ABSTRACT

Because of the association between the incorporation of 5-fluorouracil (FUra) into RNA and cytotoxicity, uridine was examined for potential selective reduction of host toxicity. Male BALB/c × DBA/2 F1 mice (tumor free or bearing advanced colon tumor 26) were used. Two uridine schedules (each beginning 2 hr after FUra) have been successful: (a) uridine at 800 mg/kg every 2 hr for three doses followed 18 hr later by uridine at 800 mg/kg every 2 hr for four doses; or (b) two doses of uridine at 3500 mg/kg separated by an 18-hr interval. With two doses of uridine at 3500 mg/kg, the 50% lethal dose for a single dose of FUra in tumor-free mice was increased 68% from 190 to 320 mg/kg. White blood cell levels were depressed 67% after a single dose of FUra at 200 mg/kg, whereas white blood cells were depressed by only 39% when the same dose of FUra was followed by uridine rescue.

In tumor-bearing mice, uridine rescue reduced FUra-induced host toxicity without significant loss of antitumor activity. Even more striking results were obtained with a combination containing N-(phosphonacetyl)-L-aspartate (200 mg/kg) and 6-methylmercaptopurine riboside (25 mg/kg), administered 24 hr before FUra. In this drug combination, the maximum tolerated dose of FUra is 40 mg/kg on a weekly schedule. With uridine rescue, FUra can be doubled to 80 mg/kg without increasing toxicity, resulting in significantly improved antitumor activity. Examination of the effect of uridine rescue on the incorporation of FUra into RNA and the subsequent recovery from inhibition of DNA synthesis in bone marrow versus tumor revealed that the uridine rescue schedule resulted in relatively faster clearance of FUra from RNA of both tissues but a striking enhancement of the rate of recovery of DNA synthesis only in the bone marrow.

INTRODUCTION

The successful utilization of biochemical modulation to improve the selectivity of a known drug, i.e., to preserve or enhance its tumor cell kill while minimizing its toxicity to sensitive normal tissues, depends on at least some knowledge of the biochemical determinants of the cytotoxicity of the drug. FUra is known to interfere with cellular metabolism through two different pathways. One involves its conversion to FdUMP, which inhibits thymidylate synthetase, leading to suppression of DNA synthesis. The second mechanism of action for FUra, via its incorporation as 5-fluorouridine 5'-monophosphate into RNA in place of UMP, can result in a variety of adverse effects on key cellular mechanisms involving RNA function and processing (14, 15, 18, 23, 40–43).

Our previous studies (24–27, 33, 34, 37–39) and those of others (3, 6, 7, 16, 18, 20–22) have led to an appreciation of the correlation between the incorporation of FUra into RNA and cytotoxic activity and provided the rationale for the selection of the natural competitive pyrimidine, uridine, as a potential rescue agent. In these earlier studies, exogenous uridine was administered both shortly before (0.5 hr) and after (0.5 hr) FUra administration, to see if normal tissue FUra toxicity could be prevented without concomitantly causing significant reversal of antitumor activity. A differential susceptibility of tumor and normal host tissue was observed in these murine studies (24–27).

In some of the latter studies, PALA and MMPR were used as metabolic modulators to increase the activity of FUra. PALA, an inhibitor of de novo pyrimidine synthesis, can increase the incorporation of FUra into RNA (34, 37, 38) by lowering the levels of the competing normal pyrimidine nucleotide of FUra, UTP. In many cells, the de novo pyrimidine pathway enzyme, OMP PRPP Tase, is believed to be the major enzyme responsible for converting FUra to its active nucleotides. 5-Phosphoribosyl-1-pyrophosphate, a necessary substrate for the conversion of FUra by OMP PRPP Tase to its active nucleotide, is also utilized in de novo purine biosynthesis. It has been shown that MMPR, by inhibiting de novo purine synthesis, can increase the level of 5-phosphoribosyl-1-pyrophosphate, allowing for greater conversion of FUra by OMP PRPP Tase to its active nucleotide, and that PALA and MMPR given together can increase the incorporation of FUra into RNA, resulting in a marked potentiation of FUra cytotoxicity, including its antican-
MATERIALS AND METHODS

Murine Tumor System. Colon tumor 26 was obtained from Dr. Joseph Mayo at the National Cancer Institute and maintained in our laboratory by s.c. brei transplant in BALB/c mice also supplied by Dr. Mayo. For experimentation, 0.3 ml of a 5% tumor brei (in modified Dulbecco’s phosphate-buffered saline at pH 7.2) prepared from 4 to 6 tumors was transplanted s.c. into 2- to 3-month-old male CD2F1 mice. Approximately 7 to 9 days later, when tumors were approaching 100-mg size, they were measured and the animals were distributed among the experimental groups of 10 mice each so that animals carrying approximately equal-sized tumors were represented in each group.

Drug and Chemicals. FURA, PALA, MMPR, and uridine were obtained from the Drug Synthesis and Chemistry Branch of the National Cancer Institute. FURA, MMPR, and uridine were dissolved in 0.85% NaCl solution immediately before use and were administered i.p. so that the desired dose was contained in 0.1 ml/10 g of mouse weight. PALA was dissolved in 0.85% NaCl solution; the pH was adjusted to 7.2 to 7.5 with 1 N NaOH before adjustment to final volume. PALA, too, was administered i.p. in a volume of 0.1 ml/10 g of mouse weight. [6-3H]FURA was obtained from Moravek Biochemicals, Inc., Brea, Calif.; [32P] from New England Nuclear, Boston, Mass.; cesium sulfate from J. T. Baker Chemical Co., Phillipsburg, N. J., and Sigma Chemical Co., St. Louis, Mo.

Tumor Measurements. Two axes of the tumor (the longest axis L and the shortest axis W) were measured with the aid of a vernier caliper. Tumor weight was estimated according to the formula:

\[
tumor \text{ weight (mg)} = L \ (mm) \times W \ (mm)^2 / 2.
\]

Statistical Evaluation. Student’s t test was used for statistical evaluation of differences in mean tumor size between groups of treated mice. Differences between groups with a statistical probability of 0.05 or less were considered significant.

Toxicity Measurements. Animal body weights were recorded immediately before and biweekly after the initiation of treatment. Weight change was calculated as a percentage of the initial body weight of the animals.

WBC levels in tail vein blood were determined electronically with a Fisher cell counter.

Incorporation of Radiolabeled Precursors into RNA and DNA. Male CD2F1 mice bearing transplanted colon 26 tumors received PALA (200 mg/kg) and MMPR (25 mg/kg) 24 hr prior to [3H]FURA (80 mg/kg containing 12 mCi [6-3H]FURA per kg). The animals were divided into 2 groups. One group received no further treatment. The second group received the uridine rescue schedule: uridine at 800 mg/kg every 2 hr for 4 doses, beginning 2 hr after FURA, followed by uridine at 800 mg/kg every 2 hr for 4 doses starting 24 hr after FURA. At various intervals thereafter, mice from both groups received [32P] (10 mCi/kg) 1 hr prior to sacrifice. The animals were sacrificed by cervical dislocation, and the tumors and bone marrow were removed. Bone marrow was collected by flushing the femurs with ice-cold 0.85% NaCl solution.

The tumor was homogenized in TNE buffer containing 1% Triton X-100. The homogenate was further diluted with TNE buffer, NaCl, and sodium dodecyl sulfate, sonicated, treated with Pronase (0.2 mg/ml, digested for 2 hr at 37°C), and extracted with chloroform:isoamyl alcohol (24:1, v/v). Samples of this material were precipitated with trichloroacetic acid and collected on glass fiber filters to determine total acid-precipitable radioactivity. Other aliquots were first treated with alkali (0.4 N NaOH, 90 to 120 min, 37°C) prior to trichloroacetic acid precipitation to determine alkali-stable, acid-precipitable radioactivity. The differences between the total and alkali-stable radioactivity were assumed to represent RNA. DNA was determined by the Burton modification of the diphenylamine color reaction (5).

Nucleic acids from bone marrow were analyzed by cesium sulfate equilibrium density centrifugation. The bone marrow pellet was suspended in TNE buffer and sodium dodecyl sulfate, sonicated, and extracted twice with phenol:cresol (7:1, v/v) and once with chloroform:isoamyl alcohol. Nucleic acids were recovered by ethanol precipitation. The precipitated material was dissolved in 0.5 ml of 50% formamide in 5 mM EDTA, heated to 85°C for 20 min, and then quick chilled to denature the nucleic acids. The sample was mixed with 4.5 ml of water and 5.5 ml of a saturated cesium sulfate solution containing 5 mM EDTA. Centrifugation was for 60 to 66 hr in a Beckman Ti 50 rotor at 45,000 rpm and 20°C. The gradients were fractionated from the bottom, and the RNA and DNA regions were located by refractive index and A260. The amount of DNA was estimated from the A260. Nucleic acids in individual fractions were then precipitated with trichloroacetic acid.

Plasma Uridine Measurements. Normal male CD2F1 mice received a single dose of uridine (3500 mg/kg) i.p. At various times, the animals were anesthetized with ether, and blood was collected in heparinized capillary tubes. Plasma was deproteinized with perchloric acid. Perchlorate was removed with KH2PO4. Uridine levels were measured by high-pressure liquid chromatography using a Dupont 850 system and a Spectra Physics calculating integrator. Assay conditions were 0.01 M KH2PO4·K2HPO4 buffer, pH 6.0, and a Dupont C8-column.

RESULTS

The potential for amelioration of FURA-induced toxicity first was investigated in normal 2- to 3-month-old male CD2F1 mice. In a representative experiment, 2 groups of 7 mice each were treated with a single course of FURA at 200 mg/kg (i.e., the approximate 50% lethal dose). One of these groups received no further treatment, while the other received a rescue schedule beginning 2 hr after FURA and consisting of uridine at 800 mg/kg every 2 hr for 3 doses followed after an interval of 19 hr with uridine at 800 mg/kg every 2 hr for 4 doses. WBC levels were measured periodically after treatment, and the results obtained at 7 days after FURA (nadir levels) are presented in Table 1 for the individual mice in both groups along with the ultimate fate of each mouse at 30 days after treatment.

Although the range of WBC level was fairly large before treatment (9100 to 21,000/cu mm), the mean level was quite similar in both groups (15,029 and 15,614/cu mm). One week after treatment with FURA alone, WBC levels were depressed significantly.
Selective rescue of FUra-induced host toxicity with uridine

Table 2

<table>
<thead>
<tr>
<th>Group</th>
<th>Treatment</th>
<th>% of body wt change</th>
<th>Dead/total</th>
<th>Av. tumor wt (mg)</th>
<th>Statistical probability</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>0.85% NaCl solution</td>
<td>- 19</td>
<td>4/10</td>
<td>3008</td>
<td></td>
</tr>
<tr>
<td>2</td>
<td>FUra (100 mg/kg)</td>
<td>- 19</td>
<td>3/10</td>
<td>1437</td>
<td>0.09</td>
</tr>
<tr>
<td>3</td>
<td>FUra (125 mg/kg) → 2 hr uridine (3500 mg/kg) → 18 hr uridine (3500 mg/kg)</td>
<td>- 5</td>
<td>0/10</td>
<td>1038</td>
<td></td>
</tr>
</tbody>
</table>

In Table 2, a dose of 100 mg of FUra per kg alone (Group 2), a maximally tolerated weekly dose in nontumored mice, appears somewhat toxic in colon 26 tumor-bearing mice, as evidenced by a 30% mortality at 5 days after the third course of treatment despite a substantial inhibition of tumor growth. In contrast, even with an elevated dose of FUra (125 mg/kg) plus the uridine rescue regimen (Group 3), there was a marked reduction in body weight loss from -19 to -5% (note that the weight loss seen here is a characteristic feature in mice carrying colon tumor 26, as may be seen in the 0.85% NaCl solution control group, and is not a reflection of drug toxicity) and no mortality, and the antitumor activity was increased substantially. These findings have also been confirmed in another laboratory.

The most striking selective host rescue by uridine to date has been achieved in the colon 26 tumor system with a FUra-containing combination of anticancer drugs. PALA at 200 mg/kg and MMPR at 25 mg/kg have been added to FUra to form this combination because each of these agents has been shown to increase the activation of FUra leading to increased incorporation into RNA (3, 6, 7, 20–22, 25, 26, 34, 38). In the presence of these 2 metabolic modulators (at the indicated doses), the maximum tolerated dose of FUra in tumor-bearing mice is 40 mg/kg. If the FUra dose in this 3-drug combination...
82. 20, and 110 mg, respectively) were used. All injections were given i.p. once a week for 3 doses. Observations were recorded 4 days after the third course of high-dose FUra-containing chemotherapy.

When one of either of the 2 uridine rescue schedules was administered following each dose of FUra. Most importantly, in each of the 4 experiments, the increased activity of the high-dose FUra combination was manifest in significantly greater antitumor activity in the uridine rescue groups. These data demonstrate clearly that an improved therapeutic index can be attained in a particular tumor-host system with uridine "rescue" of high-dose FUra-containing chemotherapy.

FUra cytotoxicity probably is caused by the disruption of RNA metabolism due to the presence of FUra in the RNA which results ultimately in the inhibition of DNA synthesis (34, 37, 38). Therefore, we attempted to correlate the enhanced selectivity observed in vivo with the uridine rescue with the clearance of FUra from RNA and the recovery of DNA synthesis from FUra-induced inhibition in tumor tissue versus bone marrow. A large group of tumor-bearing CD2F1 mice were given PALA (200 mg/kg) and MMPR (25 mg/kg) 24 hr prior to the augmented chemotherapeutic dose of FUra containing [3H]FUra (80 mg/kg, 12 mCi/kg). One-half of the animals then received a uridine rescue schedule (uridine at 800 mg/kg every 2 hr for 3 doses, starting 2 hr after FUra, followed by uridine at 800 mg/kg every 2 hr for 4 doses, starting 24 hr after FUra). In these experiments, the day the animals received the [3H]FUra was designated "Day 0."

At various times thereafter, 4 animals from each group received 32P 1 hr prior to sacrifice. The tumors and bone marrow were removed, and the level of FUra in RNA (and bone marrow DNA) and the incorporation of 32P into DNA were determined. Animals which received no treatment served as controls for DNA synthesis. Data for clearance of FUra from tumor and bone marrow RNA and bone marrow DNA are presented in Chart 2. Two hr after FUra administration is taken as the starting point since both groups are allowed 2 hr of exposure to FUra before any uridine is administered. In both tissues, the level of FUra in RNA was increased at 8 hr as compared to 2 hr in the absence of uridine treatment. The uridine rescue schedule strongly competes with this continued incorporation such that in both tissues from these animals the level of FUra RNA is reduced at 8 hr as compared to 2 hr. Additionally, we noted a significant level of FUra apparently incorporated into bone marrow DNA. In some experiments, as much as 60% of the acid-insoluble radioactivity derived from [3H]FUra appears to be in bone marrow DNA. The presence of [3H]FdUMP in bone marrow DNA was confirmed by alkaline digestion, cesium sulfate equilibrium density centrifugation, and nucleotide analysis. The uridine rescue also reduces the levels of putative bone marrow FUra DNA. At later time points, the differences in (FUra) RNA levels are less striking, indicating that the most pronounced effects of uridine are during the first 2 to 48 hr.

This faster clearance of (FUra) RNA from marrow results in a striking enhancement of the rate of recovery of DNA synthesis. In both the uridine- and nonrescued animals, the PALA-MMPR-FUra schedule causes both severe inhibition of bone marrow DNA synthesis (Chart 3A) and loss of bone marrow itself as judged by the reduction in bone marrow DNA (Chart 3B) during the first 48 hr after the initial FUra therapy. After 48 hr of exposure to FUra, the bone marrow from uridine-treated animals begins to recover its ability to incorporate 32P into DNA. By Day 3 (72 hr after FUra), the uridine-rescued bone marrow have DNA synthesis rates comparable to or

\[ \text{Table 3} \]

<table>
<thead>
<tr>
<th>Experiment</th>
<th>Group</th>
<th>Treatment</th>
<th>% of wt change</th>
<th>Antitumor activity</th>
</tr>
</thead>
<tbody>
<tr>
<td>136</td>
<td>1 PALA (200 mg/kg) + MMPR (25 mg/kg)</td>
<td>24 hr FUra (40 mg/kg)</td>
<td>-11</td>
<td>648 0.03</td>
</tr>
<tr>
<td>139</td>
<td>2 PALA (200 mg/kg) + MMPR (25 mg/kg)</td>
<td>24 hr FUra (80 mg/kg)</td>
<td>-25</td>
<td>538 0.01</td>
</tr>
<tr>
<td>140</td>
<td>3 PALA (200 mg/kg) + MMPR (25 mg/kg)</td>
<td>24 hr FUra (80 mg/kg)</td>
<td>-6</td>
<td>45</td>
</tr>
<tr>
<td>142</td>
<td>2 PALA (200 mg/kg) + MMPR (25 mg/kg)</td>
<td>24 hr FUra (80 mg/kg)</td>
<td>-6</td>
<td>116 0.0002</td>
</tr>
</tbody>
</table>

\footnote{Student’s t test. Significance: p = 0.05 or less.}

\footnote{Uridine rescue was uridine at 800 mg/kg every 2 hr for 3 doses followed 18 hr later by uridine at 800 mg/kg every 2 hr for 4 doses.}

\footnote{Uridine rescue was uridine at 1000 mg/kg every 2 hr for 3 doses followed 18 hr later by uridine at 1000 mg/kg every 2 hr for 4 doses.}

is increased to 80 mg/kg, 100% mortality ensues after 2 weekly courses of treatment (Table 3, Exp. 140, Group 2). However, in each of 4 experiments presented in Table 3, we were able to administer 3 weekly courses of the 3-drug combination with the otherwise toxic dose of FUra (80 mg/kg), when one of either of the 2 uridine rescue schedules was administered following each dose of FUra. Most importantly, in each of the 4 experiments, the increased activity of the high-dose FUra combination was manifest in significantly greater antitumor activity in the uridine rescue groups. These data demonstrate clearly that an improved therapeutic index can be attained in a particular tumor-host system with uridine "rescue" of "high-dose" FUra-containing chemotherapy.

\[ 3 \text{ PALA (200 mg/kg) + MMPR (25 mg/kg) — } \text{FUra (80 mg/kg) — } \text{uridine} \]
greater than those in the control animals. An "overshoot" of from 200% of control or more was not uncommon by Day 5 or 6. In contrast, the bone marrow from animals which did not receive the uridine rescue incorporate significantly less \(^{32}P\) into DNA than uridine-rescued animals until Day 6 (the day these animals would receive PALA and MMPR for a second course of therapy). The enhanced DNA synthesis observed with uridine rescue is reflected in the increased recovery of DNA from the bone marrow of these animals (Chart 3B). We estimate that this FUra therapy results in an 80 to 90% loss of bone marrow as judged by the recovery of DNA (Chart 3B).

Thus, while bone marrow cells from both groups have the same rate of \(^{32}P\)DNA synthesis at Day 6, the nonrescued bone marrow contains far fewer cells. The faster recovery of the uridine-rescued animals allows replacement of the bone marrow to near pretreatment levels. The nonrescued bone marrow remains 80 to 90% depleted at the start of the second course of therapy. This undoubtedly is a major cause of the severe mortality which begins within 1 day after the second dose of FUra.

In contrast, recovery of DNA synthesis in the tumors was variable (Chart 4). In some transplants recovery was complete by Day 6, even in the absence of uridine, while in others it was not. However, in contrast to the effect in bone marrow, in only one of 6 experiments was the recovery of tumor DNA synthesis enhanced by the addition of the uridine rescue schedule.

**DISCUSSION**

Because of the association between the incorporation of FUra into RNA and cytotoxicity, we have utilized its natural competitor, uridine, as a potential tool for biochemical modulation with the goal of selective reduction of host toxicity. In these studies, initiation of uridine rescue was delayed for 2 hr after the administration of FUra to insure that FUra had been completely eliminated from the plasma (mean plasma t\(_{1/2}\) of FUra, 10 to 20 min) (9, 10, 12, 36). Under these conditions, uridine does not interfere with the normal catabolic degradation of the bulk of the administered dose of FUra.

Two uridine rescue schedules have reduced FUra-associated host toxicity in CD2F\(_1\) mice. In the first schedule, uridine...
was administered at 800 mg/kg every 2 hr for 3 doses followed
18 hr later by uridine at 800 mg/kg every 2 hr for 4 doses.
The second schedule consisted of 2 doses of uridine at 3500
mg/kg separated by an 18-hr time interval. High-pressure
liquid chromatographic analysis of uridine levels shows that,
with both of these schedules that are successful for ameliora-
tion of FUra host toxicity, plasma uridine is elevated during a
period of from 6 to 8 hr on the day of FUra administration
and again on the following day. Preliminary experiments indicate
that UTP pools can be expanded only to a maximum of 2- to 3-
fold in vivo regardless of the dose of uridine administered and
that this increase is achieved at approximately a 1 nm level of
plasma uridine (data not presented). Therapeutic results indi-
cate that either high-dose bolus uridine (3500 mg/kg) or
repeated administration of lower doses (800 mg/kg every 2 hr
for 4 doses) can produce similar protection against FUra tox-
icity. Therefore, it appears that the maintenance of elevated
plasma uridine (i.e., millimolar levels) for extended periods is
the critical factor rather than the peak plasma uridine level
achieved following administration.

Using the 3500-mg/kg uridine rescue schedule, the 50%
lethal dose for a single dose of FUra in tumor-free mice was
increased 68% from 190 to 320 mg/kg. Amelioration of FUra
toxicity was associated with increased levels of peripheral WBC
in uridine-treated mice (Table 1) and with more rapid recovery
from FUra-induced inhibition of DNA synthesis in bone marrow
from uridine-treated mice (Chart 3).

In tumor-bearing mice, the uridine rescue permitted safe
elevation of the dose of FUra which resulted in both slightly
improved antitumor activity and substantially reduced host
toxicity (Table 2). The most striking therapeutic results to date
were obtained with a chemotherapeutic combination containing
FUra plus PALA and MPPR. In this drug combination with
PALA at 200 mg/kg and MPPR at 25 mg/kg, the maximum
tolerated dose of FUra was 40 mg/kg on a weekly schedule.
However, in 4 separate experiments (Table 3), we were able to
double the dose of FUra to 80 mg/kg without increased toxicity
when a uridine rescue regimen was added, and in each exper-
iment, this resulted in significantly improved antitumor activity.

Although uridine was selected because of its ability to in-
crease nucleotide pools, the exact biochemical mechanism of
the rescue is not yet understood. The rescue schedule used
here would not be expected to compete with the initial incor-
poration of FUra into RNA, the majority of which takes place in
the first 2 hr after FUra administration. However, once 5-
fluorouridine 5'-triphosphate has been incorporated into RNA,
high intracellular concentrations of UTP may effectively com-
pete with 5-fluorouridine 5'-triphosphate derived from degra-
dation of FUra-containing RNA during continuing RNA synthe-
sis. Faster replacement of (FUra) RNA with normal RNA
would not be expected to compete with the initial incor-
poration of FUra into RNA, the majority of which takes place in
the first 2 hr after FUra administration. However, once 5-
fluorouridine 5'-triphosphate has been incorporated into RNA,
high intracellular concentrations of UTP may effectively com-
pete with 5-fluorouridine 5'-triphosphate derived from degra-
dation of FUra-containing RNA during continuing RNA synthe-
sis. Faster replacement of (FUra) RNA with normal RNA
would be expected to allow faster recovery from FUra damage.

Indeed, the uridine rescue did not affect the maximum inhibition
of DNA synthesis produced by FUra in bone marrow or the loss of
bone marrow as measured by the amount of DNA (and protein)
recovered. It did, however, lead to significantly improved
recovery of bone marrow [32P]DNA synthesis and total bone
marrow DNA, differences that are not seen until 2 to 3
days after the end of the uridine rescue.

Another possibility is that the administration of excess uridine
may lead to a marked expansion of the dUMP pool. It has been
reported that under in vivo conditions, expansions of the dUMP

pool can lead to reversal of FUra inhibition of thymidylate
synthesis by fluorodeoxyuridine (FdUMP). (30) Moreover, un-
der in vivo conditions, recovery of thymidylate synthesis after
FUra is reported to result from both a fall in intracellular levels
of the inhibitor, FdUMP, and accumulation of its competitive
substrate, dUMP (29–30). Since inhibition of thymidylate syn-
thetase by FdUMP is essentially irreversible, recovery from
FUra must require new enzyme synthesis; the latter could be
protected from residual FdUMP by competition from the ur-
dine-induced high intracellular levels of dUMP.

The significant feature of the uridine rescue demonstrated in
the present study is the degree of selectivity between tumor
and host tissues that can be attained by proper dose and
scheduling of FUra and uridine. The basis for this selectivity
remains to be determined, and it is likely that quantitative
biochemical differences between cancer and normal host tis-
ues will be uncovered. It is known, for example, that tissues
differ in reparative activity (2, 8, 32), and these differences
include the ability of cells to degrade FUra-altered RNA mole-
cules (2).

The clinical utility of chemotherapeutic rescue techniques
remains to be clearly established. The possibility that the
clinical therapeutic index of methotrexate has been improved
by leucovorin "rescue" of high-dose methotrexate still is contro-
versial. More recently, enhanced clinical efficacy of using
sequential methotrexate and FUra followed by leucovorin "res-
cue" has been reported (13, 39, but these results too are pre-
liminary.

At present, the efficacy of FUra chemotherapy in humans is
limited by the small differential in cytotoxic susceptibility be-
tween normal and neoplastic tissue. Thus, it has been neces-
sary to administer FUra in suboptimal clinical doses in order to
avoid serious host toxicity. Previous attempts to reduce FUra-
associated toxicity with allopurinal (11, 35) or pyrazofurin (17)
have resulted in reversal of the antitumor activity of FUra.
However, with uridine rescue, it appears to be possible to
utilize increased FUra dosage with attendant beneficial therapeu-
tic effect. This would be of special value in current clinically
important FUra-containing chemotherapeutic combinations
(e.g., CMF: cytoxan + methotrexate + FUra) where host
toxicity is reportedly a limiting factor. The ability to reduce
the host toxicity associated with as important a clinical agent as
FUra by uridine without loss of anti-tumor effect would be a
significant clinical advance deserving of high-priority clinical
attention.

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