Massive Nitrogen Mustard Therapy in Hodgkin’s Disease with Protection of Bone Marrow by Tourniquets

By Marcel E. Conrad, Jr. and William H. Crosby

The beneficial effects of nitrogen mustard in the treatment of Hodgkin’s disease have been well documented. Unfortunately, as the disease progresses increasing dosage and frequency of administration are necessary to produce tumor regression. The amount of therapy administered is limited by severe depression of the bone marrow with the development of pancytopenia.1,2

The purpose of this article is to report a method for protection of normal hemopoietic tissue without modifying the destructive effect of nitrogen mustard on neoplastic tissue. In performance of this study a method of isolating a portion of the marrow, in vivo, was developed. The gastrointestinal toxicity of nitrogen mustard was reevaluated. Evidence of neurologic toxicity occurred which has not been previously reported following the systemic administration of this chemotherapeutic agent in man.

Materials and Methods

The patients ranged in age from 22 to 44 years. There were 6 males and 2 females. In each case the diagnosis of Hodgkin’s disease was established by microscopic examination of biopsied tissue. The diagnosis had been established one to nine years prior to therapy. Seven patients had received x-radiation; six, nitrogen mustard; four, chlorambucil and one, cyclophosphamide. Two patients were receiving prednisone at the time of therapy. There was no evidence of bone marrow involvement by Hodgkin’s disease in any patients and roentgenologic examination of arms and legs showed no evidence of osseous involvement.

The patients were premedicated with pentobarbital, meperidine and atropine 30 minutes before being taken to the operating room. Once in the operating room an intravenous infusion of saline was started through a 15 gauge needle in the antecubital vein of one arm. The other arm and both legs were elevated and massaged centrally to empty them of blood. With the extremities elevated, elastic bandages were applied tightly from the toes and fingers to the upper third of the three extremities. At the edge of the bandages orthopedic pneumatic tourniquets were applied but not inflated. Anesthesia was induced with sodium thiopental and maintained with oxygen and nitrous oxide. Endotracheal intubation was employed. Once adequate anesthesia was attained the orthopedic cuffs were inflated to a pressure of 575 mm. Hg. over the thighs and 375 mm. Hg. over the arm.13,16 The extremities were then lowered to the operating table. Nitrogen mustard, 60 to 90 mg., was administered in 10 mg. increments, through the intravenous infusion tubing. The cuffs were deflated 15 minutes after the administration of the nitrogen mustard. The patients were allowed to recover from anesthesia and were returned to a room in which all surfaces and fixtures had been washed with hexachlorophene detergent. Strict isolation technic was maintained until the patient recovered from the induced granulocytopenia.

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Table 1.—Patients Treated with Nitrogen Mustard with Orthopedic Cuff Protection of Marrow

<table>
<thead>
<tr>
<th>Pt.</th>
<th>Age</th>
<th>Sex</th>
<th>Duration disease (mos.)</th>
<th>Previous therapy</th>
<th>Manifestations at time of therapy</th>
<th>Dosage HN₂ (mg. per Kg.)</th>
<th>Recovery from HN₂ (days)</th>
<th>Duration of remission (days)</th>
<th>Complications</th>
</tr>
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<tbody>
<tr>
<td>1</td>
<td>44</td>
<td>M</td>
<td>81</td>
<td>Radiation, HN₂</td>
<td>Generalized lymphadenopathy,</td>
<td>1.08</td>
<td>17</td>
<td>41</td>
<td>Headaches, tinnitus, local thrombophlebitis</td>
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<td></td>
<td></td>
<td></td>
<td></td>
<td>chlorambucil spleenectomy</td>
<td>hepatosplenomegaly, fever, wt. loss, anemia.</td>
<td></td>
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<tr>
<td>2</td>
<td>22</td>
<td>M</td>
<td>30</td>
<td>Radiation, HN₂</td>
<td>Generalized lymphadenopathy,</td>
<td>1.0</td>
<td>20</td>
<td>58</td>
<td>Headaches, tinnitus, deafness, alopecia, local thrombophlebitis</td>
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<td></td>
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<td></td>
<td></td>
<td>chlorambucil</td>
<td>hepatosplenomegaly, fever, wt. loss, anemia.</td>
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<td></td>
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<tr>
<td>3</td>
<td>31</td>
<td>F</td>
<td>10</td>
<td>Radiation chlorambucil</td>
<td>Mediastinal-pulmonary-pericardial involvement, superior vena caval obstruction, Horner's syndrome, fever and wt. loss.</td>
<td>0.95</td>
<td>16</td>
<td>56</td>
<td>Headaches, tinnitus, local thrombophlebitis</td>
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<tr>
<td>4</td>
<td>28</td>
<td>M</td>
<td>47</td>
<td>Nitrogen mustard</td>
<td>Pulmonary lesion, general lymphadenopathy, hepatosplenomegaly, fever, wt. loss.</td>
<td>1.2</td>
<td>13</td>
<td>240</td>
<td>Headaches, tinnitus, local thrombophlebitis</td>
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<tr>
<td>5</td>
<td>30</td>
<td>F</td>
<td>89</td>
<td>Radiation chlorambucil prednisone</td>
<td>Retroperitoneal and infraclavicular nodes, fever, wt. loss.</td>
<td>1.3</td>
<td>32</td>
<td>139</td>
<td>Headaches, tinnitus, deafness, alopecia, local thrombophlebitis, pneumonia</td>
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<td>6</td>
<td>22</td>
<td>M</td>
<td>37</td>
<td>Radiation, HN₂</td>
<td>Mediastinal-pulmonary-pericardial involvement, S.V.C. obstruction, lymphadenopathy, hepatosplenomegaly, fever, wt. loss.</td>
<td>1.4</td>
<td>28</td>
<td>50</td>
<td>Headaches, tinnitus, deafness, alopecia, local thrombophlebitis. Died 78 days postoperatively with internal and external hydrocephalus</td>
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<td></td>
<td></td>
<td></td>
<td></td>
<td>cyclophosphamide</td>
<td></td>
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<td>36</td>
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<td>82</td>
<td>Radiation, HN₂</td>
<td>Retroperitoneal, mediastinal, intrapulmonary masses, general lymphadenopathy, fever, cyanosis.</td>
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<td></td>
<td>Died 2 days postoperatively of pulmonary embolism</td>
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<tr>
<td>8</td>
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<td>M</td>
<td>87</td>
<td>Radiation, HN₂</td>
<td>Spastic paralysis, fever, inanition, wt. loss, lymphadenopathy.</td>
<td>1.5</td>
<td></td>
<td></td>
<td>Died 3 days postoperatively of aspiration pneumonia</td>
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Fig. 1.—Case 2. Bone marrow aspirates from the posterior iliac crest (upper row) and tibial plateau (lower row) X230.

The hyperplastic iliac crest marrow was aplastic 20 days after the administration of 1.0 mg./Kg. of nitrogen mustard. Thirty days post therapy it was hypercellular. The fatty tibial marrow was hypercellular twenty days following therapy.

Prior to all injections the skin was prepared with iodine. Each patient received an intramuscular injection of 10 ml. of gamma globulin once weekly. Antibiotics were not used prophylactically.

Blood counts were performed three times weekly. Liver function studies and measurement of blood urea nitrogen were performed once a week. Bone marrow aspirates and marrow biopsies (Vim-Silverman needle) were obtained twice weekly. Gastric and small intestinal biopsies were performed with an intra luminal intestinal biopsy capsule.

**Case Reports**

**Case 1**

A 44 year old white male was admitted to Walter Reed General Hospital (WRGH) in August 1958 with fever, generalized lymphadenopathy, hepatosplenomegaly and anemia. He had noted a right axillary mass in February 1952, which was excised the following year and revealed Hodgkin’s disease. In April 1958 he had recurrence with generalized adenopathy, weight loss, fever and weakness. Nitrogen mustard, 25 mg., given intravenously produced a one month remission. In September 1958 he received radiation therapy to the involved retroperitoneal nodes without improvement. On October 14 he received 60 mg. of nitrogen mustard (1.08 mg. Kg.) intravenously with protection of three extremities by orthopedic tourniquets. Laboratory studies prior to treatment indicated: white count 3200 with 89 neutrophils, hemoglobin 8.7 Gm. and platelets 137,000. The white count began to drop on the third postoperative day, reaching the lowest count of 100 cells per cu. mm. on the eighth day. The lowest platelet count, 1500 per cu. mm., was seen on the fifteenth postoperative day. By November 7, the white count was 3200 with 28 neutrophils, hemoglobin 8.8 Gm. and platelets 29,500. At this time he was released from isolation. Marrow aspirated from the iliac crest, which was hyperplastic prior to the procedure, became hypoplastic on November 4 and normocellular on November 12. Marrow from the tibial plateau, which was initially fatty, showed increasing cellularity, while the unprotected marrow had become aplastic. The patient complained of tinnitus and headaches for two weeks following the procedure. He became afebrile almost immediately with almost complete disappearance of his palpable nodes, liver and spleen. On November 24 he developed fever and pain over the distal ends of the femurs. Roentgen-
Fig. 2.—Case 2. There was a rapid fall in the white cell count with a maximum depression 6 days after receiving 1.0 mg./Kg. of nitrogen mustard. Maximum platelet depression occurred on the tenth postoperative day. There was rapid recovery.

Oligographic examination showed lytic lesions which were not previously demonstrable. Subsequently he received radiation therapy to these lesions with temporary improvement. He had a recurrence of widespread disease and died on January 11, 1959 with a suppurative bronchopneumonia.

Case 2 (Figs. 1 and 2)

A 23 year old white male was admitted to WRGH in March 1959 with generalized adenopathy, hepatosplenomegaly, anemia and a Pel-Ebstein fever. He had first noted a right cervical node in October 1956. Biopsy revealed Hodgkin’s disease. A superior mediastinal mass was successfully radiated in July 1957. In November he developed malaise, fever, anemia and weight loss. He had generalized lymphadenopathy with hepato-
splenomegaly. From February to April 1958 the patient received 50 mg. nitrogen mustard in divided doses, cervical and axillary radiation, and a splenectomy was performed. He was asymptomatic until January 1959, when his findings and symptoms recurred. Cholorambucil, 6 mg. per day, was started. Initially he had improvement but while on therapy his disease progressed and he developed three intrapulmonary lesions. On April 3, the patient received 60 mg. (1.0 mg./Kg.) of nitrogen mustard intravenously with orthopedic cuff protection of three extremities. The lowest white cell count, 1225 was seen on the sixth postoperative day and the lowest platelet count, 66,000 on the ninth postoperative day. On April 20 the white cell count was 4000, hemoglobin 9.3 Gm. and platelet count 101,000. Iliac crest marrow was hypercellular preoperatively, hypocellular five days postoperatively and normocellular on the twentieth day. Tibial marrow which was initially fatty became strikingly hyperplastic in the postoperative period (fig. 1). For two weeks following the procedure the patient had headache and tinnitus. He developed moderate alopecia and a partial, permanent, perceptive deafness. The patient's pulmonary lesions, adenopathy and hepatomegaly could no longer be demonstrated. He subsequently gained 25 pounds. In early June 1959 he developed pain over the femurs followed by inguinal adenopathy, retroperitoneal fulness, cervical adenopathy and a right intrapulmonary lesion. Radiation therapy to each of these regions produced a slight decrease in size, though the patient's systemic complaints continued. In September 1959 the patient was given cyclophosphamide, 150 mg. per day. This was discontinued in early November when he developed granulocytopenia without improvement in his symptoms or findings.

Case 3

A 31 year old white female was admitted to WRGH in May 1959 with fever, severe orthopnea, generalized adenopathy, evidence of superior vena caval obstruction and a right Horner's syndrome. She had noted fever, chest pain, dyspnea and pedal edema in July 1958. When she saw a physician in November she had bilateral pleural effusion and

Fig. 3.—Case 6. Bone marrow aspirates from the posterior iliac crest (upper row) and tibial plateau (lower row) X230.

The cellular iliac crest marrow was aplastic five days after receiving 1.4 mg./Kg. of nitrogen mustard. Only occasional mature cells, stem cells and plasma cells were present. Intermediate erythroid and myeloid elements were notably absent. Twenty-eight days postoperatively the marrow was hypercellular. The protected fatty tibial marrow shows increasing cellularity following therapy beginning at a time when the unprotected marrow was becoming less cellular. (Vim-Silverman needle biopsies.)
Fig. 4.—Case 5. Gradual fall in the white cell count with maximum depression on the eighth day after receiving 1.3 mg./Kg. of nitrogen mustard intravenously.

There was a transient rise in the white cell count when the patient developed pneumonia. The platelet count was maximally depressed on the fourteenth postoperative day. Unlike Case 2 there was slow recovery from the granulocytopenia and thrombocytopenia. This patient had received extensive radiation therapy.

pericardial effusion. A biopsy at thoracotomy established the diagnosis of Hodgkin's disease. Radiation to the mediastinum produced a slight decrease in tumor size and improvement in symptoms. From February to April 1959 the patient received 428 mg. chlorambucil with little benefit. On May 15 she was given 60 mg. (.95 mg. Kg.) of nitrogen mustard intravenously with orthopedic cuffs on three extremities. The white count fell from 12,000 preoperatively to 200 cells on the eleventh postoperative day. The lowest platelet count, 18,000, occurred on the thirteenth postoperative day. Laboratory studies on June 10 revealed: white count 4300, hemoglobin 11.2 Gm. and platelets 328,000. Bone marrow aspirates of the tibia and iliac crest showed changes similar to those in case 2. The patient complained of tinnitus and headaches for two weeks following the procedure. There was improvement in her symptoms, lymph node enlargement and superior vena caval obstruction. In late July 1959 the patient developed abdominal pain and nausea. Her superior vena caval obstruction returned and radiation to the mediastinum was again employed with transitory improvement. With subsequent recurrence the patient was treated with cyclophosphamide.

Case 4 (Fig. 5)

This 28 year old white male was admitted to WRGH because of generalized lymphadenopathy, hepatosplenomegaly and a right intrapulmonary lesion associated with weakness, fatigue, 20 pound weight loss and a Pel-Ebstein fever. He had noted cervical swelling in May 1955. Lymph node enlargement became generalized and was associated with fever. Biopsy in May 1956 revealed Hodgkin's disease. He had a good remission following
30 mg. of intravenously administered nitrogen mustard. Since then he had five similar recurrences which were treated with nitrogen mustard. His periods of remission have decreased, the last persisting only one month. On June 9 the patient received 80 mg. (1.2 mg./Kg.) of nitrogen mustard with orthopedic cuffs on three extremities. His white count slowly decreased from 4300 preoperatively to a low of 800 cells on the eighth day. The maximum platelet depression, 60,000, occurred two days later. The patient was released from isolation on June 25. Headaches and tinnitus lasted for two weeks following the procedure. His enlarged lymph nodes disappeared and his intrapulmonary lesion could not be demonstrated roentgenographically. The liver and spleen decreased in size. The patient gained weight and remained afebrile. There was no recurrence of disease for eight months following therapy.

**Case 5 (Fig. 4)**

This 30 year old white female developed cervical and mediastinal adenopathy in January 1952. Biopsy of left cervical nodes revealed Hodgkin’s disease. By May 1958 the patient had received repeated courses of radiation to the mediastinum, retroperitoneal nodes, cervical, axillary and inguinal regions. Over the next year she received three courses of therapy with chlorambucil. The last course of therapy did not control her fever or back pain and there was persistent left intraclavicular adenopathy. Prednisone, 15 mg. per day, improved her symptoms. Any attempt to reduce the dosage was accompanied by a return of pain and fever. On June 18 the patient received 75 mg. (1.3 mg./Kg.) of nitrogen mustard intravenously with orthopedic cuffs on three extremities. The white cell count fell from 7000 to a low of 150 on the eighth postoperative day. The platelet count was maximally depressed to 1500 on the fourteenth postoperative day. Severe leukopenia and thrombocytopenia persisted until July 20, 1959, when the patient was removed from isolation. During the period of isolation, attempts to reduce steroid dose were unsuccessful because the patient developed symptoms of hypoadrenalism. On June 28 she became febrile with a right lower lobe pneumonia. Pneumococcus and Friedlander’s bacillus were cultured from the sputum. Treatment with penicillin, chloromycetin and erythromycin produced clearing of the infection. The patient complained of severe headaches and tinnitus. She developed a persistent perceptive deafness and alopecia. The latter lasted three months. Following discharge from the hospital prednisone was discontinued. She had no evidence of recurrence of Hodgkin’s disease until mid-November 1959, when she again became febrile with back pain and adenopathy.

**Case 6 (Fig. 3)**

A 22 year old white male was admitted to WRGH in October 1959 because of orthopnea, recurrent superior vena caval obstruction, fever, weight loss, right cervical and axillary lymphadenopathy and hepatosplenomegaly. In September 1956 he had been found to have a superior mediastinal mass. A biopsy at thoracotomy established the diagnosis of Hodgkin’s disease. The tumor responded dramatically to mediastinal radiation. There was decreasing anti-tumor effect with mediastinal x-radiation in 1957 and 1958. In May 1959 there was a recurrence of mediastinal involvement with extension into the left lung, pericardium and superior vena cava. Nitrogen mustard, 30 mg. intravenously, produced transitory improvement. Radiation therapy produced a decrease in signs of superior vena caval obstruction but not in tumor size. In August 1959 cyclophosphamide was given, 100 mg. per day intravenously. There was transitory improvement with recurrence while on therapy. On October 11, the patient received 90 mg. (1.4 mg./Kg.) of nitrogen mustard intravenously with orthopedic cuffs on three extremities. There was improvement in orthopnea and evidences of superior vena caval obstruction within 24 hours. The white cell count fell from 8525 preoperatively to 500 cells on the eighth postoperative day. It remained below 500 until November 2. The lowest platelet count, 7000, occurred on the tenth postoperative day. The patient was isolated until November 9 when the white cell count was 2100 with 61 neutrophils, platelets 173,000, hemoglobin 8.2 Gm. with 8 per cent reticulo-
Radioactive iron 59 studies were performed during bone marrow recovery (fifteenth day) from 1.2 mg./Kg. of nitrogen mustard. Fe\textsuperscript{59} (10 microcuries) was injected into the long saphenous vein at the ankle. Blood specimens were collected by venipuncture from the other ankle. There was a normal half clearance from the plasma (70 mins.). Fe\textsuperscript{59} clearance from the upper extremities was measured in a 4 pi scintillation (small animal) chamber counter. Fe\textsuperscript{59} was incorporated into the bone marrow of the protected extremity (LUE) more rapidly than it was cleared from the blood in the extremity. The increased clearance from the unprotected extremity (RUE) is indicative of fewer erythroid cells. Similar findings were observed in Case 2.

cytos. The patient complained of headaches and tinnitus. He developed a partial perceptive deafness on the third postoperative day. An ecchymosis of the left periorbital region accidentally occurred on October 11. The discoloration remained unchanged until October 30. Then concomitantly with the reappearance of monocytes in the peripheral blood the normal green-yellow color transformation began, indicating local breakdown of hemoglobin pigment in the tissue. On the fourteenth postoperative day the patient became aphasic and developed a right hemiparesis. This occurred again two days later. Each episode lasted less than five minutes with complete recovery. In late November the patient developed
papilledema with progressive mental confusion, somnolence and weakness. There had been a regression of all tumor masses. In early December the mediastinal mass began to increase in size and grew rapidly. The patient became comatose on December 20 and expired eight days later. Postmortem examination showed internal and external hydrocephalus. There was mediastinal Hodgkin's disease with infiltration of the left lung and pericardium.

Case 7

This 36 year old white male was admitted to WRGH in October 1959 with severe orthopnea. He first had fever, cervical adenopathy and a mediastinal mass in January 1953. Biopsy of a lymph node showed Hodgkin's disease. He received 40 mg. of nitrogen mustard followed by x-radiation of the mediastinum and cervical nodes with marked improvement. Between 1953 and 1957 he received radiation therapy to the mediastinum, axillae, cervical, inguinal and retroperitoneal areas. Osteolytic lesions of the ribs and skull were similarly treated. In July 1958 esophageal involvement was treated by radiation with improvement. During that hospitalization pulmonary tuberculosis was demonstrated by culture. The patient was maintained on anti-tuberculous therapy during the remainder of his life without recurrence. In August 1959 he developed a large abdominal mass with right axillary and supraclavicular nodes. These did not decrease in size with radiation therapy. On October 29 he received 90 mg. (1.5 mg./Kg.) of nitrogen mustard intravenously with protection of three extremities. Postoperatively he became more cyanotic and a tracheotomy was performed. During the next 36 hours the patient improved dramatically. On October 30 he suddenly expired. Postmortem examination revealed a large embolus filling the right ventricle and extending into both pulmonary arteries. Nodes involved with Hodgkin's disease were markedly necrotic.

Case 8

This 41 year old white male was admitted to WRGH in October 1959 with a spastic paralysis of three extremities, emaciation and Pel-Ebstein fever. He had first developed cervical nodes in July 1952. Biopsy revealed Hodgkin’s disease. Radiation therapy then and in 1955 produced a remission. In 1957 and 1958 recurrences manifest by malaise, weakness, fever and generalized lymph node involvement were successfully treated with intravenous nitrogen mustard. In February 1959 moderate improvement in systemic involvement was achieved with chlorambucil. Only partial and transitory improvement occurred in August following 30 mg. of intravenous nitrogen mustard. In September 1959 the patient developed a spastic paralysis of three extremities with urinary incontinence, weight loss and fever. This was unimproved following 30 mg. of nitrogen mustard and oral prednisone administration. On 28 October the patient received 90 mg. (1.5 mg./Kg.) of nitrogen mustard through a polyethylene catheter inserted through a femoral vein into the inferior vena cava. Four extremities were protected by orthopedic tourniquets. He did well until November 1, when he suddenly became comatose and expired. Postmortem examination revealed the cause of death to have been acute aspiration of gastric contents.

Analysis of Cases

Eight patients with far advanced Hodgkin’s disease, refractory to conventional therapy, were given large dosages of nitrogen mustard intravenously. An attempt was made to protect the bone marrow of the extremities by the application of orthopedic tourniquets during and for 15 minutes following the injection. General anesthesia was utilized because of the pain produced by tourniquets at pressures necessary to prevent arterial blood flow into the extremities (575 mm. Hg in thigh and 375 mm. Hg in arm).15,16 Endotracheal intubation was employed to prevent aspiration of vomitus or saliva.

Nausea and vomiting occurred during recovery from anesthesia and lasted
12 to 24 hours. No patient vomited prior to recovery of the gag reflex. Vomiting was no more severe than is encountered with conventional dosages of nitrogen mustard. Patients (cases 3, 6 and 7) with obstruction of their respiratory tract or superior vena cava showed improvement within 12 hours, postoperatively. Palpable tumors decreased markedly in all patients within 48 hours after the injection.

Headaches occurred in all patients and persisted for several weeks following therapy. They were relieved by elevation of the head and the administration of vasodilators. Permanent, moderate, perceptive deafness developed in three patients (cases 2, 5 and 6). Tinnitus lasted for several weeks and in one patient for three months (case 5). It was unresponsive to medication. Severe temporary alopecia occurred in three patients (cases 2, 5 and 6). Chemical thrombophlebitis at the site of injection of nitrogen mustard (cases 1-7) encouraged us to use a femoral catheter for injection of the nitrogen mustard into the inferior vena cava in case 8.

All patients were isolated. Infection occurred in one patient (case 5). She was receiving steroids and developed pneumonia due to organisms that had been cultured from her oropharynx prior to the administration of the nitrogen mustard. Her response to antibiotics was prompt. Her white count increased during the infection despite severe depression of her marrow at that time. (fig. 4).

One patient (case 6) had two episodes of aphasia and paresis during the period when bone marrow depression was severe. Each episode lasted several minutes and was followed by complete recovery. Later he was shown to have internal and external hydrocephalus. The etiology has not been demonstrated.

Two patients (cases 7 and 8) died in the postoperative period. Both were moribund when the procedure was performed and had shown clinical improvement postoperatively. Case 7 died of pulmonary embolism. Case 8 expired with an aspiration pneumonia. At autopsy their tumor masses were necrotic.

Tumors recurred in the patients after 41 to 240 days following the administration of the large doses of nitrogen mustard. In two of the patients (cases 1 and 3) the recurrence was first noted as an osteolytic lesion distal to the point of tourniquet application in the protected extremities. The lesions had not been roentgenographically demonstrable prior to therapy.

All patients (cases 1-6) developed severe thrombocytopenia and granulocytopenia postoperatively. The white blood cells reached the lowest values in 6 to 8 days and platelets between the 9th and 15th day. Lymphocytes disappeared from the peripheral blood first, followed by the monocytes, neutrophils and platelets. During the recovery period the cells reappeared in the peripheral blood in the same order. This is probably a function of the life expectancy of each cell type. Bone marrow aspirates from the iliac crest three days after therapy showed only mature cells and stem cells. There was almost complete absence of intermediate erythroid and myeloid elements. Severe aplasia developed in each patient. The marrows became normocellular by the thirtieth postoperative day. Abnormal erythroid precursors (megaloblastoid) were frequently observed during this period. Aspirates from the
tibial plateau became progressively more cellular in the postoperative period. Increasing cellularity of this protected bone marrow occurred during the period when the unprotected sternal marrow was becoming progressively hypoplastic (figs. 1 and 3). Preservation of functioning erythroid tissue was confirmed by radioactive iron studies (fig. 5).

Biopsies of the stomach and jejunum were obtained in three patients at regular intervals from 12 hours to 8 days postoperatively. The mucosa of the intestine remained intact. Mitotic activity was decreased in the crypts of Lieberkühn. There was moderate hyperemia of the lamina propria. The cytoplasm of the epithelial cells showed vacuolization (fig. 6).

Liver function tests and measurement of blood urea nitrogen were performed at weekly intervals. There was no functional deterioration of the liver or kidneys demonstrated by these studies.

**Discussion**

Early in the course of Hodgkin's disease nitrogen mustard usually produces dramatic and often long-standing remissions. As the disease progresses, larger and more frequent therapy is necessary to produce briefer periods of tumor regression. The lack of specificity for neoplastic tissue with the injurious effect to normal hemopoietic tissue limits the amount of nitrogen mustard that can be used therapeutically. In an attempt to avoid the systemic toxicity and at the same time increase

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**Fig. 6.—Case 6. Biopsies of the jejunum (left) and gastric mucosa (right) obtained 72 and 48 hours, respectively, after the administration of 1.4 mg./Kg. of nitrogen mustard.**

The mucosa was intact as in prior and subsequent biopsies from both sites. The epithelial cytoplasm shows vacuolization. There was a decrease in mitotic cells in the crypts (not shown). Degenerative changes were seen only in the epithelial cells of the bases of the crypts of Lieberkühn and the necks of the gastric glands. The mucosa remained intact in these areas. Biopsies were performed with an intraluminal intestinal biopsy capsule. X780.
the dosage of nitrogen mustard for its anti-tumor effect several methods have been proposed. Regional administration has been employed with localized tumors (intra-arterial, extracorporeal circuits and instillation into serous cavities).\textsuperscript{4,7} Drugs are being investigated that decrease toxicity without diminution of the beneficial therapeutic effects of the agent.\textsuperscript{5} More recently, preserved autologous bone marrow has been reinfrused into patients following the administration of large doses of nitrogen mustard in hopes of repopulating the bone marrow.\textsuperscript{9} We have attempted to protect a portion of the bone marrow, in vivo, by tourniquets, during the administration of nitrogen mustard hoping that this small quantity of marrow would be sufficient to repopulate the aplastic marrow in the unprotected areas.

To insure adequate protection of the bone marrow from the effects of a chemotherapeutic agent it is essential to isolate the area to be protected until the agent is inert or tissue fixed. It has been shown by arterial ligation and perfusion experiments in animals that nitrogen mustard is tissue-fixed two to eight minutes after its entry into the circulation.\textsuperscript{7,10} Thus if the bone marrow could be isolated for this period of time in humans, adequate protection should be achieved. Tourniquet protection of the human bone marrow has been attempted by ourselves and others in the past.\textsuperscript{11} It was unsuccessful because of failure to achieve adequate compression of the tissues with regular sphygmomanometer cuffs. Observation of amputations with tourniquet compression at less than 100 to 150 mm. Hg pressure above systolic arterial blood pressure will provide adequate evidence of the patency of the deep arterial circulation at these external pressures. The application of orthopedic tourniquets at pressures of 575 mm. Hg over the thigh and 375 mm. Hg over the arm is known to provide hemostasis.\textsuperscript{15,16} Utilization of these pressures for 15 minutes in our patients provided marrow protection from systemically administered nitrogen mustard. Increasing cellularity and function of the protected marrow was demonstrated at a time when marrow proximal to the tourniquets was severely aplastic (figs. 1 and 3).

It is evident from the granulocytopenia and thrombocytopenia seen in our patients that the bone marrow of the extremities is insufficient to supply an adequate number of cells to populate the peripheral blood. The only benefit derived from preservation of the extremity marrow is that it may seed the aplastic marrow spaces and decrease the period of pancytopenia. But this is difficult to prove.

The infusion of small amounts of autologous marrow in lethally radiated animals has been shown significantly to enhance survival.\textsuperscript{12} It would seem that in vivo preservation of the marrow would provide more physiologic means of repopulation. Comparison of our patients with reported cases receiving comparably large dosages of alkylating agents without bone marrow protection shows our patients to have an earlier reappearance of marrow function.\textsuperscript{5,13} Lastly, the course of our patients compares favorably with the patients reported by McFarland et al. who received autologous bone marrow infusion after "super-dosages" of nitrogen mustard.\textsuperscript{9}

The lethal dose of nitrogen mustard in humans, expressed as an LD\textsubscript{50}, is not known. It has been thought to be approximately 1.0 mg. per Kg. of body
weight. There have been survivals with doses of 1.1 mg. per Kg. using autologous marrow infusions after treatment with nitrogen mustard. We have had survival with repopulation of the marrow after 1.4 mg. per Kg. of body weight. This expression of dosage is probably significantly smaller than the effective dose to the treated tissue because of the exclusion by the tourniquets of approximately 25 per cent of the body tissues from the circulation. This is supported by the evidence of toxicity (eighth nerve damage with tinnitus and deafness, vascular headaches and alopecia) which occurred with this dose. These phenomena were not previously noted following the intravenous administration of large doses of nitrogen mustard in humans.

It has been thought that gastrointestinal toxicity would limit the dosage of nitrogen mustard once the problem of hemopoietic depression was circumvented. Based on autopsy findings this was postulated to occur in the dosage range of 1.1 to 1.4 mg. per Kg. of body weight. However, the serial biopsies of stomach and jejunum during the week following the administration of 1.2 to 1.4 mg. per Kg. of nitrogen mustard revealed an intact intestinal mucosa with only minor histologic changes (fig. 6). The maceration of the gastrointestinal tract previously reported was probably due to postmortem autolysis. Severe gastrointestinal toxicity is probably not a major problem.

The period of pancytopenia was especially prolonged in patients who had received repeated courses of radiation therapy (fig. 4) (cases 1, 5 and 6). Previous therapy with nitrogen mustard did not increase the period of bone marrow aplasia (cases 2 and 4). Radiation therapy probably produces permanent damage to the bone marrow hindering repopulation with functional tissue. The duration of pancytopenia can be predicted by periodic bone marrow examinations. We found that an ecchymotic area can likewise be used for prognostication (case 6). The normal blue-green-yellow color change due to the local degradation of hemoglobin to bile pigment by tissue phagocytes does not occur until marrow regeneration begins and monocytes reappear in the peripheral blood.

It is believed that further investigation of this method of treatment is necessary before its value in the general treatment of Hodgkin’s disease and the lymphomas can be ascertained. It is thought that larger doses of nitrogen mustard may be feasible by our technic, though they may be complicated by irreversible neurologic damage. There is suggestive evidence that patients who have received extensive radiation and those who are receiving steroid therapy are poorer risks for this procedure. Future patients treated by this method will be heparinized because of the occurrence of thrombembolism in one patient. Whether this was due to therapy or occurred incidentally to it is not known.

Three additional patients with advanced Hodgkin’s disease have been treated with divided doses of nitrogen mustard. One received 35 mg. thrice during eight days (1.68 mg./Kg.), the second received 50 mg. twice (1.75 mg./Kg.) with a 48 hour interval between doses and the third, an obese female, received 65 mg. twice (1.6 mg./Kg.) with 96 hours between doses. Three extremities were protected by orthopedic tourniquets during administration of each dose. The patients were heparinized during treatment to
prevent thromboembolism. Each patient had previously received moderately extensive radiation therapy, yet there was less hematopoietic depression and more rapid recovery than in the patients who had received a single massive dose. Only mild headaches and tinnitus occurred and there was no deafness or alopecia. There was rapid regression of demonstrable tumor. Whether the antitumor effect of the divided dose schedule is less or shorter, paralleling the diminished toxicity, remains to be seen.

Summary

1. Eight patients with far advanced Hodgkin's disease were treated with massive single doses of nitrogen mustard (0.95-1.5 mg./Kg.) with tumor regression in each case.
2. An attempt was made to protect the bone marrow of the extremities from the effects of the nitrogen mustard by applying orthopedic tourniquets during the injection. There was definite evidence that the marrow was protected by this procedure. Marrow in the tibia became progressively more cellular during the time that the marrow in the torso degenerated to almost complete aplasia. During the period of recovery, radioactive iron was given and more of it localized in the protected arm than in the unprotected, evidence of greater erythropoietic activity.
3. Neurologic complications occurred at the doses employed.
4. Serious gastrointestinal pathology was not demonstrated at these doses.
5. Prior extensive radiation therapy prolonged the hemopoietic depression due to nitrogen mustard. This was not produced by previous mustard therapy.
6. Color change in ecchymotic areas may be used to predict bone marrow recovery.
7. Dividing the massive dose seems to result in less toxicity.

Summary in Interlingua

1. Octo patientes con avantiatissime morbo de Hodgkin eseva tractate con massive doses unic de mustarda de nitrogeno (0,95 a 1,5 mg per kg), resultante in un regression del tumor in omne le casos.
2. Esseva facite le essayo de proteger le medulla ossee del extremitates contra le effectos del mustarda de nitrogeno per le application de tourniquets orthopedic durante le injection. Esseva notate definite indicios que le medulla eseva protegite per iste mesura. Le medulla in le tibia deveniva progressivemente plus cellular in le spatio de tempore in que le medulla del ossos del torso se degenerava usque a un aplasia quasi complete. Durante le periodo del restablimento ferro radio-active eseva administrate, e un plus grande quantitate de illo se localisava in le protegite bracio que in le non-protegite bracio, lo que reflecteva un plus grande activitate erythropoietic.
3. Complicationes neurologic occurreva con le doses que eseva empleate.
4. Serie formas de pathologia gastrointestinal non eseva demonstrate con iste doses.
5. Un antecedente extense therapia radiational prolongava le depression
MASSIVE NITROGEN MUSTARD THERAPY IN HODGKIN'S DISEASE

hematopoietic causae per le mustarda de nitrogeno. Un antecedente therapia a mustarda non habeva iste effecto.

6. Alterationes del color in areas ecchymotic pote esser usate pro predicer le restablimento del medulla ossee.

7. Le division del dose massive pare resultar in un reducite toxicitate.

REFERENCES


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—R. M. H.
Massive Nitrogen Mustard Therapy in Hodgkin’s Disease with Protection of Bone Marrow by Tourniquets

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