Oral Oncol, Eur J Cancer, Vol. 29B, No. 2, pp. 113–117, 1993
Printed In Great Britain

0964–1955/93 $6.00 + 0.00

Papers

Histopathological Findings After Preoperative Chemo-radiotherapy in Oral and Oropharyngeal Carcinoma. A Study of 28 Resected Specimens

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Serial sections of whole surgical specimens from 28 patients with locally advanced oral carcinoma were examined to assess the histopathological effects of a preoperative intravenous (i.v.) chemotherapy and radiotherapy protocol. In 15 cases (54%), no residual neoplastic tissue was found: in 9, the tumour area showed a dense hyalinised connective tissue, whereas in the other 6 cases foci of keratotic material were surrounded by a foreign body-type reaction. In 13 patients (46%), residual neoplastic tissue was observed: small fields of tumour were present in 6 cases, whereas tumour was widely persistent in 7. Lymph node metastases were observed in 9 cases (32%), with extracapsular spreading in 6 (21%). In 93% of cases, pathological staging on the specimen was more favourable than the clinical preoperative staging. 23 patients (82%) are alive without evidence of disease 30–55 months after surgery; 10 (43%) did not show any residual neoplastic tissue in the specimen; 10 patients (43%) showed persistence of tumour, but in 5 (21%) only small fields of tumour were observed. 4 patients (14%) died due to their tumour. The pathological data show that this protocol is successful in the treatment of advanced oral carcinoma; furthermore, they provide reliable information about the response to therapy and the subsequent course of the disease.
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INTRODUCTION

REMARKABLE CLINICAL improvements have been achieved with more active drugs, especially cisplatinum, in the management of advanced head and neck squamous cell carcinomas. Surgery and radiotherapy are still the treatments of choice in most cases. Even so, there have been several reports of the use of both single-agent and combination chemotherapy not only palliatively, but also as neoadjuvant therapy in locally advanced cases [1–4]. A significant reduction in the number of deaths [5] and longer survival [6], however, have not been achieved. Previous trials have shown that chemotherapy, radiotherapy and surgery can be employed to extend the disease-free interval in advanced oral cancer [7–9]. The histopathological effects of combination protocols, however, have received little attention and most studies have been concerned with chemotherapy alone [10–12]. This paper describes such effects in 28 patients with advanced squamous cell carcinoma (SCC) of the oral cavity and oropharynx who received a new protocol consisting of an initial course of neoadjuvant chemotherapy, followed by a second course with radiotherapy and then surgery and postoperative radiotherapy. An assessment is made of the histological parameters indicative of the response to therapy and the subsequent course and survival.

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Received 4 Sep. 1992; revised manuscript accepted 20 Oct. 1992; revised manuscript received 6 Nov. 1992.

PATIENTS AND METHODS

28 patients with advanced SCC of the oral cavity and oropharynx (Table 1) were included in this study for treatment according to the following eligibility criteria: (1) no previous treatment for oral cancer and/or for tumours in other sites; (2) a performance status (KPS) >50 compatible with the employment of chemotherapy; (3) presence of biopsy-proven stage III–IV carcinoma [13]; (4) absence of clinically observed visceral metastases; (5) histological diagnosis of SCC.

Treatment schedule

After informed consent was obtained, all patients received the same combined treatment: 100 mg/m^2 cis-platinum bolus intravenously (i.v.) on the first day and 1000 mg/m^2 5-fluorouracil (5-FU) in a 24 h i.v. infusion from the second to

Table 1. Clinical details of 28 patients with oral and oropharyngeal squamous cell carcinomas treated with preoperative chemo-radiotherapy

Case	Sex	Age	Site	TNM	Grading	Surgical treatment
1	M	58	FM	T3N1M0	2	Commando + FND
2	M	46	FM	T4N3M0	2	PT + FND
3	M	61	FM	T3N3M0	2	PT + FND
4	M	24	T	T2N2M0	2	Cryosurgery + FND
5	M	69	FM	T4N3M0	2	PT + SOD
6	M	45	SP	T4N1M0	2	Local excision
7	M	46	RT	T2N1M0	2	PT + FND
8	M	63	Lip	T2N1M0	1	PT + bilateral SOD
9	M	66	T	T2N1M0	2	Local excision
10	F	59	AM	T4N0M0	3	Emimaxillectomy + FND
11	M	30	RT	T4N3M0	3	Commando + FND
12	M	46	T	T3N1M0	2	PT + FND
13	M	49	RT	T4N3M0	2	Commando + FND
14	M	69	T	T2N1M0	2	Local excision
15	M	57	FM	T2N1M0	2	PT + SOD
16	M	72	FM	T4N1M0	2	Commando + bilateral SOD
17	M	60	CH	T4N3M0	2	Commando + FND
18	F	44	TR	T3N2M0	2	PT + FND
19	M	56	FM	T4N3M0	3	Commando + FND
20	M	57	T	T3N2M0	3	PT + FND
21	M	63	FM	T3N1M0	2	PT + FND + SOD
22	M	51	AM	T4N0M0	2	Emimaxillectomy
23	M	71	RT	T3N1M0	2	PT + FND
24	M	69	RT	T3N2M0	2	PT + FND
25	M	53	FM	T3N1M0	2	PT + FND + SOD
26	M	79	FM	T4N3M0	3	PT + FND
27	M	61	FM	T4N2M0	2	PT + FND + SOD
28	M	65	FM	T3N1M0	2	PT + FND

AM = alveolar mucosa; CH = cheek; FM = floor of the mouth; RT = retromolar trigone; SP = soft palate; T = tongue; TR = tonsillar region; FND = functional neck dissection; PT = pull-through operation; SOD = supra-omohyoid dissection.

the fifth day, followed by the same course 15 days after the first, plus 20 Gy of transcutaneous radiotherapy in 10 daily fractions over 2 weeks. Radical surgery was performed 15–20 days after completion of this second course. An additional 40 Gy in 20 daily fractions over 2 weeks were given 20–40 days after surgery.

Clinical evaluation

Preoperative responses were evaluated as follows: (1) complete response; (2) partial response > 50%. Tumour size was measured prior to therapy and then just before surgery.

Pathology

All surgical specimens were placed in a transport medium (RPMI), immediately measured and fixed for 1 day in 10% formalin. The tumour (or the tumour area in cases with complete response) was macroscopically evaluated and measured. Each specimen was then cut serially into slices (maximum 0.5 cm thick) and fixed for another 2 days. A routine procedure was employed to embed and section the slices (10–25 blocks for each case). Sections were stained with haematoxylin–eosin (H&E). Positivity for vimentin was investigated in some cases with ABC immunoperoxidase [14]. Each lymph node from the neck dissection specimen was measured and processed as above.

The pTNM and extent of the residual neoplastic tissue (R1 or R2) were indicated in each case [13]. The histological characteristics of the tumours and regressive lesions were classified into four patterns (Table 2): (i) complete absence of tumour; (ii) absence of neoplastic cells with presence of keratotic material and foreign-body type reaction; (iii) small fields of residual neoplastic tissue; (iv) extensive persistence of tumour.

RESULTS

Tumour specimens (Table 2)

In 21/28 (75%) cases, the area of the primary tumour showed roughness of the surface, firm consistency and sclerotic appearance. In 15 of these cases, no residual neoplastic tissue was found, whereas in 6 cases small tumour foci were observed histologically. In the remaining 7 cases, the gross appearance was consistent with a classical SCC of the oral cavity: usually a plaque-like infiltrating or slightly vegetating ulcerated lesion was observed.

In 15 cases macroscopically negative for tumour (54%), the oral mucosa was lined by normal or slightly hyperplastic squamous epithelium, and the stroma showed an extensive, diffuse sclerosis with slight infiltration by lymphocytes and plasma cells (Fig. 1). Congestion of blood vessels was occasionally observed. The very few stromal cells always displayed atypical, bizarre nuclei (Fig. 2). Their mesenchymal nature was confirmed by cytoplasmic positivity for vimentin. In 6/15 (40%) cases, residual tumour was also absent, but a varying number of foci with keratinised material surrounded by multinucleated giant cells of foreign-body type were scattered in

Table 2. Clinicopathological findings in 28 cases of oral and oropharyngeal squamous cell carcinoma treated with preoperative chemo-radiotherapy

Case	Site	TNM	pTNM	R	Extracapsular spreading	Pathological pattern	Outcome
1	FM	T3N1M0	pT0N0	0	−	II	NED
2	FM	T4N3M0	pT0N1	0	−	II	NED
3	FM	T3N3M0	pT1N0	1	−	III	NED
4	T	T2N2M0	pT0N2b	0	+	I	DOD
5	FM	T4N3M0	pT0N0	0	−	II	NED
6	SP	T4N1M0	pT0Nx	0	/	II	NED
7	RT	T2N1M0	pT0N0	0	−	I	NED
8	Lip	T2N1M0	pT1N0	2	−	IV	NED
9	T	T2N1M0	pT0Nx	0	/	I	NED
10	AM	T4N0M0	pT2N0	2	−	IV	DOD
11	RT	T4N3M0	pT4N1	1	+	III	DOD
12	T	T3N1M0	pT0N0	0	−	II	NED
13	RT	T4N3M0	pT0N0	0	−	I	NED
14	T	T2N1M0	pT0Nx	0	/	I	NED
15	FM	T2N1M0	pT0N0	0	−	I	NED
16	FM	T4N1M0	pT4N0	1	−	III	NED
17	CH	T4N3M0	pT2N2b	2	+	IV	NED
18	TR	T3N2M0	pT1N0	1	−	III	NED
19	FM	T4N3M0	pT0N0	0	−	I	NED
20	T	T3N2M0	pT2N2b	2	+	IV	DOD
21	FM	T3N1M0	pT1N0	1	−	III	NED
22	AM	T4N0M0	pT3Nx	2	/	IV	NED
23	RT	T3N1M0	pT0N1	0	−	I	NED
24	RT	T3N2M0	pT1N0	2	−	IV	NED
25	FM	T3N1M0	pT1N1	1	−	III	NED
26	FM	T4N3M0	pT0N2b	0	+	II	DWD
27	FM	T4N2M0	pT2N2b	2	+	IV	NED
28	FM	T3N1M0	pT0N1	0	−	I	NED

AM = alveolar mucosa; CH = cheek; FM = floor of the mouth; T = tongue; RT = retromolar trigone; SP = soft palate; TR = tonsillar region; NED = no evidence of disease; DOD = dead of disease; DWD = dead without disease; R0 = no tumour residue; R1 = microscopical tumour residue; R2 = macroscopical tumour residue; + = present; − = absent; pathological patterns: I = complete absence of tumour; II = absence of tumour with foreign-body type reaction; III = small tumour fields; IV = extensive persistence of tumour; / = no lymph node dissection.

the specimen (Fig. 3). In these cases, inflammatory reaction was present, though not usually intense.

In 13/28 patients (46%), residual neoplastic tissue was observed; in 6, macroscopically negative, small neoplastic epithelial structures were invading hyalinised stromal areas (Fig. 4); a foreign-body type reaction was noted in one case. In the other 7 cases, massive tumour infiltration was still evident on gross inspection: these cases showed varying degrees of sclerosis and inflammatory reaction, and inconspicuous necrotic foci; in 2 cases, neoplastic tissue involved the resection margins. In all cases with residual tumour, no substantial differences in histological grading were observed between the surgical specimen and the pre-operative biopsy.

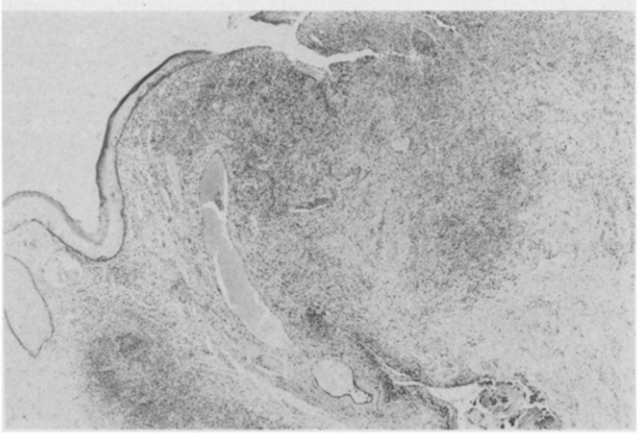

Fig. 1. Complete response after preoperative chemo-radiotherapy: the neoplastic area is occupied by a dense connective tissue with a conspicuous inflammatory reaction. Haematoxylin-eosin (35 ×).

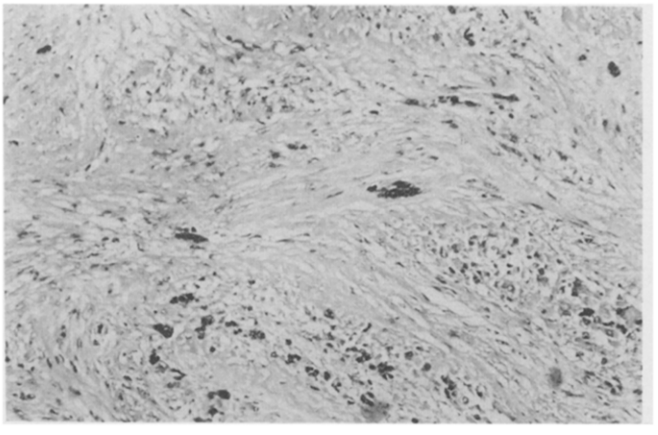

Fig. 2. As Fig. 1. Stromal cells with regressive changes. Haematoxylin-eosin (200 ×).

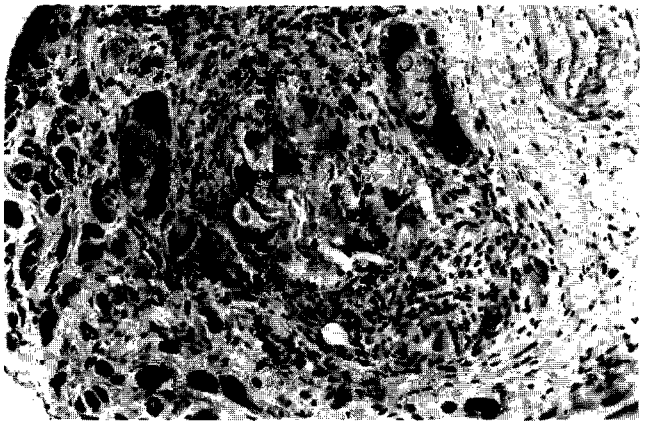

Fig. 3. As Fig. 1. Absence of neoplastic cells and residual keratotic material with foreign body-type reaction. Haematoxylin–eosin (200 ×).

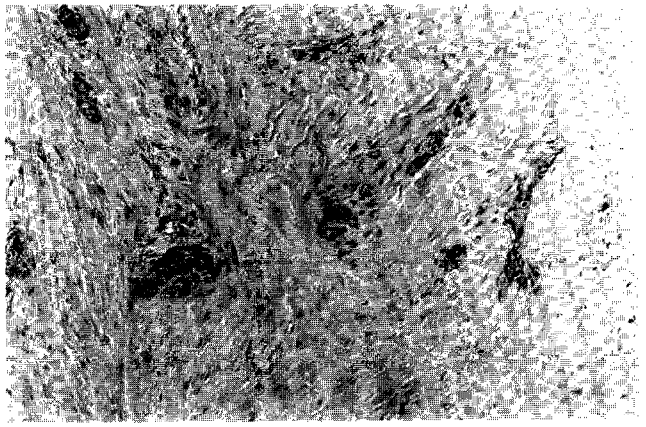

Fig. 4. Partial response after preoperative chemo-radiotherapy: foci of neoplastic tissue in a dense hyalinised connective tissue. Haematoxylin–eosin (90 ×).

Lymph node metastases were found in 9 cases, accompanied in 6 by extracapsular spreading.

Comparison between clinical and pathologic staging

In 26/28 (93%) cases, the pathological tumour stage had improved compared with the clinical stage prior to treatment (Table 2); the most dramatic differences were 6 from T4 to pT0 and 4 from T3 to pT0; in 2 cases (patients 11, 16), there was no change (T4 and pT4). In 17/24 (71%) cases, there was an improvement in lymph node staging; in another case, clinical NO was confirmed histologically; in 4 cases, no lymph node dissection was carried out. In 5 cases, absence of neoplastic tissue was accompanied by the presence of lymph node metastases.

Clinical follow-up

23/28 (82%) patients are alive with no evidence of disease 30–55 months after surgery (Table 2). In 10 cases, no neoplastic tissue was observed in the site of the primary tumour and lymph nodes. In another 10 patients, persistence of tumour was found, focally in 5 patients and extensively in the other 5. In the other 3 patients, no tumour was observed in the specimen, but lymph node metastases without extracapsular spreading were found. 4 patients died due to their tumour: in 3, residual neoplastic infiltration was observed in the specimen, this was extensive in 2 cases and focal in one; in 2 cases lymph node metastases with extracapsular spreading were observed. 1 patient died from a vascular accident a short time after completing the treatment schedule.

DISCUSSION

Pathological assessment of whole surgical specimens serves to determine the effect of medical therapy. Chemotherapy and radiotherapy give rise to regressive changes (sclerosis, necrosis, haemorrhage, atypia of stromal cells) that may mask the presence of neoplastic tissue, which can also be missed on partial examination if it occurs as microscopic fields in a prominent sclerotic reaction. Tumour residues were absent in the whole specimen in 54% of our series. The lower percentages reported by Micheau *et al.* (34%) [11] and Sulfaro *et al.* (16.1%) [12] may be due to their use of intrarterial chemotherapy only, as opposed to our simultaneous i.v. chemotherapy and radiotherapy with their possibly potentiating effect. A high rate of CR (62%) with a similar protocol was reported by Earle *et al.* [9], though no detailed histological findings were presented.

Distinguishing between complete absence of tumour and R1 or R2 may have an impact in predicting the outcome: only 7% of patients with complete absence of neoplastic tissue on the specimen died due to their tumour, whereas 23% of those with persistence after surgery died within 2 years. The outcome was equally poor in patients with minimal or extensive tumour residue: 17% vs. 18%. Other pathological findings associated with treatment failure were lymph node metastases and extracapsular spreading: 30% of patients with metastases died due to their tumour, and 50% with extracapsular spreading. The latter are expected: advanced nodal disease is an indication that a good survival will not be achieved with cisplatinum combination chemotherapy [15].

Regressive changes were observed in all specimens negative for tumour, and in cases with minimal as opposed to extensive persistent tumour. These changes seem to correspond with a greater response, whereas their absence may reflect more aggressive neoplasia. The most frequently observed changes were stromal sclerosis (68% of cases) and keratinised material with foreign-body type cell reaction (32%). The latter may be the result of extensive destruction of neoplastic cells rather than therapy-induced tumour differentiation, as suggested by Shapshay *et al.* [10]. In some cases, this finding was not associated with the presence of neoplastic cells in the whole specimen: in agreement with Micheau *et al.* [11], we considered this as a complete absence of tumour, being probably a direct effect of the therapy. The regressive lymph node changes noted by Earle *et al.* [9], however, were absent in our series.

As expected, patients with complete absence of tumour after surgery had a better outcome than those with persistence of neoplastic tissue, though the difference was not significant, because all patients received 40 Gy of postoperative transcutaneous radiotherapy resulting in further destruction of residual neoplastic tissue.

23 of our patients (82%) are currently alive and well without evidence of tumour 30–55 months after completing the therapy. A slightly better result has been reported by Sulfaro *et al.* [12], though comparison is difficult because they used intra-arterial chemotherapy. The difference may be due to the more

advanced stages of our cases (85.7% of patients with clinically positive lymph nodes).

The impact of some histological parameters on the prognosis of head and neck SCC has been demonstrated [16] and is rendered evident on examination of surgical specimens obtained after therapy. Our results and previous reports indicate that evaluation of the pathological findings after chemotherapy and radiotherapy is fundamental in management of oral cavity SCC. Histological examination of the whole surgical specimen provides reliable information about the response to therapy and the subsequent course and is recommended. The combined pathological and clinical data suggest that a combination of chemotherapy and radiotherapy before surgery is an important alternative to conventional protocols in head and neck SCC.

1. Mead GM, Jacobs C. Changing role of chemotherapy in treatment of head and neck cancer. *Am J Med* 1982, **73**, 582–595.
2. Rooney M, Kish J, Jacobs J, *et al.* Improved complete response rate and survival in advanced head and neck cancer after three course induction therapy with 120-hour 5-FU infusion and cisplatin. *Cancer* 1985, **55**, 1123–1128.
3. Vokes EE, Panje WR, Mick R, *et al.* A randomized study comparing two regimens of neoadjuvant and adjuvant chemotherapy in multimodal therapy for locally advanced head and neck cancer. *Cancer* 1990, **66**, 206–213.
4. Richard JM, Kramar A, Molinari R, *et al.* Randomised EORTC head and neck cooperative group trial of preoperative intra-arterial chemotherapy in oral cavity and oropharynx carcinoma. *Eur J Cancer* 1991, **27**, 821–827.
5. Stell PM, Rawson NS. Adjuvant chemotherapy in head and neck cancer. *Br J Cancer* 1990, **61**, 779–787.
6. Jacobs JR, Fu KK, Lowry LD, Scotte Doggett RL, Pajak TF, Al-Sarraf M. 5-year results of cisplatin and fluorouracil infusion in head and neck cancer. *Arch Otolaryngol Head Neck Surg* 1991, **117**, 288–291.
7. Marcial VA, Pajak TF, Mohiuddin M, *et al.* Concomitant cisplatin chemotherapy and radiotherapy in advanced mucosal squamous cell carcinoma of the head and neck. Long-term results of the Radiation Therapy Oncology Group study 81–17. *Cancer* 1990, **66**, 1861–1868.
8. Adelstein DJ, Sharan VM, Earle AS, *et al.* Simultaneous versus sequential combined technique therapy for squamous cell head and neck cancer. *Cancer* 1990, **65**, 1685–1691.
9. Earle AS, Adelstein DJ, Vlastou C, Sharan VM, Indresano AT. Treatment of oral squamous cell carcinoma with simultaneous chemotherapy and radiation: results and surgical implications. *J Oral Maxillofac Surg* 1990, **48**, 367–372.
10. Shapshay SM, Hong WK, Incze JS, *et al.* Histopathologic findings after cis-platinum bleomycin therapy in advanced previously untreated head and neck carcinoma. *Am J Surg* 1978, **136**, 534–538.
11. Micheau C, Richard J. Actions tissulaires de la chimiothérapie intra-artérielle dans les cancers pelvi-linguaux et leurs métastases ganglionnaires: A propos de 47 cas. *Ann Otolaryngol* 1975, **92**, 499–508.
12. Sulfaro S, Frustaci S, Volpe R, *et al.* A pathologic assessment of tumor residue and stromal changes after intrarterial chemotherapy for head and neck carcinomas. A study on serial sections of the whole surgical specimen. *Cancer* 1989, **64**, 994–1001.
13. UICC: TNM *Classification of Malignant Tumors*. 4th ed., Berlin, Springer 1987.
14. Hsu SM, Rami L, Fanger H. Use of avidin biotin peroxidase complex (ABC) in immunoperoxidase techniques: a comparison between ABC and unlabeled antibody (PAP) procedures. *J Histochem Cytochem* 1981, **29**, 577–580.
15. Al-Kourainy K, Kish J, Ensley J, *et al.* Achievement of superior survival for histologically negative versus histologically positive clinically complete responders to cisplatin combination in patients with locally advanced head and neck cancer. *Cancer* 1987, **59**, 233–238.
16. Breuninger H, Black B, Rassner G. Microstaging of squamous cell carcinomas. *Am J Clin Pathol* 1990, **94**, 624–627.

Acknowledgements—We thank Dr John Iliffe for revising the manuscript, Dr Vincenzo Carbone for his help in collecting the clinical data and Nino Ferraro for technical assistance. This work was partially supported (40%) by grant from MURST, Italy.