**In Vivo Inhibition of the Pyrimidine de Novo Enzyme Dihydroorotic Acid Dehydrogenase by Brequinar Sodium (DUP-785; NSC 368390) in Mice and Patients**


**ABSTRACT**

Little is known about the in vivo effects of inhibition of the mitochondrial pyrimidine de novo synthesis enzyme dihydroorotic acid dehydrogenase (DHO-DH). In mice a new inhibitor of DHO-DH, Brequinar sodium (DUP-785, NSC 368390) depleted the plasma uridine concentration to 40% within 2 h, followed by a small rebound after 7–9 days. The drug was subsequently evaluated in a Phase I clinical trial, during which it was possible to follow its biochemical effects in 24 patients (27 courses). In addition to the measurement of plasma uridine concentrations, we also measured in lymphocytes of 9 patients (10 courses) the duration of DHO-DH inhibition. Brequinar sodium was administered every 3 weeks as an i.v. infusion at dose levels of 15–2250 mg/m². The biochemical effects were studied following the first administration of the drug. In sonicated extracts of lymphocytes from 7 healthy volunteers the activity of DHO-DH varied from 2.0 to 3.9 nmol/h per 10⁷ cells, while in the lymphocytes of 9 patients obtained immediately before treatment this value was between 0.5 and 4.8 nmol/h per 10⁷ cells. Within 15 min of drug administration DHO-DH activity was not detectable and was still low up to 1 week later. Duration of the inhibition appeared to be related to the extent of clinical toxicity, e.g., myelosuppression, nausea, vomiting, diarrhea, and mucositis. Severe lymphopenia was observed in patients receiving Brequinar sodium at the maximum tolerated dose. At dose levels of ≥600 mg/m², uridine depletion (40–85%) was observed between 6 h and 4 days, followed by a rebound of 160–350% after 4–7 days. The extent of the depletion and of the accompanying rebound of uridine levels and the extent and duration of DHO-DH inhibition in the individual patients could be partially associated with drug toxicity in these patients. This is the first report describing biological effects of DHO-DH inhibition in humans in relation to the degree and duration of inhibition of this enzyme.

**INTRODUCTION**

DHO-DH catalyzes the oxidation of L-DHO to orotic acid, is the only enzyme of the pyrimidine de novo synthesis pathway to be found in mitochondria (1, 2), and is located at the outer side of the inner membrane (1, 3). The role of DHO-DH in the supply of tissue pyrimidine nucleotides has only been investigated to a limited extent, although it has been postulated that it may be rate limiting in the synthesis of UMP (2, 4). Insight into the role of other enzymes involved in pyrimidine metabolism was achieved by the study of a deficiency of the next enzyme, OPRT, which is associated with certain hematological disorders (5, 6), and by the in vivo and in vitro use of specific inhibitors, for example, PALA, which is a very potent inhibitor of ATC (7–12).

Brequinar sodium is a novel very potent inhibitor of DHO-DH, with a Kᵢ of 20 nm in L1210 cells (13) and 100 nm in rat liver mitochondria (14). Brequinar sodium was the first 4-quinoline carboxylic acid which showed significant activity against experimental tumors (15) and was therefore selected for clinical Phase I investigations (16–18). In vivo, the inhibition of the de novo synthesis pathway by Brequinar sodium led to a depletion of pyrimidine nucleotides (13, 19), blocking RNA and DNA synthesis (14), leading to an accumulation of cells in the S-phase (19) and arrest of cell growth. Cell growth inhibition could be prevented and reversed by addition of the pyrimidine nucleosides, uridine and cytidine, at 1 mM (14, 19) but not by thymidine and deoxyxycytidine (19). Because of the depletion of pyrimidine nucleotides, we hypothesized that pyrimidine nucleosides might also be depleted, since the de novo synthesis pathway is one of the sources for the synthesis of uridine (20).

Furthermore, it had been observed that inhibitors of the de novo synthesis pathway, such as PALA and pyrazofurin, caused an in vivo decrease in the plasma levels of pyrimidine nucleosides in humans, mice, and rats (8, 9, 11, 21).

A therapeutic dose of Brequinar sodium decreased plasma uridine in mice as early as 1 h after drug administration. Therefore we measured plasma uridine in patients in whom plasma pharmacokinetic studies of Brequinar sodium were being performed. Peripheral lymphocytes were selected for assessment of the extent and duration of DHO-DH inhibition after drug administration. The results were related to the toxic effects observed in the individual patients.

**MATERIALS AND METHODS**

Materials. Brequinar sodium (DUP-785, NSC 368390) was synthesized and obtained from the Medicinal Chemistry Section, DuPont Pharmaceuticals (Wilmington, DE). It was formulated as a 10 mg/ml solution in saline. A Lymphoprep preparation for the isolation of lymphocytes was obtained from Pharmacia (Uppsala, Sweden). Uridine, L-dihydropyrimidinol, orotic acid, and other pyrimidine compounds were obtained from Sigma (St. Louis, MO). Other compounds were of analytical grade quality.

Mice. BALB/c mice obtained from the animal breeding station "Proefdieren bedrijf TNO" (Zeist, the Netherlands) were used between 3 and 6 months of age and were randomized in a control group and one receiving 50 mg/kg Brequinar sodium i.p. Blood (about 100 μl) was sampled in heparinized tubes during the first day at the same time points in all mice, via the tail vein. All following samples were taken at the same time of day. After centrifugation (10 min at 4000 × g at 4°C), the supernatant was deproteinized with perchloric acid and subsequently neutralized, as described (22). Statistical evaluation was performed with Student’s t test for paired and unpaired samples.

Patients. The clinical study included 43 patients with solid tumors.
Written informed consent was obtained from all patients. They had normal blood cell counts and normal liver and renal function prior to entering the trial. Brequinar sodium was administered as a short term (10–60 min) i.v. infusion every 3 weeks in the dose range of 15–2250 mg/m² (23). Biochemical and pharmacokinetic studies were performed during the first chemotherapy course in 23 patients (2 women and 21 men).

Blood samples were obtained in heparinized tubes just before and after the infusion of Brequinar sodium, at 15, 30, and 60 min and 2, 4, 8, 24, 48, 96, 120, and 144 h, and were spun down immediately. Plasma was frozen at −20°C until analysis. Uridine was stable under these conditions for at least 6 months. Plasma samples were deproteinized with trichloroacetic acid and neutralized as described (24).

High Performance Liquid Chromatography Analysis of Uridine in Plasma Samples. The concentration of uridine in plasma samples was determined using reversed phase chromatography, as previously described (24). Briefly, a LiChrosorb-10 RP-18 column (length x i.d., 250 x 4 mm; particle size, 10 μm; Whatman) was eluted with 5 mm KH₂PO₄, pH 3.8, at a flow of 1 ml/min. Uridine was quantified by measurement of UV absorption at 254 and 280 nm and identified by its retention time and the 254/280 ratio.

Assay of DHO-DH in Lymphocytes. Blood samples of patients who received Brequinar sodium at doses ≥600 mg/m² and of healthy volunteers were spun down, the Buffy coat fraction containing the WBC was pipetted off and diluted 2–3 times in phosphate-buffered saline (pH 7.4), and 6 ml were layered onto 3 ml of Lymphoprep. Lymphocytes were isolated by gradient centrifugation as described earlier (25).

The interphase containing lymphocytes was washed with 10 ml phosphate-buffered saline, and the pellet was washed again with 10 ml phosphate-buffered saline. Cells were counted with a hemacytometer. With lymphocytes obtained from healthy volunteers, three procedures for the preparation of extracts were compared and the assay conditions for DHO-DH were optimized. Firstly, the lymphocyte pellet was suspended in 0.1 M Tris-HCl (pH 8.0) and immediately used for enzyme assays without further lysis. Secondly, this suspension was sonicated as described for L1210 cells (26); the third procedure included rapid freezing of the lymphocyte pellet in liquid nitrogen before suspension in the assay buffer, followed by thawing, repetition of this procedure, and suspension of the lysed lymphocytes in 0.1 M Tris-HCl, pH 8.0.

The assay for DHO-DH containing 400 μl lymphocyte extract (equivalent to 0.2–3.2 × 10⁸ cells) was initiated by addition of l-DHO (final concentration, 130 μM), performed at 37°C, and stopped after 10–30 min by addition of 40 μl 40% trichloroacetic acid. After neutralization, orotic acid was determined with high performance liquid chromatography as described (26). For determination of the Km, the final concentration of l-DHO was varied between 10 and 1000 μM and care was taken that initial rates were measured (26). For lymphocytes of patients obtained immediately or up to 1 week after treatment, longer incubation periods of up to 60 min were used, since the activity was usually very low. In addition we also increased the number of lymphocytes in the extract (up to 10⁸ cells). Protein was determined using the Bio-Rad dye-binding assay (27), with bovine serum albumin as standard.

From patient Z, who had received previous immunotherapy, we were able to obtain sufficient lymphocytes to measure the concentration of nucleoside triphosphates. This was performed as described earlier (28).

RESULTS

Plasma Uridine Concentrations in Mice. Plasma uridine concentrations were measured in control and treated mice at the same time points (Fig. 1). No variations, as compared to values found in rats (29), were observed. Brequinar sodium at the nontoxic dose of 50 mg/kg decreased plasma uridine to about 40% of initial levels after 2 h. After 24 h a partial recovery and after 7–9 days a rebound was observed. The decrease in plasma uridine levels was significant when compared with pretreatment levels in the same mouse (t test for paired samples) and with control mice (t test for unpaired samples).

Plasma Uridine Concentrations in Patients. Plasma uridine levels before treatment varied considerably between the patients. In only 2 of the 14 patients (16 courses) who received Brequinar sodium at doses of <600 mg/m² were the plasma levels of uridine decreased to values lower than 55% (data not shown). In the 9 patients (10 courses) who received Brequinar sodium at doses between 600 and 2250 mg/m², the sampling schedule was extended because of the delayed rebound in mice. In this group a significant decrease (P ≤ 0.01) of uridine was observed between 8 h and 3 days, and in 8 of 10 courses a significant rebound (P ≤ 0.01) in the uridine levels was observed, usually occurring after 2–7 days (Fig. 2). In one patient this rebound was observed with the first course (E-1) but not the second course (E-2). In one patient (Y; 2250 mg/m²), blood sampling was discontinued after 48 h. In the other patient (Z) who received the maximum tolerated dose of Brequinar sodium (2250 mg/m²), a sharp decrease in uridine levels was followed by a large rebound.

Activity of DHO-DH in Lymphocytes. The activity of DHO-DH in nontreated lymphocyte suspensions was 1.38 ± 0.40, in sonicated extracts 3.24 ± 0.68, and in extracts prepared by freeze-thawing 2.22 ± 0.15 nmol/h per 10⁶ cells (means ± SD of seven, four and three individuals). The protein content of lymphocytes was about 80 μg/10⁶ cells. It is likely that suspension of lymphocytes in assay buffer does not cause complete lysis of the cells. Thus, enzyme kinetics of DHO-DH and its inhibition by Brequinar sodium were studied in sonicated lymphocytes. The Km was about 3 μM. Activity at 130 μM L-DHO in the presence of 1.3 μM Brequinar sodium was 20% of control.

In pretreatment samples of nine patients, a large variation in DHO-DH activity was observed (Table 1, Fig. 3). Some of these samples were not treated because of the limited buffer volume in which they could be suspended. Sonication would have led to excessive heat production and (partial) inactivation of the enzyme. In subsequent samples from these patients, DHO-DH activity was also measured in nonsonicated preparations. The activity of DHO-DH was immediately completely inhibited (after all 10 courses, >95%) between 15 min and 6 h after the end of the infusion, no enzyme activity was detectable in any of the patients (Fig. 3). DHO-DH activity was below the detection limit, although a higher number of lymphocytes were used and the incubation time was longer than in pretreatment samples. In two patients some activity could be detected after 24 h. Fig. 3 also shows the duration of the inhibition and the time course in these patients.
Gl, gastrointestinal toxicity: MC, mucocutaneous toxicity; WHO, World Health Organization. Hematological toxicity

... (Thrombo). The level of lymphopenia is indicated by -, >1000 cells/mm³; +, 500-1000 cells/mm³; and -4-+.<500 cells/mm³.

... the lowest (minimum and highest (maximum) percentage compared to pretreatment levels is given. ND, not done (in this patient only DHO-DH could be assayed); given as nmol/h per 10^9 cells. The relative DHO-DH activities are given for the last day. At which activities were lower than 20% of pretreatment levels. For uridine

... times after suspension in assay buffer. Thus, at peak levels of

... residual Brequinar sodium in the cell would also be diluted several

... phocytes requires several extensive washing steps which dilute

... lymphocytes of patients was not due to residual Brequinar sodium present in blood, which would subsequently be present in the assay. Firstly, in most patients enzyme inhibition is still observed after 1 week, although Brequinar sodium was no

... patient Z) the activity of DHO-DH was back at normal levels of recovery in lymphocytes, which was variable among the patients and possibly related to toxicity. In all patients (except patient Z) the activity of DHO-DH was back at normal levels after 2 weeks. In a few patients a rebound was observed.

It can be concluded that the observed inhibition of DHO-DH in lymphocytes of patients was not due to residual Brequinar sodium present in blood, which would subsequently be present in the assay. Firstly, in most patients enzyme inhibition is still observed after 1 week, although Brequinar sodium was no longer detectable in plasma (23). Secondly, isolation of lymphocytes requires several extensive washing steps which dilute Brequinar sodium several hundred thousand-fold. Thirdly, residual Brequinar sodium in the cell would also be diluted several times after suspension in assay buffer. Thus, at peak levels of 1200 μM Brequinar sodium, its concentration in the assay mixture will be decreased to less than 2 nM, far below the Kᵢ. After 1 day, less than 0.5 nM might be present. The physiological concentration of L-DHO is not exactly known, although it has been estimated to be about 10 μM (2, 30); however, during inhibition of DHO-DH, the concentration of L-DHO may increase (2, 14).

It cannot be excluded that the apparent inhibition of DHO-DH is related to a change in cell population. The isolated cells are a mixture of mononuclear cells with more than 95% lymphocytes. However, it was not possible to determine the DHO-DH activity of subpopulations or to measure other enzymes of pyrimidine de novo biosynthesis, in order to exclude a general decrease in activity.

The observed duration of inhibition of DHO-DH may be related to the lipophilicity of the drug, which could enhance local retention of the drug in the mitochondrial membrane. It is very likely that in vivo the effective inhibition of DHO-DH in the cell is higher than we measured. We employed relatively high concentrations of L-DHO for the assay, in order to increase the detection limit. Furthermore, it can be expected that the concentration of the drug in the cell will be at least as high as in plasma. Considering the fact that after 5 days 1 μM Brequinar sodium is still present (Table 2), in vivo DHO-DH will still be inhibited to a larger extent. Table 2 shows the plasma concentrations of Brequinar sodium of those seven patients for whom either uridine or DHO-DH was measured.

Only in patient Z, with prior immunotherapy (interleukin 2 plus γ interferon), could a sufficient number of lymphocytes be obtained to measure the concentration of nucleotides before and after treatment at 2250 mg/m². A sharp depletion in the concentrations of UTP and CTP was observed after 1 day (down to 42 and 16% of control levels, respectively). A decrease in ATP and GTP concentrations was observed after 7 days (down to about 20%). After 25 days normal levels were observed.

Toxicity of Brequinar Sodium at the Various Dose Levels. Details on the toxicity of Brequinar sodium in the Phase I study have been described elsewhere (18). Here, toxic effects will be considered only in the 24 patients in whom the uridine levels and/or enzyme inhibition was determined. At dose levels lower than 600 mg/m², minor toxicity was observed. Toxicity in patients treated at doses of ≥600 mg/m² was more severe and consisted of thrombocytopenia, leukopenia (including lymphocytopenia), nausea, vomiting, mucositis, and skin rash.

The severe toxicity induced by Brequinar sodium in patients S and Z was associated with a considerable decrease in plasma uridine concentration. DHO-DH was inhibited in all patients.

![Fig. 2. Effect of Brequinar sodium on plasma uridine concentrations in patients receiving Brequinar sodium at doses from 600 to 2250 mg/m². Numbers in each subset of data, dose; arrows, end of the infusion; letters, initials of the patients; subscripts, number of the course.](image)

**Table 1** Toxic effects of Brequinar sodium during the first course of therapy at the various dose levels in relation to the biochemical effects

<table>
<thead>
<tr>
<th>Patient</th>
<th>Dose (mg/m²)</th>
<th>Uridine (%)</th>
<th>Clinical toxicity (WHO grade)</th>
</tr>
</thead>
<tbody>
<tr>
<td>E-1</td>
<td>1200</td>
<td>0.78</td>
<td>-</td>
</tr>
<tr>
<td>E</td>
<td>1200</td>
<td>1.58</td>
<td>-</td>
</tr>
<tr>
<td>H</td>
<td>1500</td>
<td>0.50</td>
<td>-</td>
</tr>
<tr>
<td>S</td>
<td>1500</td>
<td>3.20</td>
<td>-</td>
</tr>
<tr>
<td>Bo</td>
<td>1800</td>
<td>0.54</td>
<td>-</td>
</tr>
<tr>
<td>O</td>
<td>1800</td>
<td>4.80</td>
<td>-</td>
</tr>
<tr>
<td>Z</td>
<td>2250</td>
<td>1.30</td>
<td>++</td>
</tr>
<tr>
<td>L</td>
<td>2250</td>
<td>3.90</td>
<td>-</td>
</tr>
</tbody>
</table>

**DHO-DH activity**

- Minimum: 50
- Maximum: 280

**Clinical toxicity**

- Lymph: --
- Neutro: --
- Thrombo: --
- GI: --
- MC: --

**Hematological**

- Lymph: --
- Neutro: --
- Thrombo: --
- GI: --
- MC: --

Details on the toxicity of Brequinar sodium in the Phase I study have been described elsewhere (18). Here, toxic effects will be considered only in the 24 patients in whom the uridine levels and/or enzyme inhibition was determined. At dose levels lower than 600 mg/m², minor toxicity was observed. Toxicity in patients treated at doses of ≥600 mg/m² was more severe and consisted of thrombocytopenia, leukopenia (including lymphocytopenia), nausea, vomiting, mucositis, and skin rash.

The severe toxicity induced by Brequinar sodium in patients S and Z was associated with a considerable decrease in plasma uridine concentration. DHO-DH was inhibited in all patients.
Brequinar sodium inhibition of DHO-DH

Fig. 3. Extent and time course of inhibition of DHO-DH in lymphocytes of patients treated with Brequinar sodium. Baseline activity of DHO-DH of all patients before treatment was set at 100% (*). Activity after treatment was calculated relative to this baseline activity. Horizontal broken line, threshold level of 10% of initial levels; below this threshold DHO-DH was assayed (starting 15 min after the end of infusion) but was undetectable until the points indicated in the figure. Numbers, dose at which Brequinar sodium was administered; letters, initials of the patients; subscripts, number of the course.

Table 2 Pharmacokinetic parameters for Brequinar sodium at the higher dose levels

<table>
<thead>
<tr>
<th>Patient</th>
<th>Dose (mg/m²)</th>
<th>AUC (µmol h/liter)</th>
<th>Drug levels (µm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>E-I</td>
<td>1,200</td>
<td>6,215</td>
<td>1,053 44.5 4.3 3.0</td>
</tr>
<tr>
<td>Sc</td>
<td>1,200</td>
<td>7,068</td>
<td>1,073 49.5 ND NS 3.0</td>
</tr>
<tr>
<td>H</td>
<td>1,500</td>
<td>8,094</td>
<td>876 65 10 5.3</td>
</tr>
<tr>
<td>S</td>
<td>1,500</td>
<td>8,820</td>
<td>785 108.5 6 NS</td>
</tr>
<tr>
<td>Bo</td>
<td>1,800</td>
<td>16,734</td>
<td>1,228 250 5.8 1.5</td>
</tr>
<tr>
<td>O</td>
<td>1,800</td>
<td>18,538</td>
<td>978 261 17 12.4</td>
</tr>
<tr>
<td>Z</td>
<td>2,250</td>
<td>22,585</td>
<td>1,163 275 34 4.0</td>
</tr>
</tbody>
</table>

played an important role in the selection of a weekly schedule of Brequinar sodium for further clinical studies at the recommended doses of 1200, 1500, and 1800 mg/m².

For other inhibitors of pyrimidine de novo synthesis, it has also been shown that they can induce a depletion of uridine (8, 9, 21). However, the effects were less pronounced than those for the inhibition of DHO-DH. Moreover, the rebound of plasma uridine levels observed with Brequinar sodium has not been demonstrated for inhibition of ATC by PALA (8, 9, 21) and that of OPRT by pyrazofurin (21).

The inhibition of DHO-DH in lymphocytes demonstrates that Brequinar sodium enters the cells and subsequently the mitochondrion rapidly. It is unlikely that the mechanism is comparable to that of inhibition of ATC by PALA, since Brequinar sodium does not show any structural relationship to L-DHO. The duration of the inhibition of ATC in leukocytes of patients treated with PALA (10) is related to the tight binding of PALA to ATC (7). It might be that Brequinar sodium is protein bound or trapped in the mitochondrial membrane due to its lipophilicity and thus retained specifically in the mitochondrion, as has been demonstrated for another antimitochondrial drug, rhodamine-123 (31). Further investigations towards a better understanding of the uptake and retention of Brequinar sodium by mitochondria leading to inhibition of DHO-DH are warranted.

DHO-DH has not been measured previously in lymphocytes. According to literature data on other pyrimidine de novo synthesis enzymes (20, 32), DHO-DH activity is in the same range as that of carbamyl-phosphate synthetase II (20, 33), considerably lower (100 times) than that of ATC (10, 34), and 10 times lower than dihydroorotase (33). DHO-DH activity in lymphocytes is higher than that of OPRT (32), which has also been observed in rat liver (3, 34). In lymphocytes it is likely that, in vivo, the de novo pyrimidine synthesis pathway does not add significantly to nucleotide synthesis and that nucleotide supply via salvage pathways is more important; the rate of [14C]HCO₃⁻ incorporation into pyrimidine nucleotides is very low (35, 36), while uridine salvage was considerable (33, 38, 39). So, inhibition of the de novo synthesis pathway will not easily cause damage to the cells, which makes lymphocytes very suitable systems to measure inhibition of a de novo synthesis enzyme. Furthermore, lymphocytes are easily accessible cells with mitochondria. Other cells and tissues are more difficult to obtain.

In contrast, several tissues such as liver (40) have a high capacity to synthesize new nucleotides. We observed only a relatively small depletion of pyrimidine nucleotides in murine tissues after treatment with Brequinar sodium (41). This might be explained by the salvage of the relatively high uridine level which is present in tissues (42, 43). DHO-DH in these tissues was inhibited more than 90% and this lasted 4 days. After 7 days a rebound of DHO-DH activity in tissues was observed (41), which may contribute to an overproduction of nucleotides via the de novo synthesis pathway and the subsequent rebound of plasma uridine. These data indicate that DHO-DH plays an essential role in nucleotide synthesis, although overall knowledge about the role of DHO-DH is scarce. Inhibition of DHO-DH is more pronounced under hypoxic conditions (44). So, DHO-DH might be rate limiting for the synthesis of pyrimidine nucleotides in hypoxic tumors and can be an attractive target for cancer chemotherapy.

It appears that the rebound effect of uridine was observed predominantly in patients treated at doses of ≥600 mg/m². In only 2 of 10 courses given at lower doses was a clear rebound found (data not shown). In one of these courses the plasma
concentration of Brequinar sodium after 2 days was still >5 μM, while in the other patients treated at doses of ≤300 mg/m² these levels were lower than 1 μM after 2 days. It might be possible that the plasma levels of Brequinar sodium achieved at doses ≥60 mg/m² exceed a threshold level required for a manifestation of the observed biochemical effects. In addition, in most patients detectable levels of DHO-DH are observed after 4 days, possibly high enough to support pyrimidine de novo synthesis, which can contribute to enhanced synthesis of uridine.

In all patients administration of Brequinar sodium led to a clear effect on pyrimidine metabolism, which was most pronounced in patients with the most severe toxicity. Especially in those patients in which both DHO-DH activity and plasma uridine levels were affected, toxicity was more severe. Monitoring such biochemical parameters may be a valuable approach in the improvement of individual patient treatment.

In conclusion, we could evaluate the in vivo biochemical effects of a new inhibitor of DHO-DH, Brequinar sodium. This drug shows biological activity in vivo in patients, as shown by its effect on plasma uridine levels. We demonstrated that the target enzyme, DHO-DH, is inhibited in lymphocytes for at least 1 week in patients receiving the drug near the maximum tolerated dose. The retention (>1 week) and extent of inhibition (>90%) appear to be related to the toxic effects.

ACKNOWLEDGMENTS

We thank Dr. J. Wagstaff for critical reading of the manuscript.

REFERENCES


In Vivo Inhibition of the Pyrimidine de Novo Enzyme Dihydroorotic Acid Dehydrogenase by Brequinar Sodium (DUP-785; NSC 368390) in Mice and Patients


Updated version
Access the most recent version of this article at:
http://cancerres.aacrjournals.org/content/50/15/4644

E-mail alerts
Sign up to receive free email-alerts related to this article or journal.

Reprints and Subscriptions
To order reprints of this article or to subscribe to the journal, contact the AACR Publications Department at pubs@aacr.org.

Permissions
To request permission to re-use all or part of this article, use this link http://cancerres.aacrjournals.org/content/50/15/4644. Click on "Request Permissions" which will take you to the Copyright Clearance Center's (CCC) Rightslink site.