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TARGETED CANCER THERAPY: THE POTENTIAL ROLE OF TERBIUM-149

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SUMMARY

Cancer proceeds through a number of quite separate stages in the development of lethal disease. Early stages offer the potential for control if alpha emitting radioimmunotherapy (RIT) is applied. Later stages may be more appropriate for both alpha and beta RIT. In this paper the properties of various alpha and beta emitting radionuclides are examined. Prophylactic therapy for metastatic cancer requires the localisation of dose to the cancer cell and rules out radioactive beta emitting radionuclides. Alpha emitting radionuclides, however, are much more appropriate toxins, as their efficacy depends on the energy and range of the alpha particles. After matching the cancer stage, radiolabel and carrier, we find that ^{149}Tb is the radionuclide of choice in all aspects except production. We report on the production of ^{149}Tb in μCi quantities (0.1 MBq) using heavy ion reactions carried out at a tandem accelerator and in multi-mCi quantities (GBq) using spallation reaction in combination with on-line isotope separation techniques carried out at the ISOLDE facility at CERN.

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INTRODUCTION

The efficacy of radionuclide therapy depends on the type and energy of radiation, the specificity of the carrier and the nature of the target cancer [1]. Orthodox radionuclide therapy of cancer rests on a very limited quantitative basis. Traditionally ^{131}I labelled carriers are used, and more recently radio-lanthanides are being applied for palliation treatment of bone cancer. With the exception of thyroid cancer, radionuclide therapy has been markedly unsuccessful in controlling cancer. The use of monoclonal antibodies was expected to improve the situation, but responses are rare and long term control is not achieved.

CANCER TARGETS

There are four stages of cancer which require quite different approaches to effect control. These are:

i. Cells in transit
   Blood borne cancer cells from the primary tumor break away and travel through the lymphatic system or vasculature, lodging in the lymph nodes or on the walls of capillaries. These cells may be in a dormant state, i.e. the Go phase, and as such are not receptive to chemotherapy which relies on high mitotic rates to enter the cell caused damage to DNA. Thus to target these cells a short range toxicity is required with a highly selective carrier.

ii. Preangiogenic lesions
   Small nests of cells develop in appropriate sites which might stimulate cell division. However, the number of cells is insufficient to create growth factors which would induce angiogenesis in nearby capillaries.

iii. Subclinical lesions
   Sufficient cells are present to stimulate capillary growth which leads to rapid development of the tumor. However, it is still too small to be observable clinically, i.e. tumor diameter is less than say 3 mm, and the patient is symptomatic.

iv. Clinical lesions
   The tumor now manifests itself clinically with symptoms and can be readily observed by various diagnostic methods. For malignant cancers, metastatic disease is widespread, and treatment is mostly palliative in nature.

The current approach in radionuclide therapy is to use beta emitting radioisotopes bound to monoclonal antibodies for the management of clinical cancer [2,3]. We propose that different stages of cancer require different approaches.

RADIATION DOSE CONSIDERATION

The tumorcidal dose required to kill cancer cells in lesions depends on the number of viable or clonogenic cells present, i.e. nominally 60 Gy is required for 10^{12} cells, 40 Gy for 10^{8} cells, 20 Gy for 10^{4} cells and 10 Gy for 100 cells. However, the tolerance dose for bone marrow is 1 - 5 Gy, the GI tract is 1 - 5 Gy, the vascular system is 10 - 20 Gy, and for the whole body the tolerance dose is 2 Gy.

The lethal dose for a subclinical lesion of 10^4 cells is 20 Gy. In order to limit the systemic dose to 2 Gy to the body, a 10 : 1 tumor to tissue dose ratio is required. For 100 cancer cells in circulation only 10 Gy and a 5 : 1 dose ratio is required. But for beta therapy , only 2.5 % of the beta energy is deposited in a cancer cell with ^{131}I, so only 1 : 40 dose ratio is achievable. Thus control of isolated cells is not possible because there is a 5 x 40 = 200 fold short fall in dose ratio and beta radionuclide therapy cannot reasonable be expected to control cancer. For this reason alpha radionuclide therapy must examined. Alpha particles have higher linear energy transfer (LET) than betas, so fewer hits are required to kill a cell. The short range of alpha means much lower dose to surrounding normal cells if a carrier with cancer cell specific properties is used. However, cross fire is not an important aspect of dose delivery as it is with betas, and microdosimetry calculations are required to determine median cell dose and cell survival probabilities. Monte Carlo methods have been applied for determining cell survival after boron capture therapy [4,5] and similar calculations would be required for alpha radionuclide therapy.

PROPERTIES OF ALPHA-EMITTING RADIONUCLIDES

The properties of some alpha emitting radionuclides are shown in Table 1. The most efficacious of all is ^{10}B (after a neutron capture process), with a range less than a cell diameter. However, this activity must be initiated by neutron capture and as such is most suitable for local therapy, as for glioblastome multiforme. The next shortest range is that for ^{149}Tb. $^{223,224}Ra$, and ^{225}Ac have much higher alpha yields than ^{149}Tb, and therefore require lower administered concentrations. The transuranium isotopes, however, require in most cases improved chelation chemistry. ^{211}At is a halide with low in vivo stability and the alpha particle range is more than twice that of ^{149}Tb. ^{212}Bi has a very short half-life but can be made available by a generator. However, the average energy and range of alpha particles are more than twice that of ^{149}Tb.

RADIO-LANTHANIDES FOR RIT

The wide range of radio-lanthanides available enables the efficacy of radionuclide therapy to be optimised for each given stage of cancer. Lanthanides have almost identical biokinetic behaviour in vivo, as long as they are bound via bifunctional linkers to biospecific molecules as shown by Beyer et al [6] and great deal of chelating chemistry has been developed for attaching radio-lanthanides generally [7] as well as ^{153}Sm [8] and ^{166}Ho [9] to monoclonal antibodies. Chelators are for example DTPA, DOTA and TETA [10]. Thus the relative efficacy of alpha and beta emitting radio-lanthanides for killing cancer cells can readily determined by chelation to the same monoclonal antibody.

The different decay properties of radio-lanthanides are given in Tab.2. The volume factor is the cube of the range of the alpha particle emitted, or of the average range of the beta particles, and represents the effective volume of interaction. Normalised to ^{149}Tb, the volume factor varies by many orders of magnitude, even between ^{153}Sm and ^{166}Ho. Clearly, if a specific monoclonal antibody is used as a carrier which targets individual cancer cells, than the probability of cell kill relates to the fraction of energy deposited in

the cell and the hits to kill a cell. These quantities therefore determine the required dose to be administered, which may exceed the critical normal tissue tolerance dose. The number of hits to kill a cell differs by two orders of magnitude between alpha and beta particles [1].

The half-life for ^{149}Tb is very much shorter than that of beta emitters used in radionuclide therapy today. As monoclonal antibodies may take as long as 24 - 48 hours to reach peak uptake in solid tumors, such a tumors are not the target for ^{149}Tb alpha radionuclide therapy. Uptake times required for cell in transit or pre-angeogenic lesions are expected to be very short, and as such, the short half-life of ^{149}Tb may be of advantage.

PRODUCTION OF ^{149}Tb

We have two modes of production to produce ^{149}Tb. The production and decay scheme is shown in Fig.1. The first method uses a tandem accelerator to bombard Pr or Nd targets with ^{12}C ions The saturated yields curve is shown in Fig.2 for both, Pr and Nd targets. Two such measurements have been made with good agreement for Pr but the first Nd measurement suffered from poor beam alignment. Nd provides the higher yields but may require an enriched target of ^{142}Nd in order to obtain high radionuclidic purity.

Much higher yields, however, can be obtained via 1 GeV proton induced spallation reaction on Ta targets. Extremely high isotopic purity is obtained when the spallation reaction is followed by an on-line isotope separation process. We have produced first batches of multi-mCi quantities of ^{149}Tb at the ISOLDE facility [11] (ISOL = isotope separator on-line) at CERN. The 1 GeV proton beam from the CERN booster hits a 120 g/cm^2 Ta-foil target kept at a temperature close to 2400 ^{o}C. The radio-lanthanides are preferentially produced in the spallation reaction and are released very quickly from the target and ionised in high yields by a surface ionisation ion source. The ions are extracted from the target ion source unit and separated according the mass-to-charge ration electromagnetically. Theoretical yields are 2 x 10^{10} atoms s^{-1} compared with the practical production yield of 20 - 30 mCi (about 1 GBq) for a 4 hour collection time. The collected samples contain isobaric impurities which are separated by using cation exchange chromatography (Aminex A5 / α-Hydroxy-isobutyric acid, pH=5). The particle spectrum (electrons, ß$^+$ and α) and gamma ray spectrum from such a carrier-free isotopically separated clean ^{149}Tb sample is shown in Fig.3.

CONCLUSION

The alpha emitting isotope ^{149}Tb offers a more enlightened approach to the control of early stage cancer or leukaemia. If possesses properties which are superior to other alpha emitters and offers efficacy very much greater than that for beta emitting isotopes. It can be produced in quantities adequate for in vivo studies at a high energy tandem accelerator. However, at present spallation reaction combined with an isotope separation process provides already the possibility to produce quantities of this isotope required for patient therapy. The next step is the determination of efficacy in the control of cancer via *in vitro* and *in vivo* models.

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Tab.- and Fig CAPTIONS

Fig.1 Production route and the decay chains (alpha and EC/ß+) for ^{149}Tb

Fig.2 Saturated activity for the production of ^{149}Tb via ^{141}Pr (^{12}C,4n) reaction (squares) and for the ^{142}Nd (^{12}C,5n) reaction (circles) at the ANU tandem accelerator. The open symbols represent a second run for both cases. Better beam geometry improved the ^{149}Tb yield.

Fig.3 Spectra of the particles (a, e^- and ß+) (upper part, taken from [12]) and of the gamma rays (lower part) emitted from mass-separated and chemically purified ^{149}Tb samples 4 and 3.5 hours after the end of the separation, respectively. The spectra were recorded using a 80 mm^2 x 0.4 mm Si(Au) surface barrier detector (particle spectrum) and a 80 cm^3 HP-Ge detector (lower part).

NUCLIDE / daughter	$T_{1/2}$	E_α [keV]	I_α (*) [%]	range [µm]	dE/dx [keV/µm]	E_γ [keV]	I_γ(*) [%]	production route
10-B (n,α)	0	1470 1780	100	7.2 8.9	204 200	478	?	thermal neutron source
149-Tb	4.1 h	3967	17	28	142	Eγ 165 352 652 817 853	> 10 % 27 30 17 12 16	141-Pr (12-C;5n) 149-Tb 142-Nd (12-C;4n) 149-Dy HI accelerator (Tandem) Ta (p;spall)
211-At	7.2 h	5867	42			687	0.25	209-Bi (α,2n) 211-At Cyclotron
211-Po	0.5 s	7450	48	59	114	570 898	0.25 0.25	
212-Bi	60.6 m	6051 6090	25 10	74	106	727	6.75	228-Th --α--> 224-Ra -α-α-α--> 212-Pb Generator
212-Po	0.3 ms	8785	65					
223-Ra	11.4 d	5434 5540 5607 5716 5747 weak	2 9 24 53 10 2	57	115	Eγ 154 269	> 5 % 6 13.6	227-Ac --ß--> 227-Th --α--> 223-Ra Generator
219-Rn	4.0 s	6425 6553 6819	7.5 11.5 81					
215-Po	1.8 ms	7386	100					
211-Bi	2.1 m	6279 6623	16.4 83.4			351	13	
224-Ra	3.7 d	5449 5686	5 95			240	4	228-Th --α--> 224-Ra Generator
220-Rn	55.6 s	6288	99.9					
216-Po	0.15 s	6779	100	58	114			
212-Pb	10.6 h	-	-			239 300	43.4 3.2	
212-Bi	60.6 m	6051 6090	25 10			727	6.75	
212-Po	0.3 ms	8785	65					
225-Ac	10.0 d	5637 5723 5731 5791 5793 5829 others	4.5 2.9 10 8.6 18.1 50.7 5.2					U, Th (p,spall) 229-Th --α--> 225-Ra --ß--> 225-Ac
221-Fr	4.9 m	6127 6341	15.1 83.4	60	113	218	11.6	Generator
217-At	32 ms	7067	100					
213-Bi	45.6 m	5870	2			440	26.1	
213-Po	4.2µs	8376	98					
225-Fm	20.1 h	6963 7022	5 93.4	63	111			HFIR 255-Es --ß--> 255-Fm Generator

Tab.1 Selected alpha emitting radionuclides with therapeutic potential

Decay data were taken from NUCLEUS O.E.C.D./NEA Data Bank 1993,

(*) only strong relevant α and γ lines are presented

NUCLIDE	T1/2	Radiation	E(max) [MeV]	E(mean) [MeV]	range [µm]	volume factor	Eγ [keV]	Iγ (*) [%]	production route
149-Tb	4.1 h	α	3.967	3.97	28	1	s.Tab.1		see Tab.1
47-Sc	3.3 d	β , γ	0.6	0.161	300	1 200	159	70	47-Ti (n,p) 47-Sc reactor
90-Y	64.1 h	β	2.3	0.934	4 200	3 400 000	no		90-Sr --β---> 90-Y generator
137m-Ce	34.4 h	e	0.2	0.203	500	5 700	254	11	136-Ce (n,γ) 137m-Ce reactor
141-Ce	32.5 d	β, γ	0.6	0.171	400	2 900	145	48.4	235-U (n,f) fis.prod. reactor
									141-Pr (p,n) 141-Ce cyclotron
142-Pr	19.1 h	β,γ	2.2	0.809	3 500	2 000 000	1576	3.7	142-Pr (n,γ) 142-Pr reactor
143-Pr	13.6 d	β	0.9	0.315	900	33 000	no		142-Ce(n,γ)143-Ce --β-->143-Pr reactor
147-Nd	11d	β , γ	0.9	0.27	700	16 000	91	28	235-U (n,f) fis.prod. reactor
							531	13	146-Nd (n,γ) 147-Nd reactor
149-Pm	53.1 h	β	1.1	0.366	1 100	61 000	weak		148-Nd(n,γ)149-Nd--β-->149-Pm reactor
153-Sm	46.7 h	β , γ	0.8	0.269	600	121 300	103	28.3	152-Sm (n,γ) 153-Sm reactor
159-Gd	18.6 h	β,γ	1.0	0.312	800	23 000	364	10.8	158-Gd (n,γ) 159-Gd reactor
161-Tb	6.9 d	β , γ	0.6	0.195	800	26 000	75	9.8	160-Gd(n,γ)161-Gd--β-->161-Tb reactor
166-Ho	26.8 h	β , γ	1.9	0.694	2 800	1 000 000	80.6	6.2	164-Dy(2n,γ)166-Dy--β-->166-Ho reactor
169-Er	9.4 d	β	0.3	0.103	200	360	no		168-Er (n,γ) 169-Er reactor
175-Yb	4.2 d	β , γ	0.5	0.13	250	700	396	6.5	174-Yb (n,γ) 175-Yb reactor
177-Lu	6.7 d	β , γ	0.5	0.147	300	1 200	208	11	176-Yb(n,γ)177-Yb--β-->177-Lu reactor

Tab.2 Selected Radionuclides of Rare Earth Elements with therapeutic potential
Nuclear data taken from NUCLEUS O.E.C.D./NEA Data Bank 1993
(*) only the strong gamma lines are summerised
more data for 149-Tb see Tab.1 this paper

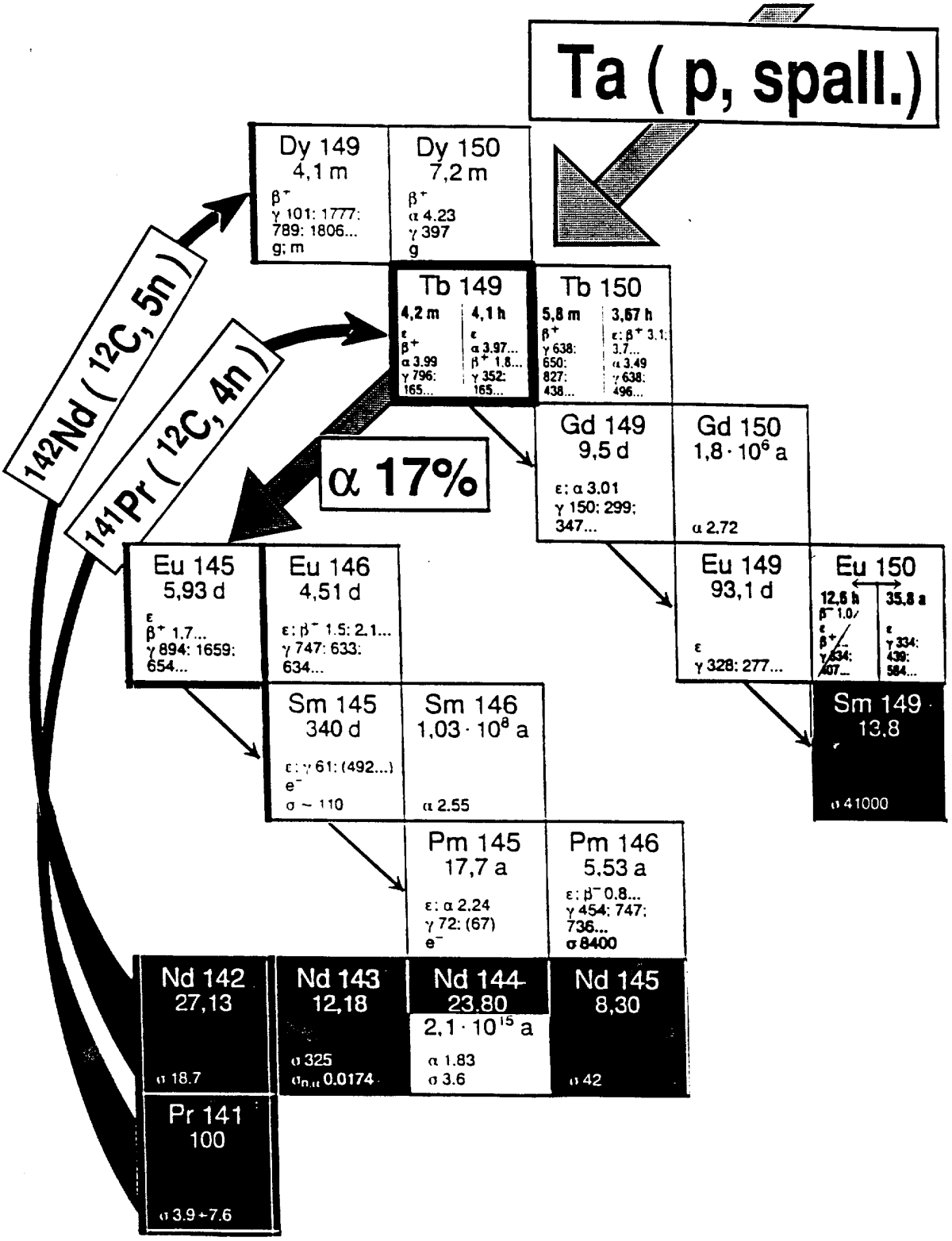

Fig.1 Production route and the decay chains (alpha and EC/β+) for ^{149}Tb

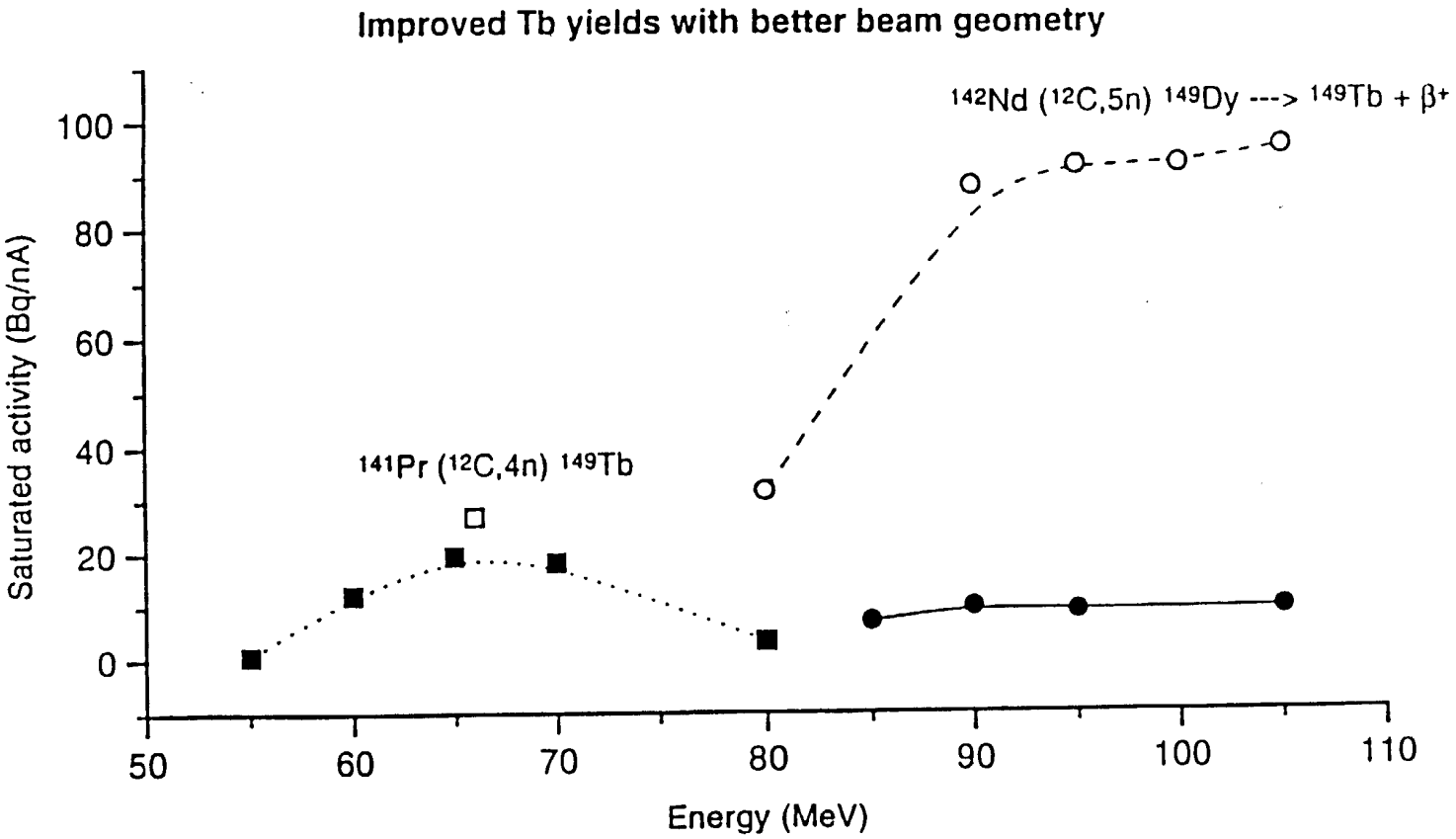

Fig.2 Saturated activity for the production of ^{149}Tb via the reaction ^{141}Pr (^{12}C,4n) ^{149}Tb(squares) and for the reaction ^{142}Nd (^{12}C,5n) ^{149}Dy (circles) at the ANU tandem accelerator. The open symbols represent a second run for both cases. Better beam geometry improved the ^{149}Tb yield.

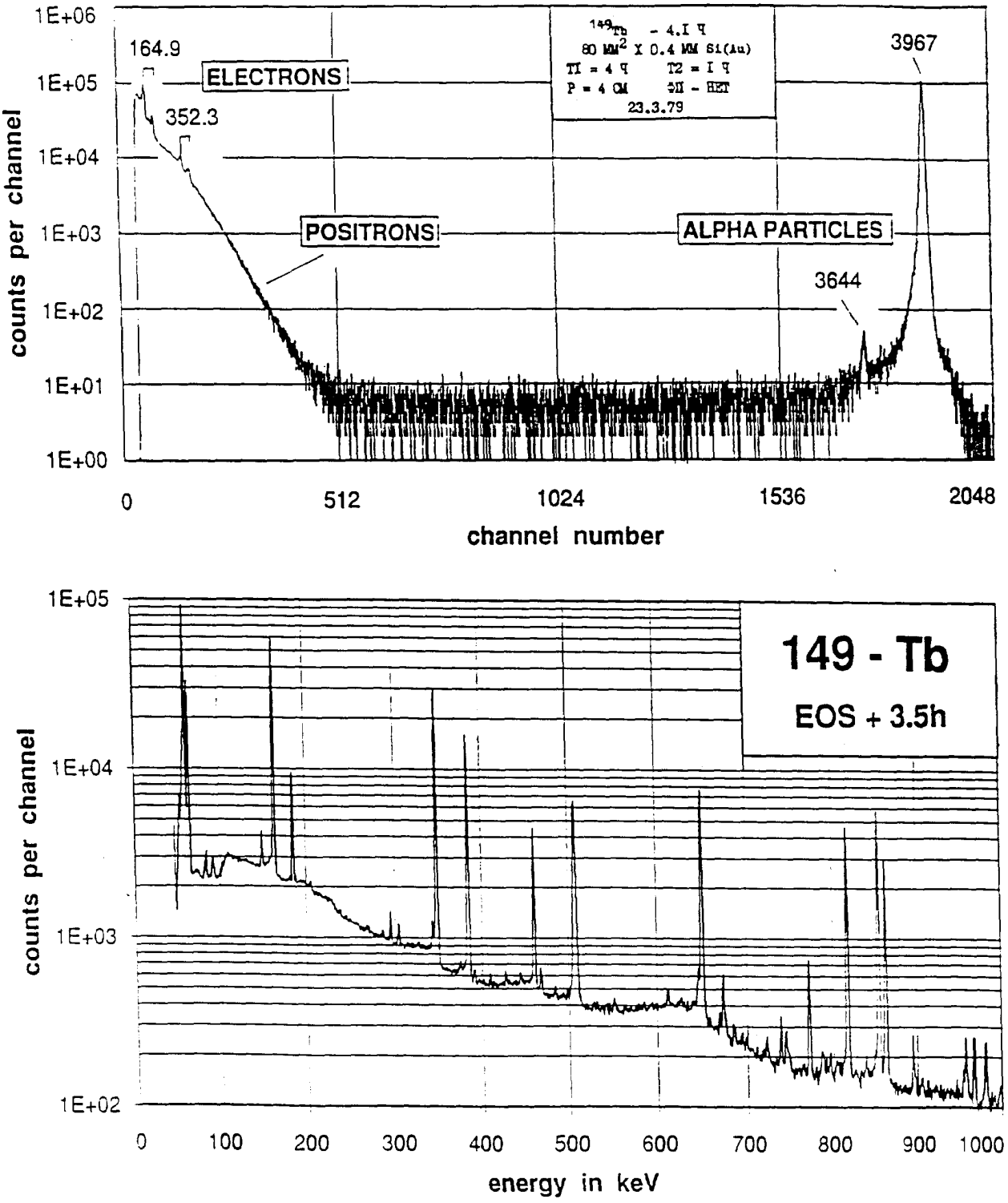

Fig.3 Spectra of particles (e^-, β^+ and α) (upper part) and gamma rays (lower part) emitted from mass-separated and chemically purified ^{149}Tb samples 4 and 3.5 h respectively after end of separation. The spectra were recorded using a 80 mm^2 x 0.4mm Si(Au) surface barrier detector (upper part) and a 80 cm^3 HP-Ge-detector (lower part)