Treatment of Early Stage Breast Cancer: NIH Consensus Development Conference Abstracts. 1990, 61.
62. Fisher B, Redmond Carol. NSABP B-13 methotrexate+5-FU in women with estrogen receptor negative, node-negative breast cancer. *Treatment of Early Stage Breast Cancer: NIH Consensus Development Conference Abstracts*. 1990. 63.
63. Cummings FJ, Gray R, Davis TE *et al.* Tamoxifen versus placebo: Double-blind adjuvant trial in elderly women with stage II breast cancer. *NCI Monogr* 1986, **1**, 119–123.
64. Castiglione M, Gelber RD, Goldhirsch A. Adjuvant systemic therapy for breast cancer in the elderly: competing causes of mortality. *J Clin Oncol* 1990, **8**, 519–526.
65. Mouridsen HT, Andersen AP, Brincker H *et al.* Adjuvant tamoxifen in post-menopausal high-risk breast cancer patients: present status of Danish Breast Cancer Cooperative Group Trials. *NCI Monogr* 1986, **1**, 115–118.
66. Goldhirsch A, Gelber RD, Tattersall MNH, Rudenstam CM, Cavalli F, for the Ludwig Breast Cancer Study Group. Endocrine adjuvant therapy for breast cancer. *Lancet* 1985, **i**, 1274.
67. Pritchard KI, Meakin JW, Boyd NF *et al.* A randomized trial of adjuvant tamoxifen in postmenopausal women with axillary node positive breast cancer. In: Jones SE, Salmon SE, eds. *Adjuvant Therapy of Cancer IV*. Orlando, Grune & Stratton, 1984, 339–347.
68. Henderson IC. Adjuvant systemic therapy: state of the art, 1989. *Breast Cancer Res Treat* 1989, **14**, 3–22.

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What has Radiation Biology Contributed to the Evolution of Radiotherapy?

J.C. Horiot

BACKGROUND

TWENTY YEARS ago, most radiotherapists would usually have thought that the only way to improve results was to increase the dose to the tumour. This was right to some extent. This concept was, of course, limited by the normal tissue tolerance. However, it reflects why most efforts occurring between 1950 and 1970 were dominated by this approach. Hence, the clinical advances in developing the performance of linear accelerators, radiation physics dose calculations and brachytherapy techniques occupied the foreground of the theatre of clinical research while radiobiologists were obscurely working in the wings.

SUMMARY OF EMPIRICAL CLINICAL EXPERIENCE

The evidence of a dose-time relationship was clearly documented from clinical experiences in most solid tumours (head and neck, breast, gynaecological): radiotherapy was preventing the growth of subclinical aggregates of tumour cells in about 90% of the cases when doses of about 50 Gy were delivered in 5 weeks and 25 fractions. Increasing the dose to 70 Gy was usually sufficient to control 70–90% of tumours of less than 3 cm but the prediction of tumour response was increasingly difficult and the failure rates much higher when tumour volumes were beyond 5 cm diameter. It was then obvious that mathematical models based upon an even damage at each fraction up to the killing of the last tumour cell after a sufficient number of fractions was not fully representative of the response of most tumours to radiation. Hence, reasons for radiation resistance were investigated and gradually emerged from radiobiology and radiobiologically oriented clinical experiments.

RADIATION RESISTANCE

Does radiation resistance exist? Why, for an equal tumour volume, does radiation therapy fail in some tumours while controlling others? Very few human tumour cell lines can survive doses of 70–80 Gy delivered under optimal conditions. Intrinsic radiation resistance is a rare phenomenon (high grade glioblastoma for instance) and even in some "radiation resistant" tumours such as malignant melanomas, very large variations of radiation response are observed, including excellent responsiveness to doses consistent with normal tissue tolerance.

Then, in most situations, the so-called "resistance to radiation" will in fact be resulting from (1) Geographical misses: either by an insufficient coverage or by ignoring the actual tumour spread (macroscopic or microscopic); (2) impossibility to deliver the required dose to the tumour without serious damage to vital normal stuctures (such as lung, heart, spinal cord, bowel, kidney); (3) radiobiological factors reducing the therapeutic gradient between normal tissues and tumours such as hypoxia and cell kinetics.

Several options are offered in modern radiotherapy to fight against each of these three major causes of failures. This paper will only concentrate on the radiobiological factors of radiation resistance.

THE OXYGEN EFFECT

Hypoxia has been known as a cause of radioresistance since 1935 [1] and was later documented in the laboratory by Gray [2] and Lacassagne [3]: most tissues irradiated in the absence of oxygen are 2.5 to 3 times more resistant than in the presence of oxygen at a normal pressure. A number of applications of this phenomenon has led to experimental work both in the laboratory and in clinical research:

- Irradiation under hyperbaric oxygen pressure (HBO).
- Irradiation using beams of a lesser oxygen dependence such as neutron beams.
- The use of oxygen mimetic radiosensitisers and bioreductive agents.

Radiotherapy and hyperbaric oxygen

Nearly all randomised trials have shown a significant improvement of local control in patients treated under hyperbaric oxygen (at an oxygen pressure of 2.5 to 3 atmospheres) as compared to

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air [4, 5]. Advanced cervix carcinoma was the most common tumour site addressed in these trials. Unfortunately, this advantage was nearly counterbalanced by an increase in severe late complications in the experimental group for two reasons: (a) the use of a larger dose per fraction due to the technical difficulties of irradiating a patient in a specially designed tank; and (b) the insufficient selectivity of the process probably enhancing the damage to normal tissues as well.

The technical burden of irradiating patients using normal fractionation in an oxygen tank was a strong limitation for starting new trials. Thus, the advent of radiosensitisers led to the abandonment of HBO experiments. However, some radiobiologists are still convinced that blood transfusion and oxygen breathing during radiotherapy sessions should be evaluated since it might be a cheap and efficient way to enhance sensitivity to radiotherapy in tumours known to have a significant hypoxic component (advanced cervix carcinoma, head and neck cancers, sarcomas).

Two very simple factors were also relevant in HBO studies: the restoration of a normal haemoglobin level prior to radiotherapy, and that patients did not smoke during radiotherapy.

Anaemic patients with cervix cancer who were transfused prior to radiotherapy showed excellent local control when treated with HBO as compared to air [4, 5]. Similar observations were made for patients treated in the Dahanca trial [6] with and without misonidazole. Anaemic patients represent a target group for selection in clinical trials whose rationale is based upon oxygen effects and related phenomenona. All patients with anaemia should be transfused prior to radiotherapy to a minimum level of 12 g of haemoglobin.

Smoking habits increase the blood concentration of barboxyhaemoglobin; patients receiving radiotherapy should at least stop smoking during radiotherapy.

Use of neutron beams

Neutrons have a 2.5 higher relative biologic effectiveness (RBE) compared to X-rays. In addition, neutrons are less oxygen-dependent and should be more active in killing anoxic tumour cells than X-rays. It also seems that sublethal injuries from neutrons show a decreased repair ability compared to X-rays. Reports from more than 10 000 patients treated with neutrons are available [7]. Promising results have been published for a number of tumour sites including salivary glands, prostate, soft tissue, bone sarcomas and neck nodes in randomised trials and historical controls.

On average, 10–30% of differences in local control are observed in favour of neutrons (or combination of photon and neutron beams) as compared to photons only. Most series are rather small and need further analysis particularly in respect of normal tissue damage. However, it can be now considered that the locations and pathologies amenable to improved results with neutron therapy are well known and that the dosimetric problems and radiobiological equivalence coefficients are understood and under control.

Use of oxygen mimetic radiosensitisers and bioreductive drugs

The incidental discovery of the radiosensitising effect of nitroimidazoles was made in 1973 with metronidazole (Flagyl) used as a trichomonacide. The first clinical work with metronidazole and misonidazole started no more than a year later in Edmonton (Canada) and Mount Vernon (UK). Very rapidly, laboratory experiments demonstrated that misonidazole was a more effective radiosensitiser [8, 9] while the first clinical applications led to the identification of the risk of severe late neurological damage (peripheral neuropathies).

The mechanism of action of these drugs is twofold: they act as oxygen mimetics restoring the sensitivity of hypoxic cells and are not easily metabolised by the cells in which they accumulate. In addition, they were shown to release cytotoxins in the hypoxic cells through a bioreductive mechanism, independently of radiotherapy.

More than 10 nitroimidazole compounds have reached the step of human clinical trials. Metronidazole is now abandoned since its radiosensitising effect was too small to produce a measurable improvement. Misonidazole was examined on the largest scale ever reported for a radiosensitiser from 1974 to the mid 1980s, thousands of patients being entered in more than 30 clinical controlled trials. A significant advantage was observed in only 4 out of 12 trials in head and neck cancers, and in only 1 out of 7 trials in glioblastomas. These disappointing results were attributed to 2 main reasons: insufficient activity at the "safe" prescription doses (< 12 g/m^2) in terms of side-effects and an overestimation of the hypoxia phenomenon as a cause of radiation resistance.

However the marginal effect observed in favour of the radiosensitisers again stressed the selection of head and neck and cervix carcinomas as the best tumour sites for clinical research with new radiosensitisers. More recently, two compounds were found to be at least 5 times more effective than misonidazole in murine and human tumours while having only minor acute toxicity and no late toxicity: SR 2508 and pimonidazole. Controlled clinical trials are underway and have confirmed the initial toxicity data. Two large trials in head and neck cancer are being simultaneously carried out with SR 2508 with closely related parameters in the USA and Europe. Conversely, the only phase III trial [10] with pimonidazole was discontinued after the entry of 178 patients with cervix cancer because of a detrimental effect on local control and survival in the experimental arm. The reasons for this remain unexplained at this time. In 1989, an unexpectedly significant result was reported by Overgaard in a Danish trial (J. Overgaard, Institute of Cancer Research, Aarhus) using nimorazole as a radiosensitiser as compared to a control arm with radiotherapy alone in head and neck cancers. Nimorazole was considered up to this point as having a low radiosensitising effect. The explanation for the favorable action in human tumours may possibly be linked to additional cytotoxicity on hypoxic cells through a bioreductive effect.

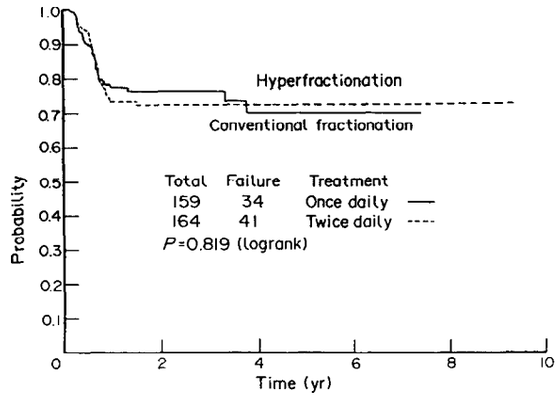
Bioreductive effects

Bioreductive activation of radiosensitising drugs in tumour cells may actually represent the best potential for improving the action of radiotherapy by the following means: a bioreductive drug (such as RSU 1069) is delivered and followed by radio-

Table 1. Tissue response when increasing the number of fractions

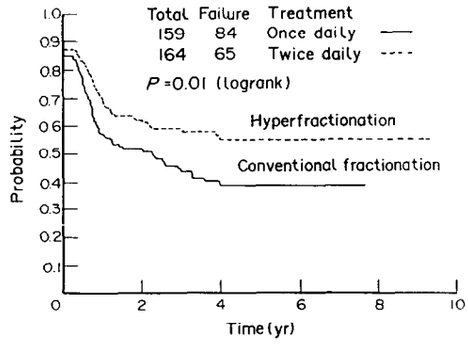
	Early responding tissues (acute)	Slowly responding tissues (late)	Tumours
Repopulation	↑	+ −	↑
Repair	+ −	↑	+ −
Reoxygenation	=	=	↑
Redistribution	↑	+ −	↑

↑ enhanced, = unlikely to be modified, + − variable.



Once daily	159	49	18	4	0	0
Twice daily	164	71	28	6	1	0

Fig. 1. Probability of avoiding late side-effects of grade 2 or 3.



Once daily	159	58	25	7	0	0
Twice daily	164	80	36	11	1	0

Fig. 2. Probability of remaining free of locoregional disease.

therapy once increased drug concentration is obtained in the tumour (simple radiosensitisation). Then, all tumour cells are made hypoxic with various manipulations of blood flow (e.g. with hydralazine) thus leaving the drug to express its cytotoxic action in the tumour only (where it is trapped). Animal experiments carried out by Adams [11] have led to a one hundred times (× 100 !) potentiation of radiation effects. Unfortunately, human experiments have shown that blood flow manipulation (with hydralazine) requires different conditions to those which apply in murine tumours and that other drugs should be used to obtain tumour anoxia. However, several trials have started with new bioreductive agents with a double action (radiosensitising and cytotoxic). These compounds may also be of interest by potentiating other chemotherapeutic agents.

OPTIMISATION OF RADIOTHERAPY VARIABLES

Up until the early 1970s, radiotherapy was delivered 5 days per week at a dose of 1.7 to 2 Gy per day (conventional fractionation). It was already clear that reducing the number of fractions and delivering a larger dose per fraction (hypofractionation) could not be used in curative treatments since it would induce a rapid increase in late complications (mostly severe fibrosis, but also bone skin and soft tissue necrosis). The demonstration of the radiobiological processes known as the four Rs of radiotherapy (repair of sublethal damage, reoxygenation, reassortment, repopulation) led to laboratory and clinical experiments delivering more than one fraction per day. The general philosophy was to try to take advantage of the ability of the normal tissues to repair sublethal damage more efficiently and more quickly than the tumour and consequently to increase the therapeutic gradient. However, delivering more than one fraction per day results in a rather complex addition of interactions (Table 1) of the various radiobiological processes. The final issue is also strongly dependent on cell kinetics: rapidly proliferating tissues (mucosa, most epithelial surfaces, skin, etc.) tolerate better these changes than slowly proliferating tissues (connective tissues, bone, lung, brain and spinal cord, etc.). It also appears that a number of tumour pathologies behave like rapidly proliferating tissues.

Two types of altered fractionation have emerged from clinical trials: hyperfractionation (HF) and accelerated fractionation (AF).

Hyperfractionation

In pure hyperfractionation, radiotherapy is given in a higher number of fractions with a smaller dose per fraction, within the same overall treatment time as in conventional fractionation (CF) regimens [12]. The division of the daily dose in two fractions (with an 8 h interval between them) allows for better recovery of normal tissues, which determines the late radiation tolerance. This gain in tolerance can be exploited by giving a 15% increment of the dose per day resulting, at the end of treatment, in a 15% higher total dose in the same overall time. Whether such an increase in dose improves locoregional control without increasing the complication rate was the question

Table 2. Comparison of parameters of different accelerated fractionation trials

Trial	Randomised	Split	Interval between courses (days)	Fraction size (Gy) (× n fr/day)	Interval between fractions (hours)	First course Gy/fr/days	Second course Gy/fr/days	Total Gy/fr/days*
22801 (EORTC)	no	yes	28	1.6 × 3	2–4	48/30/11	19.2/12/4	67.2/42/46
22811 (EORTC)	yes	yes	28	1.6 × 3	3–4	48/30/11	24/15/5	72/45/47
22851 (EORTC)	yes	yes	10/13	1.6 × 3	4	28.8/18/7	43.2/27/10	72/45/32
MGH/	no	yes	14	1.6 × 2	4	38.4/24/16	25.6/16/10	64/40/39
CHART	no	no	–	1.5 × 3	6	54/36/12	–	54/36/12

*Optimum schedules without interruption for acute toxicity, all starting on Monday.
MGH = Massachussets General Hospital, fr = fraction.

addressed by trial 22791 of the EORTC Cooperative Group of Radiotherapy.

Patients had T2 T3 N0 or N1 oropharyngeal squamous cell carcinoma under 3 cm in size (except for primaries arising from the base of the tongue). Patients were randomly allocated to CF (70 Gy in 35 fractions over 7 weeks) or HF (80.5 Gy in 70 fractions over 7 weeks). From 1980 to 1987, 356 patients were entered by 28 institutions from 7 European countries. 90% of the patients were evaluable for the final analysis.

Late damage to normal tissues was evaluated with an actuarial estimate of the freedom from grade 2 and 3 late tissue damage (Fig. 1). No difference was observed between the 2 treatment arms, which confirms the accuracy of the radiobiology prediction for normal tissue tolerance in the head and neck area. These results are of particular interest for the slower proliferating normal tissues such as bone and connective tissues.

The locoregional control was significantly higher ($P = 0.01$, log-rank) after HF compared with CF (Fig. 2). At 5 years, 56% of patients are locoregional disease free in the HF arm as compared to 38% in the CF arm. This advantage was observed only in the 217 patients with a good initial performance status (Karnovsky index 90–100%). The superiority of HF was also demonstrated in patients staged T3 N0, T3 N1 but not in T2. The Cox model confirmed that the treatment regimen was an independent significant prognostic factor for locoregional control ($P = 0.007$ log-rank).

Accelerated fractionation

Acclerated fractionation (AF) is the reduction of the overall treatment time with an increased number of fractions (multiple fractions per day). AF will reduce tumour repopulation during treatment and should improve local control providing a biological equivalent dose can be delivered to the tumour. Two types of schemes have been developed.

(1) A single continuous course of AF in which the total dose is necessarily lower than with CF. The CHART regime (54 Gy in 36 fractions and 12 days) is the best example of this scheme [13].

(2) A split course radiotherapy with the same total dose as in CF. In this case, the timing and duration of the split are essential. The split must take place at the time of maximum normal tissue repopulation and should not last more than 2 weeks in order to allow a significant reduction of the overall treatment time (e.g. 5 weeks instead of 7 weeks). EORTC protocol 22851 (72 Gy in 7 weeks versus 72 Gy in 5 weeks) is a good example of this rationale (14).

These two protocols are still on-going. Interim results of the cell kinetics study of protocol 22851 [15] in patients with advanced head and neck cancer look very promising. The median potential doubling time (T_{pot}) of these patients is of 4.2 days (range 1.5 to 15.5 days), 24% of patients having a T_{pot} lower than 2 days. Two-year local control is of 60% when T_{pot} is lower than 4 days versus 80% when higher than 4 days. Local control does not differ significantly in the two treatment arms when T_{pot} is higher than 4 days, whereas it is of 70% with AF versus 38% with CF when T_{pot} is of less than 4 days.

CONCLUSION

Radiobiology is presently offering the best insight for progress in our understanding of radiation response, leading to changes in radiotherapy parameters. These advances aim to:

- Decrease late radiation induced damage to normal tissues.
- Improve the efficacy of a given radiation dose by an optimal use of the parameters of fractionation, dose per fraction and overall treatment time.
- Modify the radiation response of the hypoxic tumour cell component by using a variety of radiosensitisers with or without blood flow modifications. This is perhaps the most promising field of investigation, although more difficult to evaluate and to monitor than fractionation changes.
- Predict radiation response from a biopsy specimen as it is already done in clinical trials: individual meaurements of tumour cell kinetics is becoming an essential component of investigations of fractionation and overall treatment time changes.

This short overview has not considered all the possible improvements of radiotherapy and has only focused on the interactions of radiobiology and radiotherapy clinical trials.

We emphasise that radiobiology is now the major source for our understanding of radiation response and has already led to significantly improved results in cancer patients by a better use of the widely available equipment producing photon and electron beams.

1. Mottram JC. Mount Vernon Hospital and Radium Institute Annual Report. Northwood, England, 1935.
2. Gray LH, Conger AO, Ebert M, Hornsey S, Scott OC. The concentration of oxygen dissolved in tissues at the time of irradiation as a factor in radiotherapy. *Br J Radiol* 1953, **26**, 638–648.
3. Lacassagne A. L'anoxi comme facteur de radiorésistance. *Acta Radiolog* 1953, (Suppl. 116), 100–107.
4. Dische S. The clinical use of hyperbaric oxygen and chemical hypoxic cell sensitizers: an overview. In: Steel GG, Adams GE, Peckham MJ, eds. *The Biological Basis of Radiotherapy*. Amsterdam, Elsevier, 1983, 225–237.
5. Dische S, Anderson PJ, Sealy R, Watson ER. Carcinoma of the cervix: anaemia, radiotherapy and hyperbaric oxygen. *Br J Radiol*, 1983, **56**, 251–255.
6. Overgaard J, Andersen AP, Jensen RH, *et al.* Misoni-dazole combined with split-course radiotherapy in the treatment of invasive carcinoma of the larynx and the pharynx: a preliminary report of the Danish Head and Neck cancer study (DAHANCA) protocol 2. *Acta Otolaryngol*, 1982 (Suppl. 386), 215–220.
7. Maor MH. Fast neutron therapy: problems and promise. In: *Innovations in Radiation Oncology*. HR Withers, LJ Peters, eds. Berlin, Springer, 1988, 279–287.
8. Asquith JC, Watts ME, Patel K, Smithen CE, Adams GE. Electron affinic sensitization. V. Radiosensitization of hypoxic bacteria and mammalian cells *in vitro* by some nitroimidazoles and nitropyrazoles. *Radiat Res* 1974, **60**, 108–118.
9. Dische S, Gray AJ, Zanelli GD. Clinical testing of the radiosensitizer Ro 07-0582. II. Radiosensitization of normal and hypoxic skin. *Clin Radiol* 1976, **27**, 159–166.
10. Medical Research Council: Trial of misonidazole in carcinoma of the uterine cervix. A report from the MRC Working Party on Misonidazole for cancer of the uterine cervix. *Br J Radiol* 1984, **57**, 491–494.
11. Adams GE, Dische S. Care with radiosensitizers. *Br J Radiol* 1979, **52**, 920–921.
12. Withers RH, Horiot JC. Hyperfractionation. In: Withers HR, Peters LJ, eds. *Innovations in Radiation Oncology*. Berlin, Springer, 1987, 223–230.
13. Dische S, Saunders MI. Continuous, hyperfractionated, accelerated fractionation (CHART): an interim report upon late morbidity. *Radiother Oncol* 1989, **16**, 67–74.
14. EORTC (European Organization for Research on Treatment of Cancer) Cooperative Group of Radiotherapy, protocol 22851: a phase III study of acclerated fractionation in the radiotherapy of advanced head and neck carcinoma. EORTC Data Centre, Brussels, Belgium, November 1985.
15. Begg AC, Hofland I, Moonen L, *et al.* The predictive value of cell kinetic measurements in an European trial of accelerated fractionation in advanced head and neck tumours: an interim report. *Int J Radiat Oncol Biol Phys.*