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ACUTE EFFECTS OF ACCELERATED RADIOTHERAPY IN COMBINATION WITH CARBOGEN BREATHING AND NICOTINAMIDE (ARCON)

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Combining accelerated radiotherapy with carbogen and nicotinamide (NAM) has been proposed as a strategy to overcome the sparing effect of tumour clonogen repopulation and hypoxia. Six patients with squamous cell carcinomas of the head and neck were given accelerated radiotherapy, carbogen breathing and high dose nicotinamide in order to evaluate the feasibility of this treatment regimen. The patients received radiotherapy in two daily fractions of 1.8–1.9 Gy, five days/week, total dose 54–57.6 Gy, in an overall treatment time of 19–22 days. The interfraction intervals were 7–8 hours between the two fractions on the same day. Carbogen breathing was started 5 minutes before and went on during each radiation fraction. A variety of NAM doses were administered orally in conjunction with radiation therapy and analyses of plasma concentrations of NAM and its metabolites were performed. The most common side-effect from NAM was nausea and vomiting, which in one case hampered further NAM administration. The side effects were not related to plasma levels of NAM or its main metabolites. Additionally, one patient with preexisting heart disease developed a severe hypotension and renal dysfunction. All acute reactions healed without further complications. The mucosal reactions were generally brisk. Thus, the combination of accelerated radiotherapy with carbogen and NAM seems to be tolerable.

In normal tissues virtually all cells are well oxygenated, whereas in tumours the presence of hypoxic cells have been implied (1). These cells are relatively resistant to radiation, and may survive, causing a treatment failure (2). Principally, the oxygenation of the tumour cells can be increased in three ways; by increasing the oxygen concentration in blood, by increasing the tumour blood flow, and by decreasing the oxygen consumption in the tumour cells. In a number of animals studies, treatment with normobaric oxygen or carbogen ($95\%O_2 + 5\%CO_2$) have shown increased tumour response to radiation (for review see (3)). Carbogen improves tumour oxygenation by increasing the amount of dissolved oxygen, enhancing tumour blood flow and may shift the haemoglobin dissociation curve to the right (4, 5). While chronic hypoxia, due to relatively poor vascular supply of the tumour, can be manipulated by changing blood flow and blood oxygenation it has been observed that a number of experimental tumours exhibit a cyclic closure of vessels leading to acute hypoxia (6, 7). This cyclic closure of tumour vessels is inhibited by nicotinamide (NAM) (8) and the primary mechanism for the radiosensitising effect of NAM, in vivo, seems to be improved oxygenation of the tumours (9). As a consequence of this, the combination of carbogen breathing and nicotinamide seems to offer a solid theoretical ground for overcoming the problem with tumour hypoxia. Indeed, animal experiments with fractionated radiotherapy and the combination of carbogen and nicotinamide have shown substantial therapeutic gain (10, 11).

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Many human tumours such as squamous cell carcinomas have potential tumour doubling times shorter than five days (12). In order to overcome tumour cell proliferation during radiotherapy, shortening of overall treatment time by increasing the number of daily fractions is a feasible way. Combining accelerated radiotherapy with carbogen and NAM was therefore proposed as a strategy to overcome the sparing effect of tumour clonogen repopulation and chronic and acute hypoxia (11).

In the present pilot study 6 consecutive patients with head and neck cancer have been selected for evaluating the feasibility of giving accelerated radiotherapy with carbogen breathing and high dose nicotinamide (ARCON).

Material and Methods

Patients. Six previously untreated patients with squamous cell carcinomas of the head and neck region were selected for ARCON therapy. Patients were included after informed consent. The study was approved by the local ethics committee. The sex, age, tumour site, and TNM-classification of the patients are shown in Table 1.

Radiotherapy. The radiotherapy schedule was based on earlier experiences where 60 Gy were given in 2 Gy fractions, twice daily, 5 days/week in an overall treatment time of 19–22 days. The interfraction intervals were 7–8 h between the two fractions on the same day. From theoretical consideration on the enhanced effect of radiation on normal tissues, by carbogen and/or nicotinamide the radiation dose was 10% lower in the present study (i.e. 1.8 Gy b.i.d., total dose 54 Gy). The treatment volumes included the tumour with a margin of 2 cm. If no lymph node metastases were present the lymph nodes closest to the tumour were included. In cases with lymph node involvement, the whole ipsilateral neck was treated. Shrinking field technique was used; after 41.4 Gy the prophylactically treated nodes were excluded and the treatment fields encompassed known tumour manifestations with a margin of 2 cm. All patients received sucralphate mixture for mouth rinsing 1 g three times daily in an attempt to decrease mucosal reaction. This was based on results from our department showing protection against intestinal reactions (13).

Nicotinamide. The doses of NAM are shown in Table 1. NAM was given orally as an aqueous solution (200 mg/ml) $1\frac{1}{2}$–2 h prior to radiotherapy, either before the first daily fraction in cases with one daily dose of NAM, or before each fraction in cases with two daily doses. The patients were fasting before the first daily dose of NAM. Samples for the determination of the concentration of nicotinamide and its metabolites in plasma were taken during the whole course of therapy, stored at −20°C and subsequently analysed by high performance liquid chromatography using the method of Stratford & Dennis (14).

Carbogen breathing. Carbogen (95%O_2 and 5%CO_2) was inhaled via a diver's nozzle. A nose clip was adapted in order to prevent nasal breathing. Carbogen breathing started 5 min before, and went on during each fraction for a total time of approximately 15 min. The flow rate of carbogen was approximately 10 l/min.

Assessment of the acute effects. Assessment of acute reactions was done weekly during the course of radiotherapy. After cessation of therapy the reactions were assessed at 2, 4, and 6 weeks and then with 4–5 weeks' interval until they had resolved. The scoring system was a four-grade scale (none—severe) where epithelitis, pain and dysphagia were assessed. At each follow-up the mucosal reaction was registered by the physician. Furthermore, a subjective estimation of pain and functional impairment was done by the patient. On each occasion a questionnaire was completed and photgraphs were taken. Liver enzymes (GOT, GPT, LD, γ-GT), serum electrolytes, and creatinine were analysed weekly. Blood pressure was measured daily.

Results and Discussion

Tolerance. In all cases there was a brisk acute reaction of the mucosa. Confluent mucositis was observed during the third week of treatment in 5/6 cases (No. 3. was excluded

Table 1

Patients included in the pilot study, tumour parameters, radiation and nicotinamide doses

Case	Sex	Age	Tumour site	TNM	Dose/fraction (Gy)	Total dose (Gy)	NAM-dose	Body weight (kg)
1	M	64	Soft palate	T2N0M0	1.8	54	3 + 3 g 5 days/week	69
2	M	59	Tonsil	T2N2M0	1.8	57.6	3 + 3 g first week 6 + 3 g second week 6 + 6 g third week	97
3*	M	57	Mesoph.	T2N0M0	1.9	57	6 + 3 g 5 days/week	66
4	M	56	Tonsil	T4N2M0	1.8	54	6 + 3 g 5 days/week	73
5	M	50	Tongue	T4N1M0	1.8	54	7 g 5 days/week	70
6**	M	46	Tonsil	T1N2M0	1.8 2.0 after 26 fractions	54.8	7 g 5 days/week	70

*Stopped NAM after 6 treatment days due to suspected side-effects.
**Stopped NAM after 13 treatment days due to nausea and vomiting.

earlier in the study due to other side effects, described below). In one patient (No. 4) the confluent mucositis remained for 3 months. The patients' estimates of pain were parallel to the grade of mucositis. 3 patients needed tube feeding for 2–8 weeks (the higher figure for No. 4). One patient had an extensive moist desquamation of the skin of the neck, the reaction resolved in 6 weeks. All acute reactions eventually healed without further complications. The reactions were similar to earlier studies on accelerated fractionation alone (2 Gy/fraction b.i.d. to a total dose of 60 Gy in 19–22 days).

In 3/6 cases (Nos. 2, 4, 6), nausea and vomiting appeared in conjunction with the intake of NAM. The symptoms did not improve when ondansetron or metoclopramide were given. In these cases naso-gastric tubes were used and the nicotinamide was administered via the tube. This improved the situation for two patients (cases 2 and 6), although the nausea and vomiting did not stop. Patient No. 6, however, had to discontinue NAM after 26 fractions due to severe nausea and vomiting. In this case carbogen breathing and radiation was continued. One patient developed severe hypotension which hampered further drug administration. This patient is discussed in detail in the following section. The dose per fraction was increased to 2 Gy when no nicotinamide was given. None of the five remaining patients had any hypotension. Weight loss was 0–15% (median 8%) over the treatment time. We have not been able to identify any adverse reactions due to carbogen breathing.

Nicotinamide concentrations. Plasma concentrations of NAM increased with dose. A summary of pharmacokinetic data and side-effects is shown in Table 2. In case 1, who received 3 g twice daily, a peak concentration of 510 nmol/ml was attained 1 h after the first dose. At 8 h (1 h after the second dose), the peak level rose to 760 nmol/ml. Similar peak levels were seen on the 14th day of treatment. Plasma concentrations 24 h after NAM dose (days 2 and 15) showed that NAM concentrations were close to zero. The estimated half-life ($T^1/_2$) of NAM was 4–5 h. Patient No. 2 received an escalating NAM dose. This patient started treatment on a Wednesday. The first week of treatment (3 + 3 g), peak levels of NAM reached 440 nmol/ml and the initial $T^1/_2$ was 5 h. During the second treatment week (6 + 3 g), a peak level of 950 nmol/ml was recorded. The plasma concentration 24 h after the first dose was 170 nmol/ml and increased to 400 nmol/ml after the second dose. Further escalation of the dose led to marked drug accumulation (peak value, 1 950 nmol/l); however the NAM was cleared following the weekend (which occurred after three days with the dose 6 + 6 g since the treatment started on a Wednesday) without drug. The $T^1/_2$ was estimated to approximately 9 h.

Fig. 1 shows the plasma concentrations of NAM observed in patient No. 3, who received 6 + 3 g on radiotherapy days. The peak concentration after 6 g on day 1 was

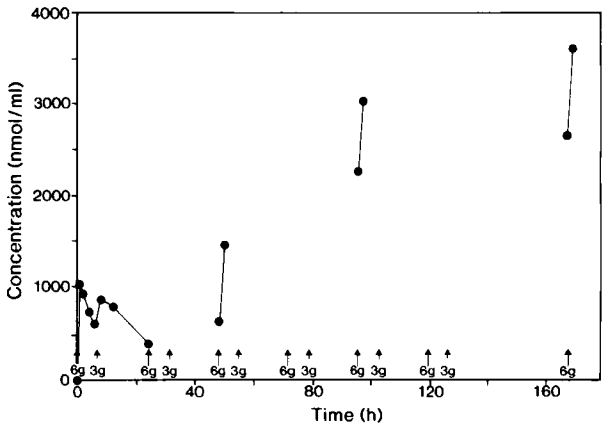

Fig. 1. The concentration of nicotinamide in plasma during the first week of treatment for case No. 3 showing the accumulation of the drug. The time and size of the nicotinamide doses are indicated by arrows.

1 030 nmol/ml, and the initial $T^1/_2$ was quite fast at 6.4 h. However, following the 3 g dose, elimination appeared to be slower. This led to significant drug accumulation after two days. Furthermore; then it appeared to be a large reduction in drug clearance over the next two days, since the value before dosing on day 5 was 2 240 nmol/ml. The patient actually received drug (but no radiotherapy) on Saturday, but the pre-dose NAM concentration the following Monday was 2 620 nmol/ml, rising to 3 570 after 6 g. At this time (total dose 54 g of NAM), the patient developed a severe hypotension (SBP = 60 mmHg), and drug administration was discontinued. The liver enzymes (GOT, GPT, LD, γ-GT) rose to 2–3 times the reference values and serum creatinine increased to about 3 times the upper reference value. The patient had a known cardiomyopathy, probably due to previous alcohol abuse, and was on treatment with enalapril, an inhibitor of angiotensin converting enzyme (ACE). The patient was referred to intensive care and the blood pressure, liver enzymes and kidney function returned to normal within two weeks without any specific therapy. The reason for this reaction is not known. The existing heart disease and the chronic medication may have interacted with the NAM clearance. It appeared that renal elimination was impaired, since the metabolism of NAM looked normal, with expected levels of the major metabolites, the 2-pyridone, and 2-methyl-nicotinamide, while the very polar N-oxide rose 5-fold over the weekend. This patient died two months after the end of therapy of cardiac failure, without any obvious relation to the earlier events.

Fig. 2 shows data on patient No. 4 in the first week of treatment with NAM, illustrating the slight accumulation of the drug over the 5 treatment days, followed by elimination over the weekend. The highest level observed was 1 430 nmol/ml following the second dose on day 2. In the second and third weeks, 24 h concentrations were slightly lower than those seen in the first week (data not shown).

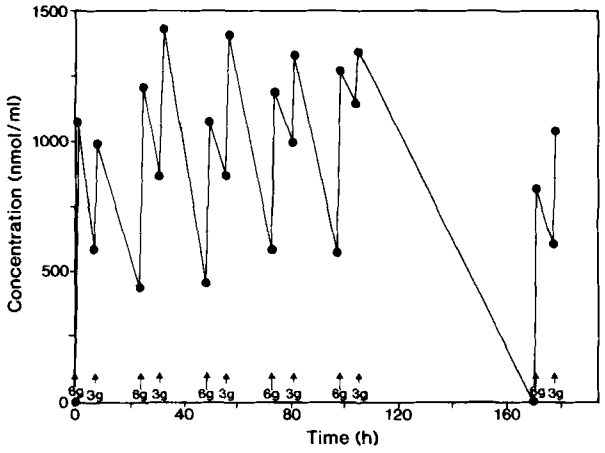

Fig. 2. The concentration of nicotinamide in plasma during the first week of treatment for case No. 4. The time and size of the nicotinamide doses are indicated by arrows.

Patient No. 5 had by day 5 accumulated very high levels of NAM, with a 24 h (pre-dose) concentration of 1 030 nmol/ml, rising to 2 090 nmol/ml at 1.9 h. This implied rather slow clearance in the first week, with $T^1/_2$ of approximately 12 h. However, in the second and third week, elimination appeared rather faster, since the 24 h levels did not rise markedly during treatment and the peak values varied between 700 and 1 400 nmol/ml. Case No. 6 did not appear to show such marked accumulation of drug during treatment with 24 h values between 0 and 360 nmol/ml and peak values between 640 and 1 150 nmol/ml.

The profiles of the main metabolites (nicotinamide N-oxide, the 2-pyridone and 1 methylnicotinamide) showed similar qualitative pattern in all the patients. For each individual, the concentration of each metabolite reached a plateau, which then remained elevated after the NAM had been cleared, but which decreased over a weekend without drug. The peak concentration for each metabolite in the six patients is shown in Table 3. There did not appear to be any correlation between the incidence of toxicity (nausea and vomiting) and the peak levels observed. However, case No. 3, who suffered the severe hypotension, had greatly elevated plasma N-oxide, and somewhat higher methylnicotinamide on the last day.

Table 3

Peak plasma concentrations of the main metabolites of nicotinamide for each patient

Case	N-oxide (nmol/ml)	2-pyridone (nmol/ml)	1-methyl-NAM (nmol/ml)
1	92	148	47
2	160	78	49
3	83	150	45
	415*		74*
4	250	54	33
5	91	57	37
6	87	84	35

*On last treatment day

To conclude, the normal tissue reactions with this regimen seems to be severe but tolerable for motivated patients. However, the patients need help with nutrition and oral hygiene during and after treatment. The most frequent side-effect from NAM was nausea and vomiting. This prevented one patient from completing the study. In two cases the use of a naso-gastric tube made it possible to continue NAM medication for a few days, in spite of nausea and vomiting. Ondansetron and metoclopramide did not alleviate these symptoms. A different form of administration might lead to improvements of the tolerance to NAM. The concentration of NAM or its metabolites, in plasma, did not seem to correlate to the degree of nausea and vomiting. Also, caution should be given if using this drug in patients with cardiovascular disease and medically treated, that may potentiate the effect of NAM. In such cases monitoring of plasma levels of NAM seems essential. At six weeks after completion of the therapy none of the patients showed any evidence of residual tumour. The

Table 2

Summary of pharmacokinetic data and side-effects

Case	Dose of NAM (g)	Peak conc. of NAM first day dose I (nmol/ml)	Peak conc. of NAM first day dose II (nmol/ml)	Accumulation of NAM	Clearing of NAM during weekend	Side effects
1	3+3	510	760	No	Yes	None
2	3+3	440	475	No	Yes	None
	6+3	950	Missing	No	Yes	Nausea, vomiting
	6+6	950	1630	Yes (see text)	Yes (see text)	Nausea, vomiting
3	6+3	1030	880	Severe	No	Nausea, vomiting, severe hypotension
4	6+3	1080	1000	Slight	Yes	None
5	7	1120	–	Severe (first week)	Yes	None
6	7	640	–	No	Yes	Nausea, vomiting

epithelial and tumour reactions were similar compared to earlier results on accelerated fractionation alone, i.e. brisk but reversible mucositis and a high proportion of complete regression of tumours.

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