gression, engendering a clone of drug-resistant cells from an original testicular infiltrate that would otherwise be susceptible to prolonged treatment with methotrexate and mercaptopurine. Finally, cyclophosphamide-induced destruction of cells within the seminiferous tubules (that is, beyond the blood-testis barrier)[11 12] may change the microenvironment to allow proliferation of leukaemic cells in that site.

The relation of testicular disease to subsequent relapse of bone-marrow and meningeal disease is considered elsewhere. Meanwhile, the fact that many of the boys with isolated testicular disease had already had bone-marrow relapses indicates the need for further chemotherapy in conjunction with local treatment to the testes and also emphasises the importance of prompt diagnosis and prevention of testicular disease. Exclusion of cyclophosphamide from routine chemotherapy seems to be important, and a full reappraisal of the value of "extra drugs" in maintenance treatment for "good-risk" patients is needed. Routine biopsy of the testes and prophylactic testicular irradiation[13] must be considered.

O B Eden is in receipt of a grant from the Leukaemia Research Fund.

References

1 Peto, R, *et al*, *British Journal of Cancer*, 1977, **34**, 585.
2 Peto, R, *et al*, *British Journal of Cancer*, 1977, **35**, 1.
3 West, R J, *et al*, *British Medical Journal*, 1972, **3**, 311.
4 Nies, B A, *et al*, *Blood*, 1965, **26**, 133.
5 Givler, L, *Cancer (Philadelphia)*, 1969, **23**, 1290.
6 Stoffel, T J, Nesbit, M E, and Levitt, S H, *Cancer (Philadelphia)*, 1975, **35**, 1203.
7 Schaison, G, *et al*, *Presse Médicale*. In press.
8 St Jude Children's Hospital, Memphis. Unpublished results.
9 Henk, J M, personal communication.
10 Berry, C L, *et al*, *Lancet*, 1972, **2**, 1033.
11 Kumar, R, *et al*, *Lancet*, 1972, **1**, 1212.
12 Dym, M, and Fawcett, D W, *Biology of Reproduction*, 1970, **3**, 308.
13 Nesbit, M, *et al*, *Proceedings of the American Association for Cancer Research*, 1977, **18**, 317.

(*Accepted 1 December 1977*)

Treatment of malignant ascitic and pleural effusions with Corynebacterium parvum

H E WEBB, S W OATEN, C P PIKE

British Medical Journal, 1978, **1**, 338-340

Summary and conclusions

Six patients with malignant effusions, five from adenocarcinomas and one from a melanoma, were treated by intrapleural or intraperitoneal Corynebacterium parvum. In each case there was a definite reduction in the effusions with a significant decrease in the number of malignant cells; in most cases the effusions stopped completely.

Although none of the patients lived for more than a year after treatment, they were undoubtedly more comfortable, as they no longer required frequent paracentesis. In some cases the patients lived for longer than originally expected in a state in which the quality of life was acceptable.

Introduction

Corynebacterium parvum is now recognised as a possible anticancer agent.[1] A preliminary study on patients with various advanced malignant conditions suggested that *C parvum* might be beneficial in the routine treatment of patients with malignant ascites. We report here the results of treatment with *C parvum* in six patients with malignant effusions.

The Rayne Institute, St Thomas's Hospital, London SE1 7EH
H E WEBB, DM, FRCP, consultant physician and senior lecturer in medicine
S W OATEN, BSC, PHD, lecturer
C P PIKE, MB, CHB, lecturer in surgical pathology (now consultant cytologist, St Luke's Hospital, Guildford, Surrey)

Patients and methods

Corynebacterium parvum strain CN 6134 (batch EZ 174) was obtained from Wellcome Research Laboratories. Preparations consisted of 7 mg dry weight per ml of formalin-killed organisms suspended in 0·01% thiomersalate. The dose, which was given intraperitoneally, intrapleurally, and occasionally intramuscularly, ranged from 0·5 ml to 2 ml.

Treatment—All patients and their relatives gave informed consent. When it was considered necessary for the patient's comfort a formal paracentesis was undertaken. Nevertheless, we always left enough fluid behind to mix the *C parvum* adequately with the effusion. Samples were taken and further injections were given using a 20-ml syringe and a three-way tap with little or no discomfort to the patient. As treatment progressed the fluid became loculated and only small amounts could be withdrawn from any one site. But it was still possible to give the *C parvum* into areas of loculated fluid. Reactions to the *C parvum* were nausea, vomiting, fever, and some pain at the site of the injection. These were controlled by analgesics and antiemetics. When there was not enough fluid to dilute the *C parvum* it was given by deep intramuscular injection of 1 ml into the buttocks. No discomfort or inflammation was observed with these injections.

Cellular studies on malignant fluid—Cytospin preparations and direct smears of the fluid were made immediately before the first intraperitoneal inoculation of *C parvum* and then at least once a week until the fluid disappeared. Careful cell counts were done and particular attention was given to malignant cells and the type of cell reaction present before and after the *C parvum* treatment.

Case reports

CASE 1

A 54-year-old woman underwent a subtotal hysterectomy and bilateral salpingo-oophorectomy for a uterine papillary adenocarcinoma. Over the next two years she was treated with chlorambucil, cyclophosphamide, and thiotepa for recurrent ascites, and when she started *C parvum* treatment she had had about 50 litres of malignant ascitic fluid removed. She was given 2 ml of intraperitoneal *C parvum* every week for four weeks. Five weeks after the start of treatment no fluid could be detected in the abdomen and it had become concave.

She was very thin and cachectic. Her morale and appetite improved and she put on weight. She needed the occasional transfusion of packed cells to maintain her haemoglobin concentration. Her ESR, which had been 133 mm in first h, fell to 5 mm in first h. The masses in her abdomen were much less easily palpable and less hard. She was discharged to convalescence, though we had thought that she would never leave hospital again. She was readmitted to hospital 11 months later in an emaciated condition; she was alert though occasionally incoherent. Her abdomen had become very distended with hard irregular nobbly masses in her lower abdomen. As it was impossible to drain her abdomen by normal paracentesis because of loculation, 2 ml of *C parvum* was given into one of the loculi. Again many adenocarcinoma cells were found. She had a further 2 ml 11 days later. The malignant cells disappeared. The abdominal distension receded and fluid became difficult to obtain. Another 2 ml *C parvum* was given one week later. The fluid disappeared and did not return. She died peacefully one month later.

At necropsy the tumour was confined to the abdominal cavity. The visceral and parietal layers of the peritoneum and both leaves of the diaphragm were extensively infiltrated by tumour deposits though none were found in the pleural surfaces. The peritoneal cavity was completely obliterated by dense fibrous adhesions and tumour tissue, which bound the loops of intestine and the other abdominal viscera to one another and to the parietal peritoneum. No metastases were found except in the para-aortic lymph nodes. No fluid was present. Histological examination showed thick layers of fibrous tissue covering the tumour and a moderate to heavy infiltration of polymorphs, macrophages, and plasma cells between the tumour cells. In the spleen there were more plasma cells than normal in the white pulp and more haemosiderin in the cells lining the sinusoids. In the bone marrow haemopoiesis appeared normal but there was an excess of haemosiderin pigment in the histiocytes.

CASE 2

A 58-year-old woman had an inoperable adenocarcinoma of the ovary with ascites; she was treated with chlorambucil. She was admitted for terminal care with gross ascites and hard masses all over her abdomen. After a formal paracentesis 2 ml *C parvum* was given intraperitoneally followed by a second dose two weeks later. After the second dose no further fluid could be removed, but she remained very ill. No more *C parvum* was given as the fluid had disappeared, and she died two months after the first dose of *C parvum*. Necropsy was not performed.

CASE 3

A 71-year-old woman had an inoperable, poorly differentiated, non-mucin-secreting ovarian adenocarcinoma and was treated with thiotepa and chlorambucil. *C parvum* treatment 2 ml intraperitoneally was started for gross ascites. A further 2 ml was given one week later. The next week no fluid could be withdrawn from the peritoneal cavity so 1 ml *C parvum* was given intramuscularly followed by 1 ml a week later. The patient was discharged but had to be readmitted after a week with severe pitting oedema of the ankles. She died two days later.

At necropsy the patient appeared superficially to have two adenocarcinomas—one of the left breast and a second of the ovary spreading to the peritoneal cavity. The abdominal contents were matted together by masses of tumour growth, which also encased the gastrointestinal tract from the second part of the duodenum to the rectosigmoid colon. Tumour was found above the diaphragm, and it had invaded the liver but no other parenchymatous organs. No fluid was found except 200 ml in one thick-walled cyst of tumour tissue. The histological examination showed the same sort of fibrous tissue reaction as described in case 1. The spleen was not examined. The invasive adenocarcinoma of the pelvis was histologically indistinguishable from the adenocarcinoma of the breast, so it was not clear which was the primary site. Possibly this patient had two primary adenocarcinomas.

CASE 4

A 38-year-old woman had widespread inoperable carcinomatosis, which was thought to have arisen from the right ovary. Histological examination showed that it was a non-secreting adenocarcinoma. Seven days after laparotomy, when the wound had healed, ascitic fluid was not obtainable from the abdomen so 1 ml *C parvum* was given intramuscularly into the buttock. One week later a further 1 ml was given. She was discharged three weeks after operation. Twenty days later she was admitted with bilateral effusions—the right very large, and the left very small. The abdomen was soft and not distended. Her right chest was drained of two litres of fluid and she went home. She was readmitted six days later with a large right pleural effusion and a peritoneal effusion. She was given *C parvum* 1·5 ml intraperitoneally and 0·5 ml intrapleurally on the right side. Eight days later 2 ml *C parvum* was given intraperitoneally followed after six days by 0·5 ml intrapleurally. Three weeks after her first intraperitoneal injection there was no fluid in her abdomen, the right pleural effusion was improving, and the small left pleural effusion remained static. She was then given 1 ml *C parvum* intramuscularly into her buttock. One month after starting treatment there remained very small effusions at her right and left bases. She had one further dose of *C parvum* 1 ml intramuscularly, and five weeks after admission went home feeling much better. During the next month her condition deteriorated; she vomited, though her bowels were working normally and there was no evidence of obstruction. She was readmitted to hospital, where she died a few days later with no increase of fluid at any site.

At necropsy the right pleural cavity was obliterated by fibrous adhesions and two small loculated areas of fluid were found containing 100 ml and 50 ml of clear yellow fluid. These areas were surrounded by thickened pleura containing tumour nodules. The left pleural cavity, untreated by *C parvum*, had a litre of blood-stained effusion with tumour nodules in the pleura but no adhesions. The fluid had 80 clumps of malignant cells per cytospin field. The lung showed evidence of pneumonia. An embolus was present in the left pulmonary artery. There were many tumour deposits throughout the abdominal cavity and vascular fibrous adhesions between portions of the gut and the abdominal wall. There were some small loculi of clear yellow ascitic fluid. Deposits were not found in the parenchymatous organs but the lymph nodes were invaded by tumour tissue.

Histologically the serosa of the intestines contained many tumour deposits in a fibrous tissue stroma, which was infiltrated by some histiocytes and plasma cells. The spleen showed a considerable increase in plasma cells, which were diffusely infiltrating the pulp cords. These and the sinusoids also contained increased numbers of polymorphs. The appearances were compatible with an immunological response. In the liver also the numbers of mononuclear cells and polymorphs in the sinusoids and portal tracts were increased. In the lung, tumour infiltrated the pleura, which was covered by a thick layer of fibrous tissue containing a few foci of tumour cells. An area of fibrinous reaction and a focus of tumour showing plasma cell infiltration was also seen.

CASE 5

A 61-year-old woman had an inoperable non-secreting adenocarcinoma arising from either the uterus or the right ovary. She was given tamoxifen 20 mg twice a day and gestronol hexanoate 200 mg intramuscularly, neither of which relieved her gross ascites. She was then given 1 ml *C parvum* intraperitoneally, and the ascites significantly resolved over the next week, when she died suddenly of a large embolus into the left pulmonary artery, which had come from an extensive deep vein thrombosis in the right leg.

At necropsy the contents of the abdomen were matted together by necrotic tumour and fibrinous tissue. Only 200 ml of ascitic fluid was present. Although tumour deposits surrounded the parenchymatous organs, there were no secondary deposits within them. There was no evidence of distant metastasis. Histological examination showed a marked fibrinous exudate, and much of the tumour on the parietal peritoneum appeared necrotic. There was no significant inflammatory cell infiltrate. In the spleen the red pulp was infiltrated with plasma cells, polymorphs, monocytes, and plasmacytoid cells.

CASE 6

A 54-year-old woman had a malignant melanoma of her right heel, which was treated by a wide incision and skin graft. She was treated then with endolymphatic phosphorus-32. Metastatic deposits that developed in her right groin were treated by a block dissection. Seven months after the original operation she developed a large left pleural effusion full of malignant melanoma cells. This was treated with intravenous dimethyl-triazeno-imidazole-carboxamide 1200 mg. She

continued to have repeated aspirations from her chest every few days with no beneficial effect so 0·5 ml *C parvum* was given intrapleurally followed by a further 0·5 ml *C parvum* three days later. The patient showed little reaction to this dose. One week later a further 1 ml of *C parvum* was given intrapleurally through an intrathoracic tube draining the effusion. Having cleared the intrathoracic tube with 10 ml of saline and then mixed the 1 ml *C parvum* in 5 ml of saline, this amount was injected followed by a further 10 ml of saline up the tube to make sure that the whole dose went into the pleural cavity. The drain was kept in for three days afterwards so that the fluid could continue to be removed. She developed severe pain in her chest. Sixteen days after the start of *C parvum* treatment the pleural fluid showed no melanoma cells but there had not been the usual degree of polymorph reaction so 2 ml *C parvum* was given intrapleurally. After this inoculation she had a higher temperature of just over 38·9 C for two days. It then subsided gradually and was normal five days later. Clearly the fluid was not reforming, although the polymorph response did not increase. The chest radiograph still showed fluid but it had not increased at all. She was discharged home feeling very much better with the radiographical appearances much improved. She was readmitted and died six weeks after starting *C parvum* treatment. Fluid had not recurred.

At necropsy the right pleural cavity contained 30 ml of clear fluid. There were scattered deposits of tumour tissue throughout the right lung. The left lung weighed 3500 g, adhered to the chest wall and diaphragm, and was encased with solid tumour tissue. No fluid was found in the left pleural cavity. Tumour was present in the parietal pleura on the anterior aspect of the chest wall. Many lymph nodes throughout the body showed evidence of tumour tissue. Histologically the spleen showed mild hyperplasia. The Malpighian corpuscles, though normal in size, contained many plasma cells scattered thinly in the red pulp. There was widespread evidence of tumour deposits.

Cellular studies on malignant fluids

All patients showed a considerable reduction in the number of malignant clumps per cytospin and cells per high-power field after treatment; in three patients no malignant cells were present 21 days after treatment, although one patient had previously shown 63 clumps. In case 1 malignant cells had again reappeared by the time of re-admission, but they disappeared after further treatment. In most cases there was an early polymorph response in the fluid after *C parvum* treatment, which changed to a predominantly lymphocyte picture in the later specimens.

Discussion

Undoubtedly *C parvum* is likely to be useful in treating malignant effusions. The most striking histopathological finding was the fibrinous exudate and formation of fibrous tissue within the peritoneal and pleural cavities. Riethmuller suggested that this might cause intestinal obstruction (personal communication, Professor G Riethmuller, Chirurgische Klinik und Poliklinik der Universität Tübingen 74, Tübingen, Germany) after he had found fibrinous deposits on the peritoneum and some stenosis of the ileum at necropsy in a patient who had had malignant melanotic ascites and had been treated with *C parvum*. His patient had developed a paralytic ileus before death. None of our patients did, in fact, develop clinical intestinal obstruction. The spleens that were examined showed a reactive state with plasma cell and polymorph infiltration. This may have indicated an immunological response.

We did not determine an optimum dose of *C parvum*, but 2 ml produced an appropriate response without an unduly unpleasant reaction for the patient. Our difficulty in case 1 was how to use the *C parvum* after the ascites had been cured. No more *C parvum* was given until she relapsed, but monthly doses of *C parvum* might have delayed the relapse. In case 4, however, *C parvum* 1 ml was given on two occasions at weekly intervals starting seven days after the operation. In spite of this a malignant pleural effusion occurred within three weeks followed very quickly by a large abdominal effusion. This suggests that *C parvum* is much more beneficial when introduced at the site of tumour activity. In case 4 the right pleural effusion was treated, with the result that it and the number of malignant cells within it were greatly reduced, whereas the untreated left pleural effusion at necropsy was large, blood-stained, and had 80 clumps of malignant cells per cytospin preparation. Certainly the *C parvum* produced the fibrinous pleural adhesions and reduced the number of malignant cells. In most cases *C parvum* produced a polymorph reaction in the ascitic fluid, which became lymphocytic. Reduction in the fluid did, however, occur without a polymorph reaction taking place. A polymorph reaction was not a feature of the response to subsequent injections of *C parvum*; this was clearly shown in case 1 on readmission, when a very distended abdomen with loculated ascites settled well on the reintroduction of intraperitoneal *C parvum*; there was no polymorph response but a clear reduction in the number of malignant cells. It therefore seems likely that *C parvum* has a more direct effect as well as the inflammatory response that inhibits the malignant effusions. This appears to make it more effective than other substances, whose chief mode of action is probably the simple irritant effect they produce.

We thank the physicians and surgeons of St Thomas's Hospital who asked our help in trying to treat these patients and also the pathologists who performed the necropsies. We are also grateful to the Wellcome Research Laboratories for supplying the *C parvum*.

Reference

[1] Scott, M T, *Seminars in Oncology*, 1974, **1**, 367.

(*Accepted 6 December 1977*)

SHORT REPORTS

Arteriovenous fistula after hair transplantation

Hair transplantation for baldness has been used for many years and is now the most common cosmetic operation for men. Various procedures have been described, including the use of rotation flaps and of bipedicle and monopedicle hair-bearing flaps and partial excision of full segments of hairless skin. The simplest, most effective, and most popular method is the punch-graft technique, in which small cylinders of autologous, full-thickness skin from hair-bearing areas are transplanted to bald areas by means of a skin biopsy punch. This technique may be carried out under local anaesthesia and is said to have relatively few complications; we, however, report a rare exception.

Case report

The patient, a 34-year-old man, had suffered from progressive premature baldness since his early 20s. For three years he had attended a private hair transplant clinic, during which he had undergone eight punch-graft operations. On the last occasion the donor area was the right temporal region.

Eighteen months after completing the treatment he presented at our vascular clinic with a nine-month history of buzzing in his right ear. This was associated with throbbing in the right auricular region. Both symptoms had gradually increased in severity, and over the previous two months he had noticed a swelling above the right ear, which was more prominent