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Clinical Experience with Radiation Enhancement by Hyperbaric Oxygen in Children with Recurrent Neuroblastoma Stage IV

P.A. Voûte, A.J. van der Kleij, J. De Kraker, C.A. Hoefnagel, M.M.C. Tiel-van Buul and H. Van Gennip

The high risk group of patients with neuroblastoma are children over 1 year with stage IV disease. Most series report a maximum of 20% survival at 5 years. For recurrent neuroblastoma stage IV, cure rates are not reported in the literature, but they are nil. Any treatment for recurrent neuroblastoma stage IV remains a therapeutic dilemma. The outcome of radiation therapy is variable. A very important factor in tumour treatment remains tumour hypoxia, and others, such as metabolic factors, also play a role. Combined application of radiation modifiers may influence the final survival rate. In an attempt to improve the survival of recurrent neuroblastoma stage IV, hyperbaric oxygen and radioionated meta-Iodobenzylguanidine (MIBG) was used in a clinical setting. Although survival may not be used as a determinant of the usefulness of a treatment for stage IV neuroblastoma disease, a better one is not available. In this study, at 28 months, a cumulative probability of survival of 32% was recorded for patients treated with [^{131}I]MIBG and hyperbaric oxygen compared to 12% for [^{131}I]MIBG treatment alone. These preliminary results are promising but further studies are needed to reveal substantial therapeutic gain.

Key words: childhood tumour, recurrent neuroblastoma, hyperbaric oxygenation, iodine-131, meta-iodobenzylguanidine

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INTRODUCTION

EXTENSIVE *in vivo* and *in vitro* studies in 1953 by Gray and colleagues [1] proved that the amount of molecular oxygen available to tumour cells at the time of irradiation has a direct influence on radiosensitivity. Several treatment modalities have been applied in clinical oncology to overcome tumour hypoxia, for example, hyperthermia, chemical modifiers of tumour blood flow, hypervolemic blood transfusion, hypoxic cell sensitisers (e.g. misonidasole), and hyperbaric oxygen (HBO). Of all the different radiation enhancement factors, oxygen possesses the highest enhancement ratio [2], 2.7–3.0, provided that molecular oxygen is present during irradiation to act as such. The efficacy of HBO in increasing tumour oxygenation has been established by several investigators [3, 4], and the efficacy of HBO in decreasing radioresistance in tumours had been shown in experimental studies [3, 5] as well as in clinical trials [6]. Controlled trials [7] with HBO have shown a 60% overall benefit, whereas in trials with misonidasole only a 21% benefit has been established. Henk and Smith [6] found a statistically significant higher survival rate and recurrence-free period in patients suffering from head and neck cancer treated with radiotherapy and HBO. They concluded in 1977 that HBO, though a complex, time consuming, and expensive procedure, should not be discarded too easily. Suit [8] reviewed the efforts to assess the clinical value of radiation enhancement. His conclusion was that "if Henk had been able to publish his results in 1962 instead of 1977, efforts would have been made to increase the efficacy of hyperbaric oxygen". In addition to the significance of improving tumour oxygenation when trying to achieve a more successful therapeutic response [2, 9], the radiation response is also influenced by biochemical factors. The metabolism of the neuroblastoma cell is hampered in such an explicit way that exposure to an environment with increased amounts of molecular oxygen leads to an increased production of oxygen-derived free radicals, and consequently to a higher efficacy of radiation therapy.

A multi-place, walk-in hyperbaric chamber for routine HBO treatment is available at the Academic Medical Centre, and HBO therapy in this setting is not felt to be a complex, time consuming procedure. So, in 1989, it was possible to carry out a phase II study for children with recurrent neuroblastoma stage IV, and to give a treatment course of [^{131}I]*meta*-iodobenzylguanidine ([^{131}I]MIBG) enhanced by hyperbaric oxygen therapy.

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MATERIALS AND METHODS

With [^{131}I]MIBG treatment, specific radiation protection measures have to be taken. Thyroid protection is necessary. The patients used potassium iodide orally for 2 weeks (e.g. oral administration of Lugol's iodine 0.2 ml three times daily, or potassium iodine 250–500 mg/m per day) starting 48 h prior to therapy. The [^{131}I]MIBG was administered through a Hickman-line or cannula. 200 mCu [^{131}I]MIBG were given for the first treatment and 100 mCu [^{131}I]MIBG for all further treatments. Prior to therapy, radiation protection restrictions, the use of protective clothing, and radiation monitors were explained to the child and parents.

The principle of hyperbaric oxygen therapy is based on Henry's Law, stating that the degree to which a gas enters into physical solution in body fluids is directly proportional to the partial pressure of the gas to which the fluid is exposed. The clinical value of hyperbaric oxygen therapy for indications such as carbon monoxide intoxications [10], gas gangrene and mixed aerobic–anaerobic infections [11], enhancement of wound healing [12], and radiation damage to bone and soft tissues [13] is well established. The hyperbaric chamber at the Academic Medical Centre at the University of Amsterdam is a multiplace walk-in chamber (98 m^3). A hyperbaric session involved pressurising the chamber from 1 to 3 ATA in 12 min followed up by 75 min at 3 ATA. The decompression profile was derived from the Canadian Forces Decompression tables. The oxygen (5–8 I.min^{-1}) was administered to the patient by a nose/mouth mask, guided in the beginning by transcutaneous polarographic pO_2 recordings ($TcpO_2$, TINA™, Radiometer). Transcutaneous polarographic pO_2 electrodes have been developed [14] for non-invasive monitoring of arterial pO_2 and their clinical value has been proven in newborn infants [15]. The oxygen tension measured by the transcutaneous pO_2 electrode closely approximates the capillary pO_2 value. Using the $TcpO_2$ electrode, we were able to adjust the administered oxygen and balance the $TcpO_2$ value between ±1000 and ±1200 mm Hg. The dip in $TcpO_2$ values on the fourth day was caused by increased activity of the patient (Figure 1). To prevent the child from feeling isolated in the hyperbaric chamber, a liquid-crystal colour monitor was available for video entertainment during HBO sessions. In addition, we observed much higher $TcpO_2$ values associated with reduced oxygen consumption. Prior to hyperbaric oxygen therapy, the patient was made familiar with the hyperbaric chamber, the oxygen nose/mouth mask, and the (de)-compression procedures. Radiation enhancement by HBO started 2–4 days after the initial treatment with [^{131}I]MIBG for a consecutive period of 4–5 days.

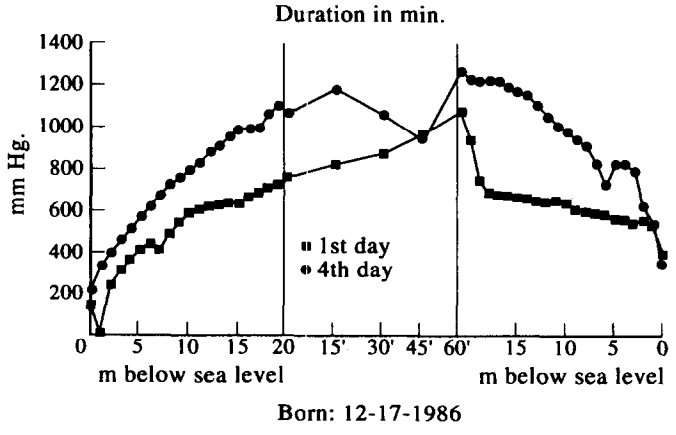

Figure 1. A representative $TcpO_2$ measurement neuroblastoma treatment: [^{131}I]MIBG + HBO.

RESULTS

Two groups of patients were compared. Both consisted of patients with a recurrent neuroblastoma stage IV after previous treatment with high dose chemotherapy treated in different centres with their own protocols, including autologous bone marrow transplantation. In a phase II study (February 1984–May 1990), the first group of patients with recurrent neuroblastoma stage IV was treated with [^{131}I]MIBG. Recurrence and metastasis were at different sites, making it unfeasible to treat patients randomly. In total 36 patients (mean age 6.7 years, ranging between 1.6 and 27.2 years) with more than one [^{131}I]MIBG course were included, because the effect of a treatment course could only be evaluated prior to the next treatment course (Table 1). A cumulative univariate survival curve [16] was constructed of this group. The mean survival of these 36 patients was 15.4 months with a minimum of 2.3

Table 1. Patients characteristics: recurrent neuroblastoma stage IV treated with [^{131}I] MIBG (February 1984–May 1990)

	Age (years)	Number of treatments	Survival after therapy onset (months)
1	3.8	2	26.5
2	1.7	2	9.1
3	26.7	2	6.0
4	3.0	2	18.8
5	3.6	2	14.0
6	3.1	2	0.3
7	6.9	2	7.2
8	27.2	2	6.9
9	1.8	2	9.3
10	4.3	2	8.6
11	4.3	3	9.8
12	3.4	3	25.6
13	1.6	3	13.1
14	2.7	3	33.2
15	4.7	3	54.1
16	4.6	3	25.2
17	8.1	3	13.4
18	2.5	3	17.9
19	2.8	3	24.7
20	7.2	3	10.7
21	8.1	3	8.1
22	5.2	3	2.3
23	4.3	3	10.6
24	2.3	3	7.9
25	3.6	4	21.0
26	2.3	4	43.0
27	4.1	4	8.4
28	9.8	4	14.1
29	7.4	5	26.2
30	13.0	5	13.7
31	7.6	5	15.4
32	4.0	5	15.9
33	2.7	6	10.4
34	14.0	6	9.5
35	3.8	8	27.4
36	22.4	12	35.6

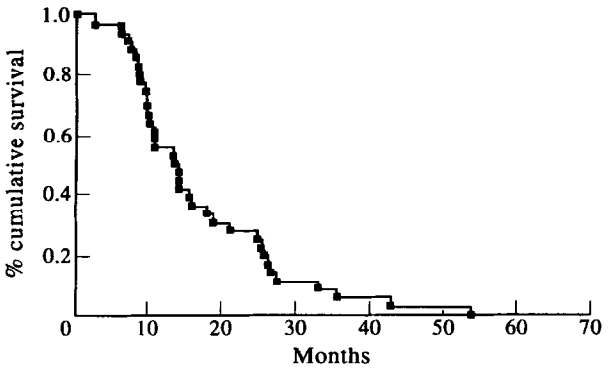

Figure 2. Recurrent neuroblastoma stage IV patients (n = 36) with <1 radioactive MIBG treatment.

months and a maximum of 54.1 months. At 28 months, a cumulative probability of survival of 12% was recorded (Figure 2). All patients have subsequently died.

The second group was treated with a radiation enhanced treatment protocol involving [^{131}I]MIBG and HBO. This phase II study started in November 1989. All patients tolerated the hyperbaric oxygen therapy without additional discomfort. Toxicity from combined treatment was no greater than with [^{131}I]MIBG treatment alone. To date, 27 patients with recurrent stage IV neuroblastoma have entered this protocol (Table 2). Their ages ranged from 1.8 to 15.3 years with a mean age of 7.1 years. From this group, a cumulative survival curve was constructed. Of 27 patients, 14 are alive. At 28 months, a cumulative probability of survival of 32% was recorded (Figure 3).

Table 2. Patients characteristics: recurrent neuroblastoma stage IV treated with [^{131}I] MIBG and HBO (November 1989–March 1994)

	Age (years)	Number of treatments	Survival after therapy onset (months)
1	4.5	2	4.7
2	10.2	2	5.1
3	15.3	2	31.7+
4	15.1	2	11.2
5	11.6	2	26.1+
6	8.3	2	4.1
7	7.7	2	1.8
8	6.6	2	4.8+
9	2.9	2	3.8+
10	5.5	2	6.9+
11	3.8	2	2.9
12	4.5	2	27.6
13	2.2	2	3.6+
14	12.9	3	13.5+
15	4.0	3	47.6+
16	5.0	3	5.0
17	3.4	3	7.1+
18	9.1	3	4.3+
19	5.9	3	9.6
20	10.6	4	4.5
21	4.0	4	61.0+
22	10.1	4	3.4
23	6.3	4	8.9+
24	5.9	4	8.5+
25	1.8	5	49.3+
26	6.3	7	27.1
27	9.0	7	15.3

DISCUSSION

The two studies were on two consecutive groups of patients and the clinical status of the patients was comparable. From the studies, it can be concluded that "unsealed source" radiation enhancement by hyperbaric oxygen is feasible provided that a large hyperbaric chamber is available. Since all patients treated with [^{131}I]MIBG and HBO had recurrent stage IV neuroblastoma after conventional therapy including bone marrow transplants, these results are promising, when compared with the results of the first phase II study on the use of [^{131}I]MIBG but without HBO in patients with recurrent neuroblastoma stage IV.

The efficacy of the combined [^{131}I]MIBG and HBO treatment may be explained by the biochemical characteristics of neuroblastoma cells. Two different defence mechanisms to free radicals can be distinguished in normal cells: the low molecular weight substance-free radical scavengers (e.g. uric acid, reduced glutathione, N-acetylcysteine, thioredoxin), and enzyme systems. A neuroblastoma cell has two defective endogenous defence enzyme systems against oxygen-derived free radicals: a reduced catalase activity, causing an intracellular accumulation of hydrogen peroxide [17] and a 2–3 times elevated ferritin content [18]. The protein bound Fe^{+++} is reduced by the superoxide radical. In the presence of Fe^{++}, hydrogen peroxide can react to form the toxic hydroxyl radical. The toxicity of the hydroxyl radical is associated with a higher peroxidation of proteins, lipids and DNA. These two factors are the main cause for the neuroblastoma cells' vulnerability to oxygen. Furthermore, increased intracellular production of free radicals is associated with hyperbaric oxygen [19]. This is a third factor which contributes to a higher intracellular content of oxygen radicals, and consequently to more damage to tumour cells.

The high specificity and sensitivity of MIBG uptake by tumours of the neural crest makes it a perfect carrier for targeted radionuclide therapy. In combination with [^{131}I], it can be used for unsealed source therapy [20]. The [^{131}I]MIBG has acquired a place in diagnostic scintigraphy and targeted therapy in neural crest tumours [21, 22]. The metabolism of MIBG is characterised by a single active uptake mechanism at the cell membrane, followed by storage in cytoplasmatic neuro-secretory granules. MIBG in itself is an inhibitor of Complex-I, which

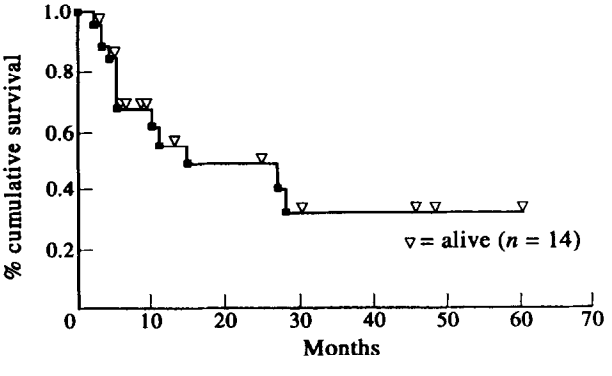

Figure 3. Recurrent neuroblastoma stage IV patients (n = 27) with <1 radioactive MIBG and HBO treatment.

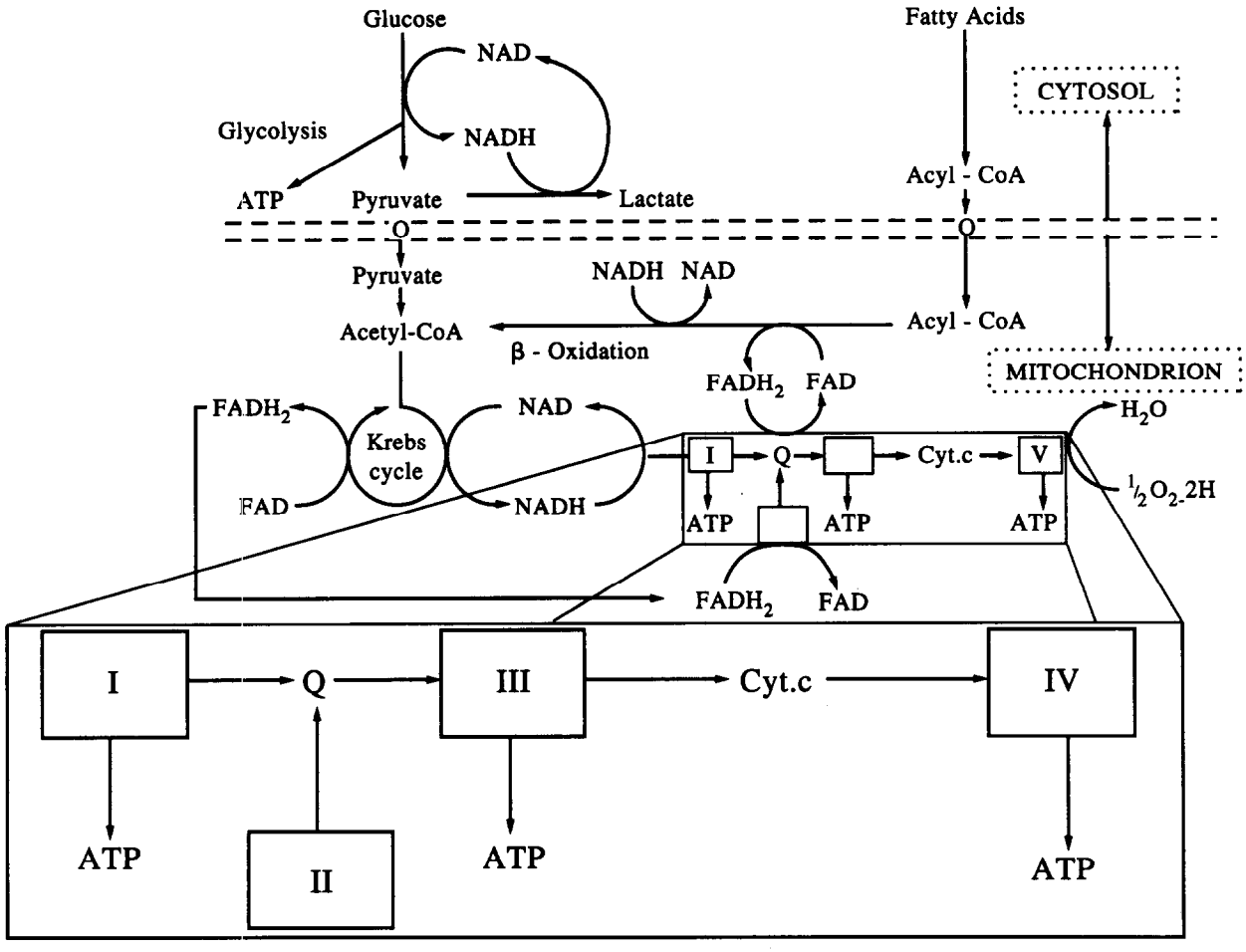

Figure 4.

is part of the enzyme system situated in the mitochondrial respiratory chain (Figure 4). Inhibition of Complex-I leads to the leak of paired electrons out of the respiratory chain, which causes an increased production of the superoxide radical. This superoxide radical is normally converted into hydrogen peroxide by the enzyme superoxide dismutase; subsequently, the hydrogen peroxide is converted into harmless water and oxygen in a reaction catalysed by catalase. However, when catalase activity is reduced, as in neuroblastoma cells, the hydrogen peroxide will partly be converted into the very reactive oxygen-derived free radical, the hydroxyl radical, which contributes to elevated contents of the free radicals: superoxide and hydroxyl. Utilising MIBG for neuroblastoma treatment, therefore, adds a fourth factor in damaging the tumour cells, whereas the radioactivity of [^{131}I]MIBG can be considered a fifth factor.

Our hypothesis is a radiation enhancement by hyperbaric oxygen in children treated for recurrent neuroblastoma stage IV with [^{131}I]MIBG is endorsed by the impaired free radical defence mechanisms of neuroblastoma cells, together with the biochemical properties of MIBG in an environment with a high availability of molecular oxygen, all these being potentially damaging to tumour cells.

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First Line Targeted Radiotherapy, A New Concept in the Treatment of Advanced Stage Neuroblastoma

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33 previously untreated advanced stage neuroblastoma patients were treated with [^{131}I] meta-iodobenzylguanidine (MIBG). The number of treatments varied between 2 and 7 per patient (mean 3). Toxicity was seldom severe. Only thrombocytopenia WHO-grade 4 was noticed. Response was documented before surgery for the primary tumour was performed. There was one complete response (CR), 18 partial responses (PR), 11 had stable disease (SD) and 3 had progressive disease (PD). After MIBG therapy and surgery, 12 of 33 patients achieved a CR. This approach is feasible, comparable to multidrug chemotherapy in efficacy and less toxic. Long term results are not known yet.

Key words: [^{131}I] meta-iodobenzylguanidine, neuroblastoma, targeted radiotherapy, paediatric
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INTRODUCTION

In 1967, Morales and associates [1] first demonstrated that C-14-labelled epinephrine and its precursors concentrated in the adrenal medulla. In the first human studies with [^{131}I]MIBG (meta-iodobenzylguanidine) in 1981 Sisson and associates [2] demonstrated successful detection of phaeochromocytoma with this radiopharmaceutical in 8 patients. The growing interest in targeting of therapeutic radiation and the already existing experience with MIBG for diagnosis and treatment of phaeochromocytoma led to its use in neuroblastoma [3, 4].

At The Netherlands Cancer Institute (1264) total body scintigrams in 418 patients with neural crest tumours have been made. [^{131}I]MIBG correctly demonstrated primary, residual and recurrent tumour, as well as diffuse bone marrow infiltration, skeletal, lymph node and soft tissue metastases [5]. 140 of these patients have subsequently received therapeutic doses of [^{131}I]MIBG. Although the exact mechanism of uptake of [^{131}I]MIBG remains unclear, it is believed to share the uptake and storage mechanism with norepinephrine, in two ways: a sodium-dependent system with a high affinity but low capacity, which is easily saturable, and a sodium-independent, apparently unsaturable process of passive diffusion [6]. Unlike norepinephrine, MIBG is not metabolised and is excreted unaltered via the kidneys. 70–90% of the administered activity is recovered in the urine within 4 days.

Since early 1984, treatment with [^{131}I]MIBG was given to patients with progressive, chemotherapy-resistant disease to achieve palliation [7, 8]. After some time, a second group of patients was included, namely those with residual disease after chemotherapy and surgery. In this group there has already been a curative intention. The results in these patients were

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