumor is calculated using the cross-sectional area of the regular region of interest encompassing the region of increased uptake of the radiopharmaceutical in the computer display and thickness from the 90° perpendicular view. The specific TcGH uptake of tumor can then be calculated.

Results

Scanning of the tumor and the blood pool

Fig. 2 depicts the time dependence of radioactive uptake of TcGH at first pass dynamic phase (Fig 2a) along with static imaging after 3 hours. In Fig. 2a the sequential images acquired 3 seconds post injection are shown representing the transit of radiopharmaceutical bolus through the cerebral circulation. The 3 hour static images of the same patient (astrocytoma Gr. III, right temporal lobe) are shown in Figs. 2b and 2c, which illustrate increased accumulation of 99m-TcGH in the tumor.

- ***(a) 3 secs. sequential images of the brain (AP view) showing the flow of the radioactive bolus in a patient (MVA, M. 52Y) with Astrocytoma Gr. III.***
- ***(b). Anterior view of 3 hour static imaging***
- ***(c) Right lateral view of the same showing the space occupying lesion of hyperconcentration***

Fig. 3 illustrates the case of another patient who was treated for astrocytoma Gr. III (right frontal lobe) tumor by surgery followed by radiotherapy. Clinically, tumor recurrence after one year was suspected. X-ray, CT scan also indicated possible recurrence. The TcGH radioisotope brain scan was positive and there was increased concentration of radiopharmaceutical on the edges of the original tumor site in the projected gamma camera images (Fig. 3). This finding correlated well with the post-operative specimen indicating recurrent, viable tumor (histopathology astrocytoma Gr. IV) around the periphery with a central hemotoma.

- ***Computer image (right lateral view) of a patient (RAM, M 29Y) with recurrent Astrocytoma Gr. IV. showing increased concentration of radioactivity in the viable peripheral part of recurrent tumor***

The accumulation of 99m-TcGH in the tumor could be due to the breakage of blood brain barrier and increase in the blood flow in the tumor or due to increased metabolic activity. To examine this further, brain blood pool imaging using in vivo RBC labelling in the same patient was carried out. Fig. 4 compares images of a tumor (malig. astrocytoma / malig. meningioma) with 99m-TcG, 99m-Tc labelled RBC (in vivo) and x-ray computerized tomography. The tumor volume with radioisotopic imaging is comparable with the tumor delineated and shown by the transverse section in CT scan. The ratio of tumor to normal brain specific uptakes from the brain blood pool images was estimated to be 1.41 against a value of 3.20 from the TcGH images. Therefore, the results indicate that the increase in the blood pool of the tumor alone cannot explain the increased TcGH concentration in the tumor and TcGH uptake may be related to the metabolic activity of the tumor.

- ***Comparison of TcGH image with blood pool image and x-ray CT scan (JAY, F, 25Y) with malignant asti - (a) TcGH image at 3 hours (Anterior view) - (b) Blood pool image of the brain at 3 hours (anterior view) - (c) x-ray CT scan showing the tumor in the right tempero parietal region***

Specific uptake of 99m-TcGH and its correlation with the degree of malignancy

The quantitative values of the specific TcGH uptakes calculated from the studied patients (in Bq/ml per injected MBq of radio pharmaceutical) are shown in Fig. 5a for various grades of brain tumours and normal brain. The ratios of tumor/normal brain specific TcGH uptakes in different types of tumours are presented in Fig. 5b.

Absolute values of 99m-TcGH specific uptakes in normal brain + and different types of brain tumours at 3 hours. • Malignant tumours Δ metatstatic tumours, O benign tumours

The ratio of tumor to normal brain specific uptake in different types of brain tumours. • Malignant tumours, Δ metastatic tumours O benign tumours

The average values of the specific TcGH uptakes in malignant primary brain tumours (Kernohens Grade III and Grade IV) were 12.85 ± 3.18 Bq/ml (n=6) as compared to 6.03 ± 2.84 Bq/ml (n=6) for metastaic brain tumours, 3.67 ± 1.39 Bq/ml (n=7) for benign tumours (astrocytoma grade I-II, oligodendrogliomas, ganglioglioma) and 2.79 ± 0.91 Bq/ml for normal brain. The ratios of tumor/normal brain specific uptake were 4.51 ± 0.84 (n=6) for high grade (malignant) brain tumours, 2.36 ± 0.95 (n=5) for metastatic tumours and 1.55 ± 0.41 (n=7) for benign tumours.

These results indicate that malignant cerebral gliomas Grades III and IV have significantly higher Tc glucoheptonate uptakes than benign tumours. In 2 patients out of 19 the scans did not show accumulation of radioactivity in low grade gliomas which agree well with a detection efficiency of about 90% for tumours reported in the literature [26].

Discussion

Present results indicate that quantitative estimation of 99m-TcGH specific uptake may permit us to differentiate between malignant and benign cerebral gliomas. The errors in the quantitation of tracer uptake from projected images acquired by a gamma camera system may arise during the process of measurement and also from assumptions and approximations made in the procedure of calculation. The measurement errors could for, example, originate from tilted acquisition of counts, matching of opposite views, variation in the gamma camera sensitivity for the entire field of view, variations in the calibration factor and statistical counting errors. The major source of errors are, however, the variations in the sensitivity of the detector and the calibration factor which have been found to be within ± 10% as estimated from repeated measurements.

The important simplifying assumptions and approximations made in the method of calculations are:

(a) normal brain radioactivity is uniformly distributed throughout the brain and the entire brain is considered as a volume source.
(b) the hyperbolic sine correction factor is applied for the total thickness of the skull.
(c) extra attenuation by skull bone is compensated by the scalp activity
(d) the brain transverse section is considered as a rectangle
(e) the measured build up factors are determined from water phantom which is taken as tissue equivalent material.

Taking into account all these factors, the reproducibility of the quantitated specific uptake is expected to be within 15 -20 % accuracy. The specific TcGH uptake has been estimated per MBq of the injected radiotracer. Variations in the labelling efficiency and individual differences in the excretion of the

radioisotope (biological half-life) can be eliminated by calculating the ratio of the specific uptakes in the tumor and the corresponding normal part of the brain.

In studies using 18-FDG positron emission tomography, glucose utilisation rates have been estimated in mg/100g of brain tissue per minute and a mean value of 1.48 ± 0.70 for both grade III and grade IV astrocytomas, normalized with a global value of glucose utilisation in the normal brain has been estimated [3], [27]. In the present study a ratio of TcGH specific uptakes of malignant cerebral gliomas with respect to normal brain in the range 3-6 at 3 hours has been observed. Assuming TcGH is taken up in the tumor as a glucose analogue, the 3 hour static image may represent an integral effect of accumulation of the radiopharmaceutical in the tumor. The exact relationship between the specific uptake of the glucose analogue TcGH and the in vivo glucose utilization needs to be investigated.

The determination of ratio of TcGH specific uptakes of tumor to normal brain may help the clinician in further classifying the tumor. The method used is relatively simple and can be carried out if facilities for conventional gamma cameras with an on line computer are available. Since the present method averages the TcGH specific uptakes by individual tissue elements over the entire volume, it is hoped that the precision of the method could be further enhanced by using cross sectional images from emission computerized tomography (ECT). For our planned clinical trials using 2-deoxy-D-glucose to improve radiotherapy of malignant cerebral gliomas, information on TcGH specific uptakes in tumours may help in selecting suitable patients, if the relationship between TcGH uptake and glucose utilization could be established. It also seems worthwhile to assess further the possible role of TcGH imaging in differentiating brain tumor recurrence from radiation necrosis of the normal brain.

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