THE USE OF CYTOTOXIC DRUGS IN THE SURGERY OF MALIGNANT DISEASE

Surgeon Captain J. Watt, Royal Navy

From the Royal Naval Hospital, Haslar, Gosport, England

Cancer chemotherapy has been practised from time immemorial and Rhazes, a Baghdad surgeon, introduced chemistry into "physic" in the ninth century A.D. In 1894 W. B. Coley used the toxins of streptococcus pyogenes and bacillus prodigiosus to treat twelve patients with inoperable bone sarcoma, and disappearance of the tumour occurred in three cases. Johnston (1962), in a carefully controlled trial designed to re-evaluate the therapeutic effects of Coley's fluid, obtained objective improvement in nine of thirty-four patients with inoperable metastatic neoplasms treated by Coley's toxin; she found no improvement in thirty-seven controls treated with typhoid vaccine. However, the stimulus to modern cytotoxic therapy came in the second world war. Gilman and Philips (1946) published an account of the biological and therapeutic effects of sulphur and nitrogen mustards. Wilkinson and Fletcher (1947) reported the results of their work in Britain, and the intense industrial and medical research of post-war years resulted in the screening of thousands of new agents. Hirschberg (1963) reviewed 2,300 papers written between 1945 and 1958. Klopp, Alford, Bateman, Berry and Winship (1950) reported the accidental discovery of intra-arterial injection as a means of treating malignant disease confined to a single region of the body, and recent results in biochemistry and microbiology have determined the nature and site of action of cytotoxins in the malignant cell.

The early 1950's brought more knowledge of cell division and the transfer of hereditary information carried by deoxyribonucleic acid (DNA). In 1953 Watson and Crick described
the DNA fibre as a double alpha helix, each strand running in opposite directions and cross-linked in step ladder fashion by weak hydrogen bonding of pairs of nucleotides, confirming the work of Chargaff and Davidson (1955) who had demonstrated mandatory base-pairing between the large purine and small pyrimidine bases in coded sequence, specific for each organism. Single-stranded messenger ribonucleic acid (RNA) transmits this coded information to the cytoplasm where it is scanned by ribosomes which, by means of transfer RNA, string together amino acids in predetermined order to form long-chain protein molecules under the influence of enzymes which are themselves protein. Cytotoxic drugs may prevent the DNA replication, essential for cell division, or disrupt cell metabolism at any site. They fall into three main groups: 1) alkylating agents, 2) antimetabolites and 3) a group of miscellaneous compounds.

Alkylating agents—Alkylating agents are radiomimetic. They have a short half-life, and bind adjacent guanine bases to short-circuit the co-axial, double helix of DNA during the early interphase stage of the mitotic cycle (Brookes and Lawley 1961) (Fig. 1). This has chromosomal implications leading to the death of the cell and is manifest by transverse splits of the centromeres, chromosome fragmentation and despiralisation, and chromatid breaks and interchanges (Koller 1958, Revell 1958, Loveless 1966).

![Chemical structures of alkylating agents]

There are four main groups of alkylating agents, all capable of adding a highly reactive alkyl group to other molecules: 1) Nitrogen mustards or chloroethylamines, which release their alkyl group after separation of the negatively charged chloride ion; 2) ethyleneimines, like thiotepa and trenimon; 3) epoxides, like epodyl, which snap open their cyclical rings to release the reactive alkyl groups; and 4) sulphonic acid esters, like busulphan (myleran).

Alkylating agents act best upon rapidly dividing cells. They are fixed by malignant cells in mitosis; their action is unselective, and also affects the dividing cells of the gastrointestinal tract and haemopoietic tissues. In fact, there is little response until the agent is given in toxic doses, and dosage must be precisely determined.
To reduce toxicity and evolve specificity the alkylating agents have become more sophisticated, and attempts have been made to design transport forms best suited to carry the twin alkyl weapons to their target area within the malignant cell (Bergel 1961).

The methyl carrier of nitrogen mustard which is a strong base causes the chloride ion to separate rapidly when in contact with acidic neoplastic tissue, whereas the weaker bases of melphalan and chlorambucil react more slowly. Cyclophosphamide (endoxana) is an enzyme-dependent mustard, inactive until the phosphoramidate linkage is broken down by intracellular enzymes. Mannitol mustard was given a sugar to facilitate its passage across cell membranes, and chlorambucil a detergent to be transferred to fat. Uracil mustard uses one of the pyrimidine bases essential to DNA synthesis to carry its reactive groups, and melphalan, the amino acid phenylalanine, which probably becomes incorporated into protein molecules to inhibit essential activity (Fig. 2).

**Antimetabolites**—The alkylating agents are rapidly fixed and rapidly inactivated, affecting only those cells about to divide, and leaving cells in other phases of mitosis largely untouched. Antimetabolites, on the other hand, act by competitive inhibition of all nucleoprotein synthesis.

In the synthesis of DNA and RNA purine and pyrimidine bases combine with the 5-carbon sugars, deoxyribose or ribose, and are then called nucleosides. These in turn are esterified with phosphoric acid to form the essential nucleotide links of the DNA and RNA chains. Folinic acid acts as co-enzyme in the synthesis of purine and pyrimidine bases and some essential amino acids. It accelerates the metaphase of the mitotic cycle and also enables messenger RNA to carry the genetic code from the DNA in the nucleus to the ribosomes in the cytoplasm (Jacobson 1966). There is evidence that folinic acid analogues can be inhibited by osteoblasts and fibroblasts, which presumably precludes their use in the treatment of sarcomas.

The antimetabolites fall into four broad groups according to their site of action: 1) analogues of purine and pyrimidine bases such as 6-mercaptopurine and 5-fluorouracil and their nucleosides; 2) folic acid analogues like methotrexate, which can be given with the corresponding metabolite, folinic acid or citovorum factor, to protect the vulnerable blood-forming organs (Sullivan, Miller and Sikes 1959); 3) glutamic acid antagonists, the alkaloids obtained from the Madagascar periwinkle, vinblastine and vincristine; and 4) antibiotics which include actinomycin D and C.

Because of similarity of chemical structure, analogues of the bases, when present in excess, become incorporated into DNA structure, causing mutations during DNA replication due to altered biochemical sequences. In an attempt to reduce toxicity, the analogue of the nucleoside rather than the analogue of the base has been used (Fig 3), a good example being FUDR or flouxuridine, but there is some doubt about its effectiveness.

The vinca alkaloids probably interfere with utilisation of glutamic acid (Johnson, Armstrong, Gorman and Burnett 1963), either preventing its incorporation into the amino-acid chain of enzyme molecules or blocking the action of glutamic acid in enzyme systems. They may also cause arrest of mitosis at the metaphase (Palmer, Livengood, Warren, Simpson and Johnson 1960). Although they depress white cells, these plant extracts have less effect upon platelets.

Antibiotics appear to exert their antimetabolite action by interfering with the synthesis of messenger RNA (Slotnick 1960, Butler 1964). DNA synthesis being quite unaffected by actinomycin D which immediately arrests RNA and protein synthesis. A scheme modified
from that of Weber (1963) provides one suggestion of how vinblastine and actinomycin D might act (Fig. 4).

**Miscellaneous agents**—A group of miscellaneous agents are used which include metaphase arrestors like colchicine and podophyllin derivatives (Stamm and Stähelin 1965), hydroxyurea (Davis and Lariomov 1964), methylhydrazines, like Natulan, an agent of low toxicity (useful in Hodgkin’s disease) (Bollag 1963), and experimental compounds like the metal chelates of copper and ruthenium (Dwyer, Mayhew, Roe and Shulman 1965). Thornes and O’Meara (1961) described a coagulative factor responsible for the invasiveness of malignant cells and which can be inhibited by a protamine derivative, prolothran. We have obtained remissions in patients suffering from widespread malignant melanoma and metastases from carcinoma of the breast by combining a prolothran infusion with an antimetabolite, after more conventional cytotoxic therapy has failed.

**INDICATIONS FOR CYTOTOXIC THERAPY**

There are probably four main indications for cytotoxic therapy in surgical practice: 1) prophylaxis, 2) pre-ablative therapy, 3) local and regional malignancy, 4) inoperable, recurrent or metastatic disease.

**Prophylaxis**—Smith, Thomas and Hilberg (1958) demonstrated cancer cells in wound washings from 26 per cent of patients undergoing definitive cancer surgery, and Morgan (1955), using
1:500 perchloride of mercury irrigations, reduced suture line recurrences after restorative operations for carcinoma of the rectum from 21.4 to 2.09 per cent. Freshly prepared 0.5 per cent sodium hypochlorite solution is, however, safer and more effective.

Forrest (1961), Sellwood, Kuper, Wallace and Burn (1965) and others isolated malignant cells in the peripheral blood during operation. A quick acting alkylating agent such as cyclophosphamide, given in fractionated doses intravenously during and after operation, can deal with this problem.

Fernbach and Martyn (1966) have shown that a 57 per cent mortality rate has been converted into a 92 per cent survival rate by the addition of actinomycin D before operation, and irradiation afterwards in the treatment of Wilm's tumour.

Pre-ablative therapy—Intra-arterial injection of alkylating agents has been used successfully before pneumonectomy for lung cancer (Poulsen 1962) and excision of the rectum (Salsbury, McKinna, Griffiths and Morgan 1965). Newton (1965) reported the results of arterial infusion during the last two weeks of a course of irradiation before operation for sarcoma of the limb. Complete or massive tumour destruction occurred in several cases, but we need a controlled trial like that of Fernbach and Martyn (1966) to find out what happens about the influence of actinomycin D on Wilm's tumour. The alkylating agents, mustine, cyclophosphamide, melphalan and epodyl, and the antimetabolites, methotrexate and flouxuridine, have been used in intra-arterial techniques and, more recently, the podophyllin derivative (SPI), which is supposed to be free from toxic effects (Westbury 1967). Pratt, Betteridge and Dixon (1963) and Lentin and Nambier (1964) described techniques of regional cytotoxic perfusion before radical mastectomy.

Local and regional malignancy—Malignancy confined to a single region of the body, with a readily available blood supply, is an indication for intra-arterial therapy in conjunction with surgery, either as a continuous low-concentration infusion, or as a short-duration, high-concentration isolation-perfusion requiring temporary extra-corporeal circulation with pumps, heat exchanger and bubble oxygenator (Creech, Kremetz, Ryan and Winblad 1958; Irvine, Noon and Bastable 1962). Raising the temperature of the perfusate increases the speed of reaction. Tourniquets, hypothermia and a perfusion pressure lower than the systemic blood pressure will protect the rest of the body and reduce leakage to the general circulation. Leakage can be monitored by dye dilution methods, 51Cr labelled red cells or 125I labelled albumin. Lawrence, Clarkson, Kim, Clapp and Randall (1964) used balloon-tipped catheters passed up the femoral vessels to occlude the aorta and inferior vena cava which overcomes the need to open the abdomen in ill patients.

For arterial infusion we use a 93 centimetres fine-gauge epidural catheter. Sites of cannulation for the arm are through the radial, brachial and anterior circumflex humeral arteries into the axillary vessel; for the head and neck, through the superficial temporal or superior thyroid into the external carotid artery; for the breast, through the superior epigastric or subclavian artery into the internal mammary artery.

Percutaneous selective angiography has been most useful for infusion of the lower limb and perfusion of abdominal organs (Byron, Perez, Yonemoto, Bierman, Gildenhorn and Kelly 1961; Thomson and Foote 1963), for which flouxuridine (Rochlin, Smart and Silva 1965) is at present being tested. Campbell and Green (1966) have described a technique for selective perfusion of a lumbar artery supplying a solitary vertebral metastasis.

Accurate siting of an intra-arterial catheter can be determined by dyes such as patent blue violet (Engeset, Brennhovd and Stovner 1962) and disulphine blue (Tempest 1960) or by injection of 5 per cent fluorescein and examination of its regional distribution by ultraviolet light. We have found that dyes are more satisfactory in the head and neck, and fluorescein in the limbs. Arteriography confirms siting of the catheter in abdominal perfusions. Leakage of the agent not taken up by the regional tissues is more difficult to monitor when infusion is used, and we have had difficulty in obtaining agents with a radioactive label.

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We formerly used a gravity method of arterial infusion (Espiner, Vowles and Walker 1962) with bottles held at 300 centimetres (10 feet) above the patient but we now use the Fenwal pressure cuff (Krant, Hall, Lloyd and Patterson 1961) over a plastic bag containing the solution. Plastic bags reduce the danger of air embolism.

<table>
<thead>
<tr>
<th>Author</th>
<th>Method</th>
<th>Number of cases</th>
<th>Response to therapy (per cent)</th>
<th>Agents</th>
</tr>
</thead>
<tbody>
<tr>
<td>Krementz et al. (1962)</td>
<td>Arterial perfusion</td>
<td>56</td>
<td>67</td>
<td>Melphalan, mustine thiotepa</td>
</tr>
<tr>
<td>Gerhartz (1964)</td>
<td>Intravenous infusion</td>
<td>13</td>
<td>40</td>
<td>Endoxana</td>
</tr>
<tr>
<td>Gross (1964)</td>
<td>Intravenous infusion</td>
<td>20</td>
<td>40</td>
<td>Endoxana</td>
</tr>
<tr>
<td>Rochlin (1964)</td>
<td>Arterial perfusion</td>
<td>47</td>
<td>68</td>
<td>Melphalan, actinomycin D</td>
</tr>
<tr>
<td>Ryan et al. (1964)</td>
<td>Arterial perfusion</td>
<td>41</td>
<td>71</td>
<td>Melphalan, thiopeta</td>
</tr>
<tr>
<td>Trapeznikov and Avdeeva (1964)</td>
<td>Arterial perfusion</td>
<td>22</td>
<td>?</td>
<td>Melphalan, ftorpan</td>
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</tbody>
</table>

Treatment can be continued, even as an out-patient, by means of a small clockwork pump (Watkins 1963; Barkin, Cowan, MacDonald and Mahoney 1966) or screw advancement syringe (Rush, Das and Boone 1966), the patient injecting fractionated doses at regular intervals.

Figure 5 shows the pattern of results obtained in regional perfusion for malignant melanoma, head and neck tumours, and advanced abdominal cancer, and it is seen that most workers claim a 50 to 60 per cent response. But even when patients exhibit no objective remission, relief of pain is often immediate.
Orthopaedic surgeons will be more interested in the effects of intravascular techniques on sarcoma of the limb (Table 1). Soft-tissue sarcomas did much better than bone sarcomas, but the best results were obtained by regional perfusion with melphalan followed by amputation within seven to ten days. Krementz, Creech, Ryan and Reemtsma (1962) recommended perfusion chemotherapy for sarcomas of the extremity as an adjunct to surgical excision of primary lesions and for palliation of advanced lesions.

Lymphangiography can outline metastases in lymph glands and the possibility of delivering chemotherapeutic agents by this route is attractive, but so far unreliable (Cox, Hare and Bruce 1966).

Topical cytotoxins have been found effective against local skin cancers (Belisario 1965).
Inoperable, recurrent and metastatic malignant disease—Skin metastases and their associated discomfort will often disappear with thiotepa, colchicum and prolothan creams. Fixed, inoperable, soft-tissue tumours sometimes respond dramatically to injections of thiotepa directly into the tumour. Generally such tumours are radio-resistant, but there is some evidence that the effects of radiotherapy can be enhanced by 5-fluorouracil (Foye, Willett, Hall and Roth 1960) and actinomycin D (Farber 1961). We have seen pleural and peritoneal malignant effusions respond dramatically to intracavity thiotepa, and the pleural space is often obliterated by this treatment. However, when the disease spreads to the opposite side, deterioration can be rapid (Ariel, Oropeza and Pack 1966).

When dissemination is widespread agents of known specific action may be used, or various combinations of drugs may help in a programme of combined therapy to reduce drug resistance. Figure 6 shows the course of a patient who refused forequarter amputation and demonstrates how this form of treatment can help patients to return to work for months or even years.

Alternatively, metaphase arrestors can be used to achieve mitotic synchrony, then withdrawn to allow malignant cells to complete division, and pass simultaneously into the

<table>
<thead>
<tr>
<th>1 GENERAL TOXIC EFFECTS</th>
<th>2 LOCAL EFFECTS</th>
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<tbody>
<tr>
<td>a Bone Marrow Depression</td>
<td>a Vesication – Pigmentation – Oedema</td>
</tr>
<tr>
<td>Granulocytopenia</td>
<td></td>
</tr>
<tr>
<td>Thrombocytopenia</td>
<td></td>
</tr>
<tr>
<td>Anaemia</td>
<td></td>
</tr>
<tr>
<td>b Epithelial Necrosis</td>
<td>b Tumour Sloughing – Haemorrhage – Sepsis</td>
</tr>
<tr>
<td>Skin Vesication</td>
<td></td>
</tr>
<tr>
<td>Alopecia</td>
<td></td>
</tr>
<tr>
<td>Testicular Atrophy</td>
<td></td>
</tr>
<tr>
<td>Buccal</td>
<td>c Muscle Necrosis (Uraemia)</td>
</tr>
<tr>
<td>Gastrointestinal Exfoliation</td>
<td>d Nerve Damage</td>
</tr>
<tr>
<td>c Lowered resistance to infection</td>
<td>3 VASCULAR COMPLICATIONS OF INTRA-ARterial TECHNIQUES</td>
</tr>
<tr>
<td>Bronchopneumonia</td>
<td>a Arterial Thrombosis &amp; Air Embolism (Hemiplegia)</td>
</tr>
<tr>
<td></td>
<td>b Post Perfusion Peripheral Circulator Collapse</td>
</tr>
<tr>
<td></td>
<td>c Superficial &amp; Deep Venous Thrombosis</td>
</tr>
<tr>
<td></td>
<td>d Catheter Fracture Leakage Dislodgement</td>
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<td></td>
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<td>Fig. 8</td>
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</table>


early resting phase of the cycle during which they are most sensitive to alkylating agents (Fig. 7). We also use cytotoxic therapy in conjunction with endocrine ablation when this is indicated.

Hitchcock (1966) has shown that oxygen and possibly hyperbaric oxygen may improve the results of cytotoxic therapy, and Ross (1961) has suggested that, as neoplastic tissue is more acidic than normal tissue, it should concentrate basic drugs, especially aromatic nitrogen mustards, with basic side chains. The difference in pH can be increased by preliminary treatment with glucose which causes increased accumulation of lactic acid in consequence of glycolysis.

COMPLICATIONS OF CYTOTOXIC THERAPY

Cytotoxic drugs are general tissue poisons, and strict control is necessary to avoid the more serious complications. These are set out in Figure 8, and Figure 9 shows how sudden and dramatic bone marrow depression may be.
SENSITIVITY OF TUMOURS TO CYTOTOXIC AGENTS

There remains the problem of choosing the right drug. Schmid, Cappuccino, Merker, Tarnowski and Stock (1966) published the result of selective action of cytotoxic drugs against a spectrum of animal tumours used by the Sloan-Kettering Institute, and most nearly

representing a cross-section of human tumours (Fig. 10). Cyclophosphamide, purine analogues vinblastine and actinomycins had both immediate and delayed action upon carcinoma and osteogenic sarcoma, and cyclophosphamide and floxuridine (FUDR) were most effective.
against adenocarcinoma. Cyclophosphamide was the only cytotoxic agent to have a prolonged action against every type of tumour.

However, except in the case of cyclophosphamide, clinical experience has often been at variance with such experimental findings, and Table II represents the present state of our knowledge regarding selectivity of tumour agents based upon the accumulated evidence of clinical reports.

### TABLE II

**THE ACTION OF VARIOUS CYTOTOXIC DRUGS**

<table>
<thead>
<tr>
<th>TUMOUR TYPE</th>
<th>AGENTS USED</th>
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<tbody>
<tr>
<td>Choriocarcinoma</td>
<td>Methotrexate</td>
</tr>
<tr>
<td>Wilm's tumour</td>
<td>Actinomycin D</td>
</tr>
<tr>
<td>Gastrointestinal cancer</td>
<td>5-Fluorouracil</td>
</tr>
<tr>
<td>Hodgkin's disease</td>
<td>Endoxana, vinblastine, natulan</td>
</tr>
<tr>
<td>Melanoma</td>
<td>Melphalan</td>
</tr>
<tr>
<td>Basal cell carcinoma</td>
<td>Colchamine</td>
</tr>
<tr>
<td>Skin metastases</td>
<td>Prolothan, colchamine</td>
</tr>
<tr>
<td>Myelomatosis</td>
<td>Melphalan, urethane</td>
</tr>
<tr>
<td>Ewing's sarcoma</td>
<td>Melphalan, endoxana</td>
</tr>
<tr>
<td>Seminoma</td>
<td>Melphalan</td>
</tr>
<tr>
<td>Safest, long-term, non-specific agent</td>
<td>Endoxana (cyclophosphamide)</td>
</tr>
</tbody>
</table>

**CONCLUSION**

Cytotoxic drugs have added a new and dangerous weapon to our therapeutic armamentarium. Handled with discernment, cytotoxic drugs may arrest malignant disease, relieve pain and prolong life in comfort. Used inadvisedly, they may deprive the patient of the one thing to which he or she is entitled—a peaceful passing.

**SUMMARY**

1. The history of cytotoxic treatment has been briefly reviewed.
2. The structure and possible mode of action of the various agents have been described.
3. The applications, techniques and complications of cytotoxic treatment have been discussed.

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The use of cytotoxic drugs in the surgery of malignant disease

REFERENCES


COLEY, W. B. (1894): Treatment of Inoperable Malignant Tumors with the Toxins of Erysipelas and the Bacillus Prodigious. Transactions of the American Surgical Association, 12, 183.


