Vasopressin Modulation of Peritoneal, Lymphatic, and Plasma Drug Exposure following Intraperitoneal Administration

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ABSTRACT

i.p. administration of cytotoxic drugs for the treatment of regionally confined cancers results in a greater total drug exposure [area under the concentration × time curve (AUC)] for the peritoneal fluid and regional lymphatics than for plasma. We sought to augment the relative advantage of i.p. administration further through modulation of peritoneal clearance by reduction in splanchnic blood flow.

Pigs were treated with 5-fluorouracil, etoposide (VP-16), and carboplatin (CBDCA) alone by the i.p. route or with the same drugs in combination with i.v. lypressin, a synthetic vasopressin analogue, which reduces splanchnic blood flow. Drug concentrations in peritoneal fluid, plasma, and thoracic lymph were monitored over the ensuing 6 h.

The pharmacokinetics of 5-fluorouracil were not altered by vasopressin; however, vasopressin increased the peritoneal fluid:plasma AUC ratio for CBDCA from 30.6 ± 5.6 to 70.6 ± 7.4 (P < 0.01) and increased the lymph:plasma AUC ratio from 1.1 ± 0.4 to 2.6 ± 0.22 (P < 0.05). In the case of VP-16, vasopressin increased the peritoneal fluid:plasma AUC ratio from 129 ± 35 to 350 ± 76 (P < 0.05) and the lymph:plasma AUC ratio from 2.1 ± 0.6 to 10.6 ± 3.5 (P < 0.05).

 Concurrent i.v. administration of vasopressin can increase the pharmacokinetic advantage of the i.p. route of administration of CBDCA and VP-16 markedly in the pig model. These data suggest that the strategy of concurrent i.p. administration of CBDCA or VP-16 plus an agent that reduces splanchnic blood flow may increase the dose intensity in the abdominal cavity and intraabdominal lymphatic tissue substantially without increasing systemic toxicity.

INTRODUCTION

The rationale for i.p. administration of cytotoxic drugs for the treatment of cancers confined to the peritoneal cavity is that it can yield a higher total drug exposure (AUC) for the peritoneal cavity than for plasma and thereby a potentially greater therapeutic efficacy for a given degree of systemic toxicity. The peritoneal:plasma AUC ratio for 5-FU ranges from 117 to 1066 (1–3). For CBDCA, the ratio is in the range of 15 (4), and for VP-16, it is ~65 (5). We have demonstrated previously that in the pig model, the high concentrations attained in the peritoneal cavity are accompanied by high concentrations of a drug in the thoracic lymphatics, and that this is particularly true for 5-FU (6). i.p. instillation of 5-FU resulted in a 5.7-fold increase in total drug exposure in the thoracic lymph relative to that for plasma (P < 0.05). In contrast, i.p. installation of VP-16 produced only a 2.1-fold increase in lymphatic exposure, and administration of CBDCA by this route resulted in no increased lymphatic AUC relative to the AUC for the plasma.

The concentration of a drug in the peritoneal cavity relative to that in the plasma at steady state is a function of the clearance of the drug from the peritoneal cavity primarily via the splanchnic circulation (7), and thus, one strategy for improving the pharmacological advantage of i.p. therapy further is to decrease splanchnic blood flow. Vasopressin is well known for its ability to decrease splanchnic blood flow selectively in man (8), making it an attractive agent for use in combination with i.p. administration of chemotherapeutic agents. The aim of this study was, in a model system, to determine the effect of concurrent i.v. administration of the vasopressin analogue lypressin on the pharmacokinetics of drugs representative of three classes of agents used commonly by the i.p. route in patients with ovarian carcinoma and gastrointestinal cancers.

MATERIALS AND METHODS

Animals and Procedures. The studies were carried out in pigs (Swedish land breed and Swedish Yorkshire) weighing 26–40 kg; there were six pigs in the experimental group, and eight animals presented previously (6) were used as controls. Anesthesia was induced by i.m. injection of 15–20 mg/kg ketamine (Parke-Davis, Morris Plains, NJ). After 10 min, 2 mg/kg methomidate chloride (A. B. Leo, Helsingborg, Sweden) and 0.05 mg/kg azaperon (Janssen Pharmaceutica, Berse, Belgium)
were injected i.v. The pigs were then placed in a supine position, intubated via the oropharyngeal route, and ventilated on a respirator with room air. Anesthesia was maintained by repeated i.v. injections of 2–3 mg/kg methomidate chloride and 0.05–0.10 mg/kg azaperon. The thoracic duct was exposed through a right-sided thoracotomy, identified, and cannulated with a tube of outer diameter 1.27 mm, the tip of which was positioned close to the point where the duct passes through the diaphragm. A separate catheter was placed in the right axillary artery and used for continuous arterial pressure monitoring and sampling, and another catheter was placed in the jugular vein. Approximately 30 min after catheterization, a tube of outer diameter 3.6 mm was inserted in the abdominal cavity from the region above the umbilicus for drug instillation, and a 2.5-mm tube was inserted into the peritoneal cavity from the left or right lower flank for sampling of peritoneal fluid. A continuous i.v. infusion of 5% glucose, 20 ml/kg/h, was maintained throughout the experimental period. Both body temperature and mean arterial blood pressure were found to remain stable and within normal limits during the first 360 min of the experiment.

The six experimental animals received 0.007 international units/min/kg vasopressin (lycin-8-vasopressin; Ferring, Malmö, Sweden) as a continuous i.v. infusion starting 30 min prior to i.p. instillation of cytotoxic agents. The eight control animals were treated identically, except that no vasopressin was given (6). In both groups, i.p. chemotherapy was administered by first instilling into the peritoneal cavity 0.9% saline in a volume of 25 ml/kg body weight and then instilling rapidly 25 mg/kg 5-FU (Roche, Basel, Switzerland), 25 mg/kg CBDA (Bristol-Myers, Syracuse, NY), and 25 mg/kg VP-16 (Lundbeck, Copenhagen, Denmark) suspended together in 0.9% sodium chloride in a volume of 25 ml/kg. Arterial blood and peritoneal fluid samples were obtained at frequent intervals (see below); thoracic duct lymph was collected continuously by gravity drainage. The heparinized blood samples were centrifuged immediately to remove corpuscular elements. The plasma, peritoneal fluid, and lymph samples were frozen at -70°C until analyzed. At the conclusion of the experiment, the pigs were sacrificed with an overdose of saturated potassium chloride.

**Measurement of Drug Concentration and Content.** Total VP-16 was measured by high-performance liquid chromatography as described (5). Non-protein-bound platinum was determined in ultrafiltrates (Centrfree micropartition cartridge; Amicon Corp., Beverly, MA) by flameless atomic absorption spectroscopy using a Perkin-Elmer (Norwalk, CT) graphite furnace (9). 5-FU was measured in deproteinized plasma samples by high-performance liquid chromatography using absorption ratios and external standards to identify and quantitate the 5-FU peak (10). The CBDCA content of tissues was measured as elemental platinum by atomic absorption spectroscopy using external standards (9) after digestion of 200 mg tissue in 0.5 ml hyamine hydroxide for 16 h at 55°C. Samples were diluted to 5 ml with 0.15 N HCl and 25-μl aliquots were used for analysis.

**Calculations and Statistics.** The AUC of each pig for each of the three compartments (peritoneal, lymph, and plasma) was calculated for the first 180 min following i.p. drug instillation using the trapezoidal and Simpson’s rule. Although drug concentrations were measured out to 360 min, the variance associated with the data permitted accurate calculation of the AUC only to 180 min. The peak plasma concentration, the time to peak concentration, and half-life were calculated from data acquired during the first 360 min. Half-lives were calculated from the slope of the terminal portion of the decay curve determined by linear regression. Peritoneal clearance (PA) was calculated according to the formula PA = −ln(Ct/C0)(Vipt−1), where Ct is the peritoneal concentration at time t, C0 is the peritoneal concentration at time 0, Vπ is the peritoneal volume (2l), and t is time.

**RESULTS**

Following instillation of 5-FU, CBDCA, and VP-16 by the i.p. route, body temperature remained stable and within normal limits throughout the 360-min sampling period. The mean arterial blood pressure in the group receiving vasopressin infusion was significantly higher (P < 0.003) than in the control group (Fig. 1).

In the peritoneal cavity, the pharmacokinetic profiles were different for each of the three chemotherapeutic agents (Fig. 2). 5-FU reached a higher peak concentration than CBDCA or VP-16, and the drugs each had different half-lives, but the addition of vasopressin did not change the pharmacokinetic profile for any of the three drugs in the peritoneal compartment.

The pharmacokinetic profiles for each drug in the lymphatic compartment are presented in Fig. 3. Although all three drugs became measurable in the lymph shortly after i.p. instillation, the concentrations of 5-FU and VP-16 peaked within the first hour and decreased thereafter with time, whereas the concentration of CBDCA in the lymph declined more gradually.
concentration of CBDCA remained relatively constant after reaching a plateau within the first 2 h. Vasopressin administration had no effect on the peak concentration, time to peak concentration, or half-life in the lymph for 5-FU or CBDCA, but it did increase the concentration of VP-16 consistently over the whole period of sampling by a factor that averaged 164% (Table 1).

Fig. 4 shows the drug concentrations in the plasma compartment as a function of time. 5-FU appeared in the plasma compartment more rapidly than CBDCA or VP-16, but, whereas the concentration of 5-FU peaked early and declined thereafter, the concentrations of CBDCA and VP-16 continued to increase progressively throughout the 360-min sampling period. Vasopressin had no discernable effect on the plasma pharmacokinetics of 5-FU but had a clear impact on the kinetics of both CBDCA and VP-16, causing a substantial decrease in drug concentrations in both cases. The peak plasma concentration of CBDCA in the vasopressin group averaged only 33% of that in the control group ($P < 0.01$; Table 1).

Table 1 also presents information on the AUC. Vasopressin administration did not change the AUC for 5-FU for any of the three compartments; however, in the case of CBDCA, vasopressin reduced the mean AUC plasma to 24% of control ($P < 0.05$). Vasopressin had the interesting effect of increasing the AUC for VP-16 to 265% of control in the lymph ($P = 0.05$). In
Vasopressin Alters Pharmacokinetics of i.p. Drugs

Table 1  Peak drug concentrations and time to peak drug concentration, half-life, and AUC (180 min), mean ± SE.

<table>
<thead>
<tr>
<th>Drug</th>
<th>Peak drug concentration (µg/ml)</th>
<th>Time to peak drug concentration (min)</th>
<th>Half-life (min)</th>
<th>AUC (µg × min/ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td>5-FU</td>
<td>3,110 ± 390</td>
<td>30 ± 14</td>
<td>222 ± 40</td>
<td>363,000 ± 68,800</td>
</tr>
<tr>
<td></td>
<td>3,010 ± 690</td>
<td>29 ± 8</td>
<td>230 ± 36</td>
<td>313,000 ± 19,400</td>
</tr>
<tr>
<td>Lymph</td>
<td>40.0 ± 8.7</td>
<td>54 ± 17</td>
<td>171 ± 56</td>
<td>6,590 ± 3,680</td>
</tr>
<tr>
<td></td>
<td>23.5 ± 2.9</td>
<td>66 ± 16</td>
<td>174 ± 54</td>
<td>4,620 ± 780</td>
</tr>
<tr>
<td>Plasma</td>
<td>12.2 ± 5.7</td>
<td>48 ± 19</td>
<td>223 ± 78</td>
<td>1,000 ± 660</td>
</tr>
<tr>
<td></td>
<td>14.2 ± 3.2</td>
<td>83 ± 20</td>
<td>282 ± 63</td>
<td>1,150 ± 270</td>
</tr>
<tr>
<td>CBDCA</td>
<td>935 ± 174</td>
<td>40 ± 20</td>
<td>247 ± 34</td>
<td>74,900 ± 3,220</td>
</tr>
<tr>
<td></td>
<td>922 ± 142</td>
<td>30 ± 8</td>
<td>270 ± 50</td>
<td>107,000 ± 13,600</td>
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<tr>
<td>Lymph</td>
<td>24.5 ± 6.7</td>
<td>124 ± 30</td>
<td>NE</td>
<td>2,630 ± 720</td>
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<tr>
<td></td>
<td>42.4 ± 9.4</td>
<td>204 ± 42</td>
<td>NE</td>
<td>3,170 ± 950</td>
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<tr>
<td>Plasma</td>
<td>14.2 ± 2.4a</td>
<td>229 ± 49</td>
<td>NE</td>
<td>974 ± 212b</td>
</tr>
<tr>
<td></td>
<td>42.3 ± 7.3</td>
<td>226 ± 36</td>
<td>NE</td>
<td>4,040 ± 1,150</td>
</tr>
<tr>
<td>VP-16</td>
<td>731 ± 280</td>
<td>48 ± 22</td>
<td>525 ± 184</td>
<td>57,200 ± 7,920</td>
</tr>
<tr>
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<td>312 ± 58</td>
<td>45 ± 10</td>
<td>416 ± 105</td>
<td>42,400 ± 7,400</td>
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<tr>
<td>Lymph</td>
<td>12.9 ± 3.3</td>
<td>59 ± 13</td>
<td>129 ± 33</td>
<td>1,670 ± 550a</td>
</tr>
<tr>
<td></td>
<td>6.5 ± 1.2</td>
<td>42 ± 15</td>
<td>116 ± 53</td>
<td>630 ± 112</td>
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<tr>
<td>Plasma</td>
<td>2.3 ± 0.9</td>
<td>169 ± 30</td>
<td>NE</td>
<td>203 ± 56</td>
</tr>
<tr>
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<td>3.4 ± 1.0</td>
<td>161 ± 30</td>
<td>NE</td>
<td>407 ± 104</td>
</tr>
</tbody>
</table>

*ap < 0.01.

**p < 0.05

Fig. 4  Plasma concentrations as a function of time. A, vasopressin group; △, control group. Each data point represents the mean of values obtained in six to eight animals. Vertical lines, SE.

Table 2  Estimated clearance from the peritoneal cavity (ml/min)

<table>
<thead>
<tr>
<th>Drug</th>
<th>Coadministration</th>
<th>Clearance</th>
</tr>
</thead>
<tbody>
<tr>
<td>5-FU</td>
<td>Vasopressin</td>
<td>8.8 ± 3.0</td>
</tr>
<tr>
<td></td>
<td>Control</td>
<td>5.0 ± 2.0</td>
</tr>
<tr>
<td>CBDCA</td>
<td>Vasopressin</td>
<td>9.6 ± 1.8</td>
</tr>
<tr>
<td></td>
<td>Control</td>
<td>7.1 ± 1.5</td>
</tr>
<tr>
<td>VP-16</td>
<td>Vasopressin</td>
<td>5.1 ± 2.1</td>
</tr>
<tr>
<td></td>
<td>Control</td>
<td>5.0 ± 1.1</td>
</tr>
</tbody>
</table>

contrast, in the plasma, it decreased the AUC to a mean of 50% of control; however, this did not reach the level of statistical significance (P = 0.12). Table 2 presents data on the effect of vasopressin on peritoneal clearance. Vasopressin did not alter the peritoneal clearance rate for any of the drugs. These findings are consistent with the findings in Fig. 2 that vasopressin did not change the pharmacokinetic profile in the peritoneal cavity for any of the three drugs.
In addition to examining the effect of vasopressin on each pharmacokinetic parameter individually, it was of interest to determine how vasopressin altered the total drug exposure for each compartment relative to each other. Fig. 5 represents the peritoneal:plasma, peritoneal:lymph, and lymph:plasma AUC ratios for each of the three drugs. In the case of 5-FU, vasopressin had no significant effect on any of the ratios, but in the case of CBDCA, it produced a 2.4-fold increase in the peritoneal:plasma ratio and a 2.4-fold increase in the lymph:plasma ratio without altering the peritoneal:lymph ratio. A similar effect was observed with VP-16. Vasopressin increased the mean peritoneal:plasma ratio by 2.7-fold and the lymph:plasma ratio by 4.9-fold ($P < 0.05$ for both) but did not alter the peritoneal:lymph ratio significantly. Thus, for two of the three drugs examined, vasopressin increased the AUC ratio for both of the compartments at highest risk for containing malignant cells in patients with intra-abdominal cancers.

The data collected in this study permitted an indirect estimate of the effect of vasopressin on the rate of drug movement from the peritoneal cavity to the lymph of the thoracic duct, which is equivalent to the permeability of the barrier separating these two pharmacokinetic compartments. Table 3 presents data on the initial rate of rise in drug concentration in the lymph calculated from the first measured data point at 10 min. The results indicate that vasopressin did not influence the movement of the drug from the peritoneal cavity to the lymph for any of the three drugs, consistent with the observation that it did not alter the peritoneal:lymph AUC ratio for any of the agents.

**DISCUSSION**

i.p. instillation of chemotherapeutic agents is attractive in the treatment of patients with cancers confined to the peritoneal cavity because of the potential for exposing the peritoneal cavity to far higher concentrations and dose intensities than can be attained if the drugs are given by the i.v. route. The magnitude of the pharmacokinetic advantage varies markedly among different types of chemotherapeutic agents (11), and the variations can be linked directly to differences in the rates of clearance of the agent from the peritoneal cavity relative to clearance from the plasma (7). Our previous pharmacokinetic studies in the pig also demonstrated that for some agents, notably 5-FU, i.p. instillation also increases drug exposure for the lymph markedly when measured at the level of the diaphragm (6). What portions of the lymphatic drainage that receive the greatest drug exposure following i.p. instillation are unknown. High concentrations might be anticipated in the lymphatics of those portions of the gut that reside in the true peritoneum and in the specialized lymphatics of the undersurface of the diaphragm (12). With increasing distance from the peritoneal surface, a decreasing drug exposure can be anticipated.

In the series of experiments reported here, we have sought to improve the pharmacological advantage of i.p. chemotherapy further through pharmacological manipulation of splanchnic blood flow.

The clearance of small solutes from the peritoneal cavity is influenced by splanchnic blood flow. A 100% increase in flow has been reported to result in a 30–50% increase in drug clearance from the peritoneal cavity (13), and a decrease in blood flow would be expected to have the opposite effect (7). i.v. administration of vasopressin can reduce the portal blood flow up to 50% in man (8) and is attractive as a candidate for pharmacological modulation of peritoneal clearance because of its long history of use in association with gastrointestinal bleeding. Our results indicate that the concurrent administration of i.v. vasopressin does alter the compartmental pharmacokinetics of CBDCA and VP-16 but has no effect on 5-FU. The effects on CBDCA and VP-16 are most easily observed as changes in the ratios of total drug exposure (AUC) for the peritoneal and lymphatic compartments relative to that for plasma. The magnitude of the effects of vasopressin for the AUC ratios for CBDCA and VP-16 is substantial, >2-fold in each case. Stated another way, for a given degree of systemic exposure, the concurrent administration of vasopressin resulted in more than a doubling of dose intensity for the peritoneal cavity and, by inference, for those lymphatics lying between the peritoneal surface and the lower thoracic duct. A possible way to increase the efficacy of vasopressin further could be to instillate it directly into the peritoneal cavity together with the cytotoxic drug.

The nature of the pharmacokinetic changes produced by vasopressin could not have been predicted *a priori* and are not explained completely by the data gathered in this study. Fig. 6 presents a model of drug movement between the various pharmacokinetically defined compartments. Certainly, differences in the drugs themselves contribute to the differential effect of vasopressin. 5-FU is a small molecule with a $M_r$ of 130,000, which is <10% protein bound in plasma and cleared almost completely (85–95%) by degradation in the liver. In contrast, CBDCA is a larger molecule with a $M_r$ of 371,000, it is more extensively protein bound, and most of the drug is excreted unchanged in the urine. VP-16 has a $M_r$ of 588,000, is bound extensively to plasma proteins (94–97%), and only ~45% of it is excreted in the urine (14). Based on these characteristics, it is likely that the clearance of 5-FU from the peritoneal and lymphatic compartments is substantially less dependent on splanchnic blood flow than the clearance of CBDCA and VP-16.

The major effect of vasopressin was to increase the peritoneal:plasma and lymph:plasma AUC ratios for CBDCA and VP-16. The model in Fig. 6 indicates that this could result from either a decrease in clearance from the peritoneal cavity or lymph or an increase in clearance from the plasma. However, although it was not possible to detect any decrease in clearance from the peritoneal cavity after vasopressin, a small effect could be masked by the variation in the data. The peak concentration in the peritoneal cavity was much higher than in lymph and plasma (20–70 times higher for CBDCA and 50–320 times higher for CBDCA). Thus, very small changes in the peritoneal clearance can explain the changes in the peritoneal:plasma ratio, even if we cannot rule out an increase in clearance from the plasma. Because anesthesia was applied identically both in the vasopressin and in the control group, it seems unlikely that the changes caused by vasopressin are influenced by anesthesia. In a clinical setting it is less likely that anesthesia would be used.

The overall therapeutic benefit of i.p. chemotherapy remains unproven, but many pilot studies have demonstrated the feasibility of producing responses in patients who have failed systemic chemotherapy with the same agents, and a large,
Fig. 5 Effect of vasopressin administration on AUC ratios for 5-FU (A), CBDCA (B), and VP-16 (C). *, P < 0.05; **, P < 0.01 relative to the control group.
randomized trial of i.p. versus i.v. chemotherapy for the treatment of optimally debulked ovarian cancer has demonstrated both increased survival and reduced toxicity in the i.p. arm (15). The major limitation of i.p. therapy is that penetration into tumor nodules following i.p. administration of even very large doses is still limited for some drugs (16), and the therapeutic effect is still likely to be small if there are tumor nodules >1 cm remaining in the peritoneal cavity (17). However, i.p. chemotherapy remains pharmacologically attractive, particularly in the adjuvant setting, in part because recent work has demonstrated that it not only results in the delivery of extremely high drug concentrations to the peritoneal cavity, but also increases regional lymphatic exposure significantly for some agents (6). The further increase in the pharmacological advantage attainable with the addition of vasopressin in the pig model suggests this as a therapeutic strategy in patients in whom there is a high risk of regional lymphatic micrometastases, such as early stage gastric or pancreatic carcinoma.

Acknowledgments

We thank Dr. Tor Skärby (Department of Clinical Pharmacology, University of Lund, Lund, Sweden) for fruitful discussion on pharmacokinetic results.

REFERENCES

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