Clinical and Experimental Projects on Chemotherapy of Bladder Tumours

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SUMMARY

Our clinical and experimental experience with chemotherapy of bladder tumours is reviewed. The routes of drug administrations, drug dosages and combinations, are presented. Adjuvant radiotherapy and chemoprophylaxis of certain tumours are discussed.

S. Afr. Med. J., **48**, 631 (1974).

In spite of the fact that chemotherapy of bladder tumours was attempted at the beginning of this century in the form of topical treatment with phenol and podophyllin, it can be said that modern chemotherapy started only after the discovery that nitrogen mustard was effective in the treatment of some human tumours.

Anticancer chemotherapy became of significant value only during the past 20 years, following the preliminary screening of a large number of drugs, many of which are presently in clinical use.

The agents commonly employed can be divided into 4 main groups: alkylating agents, antimetabolites, mitosis-blocking agents and antibiotics, apart from hormones and some other drugs that cannot be included in these groups.

A more logical classification would be one that takes into account the time at which the various drugs act along the course of the reproductive cycle of the cell, namely at the S phase, during which DNA synthesis takes place; at the G1 and G2 phases, which respectively precede and follow the S phase, or in the mitotic phase. Furthermore, cycle-non-specific drugs are those that are able to act apart from the reproductive cycle, namely during the GO or interkinetic phase, at least if administered in high doses.

Chemotherapeutic agents have greatly improved the prognosis of leukaemia and malignant lymphoma, but even some solid tumours can undergo partial or total regression. Such remissions are often long-lasting and sometimes apparently permanent and definitive. However, this applies only to some tumours, such as nephroblastoma or metastatic testicular neoplasia, which display a high reproductive activity and can therefore be easily affected by drugs acting on proliferating cells.

Slowly-growing tumours such as renal, vesical and prostatic carcinomas are, however, not very sensitive to chemotherapy, especially if their reproduction rate is lower than that of normal tissues with high proliferating activity, such as the bone marrow or the intestinal crypts. The causes of the low therapeutic index, as well as the limited usefulness of chemotherapy in these tumours, are clearly understandable.

Such tumours can be affected only by using high doses for prolonged periods of time, at the risk of severe toxicity and even lethal side-effects. Medullary aplasia, stomatitis and digestive symptoms are the most frequent manifestations of drug toxicity. Many anticancer drugs have immunodepressive effects also, and may therefore give rise to a further weakening of the host defences against infection and the tumour itself.

If lack of selectivity of anticancer drugs were absolute, useful chemotherapy of slowly-growing tumours such as renal, vesical and prostatic carcinomas would be virtually impossible. There is, nevertheless, some degree of selectivity, as shown by the fact that various drugs can produce different side-effects and display a particular spectrum of activity. Selectivity may be dependent upon differences in penetration of drugs through the cell membranes, on various modalities of fixation in nuclei of different tissues, or even on different patterns of transport, activation or breakdown in various healthy and neoplastic tissues.

Particular advantages can be obtained by local or regional techniques of administration that give rise to high drug concentrations in the affected tissues, along with reduced general toxicity. Despite these methods, which cannot always be applied, chemotherapy of carcinomas still remains almost devoid of consistent efficacy.

Prostatic and vesical carcinomas are scarcely suitable for intra-arterial treatment, which usually involves the need for surgical cannulation of afferent vessels, without any possibility of limiting the escape of the drug into the systemic venous circulation. The effect of such an escape can be prevented by simultaneous haemodialysis or forced diuresis. We have shown in the dog that toxic or even lethal doses of alkylating agents (Trenimon), or antimetabolites (5-fluoro-uracil), can be administered

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Date received: 25 September 1973.

intra-arterially with only moderate leukopenia or thrombocytopenia if the animal is submitted to haemodialysis, peritoneal dialysis or intravenous infusion of mannitol.[1] The removal of protein-bound drugs can be further enhanced if parabiotic cross-dialysis with an animal of the same or of a different species is employed instead of the usual dialysing fluid.[2] These experiments have nevertheless not been confirmed in man.

A uniformly accepted schedule of treatment is still lacking even with regard to those tumours that are very sensitive to chemotherapy. A wide range of variables in the proposed treatments is indeed encountered, such as the choice of the drug, its dosage, the duration of the treatments with or without further cycles of maintenance, consolidation or reinduction, the use of a single agent or of combinations of several drugs, the adoption of continuous or of intermittent patterns of administration, etc.

The drug combinations can be employed either by the simultaneous administration of several compounds or by the consecutive use of single drugs. A recent method of sequential treatment is that of synchronisation, using drugs that act on different phases of the reproductive cycle.

It can be achieved by the administration of antimitotic agents as the initial part of the treatment, followed by drugs that act during the subsequent S phase.

Together with chemotherapy, radiation or other complementary measures can be employed with the aim of increasing the efficacy of anticancer treatment.

A synergic action can be obtained by hyperoxygenation, hyperthermia, immunotherapy, or the administration of substances such as insulin, which may increase penetration of drugs into the cells, or which may stimulate histiocytic activity or phagocytosis, such as oestrogens. To this lack of a uniformly standardised form of treatment, the continuous introduction of new drugs into the therapeutic armamentarium should be added.

Chemotherapy is therefore still in an experimental, growing phase. Any valid and lasting progress can only be achieved by controlled clinical trials. A large number of cases is necessary, with prolonged follow-ups, based on thorough knowledge of cell kinetics. This requires carefully planned and rather complex prospective studies that have only been employed in the field of bladder tumours to a small degree. Most reports deal with only a few cases, and can generally be found only within the text or tables of statistical surveys on chemotherapy in general.

It is likely that the knowledge of relatively poor responsiveness of bladder tumours to chemotherapy has discouraged most urologists, and also likely, on the other hand, that a wider clinical experience, based on controlled trials, might lead to a significant improvement of future results. There is no doubt that chemotherapy of bladder tumours is difficult and only seldom rewarding, but it would be wrong to ignore the value of certain achievements, some of which are of the greatest interest. We emphasise, in particular, the following aspects of this topic: topical therapy of papillary growths; systemic chemotherapy of inoperable carcinoma, either alone or in association with radiotherapy; and chemoprophylaxis.

INTRAVESICAL TREATMENT

The rationale for a topical treatment of papillary neoplasms with chemotherapeutic agents has already been discussed in many reports. Its practical value has been established by the results reported in most series.

The literature is particularly exhaustive with respect to Thio-tepa, a multifunctional alkylating agent. This substance has generally been employed with success, and partial or even total regressions of papillary bladder tumours have been observed.

Other drugs have also been employed for this purpose, with rather good results, such as other alkylating drugs (Epodyl—a peptidic complex of phenylalanine mustard named Peptichemio, mechlorethamine), some antimitotic agents (colchicine, podophyllin and their derivatives such as SP-I), or antibiotics (adriamycin, daunomycin, bleomycin and Mitomycin-C). Antimetabolites, such as 5-fluoro-uracil (5-FU), and methotrexate display very little effectiveness if employed by intravesical instillation.

Our results, which are not unsatisfactory, were reported in a recent article.[3] It has been confirmed that chemotherapy can lead, in some cases, to the complete disappearance of papillary bladder growths, whether single or multiple or diffuse. When regression is obtained it is particularly valuable in the case of diffuse papillomatosis, where it can prevent a mutilating and serious procedure such as cystectomy.

Partial regressions can also be very useful since they may allow the performance of conservative transurethral or surgical interventions that were impossible before. In other instances they lead to subjective improvement or to a reduction in blood loss. It should be recognised that such therapeutic endeavours are still empirical and incompletely understood. The doses have been arbitrarily selected so that the amount administered by intravesical instillation is usually equal to a daily parenteral dose or somewhat less. It is not certain that this represents the optimum dose and that lower amounts cannot be equally effective.

The usual technique is that of administering, at weekly intervals, and for a variable number of times, intravesical instillations of a given drug, diluted in an average volume of 30 ml, to be kept in the bladder for 1 - 2 hours. A change in position is also suggested, from the prone to the supine, and to the lateral positions on both sides. This step has proved unnecessary in our experience and can be safely omitted. We prefer instead to avoid any restriction of activity, so that the treatments can be given on an outpatient basis.

It should be noted that intravesical treatment is not without risk. Some cases of severe, or even lethal myelotoxicity have been described after topical administration of Thio-tepa. In addition it should be stressed that therapeutic activity is not constant, being totally absent in about one-half of treated patients, especially in those with tumours of comparatively large size or in an advanced stage.

These considerations have led us to investigate the following points:

(a) Use of less toxic drugs, different from Thio-tepa.
(b) Different ways of administration.

(c) Preliminary study of the responsiveness of a given tumour to various agents.
(d) Fare and distribution of various drugs after intravesical administration under normal and pathological conditions, with special reference to reabsorption into systemic circulation and fixation to the tumour and to normal tissues, including the deeper layers of bladder wall.

Our investigations are still in progress, but some preliminary impressions can be anticipated.

Use of Less Toxic Drugs

Drugs other than Thio-tepa can be used intravesically with some good results. The other alkylating agents employed, namely cyclophosphamide, Trenimon, Peptichemio and mechlorethamine, often produce severe symptoms of chemical cystitis. Adriamycin and daunomycin are instead well tolerated by the bladder but it should be noted that intravesical treatment with drugs other than Thio-tepa has never given rise to complete regression of bladder tumours, except in one case treated with mechlorethamine.

Different Ways of Administration

Prolonged bladder perfusions with some drugs (demecolcine, adriamycin, Peptichemio, 5-FU), either alone or in sequential association by means of a three-way catheter, were well tolerated locally and systemically. This holds true even when for several consecutive days daily doses were used that were as high as those employed for a single instillation, and even when the treatment was started soon after a transurethral procedure. No toxic effects were ever observed, due to low concentration of drugs (the daily dose being dissolved in 1 - 2 litres), so that inflammatory reaction is practically absent and reabsorption minimal, despite prolonged contact. The number of treated cases is still small, but sufficient to show that objective and even complete regressions of papillary tumours can be obtained by continuous vesical perfusion. Furthermore, this method allows synchronisation therapy without any increase of its toxic side-effects. It can also be associated with hyperthermia and local hyperoxygenation. In addition, it is possible to utilise the antimetabolites that are ineffective if employed for short contact periods.

Tumour Responsiveness to Various Agents

The observation that neoplasms, apparently identical from either a cystoscopic or a histopathological standpoint, can display a completely different response to a topical treatment, has stimulated us to study the possibility of obtaining preliminary information on the sensitivity of a given tumour to various drugs, so that the most effective one might be selected.

Heterotransplantation into the hamster cheek-pouch, followed by the measurement of growth curves of grafted tumours in groups of animals treated with various drugs is certainly a fascinating technique, but cannot be employed in clinical practice due to its length, complexity and cost.[4]

In vitro cultures of neoplastic cells in liquid media, associated with the measurement of inhibition of the uptake of tagged precursors (tritiated thymidine and leucine), are of value in detecting which substances are most effective in blocking DNA and protein synthesis. Expensive equipment is required and the technique is rather complex and therefore not amenable to wide clinical use.[5]

Another method is available, which is simpler, more rapid and can be adopted in every average laboratory. It is based on the property of active drugs to inhibit the dehydrogenase activity of tumour cells, as shown by lack of decoloration of methylene blue around the discs containing the effective drugs.[6] Unfortunately, this simple method is not constantly reproducible. Other simple methods can be employed, which detect cytotoxic effects.

According to Lunglmayr's technique,[7] a comparative cytological examination is performed, before and after an intravesical treatment. Our experience with this method has been rather unrewarding, despite some modifications. Neither differential counts with respect to the extent of cytoplasmic or nuclear changes, nor fluorescence microscopy or vital staining seem to improve the reliability of this method, as we have observed by examining a large number of smears of either exfoliative or contact cytology as well as histological sections.

According to our experience, we consider that a wide range of error cannot be avoided by using methods based on cytotoxicity, so that the criteria for evaluation are gross and of limited reliability.

We found it especially interesting to study the actual penetration of chemotherapeutic agents into the nuclei of tumour cells. This can be easily achieved with fluorescent drugs, such as adriamycin, daunomycin or other substances such as Peptichemio, which can be chemically bound to fluorescent compounds (tetracycline, fluorescein).

Autohistoradiographic methods can also be employed in order to demonstrate the entrance of tagged anticancer drugs into neoplastic cells following intravesical treatment. It should be noted, on the other hand, that evidence is lacking that the morphological demonstration of the presence of drugs in the nuclei is proof of its anticancer activity, in that particular setting.

The conclusion, at the present stage of our experience, is that the reliability of any of the single methods is limited, but that they may acquire a greater significance if their results are concurrent. We must admit, however, that no universal antimitogram is available, that can be applied as a routine in any laboratory for the study of bladder tumours.

Drug Reabsorption and Fixation

The difference between the amount of a given drug introduced into the bladder cavity and the amount that is recovered after one or two hours (by voiding and subsequent repeated vesical rinsing), can be easily established. The amount of the drug that has disappeared

is not only related to reabsorption through the bladder wall and to escape into the systemic circulation, but also to fixation onto the tissues. This can be demonstrated again by the use of fluorescent or tagged drugs after variable periods of contact. Using adriamycin we were able to observe that substantial traces of the drug were detectable not only in the tumour but also in the deep layers of the bladder wall.

The amount of Thio-tepa and other alkylating agents that disappears after intravesical instillation is instead measurable by colorimetric methods. It is rather small in the healthy bladder, but it can reach 80% or even more in cases of multiple and large tumours, as well as after transurethral or open surgical procedures. The same behaviour can be demonstrated for adriamycin, although the extent of reabsorption seems to be comparatively lower.

SYSTEMIC CHEMOTHERAPY

Systemic chemotherapy of infiltrating carcinoma of the bladder is followed by only limited success, according to the experience of several authors. The use of alkylating agents has produced no consistent results, in spite of the report by Fox,[8] that some objective regression and symptomatic relief can be obtained if very high doses of cyclophosphamide are employed.

Relatively better results can be obtained by the use of antimetabolites. Methotrexate has been employed by infusion into the internal iliac artery with some good results, although the value of this method in pelvic tumours is limited by the extensive escape of the drug into the systemic circulation, and still remains to be assessed. It cannot be denied, however, that striking results were obtained by Carbone *et al.*[9] by intra-arterial infusion of a cocktail of drugs into the hypogastric artery after ligation of all collateral branches, except the vesical ones.

A wider experience of the use of 5-FU as the only treatment, has failed to confirm the encouraging results described in the early reports. Statistical studies would even suggest that it is not superior to a placebo, at least following the protocol developed by Prout *et al.*[10] Contrariwise, the association of 5-FU to cobalt therapy has proved successful in our hands as well as in other series, not only as a pre-operative measure, but also in the treatment of inoperable cases.[11,12]

As far as antibiotics are concerned, no conclusions can be drawn regarding daunomycin, as the experience is too limited.

Adriamycin appears, however, to be one of the most promising drugs, as shown by the high number of regression—even greater than 50%—reported by Frey *et al.*[10] This is at variance with the limited success in Whitmore's[14] experience.

Objective regressions of the original tumour or of its metastases, apart from relief of pain and other distressing symptoms, can be obtained in some of the patients treated with Mitomycin-C. This antibiotic was found, on the other hand, to be the most effective drug in experiments performed on a vesical cancer transplanted in the hamster. Its toxicity in clinical use can be greatly reduced by slow intravenous drip infusion.

The preliminary favourable experience reported by Japanese work on carcinophyllin has not yet been confirmed.

Bleomycin is another drug that deserves careful consideration, in spite of the fact that reports on its use are still contradictory. Very recent information about the experience of the co-operative group on urological oncology of the European Organization for Research on the Treatment of Cancer (OERTC), shows the following data out of 30 bladder cancers treated with bleomycin: 1 interruption of therapy; 1 death; 2 total regressions; 3 partial regressions greater than 50%; 7 partial regressions of more limited magnitude; 2 subjective improvements; and 13 failures. The percentage of objective regressions was therefore equal to 40%.

Among mitostatics, vinca alkaloids are almost totally ineffective, whereas preliminary data from the OERTC urological co-operative group seem to indicate that VM-26, a new podophyllin derivative, is another promising drug, worthy of being extensively evaluated in the treatment of bladder cancer.

Hydroxyurea was also considered a promising drug. The excellent results reported in over three-quarters of their patients by workers from USA and Italy are not in keeping with the experience of other authors. At any rate, even according to the most optimistic reports, regressions have always been transient and no patient has been cured. Little is known about the value of the various drug combinations and little can be found in the literature.

Our personal experience reported in 1970[6] refers to 102 systemic treatments using various forms of chemotherapy. Follow-ups were obtained in only 90 cases, and statistical analysis was performed as a part of a more extensive study with the help of a computer. It was concluded that the percentage of remissions was relatively good, being as high as 41% of cases, although it should be stressed that in 30% such remissions were only partial and short-lived, whereas regressions greater than 50% of the original tumour size and lasting for at least one month were less than 10%.

No drug has given consistent results or shown a clear-cut superiority over other available agents. Mitomycin-C, which was among the drugs that we had more extensively employed, yielded, however, some good results within the limited range of success that can be anticipated.

In the whole group, objective regressions were exceptional in stage T4, rare in T3, relatively more frequent in stage T2. Our best results were obtained in the patients treated with 5-FU, together with cobalt teletherapy. The number of patients is comparatively high, with follow-ups longer than 3 years in two-thirds of cases. A retrospective analysis showed that the mortality rate was higher and remissions less frequent in a comparable group of patients treated by radiation alone. The higher mortality in this latter group is very likely to be related to the fact that the average dose of radiation was significantly higher, especially because many patients had been submitted to repeated cycles of cobalt therapy.

The patients in both groups, except 2, were in a very advanced stage, either T3, or mostly T4. Our results support the contention of a renal synergism between

5-FU and radiation therapy, and are in accordance with the suggestion that, in such cases, focal radiation dose can be successfully limited in a range between 4 000 and 6 000 rads. The combined treatment, 5-FU plus radiation, was followed in 7% of cases by an apparent cure, which persisted after a follow-up of at least 3 years. In another 6% the treatment brought about a partial regression which was of such extent as to allow a radical surgical or transurethral procedure in patients who were considered locally inoperable before the treatment. A partial regression of the tumour was obtained in 47% of patients.

The duration of the remissions was prolonged for at least 3 months in about half of the responsive cases. It was clearly shown that the results were much better if the patient had not been submitted to previous irradiation or surgery, where complete failures were uniformly observed.

Our further experience is substantially in agreement with our earlier results. New drugs have been recently checked and clinical trials with others, such as VM-26, are still in progress.

Peptichemio (peptidic complex of phenylalanine mustard), has yielded some successful results, both palliative and objective, among which was the regression of pulmonary metastases of a bladder cancer.

Our experience with adriamycin is not as good as that of our colleagues from Texas.[14] We have nevertheless observed some good results. The almost complete regression of an anaplastic carcinoma in a young patient permitted the radical removal of a bladder that was initially fixed to the pelvic wall.

No demonstrable results were obtained in any of our 3 patients treated with bleomycin.

Controlled clinical trials are presently in progress, including sequential polychemotherapy with VM-26, adriamycin, 5-FU, Mitomycin-C, and Peptichemio. This seems to be a very useful treatment, but it is too early for a definite opinion. It can be concluded that, in a difficult field such as the treatment of advanced bladder cancer, the best results can be obtained by the use of all available therapeutic measures.

Surgery and radiation still remain, of course, of primary importance. Chemotherapy, on the other hand, can be combined with both and improve the results of irradiation. It may offer valuable results when other methods of therapy cannot be successfully employed, and remains the only treatment available in patients with distant metastases. It should be remembered that in such cases, as well as with inoperable patients, chemotherapy can offer, if not a permanent cure, at least some valuable palliation and temporary regression.

In view of the fact that in the latter category the aim of chemotherapy is merely to achieve palliation, (permanent cures being impossible at the present time), it is not advisable to push chemotherapy to the highest possible doses which jeopardise life in poor-risk patients, and produce severe and distressing toxic symptoms. This is contrary to the goal aimed at. Subjective remission can usually be obtained with relatively low doses that do not lead to the level of toxicity that should necessarily be reached in other fields of oncology, where a curative action can be anticipated.

CHEMOPROPHYLAXIS

The most useful indication for the use of chemotherapeutic agents in urology can probably be found in prophylaxis after adequate treatment of non-infiltrating papillary bladder tumours. This has been clearly shown in several reports dealing with Thio-tepa, mechlorethamine, podophyllin derivatives (all used intravesically), and confirmed in our own experience during the past 8 years.[15]

Successful treatment of papillary bladder tumours by transurethral or open surgery was followed in our control group by a rather high recurrence rate (60% within two years). We have used various prophylactic measures: addition of Trenimon or other anticancer drugs to the irrigating fluid during and after transurethral resection; intravesical instillation of Thio-tepa, 5-FU, daunomycin, adriamycin, Peptichemio or SP-1; administration of various drugs intravenously (5-FU, Mitomycin-C) or orally (cyclophosphamide, hydroxyurea, methotrexate, procarbazine, podophyllin glucosides, ascorbic acid and others).

The oral drugs that were more frequently employed were administered once a week in a single dose (hydroxyurea: 2 g, procarbazine, paraphyllin 200 mg, or methotrexate 10 mg). Many patients were lost to follow-up, and up to May 1971 only 134 could be evaluated, 94 of whom were given prophylactic treatment and 40 controls were given no treatment.

Intravesical prophylactic treatment has given the best results. Recurrences were observed only 3 times out of 14 patients treated with Thio-tepa, administered initially at weekly, then at monthly intervals. Only 1 recurrence was discovered out of 7 patients treated with daunomycin, and 2 recurrences occurred in 8 patients given adriamycin, both by intravesical instillation.

The prophylactic use of antiblastic drugs added to irrigation fluids is not without some efficacy, though the action is less evident. When intravesical prophylactic instillations are interrupted, recurrences often appear, so that it is advisable to continue treatment for one or even two years. Oral drugs were shown to produce valuable preventive effects and their use is particularly advocated in cases where intravesical instillations are impractical or impossible. They can also be employed for maintenance treatment after the first months of intravesical medication.

Oral treatment is limited by the low urinary excretion of most substances. The alkylating agents, in particular, are very poorly excreted by the kidneys. In keeping with the theory, a number of drugs that reach relatively high urinary concentration display a good prophylactic action. This is shown by the low number of recurrences in patients treated with hydroxyurea (80 - 90% of the administered dose excreted in the urine in 24 hours)—3 recurrences out of 13; procarbazine (70% urinary excretion)—1 recurrence out of 11; methotrexate (75% excretion)—1 recurrence out of 14.

Our experience with other drugs, administered either orally or intravenously, was inconclusive. No recurrences were observed in 4 patients with superficial muscle invasion, where topical Thio-tepa was supplemented by intravenous Mitomycin-C and 5-FU. Follow-up exceeded

5 years in one case. Some toxicity cannot be avoided with this combined treatment, but we believe that such vigorous treatment may be justified in cases where some doubt is present in the surgeon's mind about complete removal of the tumour. In all the other cases where only one drug was employed, no significant toxic effect was ever observed, apart from mild gastric disturbances after intake of Natulan. From our experience during the past year (the analysis is not yet complete), it can be anticipated that the established trends will be confirmed, in spite of a somewhat higher percentage of recurrences occurring in the patients given oral hydroxyurea.

It is likely that this reflects a different selection of cases, since we have encountered a higher number of patients with marked urothelial instability, as shown by a tendency towards multiple or diffuse papillomatosis with a past history of frequent recurrence. Patients showing this pattern of urothelial disease should probably be placed in a different category from the more usual type of papilloma, and be treated with a higher dose of oral chemoprophylactic agents, e.g. 2 g of hydroxyurea two to three times a week.

The theoretical background for oral prophylactic treatment appears to be sound, not only because a drug that reaches a higher urinary concentration shortly after oral administration can display, without a need for urethral catheterisation, an action that is similar to that of a drug directly introduced into the bladder, but also because it acts on the whole urothelial surface, and not only on the bladder mucosa. It should be emphasised, in addition, that the toxicity of hydroxyurea is minimal, even for protracted treatments using high daily doses.

Our preliminary experience with chemoprophylaxis for papillary bladder tumours is very encouraging, but greater experience is needed. We have started a polycentric controlled randomised trial and would be very grateful to any colleague who would join this programme, so that more valuable cumulative experience may be obtained.

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