Irinotecan Combined with Gemcitabine, 5-Fluorouracil, Leucovorin, and Cisplatin (G-FLIP) is an Effective and Noncrossresistant Treatment for Chemotherapy Refractory Metastatic Pancreatic Cancer

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ABSTRACT

Background. Single agents have only modest activity as treatment for metastatic pancreatic cancer with response rates of less than 10% and median survivals of less than 6 months. Evaluations of single-agent gemcitabine and rubitecan as second-line treatment for relapsed pancreatic cancer have reported good patient tolerability and median survivals of 3.85 months and 4.7 months, respectively. Regimens incorporating two drugs have demonstrated encouraging activity and clinical impact compared with single-agent therapy. G-FLIP is a regimen designed to incorporate four active single agents into a tolerable and active combination. This analysis is a retrospective evaluation of the efficacy and safety of the G-FLIP regimen as second-line chemotherapy in a series of consecutively treated patients with metastatic pancreatic cancer.

Methods. G-FLIP was administered over 48 hours and repeated every 2 weeks. Day 1 treatment consisted of sequentially administered gemcitabine 500 mg/m², irinotecan 80 mg/m², leucovorin 300 mg, 5-fluorouracil (5-FU) 400 mg/m² bolus followed by infusional 5-FU 600 mg/m² over 8 hours. Day 2 treatment consisted of leucovorin 300 mg and 5-FU 400 mg/m² bolus, followed by cisplatin 50 to 75 mg/m², and then infusional 5-FU 600 mg/m² over 8 hours.

Results. Thirty-four patients with histologically confirmed metastatic pancreatic cancer were consecutively treated. The median patient age was 64.5 years (range 41-82 years) and all patients had objective disease progression on prior therapy: 32 patients had disease progression with gemcitabine and 31 had disease progression with a gemcitabine/5-fluorouracil/cisplatin combination. Grade 3-4 hematological toxicities included anemia (23%), thrombocytopenia (53%), and neutropenia (38%). There were no grade 3-4 neutropenic fevers, treatment-related mortalities, or withdrawals. Nonhematological grade 3-4 toxicities were rare: nausea/vomiting (3%), neurotoxicity (3%), nephrotoxicity (6%), and diarrhea (3%). Based on RECIST criteria a partial response (PR) was attained in eight patients (24%) and seven patients had stable disease (SD). Seven and six patients who attained a PR or SD, respectively, had disease progression with prior gemcitabine-based therapy. The median time to disease progression for all 34 patients was 3.9 months and 5.9 months for the eight patients who attained a PR. Median overall survival for all 34 patients was 10.3 months.

Conclusion. Adding a single new drug such as irinotecan to the same first-line chemotherapy combination upon disease progression may be an important alternative to switching to different drug classes for treatment of relapsed/resistant cancer. The promising clinical outcomes and moderate toxicity associated with G-FLIP in this heavily pretreated group warrant development of this novel regimen including tests as first-line therapy in patients with diseases likely to be responsive to the drugs contained in this combination. The Oncologist 2001;6:488-495

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median survival of 5.6 months, and 1-year survival of 18% [1]. Alternative single agents have been investigated in patients with metastatic pancreatic cancer. Irinotecan (CPT-11, Camptosar) is well tolerated and associated with response rates of close to 10% in both weekly and once every 3 week schedules [2, 3].

The feasibility and efficacy of most conceivable doublet combinations of gemcitabine, irinotecan, cisplatin, and 5-fluorouracil (5-FU) have been demonstrated in patients with metastatic pancreatic cancer. Phase II trials of these doublets consistently have reported response rates and median survivals that exceed the outcomes associated with single-agent therapy. Irinotecan in combination with gemcitabine has been associated with response rates of 15% to 20%, time to tumor progression (TTP) of 30 weeks in one trial, and median survival of at least 6 months [4]. Response rates of 12% to 30% and median survivals of 8 or more months have been attained with gemcitabine/cisplatin combinations [5]. Gemcitabine/5-FU and 5-FU/cisplatin combinations have been associated with CBRs ranging from 50% to 60%, stable disease (SD) and partial response (PR) rates varying from 20% to 65%, TTP ranging from 12 to 30 weeks, and median survivals of up to 11 months [6-10].

Options for patients with relapsed pancreatic cancer are of limited benefit. Evaluations of single-agent gemcitabine or rubitecan salvage therapies for metastatic pancreatic cancer have reported good patient tolerability but median survivals of only 3.85 and 4.7 months, respectively [11, 12].

Based on the reported interaction among all of these drugs given as doublets, we hypothesized that a four-drug combination of gemcitabine, 5-FU/levucovorin, cisplatin, and irinotecan (G-FLIP) would be an effective salvage regimen for patients whose disease had progressed on gemcitabine-based regimens. Laboratory observations supported this hypothesis. Our laboratory compared combinations of gemcitabine, cisplatin, 5-FU, mitomycin C, and irinotecan in ex vivo untreated gastrointestinal tumors using an adenosine triphosphate inhibition assay. Favorable drug interactions were demonstrated between gemcitabine coupled with either cisplatin, irinotecan, or 5-FU [13]. The G-FLIP regimen was designed to approximate sequence-dependent synergistic or additive interactions while attempting to minimize sequence-dependent toxic effects among the four drugs. The dosages, schedule, and sequence were based on preclinical as well as phase I and II trials of the components as doublets [4-10]. This analysis also investigated whether resistance to an active up-front regimen could be reversed by inserting irinotecan rather than switching therapy to entirely different drug classes.

**Patients and Methods**

**Patients**

This retrospective analysis examined the outcome of 34 consecutive patients treated with G-FLIP. All patients had histologically confirmed metastatic adenocarcinoma of the exocrine pancreas. Institutional Review Board exemption was granted for this study, which used existing data without associated patient identifiers. Patients were treated between April 1999 and September 2000. All patients had measurable disease with at least one lesion measurable by computed tomography (CT). Measurable objective disease progression on prior chemotherapy was documented in all patients.

**Toxicity and Response Criteria**

Patients were assessed with weekly CBCs, liver function profiles, serum electrolytes, and serum creatinine while receiving treatment. History and physical exam were performed prior to each biweekly chemotherapy course. Results of these laboratory and clinical evaluations were graded according to the Common Toxicity Criteria of the National Cancer Institute Version 2.0.

CT scans of the chest, abdomen, and pelvis as well as tumor markers (CA 19-9, carcinoembryonic antigen, and CA-125) were routinely obtained after every third or fourth chemotherapy course or sooner if disease progression was clinically suspected. Responses were graded according to RECIST criteria. A measurable target lesion must have been identified and have been at least 10 mm by spiral CT scan in one dimension in order for RECIST criteria to have been applied. RECIST response criteria take into account the measurement of only the longest diameter of all target lesions. Per RECIST criteria, complete response was defined as the disappearance of all target and non-target lesions, no new lesions, and normalization of all tumor markers. A PR was defined as at least a 30% decrease in the sum of the longest diameters of the target lesions from baseline, non-progressive disease in the nontarget lesions, and no new lesions. Tumor markers may have remained above normal. Progressive disease was defined as at least a 20% increase in the sum of the longest diameters of the target lesions taking as reference the smallest sum of long diameters recorded since treatment started or the appearance of new lesions. Tumor markers may have remained above normal. Progressive disease was defined as at least a 20% increase in the sum of the longest diameters of the target lesions taking as reference the smallest sum of long diameters recorded since treatment started or the appearance of new lesions. Tumor markers may have remained above normal. Progressive disease was defined as at least a 20% increase in the sum of the longest diameters of the target lesions taking as reference the smallest sum of long diameters recorded since treatment started or the appearance of new lesions. Tumor markers may have remained above normal.
Treatment

The G-FLIP treatment schema is outlined in Figure 1. Day 1 treatment consisted of sequentially administered gemcitabine 500 mg/m², irinotecan 80 mg/m², and then leucovorin 300 mg, 5-FU 400 mg/m² bolus followed by infusional 5-FU 600 mg/m² over 8 hours. Day 2 treatment consisted of leucovorin 300 mg and 5-FU 400 mg/m² bolus, followed by cisplatin 50 to 75 mg/m², and then infusional 5-FU 600 mg/m² over 8 hours. Previously treated patients who had developed National Cancer Institute (NCI) grade 3 or worse thrombocytopenia were assigned to receive cisplatin 50 mg/m². All other patients received cisplatin 75 mg/m². Treatment was repeated every 14 days. Kytril 2 mg orally and decadron 10 mg i.v. were given as antiemetic prophylaxis 30 minutes prior to chemotherapy on days 1 and 2. Intravenous normal saline at 200 cc/hour for 4 hours with lasix 10 mg i.v. was given to ensure a urine output of at least 100 cc/hour prior to cisplatin administration. Normal saline hydration was continued for 6 hours at 125 cc/hour after the completion of cisplatin.

Patients were evaluated weekly for nonhematological toxicity and once or twice weekly for hematological toxicity. Chemotherapy was dose reduced or discontinued if intolerable toxic side effects developed. Dose reductions and/or dose deletions were tailored to address toxicities specific to a particular drug. In particular, renal toxicity, vomiting, and neurotoxicity prompted cisplatin dose reduction/deletion, and mucositis prompted modification of 5-FU dosages. Filgrastim and erythropoietin were used as required to promote dose intensity.

Statistics

Survival and TTP were measured from the date G-FLIP treatment was initiated. Survival and TTP curves were constructed using Kaplan-Meier estimates.

RESULTS

Pretreatment characteristics are summarized in Table 1. The median patient age was 64.5 years (range: 41-82) and 25 patients were men. All patients had metastatic disease with at least one lesion measurable by CT. Most patients had extensive metastatic spread: 26 patients had liver metastases, five had lung metastases, five had peritoneal metastases, and 15 patients had more than one site of metastatic disease. Most patients were heavily pretreated (Table 2). All patients had measurable objective disease progression on prior chemotherapy: 32 patients had failed gemcitabine and 31 had disease progression while receiving a combination of gemcitabine, 5-FU, and cisplatin (GFP). The GFP regimen was identical to the G-FLIP regimen described in Figure 1 but without irinotecan.

Treatment administration is summarized in Table 3. A total of 476 cycles of G-FLIP were administered. The median number of cycles per patient was five (range: 1-25). The median dose intensities for each drug expressed as mg/m²/2 weeks were as follows: gemcitabine 433, irinotecan 60, 5-FU bolus 600, 5-FU infusion 1,200, and cisplatin 36.

Grade 3-4 toxic side effects were largely hematological and are summarized in Table 4. Grade 3-4 toxicities per

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Figure 1. Gemcitabine, 5-FU/leucovorin, irinotecan, and cisplatin (G-FLIP).

<table>
<thead>
<tr>
<th>Table 1. Patient characteristics on study entry</th>
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<td>Total patients</td>
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<td><strong>Sex</strong></td>
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<tr>
<td>Female</td>
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<td><strong>Median age (years/range)</strong></td>
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<tr>
<td><strong>Metastatic disease</strong></td>
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<td><strong>Sites of metastases</strong></td>
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<td>Peritoneum</td>
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<td>Other</td>
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<td>Two or more sites</td>
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<th>Table 2. Prior systemic therapies</th>
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<td>Total patients</td>
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<tr>
<td><strong>Treatment</strong></td>
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<tr>
<td>Gemcitabine</td>
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<td>5-fluorouracil/LV</td>
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<tr>
<td>Gemcitabine/irinotecan</td>
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<td>Gemcitabine/cisplatin</td>
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<td><strong>Combinations</strong></td>
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<td>GFP</td>
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GFP = gemcitabine, 5-FU/leucovorin, and cisplatin
patient included thrombocytopenia (53%), neutropenia (38%), and anemia (23%). These toxicities were easily managed and there were no grade 3-4 neutropenic fevers or hemorrhagic events. Grade 3-4 nonhematological toxic side effects were rare. Grade 3-4 paresthesias occurred in 3% of patients, nephrotoxicity occurred in 6%, vomiting in 3%, and diarrhea in 3%. Less severe grade 1-2 nonhematological toxicities occurred more frequently and included vomiting (47%), diarrhea (18%), mucositis (12%), paresthesias (24%), and nephrotoxicity (12%).

Response and survival outcomes are summarized in Table 5. Eight patients attained a PR, seven patients had disease stabilization, and 19 had disease progression. The median TTP for all 34 patients was 3.9 months (Fig. 2). TTP for the eight patients who attained a PR was 5.9 months. Median overall survival measured from the start of G-FLIP was 10.3 months (Fig. 3). Thirty-one patients received G-FLIP after demonstrated disease progression on GFP. In this sequentially treated group there were seven patients who each attained either a PR or stable disease (Table 6). Median TTPs were 2.3 months for all 31 patients and 5.8 months for the seven patients who attained a PR. Measured from the initiation of GFP, the median survival for the 31 sequentially treated patients was 11.8 months, 1-year survival was 47%, and 2-year survival was 24%. Eight of the 14 patients who attained a PR or SD with G-FLIP had disease progression as a best response to prior GFP (Table 6).

**DISCUSSION**

Gemcitabine is the first drug to be approved for the treatment of pancreatic cancer on the basis of clinical benefit. Gemcitabine’s favorable toxicity profile and numerous
cellular targets such as inhibition of ribonucleotide reductase, DNA repair mechanisms, and thymidylate synthase make it an excellent candidate for use in combination therapy. Gemcitabine-based doublets with 5-FU, cisplatin, and irinotecan are feasible and there is a consistent suggestion of improved response rates, response duration, overall survival, and quality of life compared with contemporary reports of gemcitabine alone [4-10]. Palliative chemotherapy options for relapsed disease have minor clinical benefit. Single-agent second-line therapy for metastatic pancreatic cancer following gemcitabine failure has been associated with a median survival of less than 5 months [12].

The 1- and 2-year survival outcomes associated with the sequential use of GFP and G-FLIP compare favorably with both single-agent and combination therapies. While quality-of-life assessments were not done, the majority of grade III and IV toxic side effects were hematological, easily managed, and did not translate into complications requiring hospitalization or compromised performance status. The tolerability and efficacy of the G-FLIP regimen may in part be due to the rational sequencing of the drugs. G-FLIP was designed to approximate the known schedule-dependent synergistic relationships among the four drugs and to minimize sequence-dependent toxic effects.

Irinotecan was administered prior to 5-FU in the G-FLIP regimen. In cell cultures, 5-FU sequenced before or administered simultaneously with SN-38 (the active metabolite of irinotecan) demonstrated antagonistic activity. However, additive antitumor activity was seen when 5-FU followed SN-38 administration [14, 15]. Furthermore, the sequence of 5-FU following irinotecan has been associated with less diarrhea and neutropenia [16]. Pharmacodynamics, kinetics, and toxic side effects were not affected by the sequence of irinotecan-cisplatin administration [17, 18].

Pharmacokinetic (PK) studies have not demonstrated a difference between sequences of gemcitabine and cisplatin when the two drugs are given 4 hours apart. However, gemcitabine 24 hours prior to cisplatin was associated with an increase in 24-hour retention of platinum-DNA adducts [19]. Clinically, cisplatin 24 hours prior to gemcitabine was associated with significantly more leukopenia than the reverse sequence. Nonhematological toxicity consisted of grade 1-2 nausea, vomiting, and fatigue, and was schedule independent [20].

Data regarding sequence dependent efficacy or toxicity between gemcitabine and irinotecan are limited. Preliminary PK studies of a trial evaluating weekly irinotecan and gemcitabine showed no sequence-dependent differences. The maximal tolerated dose (MTD) when gemcitabine was followed by irinotecan was 1,000 and 60 mg/m², respectively. The MTD of the reverse sequence has not been determined [21].

In vitro studies support the sequence-dependent synergistic relationship between 5-FU and cisplatin. Using a HST-1 human squamous carcinoma cell line, 5-FU followed by cisplatin was more active than the reverse sequence. Synergistic activity was maximal when the interval between 5-FU and cisplatin administration was at least 24 hours. A significant reduction in DNA interstrand cross-link removal occurred in cells exposed to 5-FU 48 hours prior to cisplatin compared with cells exposed to cisplatin alone or immediately preceded by 5-FU. This finding suggests that 5-FU modulates the repair of platinum-DNA adducts thereby potentiating antitumor activity [22].

There have been no published analyses comparing sequences of 5-FU and gemcitabine. As a potential inhibitor of ribonucleotide reductase, gemcitabine may inhibit the formation of 2-deoxyuridine-5-monophosphate (dUMP) thereby enhancing the activity of 5-FU and leucovorin.

The response and survival outcomes associated with G-FLIP in this analysis of patients with previously treated adenocarcinoma of the exocrine pancreas exceed outcomes that would be expected with irinotecan alone. The reinduced
efficacy of gemcitabine, 5-FU, and cisplatin as second-line therapy in this series challenges the traditional practice of switching to different drug classes upon disease progression. Reinducible activity may be due to drug-class-independent mechanisms such as activation of blocked apoptotic pathways. Defects in the core of apoptosis, the caspase cascade, permit tumor cells to live despite an accumulation of chemotherapy-induced chromosomal and microtubule injuries [23]. Despite defective caspase pathways, chemotherapy still may cause apoptotic cell death by mitochondrial damage or activation of stress activated protein kinases (SAPK) [24]. Whether these events are independent of drug-class specific mechanisms of action and induced by adding a drug to a first-line treatment is a theoretical consideration.

Complementing these preclinical observations is emerging clinical evidence that modifying an upfront chemotherapy regimen by inserting an additional drug or altering administration schedule may overcome apparent resistance. Gutierrez et al. recently demonstrated the efficacy of retrying upfront agents via a prolonged intravenous infusion schedule rather than utilizing a salvage regimen of noncrossresistant drugs for relapsed and resistant non-Hodgkin’s lymphoma (NHL) [25]. Of 125 assessable patients, 24% attained a complete response and 50% attained PRs with infusional EPOCH (etoposide, doxorubicin, vincristine via 96-hour infusion, bolus i.v. cyclophosphamide, and prednisone). Fifty-seven percent of patients whose disease had no response to last chemotherapy (resistant disease) responded to EPOCH and nearly 20% of patients with aggressive NHL had durable responses, disease free at 36 months. The use of a fifth “new” drug added to four “upfront” drugs as well as use of a prolonged infusion schedule were associated with partial reversal of drug resistance. By comparison, second-line G-FLIP utilized a sequence and schedule of gemcitabine, 5-FU, leucovorin, and cisplatin identical to first-line combination GFP. The only modification was the addition of irinotecan, inserted after gemcitabine on day 1.

Similar reinducible activity was seen when oxaliplatin was added to the same 5-FU/leucovorin schedule upon progression of metastatic colorectal cancer. Oxaliplatin as a single agent has demonstrated response rates of 10% in patients with 5-FU resistant disease [26]. However, when oxaliplatin was added to the same 5-FU/leucovorin regimen upon disease progression, the response rate was 46% [27].

Irinotecan may overcome specific chemotherapy-tolerance mechanisms. Some cisplatin-resistant human ovarian cancer cell lines have an increased ability to synthesize DNA beyond platinum adducts [28]. Irinotecan may overcome this mechanism of cisplatin resistance by inhibiting DNA replication/repair complexes from working past or beyond platinum adducts. Irinotecan may also reverse platinum efflux mechanisms thereby restoring cisplatin sensitivity. The canalicular multi-specific organic anion transporter (cMOAT) has been shown to be associated with decreased cisplatin accumulation and sensitivity and is believed to act as an active efflux pump for platinums [29]. Decreased cMOAT expression has been associated with a significant increase in cisplatin sensitivity [30]. Interestingly, SN-38, the active metabolite of irinotecan, is primarily excreted by cMOAT [31, 32]. Irinotecan use may therefore saturate the platinum efflux pump and restore cisplatin sensitivity.

The addition of a new agent to the “upfront” drugs in the above second-line regimens may have precipitated irreversible mitochondrial damage or activated SAPK to overcome defective apoptotic pathways. Our analysis provides additional support to the concept that retrying an upfront regimen by adding a single new drug may be an important alternative to switching to different drug classes for treatment of relapsed or resistant cancer. Alternatively, irinotecan may overcome pancreatic cancer drug-specific resistance mechanisms to either 5-FU, gemcitabine, or cisplatin.

Three- and four-drug regimens have recently reported good patient tolerability and clinical benefit and deserve further evaluation as front-line therapy for patients with metastatic pancreatic cancer. Cisplatin, epirubicin, gemcitabine, and 5-FU continuous infusion has been associated with World Health Organization criteria PRs in 69% and minor response/disease stabilization in 23% of 26 evaluable patients. Median survival and TTP had not been reached at 8+ and 5+ months, respectively [33]. A 26% response rate, 39% CBR, median disease-free survival of 5.4 months, and median overall survival of 6.9 months has been attained with 5-FU bolus and infusion, leucovorin, gemcitabine, and oxaliplatin [34]. Further prospective trials will explore the use of the G-FLIP regimen as initial therapy for patients with metastatic pancreatic cancer. In addition, the feasibility and efficacy of G-FLIP in gastric cancer, cholangiocarcinomas, and other bile duct tumors will be explored.

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